U.PORTO

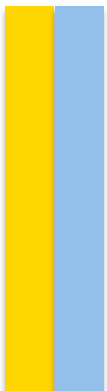
DOUTORAMENTO
CIÊNCIAS BIOMÉDICAS

Facilitated Genome Editing as Responsible Research and Innovation

Pedro André Dias Falcão Ramos

D

2023



INSTITUTO DE **CIÊNCIAS BIOMÉDICAS ABEL SALAZAR**

Pedro André Dias Falcão Ramos

Facilitated Genome Editing as Responsible Research and Innovation

Tese de Candidatura ao grau de Doutor em
Ciências Biomédicas;
Programa Doutoral da Universidade do Porto (Instituto de Ciências Biomédicas de Abel Salazar)

Orientadora
Doutora Anna Olsson
Investigadora Principal
Instituto de Investigação e Inovação em Saúde (i3S)
Universidade do Porto

Co-orientador (es)

Doutora Maria Strecht Almeida
Professora Auxiliar
Instituto de Ciências Biomédicas Abel Salazar
Universidade do Porto

Doutor Cord Brakebusch
Professor Catedrático
Biomedical Research Institute of Copenhagen
University of Copenhagen

This thesis includes the following scientific contributions:

Accepted, in press (Manuscript 1)

Ramos PD, Almeida MS, Olsson IAS. Safe and purposeful genome editing under harmonized regulation for responsible use: views of research experts. *New Genetics and Society* (2023)

The first author contributed to the study conception and design, data collection, analysis and interpretation of results. Informed consent given to participants was also prepared by the first author. Lastly, the first author also contributed to manuscript preparation, writing, proofreading and revision following submission.

To be submitted (Manuscript 2)

Ramos PD, Almeida MS, Olsson IAS. What do people think about genetic engineering: A systematic review of questionnaire surveys before and after the introduction of CRISPR.

(To be submitted to Frontiers in Genome Editing – July 2023)

The first author contributed to the study conception and design, data collection, analysis and interpretation of results. The first author also contributed to manuscript preparation, writing and proofreading.

Published (Book chapter)

Ramos, P.D., Marques, F., Lopes, M.M., Santos, J.B., Tavares, F., Almeida, M. S. & Olsson, I.A.S. 2022. Biolab – uma proposta para o desenvolvimento de projectos em ambiente multidisciplinar. In Cadernos de Inovação pedagógica nº2, Unidades curriculares INOVPED da Universidade do Porto, Transversalidade e Criatividade, UPorto Press, Universidade do Porto, 1ª edição, pp: 125-139

The first author contributed to the project conception and design, data collection, analysis and interpretation of results. The first author also contributed to book chapter writing and proofreading before submission.

The papers listed above, integrating this thesis were or will not be included in other thesis or similar work.

Agradecimentos

Após 5 anos desde o início do doutoramento, é chegado o momento da entrega da tese e com ele de fazer um caminho inverso para expressar a minha gratidão a todos os que me acompanharam ao longo deste processo.

Em primeiro lugar, à Doutora Anna Olsson, minha orientadora, que desde o dia zero foi alguém que contribuiu muito para o meu crescimento pessoal e profissional. Desde tudo o que me ensinou, a forma como me motivou e os constantes desafios que me lançou, fizeram de mim alguém muito mais conhecedor, crítico e preparado na vida profissional académica e científica. Agradeço ainda a oportunidade de fazer parte de um projecto tão entusiasmante e sempre com grande liberdade para poder ser activo em diferentes frentes e iniciativas ao longo destes anos, abrindo-me sempre caminho para expandir a minha carreira científica. Agradeço ainda todas as discussões frutíferas, conversas e confiança depositada que permitiram que o meu empenho e desempenho pudessem ser sempre melhores do que piores, mesmo em tempos mais complicados quer a nível profissional ou pessoal.

Em seguida, agradecer à Professora Maria Strecht, minha co-orientadora, pela disponibilidade e motivação nas mais diversas fases dos projectos em que estivemos envolvidos. Agradecer a possibilidade de poder contribuir significativamente para o mundo académico e educacional, o qual muito me deu prazer em ser parte activa. Agradecer ainda todas as discussões de carácter científico e educacional que permitiram reforçar o meu percurso na vida académica.

I would like to also thank Professor Cord Brakebusch, my co-supervisor for the opportunity to be recruited for IMGENE as a PhD fellow and for all the guidance and challenging mindset. I would like to thank Professor Cord for making me confident that there is a space for being a transdisciplinary researcher and that such unique path despite difficult may become very fruitful along the way.

Agradeço também a todos os elementos do LAS 211 que em muito contribuíram para o meu percurso durante estes anos. Em especial ao Jorge, Sara, Gabi, Andreia, Catarina, Nuno e Sara Jorge, que quer fosse através das conversas e reflexões conjuntas, em horas de café, reuniões do LAS (online e em pessoa) e demais momentos, permitiram perceber que o caminho conseguir-se-á sempre fazer com o equilíbrio certo de acidez e jovialidade necessárias.

To the IMGENE fellows for all the scientific discussions and all the good environment that made me enjoy this journey better than ever. I would also like to thank to the Horizon 2020 research and innovation program for the funding of the project and the opportunity to be part of the European culture under the Marie Skłodowska-Curie grant agreement.

À Rita, a minha mulher, que ainda na designação de namorada iniciou esta caminhada em conjunto comigo na longínqua Dinamarca. Agradecer todo o amor incondicional, o incentivo constante, motivação e demonstração de confiança em mim e de que tudo correrá bem se estivermos juntos no percurso. À Maria Luísa, a minha filha, que é também um fruto desta caminhada e que veio dar a motivação extra para chegar até aqui demonstrando que as últimas páginas podem ser também as mais felizes. Aos meus pais, quero agradecer-lhes por me terem incutidos valores e dado todas as condições para me tornar realizado na minha vida profissional. Por todo o amor incondicional que sempre me deram e que se reflectiu na liberdade em poder ser o que quisesse e trilhar o meu percurso à minha maneira. Aos meus sogros e cunhada pelas contínuas palavras de motivação e admiração. A todos, obrigado por me fazerem acreditar que seria possível chegar até aqui!

Agradecer por fim, a todos os meus amigos e familiares mais próximos que de uma maneira ou de outra foram importantes nas diferentes fases do percurso e me fizeram chegar ao fim com a ideia de dever cumprido.

Imgene (@Imgene.bric)

@ImgeneBric

https://imgene.ku.dk

"This project has received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No 765269."

"Nothing in life is to be feared, it is only to be understood. Now is the time to understand more, so that we may fear less."

Marie Skłodowska-Curie

Abstract

In summary, the objective of the research from this PhD project was to investigate what experts in biomedical and animal science think about genome editing (GE) in an in-depth way and to investigate the attitude of citizens to this technology. RRI was chosen to address GE as an emergent technology by collecting the anticipative reflections – through the inclusion of sociotechnical imaginaries as conceived within the Science, Technology and Society (STS) field. Points-of-view of researchers who work with the technology daily and opinions of citizens who will be the recipients of such technology were studied. This will help in heading towards a broader stakeholder engagement to address GE, a common request from the scientific community.

To investigate the views of experts, I used in-depth semi-structured interviews with biomedical and animal science researchers. The interviewed researchers are supportive of applications that intend to correct genetic defective genes and agree that the technology is not ready for all types of interventions in humans based on safety of the technology. Well-defined organs and tissues that already have proven to be safer are the ones to be used towards the development of new therapeutic solutions. As long as safety concerns have not been solved, no somatic interventions should be brought to the clinic. Researchers underline the importance of harmonizing regulation of GE worldwide. They are supportive of engaging with the public, to provide information and engage in a dialogue about emergent technologies like GE.

A systematic review of the literature reporting surveys provided an overview of the opinion of citizens worldwide about genome editing and how it evolved over 35 years. The subjects covered in the research studies explored disease, prevention of disease and enhancement of characteristics in humans as well as medicines, derived foods and welfare issues in animals. Citizens' views are most of the times in agreement with the ones from researchers for human applications but differ for animal applications. Citizens are more inclined to genome editing of individuals that have serious diseases and they also agree more with it in germline cells. On the other hand, they disagree with genome editing in scenarios of human enhancement. They also distinguish clearly that medical products derived from animals are more welcome than food derived from them and concerns with animal welfare increased over the last 35 years.

The studies on expert views and on the public perception seem to highlight the importance of reflecting on the meaning of responsible GE and are discussed in this perspective. And this expands to the training context. I discuss a toolkit to embed RRI

as a way to prepare researchers in addressing emergent technologies like GE. The toolkit – intended to be used for research projects – is also connected to the pedagogical practice described in the third publication included in the present thesis – a course unit enacting RRI – and both fuel the discussion on the importance of training towards responsible science.

This thesis contributes to the existent knowledge about GE and emergent technologies since it helps in understanding what experts consider as priorities for a responsible GE and which citizens are more open to the technology. The thesis provides also a reflection on responsible use of GE that may help decision-makers and a discussion on ways the prepare future researchers to be responsible when addressing emergent technologies like GE.

Keywords: RRI, genome editing, STS, semi-structured interviews, systematic review, researchers, citizens

Resumo

Em suma, o objectivo deste projecto de doutoramento foi investigar o que peritos em ciência biomédica e ciência animal pensam acerca de edição de genoma (ED) de uma forma detalhada bem como quais as atitudes de cidadãos face a esta tecnologia. O conceito de Investigação e Inovação Responsável (IIR) foi escolhido como abordagem em relação à edição de genoma devido a ser uma tecnologia emergente. A investigação consistiu na recolha das denominadas reflexões de carácter antecipatório incluindo imaginários sociotécnicos que estão compreendidos no âmbito da Ciência, Tecnologia e Sociedade (CTS). Os pontos de vista de investigadores que trabalham com esta tecnologia diariamente e as opiniões de cidadãos que serão os recipientes da mesma foram assim estudados. Desta forma poderemos ir ao encontro da comunidade científica que por várias vezes pediu um envolvimento de *stakeholders* de uma forma mais abrangente para abordar a tecnologia de ED.

Para investigar as percepções dos peritos na tecnologia de ED utilizei entrevistas semi-estruturadas com investigadores de ciência biomédica e ciência animal. Os investigadores entrevistados apoiam aplicações da tecnologia de ED que sejam destinadas à correcção de genes defeituosos e por isso sinalizam que a tecnologia não está preparada para todo o tipo de intervenções no genoma humano devido às questões de segurança que levanta. Apenas órgãos e tecidos que estejam bem definidos provaram até agora serem seguros o suficiente para serem usados com o intuito de desenvolver novas soluções terapêuticas recorrendo a esta tecnologia. Enquanto tais questões de segurança associadas à tecnologia não tenham sido resolvidas, nenhum tipo de intervenção em células somáticas deve ser feito num cenário de clínica. Os investigadores enfatizam ainda a importância de harmonizar a regulação da ED mundialmente. São também apoiantes de um maior envolvimento com o público de forma a fornecer informações acerca da tecnologia e de maneira a criar diálogo acerca de tecnologias emergentes como a ED.

Uma revisão sistemática da literatura correspondente a 35 anos sobre questionários feitos a cidadãos relativamente à ED permitiu ter uma visão geral sobre as suas opiniões bem como as mesmas evoluíram ao longo do tempo. Os tópicos explorados durante o estudo de investigação recaíram sobre doença, prevenção de doença e melhoramento de características em humanos assim como tópicos relativos a produtos medicinais e comida derivados de animais editados geneticamente bem como questões acerca do seu bem-estar. Os pontos de vista dos cidadãos estão muitas das vezes em concordância com as dos investigadores no que diz respeito a aplicações de ED destinados a seres humanos, mas diferem nas que são destinadas a animais. Os

cidadãos estão também mais inclinados para a ED de indivíduos que possuem doenças graves e concordam mais com situações de ED da linha germinal do que os investigadores. Por outro lado, discordam com ED em cenários de melhoramento humano. Os cidadãos também distinguem claramente produtos derivados de animais geneticamente editados considerando os produtos medicinais de uma forma mais positiva do que comida derivada destes animais. As suas preocupações com bem-estar animal também aumentaram durante os últimos 35 anos.

Os estudos dos pontos de vista dos peritos da tecnologia de ED e da percepção do público parecem evidenciar a importância de reflectir sobre o significado de ED responsável e esta perspectiva é discutida ao longo da tese. Isto também passa pelo contexto de treino já que é abordado um *toolkit* que procura integrar a IIIR como forma de preparar futuros investigadores na abordagem de tecnologias emergentes como a ED. Este *toolkit* que se pretende que seja utilizado em projectos de investigação articula-se também com uma prática pedagógica (descrita na terceira publicação desta tese) que corresponde a uma unidade curricular que também utiliza IIR. Ambas visam impulsionar a discussão sobre a importância do treino de futuros investigadores em ciência responsável.

Esta tese contribui para o conhecimento existente sobre a ED e tecnologias emergentes pois ajuda a perceber o que peritos na área consideram como prioridades para implementar ED responsável e também elucida sobre que cidadãos estão mais predispostos a adotar esta tecnologia. A tese reflecte também sobre o uso responsável da ED, o que poderá ajudar agentes de tomada de decisão, e reflecte ainda formas de preparar futuros investigadores para serem responsáveis quando abordam tecnologias emergentes como a ED.

Palavras-chave: IIR, edição de genoma, Ciência, Tecnologia e Sociedade, entrevistas semi-estruturadas, revisão sistemática, investigadores, cidadãos

Contents

Abbreviations

BioLab – Biolaboratory – Multidisciplinary Experimentation Project

BRCA1 – breast cancer associated gene 1

BRCA2 – breast cancer associated gene 2

CasX – CRISPR associated protein X

Cas9 – CRISPR associated protein 9

CRISPR – clustered regularly interspaced short palindromic repeats

crRNA - CRISPR RNA

CTA – Constructive Technology Assessment

DNA – deoxyribonucleic acid

DSBs – double strand breaks

EC – European Commission

ELSA – Ethical, Legal, Society Aspects

ELSI – Ethical, Legal and Social Issues

FoTRRIS – Fostering Responsible Research and Innovation

GE – genome editing

GGE – germline genome editing

GM – genetic modification

HDR – homology-directed repair

HGP – Human Genome Project

NASEM - National Academies of Science and Medicine

NHEJ - non-homologous end-joining

PAM - protospacer adjacent motif

PNS - Post-Normal Science

RAC - recombinant DNA advisory committee

rDNA – recombinant DNA

RNA – ribonucleic acid

RRI – Responsible Research and Innovation

SCOT – Social Construction of Science

SwafS - Science with and for Society

TALENs – transcription activation-like endonucleases

tracRNA - transactivating RNA

List of figures

General introduction, objectives of the PhD project and thesis layout

The thesis is the result of an evolution process that was initially conceived to explore the socio-ethical implications of genome editing (GE) technology once it would be implemented in the daily lives of citizens. The project was ideated to explore the view of different stakeholders under a recently established European framework – Responsible Research and Innovation (RRI) – while simultaneously exploring this concept. Naturally, due to the range of possible definitions of responsibility, the project unfolded into a sociological approach using qualitative methodology to investigate the views of stakeholders of an emergent technology from the XXI century. The approach ended up expanded to the training context through my participation in parallel initiatives like the NewHoRRIzon project and the development of Biolaboratory – Multidisciplinary Experimentation Project (BioLab). The development of a toolkit regarding RRI and emergent technologies for the use of researchers is related to those experiences.

The PhD project is part of a European project named IMGENE which stands for Improving Genome Editing Efficiency and it was funded by the Horizon2020 programme in the European Union. The project is a result of the aim to investigate an emergent technology from the second decade of the current XXI century – GE – in the context of RRI. By being integrated in a European network, the project is inserted under a work package that aims at understanding the societal and bioethical challenges that genome editing presents for the present and future of human life.

Due to the limited work that exists in capturing the views of stakeholders in a qualitative manner, the 3-year project named “Facilitated genome editing as responsible research and innovation” had two fundamental objectives:

- to investigate the predicted harm-risk/benefit of genome editing;

- to investigate the public acceptance of genome editing.

Thesis layout

Chapter 1 presents GE as an emergent technology that can be studied in the perspective of RRI and constructive technology assessment (CTA) frameworks. The first subchapter describes the evolution of genome modification from the XX century until the discovery of GE in the XXI century and its latest breaktrough – CRISPR-Cas9 – from a

technological and bioethical perspective. The second subchapter focuses on the interconnectedness of RRI framework and the field and science and technology studies (STS) as a mean to approach an emergent genetic technology and the third and fourth subchapters focus on co-creation strategies that involve stakeholders to better anticipate the implications of GE emergent technologies for present and future generations of citizens. One of these co-creation practices refers to the educational realm.

Chapter 2 provides a theoretical framework for the qualitative methodology used in the project. The first subchapter focuses in conceptualizing in-depth interviews while the second explores the usefulness of surveys in inquiring citizens and the contribution of systematic reviews as a comprehensive form to approach these studies overtime.

Chapter 3 presents the methods that have been used throughout the project, articulating what is reported in the manuscripts.

Chapter 4 develops the work conducted with biomedical and animal science researchers where semi-structured interviews were devised to capture their views about GE technology. This chapter offers a condensed overview of the arguments and reflections on how to responsibly implement the technology in humans. The work comes in the form of a manuscript under revision in *New Genetics and Society* journal.

Chapter 5 presents a comprehensive and detailed study where surveys administered to citizens have been the object of analysis through a systematic review. These surveys encompass a period of 35 years of inquiry about the support for genetic modification of humans and animals. The manuscript presented is in almost final form for submission.

Chapter 6 describes Biolaboratory – Multidisciplinary Experimentation Project, a course unit where students develop group projects with a biological background from ideation to concept and oftentimes prototypes. The initiative is included in the thesis as it provides space for further reflection on a toolkit developed within the thesis project on emergent technologies and RRI. The work is published as book chapter in “Cadernos de Inovação Pedagógica – Vol. II: Unidades Curriculares INOVPED da Universidade do Porto. Transversalidade e Criatividade” (Ramos et al 2022).

Chapter 7 comprises the general discussion of the PhD thesis and provides a deepened view of the outcomes from the studies in the manuscripts, the connection between the data gathered from both studies in terms of meaning and the two outputs from the project. This chapter also reflects about the natural evolution of the research process throughout the course of the project.

Chapter 8 is the last chapter of the thesis and provides its main conclusions and perspectives for the work to be done in the future.

Appendices

I. Conceptual matrix (manuscript 1)

II. Interview guide (manuscript 1)

III. Codebook. (manuscript 1)

IV. AUDIT (Audio-dictate transcribing software) method (manuscript 1)

V. Informed consent template (manuscript 1)

VI. Figures (S1 and S2) illustrating the results of ranking cases done by interviewees from manuscript 1 (manuscript 1)

VII. Table S1 summarizing the support of citizens for genome modification in pre-CRISPR and CRISPR periods in different settings (manuscript 2)

VIII. Deliverable published in the IMGENE named “D19. Guideline for ethically acceptable genome editing” (Olsson and Ramos 2021)

Chapter 1: Genome editing in the context of responsible research and innovation and constructive technology assessment

1.1 Genome editing and CRISPR-Cas9 biotechnology

The 2020 Nobel Prize in Chemistry came perhaps without surprise to the renowned inventors Jennifer Doudna and Emmanuelle Charpentier for their work regarding “the genetic scissors: a tool to rewrite the code of life” whereby “using these, researchers can change the DNA of animals, plants and microorganisms with extremely high precision. This technology has had a revolutionary impact on the life sciences, is contributing to new cancer therapies and may make the dream of curing inherited diseases come true” (Royal Swedish Academy 2020). The “genetic scissors” are part of the genome editing technology that has been dubbed one of the breakthroughs in molecular biology (Regalado 2014) already transforming medicine and biotechnology fields (Adli 2018) and considered one of the greatest inventions that may revolutionize health in the 21st century (Taylor 2021).

1.1.1. Genome editing breakthrough history: adaptation of CRISPR-Cas9 system from prokaryotic to eukaryotic cells

To reach this breakthrough stage, a long-term and sinuous research path took place since the 1980s after gene targeting has been successfully achieved in mice embryonic stem (ES) mammalian cells (Thomas and Capecchi 1987, Urnov 2018). Parallel to this achievement, a set of tandem palindromic repeats were identified for the first time in *Haloferax mediterranei* bacterial species, where these sequences were seen to be coherently dispersed throughout their genome, although no function had been understood at the time (Lander 2016). These were the first steps to identify what later became named as clustered regularly interspaced short palindromic repeats or as we prefer nowadays, CRISPR (Mojica and Montoliu 2016). While this brought no enthusiasm among the scientific community at the time, several research groups working with diverse bacterial species harboring such sequences, postulated and confirmed for the first time that these were part of a mechanism of defense of bacteria enacted to avoid virus attacks, a type of acquired immunity response (Lander 2016, Barrangou and Horvath 2017). Such confirmation essentially brought a sound of revival of a field that has been silently moving since before the 90s decade mainly due to the identification of the mechanism used by *Streptococcus termophilus* bacteria when defending themselves from bacteriophages by relying in both adaptation through the acquisition of spacers that resemble viral DNA and through interference by arming themselves of bacterial proteins capable of cleaving DNA (Barrangou and Horvath 2017). In fact, CRISPR-associated protein 9 or Cas9 has been the first to be identified as the protein harboring a domain with cleaving activity able to induce double strand breaks (DSBs) (Bolotin, 2005, Lander

2016, Barrangou and Horvath 2017). Upon discovery that the CRISPR-Cas9 system could be transferred to heterologous bacterial strains such as *Escherichia coli, Staphylococcus aureus and Staphylococcus epidermis* (Sontheimer and Marrafini 2008, Sapranauskas et al 2011, Lander 2016), two additional breakthroughs made the hallmarks of CRISPR history. Firstly, the identification of a protospacer adjacent motif (PAM) that is conserved among CRISPR and which enables the blunt DNA cleavage driven by Cas9 by being in the close vicinities of the cutting site (Barrangou and Horvath 2017). Secondly, the finding of a guide RNA (gRNA) as the result of the hybridization of a CRISPR RNA (crRNA) and a transactivating RNA (tracrRNA) which is then able to pair with Cas effector nucleases such as Cas9 and direct its cleavage activity to the target site DNA upon recognition of PAM sequence (always constituted by a NGG sequence) lying 3-5 nt beyond the target site (Jinek 2012, Lander 2016, Sander and Joung 2014). These discoveries together with the efficient reprogramming of CRISPR-Cas9 system in both *Streptococcus termophilus* (Gasiunas al 2012) but mainly in *Streptococcus pyogenes* bacterial platforms in order to induce specific double-strand breaks led for the first time to the thinking that an adaptation from prokaryotic to a eukaryotic system to target DNA activity could soon become a reality (Barrangou and Horvath 2017, Sander and Joung 2014).

1.1.2. CRISPR-Cas9 breakthrough following adaptation to mammalian cells: potential applications and human therapy

An important landmark that glues the discoveries attained along CRISPR-Cas9 history with its adaptation to mammalian cells has to do with the discovery of DSBs repairing mechanism. The discovery that DSBs are editable in mammalian cells either through non-homologous end-joining (NHEJ) and homology-directed repair (HDR) mechanisms (Rouet, Smith and Jasin 1994, Urnov 2018) allied with the fact that zinc-finger (ZFNs) and transcription activating-like effector nucleases (TALENs) were able to foment them (Lander 2016, Urnov 2018) allowed scientists to truly hypothesize about this idea. These nucleases rely on the ability to be very specific in the recognition of DNA sites to be targeted and they enable the tipping of the editing balance in favor of HDR when generating DSBs following the cleaving event (Sander and Joung 2014, Lander 2016, Urnov 2018). Eventually, several groups demonstrated the ability of these nucleases to target the mammalian and most strikingly, human cells (Urnov 2005, Lander 2016, Urnov 2018). The main obstacles for wider uptake of these approaches were the laborious, slow and costly processes to build the nucleases (Camporesi and Cavaliere 2016, Lander 2016).

One of the first breakthroughs in the field came by solving these challenges through the adaptation of the CRISPR-Cas technology from bacterial to mammalian cells in 2013 in Feng Zhang laboratory (Lander 2016). This technology breakthrough contributed to the rapid uptake of the technology by laboratories worldwide after the first publications in the genome editing field. As this allowed the technology to become cheaper, it was soon developing at a fast pace with reports of multiple discoveries in a high number of organisms (Birling et al 2017, Moreno-Mateos et al 2015, Sekine et al 2018, Yan et al 2018). The first reports of application of the technology in human cells of different tissues (Lyu et al 2018, Xie et al 2017, Zhang et al 2017) and animals (Bjursell et al 2018, Nelson et al 2015, Singh et al 2018) with potential clinical applications and for the editing of livestock (Carlson et al 2013) started to be published. Even research conducted with germline cells from animals (Lillico et al 2013, Long et al 2014, Niu et al 2014, Young et al 2017) and humans (Kang et al 2016, Liang et al 2015, Ma et al 2017) have been conducted and published in academic journals.

1.1.3. Before CRISPR: bioethics of genetic modification in the 20th century

It is tempting to believe that it all started with genome editing, but in fact the history of debate about genetic modification can be traced back to the 1970s following the advent of recombinant DNA (rDNA) and its application as the first genetic engineering tool in mammalian cells (Thomas and Capecchi 1987, Urnov 2018). This event led to the first international conference covering rDNA – the Asilomar Conference in 1975 – and to the subsequent creation of the recombinant DNA advisory committee (RAC) with the intention to discuss major technical, societal and bioethical implications that such revolutionary technology would bring (Hurlbut, Saha and Jasanoff 2015, Rufo and Ficorilli 2019). This was the first time scientists were confronted with having to consider the potential of genetic engineering to be extrapolated to eukaryotic organisms and how this new tool might make it possible for the first time to change the DNA of multicellular and complex organisms such as vertebrate animals and especially humans (Hurlbut, Saha and Jasanoff 2015). The series of guidelines which emerged from the conference (Berg et al 1975) show the different dimensions of the set of challenges arising from the rDNA technology: the biological risks for human health and the strategies to avoid them, the necessary education that scientists and staff needed to have regarding safety of the materials being manipulated and the responsibility that researchers and private companies needed to assume in face of certain experiments proven to be dangerous enough and be excluded or abandoned due to safety risks (Berg et al 1975, Rufo and Ficorilli 2019).

This was perhaps the first time a non-medical issue gave rise to such a wide bioethical discussion. The first steps towards building ethical guidelines for the international research community focused on research with human subjects, and started with the drafting of the Nuremberg code in 1947, followed by the declaration of Geneva in 1948 contributing to the revision of the Hippocratic Oath and reformulations of physicians' ethical duties regarding their patients (Declaration of Geneva 1948, Nuremberg Code 1996), and the Declaration of Helsinki in 1964 that established careful assessment of benefits and risk for participants in research as a priority (Declaration of Helsinki 1964). At the same time as the Declaration of Helsinki was first revised in 1975 with the introduction of Institutional Review Bodies (IRBs) as mechanisms of oversight of research with human subjects (Declaration of Helsinki 1975), the US government established the National Commission for the Protection of Human Subjects of Biomedical and Behavioral Research in 1974 (Franklin 2019, Jasanoff 2003). This preceded the first draft regarding ethical principles and guidelines for basic research with human subjects in the Belmont Report in 1978 based in respect, beneficence and justice principles as well as making informed consent mandatory when involving participants in studies (United States 1978). It was against this background of establishment of ethical principles that the discussion of genetic modification of humans in the Asilomar Conference in 1975 took place, involving multiple stakeholders where not just scientists were invited (Berg et al 2015). The outcome was a precautionary resolution based in a moratorium for the continuing of research with cells from non-human organisms (Hurlbut, Saha and Jasanoff 2015, Rufo and Ficorilli 2019).

No other topic has given rise to such a fierce bioethical discussion as the Human Genome Project (HGP) in 1990 (Atkinson, Glasner and Lock 2009, Bucchi 2004, Franklin 2019). HGP that envisaged the sequencing of the whole human genome led to the creation of the largest bioethics program ever to be funded – the Ethical, Legal and Social Issues (ELSI) program, which used around 5% of the HGP funds to establish a research program and a working group (Franklin 2019, Jasanoff 2003). The ELSI research program and the working group met different challenges and recognition along the way and also underwent some transformation as regards its composition, with professionals familiar with genetics and ethics discussions being replaced by analysts of genetics and society, and finally the program was dissolved and its activities distributed through other decision-bodies (Jasanoff 2003). In the end, the involvement of different professionals from ethics, philosophy and social science contributed to the debate about the sequencing of human genome and its potential impacts on society regarding genetic privacy, biobanks and behavioral genetics. This discussion already included topics such

as modification of human DNA in germline settings and the appearance of designer babies, which we have seen reemerging in the more recent discussion (Bucchi 2004, Franklin 2019). One of the most important changes in science policy resulting from the ELSI research program is arguably the fact that it produced a shift from self-regulatory science to an anticipative one by trying to predict the impacts of research and this was meant to improve public trust in science (Jasanoff, Hurlbut and Saha 2015). The idea that oversight of science needed to pass from self-regulation to one that intends to anticipate problems and establish binding regulatory codes gained momentum since the Asilomar event and is evident in subsequent programs, guidelines and agreements such as the ELSI program throughout HGP, the Oviedo Convention in 1997 (Council of Europe 1997) and the Universal Declaration on the Human Genome and Human Rights in the same year (UNESCO 1997), later ratified to Universal Declaration on Bioethics and Human Rights in 2005 (Langlois 2008).

Following the long debate that took place after the Asilomar conference and a period of embargo to the development of the technology, the moratoria was lifted and the research proceeded smoothly While the HGP is widely considered a success in its all dimensions, from the achievement of its objective by 2003 (Bucchi 2004) and to the debate that started with questions concerning genomic data privacy and the use of biobanks for the deposit of human genome data (Atkinson, Glasner and Lock 2009), gene therapy suffered a backlash following the death of Jesse Gelsinger in 1998 attributed to the use of genetically-altered virus carrying a copy of gene to correct for a metabolic disorder (Caplan 2019) and which ignited a wave of distrust and debate about the use of genetically-altered virus into human patients. This left unanswered many societal and bioethical questions in the genetic modification field by the end of the 20th century remaining this way until the second decade of the 21st century

1.1.4. Ethical, regulatory and societal questions to address CRISPR-Cas9 technologies

Fast-forward to the year of 2015 when the discussion of genetic modification was re-ignited in the context of genome editing technology. with two major events bringing back old questions: the revision of the UK law which gave green-light to the first three-parental embryo following mitochondrial DNA donation (Camporesi and Cavaliere 2016, Rosemann et al 2019) and the first genome-edited embryos in the laboratory (Liang et al 2015). These events were followed by a discussion in media and the research community drawing attention to the likely and imminent creation of the first-genome edited human beings (Baltimore et al 2015). Of a lesser but still remarkable magnitude

were the first clinical trials in China with lung cancer human patients that were subjected to ex-vivo GE of PD-1 receptors (albeit narrowly documented by the media) (Clinical trials gov 2016, Brown 2018) and the first genome-editing of multiple organisms from mice (Nelson et al 2015) to primates (Niu et al 2016) and mosquitoes (Caplan et al 2015, Oye et al 2014, Scudellari 2019) that motivated the notion of the dispersion of GE in research environments worldwide.

Against the background of these events, a first International Summit on Human Genome Editing was held by the National Academies of Science and Medicine (NASEM) in the US in December 2015 to discuss the most pressing issues identified by the research community in the context of genome editing, with focus on germline gene editing. This led to the release of first criteria recommendations if to proceed with it (On Human Gene Editing 2015) and simultaneously a moratoria request to stop its use (Baltimore et al 2015) and recall on article 13 and 28 of Oviedo Convention (Council of Europe 1997) by the European committee on bioethics (DH-BIO 2015). In parallel with the concerned attitude towards the use of genome editing in germline cells, a series of impactful announcements took the world of genome editing by storm, including the first clinical trials to treat patients with genetic diseases like beta-thalassemia, cancer patients with sarcoma and eye congenital disorders (Reardon 2016, Clinical trials gov 2016, Clinical trials gov 2018, Clinical trials gov 2019) and the use of GE to eliminate PERV-viruses from pig hearts to facilitate xenotransplantation into human beings (Reardon 2015). It was not until the 26th of November of 2018, when He Jiankui released a YouTube video "About Lulu and Nana" (Greely 2019) claiming the birth of the first genome-edited babies in China at the Shenzen HarmoniCare hospital (Cyranowski 2019, Greely 2019) and which immediately before had been exclusively reported in the media (Regalado, 2018) that the second wave of concerned reaction of the scientific and also non-scientific community took place (Baltimore et al 2015, Bosley 2015, NASEM 2017). Up to this point, many authors had already explored the ethics and responsibility of using an innovative technology for the purpose of editing human cells and these were intimately linked to safety of somatic genome editing applied to patients in the clinic (Ishii and Araki 2016, Cathomen et al 2018) safety and "safe enough" criteria (Baylis and MacLeod 2017), the enhancement of human characteristics in both somatic and germline genome editing settings (Brokowski and Adli 2018), definition of therapy and prevention as criteria to apply genome editing in germline cells (Juengst et al 2018), the need for a moratorium in the application of genome editing in germ cells that may lead to genome-edited newborns and a new class of individuals (Nordberg et al 2018), the impact of diverse regulatory and legal frameworks worldwide contributing to inequality in the access to

genome editing technology (de Lecuona et al 2017, Mulvihil et al 2017) and the overall impact in equity and human dignity when accessing such genetic interventions (Howard et al 2018). In animals, authors mainly explored issues such as the harm-risk/benefit balance of creating newer animal models of disease in the laboratory through genome editing (Ohl and Meijboom 2015), the balance between suffering of animals and knowledge gained from the use of these models particularly in the case of non-human primates (Neuhaus et al 2018) and the impact on welfare of genome-edited farm animals depending on the traits modified (Eriksson et al 2018, Ishii 2018). At the intersection of these two fields, there is also space for some technical and ethical reflections about the implantation of genome-edited organs originally from animals into humans such as the naturalness (Luna, 2017) and the utilitarianism of using animals for such type of research as driven by scientists (Jasanoff 2018). Out in the wild, applications with possible impacts in the environment related with biosafety and efficacy concerns of gene drive released mosquitoes with potential for widespread were explored (Caplan et al 2015, Lunshof 2015). Adding to this, the resilience and productivity of GE crops together with public trust and regulatory barriers to its commercialization based in risk approach (Casacuberta and Puidomènech 2018, Conko et al 2016) were explored from an ethical and sociopolitical perspective. This counted with previous works from authors in the field of genetic modification and genetic engineering of species assessing public perceptions in face of these applications (Horst 2007, Knight 2009).

Following the unexpected and bizarre announcement of the first genome-edited unborn babies, a second moratorium request was disseminated (Lander 2019). The next years were marked by works that highlighted once again the social and ethical concerns raised by the adoption of genome editing, but also focusing on the broad societal engagement of stakeholders in the decision-making process (Braun and Meacham 2019, Halpern et al 2019, Meagher et al 2020, EGE 2021). Moreover, the first recommendations from World Health Organization (WHO) towards the governance and oversight on the use of human GE (somatic and germline) for clinical, research and enhancement purposes were released, directed particularly towards the governance and oversight on the use of human GE in somatic and germline scenarios in both clinic, research and enhancement settings. These were building on previous reports but also provided insights about the need for international collaboration as a means to effective policy-making, the creation of human genome editing (HGE) registries and a Science Council to track unsafe, unethical and illegal GE and finally, the focus on education, engagement and empowerment of citizens for informed decision-making about genome editing (WHO 2021). (Bio)ethical and social issues in non-human GE applications were also explored

in light of human responsibility towards animals (MacLeod and Hartley 2018) and deeply related with animal welfare, dignity, harm-risk/benefit and sentience (de Graeff et al 2019). Authors in the field of Science and Technology Studies (STS) have also elaborated about the broad societal engagement of stakeholders in order to have a successful and global discussion about genome editing (Jasanoff and Hurlbut 2018, Wirz, Scheufele and Brossard 2020). Additionally, reflection over the regulatory landscape of genome editing worldwide and guidelines to follow international governance of genome editing were discussed (Baylis et al 2020, Boggio et al 2018).

In this thesis I will discuss how these concerns alongside the technical challenges and powerful applications of genome editing are seen by a multitude of actors. This will contribute to the envisioning of the predicted harm-risk/benefit of its use and ultimately help in establishing a harmonized integral vision of what genome editing biotechnology can be in the present and should be in the near and furthest future.

The next two chapters will explore the role of RRI in contextualizing genome editing technology and the framework of STS for the implementation of the work followed throughout this project.

1.2. Responsible Research and Innovation (RRI) embedded into Science, Technology and Society (STS) studies as an approach to emergent technologies

RRI came out as a concept at European Comission (EC) Directorate-research headquarters with its origins tracing back to Ethical, Legal, Society Aspects (ELSA) or Issues (ELSI) programs established at the time of Human Genome Project (HGP) start (Owen, MacNaghten and Stilgoe 2012, Shelley-Egan et al 2020). While confusion over if this would represent a mere concept or a principle of action, a holistic definition of RRI has been discussed during several actions promoted by the EC with the aim to push forward the applicability of the concept and its effective translation according to the needs established by the policy group (de Saille 2015, Owen, Macnaghten and Stilgoe 2012). In this sense, policy makers René von Schomberg and European Commissioner Geogeghan-Quinn in 2011 and 2012, respectively contributed to the first concrete definitions and principles by arguing for incorporating RRI both at the level of EC framework and under the funding policy of applications derived from it. Von Schomberg signalized RRI as a

> […] transparent, interactive process by which societal actors and innovators become mutually responsive to each other with a view on the (ethical)

> acceptability, sustainability and societal desirability of the innovation process and its marketable products (in order to allow a proper embedding of scientific and technological advances in our society, (von Schomberg 2011 page 50)

Likewise, Geogeghan-Quinn suggested that

> […] Researchers, policy makers, business people, innovators and most of all, the general public, have difficult choices to make as regards how science and technology can help tackle our different societal challenges" where "(…) we can only find the right answers by involving as many stakeholders as possible in the research and innovation process. Research and innovation must respond to the needs and ambitions of society, reflect its values and be responsible" while underlining their "(…) duty as policy makers (is) to shape a governance framework that encourages responsible research and innovation. (Owen, MacNaghten and Stilgoe 2012 page 753)

The idea of embedding RRI in European framework programs such as Framework Programme 7 (FP7) and Horizon2020 (H2020) did not immediately convince scholars within the Science and Society programme because they initially aimed for RRI to be a normative and not an ambiguous concept (Owen, Macnaghten and Stilgoe 2012, von Schomberg 2013). Another challenge was that targets of the EU treaty that involve the promotion of scientific and technology advances, generation solidarity and social justice need to be aligned with the impacts of research by weighing the harm-risk/benefit ratio of certain technologies in the quest to find how righteous they are and what implications may be translated into citizens' life as well as towards the "high level of protection and improvement of the quality of the environment, the raising of the standard of living and quality of life" (von Schomberg 2011 page 42). Likewise, a duality exists if citizens should be informed because of their contribution to publicly funded research or because of their real involvement at the decision-making process (de Saille 2015). It is here that RRI emerges as an instrument to organize the discussion for what do citizens want science and innovation to do, for what purposes specifically and what values will be added at public level (Owen, Macnaghten and Stilgoe 2012). The idea of co-creation of RRI (co-RRI) and the projection of research based on normative assumptions in order to take responsibility as a mean to solve "grand environmental and societal challenges" has been one of the proposal of groups such as the FoTRRIS (Fostering Responsible Research and Innovation) network with the project betting on reflexivity to address glocal challenges both at the level of ecosystems sustainability and social inclusivity with approaches that would necessarily bring ethical and political consequences after

decisions are made (Ruszanov and Haese 2015). This also led to the funding of European programs such as the Science with and for Society (SwafS) programmes and other educational strategies where RRI is embedded aiming at tackling “Grand societal challenges” with economic and societal impacts (RRI 2020). Additionally, a co-creation approach bridging scientists with innovation facilities such as industries and technology developers would push responsible innovation to a new level, where research institutions would be providers but also managers of innovation with a commitment to open it to the public (Lippens et al 2019). The strategy for H2020 was to “build effective cooperation between science and society, to recruit new talent for science and to pair scientific excellence with social awareness and responsibility” (RRI 2020). The latter recalls for the last but not the least important anchor point that is embedded in RRI culture: social desirability. Importantly, such an anchor point is directly linked not just to the above-mentioned improvement of life quality but also to inclusive deliberation allowing for the occurrence of two main events: the involvement of multiple stakeholders and the engagement of the public upstream of the decision-making process (de Saille 2015, Owen, Macnaghten and Stilgoe, 2012, von Schomberg 2013).

I will explore these two topics in a greater detail along the next two sub-chapters as part of the STS framework particularly when addressing emerging biotechnologies like genome editing biotechnology.

1.2.1. Science and Technology with Society: when inclusivity meets deliberation and the link between RRI and CTA

Democracy is a hallmark of society evolution since of ancient Greece civilizations meaning “power of the people” and embedded in the Universal Rights Chart as a fundamental principle (United Nations 1997). For that to be a reality, inclusivity needs to be respected and also ascertained as a human right (United Nations 2019). Inclusive deliberation is embedded in democratic principles and supposes a process that is also common to RRI principles: discussion and negotiation between actors to implement or adopt certain policies over a given process and/or products (Owen, Macnaghten and Stilgoe 2012). Around this negotiation process, issues over social, ethical and political dimensions are often evoked when the subject of discussion is the embracement of a given technology or scientific tool taken into consideration the range of applications where it may be implemented (Owen, Macnaghten and Stilgoe 2012, von Schomberg 2013). Here, a set of dimensions that are inherent to RRI come into play, namely: inclusivity, reflexivity, anticipation, and responsiveness (Owen, Macnaghten and Stilgoe 2012). Anticipation concerns a deep analysis of consequences of a certain technology

related simultaneously with its purpose and therefore intended impacts as well as its unintended consequences or so-called unforeseeable impacts which are generally of negative nature (Owen, Macnaghten and Stilgoe 2012, von Schomberg 2021). The analysis involves reflection based in technology assessment and foresight by assessing potential impact of implementation of certain products in society (Owen, Macnaghten and Stilgoe, 2012, von Schomberg 2013). Anticipatory governance is in fact an opportunity to reach greater technology robustness through enhanced reflection bringing out benefits and risks as well as ethics and regulation so that they can be clearly considered in decision-making (Owen, Macnaghten and Stilgoe 2012, Konrad et al 2016). A central character of STS relates with “collective expectations” in “mobilizing, guiding, and coordinating diverse sets of actors involved in techno scientific fields and it requires “expectations which are to some degree common, shared reference points” (Konrad et al 2016 page 469). This entails a process dimension where multiple stakeholders with a share of social responsiveness interact, communicate and in the end, take actions to decide the conditions where such a product or tool will be applied (von Schomberg 2013). Expectations are therefore a way to convey this process dimension in the sense that these are “statements about future conditions or developments that imply assumptions about how likely these are supposed to be and that travel in a community or public space” (Konrad et al 2016 page 470). Within the sociotechnical dynamics of innovative technologies, technology assessment and foresight strategies can be exercised recurring to sociology of expectations or sociotechnical imaginaries frameworks and involving both producers of technology and other actors linked with society, business and regulatory bodies (Rip 2018). In fact, the implementation of such reflexive tools may happen under the form of co-creation processes with scientific producers within institutional setups (Lippens et al 2019), in studies with bioethicists about innovative methodologies like pharmacogenetics (Hedgecoe 2010) and in workshops with diverse stakeholders about the governance of nanotechnologies (Rip 2018). For emergent technologies, expectations or the sociology of their interpretation are usually expressed as promises and opportunities or fears and concerns and may be converted in optimistic and beneficial outputs or pessimistic and risky outcomes, respectively (Konrad et al 2016). This is branded in STS through the sociotechnical imaginaries where expectations and anticipation may lead to a policy driven and sociopolitical action (ibidem).

1.2.2. RRI as a framework for assessment of emergent and innovative technologies with a policy-making goal

In the end of the 1990s, RRI surfaced as a successor of ELSA/ELSI programmes which aim to integrate science, society and technology assessment when addressing emergent technologies in the post-genomic era (Shelley-Egan et al 2020). Similarly, RRI has in its own core a clear demand for stakeholders' participation in a broad, engaging, responsible and anticipative manner (Delgado and Åm 2018). A clear demarcation of RRI from other frameworks is the integration of a social sciences approach with science and engineering through collaborations that can be characterized as science with and for society (Delgado and Åm 2018, Bruce and Bruce 2019). RRI as a public policy envisages knowledge production between several actors in order to reach more responsible techno-scientific futures without promising more than they may really offer and working in anticipative terms that have in mind unpredicted outcomes (Delgado and Åm 2018, Felt 2018). It became relevant to consider it as an important framework when addressing emergent technologies such as genome editing with its multi-level impact in society (Felt 2018). The notion of responsibility entered the domains of science and technology with due force through the amendment of the social contract between emergent scientific developments and society allowing scientists to talk about these in promising terms with hype and disappointment involved and this led to a reorganization of the division of moral labor premise with newer types of stakeholder interaction and arrangements (Rip 2018). Policy-makers for example have often to opt for hybrid suggestions due to a huge complexity that surround new or breakthrough technologies due to the difficulty in gathering all the knowledge available which advances at a fast speed hard to keep up and also due to the unavailability of knowledge on future impacts of its adoption in society at the time of deference (Sucha and Dewar 2020). As technology assessment, often evaluations of new technologies intend to observe both intended and non-intended consequences of its introduction related with environmental and sustainability (von Schomberg 2011). If we take examples such as climate change, we see that energetic transition needs to avoid accelerated global warming, prices of primary sources and its access by citizens, the building of ship and aviation fleets that could work with more sustainable fuels, the assessment of industry businesses that rely on already instated processes that ensure profit margins and their obligation to fulfill decarbonization represent already a part of a very complex intertwined relationship between factors that deserve attention by policy-makers when defining guidelines to be presented to countries' counterparts (Calenbuhr 2020).. RRI resonates with the scenario of constructive technology assessment (CTA) in evidence anticipation, reflexivity and

responsiveness and help in establishing the “better technology for the better society” through participation of institutional and societal actors either in an interactive or “division moral labor” way (Rip 2018).

Defining research questions in a policy context needs bidirectional communication between scientists and policy-makers with the will to provide the best translation possible of scientific evidence of innovative technologies for non-scientific audiences (Sinkiewicz 2020). The involvement of stakeholders such as scientists and policy-makers at the same table and their joint action in framing the problems, questioning the (questions and legitimating) policies is very relevant to help in drafting guidelines that may be essential for the application of certain scientific or technological tool for society (ibidem) When we are moving towards a technology that presents us complex challenges that usually do not have definitive answers, constant dialogue between scientists and policy-makers is absolutely essential, constituting the basis for co-creation and resembling a new face of Science for Policy – the 2.0 version (Sucha and Dewar 2020).While this reflection does not pretend to be about policy-makers or researchers specifically, one should emphasize the adoption of such an approach when tackling challenges posed by an emergent technology. Similar to the process of going from a policy problem to a research question, a comprehensive approach that seeks to ask non-neutral questions in order to define a problem and performing a diagnosis of the technology while providing possible and unlimited solutions should be the core of such reflexive exercise for scientific experts (Sinkiewicz 2020). Another category of actors who have an important but not always clearly defined role at this scientific-policy-making interface are bioethicists due to their relevance in framing questions about the implications for society that emergent technologies may present and how this may change the view of the world where we live (Hedgecoe 2010). Multidisciplinary co-creation consists in the active engagement of stakeholders including citizens in a process that emphasizes the reflection about desirable futures (Topp et al 2020). This may as well be achieved by a “division moral labor” where each actor assumes its own role and indicates other actors that may give their expectations or visions about a certain innovative technology (Rip 2018). What matters here is that either by adopting a co-creation or division moral labor strategy we become closer to de facto governance of science and technology (Konrad et al 2016). For this, foresight strategies through strategies such as horizon scanning and scenario building contribute to a speculative design that illustrates future applications of technologies and analyzes complex issues helping in creating anticipatory governance capacity (Störmer et al 2020). Ultimately, foresight tools are characteristic of sociological analysis, a methodological approach that policy-makers may also use to understand

impacts of emergent technologies allowing them to have a more distanced overview of the complexity of its systemic ramifications for society (Calenbuhr 2020).

1.2.3. Emergent technologies in policy-making design context: the case of genome editing

The notion of Post-Normal Science (PNS) is relevant for considering emergent technology features characterized mainly by its uncertainties:

> The contribution of PNS to our understanding of science in society can be summed up quite simply: In the social process of science, quality is achieved, not by a delusory pursuit of certainty, but by the skilled management of its uncertainties, involving all who have a concern for the issue. (Funtowicz and Ravetz 2020 page 18)

A definition for emergent technology has been proposed as:

> […] a radically novel and relatively fast growing technology characterized by a certain degree of coherence persisting over time and with the potential to exert a considerable impact on the socio-economic domain(s) which is observed in terms of the composition of actors, institutions and patterns of interactions among those, along with the associated knowledge production processes. Its most prominent impact, however, lies in the future and so in the emergence phase is still somewhat uncertain and ambiguous. (Rotolo et al 2015 page 1828)

Emergent technologies in a PNS environment resemble high decision stakes and high uncertainty in facts being also characterized as having values in dispute and decisions needing to happen urgently (Funtowicz and Ravetz 2020). Breakthrough technologies such as GE have therefore an effect in citizens' lives and the acceleration of its assimilation occurs in such a democratized way that it takes the decision-making usually held by high political members of the elite closer to members of society, public in general and other stakeholders to decide about it in a collective manner (Sucha and Dewar 2020).

GE can be included in the contemporaneous definition of emergent technology by featuring within the five categories identified. The radical novelty traces back to the beginning of last decade when in 2013 it has been first described in the literature as a straightforward and equally or more precise tool to edit the genome of cells (Jinek et al 2012). The fast-growing characteristic has been pushed by its rapid adoption in labs

derived from its low cost and observed by the change in research landscapes worldwide (Lander 2016, Camporesi and Cavaliere 2016). The degree of coherence over time has been given by multiple advances in facing the technicalities of genome editing and its implementation among research in multiple organisms (Liang et al 2015, Nelson et al 2015, Niu et al, 2016, Sekine et al 2018, Greely 2019) whereas potential to exert socio-economic impact is exacerbated by the "editability" of multiple species (humans included) and the differential access to therapies by citizens (Mulvihil et al 2017, Halpern et al 2019). Finally, the ambiguity and uncertain nature of these type of technologies have as the main object of CTA approach where foresight is a crucial premise when addressing stakeholders' views about it (Rip 2018, Störmer et al 2020) and which have been also done by others throughout history (Horst 2007, Knight 2009, Hedgecoe 2010).

1.3. Social construction of theory or of reality? Imagining scenarios as a co-creative step to address genetic emergent technologies

CTA requires a network of individuals, institutions and organizations in continuous communication with the goal to reach stabilization of the technology in terms of knowing what are the real consequences, challenges and opportunities that it may present for the society (Atkinson, Glasner and Lock 2009). CTA supposes an action that may be reflected in political action, citizen participation or drafting of guidelines to advise decision bodies (Jasanoff and Kim 2015). This contrasts with other frameworks from STS like social construction of technology (SCOT) where a more neutral view about potential consequences of science of technology is usually binded to (Wiesner 2018) While both frameworks are characterized by risk and benefits weighting as well as consideration of human behavior towards a given invention (Jasanoff and Kim 2015), I will focus mainly in the more proactive and straightforward mindset of CTA in the sense that it pertains to not be neutral and it intends to influence governance of emergent technologies (Konrad et al 2016, Rip 2018). There are several examples from the last 50 years of bringing experts and non-experts together at the same table and public participative initiatives like citizen groups and citizen panels that reflect over "unforeseen and unwanted effects of scientific and technological progress on the environment and human health" (Bucchi 2004 page 139, see also Hurlbut, Saha and Jasanoff 2015). Examples include what were emergent technologies in their own eras such as the discovery of recombinant DNA and the sequencing of the human genome, with the respective science-society projects represented by the Asilomar Conference in 1975 and the birth of ELSI/ELSA studies embedded in the Human Genome Project (HGP) in 1990 (Hurlbut, Saha and Jasanoff 2015, Rip 2018, Rufo and Ficorilli 2019). The Asilomar Conference is often considered

an example of looking at science as a mean to more than producing knowledge by emphasizing the need to regulate it and draft ideal guidelines that would express possible effects in society and using tools such as moratoria to prevent the use of emergent technologies under a “governable emergence” narrative (Hurlbut 2015, Rufo and Ficorilli 2019). The HGP on the other hand, constitutes an example of a successful approach of this type of strategy by involving both public institutions like the National Institutes of Health (NIH) and a private company (Celera Genomics) both in the US, but fundamentally due to the public free dissemination of what later became known as “the book of life” (Bucchi 2004). Moreover, scientific knowledge depends on opening-up and re-contextualization of science in society by the intervention of multitudes of visions and the modification of spaces like funding agencies and universities as well as by the robustness associated with the process of knowledge production challenged by cultural backgrounds and innovation (Rip 2018). At the time of HGP, a paradigm change took place by moving towards a “post-academic science” that conjugated several disciplinary fields from cybergenetics to information science and the inclusion of stakeholders like philosophers and social scientists to “study its implication for ethics and society” (Bucchi 2004). Informing and demonstrating to the public about certain technologies is a matter of accountability to which scientists are subjected, but this also defies the communalism pillar of modern science by stressing the complexity of results dissemination since it advocates both invention and intellectual property rights versus tax payer ownership rights (Jasanoff and Kim 2015, Bucchi 2004). Moreover, the European value of bilateral communication with the public came out as materialization of this dialogue as a mean to answer the societal challenges and the need to be impregnated at the core of EU policy framework leading to the Horizon 2020 legal framework that included participatory engagement of citizens and civil society (de Saille 2015). The advocacy for global citizen deliberation on GEs a matter of policy claiming for inclusive participation and the development of a global citizen assemblies and initiatives is defined with the purpose to increase trust in experts and decision-makers as well as a mechanism for collective anticipation of public responses to an emergent technology (Jasanoff and Hulrbut 2018, Dryzek et al 2020).

Emergent technologies are therefore an opportunity to converge at the same table multiple actors to discuss in a multidisciplinary manner the way to go from technology to policy and from the scientific environment to society. Here, CTA and sociology of expectations have a role in the building on narratives that may lead to envisioning of scenarios that will ultimately shape imaginary solutions for innovative technologies (Borup et al 2006).

1.3.1. Sociotechnical imaginaries are part of CTA and the basis of sociological analysis to address emergent biotechnologies like genome editing

A way to address an innovation in the field of Science and Technology is through sociotechnical imaginaries. Using this approach where desirable futures are considered, these are often faced with a more positive and progressive mind (Jasanoff and Kim 2015). It might not sound as a novelty that when humans imagine and give more time to their creative process, they tend to anticipate better scenarios in their daily life (Störmer et al 2020). By adopting such an imaginative posture as practice with regards to innovative biotechnologies, one is able to visualize the potential futures that they may or will bring (Jasanoff and Kim 2015). If this has already value when done by a single individual, collective imagination is of greater value because it emphasizes the social aspect of construction in the sense that it (has the ability to) unites communities to think thoroughly about an innovative biotechnology and such an intervention will lead to higher communication with indirect parties during technology development (Jasanoff and Kim 2015, Rip 2018). STS theory itself puts science and technology at the core with the aim to stabilize collective sociotechnical imaginaries by constructing political and sociotechnical systems and this counts with diverse actors elaborating on the collective good with a communal sense (Jasanoff and Kim 2015). For years, STS studies served itself of the use of expectations in innovative technologies like nanotechnology and gene therapy. In this context, scholars have analyzed the intervention of multiple stakeholders during this process of constant envisioning and foresight practices (Borup et al 2006). For interventions that suppose embedding of a technology in society, CTA actors play an important role when they apply a reflexive tool methodology in a foresight manner as they use socio-technical scenarios to empower them by interacting and by moving their perspective from fiction to forthcoming (Rip 2018). This builds strongly on the idea evoked by the primordial mindset of sociology of science illustrating the ambivalence of scientific inquiry, in that CTA allows scientists to reflect over their actions, making them more accountable in face of their peers, governance actors and the public sphere (Bucchi 2004). STS usually takes a constructivist approach where expectations *versus* realistic achievements of a novel technology have a value for anticipative reflection, although these may also lead to the generation of technology hype, in a way that would make those who announce its promises accountable in case expectations are not met (Borup et al 2006). In practice, social actors or stakeholders are the immediate producers of imaginative futures to a point that leads to translation into policies and governance measures with regards to such a technology that will ultimately bring impact over citizens in their daily routine, namely upon development of biotechnology products that reach the

market and face public acceptance or opposition based on safety and regulatory issues (Jasanoff and Kim 2015, Rip 2018). In the early stages of technology development when the level of uncertainty is high, sociological construction of the technology with multiple stakeholders is therefore particularly relevant, as it intends to close the gap between expectations and reality (Borup et al 2006). For a democratized emergent technology as GE presently is, these gaps or asymmetries of what is socially constructed in the present and for the future may be narrowed by recurring to CTA approaches. By challenging institutions of knowledge and societies by picturing sociotechnical imaginaries or scenarios, this approach is contributing to the dimensions of RRI framework, in particular, anticipation, reflexivity and responsiveness (Rip 2018).

1.3.2. Co-construction of technology between stakeholders is a predecessor of anticipatory governance for new genetic emergent technologies

For innovation to mobilize actors, it requires co-construction of knowledge involving humans as sociological thinkers of the technical production and this commonly intends to reach a point of stabilization (Atkinson, Glasner and Lock 2009). Stabilization means that knowledge production is dependent in its societal context including expectations and values which can change over time (Horst 2007) This often involves debates between scientific and non-scientific actors because a broader participation confers more accountability on the use of new genetic technologies by assessing benefits and risks which may translated into societal consequences (Jasanoff 2003). Stem cell technologies, tissue regeneration, organ transplantation, cloning and genome sequencing are the biotechnological breakthroughs of the last thirty years that are challenging cultural norms, beliefs and naturalness conventions and being a subject of sociological analysis (Atkinson, Glasner and Lock 2009). Emergent genetic technologies are therefore in a very similar position and the amplitude of new thoughts evoked are somewhat since they intend to answer to the same questions posed at the time of these extemporaneous technologies and common to prior ones such as the recombinant DNA technology at the time of Asilomar Conference (Hurlbut, Saha and Jasanoff 2015, Hurlbut 2015, Rufo and Ficorilli 2019, Mitchell et al 2022). Earlier discussions of genetic engineering related with genetic enhancement, equity of access by citizens and genetic discrimination might be drawn on for newer genetic emergent technologies and these comprise simultaneously questions for democracy (Jasanoff 2003). As societies of knowledge, it is no longer possible to dissociate these from accountability and deliberation under democratic collective decisions (Jasanoff 2005). One of the challenges central for science governance is therefore how to ensure that citizens are not excluded from scientific knowledge production sites (Jasanoff 2003). This is

particularly challenging considering that the major science and technology innovations, also in the genetic field, are still mostly influenced by organisms that are usually not object of direct citizen participation such as the case of regulatory, legislative and even lobbying bodies (Jasanoff 2005). Public participation through dialogue about science and technology constitute one pillar of CTA by influencing the design of technology with a more proactive approach with an emphasis into its regulation (Einsiedel 2008). While reporting scientific results to the public is essential to ensure transparency and trust in scientists (Hans Peter 2008), one still needs a way to comprehensively evaluate the changes that a given technology can bring to societies and this can be achieved by creating new imaginaries through human inter-subjectivity, envisioning simultaneously its reality and potential (Jasanoff and Kim 2015). For example, the use of metaphors like "the book of life" or "genome map" during HGP era was able to drive human imagination and this was thought to bring the public closer to what the discovery of such a "map" would mean for their life in societal terms (Bucchi 2004). In a different way, coproduction or co-construction is an alternative to collectively imagine realities of an emergent technology by serving itself of the intervention of several actors in looking at how they are already transforming the world where we live and on what to expect from them in the future (Jasanoff and Kim 2015). As an example, the envisioning of consequences such as the leakage of genetic information of individuals that could lead to discrimination and other reflections associated with the use of mastectomies motivated by susceptibility of inherited breast cancer by prevalence of mutations in BRCA1 or BRCA2 genes remains one of the most successful application of ELSA/ELSI study to an emergent technology (Bucchi 2004).In the field of genomic sequencing during HGP led to activist movements after the identification of the predisposition of mutations in these genes in increasing breast cancer risk among women (Bucchi 2004, Atkinson, Glasner and Lock 2009). Citizens cannot be excluded from the progress of science and technology and they have a stake in collectively imagining scenarios of its advancement since they deposit trust in scientific institutions and support for publicly funded projects such as the already mentioned HGP (Jasanoff 2005). Despite the crystalized figure of "Asilomar-in-memory" in time, there are still reminiscences that will help in rebuilding emergent technologies assessment through the direct influence of the scientific community as the responsible and competent stakeholders to construct the governance of the technology future under restrictive lawful premises (Hurlbut 2015), it is not expected that we as a society defer this decision role to scientific experts in the twentieth-first century (Howell et al 2020) since transparency and accountability have to be represented across such authorities (Einsiedel 2008). In summary, effective mobilization of stakeholders through co-construction or coproduction may lead to more than the mere imagining of promises and

expectations of technologies and sociotechnical imaginaries can be a step forward in being an instrument for effective policy-making and shaping of forms of governance in regards to emergent technologies in genetics that allow engagement with the public to understand their hopes and desires (Atkinson, Glasner and Lock 2009, Borup et al 2006, Jasanoff and Kim 2015).

1.3.3. Scientists, ethicists, policy-makers and the general public as the chosen representatives for CTA of genome editing

For long, scientists have been active participants in the construction of social reality for science and technology whether it is about food biotechnology or stem cell technologies (Peters 2008). Making sense of the dimensions of social construction of Science has been object of work of some authors like Sheila Jasanoff which suggested the premise of "technologies of humility" when focusing in the role of multiple stakeholders regarding the uses and applications of knowledge as a path to exercise foresight (Jasanoff 2003). Stabilization may be the goal of SCOT or CTA in the first place, but acknowledgment of it as "continually co-constructed process, and therefore a temporary (though sometimes long-lasting) phenomenon" has also been emphasized as needed (Atkinson, Glasner and Lock 2009 page 2). Achieving this scenario of continuous rationalization and critical mindset with an inquisitive posture overlaps with the equal uncertain, unknown and unpredictive discomfort which constitutes the basis of "technologies of humility" rationale by defying the problem of lack of foresight (Jasanoff 2003). It has been common in the past that different views between the public and decision-makers about topics related with genetic technologies such as the one regarding GM crops in Europe in the beginning of 2000s decade led to unnecessary rioting events (Tait 2009) and anti-technology campaigns catapulted by Monsanto Company exclusion of farmers from deliberative participation over the "Terminator Gene" experience (Jasanoff 2003). These unsettlements had an impact in the route followed for the implementation of those technologies and this has usually an impact within specific bio-economies (Atkinson, Glasner and Lock 2009). Therefore, creative and effective forms of engagement between scientific experts, decision-makers and the public are necessary and these may create an opportunity for newer participatory mechanisms of citizens with real repercussions for their own lives under democratic governance of science (Jasanoff 2003). Moreover, the discussion of scientific knowledge supposes a discussion in the public sphere with citizens under democratic conditions (Rehmann-Sutter 2010). Scientists are *connoisseurs* of their research aiming at communicating it for an audience with the help of media (communicators). They may also play a role as scientific advisors for policy to

reflect in the interaction between technologies and its societal impacts once it reaches that stage (Peters 2008). Stakeholder analysis of the technology and a dialogue approach between those who develop the technology and the public who may or may not embrace it constitute a continuous feedback process where scientists, decision-makers and citizens become part of a CTA framework in order to anticipate unintended impacts, reflect over expectations and concerns as well as socially learning and constructing an innovative technology (Einsiedel 2008). We also have to recognize that such an enactment of a network of actors might be less interactive and more "pointed to", an expression known as "division moral labor" that supposes that one type of actors (e.g. scientists) indicate other experts in other fields (e.g. regulators) to act over issues that are more likely for them to act through (Rip 2018). This may therefore be a prelude to better socially construct an emergent technology like genome editing and the more rational way to govern the CRISPR world where we live nowadays (Hurlbut 2018).

It is therefore absolutely necessary to convey multiple visions about a technology (regardless of its format) that will stay with us for the next decades and which at some point will need to reach a certain "stabilizing" point. Whether we are prepared to embrace it and in which forms we can do so, it will depend in the work done together by conjugating efforts between the so-called research experts in the technology, decision-makers, clinical staff, industry partners, farmers and ultimately, citizens.

In order to effectively infer about the perceptions of stakeholders regarding genome editing as a predominant emergent biotechnology of the twentieth-first century that already motivated punchy headlines in media (Ledford 2016, Regalado 2019, Regalado 2014), innovative clinical trials (Frangoul 2021, Stadtmauer 2020), drug formulations based in genome editing (Mhukerjee 2022), the resurgence of narratives about eugenics (Bosley 2015, Ishii 2017, Nordberg et al 2018, Rosemann et al 2019), and "disease-free" (Wahlberg 2021), it is fundamental to decide the best route to take when implementing it in society for citizens and which conditions should be ensured when doing so. After all, predicting the harm-risk/benefit of a technology with so many promises, hopes and fears needs the participation of everyone that will be impacted by the technology in the short- or long-term future and if one of the purposes is to better advise decision-makers about its use, understanding what experts and citizens have to say is a work for today while envisioning tomorrows.

1.4. RRI as an actionable principle in educational community-based environments

In higher education the proposal of a connected curriculum has been drawing attention in recent years (Fung 2017). Of particular interest in the context of the present thesis the idea of the connection between areas of expertise and with society. I can also stress from the connected curriculum proposal that when such a connection is reinforced there is an orientation of learning to research – the so-called inquiry-based learning – which is a practice internationally recognized involving dialogue and collaboration between students and their teachers or peers. The idea of testing and more specifically of pushing the limits of RRI needs therefore a space where future researchers are able to practice over its dimensions, principles and processes associated with real situations with clear applications and brainstorming processes. The idea of enticing questions that promote learning for research is the core of Course-based Undergraduate Research Experience (CURE), which has been highlighted in recent years (Bell et al 2016), and the association of RRI to this type of initiatives will reinforce it. This approach is pertinent in academic environments and particularly with future researchers who are in their early years of education and in the verge of transition from academia to research (Almeida and Quintanilha 2016, Gerrits, Breedenord and van Mil 2020).

RRI supposes co-creation and therefore a collective effort to address challenges that are dressed of social desirability and assumes the role of establishing a dimension of social responsibility from individuals linked to Science to citizens (von Schomberg 2013, Owen, MacNaghten and Siltgoe 2012). To address contemporaneous issues related with sustainability and health, there are initiatives that may be used conjugating both a reflexive and a practical approach. Some initiatives such as the already mentioned FoTRRIS project which addresses “grand environmental and societal challenges” by reflecting on glocal challenges imposed by ecosystems sustainability and social inclusivity (Ruszanov and Haese 2015) or the project of X that does Y are successful examples of these type of initiatives. Foundations are therefore thrown in order to promote courses and educational resources that emphasize the connection between academia, research and RRI shifting the concept to practice and allowing students that will be future researchers to be agents of change through their own initiatives. Some initiatives have therefore been laid out in recent times such as the Higher Education Institutions Responsible Research and Innovation (HEIRRI) project and RRI tools that integrate RRI in the pedagogical context (Tassone and Eppink 2016, RRI tools 2016).

Science, Technology, Engineering, Arts and Mathematics (STEAM) has been advocated as the way to link Science with other disciplines (European Commission 2015). In the

last years, this led to the creation of informal environments with a strong component of scientific education gained relevance over the last years both in universities and community-based spaces that are sometimes side projects or side features of universities and that exist between these and research institutions (Gerrits, Bredenoord and van Mil 2020, Scheifele and Burkett 2016).

Chapter 2. Qualitative and quantitative research to investigate stakeholders' views about genome editing technology

2.1. How interviews with stakeholders may contribute to the perception of a biotechnological revolution led by CRISPR-Cas technology

Qualitative inquiry as a social science method has the purpose to make sense of the world where we live in and it studies people and how they make sense of things, in other words, how people construct meaning (Patton 2015). Qualitative researchfocuses on exploring the way people perceive how things work in the real world and aims at capturing such understandings and personal or professional meanings which ultimately will also be relevant for collectives (VanderStoep and Johnston 2009). Qualitative inquiry intends to capture stories, perspectives, experiences and behaviors from people while noticing patterns by observation or by listening to individuals and then extracts possible repercussions from those views (Patton2015). In contrast to quantitative research where it is often assumed that the knowledge just needs to be "discovered", qualitative research assumes the need to "construct" such knowledge and that this may only happen in a co-creative way, i.e. through interaction between individuals that communicate and express their own meanings (VanderStoep and Johnston 2009). In fact, qualitative research has a holistic overview of the world looking into context and accounting for factors that are much more dependent on the community level rather than dictated by the individual alone (Patton 2015). This rationale where individuals are somewhat looked into and analyzed in a "big picture" mode contributes to the concept of social construction of reality (VanderStoep and Johnston 2009). More than summarizing data in numbers that may be statistically assessed in order to find significant changes between certain variables as in the case of quantitative research, qualitative inquiry uses in-depth methodology to go from the individual contextually inserted in their own world and this of course renders descriptive and heavy data (Miles, Huberman and Saldaña 2014, Patton 2015). Qualitative data also brings a personified view of the world and this usually has more impact for decision-makers in the ground of the field that is being explored (Miles, Huberman and Saldaña 2014). Qualitative research supposes then that only by approaching others that are there, in the field, in a closer way, one will be able to really understand what they understand and by sensing those experiences, the unanticipated consequences might be assessed more faithfully (Patton 2015). Resultant qualitative data is an aggregate of individual's view and it aims at influencing those same individuals in the first place (VanderStoep and Johnston 2009). The issue of data generalizability or aggregated generalizations may be considered a limitation in certain contexts (Miles, Huberman and Saldaña 2014). Nevertheless, it provides a different and why people think what they think and provide meanings for findings than quantitative studies do since these are not self-explanatory either if we are talking about surveys or other research

data in the field of life sciences (Patton 2015, VanderStoep and Johnston 2009). Qualitative data is, in this sense, more prone to yield unpredicted conclusions and abler to identify unanticipated consequences because it often doesn't have a predictive rationale behind (Patton 2015, VanderStoep and Johnston 2009). You simply cannot predict what you still don't know what you are going to find out and therefore, digging the unexpected and exploring the unknown is in itself a process of inquiring both the intended and the unintended (Patton 2015).

Within qualitative research, different methods might be applied to obtain an in-depth overview of individuals' meanings, but all-in-all, those methods resemble an inductive approach which emphasizes a trajectory from observations or conclusions to theory and hypothesis formulation (VanderStoep and Johnston 2009, Braun and Clarke 2006). This methodological logic has its roots in grounded theory where fieldwork is carried out with in the real world in direct contact with other scholars and people directly involved in decision bodies (Patton 2015). For Glaser and Strauss, the theory is grounded in the data that will be extracted from interaction with participants in a study and these will precede the formulation of hypothesis (Glaser and Strauss 1967). Grounded theory supposes the use of coding, a method that relies on the attribution of codes which work as categories, dividing the data to better identify patterns (Miles, Huberman, and Saldaña 2014, Saldaña 2009). This process is a continuous step part of content and thematic analysis, where those patterns will lead to the emergence of themes reflecting the meaning of individuals in a structured way that will answer to research questions that have surfaced or have been questioned beforehand (Miles, Huberman and Saldaña 2014, Braun and Clarke 2006). One may as well adopt a mixture of methodology like mixed-methods research where both quantitative and qualitative methodology will be used (Patton 2015, VanderStoep and Johnston 2009). Different approaches throughout the phases of qualitative research seem also an equally relevant tactic leading to an interchange between subjective and objective paradigms since this may also demonstrate the ability to vary within the presence of an initial research question or not in the beginning and the need to achieve scientific rigor, in other words, an intertwining between inductive and deductive approaches (Mayring 2014). In the end, what really counts for a good, qualitative research relates to the quality of the data in the sense that it really needs to reflect the conceptions of participants of a study faithfully and this needs to be supported by "rich descriptive examples that persuade the reader to adopt the researcher's interpretation of the text" (VanderStoep and Johnston2009). For this to happen, collection of data based on skillful inquiry and systematic and rigorous analysis are fundamental steps that have to be carefully designed and executed in order to either

reply to a research question initially conceived or to discover research questions (Patton 2015). Some scholars support a narrative of traditional methodology basing its research process in an initial research question that will be linked to a theory and from where all the design flows independent of the adoption of a more exploratory or causal one (Mayring 2014). Others, however, consider themselves “shameless eclectic” not obeying to any genre of qualitative research specifically and focusing more in the process of qualitative analysis rather than the method of data collection and paradigm that abides to (Miles, Huberman and Saldaña 2014). Nevertheless, qualitative research has a strong purpose: making sense of the world contributing to knowledge by generating or testing a theory with the likely aim to protrude to fields out of social research, namely to policy-making environments (Patton 2015). Understanding the world of participants that are inquired is part of the holistic overview of the researcher and this may stay in the scientific domain of academic journals or it may also resemble an action-driven research in the sense that problem-solving is embedded in the mind of the researcher conducting the study and as an agent of change (Miles, Huberman and Saldaña 2014, VanderStoep and Johnston 2009). The researcher’s role in qualitative research is in fact one of the most important one since itis his or her ability in interacting and engaging with participants and depends of a prolonged contact between them to gather an in-depth understanding of their meanings of the world where they are socially embedded (Miles, Huberman and Saldaña 2014, Patton 2015). Qualitative researchers are also looking for depth rather than breadth and although many interpretations of the data are possible, not all interpretations may in fact be the result of the research conducted. When adopting qualitative methodology in practice, different inductive methods such as in-depth interviews, observations, and written communications may be used (Patton 2015). In fact, there are more than twenty qualitative research genres, but analytically-speaking, they all have in common the attribution of codes or themes, meaning the identification of patterns in the collected data that resemble a given category and the process of isolation of those patterns as a way to distinguish similarities and differences to make generalizations that are consistent and which allow for comparing with existent theories (Miles, Huberman and Saldaña 2014). The use of open-ended or in-depth inquiries allow the generation of open-ended responses that are detailed and this attributes a holistic and rich character to the qualitative data collected which is definitely one of its biggest strengths (Miles, Huberman and Saldaña 2014, Patton 2015). It is also acknowledged that the major way where qualitative researchers may collect this in-depth data is usually through intense interviewing (Patton 2015).

2.1.1. Interviews as qualitative research yield comprehensive themes and help in theory construction

Open inquiry is a form of qualitative methodology that uses open-ended questions to make meaning of what is happening in the real world (Patton 2015). The idea that data arises from people's observations and explanations traces back to the grounded theory of Glaser and Strauss which assumes that data is grounded and can be extracted in an inductive manner. In their own words:

> […] we suggest that it is likely to be a better theory to the degree that it has been inductively developed from social research… Generating a theory from data means that most hypotheses and concepts not only come from the data, but are systematically worked out in relation to the data in the course of the research. (Glaser and Strauss 1967 page 5 and 6).

Even then, the use of in-depth inquiry cannot be dissociated from interviewees that speak about the features and consequences of a given reality where they inhabit or have contact with even if a scenario-making is behind that process assuming therefore a connotation of social constructionism or constructivist framework (Patton 2015). Open qualitative inquiry in the form of in-depth interviews resemble both frameworks since while inquired people construct reality at the same time they speak, it is the fact of making sense of this data following its extraction that results in a process of rediscovering the what is grounded on it and that one keeps observing continuously leading to the identification of patterns in the data that when categorized may result in the emergence of themes that help answering research questions (Braun and Clarke 2006, Patton 2015). Qualitative research is well-suited of different methodology to perform comprehensive approaches when collecting data from participants, namely through interviews, focus groups, observation, discourse analysis and even study of audio and written documents (Braun and Clark 2006). Interviews are the most frequent method used in qualitative research (Patton 2015) resembling a constructivist theory paradigm due to its inductive nature which alludes to a process of social construction of reality (Braun and Clarke, 2006, Mayring 2014, Van der Stoep and Johnston 2009). This type of research method entails the collection of data voiced by participants which are later transformed in written excerpts that are usually rich and meaningful for researchers during and after inquiry process (Chapter 1 Braun and Clarke 2006). Qualitative inquiry allows to understand context on how and why a given subject (person or topic) matters, to identify unanticipated consequences of such subject and (to identify) categories and patterns within case-studies which will ultimately lead to generation of themes (Braun and Clarke 2006, Miles, Huberman and Saldaña 2014, Patton 2015). Likewise, not just

consensual inductions from the data are accommodated but also the divergent features of it (Braun and Clarke, 2006). Researchers aim at finding intended and unintended patterns and, in the end, construct a theory (Braun and Clarke 2006, Patton 2015).

Data collection is important for qualitative inquiry because of the fidelity of the data and structure of the method used for data collection (Van der Stoep and Johnston 2009). Both these features influence the strength of qualitative data in the sense that they are stronger if data is naturally occurring avoiding biased collection and complex due to its soundness and richness of it (Miles, Huberman and Saldaña 2014). To strive for fidelity is to strive for the most accurate representation of what inquired ones tell and structure relies in the flexibility of interviewer approach in the sense that a higher flexibility or lower structure intends to capture deeper data by providing the ability to adapt questions, question questions that were not predicted initially in a guide and therefore contribute to higher richness of the data (VanderStoep and Johnston 2009).

An approach using semi-structured or guided interviews where it is aimed for the researcher to obtain comparable data between inquired ones while still holding flexibility in the flow of the interview by having the ability to make questions out of the prepared guideline is a compromised approach between flexible and structured interviews (VanderStoep and Johnston 2009, DiCicco-Bloom and Crabtree 2006) and which may be of use in social constructivist frameworks of qualitative inquiry (VanderStoep and Johnston 2009, Patton 2015).

2.1.2. Qualitative research: the purpose of semi-structured in-depth interviews

Low structured interviews also called informal interviews are usually held for exploratory themes to feature in soon-to-be projects not requiring a preset of questions (a go-with-the-flow approach) and leading to more naturally occurring questions and unexpected theories while highly structured ones intend to be rigid with the interviewer following guide and to capture only what has been designed in the list of questions prepared in the first place leading to less unexpected outcomes and leaving the interviewer also out of pursuing serendipitous discoveries retrieving more comparable data suitable for generalization (Patton 2015, VanderStoep and Johnston 2009). In order to respect the dimensions of social constructionism where different groups of people construct multiple realities about the real world and unlike phenomenology paradigm intend to capture intersubjectivity of real people, the researcher is expected to retain malleable guidelines while collecting data and should be engaging at the same time in order to consider additional perspectives from the ones initial laid out in the preparation of the qualitative inquiry (Patton 2015).

Semi-structured interviews are an instrument that incorporates the mean to achieve the overlap between inductive and deductive analysis that will follow. This is due to its flexible and hybrid nature since it includes open questions that are initially set in a guide but they also have space to include questions that may come from the interaction with participants that are being interviewed (DiCicco-Bloom and Crabtree 2006). This interview format intends to capture people answers about a topic but also reflections towards central questions that are presumed to guide the interviewee (Jamshed 2014). Semi-structured interviews can be conducted individually or in a group and they have a time estimation that may vary between 30 minutes to several hours due to its flexible nature (DiCicco-Bloom and Crabtree 2006, Jamshed 2014). Therefore, recording of this type of interviews is advised in the sense that notes and memos during the interviews may be insufficient to capture all dimensions of answers from participants (Jamshed 2014). These recordings suppose a following step of data transcription that occurs generally in an *ad verbatim* mode with the aim to transcribe everything from participants to a text that may later be easily and thoroughly analyzed (Jamshed 2014, Saldaña, Miles and Huberman 2014). This way, content analysis of what is grounded in the data will allow one to make sense of participants answers and argumentation about the topics explored throughout the whole interview (Miles, Huberman and Saldaña 2014).

Semi-structured interviews comprise an in-depth interviewing methodology that have sufficient length, flexibility and complexity to help in capturing arguments and rationale of participants (Jamshed 2014). This type of interview is therefore adequate for data collection from participants with the aim to extract intended and non-intended views and with that, help in theory construction (Patton 2015). Additionally, semi-structured interviews result in the generation of data that may analyzed in terms of content and these are usually object of thematic analysis (Braun and Clarke 2006).

2.1.3. Grounded theory, qualitative content analysis and thematic analysis

As previously explored, grounded theory is a usual line of thinking of qualitative research that seeks to identify theories hidden in the data that is collected in a mostly inductive logic allowing for connection with deductive ones if to test the theory that is emerging from it by performing certain types of fieldwork (Braun and Clarke 2006, Patton 2015). In order to unveil the theory that data conceals, qualitative content analysis through a series of tentative codes in early collected datasets aims at sorting and synthesizing them in a clear way (Patton 2015, Miles, Huberman and Saldaña 2014). Coding is a research action to develop categories that were not initially drafted before the qualitative inquiry takes place (Patton 2015). The approach supposes the use of words, expressions,

gerunds or any similar to data extracts such as interview transcripts to better identify similar excerpts that resemble similar categories and allows for distinction of relationships of some of these categories that may fall under different groups or subgroups within these (Miles, Huberman and Saldaña 2014). Using memos or the use of notes while a qualitative inquiry takes places may be a complement to be added and that may ease the connection of categories identified during the process of data coding and help in sorting patterns out of it which when zooming out from the data may make similarities and differences among codes and patterns clearer and more evident (Miles, Huberman and Saldaña 2014, Patton 2015). In this sense, we are moving from an inductive to deductive approach and undulating in the field of grounded theory where theory is assumed to be grounded in the data and not forced to appear illustrating what is perceived by participants in reality (Glaser and Strauss 1967, Patton 2015).

Thematic analysis corresponds to the process of discovering themes through common patterns that start to emerge from data (Braun and Clarke 2006). Once one starts to see a repetitive idea across excerpts with high frequency and emphasis, one is starting a theme identification (Braun and Clarke 2006, VanderStoep and Johnston 2009). The process of theme discovery is not closed since there are themes that may eventually be deconstructed into shorter or complex ones with the aim to develop a consistently ramified pathway that will help in answering the research questions (Braun and Clarke 2006). It is after theme identification that a theory or a research hypothesis can be formulated and be evaluated in terms of its consistency and reliability due to the level of correlations that can be established with the remaining data (Patton 2015). Moreover, a continuous process of going back and forth between the discovered themes from the data and the interview transcripts ensures the inductive character of the research and directs the research to newly formulated theories that are stronger in describing a given phenomenon or reality (Patton 2015, Braun and Clarke 2006).

This consolidates the essence of approaching stakeholders through in-depth interviewing methods in order to construct the reality by hearing their reflections and anticipative thinking (Patton 2015, VanderStoep and Johnston 2009) towards an emerging technology such as genome editing and this way, to better understand the (un)anticipated consequences that the technology poses to current and future generations.

In a study that involves capturing views of experts in both an exploratory and precise way, semi-structured interviews are an adequate option and they will be the instrument used for addressing researchers.

2.2. Surveys to citizens as a mean to address public opinion

Surveys or questionnaires, are closed and structured instruments employed in research to obtain a well-defined and precise picture of a certain topic in a specific period of time (Stockerner 2019). Publics are dynamic and may have opinions and positions passive of discussion in the support and approval of subjects that will impact them (Pereira and Völker 2020). Opinion surveys intend to analyze the views in a given context regarding a specific topic and they may happen in the form of a cross-sectional survey whose objective is to draw a picture of a precise point in time or as longitudinal surveys that follow individuals' opinion over time (Stockerner 2019). Additionally, drawing on attitudes of populations about subjects features in the materialization of "explicit measures" where citizens are inquired to answer research questions (Haddock and Maio 2008). Cross-sectional surveys are intended to answer any kind of research questions and a mean to test a theory due to its singular nature and the usual option that researchers adopt when drawing on populations' view about a specific subject (Stockerner 2019). The limitations of these surveys are therefore related with the difficulty in illustrating the change in behavior of individuals towards specific subjects over time and the challenge of not being able to infer about causality due to the same reason (ibidem). Quantitative research falls under the positivist theoretical paradigm where the world is seen by researchers in an objective form and where a hypothesis has a binary result: accepted or rejected (VanderStoep and Johnston 2009). Surveys are instruments in social research that help in the objectivity of quantitative research because they are constructed with closed options aiming to capture individuals' answer in a delimited form (Stockerner 2019, VanderStoep and Johnston 2009).

2.2.1. Surveys as instruments of stabilization of GE technologies

Stabilization is one of the objectives of STS approach and a central role of collective foresight when it comes to emergent technologies (Atkinson, Glasner and Lock 2009, Konrad et al 2016). This process of stabilization concerns the way these technologies stand in people's eyes and how they diagnose regarding its expansion, generalization and fears or opportunities they might present (Atkinson, Glasner and Lock 2009). In order to infer on how closer emergent technologies are or not of reaching stabilization, surveys are therefore one of the common instruments of choice to measure the status of emergent technologies and the future directions that these should take (Braun and Meacham 2019). By constructing valid and reliable surveys that measure citizens' opinion about technologies of genetic modification, one will have the chance to understand in which terms the technology may or not develop and be or not embedded

in society as soon as social stakeholders are involved in consultation phases (CoGEM 2018). Citizens may participate as sensors in a data collection phase, in stages of problem definition and data collection under the form of participatory science, in data collection and analysis interpretation phases (distributed intelligence) and under an extreme citizen science where they participate all the way from problem definition to communication engagement (Catalão Alves 2020). Consultation of citizens under the form of surveys and polls may be less engaging despite being a more direct way to measure public opinion (Scheufele et al 2021) falling under a low structured decision-making axis but with considerably high level of public transparency (Catalão Alves 2020). Moreover, recent surveys have shown that it is indeed possible to provide a view of citizens about emergent genetic technologies and therefore draw a picture of the technology status and possible paths for adoption based on support and rejection as well as to understand the reasoning behind those attitudes (Busch et al 2021, Jedwab et al 2020, Sawai et al 2017, Watanabe et al 2020). Furthermore, results of these consultation processes already led to considerations by authors in the field of STS advocating for citizens to be placed in a more upstream and central position of deliberation and intervention under the form of citizens' assembly and civic science initiatives so that decisions made are in the best interest of society, ensuring trust, transparency and respecting citizens' will (Dryzek 2020, Scheufele et al 2021, Wirz, Scheufele and Brossard 2020).

2.2.2. Systematic review as the method of choice to make sense of data collected in surveys with citizens about GE

A systematic review is predominantly described as the gold standard approach to establish scientific evidence due to its exhaustive, comprehensive and complete nature in comparing the studies that have been done about a certain topic throughout a period of time (CRD 2008, NHMRC 1999, Moher et al 2009, Petticrew and Roberts 2005). Systematic reviews also have a strong purpose for social policy environments (Petticrew and Roberts 2005). Cross-sectional surveys present limitations in drawing a picture of the behavior change of populations that are inquired (Stockerner 2019), a limitation that can be partly overcome by systematically reviewing a number of cross-sectional surveys carried out at different time points. Systematic reviews constitute a method able to map existing studies where research about a topic has been made as well as a method that would make sense of that large body of evidence (Petticrew and Roberts 2005). Moreover, they aim at identifying, assessing and synthesize the evidence from the group of studies included to provide an overall idea of the topic that is being researched to feed

decision-makers (CRD 2008). Furthermore, systematic reviews are also a mean to conclude if there is the need of more studies to undertake regarding that certain topic where knowledge might be absent (CRD 2008, Petticrew and Roberts 2005). Overall, this is a type of method which strives for context about populations and elevates the statistical significance to social significance (Petticrew and Roberts 2005).

Systematic reviews suggest a reproducible and strict methodology, aiming for a rigorous process and mapping of the most complete body of evidence as possible (CRD 2008). In addition, they should aim to critically assess the methodological quality of the individual studies that are analyzed and therefore they suppose methods to assess such quality (CRD 2008, Petticrew and Roberts 2005). These comprise methods such as the meta-analysis of the data that is presented in the individual studies analyzed as well as a critical appraisal of the methodology employed (CRD 2008, Petticrew and Roberts 2005, Moher et al 2009).

Critical appraisal of the questionnaires suggests a measure of quality of the studies due to its type of assessment of parameters that go from the content of the questionnaires, to its validity and reliability (Boateng et al 2018, Hair et al 2019) ending in parameters that are related with the sampling method used, the type of bias that may occur (Stockerner 2019) and the compliance with ethical and privacy standards (Fanelli et al 2009). Face validity intends to measure what is supposed regarding the object of the study, content validity assesses how understandable is the language and the questions for respondents and construct validity concerns the efficacy of measurement of the instruments used (Boateng et al 2018, Bolarinwa 2015, Hair et al 2019). Instruments like scales may be variable (Likert, forced, non-forced, etc) but they always have the objective of measuring respondents' view about a subject and this may be done for unstructured or structured questions (Malhotra 2006). Moreover, the instruments always have to be validated before being implemented in the surveys so that all inquirers know what they will measure and if the measure will have an explicit outcome (Boateng et al 2018). Tests of validation and reliability of scales usually take place in the final stage of questionnaire preparation followed by pre-testing of the questions in pilot studies (Boateng et al 2018, Malhotra 2006). Furthermore, questions' structure and wording should be as unambiguous, simple and avoid biasing respondents (Malhotra 2006).

Systematic reviews of questionnaires applied in some of these settings have also been done with the aim to infer about the quality of the surveys administered to respondents and how well these answered to initially conceived research questions (CRD 2008, Petticrew and Roberts 2006). The adequacy of instruments to answer research

questions is the most important aspect of questionnaire design (Malhotra 2006) and for that to be a reality, critical appraisal of questionnaires is part of these type of scientific article and considered an essential requirement that resembles quality (Moher et al 2009). Moreover, one has to consider how knowledgeable and willing are respondents to answer a questionnaire and therefore questionnaire design and efficacy has to be taken into account (Malhotra 2006) and this elevates the objective of systematic reviews that intend to assess these instruments in surveys (Pace et al 2012). When applied to cross-sectional surveys, systematic reviews are then able to not just collect the answers of a group of respondents about a subject in a more longitudinal way due to the insertion of a time variable but these are also able to assess the quality of the surveys that have been administered to respondents during the same time period.

Chapter 3. Methods

This PhD thesis addresses different materials and methods that were already explored in theory in the chapter 2.

The two studies presented in manuscripts 1 and 2 are the result of the use of qualitative methodology as methods. The study in manuscript 3 is the result of the implementation of course unit following a CURE approach already described in chapter 1.

In manuscript 1, a qualitative in-depth study in the form of semi-structured interviews was used with the aim to capture expert views about genome editing. These semi-structured interviews were devised by building an interview guide resulted from a conceptual matrix (Appendix I and II). The conceptual matrix was divided in concepts, dimensions, sub-dimensions and expected outcomes which later translated in the form of open-ended questions to be integrated in the interview guide. The interviews were done with researchers recruited worldwide in an online format. All transcripts were transferred to NVivo 11 software to be coded and data was later analyzed and organized in themes with the aim to present the whole relevant data in the findings section of manuscript 1. The generated codebook can be found in the appendix III. Interview transcripts were generated by recurring to an automated transcribing method named AUDIT (Audio-dictated transcribing software) presented at the Cool Tools for Science event in 2021 (Appendix IV). All specific details are presented in the materials and methods section of manuscript 1.

Manuscript 2 introduces the use of a systematic review of studies reporting surveys that were administered over the last 35 years to citizens with the aim to capture their opinion towards genome modification in a period before CRISPR technology was known (pre-CRISPR) and after its advent (CRISPR period). Surveys with citizens were searched, selected generating 53 studies that were further explored. These studies were analyzed in terms of data with the aim to collect the views about support for genetic modification of humans and animals and a critical appraisal was performed to assess quality of the studies. This analysis resulted in figures 1-5 of manuscript 2 and in table S1 that belongs to Appendix VI.

Manuscript 3 presents the strategy and methodology used for the BioLab divided in three stages: capacitation, engagement and action. Capacitation supposes lectures on diverse topics from teachers, role-playing and engaging activities. Engagement focuses on a call for ideas and a call for projects to which students had to adhere as well as on lectures from external stakeholders and study visits. And action involves steps from ideation, to weekly pitches and discussions, project development, and finally to presentation, prototyping and conceptualization of projects.

Participants

Participants from the study described in manuscript 1 are all biomedical and animal science researchers with a PhD and active research in genetic modification technology in a total of 22 interviewees of different nationalities and cultural backgrounds living in different parts of the world. They were recruited in the period between 2018-2019 and the sampling method used is further described in the materials and methods section of manuscript 1. All participants of the study signed informed consents (Appendix V).

Participants in the study described in manuscript 2 are indirect participants since these are citizens that had been surveyed already by other teams of researchers in the period 1987-2020. Participants were recruited by random and purposeful sampling depending on the studies.

Participants from BioLab (manuscript 3) are university students with multiple backgrounds (from natural sciences to media) and from different study cycles (from bachelor to PhD) in a total of 12 participants. They applied to be part of the curricular unit and they were all accepted.

Ethical approval

The study displayed in manuscript 1 received ethics approval from the Ethics Committee of Institute of Biomedical Science Abel Salazar, in May 2018 (process number 265/2018/CETI). All participants signed an informed consent form at the time of their participation in the study, but no personal data besides the name was collected.

Manuscript 2 reports a systematic review of published literature and ethical approval does not apply.

The assessment of pedagogical practice implemented in BioLab (2020 edition), included in manuscript 3, was done by a questionnaire applied to students that participated in the course and received ethics approval in the end of 2021 ICBAS (process number 2021/CE/P28).

Chapter 4. MANUSCRIPT 1. Safe and purposeful genome editing under harmonized regulation for responsible use: views of research experts

Manuscript 1 (Accepted, in press)

Pedro Dias Ramos[a,b]*, Maria Strecht Almeida[b] and I. Anna S. Olsson[a,b]

[a] *i3S – Instituto de Investigação e Inovação em Saúde, Universidade do Porto, Porto, Portugal;* [b] *ICBAS – Instituto de Ciências Biomédicas Abel Salazar, Universidade do Porto, Portugal*

pedro.ramos@ibmc.up.pt *corresponding author

Pedro Dias Ramos, ORCID: 0000-0003-3389-1682
Maria Strecht Almeida, ORCID: 0000-0002-8032-7390
I. Anna S. Olsson, ORCID: 0000-0002-4369-8723

Safe and purposeful genome editing under harmonized regulation for responsible use: views of research experts

Abstract

CRISPR-Cas9 revolutionized the precise editing of mammalian cells genome. The present study explores genome editing (GE) in the context of the Responsible Research and Innovation framework for emerging technologies, through semi-structured interviews with life sciences researchers worldwide.

Our study demonstrates that for researchers in the field, GE technology is viewed as promising but also harboring unsolved challenges. These experts call for complementary research to improve the technology and increase knowledge of the genome function. They clearly do not support what they perceive as unsafe, unpredictable and irrelevant applications, and they view the lack of international harmonization of regulation in combination with cultural differences in public attitude as difficult challenges. Interviewees see public misconceptions as a problem while recognizing the need to foster a clear science-society dialogue with informed citizens.

This study with scientists provides insight into the science-based priorities for GE to be a technology that can be responsibly applied.

Keywords: CRISPR-Cas9; genome editing, RRI; expert views; science and technology studies, thematic analysis

Introduction

Clustered Regularly Interspaced Palindromic Repeats and its associated nuclease Cas9 (CRISPR-Cas9) was first presented as a revolutionary biotechnology tool for precise genome editing of mammalian cells in 2012 (Barrangou and Horvath 2017). Since then, numerous labs have shifted from traditional genetic modification systems to CRISPR-Cas9 genome editing (GE) technologies (Adli, 2018), which are cheaper and more straightforward at equal or higher precision (Adli 2018; Camporesi and Cavaliere 2016). The rapid uptake of the technology is accompanied by ethical (Brokowski and Adli 2018), regulatory (Isasi, Kleiderman and Knoppers 2016), policy (Nordberg et al. 2018) and societal (Howard et al. 2018) concerns including safety and ethical implications, making GE a highly relevant case within science and technology studies.

The research community's efforts to deal with the wider implications of GE has translated into events, guidelines and even requests for a moratorium for certain applications (de Lecuona et al. 2017). When participating in this discussion, scientists are effectively engaging in constructive technology assessment (CTA) and Responsible Research and Innovation (RRI) even though they may not be familiar with these concepts. Both CTA and RRI are based on the understanding that different actors can co-produce sociotechnical future scenarios (and imaginaries) to facilitate reflection over science and technology and how to use it responsibly for society (Borup et al. 2006; Jasanoff and Kim 2015). RRI builds on the ethical, legal and social implications (ELSI) framework developed by philosophers, social scientists and molecular biologists (Bucchi 2004; Felt 2018) and it is a public policy that envisages knowledge production between several actors for more responsible techno-scientific futures (Rip 2018). RRI enacts the four dimensions of inclusion, anticipation, responsiveness and reflexivity (Owen, Macnaghten and Stilgoe 2012), which translate into a multi-stakeholder effort to anticipate scenarios, critically reflect and timely respond in an in-depth manner to positive and negative implications of emerging technologies (Felt 2018; von Schomberg 2013). Discussing challenges, potential and applications is important for effective policy-making and responsive anticipatory governance with regards to GE as an emergent technology (Hurlbut 2018; Hurlbut, Saha, and Jasanoff 2015). Including multiple stakeholders promotes foresight and technology assessment in the name of "collective expectations" (Konrad et al, 2016), somehow addressing the premise of "technologies of humility" as a mean to deal with uncertainties as part of this debate (Jasanoff 2003). Within this theoretical framework for technology anticipation, exploring expert views in a particular

topic with the aim of developing arguments and reasoning on technology assessment seems a significant approach.

The discussion of the societal consequences of GE has a history of several decades, but the emergence of CRISPR-Cas9 is changing the technological potential in a way that makes the discussion much more urgent. This became evident in the course of the present study when the birth of the first genome-edited human babies was announced (Regalado 2018) and since then the significance and implications of the case (He Jiankui's case) have been explored (e.g., Meyer, 2022; Wahlberg et al. 2021).

In the present study, we aim to understand how scientists using CRISPR-Cas technology reflect on the technology and its use. Using an in-depth qualitative approach, we explore what researchers see as realistic, desirable and concerning in developing and applying GE and the societal and governance consequences of its implementation for human life. The imaginaries constructed by researchers are structured into three overarching themes where they provide both problematization and ideas to deal with the development and the future of the technology from bench to bedside across species and across the world.

Methods

Interview guide design, and recruitment of participants

The interview guide included open-ended questions combined with a ranking exercise representing hypothetical applications of GE (Supplementary material). The interview guide was triangulated within the research team and tested in two pilot interviews with researchers.

Participants were recruited using convenience and snow-balling non-random sampling as well as purposeful sampling. They were recruited from different parts of the world to ensure geographic diversity of perspectives and experience. All participants are researchers from biomedical (BSR) and animal sciences (ASR) holding at least a PhD and with previous and/or current experimental practice and published records on genetic engineering, preferably on GE. Recruited researchers could be working with GE in human/animal cell models or in vertebrate organisms. From a total of 32 invitations, 22

researchers accepted to be interviewed, including researchers from Europe, Brazil, North America and China. The interviews were done online or by telephone and lasted 60-120 min, taking place between July 2018 and March.

Interviewing and thematic analysis

Audio records of interviews were collected. Transcription was achieved resorting to an automated method – AUDIT – which simultaneously allowed preservation of participant's anonymity (redacted for blinding). Transcripts were imported to NVivo 11 software (NVivo, QRS International (Americas), Burlington, MA) for thematic analysis.

Our study followed a hybrid inductive and deductive coding process. Initially, a subset of transcripts were coded manually and separately by the first and last authors. Through a first triangulation step, a codebook was sketched (Supplementary material). A second coding step was performed by the first author covering all transcripts. A sub-coding process retrieved additional refined codes stemming from some of the first ones. Following this coding process, the three authors worked together identifying patterns in the data leading to the generation of themes.

Ethics compliance

This study received ethics approval from the Ethics Committee of our institution, in May 2018 (process number 265/2018/CETI). All participants signed an informed consent form at the time of their participation in the study (Supplementary material). No personal data other than name was retrieved and the results are presented using pseudonyms.

Findings

The main story emerging from the qualitative analysis of the interviews with scientists is one about how to develop gene editing as a technology so that it can safely be used outside the laboratory, about its acceptance and about the challenges of governance internationally in a diverse context in terms of societal acceptance and access. In the following, the results of the analysis are organized according to the overarching themes technology, governance and society.

Technology: Technical challenges of GE need to be solved to make it safe enough to proceed to in vivo applications

Of the many different specific challenges of GE identified by the interviewees, the vast majority have implications for safety, with consequences for the different applications of the technology. The main challenges identified are the efficacy of introduction of genetic traits in species, the delivery of the CRISPR-Cas components, the efficiency of the homology-directed repair (HDR) pathway to introduce gene segments as well as the fidelity of its insertion (adverse on-target effects) and finally the undesired effects of off-targets of the CRISPR-Cas components and the genome complexity of organisms. Figure 1 shows an overview of the challenges and how they relate to each other, and to the potential implementation of GE technology in different contexts. Whereas the interviewees repeatedly highlight the need for researchers to improve technology in order to overcome these challenges, they also express that if the technology is considered as "not safe enough" it is preferably to explore GE *in vitro* and avoid using it for *in vivo* applications.

Technology development and application in multiple organisms

GE technology has been advancing at a fast pace. As noted by one of the interviewees: "[…] the beauty of Cas9 is that there are no technical hurdles, there are virtually no technical hurdles to getting the genome to edit." (Megan-ASR). This opens the door to a wide range of applications: "[…] with the technical level that we have now it's clear that [it] can be applied to all kind of species." (Jay-BSR). With the possibility to correct multiple genes simultaneously, CRISPR-Cas9 reduces the time for gene correction:

> And that means not work only at one at a time. But 10 genes or just any number we want, let's say something between one and 10 […] to finally become faster. Faster in terms of introducing more genetic correction […]. (Jay-BSR)

A prevailing view among the interviewees is that CRISPR-Cas9 has facilitated the implementation of GE which they point out might broaden possibilities of the application of the technology.

Efficiency, specificity and fidelity crucial for safety

Efficiency of GE is fundamental: "[…] to increase a lot the ability of the system to get into cells and not only to get into cells but to correct the sequence as we wanted" (Ruben-

BSR). Likewise, improving specificity of Cas9 and fidelity of DNA template integration are seen as important steps for future CRISPR technology development:

> So, there's the good where the foreign DNA is inserted at the target site, then there's the bad where the foreign DNA inserts itself somewhere else in the genome and then there's the ugly, the ones where the DNA is inserted on the target site, however insertion is not precise. (Irvin-BSR)

Interviewees stress that the type of application defines the required level of fidelity of CRISPR-Cas9: "As we are talking about the somatic cells, we need to control a lot of insertions; we cannot do this in germ cell lines for now" (Lisa-BSR). In the same way, participants mention that it is easier to deliver CRISPR-Cas9 to certain type of cells *in vitro* than *in vivo*:

> For example, defective genes in muscles and nervous system tissue can be corrected with amazing creativity and efficiency in cell culture using the new abilities of CRISPR. However, those corrections often cannot be delivered to tissues in an effective way that is meaningful for patient health. (Charles-BSR)

Broadly, the interviewed CRISPR experts highlight diverse technical challenges – with regards to efficiency of repairing genes, specificity of the CRISPR components used and fidelity of the integration of the new DNA material – that must be solved in view of safety of the process before its application in patients. In this way, technical aspects relate to risk acceptance, further developed in another sub-section.

Delivery and off-target effects influences safety

Several interviewees refer to anatomical characteristics making specific tissues particularly apt targets – "a possible entry point for *in vivo* GE […] will be in organ systems that are easily reachable and well and anatomically well-defined like the eye" (Irvin-BSR) –, suggesting that these organs will be the first on which somatic GE will be applied. For *in vivo* delivery of GE technology, viral vectors rather than nanoparticles are preferred, and the need to avoid a potential immune response from patients is emphasized. So, for participants that see delivery as a technical challenge, they consider that before moving to clinical *in vivo* applications, essential considerations on "how to deliver CRISPR?" should be included:

> […] basically new methods should be found and presently [we] have an efficient viral approach where you have to inject enormous titers of viral particles, that is still not very efficient […] it inflates the tissue with these second viral vectors copies. That's totally unexplored whether this will be for used for human therapy

> […] So, delivery for gene editing [in] somatic cells; that is one big thing. (Jay-BSR)

Participants also identify off-target effects (OTEs) as fundamental technical challenges that need to be solved before moving to clinical therapy. OTEs happen when a gene correction meant to happen in one gene ends up taking place in another gene affecting critical functions. For example, if the affected gene has impacts in cell death and cell proliferation, it may lead to complicated problems related with tumor formation and progression. Therefore, specificity of Cas9 nuclease cutting function and fidelity of the insertion of the DNA replacing material are identified as the main causes for OTEs. Irvin-BSR highlights the need to tackle: "Not only to the off-target activity at the nucleases themselves but to something that we want to act through, which is the importance of the specificity […] of which your foreign DNA is inserted into the genome and the fidelity". In general, participants note that a system to detect OTEs is essential: "so we're going to have to figure out a way to somehow assess off-targets or develop reagents that we're confident do not have off-target for the use in the clinic" (Megan-ASR). Not only GE for clinical applications but also germline GE needs OTEs assessment as pointed out by Benjamin (BSR): "[…] birth of an *in vitro* genome-edited embryo. I mean, it can be done but until the off-target and also many other scientific problems associated with this [are solved] it [is] still really [a] far away approach to do this.". Simon (ASR) alludes to He Jiankui's case of the genome-edited babies raising the question if the researchers had even been successful in knocking-out the intended gene (CCR5) to emphasize OTEs relevance for safety: "[…] we don't know the effects of injecting CRISPR in human people or sperm or oocyte. And I think we even don't know for example, that the Chinese [researcher] performed the knockout of the CCR5 [...]. And… could it have [had] another unexpected effect?". Another important observation is the need to assess effects in a wider context. This includes over time and across organs:

> […]if you [do] knock-in or knock-out [in] cell lines maybe the only phenotype you see is only a small part you can get from cells but you do not know what happens for long times growing for this kind of cells and organs. (Dean-BSR)

Delivery to the right type of cells and the right gene, the latter hampered by the possibility of OTEs, are thus additional technical challenges to be solved before the application of the technology in view of safety.

Greater understanding of the genome

According to participants, next-generation sequencing can be used to to detect OTEs, but they also stress the limitation of current methods focusing only on the most likely predicted off-target sites:

> "Currently, what is done is actually [that] people are [using] the predictive path. It's made by sequencing, however, nobody is […] interested about the possible sites where it can be interacting or about additional controls […] you cannot predict all the changes" (Liam-BSR).

The need for greater understanding of the genome, and the function and interconnectedness of genes, is also highlighted:

> Do we have enough knowledge about the genome to let's say: 'OK, for complex diseases we need to edit these in these genes' […] We need a whole research infrastructure […] on finding causal variants but people have been looking for [that] almost 30 years already. [And] still, we only have a couple handful of genes that we know. (Owen-ASR)

In general, participants also mention that greater understanding of genome complexity and genetic diversity evolves due to research with non-human species: "You can address questions in a mouse, but I have to say the cliché, a mouse is not a man. Therefore, if you can do in complementary species experiments, complement those results all together or if you can focus on other species, I think the insights will be better." (Irvin-BSR).

Risk acceptance

Again, the relevance of technical challenges in terms of safety and the level of risk that may be accepted varies with applications. About this, Vince-BSR, says: "[…] if you define *in vitro* as something that cannot survive outside the lab, well perhaps is relatively safe and not so much of an ethical concern. But if it's real organisms that can survive in the environment…then it might be different". Nevertheless, controlling for every factor when using GE technologies is considered utopic despite the absolute need of minimizing side effects and Steve (BSR) completes: "[…] of course you want the translation of the technology to really [be in] clinical use for human traits […] but these have to be really controlled and be in certain place like only in research use, in the laboratory use." Reflections over interventions in germline cells also express worries about genetic enhancement of future humans as noted by another interviewee:

> We cannot foresee what happens in the future, the future generations. We should never do stuff where we don't know what will happen. Once we delve [into] all the

technology for germline modification that can be used to make artificial enhancement elements, that could create the nightmare scenario of designer babies. (Phillip-BSR)

The importance of considering the type of application in dealing with technical challenges, seems a common concern for these experts, independently of their field of research. This concern around technical aspects and the emphasis on their impacts on the safety of applications provides robust support to the concerns already raised by STS scholars and which motivated the call for a global observatory to follow the main outcomes of research into GE of organisms (Jasanoff and Hurlbut 2018). Questions about how to take technological challenges into account at the regulatory level and how this may limit the acceptable applications of GE were also addressed by the interviewees, as explored in the following section.

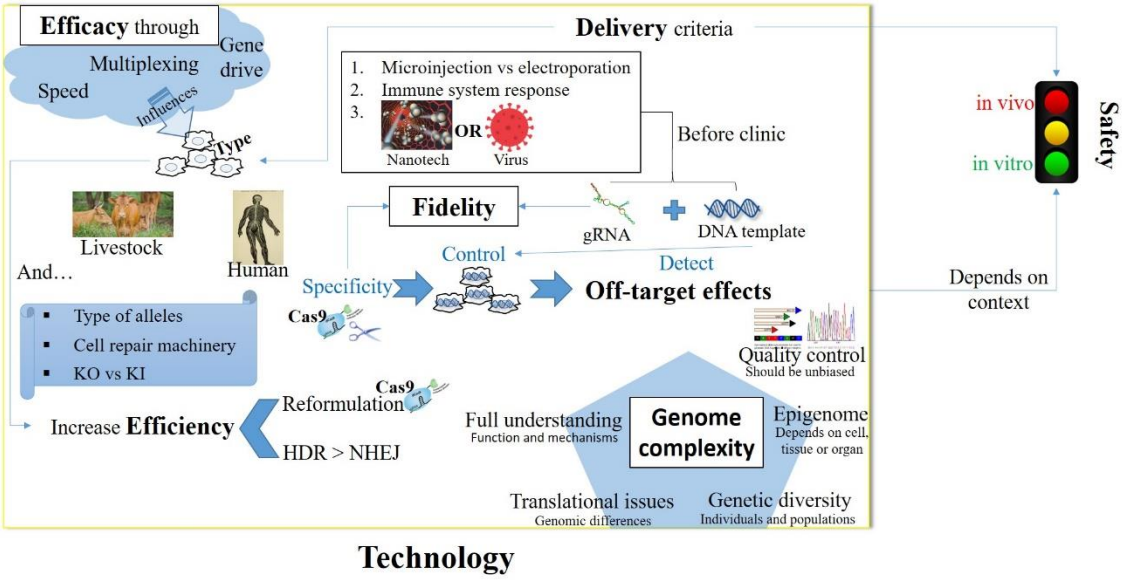


Figure.1. Technology as an overarching theme comprising multiple challenges that may influence safety. The different issues around technology brought up by the interviewees, and how these issues relate to each other are illustrated. Delivery of genome editing components such as CRISPR-Cas9 depends on different factors and different type of cells may be influenced by the efficacy of gene correction. Together, these influence efficiency of technology which needs to be improved by different basic research methods. Fidelity and control of DNA target integration and specificity of Cas9 cutting influence the appearance of off-target effects which need to be detected and monitored. Genome complexity in combination with these technical issues determine if the technology is to be considered safe enough for application *in vivo*. The arrows indicate relationships between technical aspects

Governance: ***Purpose of GE interventions is important for regulation of the technology and criteria must be defined in order to implement it***

When discussing different applications, it is clear that for the interviewees, acceptability is directly related to how purposeful an application is perceived to be. They also stress the importance that criteria are established for when GE can be applied. Figure 2 offers an overview of the different topics explored by researchers at this stage. Participants suggested different questions that should be answered in the first place.

> I think that the big question would be like: When is it OK to modify a genome and when is [it] not? You have a child that has a particular mutation that you know that increases the chances of having cancer. But that doesn't mean that will have cancer. Because we have other environment factors. So…when is it OK to say 'OK, so this is a situation where I can change the genome of the embryo and this is not the case'. So to draw that line – I think it's gonna be particularly hard to draw that line. […] not just disease susceptibility, it is also diseases, different types of diseases. So for example is it OK to change everyone's CCR5 to avoid HIV infection? Is it OK to change other types of receptors? (Jay-BSR)

The excerpt illustrates the criteria that the life sciences experts envision to delimit acceptable purposes - therapy, prevention of diseases and reduction of disease susceptibility – as a first step towards governance of GE.

Access to genome editing

In addition to the question of eligibility, some interviewees also raise the question of accessibility to GE with regards to social justice. An interviewee illustrates that GGE "[…] might be creating inadvertently a new class of human beings. The ones that are genetically modified and the ones that are not. And one cannot at this stage foresee if this will introduce a societal aspect" (Irvin-BSR). He continues by saying that editing of humans may result in prejudice and asks: "Are there people that will be genetically modified? Is there a status that people will acknowledge? Is something that is supposed to be private? […] Will there be any discrimination in favor of people that underwent germline genetic modification?" The interviewee finishes by saying that access might be dictated by economic constraints, which may result in medical tourism:

> And related to that, will some wealthy people have access to these technologies almost by default? Just because they have money and they can travel from the countries where these technologies are not available to a country where these technologies are available. (Irvin-BSR)

The general feeling expressed here is that accessibility to the technology is key and that avoiding/preventing different kinds of discrimination of citizens or group of citizens will be crucial.

Purpose affects acceptability

Editing of germline cells is generally not seen as acceptable by researchers unless as Vince (BSR) points out "[…] if one can prevent a life-threatening disease, perhaps I could give it a thought. But if it's just to increase the person's skills or something like that, then I'm against it by all means". When doing the ranking exercise (figures S1 and S2), researchers expressed preferences for applications that would help many people:

> So I'm gonna again put the food things and food security and getting rid of human pathogens that affect the most people up at the top. I really think that's moral imperative is to do [as] most good [as] possible. I guess cancer and genetic diseases would be next because lots of people get cancer and have genetic diseases. (Bruce-ASR)

In general, interviewees also consider some applications like GE horses irrelevant (figures S1 and S2). Moreover, an application that initially is reasonable may also represent a slippery slope:

> Once you start it never ends so even if you're trying to correct the genetic disease that's fine, but the next year, someone would say 'but my child is not intelligent enough. This is a disease'. Just a normal intelligence. 'We know how to improve it, please do this'. So then there's the enhancement. (Jay-BSR)

The existence of alternatives is another criterion that researchers consider: "In the midst of existing highly effective prenatal genetic diagnosis methods, there is very little (near zero) legitimate need for CRISPR germline correction techniques to be used [for gene editing human embryos]" (Charles-BSR). The interviewees see a gap between technology potential and regulation with implications for misuse of the technology as suggested by Michael (BSR): "I think if there would be a way under to control that it's not misused you know, abused, um… for eugenic reasons. Then I think it could be OK. But who is controlling that and who gets who gets control and who draws the line so… I think that's an issue".

The purpose of the application of GE is viewed as major aspect for the acceptance of the technology in a way that also provide the foundation for the regulation of the technology.

Criteria to harmonize regulation

The diverse regulatory landscape of GE may open the way for the genetic tourism issue:

> […] let's say that that editing is allowed in North America or in China or South America or whatsoever. And that we would be able to buy genetic material from those parts whereas gene editing in Europe is still not allowed. I mean, [this] could happen, that is mismatch […] in legislations related to editing. (Owen-ASR)

To tackle this, researchers evoke the need to apply regulatory criteria by defining: "[…] for which application would it be allowed and for which applications not? […] like one of the conditions could be: Is there an alternative or not?" (Owen-ASR).

Researchers acknowledge that stakeholder interaction helps in establishing such criteria when drawing guidelines for the use of the technology as noted by another interviewee:

> […] I don't say that we should use or take advantage of the extreme situation of a given technology, but we should know the limitations and then the society such [as] politicians and ethical people should draw the limits of where we should use the technology. (Frank-BSR)

The importance of a regulatory harmonization by involving different stakeholders in the process is thus a common view highlighted by interviewees. RRI means collaborative work between the different societal actors in science and innovation. For the participants in this study, a desirable harmonization of regulation will be constructed together with stakeholders. The call for a more inclusive public engagement and involvement of stakeholders in science policy regarding GE (Iltis, Hoover and Matthews 2021) is backed here by researchers in the life sciences. The perceptions and attitudes of the general public will influence regulatory actions, which has consequences for international harmonization, as further discussed in the next section.

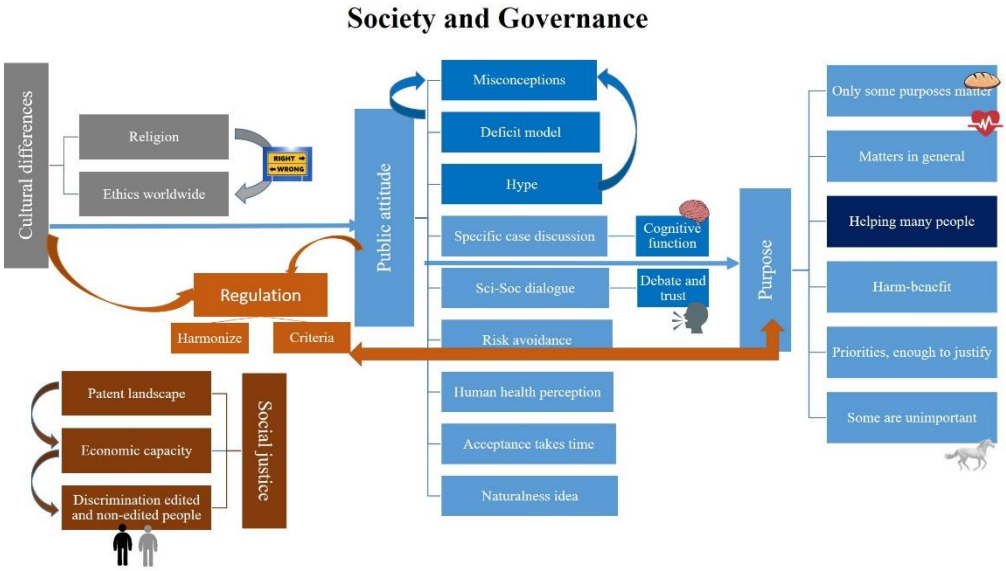


Figure 2. Governance and society are overarching themes comprising six subthemes with ethical and societal dimensions. Helping many people is the most important purpose identified over a set of applications that range from purposeful to irrelevant. Cultural differences involving ethics and religion may shape public attitudes towards the technology and together these may influence regulation worldwide which matters for governance. Public attitude concerns citizens' perception and acceptance of the technology which involve mainly public education, misconceptions and science-society dialogue features. This brings issues of social justice to the table particularly related with discrimination of individuals based on socio-economic status. The arrows indicate how issues influence each other and the colors of arrows and boxes indicate how subthemes are interrelated.

Society: ***Cultural differences influence public perception, and scientists have a responsibility to engage in dialogue***

Society as an overarching theme is presented in figure 2. When the interviewees reflect on GE technology in society, two main topics emerge: cultural differences and public attitude. Cultural differences between countries will affect public attitude to gene editing and indirectly also regulation worldwide. Some researchers point out that different ethics pattern worldwide influence public receptivity:

> […] we have still this very western centric view of the world. […] there was a very nice interview on TV […] it was about germline editing in humans. And someone asked the Chinese scientist, or talked about the European conservative view there and the guy was just laughing. (Michael-BSR)

Likewise, religion defines some of researchers' perspectives about their ethics towards implementation of GE technologies:

> […] one thing that is for me personally is a bit of an issue… I mean, I'm a Christian and so, how should I compare with this gene editing? Is this a bit like playing God? But then to this extent are we doing it anyway already with our less classical ways of breeding? […] I guess religious aspect is also important to somehow cover as well in the ethical debate […] even in cultures where there are a lot of Christian principles in it that has formed over all the centuries […] People think about it, even if they are not religious. (Owen-ASR)

Ethics and public perception play into in the distinction of concepts and applications:

> There's a huge difference between germline and somatic because there's ethical concerns with germline editing in humans. Who has the right to make their child […] what traits are they allowed to change, right? Because you're getting into eugenics. And as a society in general, I think we shivered to think about that because it's associated with nationalism and racism and bigotry. It's been represented in a lot of science fiction movies as what the future will be. So when that's ethically allowed and what traits are being changed, … maybe genetic diseases will be allowed at first, but … starting to alter humans for other reasons. We don't practice genetic improvement on our own species, typically. So that's why I see it as very distinct from germline editing in animals. (Bruce-ASR)

There may be a sequence to the acceptability of gene editing: "[…] it will be the first use in vegetables people will start to think that is not that dangerous and actually [it will be] helping them. And then [they] will probably accept better the use of CRISPR-Cas9 to therapy, drug and probably this will be done in the 5, 10 years' time" (Paul-BSR).

The influence of culture shapes the attitude of citizens as perceived by the CRISPR experts. Moreover, when ethical questions are at stake, interviewees highlight how the public discussion of GE is affected by associations with more general problems.

Fostering dialogue between scientists and society

The need for science-society dialogue is acknowledged: "[…] what is really needed is a societal debate about gene editing involving society and what you can do with the technology and then this is so I think that we can make an informed decision whether they would like or not" (Owen-ASR). Because implementation is at different stages, this debate will be different for human versus animal gene editing:

> I think for humans; the debate should be "if". […] I think [it] is really complicated, especially with humans. In animals I mean, I think that it's too late for that debate on if it should be used because it is being used (Jane-ASR)

Such a discussion in the public sphere is acknowledged by some researchers as being dependent "on who's starting this discussion, is it the large industry or is it a this is the scientist? Or is it a govern? […] the public have different perceptions or trust in different types of organizations" (Vince-BSR). It also seems to be focused on specific cases of GE applications and leaving others out. Quoting another interviewee: "[…] I don't see so many people discussing gene drive and gene drive is much closer to application than […] the birth of an *in vitro* human embryo." (Selma-BSR). Some interviewees call for a cautious and honest approach when discussing this with non-expert citizens:

> […] some recognition that we may not have all the answers and, an honest evaluation of the risks […] being open about those risks. […] I think there are going to be lots of shades of grey and so that's why I think we need to have some humility in terms of our own abilities and our own understanding of biology in general. […] Share both the facts and the uncertainties with the broader public. (Megan-ASR)

Educating the public to tackle misconceptions, hype and fear of GE is essential in researchers' general opinion:

> The more important thing is that you educate people, because the [official issue] must not only come from experts because obviously experts tend to have "bigger limits", so [probably] experts take the technology to an extreme [somehow]. We should educate people to know about it and to understand [and to] also [have] judgment about it. Judgment based on knowledge, not judgment based on belief. So I think it's important that you can educate people because at the end not all the laws are made by the [scientists]. (Paul-BSR)

Overall, scientists highlight the need to equip people with the necessary scientific understanding through dialogue in order for them to be able to participate in decision-making processes upfront. Language shapes this dialogue, as has been shown recently (Hartley et al 2023). How technologies, governance and society are intertwined within responsible science and innovation and how anticipation is a key aspect of technology assessment are the main lines of how the interviewed experts discuss GE as an emergent technology.

Discussion

Our analysis was structured into research questions related to the technology itself (challenges and expected realistic developments of genome editing technology) and its application (what applications are likely, what are desirable and what are concerning).

Safety of applications is transversal to all the research questions and the aggregating theme identified in this study. OTEs are the most worrying technical challenge for safety and interviewees consider crucial to develop methods to detect OTEs before clinical application. Literature highlights the need to avoid OTEs due to the critical consequences it may have for genome integrity and regulation (Cathomen et al. 2018). The difficulty of setting a "safe enough" standard has been a challenge since CRISPR discovery (EASAC 2017; NASEM 2017). Interviewees also consider that lack of genome knowledge increases the risk for undesired effects in human cells and that inefficiency in delivering CRISPR-Cas components may prevent a desired systemic effect of gene correction. These researchers believe that technology development will solve the issues of efficiency of CRISPR-Cas correction and fidelity of target DNA. Their belief is likely shaped by recent developments in optimization of genome editing repair pathways and the appearance of revolutionary editing strategies (Anzalone et al. 2019). Our study is concordant with a recent survey to biomedical researchers, which identified off-target mutations, efficiency levels for therapy and difficulties in targeting specific tissues *in vivo* as likely obstacles for the use of GE technologies in humans (Rocha, Braga, and Mota 2020).

Food derived from GE crops*, in vitro* GE research and GE for human therapy emerge in the interviews as desirable applications with no cause for concern. As regards human therapy, our interviewees' view that this will be hindered by public acceptance is in contrast with the evidence of high acceptance of GE for therapy purposes in some public surveys (75-90%) (Delhove et al. 2020) and citizens' vision of human GE therapy as highly likely in the next 20 years (So, Sladek, and Joly 2021). Such divergences between experts and non-experts might relate to the culture of risk perception, where scientists base their views on the scientific method whereas the general public has a more intuitive understanding that transcends science and includes societal, political, regulatory or ethical factors (Gaskell et al. 2007).

Some interviewees are worried over unequal access to GE therapies, including concern that this may lead to edited and non-edited classes of individuals. This social justice argument has been further developed by scholars who see that differentiated access to GE therapies may lead to aggravation of discrepancies and marginalization of vulnerable

citizens (Halpern et al. 2019; Hildebrandt and Marron 2018) as well as to irresponsible medical tourism only affordable to high-income citizens travelling to places where GE regulation is more relaxed (Meagher et al. 2020; Rosemann et al. 2019). Our interviewees generally consider GGE undesirable and worrying, but they also think it is unlikely and even less realistic for enhancement purposes. The interviewees emphasize that although technically easier to perform than somatic, germline editing is unlikely to satisfy safety considerations regarding physical, psychological and societal consequences for future generations. Again this is concordant with the literature where scholars explore reasons against GGE based on safety, responsibility, accountability and potential threats posed to future generations (de Wert et al. 2018; Howard et al. 2018). Nevertheless, interviewees generally acknowledge that a certain level of risk must be accepted for GE either in somatic and germline, and that it can be estimated based on preclinical animal testing which also has been suggested in the literature (Matthews and Iltis 2019; Polcz and Lewis 2016). Interviewees also stress that GGE should be avoided where alternatives such as pre-implantation diagnosis exist, considering it only acceptable for cases of life-threatening diseases and where these alternatives are not enough. Autosomal dominant and polygenic diseases have been identified as cases where there is no alternative to GGE (Gyngell, Douglas, and Savulescu 2017). Once again, scientists and general public perception seems to be discrepant since GGE for medical reasons earns considerably high public support for prevention of diseases of multiple levels of severity (Jedwab et al. 2020).

Overall, the interviewees welcome GE regulation and highlight the need to establish criteria and define crucial concepts like disease, susceptibility, enhancement and prevention. International health agencies defend that defining disease is important when regulating therapy and disease prevention (WHO 2021). A recurrent issue in the interviews is the lack of harmonization in how GE is regulated. It is known that GE regulation and legislation vary worldwide (Baylis et al. 2020) with a risk of safety and responsibility being dismissed in countries with less oversight (Boggio et al. 2019). Public opinion will influence regulation, and participants of the study emphasize the need to inform and educate the public to avoid misinformation and allow a rational discussion of GE applications.

Creative and effective forms of engagement between scientific experts, decision-makers and the public are necessary and may create an opportunity for newer participatory mechanisms of citizens with real repercussions for their own lives under democratic governance of science (Jasanoff, 2003). Interviewees acknowledge that ethics, religion

and regulation differ between countries and these cultural and normative differences affect attitude towards gene editing but their view is still distant from a culture of "civic science" (Wirz, Scheufele and Brossard 2020) based on an early engagement of the public. However, when interviewees speak about the need for education, this is closer to a dialogue model that also accounts for cultural and experimental knowledge, because focus is on making citizens knowledgeable to participate in decision-making, contrasting with a deficit model where scientific expertise is the dominant knowledge (Reincke, Bredenoord and van Mil 2020). Whereas the view that further development of the technology will overcome many of the present problems predominates, some interviewees also highlight the importance of recognizing our limited knowledge, in a way that comes closer to the "technologies of humility" concept (Jasanoff 2003). Contrary to deference to scientific authority, trusting science and its actors is a precursor of a democratic view (Howell et al. 2020) and this is a crucial factor influencing society questions about safety (Braun and Meacham 2019). The view that a broad stakeholder engagement with an informed public will be more useful reflects a culture of global public engagement for decision-making about GE (Jasanoff and Hurlbut 2018). The anticipation of scenarios in a collective manner builds on inclusive deliberation embedded in democratic principles and supposes a process of discussion and negotiation between actors (Owen, Macnaghten and Stilgoe, 2012) and participants in this study recognize that this is crucial for the emergent character of GE technology.

Limitations and strengths of the study

Our group of participants displays a mostly European, biomedical and/or animal science profile. This is a consequence of the non-random sampling method used and we recognize that other profiles of researchers could be sought for these interviews (e.g. medical or clinical background). Nonetheless, we achieved enough sample breadth and depth.

Finally, it is worth stressing that half of the interviews took place right after the He Jiankui's case. This might have influenced researchers' view and therefore we note a possible pre- and post- interpretation of the "birth of an *in vitro* genome-edited human embryo" ranking case (Supplementary material).

Conclusions and future perspectives

In summary, our study shows that for researchers in the field, GE technology is viewed as promising but with many unsolved challenges, in need of further research and of internationally harmonized regulation. They call for technology improvement and increased knowledge about genome function. They clearly do not support what they

perceive as unsafe, unpredictable and irrelevant applications, but there are some discrepancies with the view of non-scientist citizens. Whereas public misconceptions are seen as a problem by the interviewees, there is also recognition that fostering a clear science-society dialogue is needed, and that science does not have all the answers.

This study with scientists that work daily with the technology provides insight into the science-based priorities for GE to be a technology that can be responsibly applied in humans. Contextualizing this study within RRI framework, we note that a slice of the research community is represented and additional stakeholders are needed to confer inclusivity in order to better anticipate unintended consequences of resulting innovations. We are complementing the present study by interviewing ethicists and policy-makers, with focus on governance aspects of this technology.

Acknowledgments

We would like to thank all participants of the study for their essential contributions to this work and to all researchers that contributed with inputs for the interview guide during pilot tests. Finally, we would like to thank the Data Protection Officer from [redacted for blinding] for the contributions to the redaction of the final informed consent form used.

References

Adli, M. 2018. "The CRISPR Tool Kit for Genome Editing and Beyond." *Nature Communications* 9 (1): 1911. https://doi.org/10.1038/s41467-018-04252-2.

Akcakaya, P., M. L. Bobbin, J. A. Guo, J. Malagon-Lopez, K. Clement, S. P. Garcia, M. D. Fellows*, et al.* 2018. "In Vivo CRISPR Editing with No Detectable Genome-Wide Off-Target Mutations." *Nature* 561 (7723): 416-19. https://doi.org/10.1038/s41586-018-0500-9.

Anzalone, A. V., P. B. Randolph, J. R. Davis, A. A. Sousa, L. W. Koblan, J. M. Levy, P. J. Chen*, et al.* 2019. "Search-and-Replace Genome Editing without Double-Strand Breaks or Donor DNA." *Nature* 576 (7785): 149-57. https://doi.org/10.1038/s41586-019-1711-4.

Barrangou, R., and P. Horvath. 2017. "A Decade of Discovery: CRISPR Functions and Applications." *Nature Microbiology* 2: 17092. https://doi.org/10.1038/nmicrobiol.2017.92.

Baylis, F., M. Darnovsky, K. Hasson, and T. M. Krahn. 2020. "Human Germ Line and Heritable Genome Editing: The Global Policy Landscape." *CRISPR Journal* 3 (5): 365-77. https://doi.org/10.1089/crispr.2020.0082.

Boggio, A., B. M. Knoppers, J. Almqvist, and C. P. R. Romano. 2019. "The Human Right to Science and the Regulation of Human Germline Engineering." *CRISPR Journal* 2: 134-42. https://doi.org/10.1089/crispr.2018.0053.

Braun, M., and D. Meacham. 2019. "The Trust Game: CRISPR for Human Germline Editing Unsettles Scientists and Society." *EMBO Reports* 20 (2). https://doi.org/10.15252/embr.201847583.

Borup, M., N. Brown, K. Konrad, and H. van Lente. 2006. "The sociology of expectation in Science and Technology". *Technology Analysis & Strategic Management* 18 (3,4): 285-298

Brokowski, C., and M. Adli. 2018. "CRISPR Ethics: Moral Considerations for Applications of a Powerful Tool." *Journal of Molecular Biology* 431 (1): 88-101. https://doi.org/10.1016/j.jmb.2018.05.044.

Bucchi, M. 2004. Science in Society: an introduction to social studies of science. Translated by Adrian Belton. Abingdon: Routledge

Camporesi S., and G. Cavaliere. 2016. "Emerging ethical perspectives in the clustered regularly interspaced short palindromic repeats genome-editing debate". *Personalized Medicine* 13 (6): 575-586. https://doi.org/ doi: 10.2217/pme-2016-0047

Cathomen, T., S. Schule, M. Schussler-Lenz, and M. Abou-El-Enein. 2018. "The Human Genome Editing Race: Loosening Regulatory Standards for Commercial Advantage?". *Trends in Biotechnology* 37 (2): 120-23. https://doi.org/10.1016/j.tibtech.2018.06.005.

de Lecuona I., M. Casado, G. Marfany, M. Lopez Baroni, and M. Escarrabill. 2017. "Gene Editing in Humans: Towards a Global and Inclusive Debate for Responsible Research". *Yale Journal of Biology and Medicine* 90 (4): 673-681 PMID: 29259532; PMCID: PMC5733855.

de Wert, G., B. Heindryckx, G. Pennings, A. Clarke, U. Eichenlaub-Ritter, C. G. van El, F. Forzano*, et al.* 2018. "Responsible Innovation in Human Germline Gene Editing. Background Document to the Recommendations of ESHG and ESHRE." *Human Reproduction Open* 1: 1-18. https://doi.org/10.1093/hropen/hox024.

Delhove, J., I. Osenk, I. Prichard, and M. Donnelley. 2020. "Public Acceptability of Gene Therapy and Gene Editing for Human Use: A Systematic Review." *Human Gene Therapy* 31 (1-2): 20-46. https://doi.org/10.1089/hum.2019.197.

European Academies Science Advisory Council. 2017. Genome editing: scientific opportunities, public interests and policy options in the European Union. EASAC policy report 31

Felt, U. 2018. "Responsible Research and Innovation". In *the Handbook of Genomics, Health and Society*, edited by Sahra Gibbon; Barbara Prainsack; Stephen Hilgartner; Janelle Lamoreaux, 108-116. Abingdon: Routledge

Gaskell, G., N. Kronberger, C. Fischler, J. Hampel, and J. Lassen. 2007. "Consumer perceptions of food products from cloned animals: A social scientific perspective". *Prepared for the European Food Safety Authority*

Gyngell, C., T. Douglas, and J. Savulescu. 2017. "The Ethics of Germline Gene Editing." *Jounal of Applied Philosophy* 34 (4): 498-513. https://doi.org/10.1111/japp.12249.

Halpern, J., S. E. O'Hara, K. W. Doxzen, L. B. Witkowsky, and A. L. Owen. 2019. "Societal and Ethical Impacts of Germline Genome Editing: How Can We Secure Human Rights?". *CRISPR Journal* 2 (5): 293-98. https://doi.org/10.1089/crispr.2019.0042.

Hartley S., A. Stelmach, J. A. Delborne, and S. K. Barnhill-Dilling 2023. "Moving beyond narrow definitions of gene drive: Diverse perspectives and frames enable substantive dialogue among science and humanities teachers in the United States and United Kingdom." *Public Understanding of Science*, 0(0). https://doi.org/10.1177/09636625221148697.

Hildebrandt C., and J. Marron. 2018. "Justice in CRISPR-Cas9 research and clinical applications". *AMA Journal of Ethics* 20 (9): E826-833. https://doi.org: 10.1001/amajethics.2018.826.

Howard, H. C., C. G. van El, F. Forzano, D. Radojkovic, E. Rial-Sebbag, G. de Wert, P. Borry, *et al.* 2018. "One Small Edit for Humans, One Giant Edit for Humankind? Points and Questions to Consider for a Responsible Way Forward for Gene Editing in Humans." *European Journal of Human Genetics* 26 (1): 1-11. https://doi.org/10.1038/s41431-017-0024-z.

Howell, E. L., C. D. Wirz, D. A. Scheufele, D. Brossard, and M. A. Xenos. 2020. "Deference and Decision-Making in Science and Society: How Deference to

Scientific Authority Goes Beyond Confidence in Science and Scientists to Become Authoritarianism." *Public Understanding of Science* 29 (8): 800-18. https://doi.org/10.1177/0963662520962741.

Hurlbut, J. B., K. Saha, and S. Jasanoff. 2015. "CRISPR democracy: Gene editing and the need for inclusive deliberation". *Issues in Science and Technology* 32 (1): 1-14

Hurlbut, B. 2018. "In CRISPR's world: genome editing and the politics of global science". In *the Handbook of Genomics, Health and Society*, edited by Sahra Gibbon; Barbara Prainsack; Stephen Hilgartner; Janelle Lamoreaux, 121-178. Abingdon: Routledge

Iltis, A. S., S. Hoover, and K. R. W. Matthews. 2021. "Public and Stakeholder Engagement in Developing Human Heritable Genome Editing Policies: What Does it Mean and What Should it Mean?" *Frontiers in Political Science* 3. https://doi.org/10.3389/fpos.2021.730869.

Isasi R., E. Kleiderman, and B. M. Knoppers. 2016. "Editing policy to fit the genome?" *Science* 361(6271): 337-339. https://doi.org/10.1126/science.aad6778.

Jasanoff, S. 2003. "Technologies of Humility: Citizen Participation in Governing Science." *Minerva* 41: 223-244. https://doi.org/10.1023/A:1025557512320

Jasanoff, S., and J. B. Hurlbut. 2018. "A global observatory for gene editing". *Nature* 555: 435-437. https://doi.org/10.1038/d41586-018-03270-w

Jasanoff, S., and S. Kim. 2015. *Dreamscapes of modernity: sociotechnical imaginaries and the fabrication of power*. Chicago and London: The University of Chicago Press

Jedwab, A., D. F. Vears, C. Tse, and C. Gyngell. 2020. "Genetics Experience Impacts Attitudes Towards Germline Gene Editing: A Survey of over 1500 Members of the Public." *Journal of Human Genetics* 65 (12): 1055-65. https://doi.org/10.1038/s10038-020-0810-2.

Konrad, K., H. van Lente, C. Groves, C. Selin. 2016. "Performing and Governing the Future in Science and Technology". In *The Handbook of Science and Technology Studies*, edited by Ulrike Felt; Rayvon Fouche; Clark A. Miller; Laurel Smith-Doerr, 465-493. Cambridge: MIT Press

Matthews, K. R. W., and A. S. Iltis. 2019. "Are We Ready to Genetically Modify a Human Embryo? Or Is It Too Late to Ask?". *Accountability in Research* 26 (4): 265-70. https://doi.org/10.1080/08989621.2019.1617139.

Meagher, K. M., M. A. Allyse, Z. Master, and R. R. Sharp. 2020. "Reexamining the Ethics of Human Germline Editing in the Wake of Scandal." *Mayo Clinical Procedures* 95 (2): 330-38. https://doi.org/10.1016/j.mayocp.2019.11.018.

Meyer, M. 2022. "Taking responsibility, making irresponsibility: Controversies in human gene editing." *Social Studies of Science* 52 (1): 127-143. https://doi.org/10.1177/0306312721102563

National Academies of Sciences Engineering and Medicine (U.S.). 2017. Human Genome Editing: Science, Ethics, and Governance. *The National Academies Press*, Washington, DC

Nordberg, A., T. Minssen, S. Holm, M. Horst, K. Mortensen, and B. L. Moller. 2018. "Cutting Edges and Weaving Threads in the Gene Editing (Я)Evolution: Reconciling Scientific Progress with Legal, Ethical, and Social Concerns." *Journal of Law Biosciences* 5 (1): 35-83. https://doi.org/10.1093/jlb/lsx043.

Owen, R., P. Macnaghten, and J. Stilgoe. 2012. "Responsible Research and Innovation: From Science in Society to Science for Society, with Society." *Science and Public Policy* 39 (6): 751-60. https://doi.org/10.1093/scipol/scs093.

Polcz, S., and A. Lewis. 2016. "CRISPR-Cas9 and the Non-Germline Non-Controversy." *Journal of Law and the Biosciences* 3 (2): 413-25. https://doi.org/10.1093/jlb/lsw016.

[redacted for blinding].

Regalado, A. 2018. "EXCLUSIVE: Chinese scientists are creating CRISPR babies." *MIT Techology Review*, November 25

Reincke, C. M., A. L. Bredenoord, and M. H. van Mil. 2020. "From Deficit to Dialogue in Science Communication: The Dialogue Communication Model Requires Additional Roles from Scientists." *EMBO Reports* 21 (9): e51278. https://doi.org/10.15252/embr.202051278.

Rip, A. 2018. *Futures of Science and Technology in Society*. Wiesbaden: Springer

Rocha, L. F. M., L. A. M. Braga, and F. B. Mota. 2020. "Gene Editing for Treatment and Prevention of Human Diseases: A Global Survey of Gene Editing-Related Researchers." *Human Gene Therapy* 31 (15-16): 852-62. https://doi.org/10.1089/hum.2020.136.

Rosemann, A., A. Balen, B. Nerlich, C. Hauskeller, M. Sleeboom-Faulkner, S. Hartley, X. Zhang, and N. Lee. 2019. "Heritable Genome Editing in a Global Context: National

and International Policy Challenges." *Hastings Center Report* 49 (3): 30-42. https://doi.org/10.1002/hast.1006.

So, D., R. Sladek, and Y. Joly. 2021. "Assessing Public Opinions on the Likelihood and Permissibility of Gene Editing through Construal Level Theory." *New Genetics and Society*: 1-25. https://doi.org/10.1080/14636778.2020.1868985.

von Schomberg, R. 2013. "A vision of Responsible Research and Innovation". In *Responsible Innovation: Managing the Responsible emergence of science and innovation in society*, edited by R. Owen, J. Bessant and M. Heintz, 51-74. London: Wiley

WHO Expert Advisory Committee on Developing Global Standards for Governance and Oversight of Human Genome Editing. 2021. Human Genome Editing: position paper. Geneva: World Health Organization

Wahlberg, A., D. Dong, P. Song, and Z. Jianfeng. 2021. "The Platforming of Human Embryo Editing: Prospecting 'Disease Free' Futures." *New Genetics and Society* 40 (4): 367-383. https://doi.org/10.1080/14636778.2021.1997578

Wirz, C. D., D. A. Scheufele, and D. Brossard. "Societal Debates About Emerging Genetic Technologies: Toward a Science of Public Engagement." *Environmental Communication* 14 (7): 859-64. https://doi.org/10.1080/17524032.2020.1811478.

Word count: 7986 (from abstract)

Chapter 5. MANUSCRIPT 2. What do people think about genetic engineering: A systematic review of questionnaire surveys before and after the introduction of CRISPR

Manuscript 2 (To be submitted)

What do people think about genetic engineering: A systematic review of questionnaire surveys before and after the introduction of CRISPR

Pedro Dias Ramos [1,2], Maria Strecht Almeida[2], Ingrid Anna Sofia Olsson [1,2]

[1]i3S – Instituto de Investigação e Inovação em Saúde, Universidade do Porto, Portugal

[2]ICBAS – Instituto de Ciências Biomédicas Abel Salazar, Universidade do Porto, Portugal

What do people think about genetic engineering: A systematic review of questionnaire surveys before and after the introduction of CRISPR

Pedro Dias Ramos [1,2], Maria Strecht Almeida[2], Ingrid Anna Sofia Olsson [1,2]

[1]i3S – Instituto de Investigação e Inovação em Saúde, Universidade do Porto, Portugal

[2]ICBAS – Instituto de Ciências Biomédicas Abel Salazar, Universidade do Porto, Portugal

Abstract

The advent of CRISPR-Cas9 in 2012 has come to revolutionize the field of genetics by broadening the access to a method for precise modification of the human genome. It also brought renewed attention to the ethical issues of genetic modification and the societal acceptance of technology for this purpose. So far, many surveys assessing public attitudes towards genetic modification have been conducted worldwide. Here we present the results of a systematic review of primary publications of surveys addressing public attitudes towards genetic modification as well as awareness and knowledge about the technology. A total of 53 primary publications (1987-2020) focusing on applications in humans and non-human animals were identified covering countries in 4 continents. The 30 studies from until 2012 (pre-CRISPR) address gene therapy in humans and genetic modification of animals for food production and for biomedical research. The 23 studies from after 2013 (CRISPR) address gene editing in humans and animals. Across countries, respondents see gene therapy for disease treatment or prevention in humans as desirable and highly acceptable, whereas enhancement is generally met with opposition. When the study distinguishes between somatic and germline applications, somatic gene editing is generally accepted whereas germline applications are met with ambivalence. The purpose of the application is also important for attitude to genetically modified animals: modification in food production is much less accepted than for biomedical application in pre-CRISPR studies. A relationship between knowledge/awareness and attitude to genetic modification is often present. A critical appraisal of methodology quality in the primary publications as regards to sampling as well as questionnaire design, development, administration shows that there is considerable room for improvement in the reporting of methodological detail. Lack of information is more common in earlier studies, which probably reflects the changing practice in the field.

Introduction

The advent of CRISPR-Cas9 in 2012 came to revolutionize the field of genetics by democratizing the access to a method for precise modification of the mammalian genome [1,2]. Being straightforward and low-cost while precise and efficient underlies the wide uptake of CRISPR-Cas9 by research groups and industry [2,3,4]. This has resulted in an explosion of laboratories engaging in research using genetic modification of organisms, including applications in clinical practice, biomedical research, food production and for environmental purposes (3,4,5).The possibility to apply CRISPR-Cas9 to human embryos

has nonetheless raised concern among scientists as well as in society, and led to revisit previous regulation on human genetic manipulation, such as Article 13 of Oviedo Convention, The Universal Declaration on the Human Genome and Human Rights, and the EU Charter of Fundamental Rights [4]. The first years of CRISPR-Cas9 were marked by uncertainty and an international moratorium to human germline manipulation was adopted by a range of countries [3,5,6,7]. However, in 2018, the media announced the first case of human embryo manipulation following the birth of the first gene-edited babies and the expected arrival of a second gene-edited baby in the summer of 2019 [8,9]. This story initiated a frenzy of media articles, generally characterized by strong disapproval in the public sphere, conveying their concern that scientists were 'crossing the line' and almost unanimous rejection by members from the scientific community [4,10]. The discussion around CRISPR-Cas9 has also reignited concerns about gene-editing of animals, including those used for food, and their potential release into the environment and into the food supply [11].

CRISPR-Cas 9 comes across as a technology which is perceived as both promising and threatening, and as such is particularly interesting in the context of initiatives such as RRI (Responsible Research and Innovation) which aim to open up research to society [12]. The underlying objective is to align the research and development of new technologies with societal values and priorities.

Understanding public knowledge and awareness of a new technology is an important part of the process, as is the measurement of citizens' attitudes towards such development, for two main reasons. Firstly, in representative democracies, questionnaires are important sources of information about how citizens position themselves in specific questions. Secondly it is important to understand how much citizens are receptive to adopt the technology in their daily lives.

Opinion surveys measure the views of society within a given context in relation to a certain topic, often with a cross-sectional approach that measures opinion at a specific time-point and allows for comparison such as between countries or regions but not over time [13,14]. When used as research instruments, surveys of public opinion are designed to provide quantitative information that allows the researchers to answer underlying research questions by assessing the attitudes of inquired people [15]. A critical appraisal of study methodology is an important complement to a systematic review of study outcome. Although most common in reviews of studies like randomized clinical trials, critical appraisal is relevant for many types of studies either quantitative, qualitative, mixed-methods or surveys [20,21,22,23,24,25,26]. An important aspect of methodological quality is the survey

instrument, that is the questions and the accompanying measurement scales such as Likert and semantic differential scales, constructs that need to be evaluated in terms of validity and reliability before the survey is administered [15,16,17]. In systematic reviews of quantitative questionnaire studies, critical appraisal also includes the validity and how representative the sample is of the population under study, how the variables have been defined, whether potential biases are considered and other factors that may interfere with result interpretation [18].

The aim of the present systematic review is to map the existing body of evidence concerning public attitudes towards genetic modification, since the first survey on the topic was applied nearly 35 years ago. The review covers 53 primary publications covering countries in Asia, Europe, North and South America and Oceania, integrating public attitudes as well as awareness and knowledge about genetic modification. Our approach is comprehensive in that it includes cross-sectional surveys measuring public opinions on matters of biotechnology and genetic engineering when applied to humans as well as to other animals, and by introducing critical appraisal as a means to assess methodology quality surrounding questionnaire design, development and administration together with sampling of population and main limitations and successes drawn from studies in this type of analysis. This systematic review will complement existing narrative reviews and perspective papers on the topic, such as [27,28,29].

Methodology

Search

Web of Science (WOS) was selected as the primary database for scholarly publications, focusing the search to identify surveys done with citizens about three different themes: *Gene therapy*, *Genetically-modified animals (GM animals)* and *Genome editing*. The search was conducted between July and November 2019 and reviewed again in February 2020 and August 2022. This database search was complemented with Google search engine to look especially for grey literature that could not be found through WOS, namely governmental reports and other studies not published in academic journals. Although not peer-reviewed by academic scholars, their relevance for policy advising means this type of literature is worth considering [139,140]. For the WOS search, the themes *Gene therapy* and *GM animals* included only publications until 2012 since this was the year of the advent of CRISPR-Cas9 biotechnology which changed the terminology of scientific articles from “genetic modification” to “genome editing” specifically. Conversely and likewise, for the *Genome editing* theme, only studies from 2013 onwards were included. All WOS collections were investigated, namely: WOS Core Collection, Current Contents Connect, Derwent

Innovations Index, KCI-Korean Journal Database, MEDLINE®, Russian Science Citation Index, SciELO Citation Index. Pilot studies were done with different combination of keywords until the identification of the final Boolean strings to be used for the searching process (See supplementary material). For this, the numbers of publications retrieved from WOS for a specific combination of strings were analyzed and only the ones with the highest number were considered. For *GM animals*, the different combinations of strings yielded the highest number of all while for *Gene therapy* and *Genome editing* themes it was irrelevant to add more string terms since it would always yield equal or lower number of publications. These allowed us to conduct the search in a broadened way, finding the most publications possible for each theme and avoiding to discard unintended ones. As for the Google search, the terms used included the theme name adding "public" plus "attitude" terms and the search results were screened thoroughly until the titles of the links started to be redundant in the upcoming searching pages. After identification of websites conveying multiple surveys, these were also used as a source to search for additional grey literature.

Selection

The screening process is described in the PRISMA flowchart presented in figure 1. All WOS publications that featured surveys with the general public regarding genetic modification of humans or animals were included in the Endnote library. All publications only addressing genetic modification of plants or crops were excluded from the library and so were publications in the format of reviews and meeting or conference abstracts. All publications for which it was not possible to access its full-text or PDF document or not written in English were equally excluded. From the initial set of 2981 publications following duplicate removal and the exclusion criteria, 60 publications were left. After a careful reading of these, 33 publications reporting qualitative rather than quantitative studies and/or with low sample sizes (lower than 100 respondents) were excluded. To the WOS final list of 27 publications,

26 from grey literature not meeting the exclusion criteria were added equaling a total of 53 primary publications eligible for the systematic review.

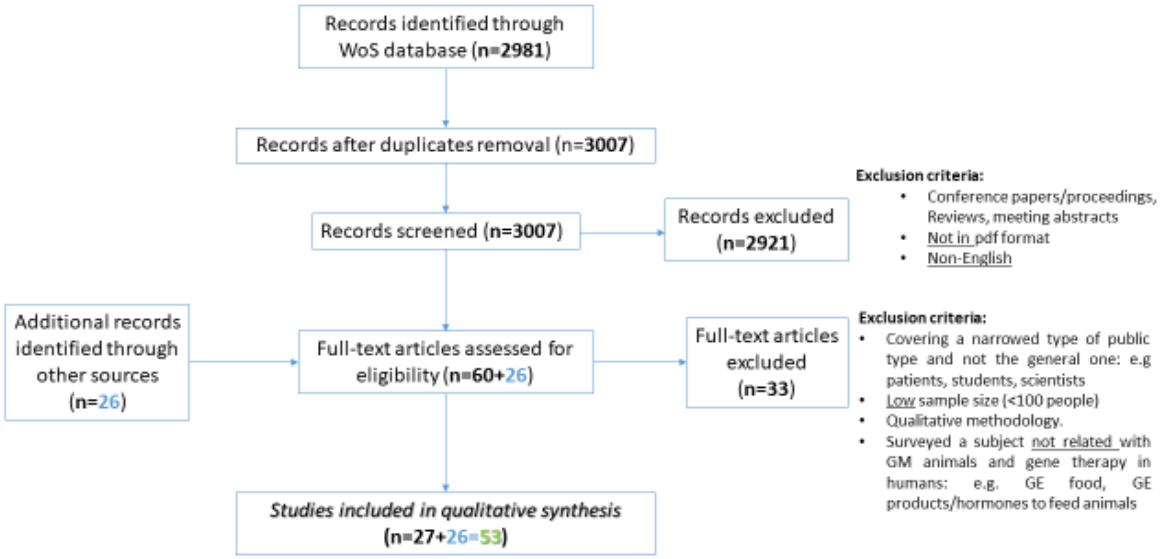


Figure 1. PRISMA Flowchart and exclusion criteria used for search and selection of primary publications in the systematic review. Records identified from WOS and additional sources (Google) were screened and refined using an exclusion criteria summarized in the text box following PRISMA guidelines [21]. In the end, a total of 53 primary publications (27 from WOS database and 26 from Google search) were included for the systematic review.

Survey parameters

The systematic review followed the PICOS guidelines (Population, Intervention, Comparator, Outcomes and Study designs) for evaluation of studies resulting from the initial search, except for the Intervention index since we were not doing any statistical or meta-analysis [30,31]. Population concerned the number of participants featuring in the surveys and the country where the surveys took place. Comparison concerned the differences and similarities of public attitudes towards genetic modification procedures between citizens of different countries, comparison between years as well as comparison of the type of questions and terminology used by surveyors. Outcomes analyzed were: percentage of agreement genetic modification in broad terms and for specific applications in humans and animals, the reasoning behind those attitudes and so as their level of knowledge and/or the level of familiarity with biotechnology and/or genetic engineering topics. For more details, please see supplementary tables S1, S2 and S3.

Critical appraisal of primary publications

All included primary publications were evaluated in regards to the methodological quality of the studies they reported. This was done by assessing if certain indicators were present or absent and by evaluating how well described and appropriate they were for the studies in question.

The critical appraisal addressed: *content of questionnaires* – whether authors generated their own items or adapted them from previous surveys; *validity* – cross-checking between authors and/or external advisers and tested with target population for both clarity and efficacy of measuring concepts; *reliability* – trustworthiness of the same items and constructs used within the surveys; *sampling* – representativeness and randomness; *risk of bias* – potential response, non-response and selection bias; and *ethical practices* – details on informed consent obtained, if there were incentives given to respondents and disclosure of any ethical statements by authors either related with ethical approval of studies or potential conflict of interests experienced.

The search, selection and first analysis were performed by the first author, obtaining feedback by the other two authors. The critical appraisal was performed by PDR and IASO while MSA did the co-authorship network analysis (see Supplementary material).

Results

Of the 53 primary publications identified in this review, the 30 conducted prior to the advent of CRISPR-Cas9 technology in 2012 represent the pre-CRISPR period whereas the 23 conducted from 2013 onwards represent the CRISPR period (table S1 and supplementary table S1). Pre-CRISPR studies were conducted between 1987 and 2010 and comprise 25 surveys with questions assessing attitudes towards genetic modification of animals (GM animals) and 14 assessing attitudes towards genetic modification of humans. In the CRISPR period, 8 survey studies addressed genetic modification of animals and 15 did the same for genetic modification of humans.

Generally speaking, the surveys conducted in the pre-CRISPR period focused on the opinion of the general public towards genetic modification of animals for use in medical applications, food products derived from them (meat and milk) and on the genetic modification of humans as gene therapy applications for the cure, prevention and reduction of risk of diseases. Some of these surveys also included additional aspects of human

genetic modification such as adults and children, prevention and therapy, and modification to change characteristics not related with diseases.

Table S1 (Appendix VI) summarizes the number of approvers of genome editing technology in both periods in a proportion of ten citizens considering the previously mentioned applications and the region where the surveys took place.

Genetically modified animals in pre-CRISPR and CRISPR periods

A) Pre-CRISPR: GM animals for food purposes are mostly rejected and medical applications are seen ambivalently worldwide

Overall, 25 of the 30 surveys from the pre-CRISPR period covered genetic modification of animals (GM animals). In a quick overview of table S1, we can see that transplants and medicines face a higher approval from respondents than food products derived from GM animals. For all cases of food derived from GM animals, either to obtain "leaner meat", "meat less fatty" or simply, meat from these animals, the approval rate is very low among respondents in almost all countries analyzed and this trend is consistent from 1987 to 2006 although there are some studies where approval for meat consumption of GM animals reaches more than half of the respondents (US in 1987, Japan in 1997, Thailand and India in 1997 and 2000 and Australia in 2000; Figure 2A). Approval of GM animals for transplant and medicines drops considerably between 1991 and 2010, in Europe. The lowest approval reaches 4 in every 10 European citizens in the 1996 and in 2002 and only 3 in every 10 citizens in 2005 Eurobarometer (fig. 2A). Conversely, medicines derived from GM cows gained approval among Europeans between 2002 and 2010 Eurobarometers (fig. 3A). Australians and New Zealanders are among the lowest approvers of GM animals worldwide for both medical and food purposes and their approval has been decreasing in surveys after 2000s (fig. 2A and 3A). A similar trend was followed by citizens from US who rejected meat derived from GM pigs in all surveys conducted after 2000 (fig. 2A). Japanese citizens are the most inquired publics in the pre-CRISPR period regarding attitudes towards GM animals and supported GM for food - meat and milk – slightly more than for medical purposes (organs for transplantation in pigs (fig. 2A) and mice for cancer research (fig. S1 – Appendix V), going against the general trend. A note of remark is their drop in approval for meat of GM pigs from 1997 to 2003 as well as for transplants and medicines (fig. 2A). GM mice for cancer research is seen as "to be encouraged" more than GM pigs for transplants among Japanese citizens (table S1 (pre-CRISPR) – See Appendix VI). In two studies of single European countries, in Germany less than half of citizens supported GM laboratory animals

for cancer research and Swedish citizens categorically rejected GM salmon for food consumption (fig. S1 – Appendix V) similar to what they did with GM pigs (fig. 2A) .

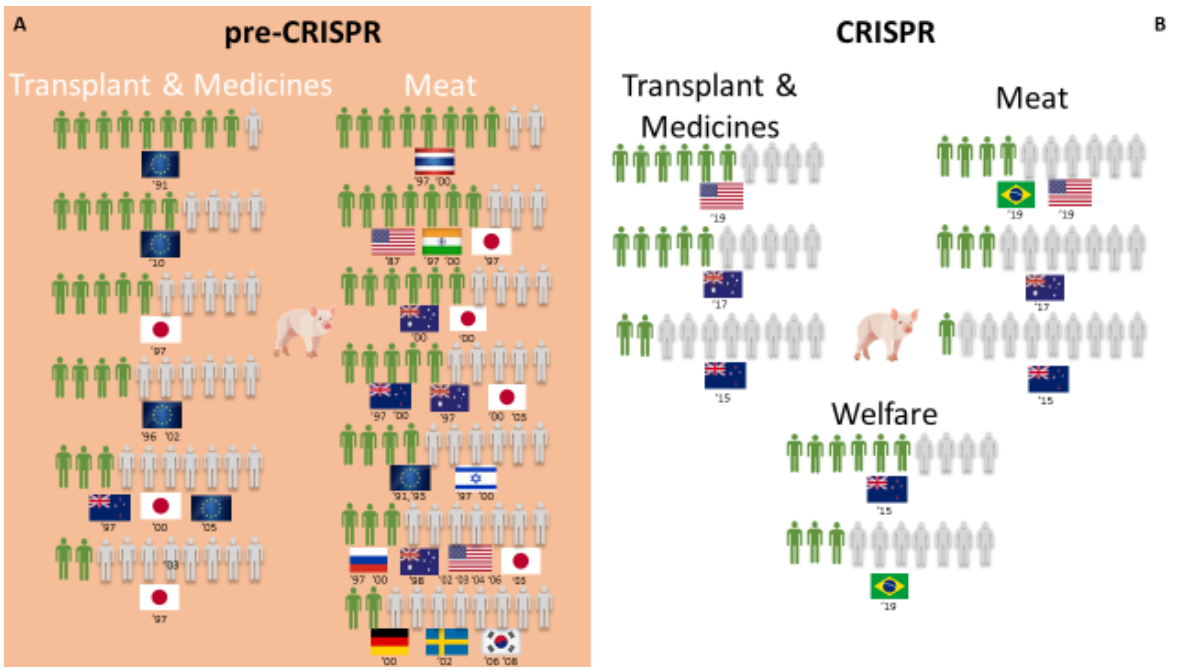


Figure 2. Public support for gene modification in pigs worldwide for a proportion of 10 citizens upon survey inquiry in pre-CRISPR and CRISPR periods. The number of citizens approving GM pigs for transplant & medicines and meat in pre-CRISPR (A) period are represented by a higher number of green stickmen. The number of citizens approving GE pigs for transplant & medicines, meat and welfare purposes in CRISPR period (B) are represented by a higher number of green stickmen.

B) CRISPR: Animal welfare in focus and genome-edited animals for food applications continue to be less approved than for medical ones

The CRISPR period surveys on attitudes to GM animals are a result of a 10-year period inquiry representing a total of 8 surveys worldwide, with the highest number conducted in the US [65, 68, 66, 84]. Table S1 (CRISPR period) (see appendix VI) shows that US citizens approve genetic modification of animals for human health purposes, in this case genome-edited pigs for transplants of organs to humans (6 in 10) and genome-edited mosquitoes to eradicate spreading of diseases into humans (7 in 10). Looking into Oceanic countries, Australian citizens approve more GE cows for medicines rather than for meat and milk derived products while New Zealanders are profound rejecters of GE animals for both applications (fig. 3B). If we now report on the approval for genome-edited pigs for food consumption, Brazilian citizens are mostly rejecters of this (only 4 in 10) by opposition to US citizens where more than half support it either for derived products such as meat in GE pigs or meat and milk in GE cows (figs. 2B and 3B). A new type of question present in

surveys in post-CRISPR era has to do with the support of the public for GE animals based in welfare premises. Here, we can see that US citizens frankly approve “GE cows to become hornless” as a matter of a welfare concern (fig. 3B). For GE pigs, the majority of citizens in New Zealand approve it for better animal health and safety in opposition to Brazilian citizens where the approval for GE pigs to “reduce boar taint in pigs” is below half of the respondents (fig. 2B). The only study covering genome editing in wildlife reported a profound rejection among US citizens (table S1 and fig. S1 – CRISPR period – Appendix V and VI) because this was perceived as a risk for both humans and nature.

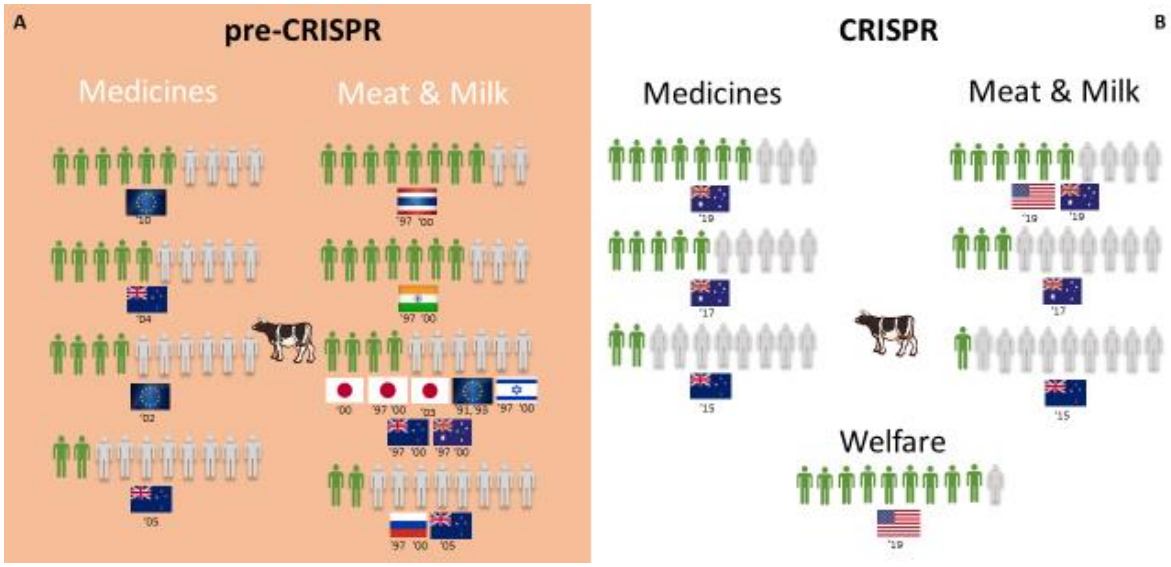


Figure 3. Public support for gene modification in cows worldwide for a proportion of 10 citizens upon survey inquiry in pre-CRISPR and CRISPR periods. The number of citizens approving GM cows for medicines and meat & milk in pre-CRISPR (A) period are represented by a higher number of green stickmen. The number of citizens approving GE cows for medicines, meat & milk and welfare purposes in CRISPR period (B) are represented by a higher number of green stickmen.

Genetic modification of humans in pre-CRISPR and CRISPR periods

A) CRISPR: Somatic genetic modification for therapy is a yes, enhancement is a no

Overall, genetic modification of humans for gene therapy purposes receives medium to high acceptance worldwide (table S1 and figure 4A). Only three exceptions can be identified, two related with disease prevention (4 in every 10 New Zealand respondents agreeing with it for “preventing stomach cancer by modifying a person’s genetic code” and 2 in every 10 UK citizens approving it to prevent baldness. The same low proportion of UK citizens approved of gene therapy to treat aggressive behavior and alcoholism identified as diseases (fig. 4A). The overall greatest support for gene therapy is found among Thai citizens followed shortly by Australian, New Zealanders, Israeli and Japanese citizens in the 90s to cure fatal diseases and UK citizens in the 2000s for genetic diseases like cystic fibrosis and heart diseases (table S1, appendix VI and fig. 4A). On the other side of the genetic modification of humans, enhancement is mostly rejected by all citizens inquired during the pre-CRISPR period with the only exceptions in 1995 and 2000 studies where Thai and Indian citizens show high approval to “make people more ethical” and the ambivalence demonstrated by US citizens in 1987 towards “changing the genetic makeup of human cells” as well as European Union respondents in 2010 regarding human enhancement (table S1 – Appendix V and fig. 4A).

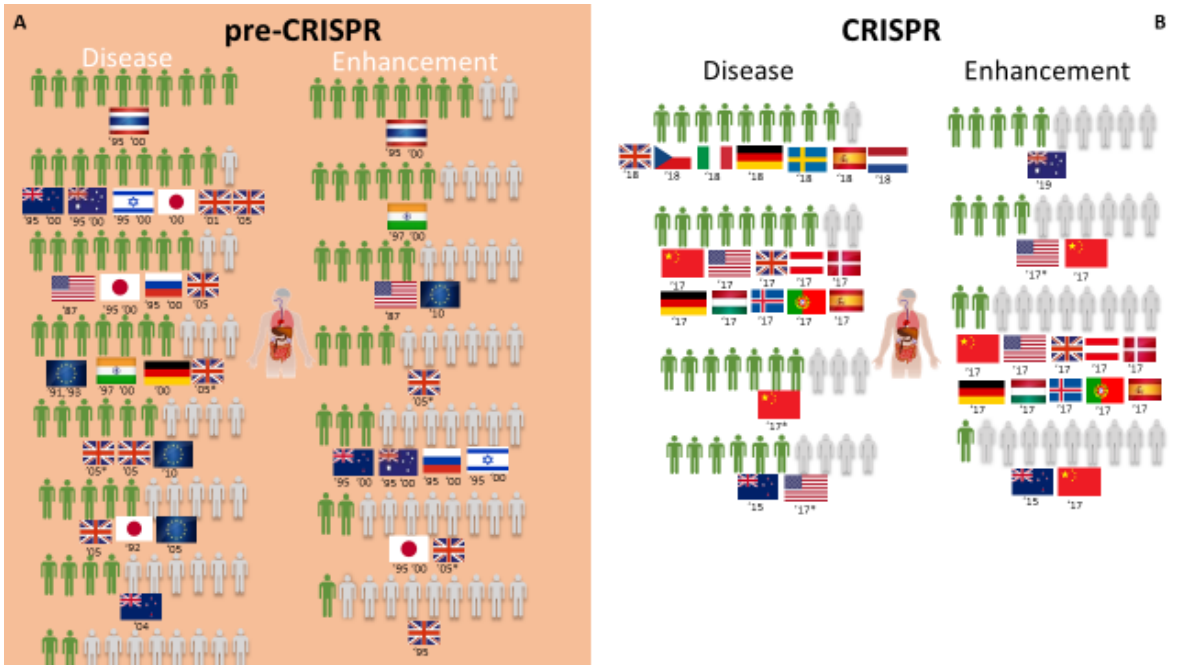


Figure 4. Public support for gene modification in human adults worldwide for a proportion of 10 citizens upon survey inquiry in pre-CRISPR and CRISPR periods. The number of citizens approving GM in human adults for disease and enhancement in pre-CRISPR (A) period are represented by a higher number of green stickmen. The number of

citizens approving GE in human adults for disease and enhancement in CRISPR period (B) are represented by a higher number of green stickmen.

Germline genetic modification for therapy purposes gathered high approval similarly to somatic genetic modification. Once again, there are exceptions and these are citizens from New Zealand in 2005 and Europeans in 2010. For New Zealanders, this represents a drop from much higher levels in the second-half of the 1990s (almost 8 in every 10 citizens supporting it to cure fatal disease (figure 5A) to 10 years later 4 in 10 citizens for approving GE for serious defect and decreasing to 2 for minor defect and to 1 in every 10 citizens to prevent aggression and violence (table S1, Appendix VI and fig. 6A). Among the most approving respondents of germline genome modification (GGM) for therapy are Thai respondents followed closely by Australian and Indian citizens (fig. 5A).

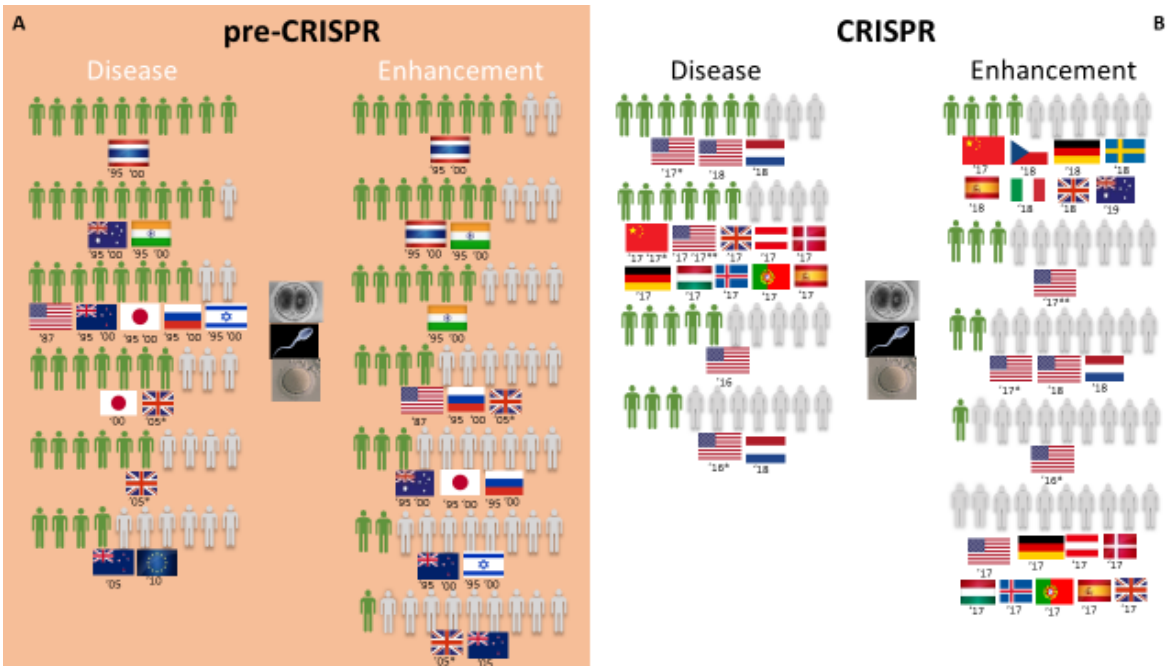


Figure 5. Public support for gene modification in human germline cells worldwide for a proportion of 10 citizens upon survey inquiry in pre-CRISPR and CRISPR periods. The number of citizens approving GM in human germline cells for disease and enhancement in pre-CRISPR (A) period are represented by a higher number of green stickmen. The number of citizens approving GE in in human germline cells for disease and enhancement in CRISPR period (B) are represented by a higher number of green stickmen.

Germline genetic modification (GGM) for enhancement purposes is approved largely by Thai and Indian citizens to improve the physical characteristics and intelligence level “that

children would inherit" (table S1 – Appendix VI and fig. 5A). All the other countries inquired about this reject those applications particularly for the improvement of intelligence, cosmetic modifications in children and determination of sex in an unborn baby (table S1 - Appendix VI).

B) CRISPR: Genome editing of humans for therapy considered more acceptable in somatic than germline but no support for enhancement

Surveys in CRISPR period inquired citizens about genome editing of humans for therapy, similarly to what happened in the pre-CRISPR period and these show strong approval worldwide. At this point, Europeans are the most approving of GE to cure diseases although by a low margin when compared with China and US citizens and with New Zealand citizens following closely. For prevention of diseases, all citizens inquired demonstrate an equally high approval rate of 8 in every 10 citizens (fig. 4B). In children, the approval rate of gene therapy was only assessed in China and showed to be similarly high among citizens (fig. S2 – Appendix V).

Similar to pre-CRISPR period, genetic enhancement of human beings was generally rejected by citizens worldwide (fig. 4A and 4B). Intelligence and change of skin color were purposes profoundly rejected by Chinese citizens (table S1 – Appendix VI). The only case with less than a majority rejecting enhancement (genome editing of "human body cell to change one's appearance") was mentioned by Australian citizens when inquired about it (fig. 4B).

Overall, GE in human germline as in the cases of unborn babies and embryos to cure serious diseases gathered approval among citizens (fig. 5B). US citizens were the most inquired public in CRISPR period and the multiple surveys conducted consecutively from 2016 to 2018 demonstrate a growth in approval of this type of genetic intervention for disease during this period, going from 3 and 5 in every 10 citizens in two surveys in 2016 to 6 and 7 in every 10 citizens in surveys conducted 2017 and 2018, respectively. The remaining studies include European and Chinese publics and approval rates fall between the highest and the lowest of the US studies (fig. 5B and – table S1, Appendix VI). In fact, 7 in every 10 citizens from the Netherlands approve GGE for neuromuscular disease while only 3 in every 10 agree with it for HIV resistance (table S1). As for prevention of diseases, Australian citizens are the most approving ones of GE in germ cells and embryos whereas US citizens have lower approval rates, between 3 in every 10 citizens in 2016 surveys and 5 in every 10 citizens in 2018 that approve genetic interventions in unborn babies (fig. 6B).

Finally, the idea of genetic enhancement of unborn babies is not approved by members of the public anywhere in the world. It was even completely rejected among Europeans and US citizens in a survey conducted in 2017 (fig. 5B), and although the surveys conducted demonstrate a higher approval rate among Europeans one year later, still less than half of the participants agree with germline genetic enhancement, which is similar to the responses of Australian and Chinese citizens (fig. 5B and table S1 – Appendix VI).

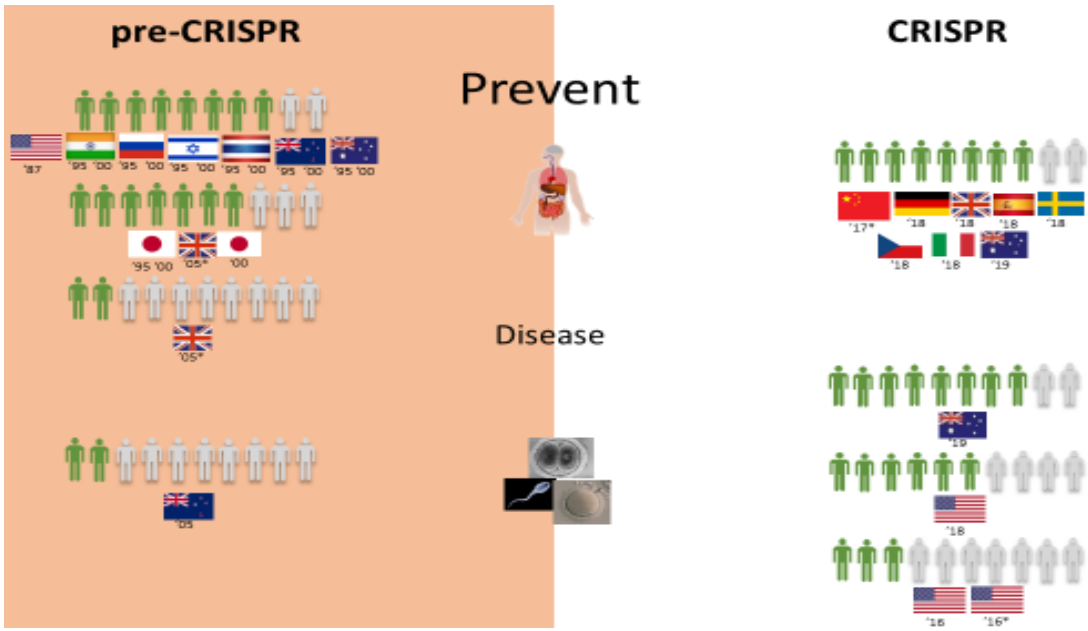


Figure 6. Public support for gene modification in human adults and human germline cells for preventing disease worldwide for a proportion of 10 citizens upon survey inquiry in pre-CRISPR and CRISPR periods. The number of citizens approving GM in human adults and human germline cells to prevent for disease in pre-CRISPR (A) period are represented by a higher number of green stickmen. The number of citizens approving GE in human adults and human germline cells to prevent for disease in CRISPR period (B) are represented by a higher number of green stickmen.

Awareness and knowledge correlation with public attitudes towards genetic modification of humans and animals

Overall, the more aware or knowledgeable inquired publics are about topics of science and technology in general, biotechnology, genetics, genetic modification and gene editing, the most approving they are of genetic modification in humans and animals. In total, 44 surveys assessed awareness knowledge of participants about genetic modification topics, and from these, 17 surveys assessed only awareness (level of familiarity) and 8 surveys assessed only knowledge (how well educated respondents are) about these topics.

In the first US survey in 1987, understanding of science was unrelated with awareness to genetic engineering; however lower levels of awareness were associated with less support for genetic modification of human cells [34]. US citizens demonstrated a somewhat higher although non-significant approval for hybrid animals after hearing about it in a survey in 2002 [60] and this also happened in a survey in 2006 that also counted with South Korean citizens who demonstrated a similar pattern [58]. In Europe, contrasting trends were observed: in 2005 Eurobarometer, the more aware these citizens were the more approvers of gene therapy but also the more rejecters for GM foods [41] whereas in a UK survey in 2007, high attentiveness, i.e. higher awareness and interest in genes and genetics is significantly correlated with lower approval of gene therapy in humans [85]. As for biotechnology and genetic engineering topics, higher awareness of citizens about these seemed to lead to lower approval among Australians, New Zealanders, Japanese, Russians and Israeli but not among Indian and Thai citizens in applications for food purposes such as meat and milk at the intercontinental survey in 1993 [54]. Conversely, when the subject was gene therapy, respondents favoring it less were also the ones showing lower awareness [36]. However, in the first surveys in Japan (1992 and 2000), there was no correlation between awareness to genetic manipulation and willingness to undergo gene therapy nor with approval for the genetic modification of mice or pig's acceptance for medical purposes [35,43]. In the case of GM pigs for human transplants, acceptance seems to have decreased even more with awareness increase over the years [56,64]. In a 2006 survey to Japanese citizens, public opinion was seen to be influenced by the level of awareness about gene therapy where citizens became more aware following the first case of success of gene therapy in humans although remaining undecided [87]. Fast-forward to the CRISPR period, it is possible to notice that the more familiar Japanese respondents were with genome editing, the more accepting they were of it for therapy purposes, but not for germline settings [86]. This contrasts with results for US citizens where germline gene editing was considered more appropriate by respondents who were more aware of it [74,77] even for cases of genetic enhancement [76]. Greater awareness was associated with more support for genetic modification of humans and animals in New Zealand [67] and overall for therapeutic applications of genome editing among European citizens [80].

In the surveys performed in the US in 2002, 2003 and 2004 that measured subjective knowledge (respondents' self-reported understanding of topics) and objective knowledge (as measured in in tests or quizzes) of biotechnology, knowledgeable respondents were found to be more accepting of animal-derived GM products even though the correlation was not strong [60,61,62], similar to what was found in the first survey conducted in the US in 1987 [34]. Among Europeans, there was also a correlation between objective knowledge

and support for biotechnology and genetic engineering (Eurobarometers in 1991 and 1993 [37,38]) and between lower objective knowledge about biotechnology and preference for traditional breeding rather than GM animals (Eurobarometer in 1996 [39]). In the only survey performed in Germany in 2000, however, there was no correlation found between subjective knowledge and support for genetic engineering; in this study most respondents assessing themselves as “ignorant” and strongly opposed several applications involving GM animals [51]. Two UK surveys at the beginning of the first decade of the 2000s also demonstrated more support for medical applications and more disapproval of genetic modification for physical attributes (e.g. baldness) among respondents with higher knowledge [49]. Already in the CRISPR period, US citizens that were more knowledgeable about science and technology were heavy accepters of gene therapy and enhancement applications, although the correlation was weak [75]. About genome editing in animals, a higher support was correlated although not significantly with high knowledge about it among US citizens and with a higher approval for GM cows to be hornless in two surveys conducted in 2018 and 2019 [65,68]. Moreover, gene editing in mosquitoes is highly approved among elements of the public in the US that have a higher level of knowledge of Zika virus [66]. Finally, Australian citizens surveyed in 2019 on gene editing showed more support for applications when their knowledge about the technology was higher [70] whereas in Brazil knowledge was not found to be related with respondents’ acceptance of gene edited pigs to “prevent boar taint” [69]. One study reported a positive association between approval and benefit perception of GE mosquitoes and this was seen as higher than the negative association found between risk perception of GE mosquitoes and its approval [66].

Methodological quality: Reporting of critical issues

This section presents the results of critical appraisal of the methodology as reported in the primary publications selected for analysis, to provide an indication of the methodological quality [19].

Questionnaire development

Surveys may be the result of original item generation or adaptation of items used in previous surveys. For pre-CRISPR surveys, there was an approximately even distribution between the 8 studies originally generating their own items and the 10 which adapted existing. For studies in the CRISPR period, generating own items was much more common (11 versus 4). As for the remaining 18 surveys that have information available both in pre-CRISPR and CRISPR periods, there has been a hybrid approach followed.

The validity of a survey instrument has to do with how well it measures what it is supposed to measure. Face validity (whether it appears to measure what it should) and content validity (if it is understandable to respondents) were the most reported types, in 29 and 37 studies respectively [16, 17]. Construct validity to check if the construct used is suitable appears mostly in the form of hypothesis-testing in 16 pre-CRISPR and 14 CRISPR surveys.

Reliability is about how reproducible survey instrument data are across different applications of the survey. Most papers (32 of the 53) included in the review did not report this parameter. Among the papers that did, the most used was Chronbach's alpha index to measure internal consistency and split-half reliability where samples are divided in halves or thirds to ensure that there isn't a significant difference between groups of individuals studied.

Sampling: method, response rate and weighing

The methodology of sampling participants for surveys is very diverse across the different surveys analyzed. CRISPR surveys were conducted mostly online, and pre-CRISPR surveys overlap between telephone, face-to-face and mail response. Quota sampling from databases (rather than random sampling) was more common in CRISPR surveys than pre-CRISPR surveys (10 vs 3). Weighing of the sample has been used to overcome potential sampling bias but was reported in less than half of the studies. In the studies where weighing was reported, the correction tool mostly used was based in demographics for both pre-CRISPR and CRISPR surveys. The majority of the studies report a medium response rate (25-75% of invited participants responded). CRISPR studies show a medium to high response rate compared with pre-CRISPR that overlaps between low to medium response rate in general. Moreover, multinational surveys such as Eurobarometers and intercontinental surveys demonstrate a different response rate per country and therefore sample weighing was used. Furthermore, an equal number of pre-CRISPR and CRISPR surveys did not report on response rate (7 to be precise).

Methodology accountability and reporting

Only half of the studies provide information on bias and this is transversal to both pre- and CRISPR studies. The most commonly referred by authors in the studies from the systematic review is recruitment bias, with under- or overrepresentation of certain demographic groups

for education, age, gender, race and socio-economic status. Some studies report techniques to avoid bias namely the use of random digit dialing to avoid inadequate telephone surveys [34], demographics comparisons to Census to avoid sample distortions [34], standardization of questionnaires and their delivery [36], use of open responses [36,44], background campaigning [72,73], online survey to have a more robust sample [83], online tools to avoid age bias [81] and not mentioning survey nature to avoid self-selection bias [68,69]. In CRISPR studies, authors report about ethical practices taken during survey conduction whereas in pre-CRISPR this is mostly non-existent. Such practices involve obtaining informed consent from participants, voluntary participation invitation, obtaining a privacy statement or even the chance of withdrawal from the study. Formal ethics approval for the study was only reported for 10 studies from the total of publications in the systematic review. Finally, incentives to participants in order to increase their willingness to participate were disclosed in 9 studies.

DISCUSSION

This systematic review of 53 primary publications on attitude to genetic modification in humans and non-human animals provide a comprehensive picture of studies in Europe, North America, Asia and Oceania over 35 years. Taken together, the review shows some variation between countries but a clear pattern in how different applications are viewed, that does not change substantially over time.

There is an overall positive attitude to gene therapy for disease treatment or prevention in humans, both for adults and children and both as treatment for a fatal genetic disease and as prevention from developing a disease that would otherwise be likely to happen. This is transversal from the early studies before the 1990s to the most recent studies, with little variation among public and regardless of their origin. This is in agreement with international and national policies [88,89,90,91,92], and indeed several clinical trials of somatic gene therapy are underway [93,94]. Key challenges on the use of these therapies in the clinic raised by scholars regard their definition and regulation [91,95], and were partly recognized in some of the public opinion surveys, including the “need for strict regulation” in somatic therapy [42] and the need for FDA approval to proceed with such [74].

The differentiation between germline and somatic cells becomes important over time. Surveys administered pre-CRISPR hardly ever distinguish between correction of genes carrying disease for the individual and the ones that can be passed onto future generations. In contrast, post-CRISPR surveys address this directly not just by questioning explicitly about germline and unborn babies but also when asking both about germline versus somatic

therapy and adult versus prenatal therapy. Overall, somatic gene therapy is widely accepted in most surveys, whereas there is much ambivalence about germline gene therapy, with higher support to prevent future health issues in still unborn babies and a lower support if the purpose are non-health related issues like physical and psychological characteristics. The ethics of germline gene editing experienced a spike of interest with the advent of the CRISPR-Cas9 technology [4,5,10,96] and the ethical issues are discussed by the general public and the scientific community in distinct ways. The public mentions unnaturalness, messing with nature and humans playing God in the creation of designer babies as main arguments to reject germline gene editing and health benefits to accept it. Researchers on the other hand primarily refer to the technical hurdles and uncertainties such as off-target effects and mosaicism as the background of ethical questions related with unintended consequences and safety, but also the problem of introducing irreversible changes to the genome of future individuals whose consent cannot be obtained [5,10,96,98,99]. Many scholars defend that, while germline gene editing will eventually be inevitable, the technology should not be pursued in the clinic except when no other alternative exists to prevent a severe or deadly genetically transmitted disease, and only after the technology has proven to be safe to proceed to clinical trials [5,10,97,98,99]. Others defend research on gene editing could improve understanding of genetic diseases and should be used for single-gene disorders and other disorders arising from polygenic traits [99]. Scholars have defended the adoption of a moratorium on germline gene editing more than once: following the first edit on human cells and after the birth of the first gene-edited babies in late November 2018, respectively [5, 93,100,101], often justified by the precautionary principle and taking into account the unpredictability of an emerging technology [4].

A third relevant point is the differentiation between therapy and enhancement. Across countries, citizens are generally opposed to genetic modification for the purpose of enhancement. When asked to distinguish between different types of enhancement, intelligence or psychological features were favored over physical abilities and appearance in US and British studies. Across the countries where there is some support for non-therapeutic gene editing, the most supported purpose is improved human health. This is line with the establishment of a purpose for genome editing beforehand and the clear distinction of what is a disease and what is a deviation from a societal-norm [5]. As for current guidelines, the US National Academies of Sciences Engineering and Medicine exclude the use of genome modification for any type of enhancement under any circumstance [5,96]. The reasons for this are also aligned with the slippery-slope argument that gene editing will ultimately lead to social harm by the creation of new genetically-

modified humans that may lead to “new forms of inequality, discrimination and societal conflict” if regulation fails to limit germline gene editing to therapeutic uses [99].

As regards GM animals, the aspect that stands out as a continued trend is the way acceptance differs between different purposes. Overall, GM animals appear as generally not acceptable for food purposes, be it for leaner or healthier meat as in the case of GM pigs or to produce more milk in the case of GM cows. Novoselova et al (2007) highlight the important role of consumers for the potential integration of GM products derived from animals into the food chain, pointing out perception of health and safe food as well as understanding of environmental and ethical concerns as key issues [103]. This perception is based on arguments that “genetic modification is intrinsically wrong” when to be applied for food applications [104], with many people even questioning the usefulness of such applications [105]. Risk and benefit perceptions regarding food are affected by many factors which interact in complex ways; specifically, as regards animals this is further complicated by the duality of the animal as friend and food [106]. As for GM pigs or GM sheep for medical purposes such as organs for transplantation and derived products to help with diseases, the acceptance is higher. Also among professionals who are involved with animal research, support for GM pigs in medical applications like xenotransplantation was greater than for food applications [107]. While this would overcome the shortage of human organs for transplantation, this discussion is again reflecting current and older moral reservations regarding the mixing of tissues from human and non-human species as well as the unnaturalness and invasiveness of the process and ultimately the risk for human health [93,108,109,110]. Similarly, it has been found over the years of public opinion surveys that public perceptions of risk are higher when they concern GM animals rather than GM crops/microorganisms and are also perceived as riskier and having more ethical concerns if the context is food rather than medical ones, as the latter tend to be evaluated on a more specific or case-by-case basis [104,111]. The two differences that appear when comparing surveys from before and after the introduction of CRISPR-Cas9 technology are largely associated with the type of questions that were asked. In pre-CRISPR surveys, most respondents see laboratory research in animal models like GM mice as useful but not morally acceptable. This reflects an ambivalence between what is perceived to be a valuable objective (the study of human disease) and the concerns over animals’ welfare [93,105,108,112]. In the post-CRISPR surveys that include animal applications, the questions are about applications where genetic modification is done to avoid animal welfare problems, and while people mention some concern, in particular about potential suffering, overall they see it as something good. However, they also reveal an unwillingness to consume products derived from these animals, similar to respondents in pre-CRISPR

surveys. This follows the usual perception of risks and ethical concerns where the public has also been found to be willing to pay less for GM foods than conventional ones [111]. Impact on human health by introduction of genetically modified species in the food chain, naturalness and potential ecosystem disturbance are also recognized as moral issues of these interventions [4,93,108,113]. Impact on biodiversity and sustainability are repeatedly identified ethical concerns about genetic modification of animals, together with animal welfare, tampering with Nature and unnaturalness [105,107,111,114]. Furthermore, GM animals are also seen more negatively than GM plants and the perception that the technology is unnatural has increased over the years [115].

Across many surveys there is a correlation with support for gene technology: the higher the awareness and knowledge levels, the higher the support as well. This lends some support for the deficit model, according to which education and improved public understanding of science would lead to a higher acceptance of food that is genetically-engineered and gene therapy subjects [33,120]. However, in most of the cases, this relationship is weak, and awareness and knowledge levels towards genetic engineering or modification and biotechnology are generally not considered predictive of public attitude [33,111,121,122,123,124].

The critical appraisal of methodological quality shows that most studies provide low to medium quality information. Only two publications [53,84] fulfill all the criteria recommended for questionnaire surveys [13,14,19,125]. Most studies report or demonstrate consideration of two to three of the criteria, but typically not on the aspects considered more relevant for ensuring methodological quality, such as item generation method and response rate. Characteristics of greater relevance such as validity, reliability, risk of bias and sampling are reported at a much lower frequency than what is desired. Poor methodological quality may justify the exclusion of studies from a systematic review. We nevertheless included all surveys into this systematic review, firstly because our priority was comprehensiveness and secondly in order to be able to highlight the issue of study quality, which is not yet receiving as much attention in reviews of social science research as it does in biomedical research. While not reporting does not necessarily mean that the practice was absent, it does at least suggest limited attention to methodology. Lack of information is more common in earlier studies, which probably reflects the changing practice in the field. One also needs to distinguish between survey reports in grey literature, which focus on reporting the results, from articles in scholarly journals with peer review, where a discussion of methods and issues such as risk of bias is expected to be an integral part of reporting.

To the best of our knowledge, our study is unique in comprehensiveness. Firstly, it includes publications covering almost 35 years and addressing attitudes to human as well as non-human genetic modification. Whereas the recent systematic review by Delhove and collaborators (2020) took a similar approach in terms of timespan and definition of primary publications, it covers only attitudes to human genetic modification [128]. The limitations of our study include the choice of databases, studies and information to include. We used WOS as source database as well as Google web search for publication retrieval. It is possible that other databases would have generated a somewhat different outcome in terms of selected publications. We chose only to include studies of the general public, excluding studies of only specific publics [123,126,127]. In terms of analysis of results, we opted to only assess the influence of awareness and knowledge in public attitudes, and did not include other parameters that could have had an influence here like trust in organizations, demographics (e.g. socio-economic status) and religious index. The reason to only include awareness and knowledge is because these variables have been continuously assessed and therefore we could have a parallel view on how they would have influenced opinions of the public towards genetic modification over time. Finally, the present paper includes only a qualitative analysis of quantitative results and did not perform a meta-analysis.

Future perspectives

Public consultation is critical in controversial matters in relation to genetics and biotechnology, especially when applications will potentially directly influence citizens' lives and therefore has to be ensure accurate representation [3,130]. While cross-sectional surveys such as the ones we analyzed are important in that they provide an overview of how public opinion evolved during the last 35 years, real comprehensive initiatives of public engagement and societal debate on genome modification beforehand are indispensable [131,132,133]. This could include a citizen policy approach such as described for climate action policy [134,135]. This would be particularly important in the context of policy making for CRISPR-Cas9 technology implementation. Design of citizen engagement initiatives with multiple stakeholders in the discussion of genome editing driven by the intervention of some associations already in place like Association for Responsible Research and Innovation in Genome Editing (ARRIGE) will elevate the dialogue and contribute to the adoption of a participatory governance framework that may resemble such reflections [8,136,137]. This path would also entail the best opportunity for scientists and policy makers to consolidate RRI practices in an era where speed of technology implementation is key but responsibility of its adoption is mandatory [12,131].

The surveys we analyzed varied widely in methodology and more standardized approaches across countries and over time would be important for such future studies. A good example to follow are Eurobarometers and international surveys that demand a higher collaboration between teams and offer a consistent overview that may transform a cross-sectional view into a more longitudinal one allowing for more robust hypothesized theories over time [13,14].

Additionally, the bioethics literature on biotechnology recognizes a wider range of issues than those that have been covered in the public attitude surveys, such as eugenics, access to technology, funding of genome technologies and social justice. These are subjects that impact the public, and which they often care about, and should be included in future studies as well [5,6,113]. Principles linked to solidarity and social justice as well as future generations welfare is a must on this quest and need to be respected and be the drive to the generation of policies concerning genome editing [3,130,138]. Finally, it is important to include assessment of technology awareness and knowledge as part of the survey. Many surveys indicate low levels of knowledge and awareness, and these factors seem to be related to opinion, at least to some extent.

References

1. Barrangou R, Horvath P. A decade of discovery: CRISPR functions and applications. Nat Microbiol. 2017;2:17092.

2. Camporesi S, Cavaliere G. Emerging ethical perspectives in the clustered regularly interspaced short palindromic repeats genome-editing debate. Per Med. 2016;13(6):575-586

3. Jasanoff S, Hurlbut JB, Saha K. CRISPR democracy: Gene editing and the need for inclusive deliberation. Issues in Sci Technol. 2015;32(1):1-14

4. Nordberg A, Minssen T, Holm S, Horst M, Mortensen K, Moller BL. Cutting edges and weaving threads in the gene editing (Я)evolution: reconciling scientific progress with legal, ethical, and social concerns. J Law Biosci. 2018;5(1):35-83.

5. Brokowski C, Adli M. CRISPR Ethics: Moral Considerations for Applications of a Powerful Tool. Journal of Molecular Biology. 2019;431(1):88-101.

6. Isasi R, Kleiderman E, Knoppers B. Editing policy to fit the genome? SCIENCE. 2016;351(6271):337-339

7. Boggio A, Knoppers BM, Almqvist J, Romano CPR. The Human Right to Science and the Regulation of Human Germline Engineering. CRISPR J. 2019;2:134-42.

8. Hirsch F, Lemaitre C, Chneiweiss H, Montoliu L. Genome Editing: Promoting Responsible Research. Pharmaceut Med. 2019;33(3):187-91.
9. Meagher KM, Allyse MA, Master Z, Sharp RR. Reexamining the Ethics of Human Germline Editing in the Wake of Scandal. Mayo Clin Proc. 2020;95(2):330-8.
10. Morrison M, de Saille S. CRISPR in context: towards a socially responsible debate on embryo editing. Palgrave Communications. 2019;5(1).
11. Caplan AL, Parent B, Shen M, Plunkett C. No time to waste--the ethical challenges created by CRISPR. EMBO Rep. 2015;16(11):1421-6.
12. Shelley-Egan C, Gjefsen MD, Nydal R. Consolidating RRI and Open Science: understanding the potential for transformative change. Life Sci Soc Policy. 2020;16(1):7.
13. Stockemer D. A short introduction to survey research. In: Stockerner D editors. Quantitative methods for the social sciences: A practical introduction with examples in SPSS and Stata. Ottawa: Springer; 2019. pp.23-35
14. Stockemer, D. Conducting a survey. In: Stockerner D editors. Quantitative methods for the social sciences: A practical introduction with examples in SPSS and Stata. Ottawa: Springer; 2019. pp.57-72
15. Haddock G, Maio GR. Attitudes: content, structure and functions. In: Hewstone M, Stroebe W and Jonas K editors. Introduction to social psychology: a European perspective. BPS textbooks in psychology. Oxford: Blackwell; 2008. pp. 112-133.
16. Boateng GO, Neilands TB, Frongillo EA, Melgar-Quinonez HR, Young SL. Best Practices for Developing and Validating Scales for Health, Social, and Behavioral Research: A Primer. Front Public Health. 2018;6:149.
17. Hair JF, L.D.S. Gabriel M, da Silva D, Braga Junior S. Development and validation of attitudes measurement scales: fundamental and practical aspects. RAUSP Management Journal. 2019;54(4):490-507.
18. The Netherlands Commission on Genetic Modification COGEM. CRISPR & Animals: Implications of genome editing for policy and society. 2018 Policy Report CGM/180501-01.
19. Petticrew M, Roberts H. Systematic Reviews in the Social Sciences – A Practical guide. 1st ed. Oxford: Blackwell Publishing; 2005
20. Crowe M, Sheppard L. A review of critical appraisal tools show they lack rigor: Alternative tool structure is proposed. J Clin Epidemiol. 2011;64(1):79-89.
21. Moher D, Liberati A, Tetzlaff J, Altman DG, Group P. Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement. PLoS Med. 2009;6(7):e1000097.
22. National Health and Medical Research Council. Appraising and selecting studies. In: National Health and Medical Research Council editors, How to review the evidence:

systematic identification and review of the scientific literature. Canberra: Commonwealth of Australia; 2000. pp. 23-25
23. National Health and Medical Research Council. Aetiology and risk factors. In: National Health and Medical Research Council editors, How to review the evidence: systematic identification and review of the scientific literature Canberra: Commonwealth of Australia;2000. pp. 71-77
24. Nolan M, Beran T, Hecker KG. Surveys assessing students' attitudes toward statistics: A systematic review of validity and reliability. SERJ. 2012;11(2):103-123
25. Oluka OC, Nie S, Sun Y. Quality assessment of TBP-based questionnaires: A systematic review. PLoS One. 2014;9(4):e94419.
26. Pace R, Pluye P, Bartlett G, Macaulay AC, Salsberg J, Jagosh J, et al. Testing the reliability and efficiency of the pilot Mixed Methods Appraisal Tool (MMAT) for systematic mixed studies review. Int J Nurs Stud. 2012;49(1):47-53.
27. Lassen J, Gjerris M, Sandoe P. After Dolly--ethical limits to the use of biotechnology on farm animals. Theriogenology. 2006;65(5):992-1004.
28. Condit CM. Public attitudes and beliefs about genetics. Annu Rev Genomics Hum Genet. 2010;11:339-59.
29. Howell EL, Yang S, Beets B, Brossard D, Scheufele DA, Xenos MA. What Do We (Not) Know About Global Views of Human Gene Editing? Insights and Blind Spots in the CRISPR Era. The CRISPR Journal. 2020;3(3):148-55.
30.
31.
30. Centre for Reviews and Dissemination. Core principles and methods for conducting a systematic review of health interventions. In: Centre of Reviews and Dissemination editors, Systematic Reviews. York: University of York; 2009. pp.1-108
31. Centre for Reviews and Dissemination. Incorporating qualitative evidence in or alongside effectiveness reviews. In: Centre of Reviews and Dissemination editors, Systematic Reviews. York: University of York; 2009, pp. 219-237
32. Sandhu JS, Keating A, Hozumi N. Human gene therapy. Crit Rev Biotechnol. 1997;17(4):307-26.
33. Gottweis, H. Gene therapy and the public: a matter of trust. Gene therapy. 2002;9:667-669
34. Office of Technology Assessment. New Developments in Biotechnology—Background Paper: Public Perceptions of Biotechnology. 1987
35. Macer DRJ. PUBLIC ACCEPTANCE OF HUMAN GENE-THERAPY AND PERCEPTIONS OF HUMAN GENETIC MANIPULATION. Human Gene Therapy. 1992;3(5):511-8.

36. Macer DRJ, Akiyama S, Alora AT, Asada Y, Azariah J, Azariah H, et al. INTERNATIONAL PERCEPTIONS AND APPROVAL OF GENE-THERAPY. Human Gene Therapy. 1995;6(6):791-803.
37. Comission of the European Communities Directorate-General Science, Research and Development . Opinions of Europeans on Biotechnology in 1991. 1991 Eurobarometer 35.1
38. Comission of the European Community Directorate-General Science, Research and Development Unit XII/E/1: Biotechnologies. Biotechnology and Genetic Engineering: What Europeans think about it in 1993. 1993 Eurobarometer 39.1
39. European Comission Directorate-General Science, Research and Development XII. European opinions on modern biotechnology. 1996 Eurobarometer 46.1
40. European Comission Directorate General for Research. Europeans and Biotechnology in 2002. 2002 Eurobarometer 58.0 QLG7-CT-1999-00286
41. European Comission Directorate-General for Research. Europeans and Biotechnology in 2005: Patterns and Trends. 2006 Eurobarometer 63.1
42. European Comission Directorate-General for Research. Europeans and biotechnology in 2010: Winds of change? 2010 Eurobarometer 73.1
43. Ng MAC, Takeda C, Watanabe T, Macer D. Attitudes of the public and scientists to biotechnology in Japan at the start of 2000. Eubios journal of Asian and international bioethics : EJAIB. 2000;10(4):106-13
44. Macer DRJ, Azariah J, Srinives P. Attitudes to biotechnology in Asia. International of Biotechnology. 2000;2:313-332. Available from: https://www.eubios.info/Papers/ASIABIO.htm
45. Evans MDR, Kelley J, Zanjani ED. The ethics of gene therapy and abortion: Public opinion. Fetal Diagnosis and Therapy. 2005;20(3):223-34.
46. Cook AJ, Fairweather JR, Satterfield T, Hunt LM. New Zealand Public Acceptance of Biotechnology. 2004 AERU Research Report n.269
47. Torgensen H, Hampel J, Bergmann-Winberg, Mv, Bridgman E, Durant J, Einsiedel E et al. Promises, Problems and Proxies: 25 years of European biotechnology debate and regulation. In: Bauer M, Gaskell G editors. Biotechnology: The making of a global controversy. Cambridge: Cambridge University Press; 2002, pp.1-62
48. Human Genetics Commission. Public attitudes to human genetic information. 2001
49. Sturgis P, Cooper H, Fife-Schaw C. Attitudes to biotechnology: estimating the opinions of a better-informed public. New Genetics and Society. 2005;24(1):31-56
50. Marteau T, Michie S, Drake H, Bobrow M. PUBLIC-ATTITUDES TOWARDS THE SELECTION OF DESIRABLE CHARACTERISTICS IN CHILDREN. Journal of Medical Genetics. 1995;32(10):796-8.

51. Hampel J, Pfenning U, Peters HP. Attitudes towards genetic engineering. New Genetics and Society. 2000;19(3):233-49

52. Norton J, Lawrence G, Wood G. The Australian public's perception of genetically-engineered foods. Australasian Biotechnology. 1998;8(3):172-81.

53. Magnusson MK, Hursti UKK. Consumer attitudes towards genetically modified foods. Appetite. 2002;39(1):9-24.

54. Macer DRJ. Ethics and public acceptance of agricultural biotechnology. In: A. Altman editor. Biotechnology in Agriculture. New York: Marcel Dekker; 1997. pp. 661-90

55. Macer D, Ng MAC. Changing attitudes to biotechnology in Japan. Nature Biotechnology. 2000;18(9):945-7.

56. Inaba M, Macer DRJ. Attitudes to biotechnology in Japan in 2003. EJAIB. 2003;13:78-89. Available from: https://www.eubios.info/EJ133/ej133b.htm

57. Small BH, Parminter TG, Fisher MW. Understanding public responses to genetic engineering through exploring intentions to purchase a hypothetical functional food derived from genetically modified dairy cattle. New Zealand Journal of Agricultural Research. 2005;48(4):391-400.

58. Nayga RM, Fishera MG, Onyangob B. Acceptance of genetically modified food: comparing consumer perspectives in the United States and South Korea. Agricultural Economics. 2006;34(3):331-41.

59. Govindasamy R, Onyango B, Hallman WK, Jang H-M, Puduri V. Public approval of plant and animal biotechnology in South Korea: An ordered probit analysis. Agribusiness. 2008;24(1):102-18.

60. Hallman WK, Adelaja AO, Schilling BJ, Lang JT. Public Perceptions Of Genetically Modified Foods: Americans Know Not What They Eat. 2002 Food Policy Institute, Research Reports 18176

61. Hallman WK, Hebden WC, Aquino HL, Cuite CL, Lang, JT. Public Perceptions of Genetically Modified Foods: A National Study of American Knowledge and Opinion. 2003 Food Policy Institute RR-1003-004

62. Puduri V, Govindasamy R, Lang JT, Onyango B. I Will Not Eat It with a Fox; I Will Not Eat It in a Box: What Determines Acceptance of GM Food for American Consumers? Choices. 2005;20(4):257-261

63, Macer D, Bezar H, Harman N, Kamada H, Macer N. Attitudes to Biotechnology in Japan and New Zealand in 1997, with International Comparisons. EJAIB. 1997;7:137-151. Available from: https://www.eubios.info/EJ75/ej75h.htm.

64. Inaba M, Macer DRJ. Japanese Views of Medical Biotechnology. In: Sang-youg S, Young-Mo K, Macer, DRJ editors. Asian Bioethics in the 21st Century. Tsukuba: Eubios Ethics Institute; 2003. pp. 178-196

65. Funk C and Heferon M. Most Americans Accept Genetic Engineering of Animals That Benefits Human Health, but Many Oppose Other Uses. 2018 Pew Research Center
66. Lull RB, Akin H, Hallman WK, Brossard D, Jamieson KH. Modeling Risk Perceptions, Benefit Perceptions, and Approval of Releasing Genetically Engineered Mosquitoes as a Response to Zika Virus. Environmental Communication-a Journal of Nature and Culture. 2019.
67. Chikhazhe TL. New Zealand public attitudes towards genetically modified food. M.Sc. Thesis, Lincoln University. 2015. Available from: https://researcharchive.lincoln.ac.nz/handle/10182/6825
68. McConnachie E, Hotzel MJ, Robbins JA, Shriver A, Weary DM, von Keyserlingk MAG. Public attitudes towards genetically modified polled cattle. Plos One. 2019;14(5).
69. Yunes MC, Teixeira DL, von Keyserlingk MAG, Hotzel MJ. Is gene editing an acceptable alternative to castration in pigs? Plos One. 2019;14(6).
70. Critchley C, Nicol D, Bruce G, Walshe J, Treleaven T, Tuch B. Predicting Public Attitudes Toward Gene Editing of Germlines: The Impact of Moral and Hereditary Concern in Human and Animal Applications. Frontiers in Genetics. 2019;9.
71. Cormick C, Mercer R. Community attitudes to gene technology. 2017 The Office of the Gene Technology Regulator J2537
72. McCaughey T, Sanfilippo PG, Gooden GEC, Budden DM, Fan L, Fenwick E, et al. A Global Social Media Survey of Attitudes to Human Genome Editing. Cell Stem Cell. 2019;18(5):569-572.
73. McCaughey T, Budden DM, Sanfilippo PG, Gooden GEC, Fan L, Fenwick E, et al. A Need for Better Understanding Is the Major Determinant for Public Perceptions of Human Gene Editing. Human Gene Therapy. 2019;30(1):36-43.
74. STAT and Harvard T.H. Chan School of Public Health. The public and genetic editing, testing, and therapy. 2016 Harvard T.H. Chan School of Public Health
75. Scheufele DA, Xenos MA, Howell EL, Rose KM, Brossard D, Hardy BW. SCIENCE AND SOCIETY U.S. attitudes on human genome editing. Science. 2017;357(6351):553-4.
76. Funk C, Kennedy B, Sciupac E. U.S. Public Wary of Biomedical Technologies to ‘‘Enhance’’ Human Abilities. 2016 Pew Research Center
77. Funk C and Heferon M. Public Views of Gene Editing for Babies Depend on How It Would Be Used. 2018 Pew Research Center
78. Gaskell G, Bard I, Allansdottir A, da Cunha RV, Eduard P, Hampel J, et al. Public views on gene editing and its uses. Nature Biotechnology. 2017;35(11):1021-3.
79. Hendriks S, Giesbertz NAA, Bredenoord AL, Repping S. Reasons for being in favour of or against genome modification: a survey of the Dutch general public. Human reproduction open. 2018;2018(3):hoy008-hoy.

80. Lakomý M, Bohlin G, Hlavová R, Macháčková H, Bergman M, Lindholm M. Public attitudes to life sciences research in six European countries. 2018 ORION
81. Wang J-H, Wang R, Lee JH, Iao TWU, Hu X, Wang Y-M, et al. Public Attitudes toward Gene Therapy in China. Molecular Therapy-Methods & Clinical Development. 2017;6:40-2.
82. Chen, C, Liang, Z. Chinese Public Attitudes on Gene Editing. 2018 Sun-Yat Sen University
83. Weisberg SM, Badgio D, Chatterjee A. A CRISPR New World: Attitudes in the Public toward Innovations in Human Genetic Modification. Frontiers in Public Health. 2017;5(117):1-9.
84. Kohl PA, Brossard D, Scheufele DA, Xenos MA. Public views about editing genes in wildlife for conservation. Conservation Biology. 2019;33(6):1286-95.
85. Barnett J, Cooper H, Senior V. Belief in public efficacy, trust, and attitudes toward modern genetic science. Risk Analysis. 2007;27(4):921-33.
86. Uchiyama M, Nagai A, Muto K. Survey on the perception of germline genome editing among the general public in Japan. Journal of Human Genetics. 2018;63(6):745-8.
87. Sato H, Akabayashi A, Kai I. Public, experts, and acceptance of advanced medical technologies: The case of organ transplant and gene therapy in Japan. Health Care Analysis. 2006;14(4):203-14.
88. Walters L. Human Gene Therapy: Ethics and Public Policy. Human Gene Therapy. 1991;2:115-122
89. Horst M. Public Expectations in Gene Therapy: Scientific Futures and Their Performative Effects on Scientific Citizenship. Science, Technology & Human Values. 2007;32(2):150-171
90. Polcz S, Lewis A. CRISPR-Cas9 and the non-germline non-controversy. Journal of Law and the Biosciences. 2016;3(2):413-25.
91. Nicol D, Eckstein L, Morrison M, Sherkow JS, Otlowski M, Whitton T, et al. Key challenges in bringing CRISPR-mediated somatic cell therapy into the clinic. Genome Med. 2017;9(1):85.
92. Committee on Bioethics (DH-BIO). Statement on genome editing technologies. 2015 Statement of Council of Europe DH-BIO/INF 13 Final
93. European Academies Science Advisory Council. Genome editing: scientific opportunities, public interests and policy options in the European Union. 2017 EASAC policy report 31
94. Karagyaur MN, Efimenko AY, Makarevich PI, Vasiluev PA, Akopyan ZA, Bryzgalina EV, et al. Ethical and Legal Aspects of Using Genome Editing Technologies in Medicine (Review). Sovremennye tehnologii v medicine. 2019;11(3).

95. Sherkow JS, Zettler PJ, Greely HT. Is it 'gene therapy'? J Law Biosci. 2018;5(3):786-93.
96. National Academy of Sciences, Engineering and Medicine. Human Genome Editing: Science, Ethics, and Governance. 2017
97. Brokowski C. Do CRISPR Germline Ethics Statements Cut It? CRISPR J. 2018;1:115-25.
98. Bosley K, Botchan M, Bredenoord A, Carrol D, Alta Charo R, Chapentier E et al. CRISPR germline engineering – the community speaks. Nat Biotechnology. 2015;33(5):478-486
99. Gyngell C, Douglas T, Savulescu J. The Ethics of Germline Gene Editing. J Appl Philos. 2017;34(4):498-513.
100. On Human Gene Editing: International Summit Statement [Internet]. 2015; Dec. 3
101. Lander S. Adopt a moratorium on heritable genome editing. Nature, 2019, Mar 13 [Cited 2021 Jan 22]. Available from: https://www.nature.com/articles/d41586-019-00726-5
102. Convention for the Protection of Human Rights and Dignity of the Human Being with regard to the Application of Biology and Medicine: Convention on Human Rights and Biomedicine, Stat. 13 (1997).
103. Novoselova TA, Meuwissen MPM, Huirne RBM. Adoption of GM technology in livestock production chains: an integrating framework. Trends in Food Science & Technology. 2007;18(4):175-88.
104. Frewer L. 10. Societal issues and public attitudes towards genetically modified foods. Trends in Food Science & Technology. 2003;14(5-8):319-32.
105. Macnaghten P. Animals in their Nature. Sociology. 2016;38(3):533-51.
106. Ueland O, Gunnlaugsdottir H, Holm F, Kalogeras N, Leino O, Luteijn JM, et al. State of the art in benefit-risk analysis: consumer perception. Food Chem Toxicol. 2012;50(1):67-76.
107. Schuppli CA, Weary DM. Attitudes towards the use of genetically modified animals in research. Public Underst Sci. 2010;19(6):686-97.
108. de Graeff N, Jongsma KR, Johnston J, Hartley S, Bredenoord AL. The ethics of genome editing in non-human animals: a systematic review of reasons reported in the academic literature. Philos Trans R Soc Lond B Biol Sci. 2019;374(1772):20180106.
109. Luna A. The Unknown of Human Animal Chimera Research. Intersect. 2017;10(3):1-8
110. Einsieddel EF. Public perceptions of transgenic animals. Rev. sci. tech. Off. In. Epiz. 2005;24(1):149-157

111. Frewer LJ, Bergmann K, Brennan M, Lion R, Meertens R, Rowe G, et al. Consumer response to novel agri-food technologies: Implications for predicting consumer acceptance of emerging food technologies. Trends in Food Science & Technology. 2011;22(8):442-56.
112. Spencer S. The animal scientist in a changing society. Domestic Animal Endocrinology. 1999;17:95-100
113. Nuffield Council on Bioethics. Genome Editing: an ethical review. 2016
114. Frewer L, Lassen J, Kettlitz B, Scholderer J, Beekman V, Berdal KG. Societal aspects of genetically modified foods. Food Chem Toxicol. 2004;42(7):1181-93.
115. Frewer LJ. Consumer acceptance and rejection of emerging agrifood technologies and their applications. European Review of Agricultural Economics. 2017;44(4):683-704.
116. Bloomfield BP, Doolin B. Imagination and technoscientific innovations: governance of transgenic cows in New Zealand. Soc Stud Sci. 2011;41(1):59-83.
117. Pfister H-R, Böhm G, Jungermann H. The cognitive representation of genetic engineering: Knowledge and evaluations. New Genetics and Society. 2000;19(3):295-316.
118. Gaskell G. Biotechnology and the European public. Nature Biotechnology. 2000;18:935-938
119. McCullum C, Benbrook C, Knowles L, Roberts S, Schryver T. Application of Modern Biotechnology to Food and Agriculture: Food Systems Perspective. Journal of Nutrition Education and Behavior. 2003;35(6):319-32.
120. Uzogara S. The impact of genetic modification of human foods in the 21st century: a review. Biotechnology Advances. 2000;18:179-206
121. Wheeler SA. The barriers to further adoption of organic farming and genetic engineering in Australia: views of agricultural professionals and their information sources. Renewable Agriculture and Food Systems. 2008;23(2):161-70
122. Priest S. US public opinion divided over biotechnology? Nature biotechnology. 2000;18:939-942
123. Chen HY, Chern WS. Willingness to pay for GM foods: Results from a public survey in the USA. Evenson RE, Santaniello V editors. 6th International Conference of the International-Consortium-on-Agricultural-Biotechnology-Research. 2004. p. 117-29
124. Saher M, Lindeman M, Hursti UK. Attitudes towards genetically modified and organic foods. Appetite. 2006;46(3):324-31.
125. Malhotra N. Questionnaire design and development. In: Grover R, Vriens M editors. The Handbook of Marketing Research: Uses, Misuses, and Future Advances. California: Sage Publications; 2006. pp. 176-202
126. Napier TL, Tucker MA, Henry C, Yang X. Ethical orientations of Ohio residents toward genetically engineered plants and animals: An urban/rural comparison. Journal of Food Agriculture & Environment. 2004;2(2):400-11.

127. Frewer LJ, Howard C, Shepherd R. Public concerns in the United Kingdom about general and specific applications of genetic engineering: Risk, benefit, and ethics. Science Technology & Human Values. 1997;22(1):98-124.
128. Delhove J, Osenk I, Prichard I, Donnelley M. Public Acceptability of Gene Therapy and Gene Editing for Human Use: A Systematic Review. Hum Gene Ther. 2020;31(1-2):20-46.
129. Bai F, Ling J, Esoimeme G, Yao L, Wang M, Huang J, et al. A systematic review of questionnaires about patient's values and preferences in clinical practice guidelines. Patient Prefer Adherence. 2018;12:2309-23.
130. Halpern J, O'Hara SE, Doxzen KW, Witkowsky LB, Owen AL. Societal and Ethical Impacts of Germline Genome Editing: How Can We Secure Human Rights? CRISPR J. 2019;2(5):293-8.
131. Tait J, Banda G, Watkins A. Proportionate and Adaptive Governance of Innovative Technologies. Case Study: Responsible Governance of Innovative Technologies. 2017 Innogen Summary Report
132. Jasanoff S, Hurlbut JB. A global observatory for gene editing. Nature. 2018;555:435-437
133. Wirz CD, Scheufele DA, Brossard D. Societal Debates About Emerging Genetic Technologies: Toward a Science of Public Engagement. Environmental Communication. 2020;14(7):859-64.
134. Wintle BC, Boehm CR, Rhodes C, Molloy JC, Millett P, Adam L, et al. A transatlantic perspective on 20 emerging issues in biological engineering. Elife. 2017;6.
135. O'Grady C. Jury duty for global warming: citizen help solve the puzzle for climate action. Science, 2020, Oct 29 [Cited 2021 Jan 16]. Available from: https://www.sciencemag.org/news/2020/10/jury-duty-global-warming-citizen-groups-help-solve-puzzle-climate-action
136. Pereira ÂG, Völker T. Engaging With Citizens. In: Sucha V, Sinkiewicz M editors. Science for Policy Handbook. Elsevier: European Union; 2020. pp. 78-95
137. Montoliu L, Merchant J, Hirsch F, Abecassis M, Jouannet P, Baertschi B et al. ARRIGE Arrives: Toward the Responsible Use of Genome Editing. CRISPR J. 2018;1(2):128-129
138. Mulvihill JJ, Capps B, Joly Y, Lysaght T, Zwart HAE, Chadwick R, et al. Ethical issues of CRISPR technology and gene editing through the lens of solidarity. Br Med Bull. 2017;122(1):17-29.

*References included later on after reviewers' advice:
139. Haddaway NR, Collins AM, Coughlin D, Kirk S. 2015. "The Role of Google Scholar in Evidence Reviews and Its Applicability to Grey Literature Searching." *PLoS One.*

17;10(9):e0138237. doi: 10.1371/journal.pone.0138237. PMID: 26379270; PMCID: PMC4574933.

140. Piasecki J, Waligora M, Dranseika V. 2018. “Google Search as an Additional Source in Systematic Reviews.” Sci Eng Ethics 24(2):809-810. doi: 10.1007/s11948-017-0010-4. Epub 2017 Dec 16. PMID: 29249022; PMCID: PMC5876410.

Chapter 6. Book chapter. Biolab – uma proposta para o desenvolvimento de projectos em ambiente multidisciplinar

BioLab – uma proposta para o desenvolvimento de projetos em ambiente multidisciplinar

Pedro Dias Ramos(*)§,

Filipe Marques(**)§,

Maria Manuela Lopes(***),

Júlio Borlido Santos(****),

Fernando Tavares(*****),

I. Anna S. Olsson(******),

& Maria Strecht Almeida(*******)

(*)i3S – Instituto de Investigação e Inovação em Saúde, Universidade do Porto; ICBAS – Instituto de Ciências Biomédicas Abel Salazar, Universidade do Porto
(*)pedro.ramos@ibmc.up.pt

(**)i3S – Instituto de Investigação e Inovação em Saúde, Universidade do Porto; ICBAS – Instituto de Ciências Biomédicas Abel Salazar, Universidade do Porto
(**)filipe.marques@ibmc.up.pt

(***)i3S – Instituto de Investigação e Inovação em Saúde, Universidade do Porto; ESE.IPP – Escola Superior de Educação, Instituto Politécnico do Porto
(***)maria.lopes@i3s.up.pt

(****)i3S – Instituto de Investigação e Inovação em Saúde, Universidade do Porto
(****)borlido.santos@i3s.up.pt.pt

(*****)FCUP – Faculdade de Ciências, Universidade do Porto
(*****)ftavares@fc.up.pt

(******)i3S – Instituto de Investigação e Inovação em Saúde, Universidade do Porto; ICBAS – Instituto de Ciências Biomédicas Abel Salazar, Universidade do Porto
(******)olsson@ibmc.up.pt

(*******)ICBAS – Instituto de Ciências Biomédicas Abel Salazar, Universidade do Porto
(*******)msalmeida@icbas.up.pt

§ – estes autores contribuíram igualmente para o trabalho

Resumo

Este capítulo descreve a implementação da unidade curricular de inovação pedagógica *Biolaboratório – Projeto de Experimentação Multidisciplinar* (BioLab). A unidade de formação contínua, que teve a sua primeira edição em 2020-21, procura explorar cruzamentos ciência/arte/sociedade. Na prática, desenvolve-se como investigação integrada em unidade curricular e procura tirar partido do ambiente informal que existe em laboratórios comunitários.

Pelos projetos desenvolvidos e pelo entusiasmo de todos os envolvidos, e principalmente dos estudantes, considera-se uma iniciativa bem-sucedida.

Abstract

This chapter describes the implementation of the curricular unit of pedagogical innovation *Biolaboratory – Multidisciplinary Experimentation Project* (BioLab). The continuing training unit, launched in 2020-21, aims to explore science/arts/society intersections by applying course-based research experiences inspired by community laboratories. Awe-inspiring projects and positive feedback from all players involved, notably the students, suggest BioLab was a successful initiative.

Palavras-Chave: STEAM; CURE; laboratório comunitário; trabalho colaborativo.

Keywords: STEAM; CURE; community lab; collaborative work.

Introdução

Tem sido defendido que a educação científica deve focar-se hoje numa mudança de STEM (*Science, Technology, Engineering and Mathematics*) para STEAM, que acrescenta as artes a STEM e assim liga a ciência todas as outras disciplinas (European Commission 2015). A UC InovPed *Biolaboratório – Projeto de Experimentação Multidisciplinar* (BioLab) cuja implementação aqui se reporta foi concebida para um contexto STEAM, explorando cruzamentos ciência/arte/sociedade.

Enfatizar a ligação entre investigação e ensino proporcionando uma aprendizagem orientada para a investigação (*inquiry-based learning*) é hoje uma prática reconhecida internacionalmente (Fung 2017). A proposta de currículo conectado (*ibidem*) considera essa ligação como uma das suas vertentes. E a essa acrescenta a das ligações entre disciplinas e entre a academia e o mundo exterior. Na prática, o BioLab desenrola-se como um CURE (*Course-based Undergraduate Research Experience*), promovendo uma aprendizagem pela investigação (Bell, Eckdahl *et al*. 2016), proporcionando aos estudantes o desenvolvimento de um pequeno projeto que, neste caso, tem a particularidade de integrar contribuições a partir de diferentes áreas disciplinares. Nesta UC InovPed procurou

ainda integrar-se a informalidade (regulada) de um laboratório comunitário (Scheifele & Burkett 2016).

A educação STEAM visa trazer o "mundo real", com a sua complexidade, para a sala de aula, ligando diferentes currículos de uma forma que se relacionem tanto com o mundo como entre si. Neste sentido, os processos de pensamento crítico, e métodos artísticos e de design foram introduzidos num processo de aprendizagem ativa centrada no estudante esperando aumentar a motivação, a autoeficácia das aprendizagens e a capacidade de resolução de problemas. Assente na lógica apresentada por Martinez (2017), o sucesso da proposta de unidade curricular dependia do caráter multidisciplinar da equipa (tanto de docentes quanto de estudantes) em criar uma experiência de aprendizagem conjunta. Assim sendo, os papeis não estavam predefinidos à priori, havendo lugar a um ajuste permanente na função de cada um no decorrer do desenvolvimento do ambiente de aprendizagem.

No que se segue, a unidade curricular é apresentada em detalhe, incluindo a motivação para a sua criação, a proposta de inovação pedagógica que representa e como funcionou de facto na edição de 2020-21. Uma análise crítica da implementação do curso reflete sobre a prática, discutindo lições aprendidas em termos de desafios e oportunidades, tentando-se, finalmente, algumas notas conclusivas.

A unidade curricular Biolaboratório – Projeto de Experimentação Multidisciplinar

Motivação

Proporcionar uma experiência de investigação que integrasse alguma da informalidade dos laboratórios comunitários foi a ideia subjacente à proposta desta UC InovPed. A iniciativa desenvolveu-se a partir de alguns eventos prévios entre os quais se destaca um workshop sobre CRISPR *&* *Biohacking* com estudantes frequentando o 1.º ano dos programas doutorais BiotechHealth (ICBAS/FFUP), MCBiology (ICBAS/FCUP) e Neurociências (FMUP/ICBAS). O *workshop* que ocorreu em janeiro de 2020 pretendeu associar o tema da edição de genoma e da sua utilização por parte da comunidade de *biohacking* em exercícios de reflexão contando para isso com a elaboração de diferentes perguntas que pretendiam levar à discussão entre os estudantes. Debateram-se a utilização de microrganismos geneticamente modificados, a que classe de segurança estariam associados no caso do seu uso e ainda, quais os mecanismos de regulação e transparência necessários para que estes pudessem ser utilizados numa lógica de laboratório aberto ou *OpenBiolab*. Após 3 horas de discussão, foi ainda possível uma

participação remota de um *biohacker* convidado do espaço OLGA (Open Lab Graz Austria) num momento interativo que proporcionou aos estudantes a oportunidade de colocarem questões sobre a atividade desenvolvida pelo mesmo, bem como as condições em que o *hackerspace* se pode organizar. O sucesso do *workshop* que contou com a presença de quase todos os elementos docentes que mais tarde acabariam por integrar o projeto da UC InovPed evidenciou a questão que todos se colocavam: seria possível a fundação de um laboratório aberto de biologia, vulgo *OpenBiolab*, em contexto académico na nossa Universidade? E se sim, estariam estudantes de diferentes áreas do conhecimento abertos à sua criação e integração? O mote estava lançado e a candidatura da InovPed esboçou-se a partir desse momento acrescentando-se à base biológica da UC e à ideia de investigação integrada num curso (internacionalmente descrita como CURE) as componentes de responsabilidade social e expressão artística, bem como o diálogo entre ciência e sociedade na definição dos objetivos para a concretização da UC.

Proposta pedagógica

No BioLab proporciona-se uma aprendizagem que integra a aquisição de conhecimentos teóricos e práticos no decurso de um processo de experimentação científica, artística e social, desenvolvido num ambiente multidisciplinar, no sentido da resolução de um problema específico. Nesse âmbito, como se apresenta na ficha de unidade curricular institucional (https://s.up.pt/kw4l) podem considerar-se como objetivos;

- reconhecer a relevância das abordagens multidisciplinares na produção de conhecimento científico e no diálogo ciência-sociedade;
- promover capacidades de diálogo e colaboração em equipa multidisciplinar;
- desenvolver a capacidade de conceber, planear e implementar um trabalho interdisciplinar de projeto, promovendo o trabalho colaborativo que potencie a complementaridade de conhecimentos e competências;
- adquirir (e/ou aprofundar) competências de trabalho laboratorial no contexto das ciências da vida e das questões em estudo;
- reconhecer a contribuição de práticas artísticas na reflexão em torno de problemas associados às ciências da vida;
- estimular competências que promovam a comunicação do projeto de grupo perante diferentes públicos, pelos seus pares e outros agentes sociais.

A capacitação dos estudantes enquanto cidadãos dotados de autonomia, pensamento crítico, capacidade de inovação e resolução de problemas é proporcionada por uma experimentação consciente e engajada em problemas societais mobilizando conceitos e

ferramentas das diferentes áreas STEAM. Assim, de forma sucinta, os resultados de aprendizagem e competências que o BioLab propõe alcançar, segundo a já mencionada ficha de unidade curricular institucional, traduzem-se:

- na aquisição de novos conhecimentos, particularmente em áreas académicas fora da formação base de cada estudante, potenciando a complementaridade de conhecimentos individuais;
- no reconhecimento da importância de recrutar conhecimentos multidisciplinares, para identificar questões ou problemas atuais e de interesse transversal que possam ser abordados numa perspetiva biológica e promovendo um olhar de perspetivas de arte/humanidades e das ciências sociais sobre questões de biologia/biotecnologia;
- no reforço de competências de trabalho em grupo no âmbito do projeto, nomeadamente na obtenção de consensos sobre um fluxograma de trabalho que identifique e hierarquize tarefas, estabeleça marcas, aponte responsabilidades individuais, valorize sinergias dentro do grupo, permita diagnosticar constrangimentos, avaliar riscos e proponha soluções alternativas;
- num aumento de competências de comunicação do saber científico que valorize as novas tecnologias de informação e vá de encontro às exigências crescentes de uma sociedade digital;
- num aumento de capacidade de pensamento crítico e criatividade que a arte/humanidades e as ciências sociais podem trazer.

Para implementar o processo de trabalho de projeto pelos grupos, a metodologia baseou-se em diferentes etapas, conforme ilustra o diagrama da figura 1. Destacam-se três vetores principais: i) capacitação dos estudantes para trabalho de projeto multidisciplinar, através de aulas teórico-práticas (que em 2020-21 decorreram *online* dadas as restrições para contenção da pandemia COVID-19) com diferentes temáticas cobertas; ii) envolvimento dos estudantes num tema integrador, incluindo participação de *stakeholders* externos, uma chamada para ideias de cada estudante naquilo que poderia ser um provável projeto dentro do contexto Biolab, e uma chamada para grupos em que cada estudante teve a oportunidade de integrar os projetos com que mais se identificasse dentro dos selecionados pelos docentes após proposta de cada estudante; iii) ação, isto é, desenvolvimento dos projetos selecionados no contexto de prática laboratorial tutoreada, com apresentações regulares semanais para discussão de ideias, uma apresentação final de cada grupo sobre o seu projeto, incluindo quais os sucessos e insucessos bem como reflexão crítica, em formato de ensaio individual, da utilidade do seu projeto a nível científico, artístico e/ou social.

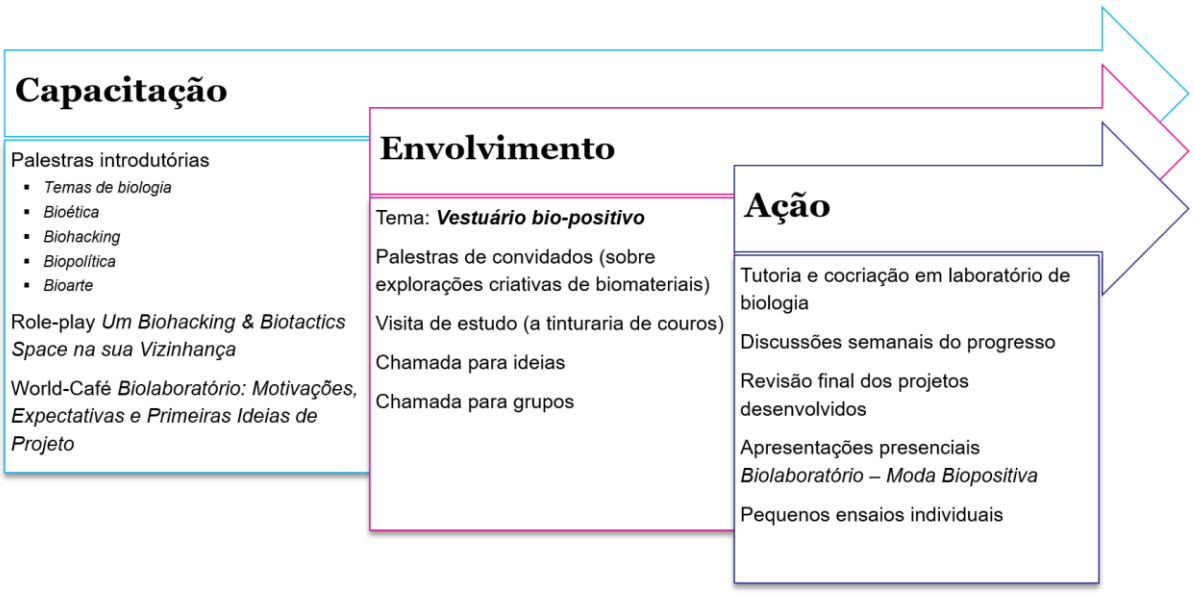


Figura 1. Vetores pedagógicos da unidade curricular BioLab com vista ao desenvolvimento dos projetos pelos grupos de estudantes

As componentes de avaliação dos estudantes incluem a participação presencial, um trabalho escrito individual bem como o trabalho de projeto, de grupo, e sua apresentação.

Edição de 2020-21

Na sua primeira edição, o BioLab captou o interesse de treze estudantes. Ainda que a alguns estudantes tenham selecionado o Biolab como Opção UP no âmbito do seu ciclo de estudos, a maior parte fê-lo fora desse âmbito. Em termos de distribuição por ciclos de estudo, a grande maioria frequentava uma licenciatura (9), com dois estudantes a frequentarem cursos de pós-graduação (1 mestrado, 1 doutoramento) na U.Porto. A quase totalidade dos estudantes (12) frequentava ou tinha frequentado um ciclo de estudos na área das ciências da vida, contribuindo assim para uma certa homogeneidade do grupo.

"Vestuário bio-positivo" foi o tema proposto aos estudantes, tendo o desafio sido colocado em termos de explorar possíveis alterações nos modelos de fabrico de vestuário evitando processos poluentes, que esgotem recursos naturais ou impliquem sacrifício de animais para a sua produção. Os estudantes organizaram-se em quatro grupos para desenvolver quatro projetos colaborativos que os próprios desenharam no BioLab em resposta ao tema proposto. Estes projetos focaram, designadamente: i) produção de celulose bacteriana, na perspetiva de obtenção de matéria-prima mais sustentável do que o algodão; ii) biodegradação de pigmentos de origem biológica potencialmente utilizáveis em vestuário, com vista à redução da poluição causada pela indústria têxtil; iii) produção

de biopeles/biocouros com base em resíduos ou excedentes alimentares frutícolas, procurando a utilização de subprodutos em matéria-prima para a indústria de moda; iv) sensibilização para a produção e uso de bioplásticos, incorporando a sua utilização em iniciativas de consciencialização sobre o impacto ambiental da moda.

Lições aprendidas

A experiência do BioLab foi, em grande medida, uma aventura em que todos os elementos da equipa docente embarcaram com entusiasmo. O trabalho colaborativo, de projeto e com contribuição de diferentes áreas do conhecimento constituiu simultaneamente um estímulo e um desafio. Na perceção dos docentes, foi assim também para os estudantes e isso faz com que esta seja considerada uma experiência bem-sucedida.

Mas vejamos em maior detalhe o que podemos retirar da experiência. Para pensar as lições aprendidas na sua globalidade, haveria que considerar três fontes principais de informação acerca da pertinência da abordagem pedagógica: os estudantes, outras pessoas que colaboraram no curso e ainda *stakeholders* nos projetos desenvolvidos. Centremo-nos em primeiro lugar nos estudantes. Fez-se uma avaliação da primeira edição do BioLab através de um inquérito por questionário implementado após a conclusão do curso. O estudo obteve parecer favorável da Comissão de Ética CHUPorto/ICBAS (referência do projeto: 2021/CE/P28(P368/CETI/ICBAS)). Uma análise detalhada dos resultados desse questionário foi incluída no artigo submetido às Atas do *7.º Congresso Nacional de Práticas Pedagógicas no Ensino Superior*, referente à comunicação que tinha sido apresentada na reunião (Marques, Ramos *et al*. 2021). As questões propostas aos estudantes incidiram sobre a experiência subjetiva dos atributos e forma do curso, a classificação da importância relativa das competências prévias e das componentes pedagógicas do curso, e a probabilidade de recomendação do curso. Sete dos treze estudantes responderam ao questionário. Os estudantes sublinharam o carácter multidisciplinar, prático e criativo do curso, o qual evoluiu num sentido que consideraram ser de aproximação a um espaço de experimentação autónoma. De entre as competências prévias sugeridas, os estudantes consideraram muito importante a capacidade de trabalho em grupo e menos importantes, quer o contacto prévio com fundamentos de química e biologia, quer a destreza laboratorial. A este respeito, a instrução entre pares foi apontada como fator mitigante das disparidades de preparação teórica e técnica entre estudantes. Em termos das componentes do curso, os estudantes avaliaram como muito importantes, entre outras, a assistência dos docentes no desenvolvimento dos projetos e o acesso a um

laboratório de biologia para realização de experiências. Por outro lado, os estudantes consideraram que haveria margem para melhorar as palestras sobre abordagens multidisciplinares, a discussão semanal do progresso dos projetos e a escolha da abordagem ao tema proposto. Pode ainda fazer-se referência a outras impressões positivas recolhidas informalmente. Entre as pessoas que externamente colaboraram no curso, contaram-se duas designers (uma nacional e outra internacional) que colaboraram pela apresentação em seminário breve do seu trabalho relacionado com o tema "Vestuário bio-positivo", bem como um representante de empresa de tinturaria do Grande Porto que proporcionou uma visita de estudo às instalações da empresa e que participou no momento das apresentações dos projetos finais. De alguma forma, este último é um *stakeholder* nos projetos desenvolvidos no curso que se mostrou entusiasta e com interesse em desenvolver colaboração.

Em 2020-21, pelas restrições da pandemia, não foi possível concretizar a planeada exposição pública final dos projetos desenvolvidos no curso. Já após a conclusão do curso, tendo sido lançado o repto para participação na *Noite Europeia dos Investigadores*, no i3S, em setembro de 2021, nove estudantes mobilizaram-se para ajudar a dinamizar uma pequena banca de demonstração dos projetos dos seus grupos. O sucesso da representação do BioLab neste evento ilustra bem a qualidade dos resultados do curso, sendo que a participação voluntária dos estudantes após conclusão do curso também parece indicar que a experiência terá sido positiva.

Considerações finais

O Biolab apresentou-se como unidade curricular formal pioneira na abordagem multidisciplinar das ciências da vida na U.Porto. Com uma abrangência no conjunto alargado das áreas STEAM, o BioLab procurou transpor as (boas) práticas de experimentação em laboratórios comunitários para um ambiente académico, multidisciplinar, de investigação baseada em projetos de índole científica e artística, respondendo a preocupações societais. O BioLab envolveu um conjunto alargado de docentes, estudantes, técnicos e convidados, representando uma diversidade considerável de áreas do conhecimento, que cocriaram o próprio processo pedagógico, através das contribuições no sentido de uma capacitação, envolvimento ou desenvolvimento efetivo dos projetos.

A elevada satisfação de todos os intervenientes, quer estudantes quer docentes leva-nos a considerar que o BioLab é uma experiência bem-sucedida. Apesar dessa perceção, reconhece-se que existe espaço para evoluir, nomeadamente através da integração de

estudantes de áreas do conhecimento ainda mais diversas além das ciências da vida, num plano multidisciplinar, e o desejável envolvimento de um conjunto mais alargado de possíveis *stakeholders* no contexto do Biolab. A exploração científica e artística de temas com impacto social continuará a ser o *motto* do Biolab. Esta UC InovPed foi essencialmente uma experiência de cocriação entre estudantes e docentes, carácter que se pretende enfatizar em próximas edições.

Agradecimentos. Os autores agradecem a Carla Oliveira (ICBAS) o apoio técnico na edição 2020-21.

Referências

Bell, J. K., Eckdahl, T. T., Hecht, D. A., Killion, P. J., Latzer, J., Mans, T. L., . . . Bell, J. E. (2016). CUREs in biochemistry – where we are and where we should go. *Biochemistry and Molecular Biology Education*, 45(1), 7-12. https://doi.org/10.1002/bmb.20989

European Commission. (2015). *Science education for responsible citizenship: report to the European Commission of the expert group on science education*. Publications Office. https://doi.org/doi/10.2777/13004

Fung, D. (2017). *A connected curriculum for higher education*. UCL Press. https://doi.org/10.14324/111.9781911576358

Marques, F., Ramos, P. D., Lopes, M. M., Santos, J. B., Tavares, F., Olsson, I. A. S., & Almeida, M. S. (2021). *BioLab – um espaço de experimentação e cruzamento disciplinar [Resumo de apresentação em conferência]* 7.º Congresso Nacional de Práticas Pedagógicas no Ensino Superior, Aveiro. https://cnappes.org/files/2021/08/Livro_Resumos_CNaPPES.pdf

Martinez, J. E. (2017). *The Search for Method in STEAM Education*. Palgrave Macmillan, Cham. https://doi.org/10.1007/978-3-319-55822

Scheifele, L. Z., & Burkett, T. (2016). The first three years of a community lab: lessons learned and ways forward. *Journal of Microbiology & Biology Education*, 17(1), 81-85. https://doi.org/10.1128/jmbe.v17i1.1013

Chapter 7. Discussion

In this chapter, I will discuss the main outcomes of the studies presented in manuscripts 1 to 3 and how these articulate with the current literature. RRI frames the project here reported and will be emphasized in the discussion since it is transversal to all the manuscripts included studies. Moreover, there will be space to discuss the accomplishment and redefinition of the objectives of this thesis. Furthermore, a critique to the methodology used in the studies and how it relates with chapter 2 from the thesis will also be explored. Finally, the details of additional outputs of this PhD project will be briefly expanded in this chapter.

Researchers and citizens are stakeholders who think about the present and the future of GE.

The research performed within this PhD project – looking at both expert views and public acceptance of the technology – aimed at answering to the new "old" questions now being raised by access to the so-called "genetic scissors" that are part of the GE technology that has been dubbed one of the breakthroughs in molecular biology. Technological breakthrough in biology makes researchers relevant stakeholders in addressing its innovativeness and applications that will be used for the good of society. We are still on the early steps of genome editing development with clinical trials happening for the first time in 2016 and with many others already planned for the upcoming years (Frangoul et al 2021, Stadtmauer 2020). In this sense, innovativeness in this field of expertise reaches scientific news media in bulk. How to make sense of this and to gather the most conciliated view of what such innovativeness might mean not just in scientific terms or activities but also what it may mean for citizens worldwide, is perhaps the insightful way of predicting the harm-risk/benefit of a technology. This becomes important because such innovative technology may protrude in our lives albeit not yet in a completely defined form due to unregulated premises. For how long, however, is unclear … Advancements during the last ten years also took place with many (bio)ethicists, policy-makers and governmental agents depicting regulatory and legislative boundaries to the use of genome editing technology (Bangkok Statement 2019, Danish Council 2016, EGE 2021, Nuffield 2016). What is definitely true is the time that we still have as citizens to be informed about its plausible use in our lives and if the logic of RRI to enact a responsiveness dimension in having the most responsible technology as possible (von Schomberg, 2013). Broad stakeholder engagement has been one of the most frequent requests from scientific community (Bangkok 2019, Rosemann 2019) and within this thesis we wanted to be as inclusive as possible to gather input from these.

STS studies aim at discussing sociological and oftentimes ethical dimensions of scientific and technological developments (Konrad et al, 2016). In this PhD project, RRI is therefore

our core approach and it is inserted in a logic of division moral labour (Rip 2018) when aggregating the view of the different actors that participated in our interviews. With this in mind, anticipation has been the main dimension of RRI to be explored combined with the reflexive and construal level of "collective imaginations" by a number of stakeholders that are intra-diverse in their own level of expertise. Methodologically, the work in manuscript 1 of this thesis is developed in line with previous qualitative research e.g Pereira et al 2015 whereas in the case of citizens' opinion detailed in manuscript 2, it relates to work of (Clayton et al 2018, Clark et al 2016, Sawai et al 2017) but with the focus in genome editing as an emergent technology in the 21st century.

Redefinition of objectives of the PhD project and RRI as a real approach to genome editing

Before we delve into the real questions (this has been the main sentence of my interviews before starting the conversation with interviewees), it is important to focus on the main goals of the PhD project, also in order to consider how they changed along the way as ideas gained clarity and there was a shift in focus and finally to reflect on how well I succeeded in meeting them. As previously mentioned, the two first objectives of this PhD project were to:

- Investigate the predicted harm-risk/benefit of GE;

- Investigate the public acceptability of GE.

Two groups of experts were interviewed. As the group of stakeholders that most likely would elucidate about the technology in itself, we interviewed researchers that are familiar with and use the technology. The second group of experts were policy-makers and ethicists. Here ethicists, bioethicists, researchers with active participation in ethical committees and researchers involved in a regulatory and legislative agenda were selected. The question about public acceptability towards genome editing technology could have been addressed empirically, but for reasons that will be discussed later, we approached this question through an extensive and exhaustive systematic review of surveys public attitudes towards genetic modification of humans and animals in two periods: before (pre-CRISPR) and after the advent of CRISPR (CRISPR period).

As outlined in the previous section, the two main objectives of the thesis were fulfilled, and although some work is currently under analysis, the three studies already generated much information. Two additional objectives were established during the course of analysis and discussion. The first was to expand the predicted harm-risk/benefit in a clearly defined dimension of responsibility for the implementation of genome editing. The second objective was to explore how to prepare future scientists to act in the context of RRI either on their

ongoing projects or when submitting new research projects to be funded as well as in pedagogical or educational environments where university students may prepare themselves to be future researchers. Both new objectives have a RRI framework behind but they unfolded differently throughout time. The first objective is research-driven and intends to elucidate on what stakeholders from different backgrounds really think about genome editing technology and, in the end, will tell what they understand as the most and least relevant and the most and least worrying features of it. Here, three different concepts were explored (Appendix I). Firstly, revisiting the present and future of the technology regarding technical challenges for biomedical and animal science researchers and ethical, regulatory challenges for (bio)ethicists and policy-makers. Secondly, feasible applications of genome editing where germline and somatic genome editing and human and animal perspective were sub-dimensions common to both interview guides done with the two group of stakeholders. Some different sub-dimensions were intended to be approached meanwhile since for biomedical and animal science researchers, more weight was put into technical compared with ethical concerns compared to (bio)ethicists which had more weight put on the latter. For these stakeholders, a sub-dimension concerning current regulatory and legislation guidelines has been added. Thirdly, GE in animal research was a strong focus of our interviews which suffered some adaptation from the interviews with the first group of stakeholders to the second one (Appendix I) demonstrates. Finally, and as already mentioned in the beginning of this discussion, throughout time, we developed more and more (based on the first interviews) the idea of responsibility in the context of genome editing. This led us to include a fourth concept in our interviews with (bio)ethicists and policy-makers, covering policies and means of broadening societal engagement, public opinion about genome editing technology and sub-dimensions related with social justice, ownership and accessibility to this technology. Here, it is clear the turnaround that the first interviews with biomedical and animal researchers provided since it helped developing different concepts for the second group of stakeholders that would be interviewed in a second phase. Reinforcing the involvement of stakeholders like scientists and policy-makers in framing the problems, questioning the questions and legitimating policies is very relevant to help drafting guidelines that may be essential for the application of scientific or technological revolutionary tools for society (Sinkiewicz 2020). While keeping the idea of comparison between groups is important, it is equally relevant to see the research unfolding into new questions due to the analysis of a group of stakeholders that are maybe in a privileged situation to discuss them. This intertwines greatly with our work performed in the systematic review of questionnaires measuring people's opinion about genetic engineering and genetic modification technologies, and of which genome editing is part since 2013. Importantly, considering the need to close the gap between research-based activities and the public

(e.g.Topp et al 2020), the idea of responsibility that has to be shared with citizens in order for them to have a participatory agenda regarding emergent technologies fits with what is already proposed by many authors in STS field (de Saille 2015, Jasanoff 2005, Einsieddel 2008) where genome editing is one that already shares this concern (Dryzek 2020, Wirz, Scheufele and Brossard 2020, Reincke, Bredenoord and van Mil, 2020, Scheufele 2021). By having all this in account, it would be easier to shift towards more governable genome editing for present and future generations (Rip 2018, Hurlbut 2018).

My work towards the second new objective took a more practical format, inspired by the NewHoRRIzon project and the idea of creating social labs as spaces for the discussion of different stakeholders in a logic of a series of workshops (Timmermans et al 2020). By embedding RRI in research projects of future researchers, one is striving for anticipative methodology about new scientific and technological developments like emergent technologies. I explore this later in this chapter when discussing the RRI toolkit from the PhD project. Additionally, an initiative with a pedagogical and educational character - the BioLab (manuscript 3) - feature as another approach that may better prepare university students that will later be researchers and incorporate the responsibility dimension in research and innovation projects where they will work on.

Main discussion points

Reaching to this point, let me focus on the most important conclusions of the study and discuss them in the light of the evolution of the technology in the last 10 years either in relation to technical, (bio)ethical, regulatory/legislative and public opinion and how are we in matters of responsibility and stabilization of this technology. Additionally, I will provide an overview of the most and least relevant priorities of the technology according with stakeholders' point-of-view and based in their arguments and finally, a general advice about the use of genome editing if this reaches decision-makers and agents of change like governmental entities. For this to happen, first and foremost, I will divide the conclusions into three overarching themes which constitute the pillars of the narrative embodied by stakeholders. I will then discriminate the themes that are explored in the manuscripts of this PhD thesis. Secondly, a critique to the methodology used during both studies will be presented. In the end, there will be space to also discuss guideline for responsible genome edting, the toolkit for researchers to address genome editing emergent technologiy and the BioLab

Research questions and overarching themes

In order to effectively infer about the perceptions of stakeholders regarding genome editing as a predominant emergent biotechnology of the twentieth-first century, our semi-structured interviews aimed at discovering themes that could answer our research questions. Throughout the process of building the matrix that would form the basis for our interview guide, we developed two main research questions:

a) What do researchers envision as the current and future challenges, concerns and potential of GE technology?

a.1) Which GE applications are more realistic and desirable in the near future for humans and for animals?

b) In the eventuality of surpassing all the challenges posed by the technology, what are the lines that need to be drawn in order for it to be implemented responsibly?

In the following, I will discuss the findings with regards to these research questions.

a) What do researchers envision as the current and future challenges, concerns and potential of GE technology?

Firstly, according with researchers in the field, one may already say that GE technology technology has many gaps to close before it will be implemented in clinical settings. Researchers encounter many scientific obstacles to the progression of this technology due to the level of uncertainty and randomness that it may bring to the humans that will be subjected to such an intervention. Eventually, this will translate into problems related with off-target effects in the genome and lack of understanding of our genome as a complex regulatory frame where multiple genes and intronic regions of the genome are still undefined in their own function. This is elucidated by biomedical researchers and reinforced by (bio)ethicists (unpublished data) because they foresee that until this challenge is solved, research has to continue but clinics have to wait for treatments based in this technology. However, this is not entirely immutable in their point-of-view, even currently, because there are already several proven genome editing platforms that show very promising results to be included in clinical settings and that are specific to very defined organs of the human body. Here, biomedical researchers include mainly the eye and the skin as organs of particular interest and feasibility and which may soon be done with the help of another rapidly developing technology named cell-assisted reproductive technology or CAR-T cell technology (Cathomen 2018, Mussolino et al 2018). When indicating the different genetic therapies that may also benefit from the symbiosis of these technologies, monogenic

diseases (not defined if dominant or recessive) often appear as those considered most realistic and desirable. Cancer however, falls short compared to genetic diseases because this type of disease has already other type of treatments that demonstrate a higher efficacy and a stronger reliability than emergent genetic technologies like chemotherapy, radiotherapy and coadjuvant drugs (Liu et al 2021). Even so, researchers find it desirable nonetheless but they agree that more defined cancers such as leukemia might be more suitable to genetic treatments based in GE technology than others. Somatic genetic intervention is also evoked by researchers as something that may be used at any point in an individual's life despite being technically more difficult but still controllable whereas germline genome editing raises many other questions about this and researchers consider its use in humans difficult to justify. Moreover, somatic genetic intervention still fits with everything mentioned before by researchers regarding potentialities and technical challenges of genome editing and it is said to be sought based on these premises before clinical or medical applications take place while germline genetic intervention does not fit into this discourse.

Another challenge that is mostly a barrier for progression of GE into the clinics relates with the efficiency of GE technology. Here, two main problems are identified: the aspect of achieving a threshold that can be considered as enough to be used in humans and the aspect of when to use it in regards to the lifetime of a human being. The interviewed researchers stress as well that efficiency means different things regarding different scenarios where patients are included. They would prefer to improve the technology before this would reach the clinical settings but some consider this to be just enough for some cases. We start already to see the balance of harm-risk/benefit assessment and the decision to proceed or not to a clinical setting based in the need to achieve high levels of correction of genetic disorders and in the most predictable way possible with minimal randomness possible (Araki and Ishii 2016, El-Enein et al 2017). Here, a confounding factor of ethical order pops up to influence what may be considered significant or not and the discourse of how enough is enough to proceed gains momentum. Limiting the age and the condition of the patient to decide whether to apply certain gene correction therapy based in genome editing is therefore the criteria that will still lead to the decision as it is already done in clinical trials of this magnitude (El-Enein et al 2017). Therefore, the level of efficiency of the technology is still not the criterion to define if one should or not proceed for treatment but rather to say that if the patient really needs it and checks certain aspects, it may be considered as valid to be exposed to a genetic intervention that it is still in its learning curve. Researchers are also very clear in what they expect since they are aware of the limitations, potential and usefulness of the technology. While they acknowledge that the technology is

not stabilized due to the constant progress and breakthroughs that is experiencing as well as due to its emergent character, researchers claim the responsibility of continuing the research without restrictions to their own work unless irregularities as the ones already mentioned are taking place.

Taking the results above into considerations, it is very clear that researchers are concerned mostly with the safety of the technology. Safety can be based in how much we can correct in terms of disease and simultaneously how much damage we may induce in the genome while doing it due to the uncertainties that this type of genetic intervention supposes (El-Enein et al 2017, Araki and Ishii 2016, Howard et al 2018). Safety preconizes the fundamental criteria to say that the technology is ready to be used and humans will not suffer from its use when it comes the time to adopt it for disease-related reasons. For researchers, it is clear that until the technology is proven to be safe or safe enough, genome editing should stay in research grounds and not be disseminated in clinical settings. The ambivalence of a "safe enough criteria" has been already discussed by bioethical and legal scholars (EGE 2021, NASEM 2017, Nordberg et al 2018). This notion of safety is directly related with the responsibility that dominates researchers' words and this falls once more in the domain of "technologies of humility" present in Jasanoff's work where researchers in fact recognize that the technology will be useful, will be needed but it is not ready for society to adopt it yet unless some more stakeholders are brought to foresight exercising about it (Jasanoff 2003). This will be explored thoroughly during the second research question.

This intertwining between technical observations of researchers and ethical concerns that are described by them and citizens when inquired about somatic and germline genome editing were extensively continued for more applications in somatic and germline topics during interviews and extended to applications that would reach non-human species.

a.2) Which genome editing applications are more realistic and desirable in the near future for humans and for animals?

While it wasn't an objective of our research since the beginning, the recurrent overlap between what is realistic and what is desirable by people that have been interviewed assured us that putting the "R" into the "RI" is pertinent. RRI while not present in the speech

of researchers due to a likely unawareness to the concept is in fact dominating the ranking exercises in terms of their argumentation because they are anticipating and imagining scenarios if safety is ever reached at some point. Curiously, it is now 5 years from the first interviews that took place. If we now look into what has been happening since then and what they mentioned, we find a concordance although some of the applications are still on their own infancy of appearance or still to happen. In order to present their overview, I will divide these based in safety, responsibility and purpose criteria.

The safe, purposeful and responsible applications are all that are meant to correct diseases (mostly genetic ones) and to improve health in patients. Primarily, this comprises all diseases that may be gene corrected as well as the ones considered to be safer when involving monogenic diseases. Clinical trials, for example, started to happen for some genetic diseases identified as easier to perform genome editing on and the first preliminary results came to light recently (Frangoul 2016, Stadtmauer 2020, Ref). An application that is seen as technological complicated but when solved will definitely bring the necessary safety and health status to patients that are in need is the transplantation of genome-edited organs where almost all researchers conclude to be fundamental to improve health status. Equally important are also applications that involve promoting health in individuals through food improvement and here applications where genome-edited organisms such as crops will lead to better and more food for an overpopulated world are in the point-of-view of researchers what brings more good to society. Here, a regulatory differentiation happened during 2018 with the EU Court of Justice considering genetically-modified organisms (GMOs) under the same regulation premises as the ones obtained through GE (EU Court 2018). The mentality of responsibility prevails greatly and here scientists consider themselves the ones that should solve technicalities in order for these applications to become a reality and therefore desirable.

They also consider some applications as unsafe and irresponsible such as the birth of genome-edited human embryo. Here, their estimates were flawed since the birth of the in genome-edited *in vitro* human embryo by the hands of He Jiankui (Regalado 2018) was announced halfway through the study. The surprise for this event could hardly have been bigger and since there had been no scientific publication of the work so far, this led researchers to stay skeptical about whether this had really happened, in parallel with the fierce criticism of the overall scientific community (Greely 2019). Researchers, on the other hand recognize that for monogenetic disease particularly autosomal dominant ones it may be valid to use germline gene editing and it may bring more benefits than risks but they discard other diseases. Curiously, they are concerned with benefits and risks equally presenting a frequent narrative: “We don’t need it” or “We don’t need to do it”. Maybe the

fact that researchers consider themselves to work responsibly and consider it unthinkable (Martin and Turkmendag 2021) to transgress the lines of ethical reason which for them are sacred led interviewees to misjudge this of happening so soon because of their belief that the scientific community controls the research work being carried worldwide. The most worrying questions were of scientific and ethical nature and some questions laid out by the scientific community were the same as posed by researchers in interviews: why do it particularly for prevention of disease? And why do it secretly, under the radar of the scientific community? (Meyer 2022). Besides the unnecessity of performing GE in cases where HIV will likely have less than 1% chance of being passed to the newborn baby, we now know that this wasn't efficiently achieved since one of the twins had correction in only one of the genes (Greely 2019). Here too, citizens manifest a somewhat different opinion since they are supportive of these applications based on the purpose of correcting serious disease and preventing life-threatening diseases (manuscript 2). Researchers, on the other hand, speak about unnecessity and unknown effects of implementing the technology for germline genome editing and they do it because they also recognize (not all, but some) that scientifically speaking, it is easier to apply genome editing in a germline cell (embryo, sperm or egg) than it is to apply in a somatic cell that is differentiated and corresponds to a certain tissue in the human body. This situation resembles a conflict between what is easier to do it and what is harder to decide. According to researchers, it is easier to use genome editing technology in human germline but the questions are harder to answer when one does it.

Other applications considered to be unsafe and irresponsible are related with economic gain, eccentricity and individuals' preferences. Although recognized as "technically doable" in manuscript 1, many consider applications in private stances to be unnecessary and a lack of humility is raised due to the human egocentric character. These are usually applications that involve mainly the editing of non-human species that are usually owned by humans and where there are already accountable measures that may be reinforced if these applications turn out to become a reality, i.e. GE horses for sports or GE pigs to be domesticated.

Finally, applications that may be purposeful but still raise safety concerns, we considersemi-responsible and therefore object of more caution on the side of researchers. Gene drive insects for vector-borne diseases like malaria are one of these because they expect health improvement of populations but but biosafety issues for non-human species present in the ecosystems where they are released are seen as possible. Some experiences using gene drives with animals in the wild for insect species such as *Aedes aegypti* for Malaria disease with the first trials happening already in countries outside of EU regulatory premises (Camporesi and Cavaliere 2016, Scudellari 2019). From the citizens that were inquired,

there are different views about this because they usually separate the release of the species from the health impact. So, citizens that are concerned with health, approve this, but citizens that are concerned with nature, usually reject it. Other equally questionable applications involve research experiments with farm animals due to the harm inflicted to animals for the human benefit. This is a line of research already investigated over many years in terms of human consumption and the utilitarian view of animals (Frewer et al 2011, Frewer et al 2003, Schuppli and Weary 2010, MacNaghten 2004).

Citizens are also more supportive of GE animals when these have a direct medical purpose like vaccines, medicines or transplants when compared with food purposes where they usually weigh more the harm done to the animal. There are already works in this sense to obtain viable organs that may avoid rejection in patients (Boeke 2016). Researchers consider GE non-human primate as purposeful but unsafe. According with last reports, there has been an increase in the use of certain species of animals in research following the GE advent, specifically fish and non-human primates, whereas other species decreased their numbers like rodents and rabbits (EU report 2019). This behavior is observed by researchers as understandable due to the revolutionary and democratized access to the technology that has been adopted by laboratories and companies worldwide. Again, the sense of responsibility prevails when judging the control of these situations but maybe because of the proximity with their own research lines of work, not all researchers recognize if the welfare of these animals is in fact improving or decreasing and illustrating scientific impact over discoveries that are being carried worldwide (unpublished data). Such a duality of concern versus in-depth unquestioning of reality corresponds to an ethical dilemma that researchers often face where their desire to contribute for treating diseases clashes with the induced negative impact in animals once modelling disease (de Graeff et al 2019, MacNaghten 2016). As for citizens, they are usually more receptive to health products derived from GE animals rather than food and so in a way, they emphasize the more biomedical purpose of laboratory animals have compared than farm animals for food production purposes. On the other hand, researchers don't find any value in performing genome editing in human cells to be later transferred for animals. Researchers speak about the social dimension of animals that is reduced compared with humans evoking the speciesism argument and the hierarchical order of species (Olsson and Sandøe 2010, Olsson et al 2016, Kramer and Meijboom 2021, MacNaghten 2016). Fictional scenarios are resources used during collective imaginations or exercises and demonstrate the relationship between skepticism and optimistic or pessimistic feelings from citizens towards their notion of realism (Eichmeier 2023, So, Sladek and Joly 2021). Overall, these disturbing

scenarios are considered unrealistic and far out to reach in the future and with a long timeline ahead, in case it happens (20 years).

The idea of responsibility drives their own rationale and this modulates their own expectations for the technology to be adopted in lives of citizens. Technological advance also drives which application will come first, but the ethical reasoning driven by responsibility prevails during their reflection. We are also confronted with Jasanoff's work based in the "technologies of humility" since researchers recognize the strengths and weaknesses of the technology, but they also claim the responsibility to solve those problems to make the technology available for the world with the aim to promote the good for society (Jasanoff 2003). This drove their argumentation and allowed to generate a figure that represents the different dimensions and criteria that is used to draw the line about certain applications (Fig.7.1).

This brings us, then, to the second research question and it will be answered based in the main overarching theme that is divided in the two other overarching themes: human health and biosafety.

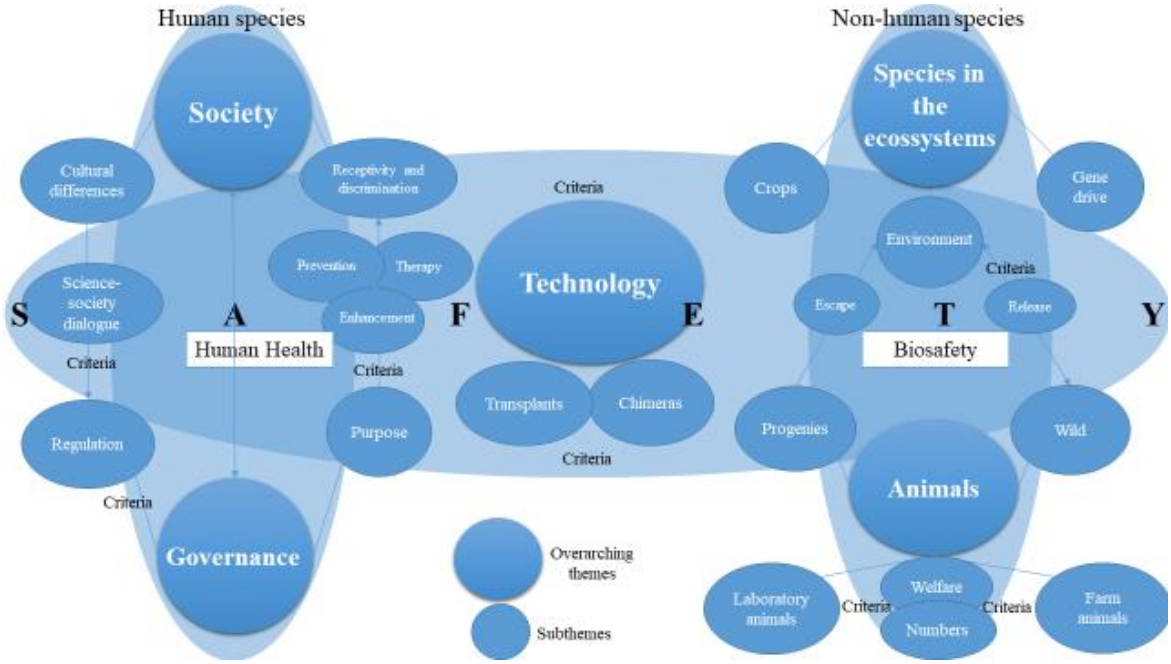


Figure 1. Overarching and subthemes discovered from thematic analysis of interviews with biomedical and animal science researchers. Safety is the main overarching theme that unfolds in three other lower overarching themes: technology, human health and biosafety. Technology aggregates overlapping issues in human and non-human species that are important for human health and biosafety, respectively. Human health is subdivided in two axes – society and governance – whereas biosafety is subdivided in species in the ecosystems and animals.

b) In the eventuality of surpassing all the challenges posed by the technology, what are the lines that need to be drawn in order for it to be implemented responsibly?

Main overarching theme: Safety

When it comes to the main argumentation of biomedical and animal science researchers regarding the lines needed to be drawn for genome editing in human and non-human species, there are two dimensions of safety that need to be attended as well: safety for human health and biosafety. For human health, there are a number of criteria that interplay to accept the technology as likely being adopted in the future and these are mainly related with the purpose of its use and establishing the limits between prevention, therapy and enhancement (figure 7.1). Many authors in scientific journals and ethics cabinets from non-governmental bodies clearly established the boundaries that exist between these (EASAC 2017, EGE 2021, Nuffield 2016, NASEM 2017, Polcz and Lewis 2017). The European Group on Ethics (EGE) opinion illustrates very coherently the difference between therapy, prevention and enhancement in its definitions and how this related with scenarios of disease and no-disease. While therapy is attributed to somatic genome editing in order to treat an existing disease, prevention and enhancement blur the line between the intervention stage – germline or somatic – and the purpose – avoiding disease and improving a 'normal feature or function' (EGE 2021). Disease and enhancement are clearly defined if the purposes are different, for example, in the cases between treating a genetic disorder versus improving genetic conditions related with appearance; this changes considerably if one is discussing life-limiting or non-limiting life conditions such as in the case between correcting blindness or improving eyesight. Here, the lines are blurred but overall, researchers disagree with the latter, supporting the first. However, if one is now considering preventing disease, a criterion of susceptibility appears because one is now in the domain of the unknown and likelihood of experiencing a certain disease. Here, researchers show more uncertainty about their own opinion but express support for cases where there is a patient rather than an individual that may develop a condition likely waiting for them to develop it in the initial stage to start treatment and not acting before that confirmation. This is once again related with the fact that one may induce unwanted off-targets elsewhere in the genome when trying to prevent which may be avoidable but also in the benefit that it may bring to the individual that may experience some type of enhancement or dis-enhancement (Juegst et al 2018, Almeida and Ranisch 2022). Enhancement is not straightforward for most traits when compared with interventions in germline for the purposes of prevention as mentioned previously for

autosomal, monogenetic and inherited diseases (Gyngell et al 2017, Juengst et al 2018). Nevertheless, enhancement for germline genome editing deserved reflections from authors due to the weighing against Pre-Implantation Diagnosis (PGD) that still do not foresee a consensual opinion (Almeida and Ranisch 2022). Researchers in the interviews are much more willing to maintain PGD as the gold-standard method of approaching avoidance of disease in unborn individuals despite the selective characteristic that it possesses when compared with the corrective one from genome editing. Moreover, fears of eugenics and designer babies are serious concerns mentioned in the literature (Bosley 2015, Danish Council 2016, Lander 2019) but quite limitedly expressed by researchers. It has been emphasized that the role of public perception and attitudes, highlighting that enhancement is recognized to be less acceptable for *'most people'* and it is also pointed that dis-enhancement can be a concerned for the *'wider public'* (EGE 2021). Once again, the purpose of disease is justified but the criteria of uncertainty or susceptibility influences decision and both factors are fundamental to the relevant axis of governance and society.

Citizens are very clear in their opinion about genome editing. This does not mean that there is no ambivalence in their approval, but it is clear what their priorities are to apply the technology. Citizens worldwide that were asked about genome editing and genome modification of human adults for disease-related scenarios give an approval always above 60% of the total number of inquired. This counterbalances with situations of enhancement of human adults where citizens reach a maximum of 50% approval worldwide. If we now look to the germline settings, one is able to see the same pattern, but a lower approval for disease-related purposes and influenced by disease type. Others more optimistic, approve enhancement for characteristics and not disease. There is an ambivalence regarding the prevention of disease in unborn babies in US citizens to reduce the risk of disease later in life where over the years there has been a tendency to consider an appropriate use of the technology. Overall, the reasons for rejecting enhancement are similar in scenarios of somatic (adults) or germline (unborn babies) settings and they are marked by the rejection of the use of genome editing for cosmetic purposes and for providing advantage of individuals over their peers either in physical, psychological or emotional characteristics, because it "messes with nature" and because it may be misused or used for wrong purposes. A sense or expectation that new technologies might not be used in the interest of the masses have been object of study and constitute a usual argument from citizens independent of which technology they are being inquired about (Hedgecoe 2010). Citizens also manifest their skeptical view that genome modification technologies in germline settings will bring benefits for society other than for disease purposes and while some don't discuss this, there is an increased risk perception that modulates genome editing

acceptance or approval. Perception of risk is associated with diverse factors ranging between the awareness, knowledge, education and socioeconomic status of individuals (Busch et al, 2021, Clark et al 2016, Davies et al 2021). Concerns of moral order and some arguments related with legal approval or insufficient regulation are also part of the questionnaires and usual arguments of citizens that don't favor genome editing technologies or when considering the inappropriateness of the technology. Here one may also find reasoning behind the cultural backgrounds of populations that are inquired. Despite the complicated analysis of the cultural backgrounds that may influence individuals in their assessment of an emergent technology to intervene in the genome, what becomes transversal to the studies done and the (bio)ethical framework that has been outlined in the literature are arguments that usually fall under the premises of morality related with unnaturalness, lack of integrity and corrupting dignity. There has been some work from EGE that related these aspects with humanness (EGE 2021). Curiously, on the side of bioethical discussions, the moral perspective seems to have suffered an alteration particularly for the case of germline interventions because the narrative shifted towards pragmatic questions related with safety and efficacy that suggest a more biomedical tone (Almeida and Ranisch 2022). Interestingly, researchers' argumentation is often not based in moral concerns by opposition to citizens because researchers are more pragmatic and usually don't see the human genome as something to be preserved but rather a biological material that needs intervention due to the high level of errors that it may encompass. Citizens, on the other hand, usually opt to argument based in moral concerns despite safety worries being also observed. This may be true due to the notion that for researchers, genetic determinism is not the paradigm anymore and the fact that genome diversity may be challenged due to epigenetic and environmental factors that are able to influence an individual's life more than his/her own genetic code (Brokowski and Adli 2018, Mulvihil et al 2017, Nerlich and Hellsten 2003). Citizens often see the genetic code as something that once altered will be changing nature and therefore observe a higher risk perception motivated by those moral concerns (Knight 2005, Clark et al 2016, Busch et al 2021). However, there is also the view that pragmatic questions cannot be the sole driver of the discussions and therefore the topics that allude to the moral side of it have been conveyed as relevant and cross with the relevant documents such as The Universal Declaration on the Human Genome and Human Rights (UDHGHR) that stress the importance of genome preservation (EGE 2021).

For a discussion of moral concerns with regards to future generations, citizens are more unclear but they still leave some tracks that concern the morality of the genetic intervention in face of the responsibility over the individuals and in the likely permanent effects that may be induced to unborn babies. This notion intersects with the requests by the scientific

community that discouraged germline genome editing over the last years even for clinical applications despite acknowledging the inevitability that accompanies it (Baltimore et al 2015, Bosley 2015, Lander 2019). Interviewed life sciences researchers also elaborated on worries of moral nature related with inability that a recently born edited baby have towards its own consent and if there is such a need from clinical staff when comparing with strategies of *in vitro* fertilization or pre-implantation diagnosis. A germline intervention is not a measure to avoid imminent death, or an urgent medical intervention that will save a patient's life. It is avoiding serious disease that will be a burden for the individual and very likely to their parents and that will diminish life quality as regards to physical capacity and level of independence considered under the World Health Organization Quality of Life (WHOQOL 2012). Usually, a physician assumes responsibility for a medical intervention when informed consent cannot be obtained, i.e. for mentally ill patients or for patients that are minors; physicians also swear the Hippocrates oath (Declaration of Geneva 1948) which stresses that they have to intervene if there is no alternative when imminent death or saving a human life is at stake and abide by other medical ethics guidelines which places the health and well-being of the patient as the primary consideration (Hajar 2017). Justification for the intervention is related to the seriousness of the condition, and these are fundamental aspects to substantiate acceptability in the opinion of researchers. An additional and critically important question seems to be then: who defines who is responsible when the affected individual cannot provide their consent? (Howard et al 2018). Safety is also a meaning of what one wants to guarantee for the patient when the intervention takes place. Does one want to guarantee higher life expectancy or immediate relief of its health status?

Two axes: Governance and Society

Society will be the ultimate level of genome editing implementation and where biomedical and animal science researchers recognize their role as science informants and as actors that may foster science and society dialogue. The deficit model that ensures that there is one that teaches to the one that listens would then be gradually replaced by the one where both actors are in equal stances and feed each other retroactively and continuously in a dialogical approach (Einsiedel 2008, Wirz, Scheufele and Brossard 2020). For this to happen, biomedical and animal science researchers say that, in the first place, citizens need to be receptive to learn about scientific and technological developments particularly emergent technologies and only then, a dialogical attitude to discuss societal impacts of the technologies. Researchers that share this view also emphasize that they need to remain

receptive to public questions and public skeptics. Researchers encounter one factor that may influence the behavior of the public towards this dialogical attitude which is related with the cultural background that exists. Cross-cultural communication remains one of the strategies already evoked for the ethical, legal and social aspects (ELSA) of technologies and this involves the dimensions of "perception, social use and significance of technology, second, the construction of social and ethical issues; and third, how social and ethical implications are investigated" (Rehman-Sutter 2010 page 89). I will not discuss people nor researchers' cultural backgrounds for the sake of keeping personal data safe and confidential but I will stress that cultural differences are perhaps deeply integrated in the regulation of technology and this has been intended to be explored in the second interviews with (bio)ethicists and policy-makers. Moreover, regulation of genome editing is strongly associated with the axis of governance which complements the understanding of the technology implementation in a chronological and geographical manner.

Regulation is a relevant factor in bridging technology and society comprising the previous step to governance of the technology. Governance of emergent technologies may happen in multiple ways with the enforcement of soft and hard legislation or through a mechanism that avoids prohibition and allowance but rather establishes criteria for the technology to be implemented, the conditions of implementation and the scenarios where such implementation should take place (Hurlbut 2018, Nordberg et al 2018). Almost all researchers demonstrate a will for technology to be regulated because they understand that this should be the first step to avoid radical legislation that might be enacted by governments regarding genome editing. This would influence their own work but also limit the research that would later be adopted by society if breakthroughs are achieved. There are some lines of thought about the use of soft and hard law that have been explored in a policy perspective regarding emergent technologies like artificial intelligence and genome editing (see https://www.sienna-project.eu/genomics/enhancing-legal-frameworks/). Researchers throw again the coin of responsibility in face of what the society might need once the technology is proven to be safe and useful for patients in therapeutic scenarios. Moreover, both biomedical and animal science researchers recognize that regulation will in fact help in clarifying the lines that need to be drawn regarding disease and non-disease purposes as well as between disease and prevention of diseases and in this case, populations will know better why certain lines of research are being conducted and with which objectives. Furthermore, researchers acknowledge the establishment of regulatory mechanisms for application of genome editing as a mean to prevent wrong practices in Science and scandals such as the He Jiankui's case. In general, researchers do not exclude that research not supervised by scientific community and governments may still happen but the

level of regulation applied may help in tracking certain misuses of the technology that include both research with germline cells, human subjects and animals and later challenge authorities to act once those practices are observed. All researchers have present in their argumentation the need to abstain humans of being submitted to any questionable medical practices which goes hand-in-hand with the Declaration of Helsinki (Declaration of Helsinki 2013). The moratorium that still stands for research in germline cells gathers ambivalent opinions due to the problem of limiting scientific research that may be later demonstrated as relevant for discoveries in genome editing. This ambivalence is constant from the beginning with many authors and experts voicing their discontent with the moratorium (de Lecuona et al 2017) and others being more radical in emphasizing the necessity to proceed with germline genome editing research (Gyngell et al 2017). Overall, researchers understand the request but many don't understand the precautionary principle rule because some foresee that this would be just a temporary measure without a proactive alternative resembling what happened in the past during the Asilomar period for recombinant DNA that gradually became a mainstay in laboratories worldwide (Rufo and Ficorilli 2019).

Several works illustrate the unclearness, absence or unknown regulation and legislation regarding germline and somatic genome editing (Isasi, Knoppers and Kleiderman 2016, Baylis et al 2020) demonstrating in some cases an uncomfortable feeling with the delay that still exists asking for inclusion of different stakeholders, broadening the societal engagement of actors and with a focus in public participation (Halpern et al 2019, Jasanoff and Hurlbut 2018). Researchers interviewed agree with this view and also validate what is meant by public participation in surveys about these topics as detailed in manuscript 2. It is fundamental to map the ethical debate that exists regarding somatic and genetic editing to see which common lines may be followed for therapy, enhancement and prevention of disease purposes. The ethical debate is currently biased towards germline genome editing research due to the He Jiankui's case, leaving less room for the debate about therapeutic and prevention of diseases scenarios in somatic settings, which is what interviewed researchers are usually more interested in and knowledgeable about (Polcz and Lewis 2016). Nevertheless, much work has been done in the (bio)ethics and STS field to extract all the ethical and societal relevant questions that are fundamental to be address around genome editing research to inform stakeholders in decision-making positions to clearly regulate and legislate it (Almeida and Ranisch 2022, Brokowski and Adli 2018, Cribbs and Perera 2018, Howard et al 2018) while others positioned themselves over the years towards trying to establish the criteria needed for the technology to be applied (Baltimore et al 2015, Caplan 2019, Cathomen et al 2018, de Wert et al 2018, Halpern e al 2019, Ishii 2017, Rosemann et al 2019).

What still needs to be discussed bridges with one of the affirmations of researchers interviewed and that is transversal to the different works that map the ethical questions or explore them: the need of a common governance for genome editing. Common governance of genome editing technology in researchers' words is also a mean to avoid discrimination of individuals. Social justice is present in their argumentation not just based in the criteria for somatic and germline genome editing, but also according to the difference in legislation and acceptance between different regions of the globe. There are countries that are permissive, countries that are prohibitive and others that remain undefined regarding the use of genome editing in these scenarios (Baylis et al 2020, Boggio et al 2019, Isasi, Kleiderman and Knoppers 2016). So far, no legislation or regulation considers purpose, e.g. disease vs non-disease, disease and prevention of disease, disease and enhancement or prevention of disease vs enhancement. Researchers differentiate the approval dependent of those scenarios (as seen in manuscript 1) similarly to what citizens did and they suggest that these steps need to be taken as soon as possible to avoid discrimination between the genome-edited individuals and the non-genome edited ones once this happens in the future. Medical tourism is also acknowledged by researchers as a future likelihood if countries or continents start legislating without reaching a common ground for it and they are concerned that genome editing may follow other examples such as the case of stem cells transplantation (Meagher et al 2020, Nordberg et al 2018, Rosemann et al 2019). They understand the challenges of regulating genome editing worldwide due to the problem of seating everyone at the same table (a proper RRI and CTA approach, I must say!) but they advise that some actions in that sense need to be happening along with measuring public opinion about genome editing technology to enact the best policies as soon as possible. Responsibility and "technologies of humility" narratives drive their discourse and their sense of reasoning demonstrating that anticipation is the best way to deal with uncertainty and the logical step towards responsiveness. This narrative covers questions about social justice, access to genome editing technology and the danger of discrimination of individuals that emerged from interviews with researchers. Those questions were predominantly mentioned by researchers once the connection between regulation of the technology with society needs was being discussed and when the lines of criteria between disease, prevention of diseases, enhancement and somatic and germline were being drawn. During these moments, it was clear the deployment of questions already discussed in the literature by authors in the field of bioethics and STS field. These relate with the preoccupation with classism through the inadverted creation of individuals that are edited versus the non-edited ones that could generate perverse social stratification of citizens and a sense of superiority between humans (Hildebrandt and Marron 2018, Howard et al 2018, Hoffman 2018). This

sense of superiority could be evidenced in different ways: through the easier accessibility by individuals of privileged socio-economic status that may consider wealth the criteria for priority particularly in therapeutic scenarios or through the superiorization of individuals that when genome-edited as unborn babies were given enhanced physical, psychological or emotional characteristics exacerbating inequalities (Brokowski and Adli 2018, Howard et al 2018). Exacerbation of inequalities may come as an intentional or acknowledged form where citizens play the privilege card to have better access to technologies or this may be unintentional and by chance of likelihood if the inequalities are a consequence of therapeutic treatment or embryo selection (Almeida and Ranisch 2022). Researchers also recognize some impact about the patent dispute that is taking place since the invention of the technology may negatively affect citizens due to social exclusion by socio-economic status. As pointed elsewhere, the economic gain will prevail in face of social accessibility excluding many to its access due to overpriced despite of the decision on who may be allowed to commercially explore it (Brokowski and Adli 2018, Mulvihill et al 2017). Researchers consider that taxpayers are interested in this discussion because of their role in enabling national health service systems to access treatments (Roemer 1993). Likewise, prospective parents should be asked about GE for genetic diseases in the case of prevention of disease. Besides health purposes, this may also reduce the burden for public health investment (Polcz and Lewis 2016). This raises again the need for a broadened public discussion and evidence narrative of "technologies of humility" (Jasanoff 2003) where researchers evidence the need to bring multiple stakeholders for the discussion because they represent part of the actors that may be involved in a network of discussion and foresight towards an emergent technology such as genome editing (Jasanoff and Hurlbut 2018).

While researchers recognize their role as primary informants of society and one of the important group of stakeholders that needs to interact with the public in a dialogical manner, they understand the need to welcome another group of stakeholders that will have the background and the authority to take matters of genome editing regulation to higher hierarchical stances in order for it be implemented. Nevertheless, similar to what they expect from the public in remaining with an active feedback posture, researchers do not exclude themselves from a similar role with policy-makers and regulators since they provide the scientific argumentation and evidence for the justification of the technology development.

Some words for animal welfare and biosafety

Overall, there is a direct relationship between animal welfare worries and the approving of genome editing by researchers and citizens. In farm animals this is associated with the

productivity index that producers want to obtain by achieving certain characteristics in their animals through breeding. Dehorning of cows increases the number of cattle per square meter and reduces bruises in animals that will be later be converted in meat; preventing boar taint in pigs avoids smell of the meat without recurring to surgical castration; increasing myoglobin protein increases muscle mass in animals' meat and all these are recurrent objectives of farm producers (Eriksson et al 2018, Ishii 2017, Kramer and Meijboom 2021, Mueller et al 2019). The balance between what producers want and what consumers expect is somewhat different and produces different opinions in the general public because this is also associated with what citizens consider as "normal" in their consuming habits whether it's meat, milk or medicines and vaccines derived from the genetic engineering of animals (Clark et al 2016, Novoselova et al 2007, Ueland et al, 2012).

Biosafety is also related with the escape or release of edited species into the environment and the effects in their evolution are voiced by researchers as the most worrying situations that may result from the implementation of this technology (unpublished data). According to researchers, a better fitness of the genome-edited species may override biology of these and cause ecosystem unbalance which may ultimately result in alterations in food chain and exaggerated proliferation or extinction of certain species (unpublished data). The species that have an easier ability to reproduce due to shorter periods of pregnancy or gestation and the species that mate the easiest are the ones to cause concern about, in researchers' overview (unpublished data). This view is driven by their awareness about applications where genome-edited insects like mosquitoes are released in the wild as a result of trial fields that are already happening in some places of the world (Caplan et al 2015, Camporesi and Cavaliere 2016, Armbruster 2019, Scudellari 2019). The line drawn by citizens is usually driven by their moral compass since they may approve the use of GE animals released in the wild to eradicate diseases based in the benefit for human health but they also envision that these may mess with nature and this is seen as unnatural.

To summarize what the work in manuscripts 1 and 2 explored, a set of guidelines for a responsible genome editing have been formulated to establish priorities for its implementation in society. This guideline is an extension of the one submitted as deliverable for the IMGENE project (Appendix VII).

The guidelines for a responsible use of genome editing are organized in the following structure:

1) Safety of genome editing applications is crucial and depending on:

- Low off-target effects of the technology to proceed for clinical applications
- Higher knowledge of genome function by developing basic research in the field

2) Efficiency of editing the desired genes in a controlled and systemic way needs to be improved to achieve better correction of genetic and cancer diseases in humans

3) Purpose of genome editing applications should be to improve human health either by targeting diseases and by improving food resources (from animals or plants)

4) Birth of genome-enhanced humans are seen as irrelevant and unpredictable applications and therefore should be avoided

5) International regulation of genome editing should be harmonized between countries and citizens need to be scientifically educated to make informed decisions about the technology in order to be applied responsibly in society

6) Citizens are receptive to interventions in adults and children with serious genetic defects and exclude any practices involving enhancement of humans in both somatic and germline settings

7) Genome-edited animals should be used as they were until now: for disease-related purposes

- Focus in monogenetic diseases to proceed for clinical trials (researchers view)
- Focus and support for serious genetic diseases (general public view)

8) Genome-edited animals in farm scenarios are ok as long as their welfare is preserved, but genome-edited wild animals should be further tested to match approval expectations

Methodologies used in the project: critics and improvements

Manuscript 1 – Strengths: design and analysis. Limitation: sampling

I will start by the manuscript 1 where a qualitative study was designed to address a group of stakeholders (biomedical and animal science researchers) in an in-depth form. Semi-structured interviews are a form of in-depth interviews that oscillates between the unstructured, exploratory and the structured, closed type (Jamshed 2014). This type of interviews corresponds to the type that may help in gathering structured points-of-view from interviewed individuals while still leaving space for additional unpredicted questions and therefore a more flexible interview that resembles a conversation and a safe and healthy space to talk about complex subjects (DiCicco-Bloom and Crabtree 2006). Moreover, they

potentiate that unintended topics could surface in during conversations and lead to the discovery of themes initially not thought to be part of the studied subject (Braun and Clarke 2006, Mayring 2014). The method that we used during the work depicted in manuscript 1 allowed us to have expected and unexpected themes and consequently, a richness of data that favors the reasoning for leaving it in a semi-structured way throughout the whole study. I will also stress that this richness of data allows one to discuss topics in light of what the researchers really think and not just be stuck to what literature describes as well as it allowed us to distinguish between what is generalizable and what should be kept as singular. Moreover, we also attend to the deductive-inductive nature of the process of content (Saldaña, Miles and Huberman 2014) and thematic analysis (Braun and Clarke 2006) that allows one to discovery themes grounded in the data due to exhaustive, repetitive and rigorous analysis of excerpts that are voiced by interviewees (Braun and Clarke 2006, Namey et al 2008).

The triangulation of the data that is organized already in initial codes resembling categories of interest has been done and it corresponds to quality control check during analysis stage (VanderStoep and Johnston 2009). Me, Anna Olsson and Maria Strecht Almeida did a constant triaging and triangulation of the data in order for analysis to be faithful to what interviewees were arguing and discussing during interviews.

An effort to accomplish sample breadth and depth has been sought and achieved throughout the designing process of the study. Breadth corresponds to the idea of generalization, inclusion and diversity of data that is usually resembled in the finding of common patterns in the data analyzed that may later provide overarching and subthemes that will answer to research questions (Miles, Huberman and Saldaña 2014, VanderStoep and Johnston 2009). Depth is directly related richness of the data *per se* meaning both the amount of data which is quantifiable and elucidates about data saturation as well as it means the complexity of the data that has been analyzed which elucidates about the quality of the data (Miles, Huberman and Saldaña 2014). The geographical and background diversity of stakeholders that came from a total of 10 countries and 4 continents intended to bring the breadth necessary and here I find that this relates directly with depth because data saturation has been achieved due to the amount of data the repetitively attributed to the same categories. Moreover, depth of data is supported by a large amount of arguments from interviewees leading to the diversification of perspectives about the topics asked during interviews. A data saturation method assessment using quantitative calculations has been published after analysis of the interviews were conducted, but this would have brought an added-value to the analysis (Guest, Namey and Chen 2020).

I should stress nonetheless that a level of representation bias of participants happened during recruitment process and this is directly related with the purposeful sampling method employed. A recruitment bias is always expected in purposeful sampling and it may lead to the overrepresentation of individuals (VanderStoep and Johnston 2009, Stockerner 2019). While this bias was intended for the level of education of interviewees (all doctorate individuals), this was not the case in terms of gender and nationalities. Some of the participants recruited for the study showed to be less comfortable with English than others particularly non-European interviewees and this is sometimes reflected in the clarity of excerpts that were extracted during analysis of data. Eventually, this situation could have been minimized if more participants from English-speaking countries had been recruited. Nevertheless, one needs to expect a certain level of rejection that one should be aware of. rejection to invitations to participate in the study are always unpredictable and needs to be taken into account as well.

Some final considerations about the qualitative paradigm adopted for this type of research may also be said at this point. In the analysis, a constructivist and post-positivist approach to strive for subjectivity and instill a dialogical behavior of participants in the interviews (Mayring 2014, VanderStoep and Johnston 2009). A more objective and less discussing approach would likely decrease the difficulty of the analysis process and be more direct to the topics approached in the literature and shorten the time of research, but that was not the interest of this approach which intended to be in-depth. Moreover, a mixed-methods research approach comprising both quantitative and qualitative methodology could have been adopted for the group of stakeholders interviewed in order to have initial simple data that would be detailed later with the aim to still keep complexity and richness of data. This would eventually lead to a more extensive analysis of the opinion of a certain group of stakeholders but this would put at risk the diversity of stakeholders analyzed also influenced by time constraints of the project.

Manuscript 2 – Strenghts: critical appraisal and systematic review guidelines. Limitations: meta-analysis and inclusion criteria

A systematic review has been specifically adopted for the study of public acceptance of genome editing and it is fully represented in manuscript 2 of this PhD study. A systematic review is predominantly described as the gold standard for synthesizing scientific evidence due to its exhaustive, comprehensive and complete nature in comparing the studies that have been done about a certain topic throughout a period of time (CRD 2008, NHMRC 1999, Moher et al 2009, Petticrew and Roberts 2006). Surveys or questionnaires, are in the

other hand, very narrowed, closed and structured instruments employed in research to obtain a well-defined and precise picture of a certain topic in a specific period of time (Stockerner 2019). In order to investigate the public acceptance of a topic like an emergent technology such as genome editing which is defined in a period of time (10 years) it is therefore necessary to employ a broadened methodological approach while at the same time looking to the methods that more recurrently and easily are used to extract people's opinion. It is in this sense that a systematic review of surveys about genome editing (later transformed in genetic modification in pre-CRISPR and CRISPR periods) has been performed looking into 35 years where citizens have given their opinion about this subject.

Questionnaires that are delivered in a precise time show a picture of that time-point but it is only when after observing the totality of surveys done in a time space that one may be able to visualize if and how opinions of respondents shifted (Stockerner 2019). Oftentimes, people that answer to surveys are not the same that will be recruited for the next unless these are longitudinal surveys that have the intention to follow the same individuals over time (Stockerner 2019). After acknowledging this, we opted to analyze an instrument that directly asks to citizens if they would use/not use, approve/disapprove or support/reject genetic modification with a defined number of answers possible while simultaneously leaving space for arguments and explanations for their options.

We acknowledge strengths and limitations of the methodology applied during the study that is detailed in manuscript 2. A scoping review as an example of comprehensive approach similar to systematic review could be used but it would exclude a critical appraisal of studies (Munn et al 2018). The critical appraisal of studies in a systematic review of surveys is a measure of quality since it also attests the quality of the studies that are included in this type of scientific writing format (CRD 2008, NHMRC 2000, Pace et al 2012, Petticrew and Roberts 2006). A critical appraisal usually supposes the analysis to studies in terms of if the research questions are answered by the research performed and if the methods used are in fact rigorous and scientifically valid to help in answering those research questions (Schlosser 2006, Petticrew and Roberts 2006). This is not a perfect method since there are acknowledged flaws to some of the tools that are constructed due to some lack of depth and difficulty in attesting the quality of studies as well as the necessity to address conformity with institutional and legal aspects that attest the quality of a study regarding the objects and the authors of the study from its design until its publication stage (Crowe and Sheppard 2011). In our case, we were also interested in the real quality of studies so that our own objective regarding public acceptance about genome editing wouldn't also be compromised. Observations about validity, reliability, item generation, risk of bias, sampling methodology, response rate and conformity with ethical and privacy requirements were part of the critical

appraisal of the surveys analyzed. This way it was possible to establish levels of quality among studies analyzed which fulfilled the remaining criteria to be part of a systematic review.

To generate the highest level of evidence synthesis, a systematic review may be combined with a meta-analysis procedure that allows comparison in a multi-level statistical approach (Moher et al 2009, Roever and Zoccai 2015). We did not do this, because we were interested in a general overview of citizens' opinions and in the reasons for their opinions rather than just comparing percentages of support. While meta-analysis in systematic reviews is not mandatory, statistical significance to compare quantitative data is a common procedure in systematic reviews in biomedical sciences (Moher et al 2009). Despite the obvious biomedical nature of the subject that is covered in the questionnaires administered to citizens and the quantitative nature of surveys, they still remain in the social research field and this approach remains valid (Campbell et al 2019). This is not a complete argument *per se* to justify the absence of this procedure, but we should stress that we still followed PICO (Population, Intervention, Comparison, Outcomes) guidelines (CRD 2008) in the sense that during the study designed in manuscript 2, we performed a qualitative comparison of outcomes from questionnaires. Such a comparison was done transversally and longitudinally by comparing populations among different countries and between different years of survey dates.

Another remark is the need to widen or narrow the criteria during study selection in order to include and exclude studies. We followed on the examples of (Cations et al 2018, Clayton et al 2018, Fei Bai et al 2018, Zhong, Darren and Dimaras 2017) that give priority to aspects related with specificity of the subject, minimum number of individuals inquired and the outcome assessed by inquirers while simultaneously following the PRISMA checklist guidelines (Moher et al 2009). The inclusion of studies that are indexed in scientific repositories and grey literature in an equal manner intended to avoid discarding relevant surveys that were administered to citizens without necessarily resulting in a publication in scientific or academic journals in the format of a publishable scientific article (CRD 2008). One of the limitations about the search of scientific empirical work from different sources relies in the use of non-consistent keywords which are dependent in the search engine used for that end (Piasecki, Waligora and Draseinka 2018). For example, in official online libraries with indexed journals such as the Web of Science, keywords need to obey to a set of guidelines in order to retrieve the most accurate scientific publications (Boolean strings) whereas google engine search relies on the use of tags (Haddaway et al 2015). Grey literature deserves recognition and a similar relevance treatment and it has been described

as an important source to find documents from official and non-official origin that comprise empirical work such as the case of surveys inquiring citizens (ibidem).

A toolkit and BioLab walk into research…

Two additional tools to equip future researchers in embedding RRI into research work and to explore emergent technologies constitute the final part of this discussion. I must also disclaim that the toolkit is intended for researchers currently in academia and BioLab is an initiatve (already a curricular unit) directed to university students that will be future researchers.

How do researchers may practice RRI within a funded project about emergent technologies? It was while thinking about this situation that two solutions were formulated. One solution is a toolkit already introduced in the deliverable submitted in the IMGENE project (Appendix VII). The solution follows the NewHoRRIzon experiences that were part of my learning about RRI in its full extent (Cohen and Loeber 2021). NewHoRRIzon project devised three series of workshops where I had the luck to participate and learn about RRI concept and about RRI as a principle of action. These workshops were the basis of formulation and simultaneously the end of the solution found to address an identified gap, a toolkit that may equip the next generation of researchers and allow them to deliver guidelines for responsible use of emergent technologies mainly focused in GE. The workshop series in the toolkit themselves would unfold in a similar timeline to the one that the NewHoRRIzon project covered although with a different approach regarding the invitation of stakeholders chosen to participate (Cohen and Loeber 2021, Timmermans et al 2020).

Biolaboratory – Multidisciplinary Experimentation Project has its roots in an initiative in the beginning of 2020 where a group of PhD students from three different PhD courses (MCBiology, NeuroScience and BiotecHealth) attending a module on Ethics and Society were presented to a workshop on CRISPR and Biohacking.

These ideas gained track when me and Filipe Marques conducted a workshop for university students after the invitation by the responsible for the teaching module and my supervisor Anna Olsson in articulation with my co-supervisor Maria Strecht Almeida. In the workshop, students were challenged with a discussion about emergent technologies and practices in the scientific domain, in the case CRISPR and biohacking. Their task was to interpret certain questions in a group and elaborate argumentation over those topics to be discussed later on and assuming a position towards it. This is one of the cases where one can say: “We went for the subject but continued because of the experience”. And in fact, it was the active

participation and constant feedback between us facilitators of the workshop and the PhD students that led to fruitful discussions about the subjects that were unknown for some, strange to others but overall, interesting to the many that were attending.

The toolkit for future researchers would be somewhat different since it would involve the guidelines for the facilitation of a series of workshops divided by the three years of a project. In this case, one workshop per year of the project would be ideal and it would allow researchers to have time for analysis and discussion of the outcomes from each workshop as well as time to prepare for the next workshop to happen regarding logistics and the scientific approach to be set for it. It is important that the researcher(s) that lead the project have some preparation in science and society interaction and dialogue through some previous research or training in scientific education and public engagement initiatives and a familiarization with workshop workflow so that the facilitation of future workshops would happen in the smoothest and natural way possible. Moreover, it is important to have a literature review in order to gather the topics that will function as the initiators of the workshop. Similarly, like NewHoRRIzon project, the workshops will follow the design of social labs where actors are joined in the same round and square tables or other discussion spaces always in a living form and where the interaction is the mean that will make the outcomes of emerge (Timmermans et al 2020). The workshops that will be included in the toolkit would be designed so that each workshop would correspond to a specific phase representing different types of actors interacting with each other. In the first workshop, a discussion between researchers involved in the project should take place followed by a debate about the most burning questions regarding genome editing in and the collection of the most important ethical issues that it faces in a second phase and finally in a third phase, the establishment of a criteria that could be used for an ethical acceptable guideline for the implementation of genome editing practices both in research and other environments that is intended to be embraced by policy-makers later on. For each workshop session, different questions would be driving the discussion and a different group of stakeholders would be joined in the co-creation space. Ideally, diversity of stakeholders from the different axis that are represented in RRI concept should be sought by the leaders of the project. The outcomes of the first workshop would have the intention to also feed on the second workshop and help in the recruitment of the set of stakeholders that would have the expertise to discuss the questions raised. Moreover, participation of institutional and societal actors highlights the logic of “division moral labor” since the questions that are raised by ones are intended to be answered by others and this will help in the stabilization and constructive technology assessment of emergent technologies (Rip 2018). A final workshop where there should be a reconvening of the different questions raised and

explored in the first two workshops should then have a closing mindset either joining internal stakeholders that are part of the project (if a large one) or joining internal stakeholders and external ones but with a policy-making role. The closure should convey the final considerations of the project to be presented either in an informal or formal way in conferences or meetings with experts that may take them up to hierarchical stances.

As for the BioLab, the experience was positive and, in this sense, it would be worth continuing and further developing. Together with three other colleagues (Júlio Borlido, Fernando Tavares and Manuela Lopes), we worked on the proposal of a Pedagogical Innovation Course Unit (UC InovPed) offered at ICBAS – School of Medicine and Biomedical Sciences in conjunction with the Faculty of Sciences and the research institute i3S (Instituto de Investigação e Inovação em Saúde). This UC InovPed is presented in detail in chapter 6 [manuscript 3]. Biolaboratory functions as a space for learning, practicing and reflection where students developed projects that ranged between scientifically rigorous and freeform experiments but where in the end, a re-focus towards the theme that was initially proposed would have to come either in a conceptual or prototype format (Marques et al 2021). So far, the course had two complete editions, and a third is ongoing at the time of writing (May 2023). The different group projects that were conducted in these two editions intended to tackle challenges within the themes bio-positive fashion and sustainability, respectively. Despite from the methodological approach with a strong biological background that all projects had to follow, there was space for anticipation of the positive and negative consequences as well the impacts that these projects would have for society (Marques et al 2021, Ramos et al 2022) and a reflection about how much they were harmonized with the 2030 agenda for the Sustainability Development Goals (UN 2015). This anticipative and reflexive behavior of students was constant throughout the whole project development having its peaks during ideation and closing stages. Such behavior is strongly embedded in RRI when postulating about Science and Technology developments for current and future generation (RRI 2020) The sharing of the space by student-researchers and teacher-researchers with multidisciplinary backgrounds with a strong community spirit and in an informal environment (Scheifele and Burkett 2016) led to the co-creation of multiple projects that have been described and presented elsewhere (Ramos et al 2022, Marques et al 2021, Ramos et al 2022b).

.

It is important to look at how this pedagogical practice was relevant in the context this PhD project on technical and ethical challenges of GE. What started as a side-kick project, also became an opportunity to be articulated with my PhD project because, in the end, within BioLab, we were promoting a RRI environment and space to train (for) RRI. Apart from the

fruitful projects that came from BioLab, it was noteworthy the linkage with the studies performed with stakeholders about an emergent technology such as GE due to the anticipative, reflexive and responsive nature that both were after. Interestingly, the BioLab is a space to train RRI and training RRI with the help of toolkits such as the one presented may be a way to be more responsible while performing research (Gerrits, Breedenord and van Mil 2020, Timmermans et al 2020). Moreover, the BioLab has the potential to be transferred to other academic institutions and study contexts (Marques et al 2021) and in illustrating how RRI can be enacted in practice and considered a relevant space to train future researchers.

Chapter 8. Conclusion and future perspectives

This thesis fills the gap in providing the point-of-view of researchers that contact with GE technology daily for its implementation once an opportunity to be embraced by society arrives. The work showed in this thesis also enlightens about the priorities for GE to be done responsibly and how different citizens around the world may be more open or skeptical about its adoption in a clearer and organized way. A clear set of guidelines for the responsible use of genome editing have been elaborated as an output from these studies so that stakeholders in decision-making bodies might use it for policy action. Two additional outputs aiming at preparing future researchers were also an opportunity to fill a gap that in approaching emergent technologies. By embedding RRI in a toolkit to perform workshops with a social lab character and by forming community space labs where RRI might be trained, I am providing tools for researchers to train anticipative, reflexive, inclusive and responsive dimensions when addressing emergent technologies.

Overall, the objectives of the PhD project have been achieved, taking into account the transformation that they have experienced along the journey. Re-definition of objectives is already a part of a medium-term project which when influenced by a third party that produces knowledge generally lead to another degree of development. Participants of the studies have direct influence in the course that the project will follow. Better yet, the central role of my research was to investigate how individuals belonging to a group of stakeholders think about genome editing in an in-depth way and therefore one could only expect this to lead to more complete information being collected and a diversification of views that would bring richness to the research work.

Interviewed researchers and citizens constitute two group of stakeholders that are usually in the beginning and end points of scientific and technological development, respectively. RRI changes the paradigm because researchers and citizens are in interchangeable positions. Researchers are also citizens and may have ethical and societal concerns while may be present in science-society dialogues and important stakeholders in the decision-making process of emergent technologies like GE. Throughout this project, RRI was considered both as a concept to theoretically define the angles through which the research work would proceed and as an action principle due to its practical application based in the analysis of the data and subsequent conception of guidelines for implementation of responsible genome editing as well as in its application as a method to increase responsibility of future researchers. The construction of a RRI toolkit able to be integrated in work packages of future projects for the practice of anticipation, reflexivity, inclusivity and responsiveness dimensions of current and future researchers is the culmination of RRI as an object of research and the complementary step that will add an ethical and societal value that is necessary and asked more and more by society.

It became clear from the outcomes of the work detailed in manuscript 1 what are the lines drawn by biomedical and animal science researchers about the use of genome editing in multiple settings. From research to clinical applications, from somatic to germline cells, from therapeutic to prevention of diseases, from treatment of diseases to enhancement of characteristics in humans and from laboratory animals to farm and wild animals for situations of disease modelling, disease eradication, health improvement and productivity, researchers provided their point-of-view. Citizens' views are most of the times in agreement with the ones from researchers for human applications but differ for animal applications and these should therefore be observed in a larger scale with more group of stakeholders being called to the discussions and further initiatives of engagement between these stakeholders to have a more comprehensive and clear view of the lines that may or may not be crossed in the short- and long-term for the application of genome editing technology.

In my research work, a series of interviews with (bio)ethicists and policy-makers were designed, reviewed and performed with the aim to capture the views of another very important group that is actually in hierarchical positions of advice, counselling and decision-making and that may engage in political milieus. This is still work in progress on their views regarding what they perceive about genome editing. Preliminary results (unpublished data) overlap with the ones extracted from interviews with life sciences researchers and citizens' surveys in the majority of topics and it assumes a global dimension due to the geographical breadth achieved once more for this group of stakeholders. There are discordant views particularly in the field of somatic and germline GE, but there is a relevant dimension explored during these interviews. This dimension further highlights what has been voiced by the life sciences researchers during interviews and in the published literature on genome editing technology. It encompasses the need to proceed with a more dialogical attitude with citizens, the acknowledgement of their involvement in a more upstream stage of the discussion and the inclusion of groups of stakeholders that are usually marginalized in an equal degree as scientists and policy-makers.

Once again, the contribution of the different societal actors (researchers included) will be valuable in improving the guidelines for implementation of genome editing technology conferring a more conciliated and complete view about what society needs from this emergent technology. This will also be useful to inform stakeholders that participated in the studies of this PhD thesis since they might be unaware or oblivious about how other stakeholders view genome editing. This may also influence their attitudes towards other groups of actors now that they understand how they think about those technologies and be more willing to promote dialogical attitudes and public engagement initiatives as well as this

might change their approach to the design of their research projects by including more stakeholders that before were not immediate to them.

All in all, despite its theoretical conceptualization as an STS research work that looks deeply into genome editing as an emergent technology, this PhD project intends to have a practical and actionable dimension. This project is not an end in itself with limited and final answers but rather a contribution to the acknowledgement that a lot of work is still necessary and crucial to be done in investigating the span of genome editing in its different multitudes before any decisions are made when implementing it for society.

RRI and CRISPR/CasX are both in their infancy with both celebrating 10 years and one thing is certain: they should not lag behind one another but rather feed one another continuously so that a responsible, humble, desirable and revolutionary technology can be delivered to society and make serious problem from the past, mere trivialities in the 21st century.

References

Abou-El-Enein M, Cathomen T, Ivics Z, June CH, Renner M, Schneider CK, Bauer G. 2017. “Human Genome Editing in the Clinic: New Challenges in Regulatory Benefit-Risk Assessment. Cell Stem Cell”. 21(4):427-430. doi: 10.1016/j.stem.2017.09.007. PMID: 28985524.

Adli, M. 2018. "The CRISPR Tool Kit for Genome Editing and Beyond." Nature Communications 9 (1): 1911. https://doi.org/10.1038/s41467-018-04252-2.

Almeida, M., Ranisch, R. 2022. “Beyond safety: mapping the ethical debate on heritable genome editing interventions.” Humanit Soc Sci Commun 9, 139. https://doi.org/10.1057/s41599-022-01147-y

Araki M, Ishii T. 2016. “Providing Appropriate Risk Information on Genome Editing for Patients”. Trends Biotechnol. 2016;34(2):86-90.

Atkinson, P., Glasner, P., & Lock, M. (Eds.) 2009. The Handbook of Genetics & Society: Mapping the New Genomic Era (1st ed.). Routledge. https://doi.org/10.4324/9780203927380

Bai F, Ling J, Esoimeme G, Yao L, Wang M, Huang J, et al. 2018. “A systematic review of questionnaires about patient's values and preferences in clinical practice guidelines”. Patient Prefer Adherence.;12:2309-23.

Baltimore, D., P. Berg, M. Botchan, D. Carroll, R. A. Charo, G. Church, J. E. Corn, et al. 2015. "Biotechnology. A Prudent Path Forward for Genomic Engineering and Germline Gene Modification." Science 348 (6230): 36-8. https://doi.org/10.1126/science.aab1028.

Bangkok Statement on Human Gene Editing and Human Genetic Engineering. 2019. Eubios Journal of Asian and International Bioethics 29 (1): 1-4

Barrangou, R., and P. Horvath. 2017. "A Decade of Discovery: CRISPR Functions and Applications." Nature Microbiology 2: 17092. https://doi.org/10.1038/nmicrobiol.2017.92.

Baylis, F., M. Darnovsky, K. Hasson, and T. M. Krahn. 2020. "Human Germ Line and Heritable Genome Editing: The Global Policy Landscape." CRISPR Journal 3 (5): 365-77. https://doi.org/10.1089/crispr.2020.0082.

Baylis F, McLeod M. 2017. “First-in-human Phase 1 CRISPR Gene Editing Cancer Trials: Are We Ready?” Curr Gene Ther. 17(4):309-19.

Bell, J. K., Eckdahl, T. T., Hecht, D. A., Killion, P. J., Latzer, J., Mans, T. L., . . . Bell, J. E. 2016. “CUREs in biochemistry – where we are and where we should go.” Biochemistry and Molecular Biology Education, 45(1), 7-12. https://doi.org/10.1002/bmb.20989

Berg, P., Baltimore, D., Brenner, S., Roblin, R and Singer, M. 1975. “Summary statement of the Asilomar conference on recombinant DNA molecules.” PNAS. 72 (6) 1981-1984. https://doi.org/10.1073/pnas.72.6.1981

Bjursell M, Porritt MJ, Ericson E, Taheri-Ghahfarokhi A, Clausen M, Magnusson L, et al. 2018. “Therapeutic Genome Editing With CRISPR/Cas9 in a Humanized Mouse Model Ameliorates alpha1-antitrypsin Deficiency Phenotype”. EBioMedicine. ;29:104-11.

Boggio, A., B. M. Knoppers, J. Almqvist, and C. P. R. Romano. 2019. "The Human Right to Science and the Regulation of Human Germline Engineering." CRISPR Journal 2: 134-42. https://doi.org/10.1089/crispr.2018.0053.

Boateng GO, Neilands TB, Frongillo EA, Melgar-Quinonez HR, Young SL. 2018. “Best Practices for Developing and Validating Scales for Health, Social, and Behavioral Research: A Primer”. Front Public Health.6:149.

Boeke JD, Church G, Hessel A, Kelley NJ, Arkin A, Cai Y, Carlson R, Chakravarti A, Cornish VW, Holt L, Isaacs FJ, Kuiken T, Lajoie M, Lessor T, Lunshof J, Maurano MT, Mitchell LA, Rine J, Rosser S, Sanjana NE, Silver PA, Valle D, Wang H, Way JC, Yang L. GENOME ENGINEERING. 2016. “The Genome Project-Write.” Science. 8;353(6295):126-7. doi: 10.1126/science.aaf6850. Epub 2016 Jun 2. PMID: 27256881.

Bolarinwa OA. 2015. “Principles and methods of validity and reliability testing of questionnaires used in social and health science researches.” Niger Postgrad Med J., 22(4):195-201. doi: 10.4103/1117-1936.173959. PMID: 26776330.

Bolotin A, Quinquis B, Sorokin A, Ehrlich SD. 2005. “Clustered regularly interspaced short palindrome repeats (CRISPRs) have spacers of extrachromosomal origin. Microbiology” 151(Pt 8):2551-2561. doi: 10.1099/mic.0.28048-0. PMID: 16079334.

Borup, M., N. Brown, K. Konrad, and H. van Lente. 2006. "The sociology of expectation in Science and Technology". Technology Analysis & Strategic Management 18 (3,4): 285-298

Bosley K, Botchan M, Bredenoord A, Carrol D, Alta Charo R, Chapentier E et al. 2015. “CRISPR germline engineering – the community speaks.” Nat Biotechnology.;33(5):478-486

Braun, M., and D. Meacham. 2019. "The Trust Game: CRISPR for Human Germline Editing Unsettles Scientists and Society." EMBO Reports 20 (2). https://doi.org/10.15252/embr.201847583.

Braun, V., and V. Clarke. 2006. "Using Thematic Analysis in Psychology." Qualitative Research in Psychology 3 (2): 77-101. https://doi.org/10.1191/1478088706qp063oa.

Brokowski, C., and M. Adli. 2018. "CRISPR Ethics: Moral Considerations for Applications of a Powerful Tool." Journal of Molecular Biology 431 (1): 88-101. https://doi.org/10.1016/j.jmb.2018.05.044.

Brown, C. 2018. “China Has Already Gene-Edited 86 People With CRISPR”. Gizmodo, January 22

Bruce, A., and D. Bruce. 2019. "Genome Editing and Responsible Innovation, Can They Be Reconciled?". Journal of Agricultural and Environmental Ethics 32 (5-6): 769-88. https://doi.org/10.1007/s10806-019-09789-w.

Bucchi, M. 2004. Science in Society: an introduction to social studies of science. Translated by Adrian Belton. Abingdon: Routledge

Busch, G., E. Ryan, M. A. G. von Keyserlingk, and D. M. Weary. 2021. "Citizen Views on Genome Editing: Effects of Species and Purpose." Agriculture and Human Values 39 (1): 151-64. https://doi.org/10.1007/s10460-021-10235-9.

Calenbuhr, V. 2020. “Complexity Science in the Context of Policymaking”. In: Sucha V, Sinkiewicz M editors. Science for Policy Handbook. Elsevier: European Union; pp. 119-127

Campbell M, McKenzie JE, Sowden A, Katikireddi SV, Brennan SE, Ellis S, Hartmann-Boyce J, Ryan R, Shepperd S, Thomas J, Welch V, Thomson H. 2020. “Synthesis without meta-analysis (SWiM) in systematic reviews: reporting guideline”. BMJ 16;368:l6890. doi: 10.1136/bmj.l6890. PMID: 31948937; PMCID: PMC7190266.

Camporesi S., and G. Cavaliere. 2016. "Emerging ethical perspectives in the clustered regularly interspaced short palindromic repeats genome-editing debate". Personalized Medicine 13 (6): 575-586. https://doi.org/ doi: 10.2217/pme-2016-0047

Caplan, A. L., B. Parent, M. Shen, and C. Plunkett. 2015. "No Time to Waste--the Ethical Challenges Created by CRISPR: CRISPR/Cas, Being an Efficient, Simple, and Cheap Technology to Edit the Genome of Any Organism, Raises Many Ethical and Regulatory Issues Beyond the Use to Manipulate Human Germ Line Cells." EMBO Reports 16 (11): 1421-6. https://doi.org/10.15252/embr.201541337.

Caplan A. 2019. “Getting serious about the challenge of regulating germline gene therapy. PLoS Biol 17(4): e3000223. https://doi.org/10.1371/journal.pbio.3000223

Carlson DF, Tan W, Hackett PB, Fahrenkrug SC. 2013. “Editing livestock genomes with site-specific nucleases”. Reprod Fertil Dev. 26(1):74-82. doi: 10.1071/RD13260. PMID: 24305179.

Catalão Alves, C. 2020. “From Science to Society: Strengthening Citizenship”, In: Patrão Neves, M.C. editor. Ethics, Science and Society: Challenges for BioPolitics, Fundação Luso-Americana

Carvalho C, Gaspar A, Knight A, Vicente L. 2018. “Ethical and Scientific Pitfalls Concerning Laboratory Research with Non-Human Primates, and Possible Solutions”. 2018. Animals (Basel).;9(1).

Cathomen, T., S. Schule, M. Schussler-Lenz, and M. Abou-El-Enein. 2018. "The Human Genome Editing Race: Loosening Regulatory Standards for Commercial Advantage?". Trends in Biotechnology 37 (2): 120-23. https://doi.org/10.1016/j.tibtech.2018.06.005.

Centre for Reviews and Dissemination. 2009. Core principles and methods for conducting a systematic review of health interventions. In: Centre of Reviews and Dissemination editors, Systematic Reviews. York: University of York;. pp.1-108

Centre for Reviews and Dissemination. 2009. Incorporating qualitative evidence in or alongside effectiveness reviews. In: Centre of Reviews and Dissemination editors, Systematic Reviews. York: University of York;, pp. 219-237

Court Justice of the European Union. 2018. “Organisms obtained by mutagenesis are GMOs and are, in principle, subject to the obligations laid down by the GMO Directive”. PRESS RELEASE No 111/18, Luxembrourg 25 July

Chikhazhe TL. 2015. “New Zealand public attitudes towards genetically modified food. M.Sc. Thesis, Lincoln University”. Available from: https://researcharchive.lincoln.ac.nz/handle/10182/6825

Clayton EW, Halverson CM, Sathe NA, Malin BA. 2018. “A systematic literature review of individuals’ perspectives on privacy and genetic information in the United States.” PLOS ONE 13(10): e0204417. https://doi.org/10.1371/journal.pone.0204417

Clark, B., Stewart, G.B., Panzone, L.A. et al. 2016. “A Systematic Review of Public Attitudes, Perceptions and Behaviours Towards Production Diseases Associated with Farm Animal Welfare.” J Agric Environ Ethics 29, 455–478. https://doi.org/10.1007/s10806-016-9615-x

ClinicalTrials.gov [Internet]. Sichuan University; Chengdu Medgencell (MD): National Library of Medicine (US). 2016 June 8 - . Identifier NCT02793856, PD-1 Knockout

Engineered T Cells for Metastatic Non-small Cell Lung Cancer; 2016 June 8 [cited 2021 Aug 3]; [about 6 screens]. Available from: https://www.clinicaltrials.gov/ct2/show/NCT02793856?term=CRISPR&cond=cancer&cntry=CN&rank=5

ClinicalTrials.gov [Internet]. Chinese PLA General Hospital (MD): National Library of Medicine (US). 2018 June 4 - . Identifier NCT03545815, Study of CRISPR-Cas9 Mediated PD-1 and TCR Gene-knocked Out Mesothelin-directed CAR-T Cells in Patients With Mesothelin Positive Multiple Solid Tumors.; 2018 June 4 [cited 2021 Aug 3]; [about 4 screens]. Available from: https://www.clinicaltrials.gov/ct2/show/NCT03545815?term=CRISPR&cond=cancec&cntry=CN&rank=1

ClinicalTrials.gov [Internet]. Editas Medicine (MD): National Library of Medicine (US). 2016 Feb 29 - . Identifier NCT03872479, Single Ascending Dose Study in Participants With LCA10; 2019 Mar 13 [cited 2021 Aug 3]; [about 5 screens]. Available from: https://www.clinicaltrials.gov/ct2/show/NCT03872479?term=NCT%2303872479&rank=1

Cohen, JB and Loeber A. 2021. “NewHoRRIzon CHANGING THE RESEARCH AND INNOVATION SYSTEM THROUGH DEMOCRATIC EXPERIMENTATION: A guide to good practices for Responsible Research and Innovation.”, Amsterdam

Committee on Bioethics (DH-BIO). Statement on genome editing technologies. 2015 Statement of Council of Europe DH-BIO/INF 13 Final

Convention for the Protection of Human Rights and Dignity of the Human Being with regard to the Application of Biology and Medicine: Convention on Human Rights and Biomedicine, Stat. 13 (1997).

Cormick C, Mercer R. 2017. “Community attitudes to gene technology”. 2017 The Office of the Gene Technology Regulator J2537

Cribbs A, Perera S. 2018. “Science and Bioethics of CRISPR-Cas9 Gene Editing: An Analysis Towards Separating Facts and Fiction”. Yale Journal of Biology and Medicine 2017 Dec 19;90(4):625-634

Critchley, C., D. Nicol, G. Bruce, J. Walshe, T. Treleaven, and B. Tuch. 2019. "Predicting Public Attitudes toward Gene Editing of Germlines: The Impact of Moral and Hereditary Concern in Human and Animal Applications." Frontiers in Genetics 9: 704. https://doi.org/10.3389/fgene.2018.00704.

Crowe M, Sheppard L. 2011. “A review of critical appraisal tools show they lack rigor: Alternative tool structure is proposed.” J Clin Epidemiol. ;64(1):79-89.

Cyranoski, D. 2019. “The CRISPR-baby scandal: what’s next for human gene-editing?”. Nature 566 Feb 28

Davies G, Gorman R, McGlacken R, Peres S. 2022. “The social aspects of genome editing: publics as stakeholders, populations and participants in animal research”. Lab Anim. 56(1):88-96. doi: 10.1177/0023677221993157. Epub 2021 Feb 17. PMID: 33596730; PMCID: PMC8918870.

de Graeff, N., K. R. Jongsma, J. Johnston, S. Hartley, and A. L. Bredenoord. 2019. "The Ethics of Genome Editing in Non-Human Animals: A Systematic Review of Reasons Reported in the Academic Literature." Philosophical Transactions of the Royal Society of

London B Biological Sciences 374 (1772): 20180106. https://doi.org/10.1098/rstb.2018.0106.

de Lecuona I., M. Casado, G. Marfany, M. Lopez Baroni, and M. Escarrabill. 2017. "Gene Editing in Humans: Towards a Global and Inclusive Debate for Responsible Research". Yale Journal of Biology and Medicine 90 (4): 673-681 PMID: 29259532; PMCID: PMC5733855.

De Saille, S. 2015. “Innovating innovation policy: the emergence of ‘Responsible Research and Innovation”. Journal of Responsible Innovation 2:2, 152-168, DOI: 10.1080/23299460.2015.1045280

de Wert, G., B. Heindryckx, G. Pennings, A. Clarke, U. Eichenlaub-Ritter, C. G. van El, F. Forzano, et al. 2018. "Responsible Innovation in Human Germline Gene Editing. Background Document to the Recommendations of ESHG and ESHRE." Human Reproduction Open 1: 1-18. https://doi.org/10.1093/hropen/hox024.

Declaration of Geneva. 1948. General Assembly of The World Medical Association at Geneva. September, Switzerland

Declaration of Helsinki. 1964. Recommendations guiding doctors in clinical research. 18th World Medical Assembly, June, Helsinki, Finland

Declaration of Helsinki. 1975. Recommendations guiding medical doctors in biomedical research involving human subjects. 29th World Medical Assembly, Tokyo, Japan

Declaration of Helsinki. 1996. Recommendations guiding physicians in biomedical research involving human subjects. 48th General Assembly, Somerset West, Republic of South Africa

The World Medical Declaration of Helsinki. 2000. Ethical Principles for Medical Research Involving Human Subjects. 52nd WMA General Assembly, Edinburgh, Scotland

Delgado, A., and H. Å,. 2018. "Experiments in interdisciplinarity: Responsibleresearch and innovation and the public good". PLoS Biology 16 (3): e2003921. https://doi.org/10.1371/journal.pbio.2003921

Delhove, J., I. Osenk, I. Prichard, and M. Donnelley. 2020. "Public Acceptability of Gene Therapy and Gene Editing for Human Use: A Systematic Review." Human Gene Therapy 31 (1-2): 20-46. https://doi.org/10.1089/hum.2019.197.

DiCicco-Bloom, B., and B. F. Crabtree. 2006. "The Qualitative Research Interview." Medical Education 40: 314-21. https://doi.org/10.1111/j.1365-2929.2006.02418.x.

DIVISION OF MENTAL HEALTH AND PREVENTION OF SUBSTANCE ABUSE WORLD HEALTH ORGANIZATION 1998. WHOQOL User Manual. Rev 2012.03

Dryzek, J et al. 2020. “Global citizen deliberation on genome editing”. Science 369,1435-1437(2020).DOI:10.1126/science.abb5931

Eichmeier, A. A., Bao, L., Xenos, M. A., Brossard, D. and Scheufele, D. A. 2023. “Fictional scenarios, real concerns: science fiction and perceptions of human genome editing”. JCOM 22(01), A08. https://doi.org/10.22323/2.22010208

Einsiedel, E. 2008. “Public participation and dialogue”. In: Bucchi, M & Trench, B editors. Handbook of Public Communication of Science and Technolog. Routledge: USA and Canada; pp. 131-146

European Academies Science Advisory Council. 2017. Genome editing: scientific opportunities, public interests and policy options in the European Union. EASAC policy report 31

European Group on Ethics in Science and New Technologies. 2021. Opinion on ethics of Genome Editing. Opinion no.32

European Commission. 2015. Science education for responsible citizenship : report to the European Commission of the expert group on science education. Publications Office. https://doi.org/doi/10.2777/13004

Fanelli D. 2009. “How many scientists fabricate and falsify research? A systematic review and meta-analysis of survey data”. PLoS One.4(5):e5738. doi: 10.1371/journal.pone.0005738. PMID: 19478950; PMCID: PMC2685008

Felt, U. 2018. "Responsible Research and Innovation". In the Handbook of Genomics, Health and Society, edited by Sahra Gibbon; Barbara Prainsack; Stephen Hilgartner; Janelle Lamoreaux, 108-116. Abingdon: Routledge

Frewer L. 2003. “Societal issues and public attitudes towards genetically modified foods.” Trends in Food Science & Technology.14(5-8):319-32.

Fereday, J., and E. Muir-Cochrane. 2006. "Demonstrating Rigor Using Thematic Analysis: A Hybrid Approach of Inductive and Deductive Coding and Theme Development." International Journal of Qualitative Methods 5 (1): 80-92 https://doi.org/10.1177/160940690600500107

Frangoul H, Altshuler D, Cappellini MD, Chen YS, Domm J, Eustace BK, Foell J, de la Fuente J, Grupp S, Handgretinger R, Ho TW, Kattamis A, Kernytsky A, Lekstrom-Himes J, Li AM, Locatelli F, Mapara MY, de Montalembert M, Rondelli D, Sharma A, Sheth S, Soni S, Steinberg MH, Wall D, Yen A, Corbacioglu S. CRISPR-Cas9 Gene Editing for Sickle Cell Disease and β-Thalassemia. N Engl J Med. 2021 Jan 21;384(3):252-260. doi: 10.1056/NEJMoa2031054. Epub 2020 Dec 5. PMID: 33283989.

Franklin S. 2019. “Ethical research - the long and bumpy road from shirked to shared”. Nature. 574(7780):627-630. doi: 10.1038/d41586-019-03270-4. PMID: 31664214.

Fung, D. 2017. “A connected curriculum for higher education”. UCL Press. https://doi.org/10.14324/111.9781911576358

Funk C and Heferon M. 2018. Most Americans Accept Genetic Engineering of Animals That Benefits Human Health, but Many Oppose Other Uses. Pew Research Center

Funtowicz, S and Ravetz, J. 2020. “Post-Normal Science: How Does It Resonate With the World of Today?”. In: Sucha V, Sinkiewicz M editors. Science for Policy Handbook. Elsevier: European Union; pp. 15-18

Gasiunas G, Barrangou, R., Horvath, P., and Siksnys, V. 2012. “Cas9–crRNA ribonucleoprotein complex mediates specific DNA cleavage for adaptive immunity in bacteria”. PNAS. 109 (39) E2579-E2586. https://doi.org/10.1073/pnas.1208507109

Gerrits, E.M., Bredenoord, A.L. & van Mil, M.H.W. 2022. “Educating for Responsible Research Practice in Biomedical Sciences”. Sci & Educ 31, 977–996. https://doi.org/10.1007/s11191-021-00295-y

Glaser, B., and Strauss, A. 1967. “The Discovery of Grounded Theory: Strategies for Qualitative Research”. In: Glaser, B and Strauss, A. Mill Valley, CA: Sociology Press

Greely H. 2019. “CRISPR’d babies: human germline genome editing in the ‘He Jiankui affair”. Journal of Law and the Biosciences 6(1): 111–183,

Guest G, Namey E, Chen M. 2020. “A simple method to assess and report thematic saturation in qualitative research.” PLoS One,15(5):e0232076. doi: 10.1371/journal.pone.0232076. PMID: 32369511; PMCID: PMC7200005.

Gyngell, C., T. Douglas, and J. Savulescu. 2017. "The Ethics of Germline Gene Editing." Jounal of Applied Philosophy 34 (4): 498-513. https://doi.org/10.1111/japp.12249.

Halpern, J., S. E. O'Hara, K. W. Doxzen, L. B. Witkowsky, and A. L. Owen. 2019. "Societal and Ethical Impacts of Germline Genome Editing: How Can We Secure Human Rights?". CRISPR Journal 2 (5): 293-98. https://doi.org/10.1089/crispr.2019.0042.

Haddaway NR, Collins AM, Coughlin D, Kirk S. 2015. “The Role of Google Scholar in Evidence Reviews and Its Applicability to Grey Literature Searching.” PLoS One. 17;10(9):e0138237. doi: 10.1371/journal.pone.0138237. PMID: 26379270; PMCID: PMC4574933.

Haddock G, Maio GR. 2008. “Attitudes: content, structure and functions. In: Hewstone M, Stroebe W and Jonas K editors. Introduction to social psychology: a European perspective. BPS textbooks in psychology. Oxford: Blackwell;. pp. 112-133.

Hair JF, L.D.S. Gabriel M, da Silva D, Braga Junior S. 2019. “Development and validation of attitudes measurement scales: fundamental and practical aspects.” RAUSP Management Journal.54(4):490-507

Hajar R. 2017. “The Physician's Oath: Historical Perspectives.” Heart Views. Oct-Dec; 18(4):154–159.

Hedgecoe A. 2010. “Bioethics and the reinforcement of socio-technical expectations.” Soc Stud Sci. 40(2):163-86. doi: 10.1177/0306312709349781. PMID: 20527320.

Hildebrandt C, and J. Marron. 2018. "Justice in CRISPR-Cas9 research and clinical applications". AMA Journal of Ethics 20 (9): E826-833. https://doi.org: 10.1001/amajethics.2018.826.

Hirsch F, Lemaitre C, Chneiweiss H, Montoliu L. 2019. “Genome Editing: Promoting Responsible Research. Pharmaceut Med.33(3):187-91.

Hofmann, B. 2018. “The gene-editing of super ego.” Medicine, Health Care and Philosophy. https://doi.org/10.1007/s11019-018-9836-z

Horizon 2020 work programme 2018-2020. 2018. “Science with and for Society.” European Commission Decision C(2018)4708 of 24 July.

Horst M. 2007. “Public Expectations in Gene Therapy: Scientific Futures and Their Performative Effects on Scientific Citizenship. Science, Technology & Human Values.;32(2):150-171

Howard, H. C., C. G. van El, F. Forzano, D. Radojkovic, E. Rial-Sebbag, G. de Wert, P. Borry, et al. 2018. "One Small Edit for Humans, One Giant Edit for Humankind? Points and Questions to Consider for a Responsible Way Forward for Gene Editing in Humans." European Journal of Human Genetics 26 (1): 1-11. https://doi.org/10.1038/s41431-017-0024-z.

Howell EL, Yang S, Beets B, Brossard D, Scheufele DA, Xenos MA. 2020. “What Do We (Not) Know About Global Views of Human Gene Editing? Insights and Blind Spots in the CRISPR Era.” The CRISPR Journal.3(3):148-55.

Howell, E. L., C. D. Wirz, D. A. Scheufele, D. Brossard, and M. A. Xenos. 2020. "Deference and Decision-Making in Science and Society: How Deference to Scientific Authority Goes Beyond Confidence in Science and Scientists to Become Authoritarianism." Public Understanding of Science 29 (8): 800-18. https://doi.org/10.1177/0963662520962741.

Hurlbut, J. B., K. Saha, and S. Jasanoff. 2015. "CRISPR democracy: Gene editing and the need for inclusive deliberation". Issues in Science and Technology 32 (1): 1-14

Hurlbut, B. 2018. "In CRISPR’s world: genome editing and the politics of global science". In the Handbook of Genomics, Health and Society, edited by Sahra Gibbon; Barbara Prainsack; Stephen Hilgartner; Janelle Lamoreaux, 121-178. Abingdon: Routledge

Isasi R., E. Kleiderman, and B. M. Knoppers. 2016. "Editing policy to fit the genome?" Science 361(6271): 337-339. https://doi.org/10.1126/science.aad6778.

Ishii T. 2017. “Genome-edited livestock: Ethics and social acceptance”. Animal Frontiers 7(2):24-32

Jasanoff, S, 2018. “Bodies in Transition: Ethics in Xenotransplantation Research,” Defining Death: Organ Transplantation and the Fifty-Year Legacy of the Harvard Report on Brain Death special report”. Hastings Center Report 48(6): S67- S69. DOI: 10.1002/hast.960

Jasanoff, S. 2003. “Technologies of Humility: Citizen Participation in Governing Science.” Minerva 41: 223-244. https://doi.org/10.1023/A:1025557512320

Jasanoff, S., and J. B. Hurlbut. 2018. "A global observatory for gene editing". Nature 555: 435-437. https://doi.org/10.1038/d41586-018-03270-w

Jasanoff, S., and S. Kim. 2015. Dreamscapes of modernity: sociotechnical imaginaries and the fabrication of power. Chicago and London: The University of Chicago Press

Jasanoff, S. 2005. Designs on Nature: Science and Democracy in Europe and the United States In: Jasanoff, S editors. Princeton, USA

Jedwab, A., D. F. Vears, C. Tse, and C. Gyngell. 2020. "Genetics Experience Impacts Attitudes Towards Germline Gene Editing: A Survey of over 1500 Members of the Public." Journal of Human Genetics 65 (12): 1055-65. https://doi.org/10.1038/s10038-020-0810-2.

Jinek, M., K. Chylinsky, I. Fonfara, M. Hauer, J.A. Doudna, and E. Charpentier. 2012. "A programmable dual-RNA-guided DNA endonuclease in adaptive bacterial immunity". Science. 337 (6096): 816-20. https://doi.org/10.1126/science.1225829.

Juengst, E. T., G. E. Henderson, R. L. Walker, J. M. Conley, D. MacKay, K. M. Meagher, K. Saylor, et al. 2018. "Is Enhancement the Price of Prevention in Human Gene Editing?". CRISPR Journal 1: 351-54. https://doi.org/10.1089/crispr.2018.0040.

Kang X, He W, Huang Y, Yu Q, Chen Y, Gao X, et al. 2016. “Introducing precise genetic modifications into human 3PN embryos by CRISPR/Cas-mediated genome editing.” J Assist Reprod Genet. 33(5):581-8.

Knight AJ. 2009. “Perceptions, knowledge and ethical concerns with GM foods and the GM process.” Public Underst Sci. 18(2):177-88. doi: 10.1177/0963662507079375. PMID: 19579682.

Kohl PA, Brossard D, Scheufele DA, Xenos MA. Public views about editing genes in wildlife for conservation. Conservation Biology. 2019;33(6):1286-95.

Konrad, K., H. van Lente, C. Groves, C. Selin. 2016. "Performing and Governing the Future in Science and Technology". In The Handbook of Science and Technology Studies, edited by Ulrike Felt; Rayvon Fouche; Clark A. Miller; Laurel Smith-Doerr, 465-493. Cambridge: MIT Press

Laber K, Newcomer CE, Decelle T, Everitt JI, Guillen J, Bronstad A. 2016. “Recommendations for Addressing Harm-Benefit Analysis and Implementation in Ethical Evaluation - Report from the AALAS-FELASA Working Group on Harm-Benefit Analysis - Part 2”. Lab Anim. 50(1 Suppl):21-42.

Lander S. Adopt a moratorium on heritable genome editing. Nature, 2019, Mar 13 [Cited 2021 Jan 22]. Available from: https://www.nature.com/articles/d41586-019-00726-5

Lander ES. The Heroes of CRISPR. Cell. 2016;164(1-2):18-28.

Langlois A. 2008. “The UNESCO Universal Declaration on Bioethics and Human Rights: perspectives from Kenya and South Africa”. Health Care Anal. 16(1):39-51. doi: 10.1007/s10728-007-0055-7. Epub 2007 Jun 28. PMID: 18240025; PMCID: PMC2226192.

Lassen J, Gjerris M, Sandoe P. 2006. “After Dolly--ethical limits to the use of biotechnology on farm animals.” Theriogenology.65(5):992-1004.

Ledford H. 2016. “The unsung heroes of CRISPR”. Nature, July 21 [Cited 2021 Mar 2021]. Available from: https://www.nature.com/articles/535342a

Ledford H. 2016. “CRISPR: gene editing is just the beginning”. Nature, March 2016

Liang P, Xu Y, Zhang X, Ding C, Huang R, Zhang Z, et al. 2015. “CRISPR/Cas9-mediated gene editing in human tripronuclear zygotes.” Protein Cell.;6(5):363-72.

Lillico SG, Proudfoot C, Carlson DF, Stverakova D, Neil C, Blain C, et al. 2013. “Live pigs produced from genome edited zygotes”. i.3:2847.

Lippens S, D'Enfert C, Farkas L, Kehres A, Korn B, Morales M, Pepperkok R, Premvardhan L, Schlapbach R, Tiran A, Meder D, Van Minnebruggen G. 2019. “One step ahead: Innovation in core facilities”. EMBO Rep Apr; 20(4):e48017. doi: 10.15252/embr.201948017. Epub 2019 Mar 14. PMID: 30872318; PMCID: PMC6446189.

Liu YP, Zheng CC, Huang YN, He ML, Xu WW, Li B. 2020. Molecular mechanisms of chemo- and radiotherapy resistance and the potential implications for cancer treatment. MedComm; 2(3):315-340. doi: 10.1002/mco2.55. PMID: 34766149; PMCID: PMC8554658.

Long C, Annally J, Shelton J, Mireault A, Bassel-Duby R, Olson E. 2014. “Prevention of muscular dystrophy in mice by CRISPR/Cas9–mediated editing of germline DNA.” Science 345(6201):1184-1188.

Luna, A. 2017. "The Unknown of Human Animal Chimera Research". Intersection 10 (3): 1-8

Lull RB, Akin H, Hallman WK, Brossard D, Jamieson KH. 2019. “Modeling Risk Perceptions, Benefit Perceptions, and Approval of Releasing Genetically Engineered Mosquitoes as a Response to Zika Virus.” Environmental Communication-a Journal of Nature and Culture.

Lyu C, Shen J, Wang R, Gu H, Zhang J, Xue F, et al. 2018. “Targeted genome engineering in human induced pluripotent stem cells from patients with hemophilia B using the CRISPR-Cas9 system.” Stem Cell Res Ther. 9(1):92.

Ma H, Marti-Gutierrez N, Park SW, Wu J, Lee Y, Suzuki K, et al. 2017. “Correction of a pathogenic gene mutation in human embryos“. Nature. 548(7668):413-9.

Macnaghten P. 2016. “Animals in their Nature.” Sociology.38(3):533-51.

Malhotra N. 2006. “Questionnaire design and development. In: Grover R, Vriens M editors.” The Handbook of Marketing Research: Uses, Misuses, and Future Advances. California: Sage Publications;pp. 176-202

Marques, F., Ramos, P. D., Lopes, M. M., Santos, J. B., Tavares, F., Olsson, I. A. S., & Almeida, M. S. 2021. “BioLab – um espaço de experimentação e cruzamento disciplinar” [Resumo de apresentação em conferência] 7.º Congresso Nacional de Práticas Pedagógicas no Ensino Superior, Aveiro. https://cnappes.org/files/2021/08/Livro_Resumos_CNaPPES.pdf

Marraffini LA, Sontheimer EJ. 2008. “CRISPR interference limits horizontal gene transfer in staphylococci by targeting DNA”. Science.322(5909):1843-5. doi: 10.1126/science.1165771. PMID: 19095942; PMCID: PMC2695655.

Martin, P. A., and I. Turkmendag, I. 2021. “Thinking the Unthinkable: How Did Human Germline Genome Editing Become Ethically Acceptable?” New Genetics and Society 40 (4): 384-405. https://doi.org/10.1080/14699915.2021.1932451

Matthews, K. R. W., and A. S. Iltis. 2019. "Are We Ready to Genetically Modify a Human Embryo? Or Is It Too Late to Ask?". Accountability in Research 26 (4): 265-70. https://doi.org/10.1080/08989621.2019.1617139.

Mayring, P. 2014. “Qualitative content analysis: theoretical foundation, basic procedures and software solution”. Klagenfurt. URN: http://nbn-resolving.de/urn:nbn:de:0168-ssoar-395173

McConnachie, E., M. J. Hotzel, J. A. Robbins, A. Shriver, D. M. Weary, and M. A. G. von Keyserlingk. 2019. "Public Attitudes Towards Genetically Modified Polled Cattle." PLoS One 14 (5): e0216542. https://doi.org/10.1371/journal.pone.0216542.

McLeod C, Hartley S. 2018. “Responsibility and Laboratory Animal Research Governance.” Sci Technol Human Values.;43(4):723-741. doi: 10.1177/0162243917727866. Epub 2017 Sep 1. PMID: 30008495; PMCID: PMC6027781.

Meagher, K. M., M. A. Allyse, Z. Master, and R. R. Sharp. 2020. "Reexamining the Ethics of Human Germline Editing in the Wake of Scandal." Mayo Clinical Procedures 95 (2): 330-38. https://doi.org/10.1016/j.mayocp.2019.11.018.

Menchaca, A., Schlapp, G., Meikle MN., and Crispo, M. 2017. “Transgenesis and gene edition in mammals.” In: Reference Module in Life Sciences doi:10.1016/B978-0-12-809633-8.09222-0

Meyer, M. 2022. “Taking responsibility, making irresponsibility: Controversies in human gene editing.” Social Studies of Science 52 (1): 127-143. https://doi.org/10.1177/0306312721102563

Miles, M.B., Huberman, M. and Saldaña, J. 2014. Qualitative data analysis: a methods sourcebook. California: SAGE Publications, Inc. – Third Edition

Mitchell, J.R., Mitchell, R.K., Hunt, R.A. et al. 2022. “Stakeholder Engagement, Knowledge Problems and Ethical Challenges.” J Bus Ethics 175, 75–94. https://doi.org/10.1007/s10551-020-04550-0

Moher D, Liberati A, Tetzlaff J, Altman DG, Group P. 2009. “Preferred reporting items for systematic reviews and meta-analyses: the PRISMA statement.” PLoS Med. 6(7):e1000097.

Mojica FJM, Montoliu L. 2016. “On the Origin of CRISPR-Cas Technology: From Prokaryotes to Mammals.” Trends Microbiol. 24(10):811-820. doi: 10.1016/j.tim.2016.06.005. Epub 2016 Jul 9. PMID: 27401123

Moreno-Mateos MA, Vejnar CE, Beaudoin JD, Fernandez JP, Mis EK, Khokha MK, Giraldez AJ. 2015. “CRISPRscan: designing highly efficient sgRNAs for CRISPR-Cas9 targeting in vivo”. Nat Methods. 12(10):982-8. doi: 10.1038/nmeth.3543. Epub 2015 Aug 31. PMID: 26322839; PMCID: PMC4589495.

Morrison, M., and S. de Saille. 2019. "CRISPR in Context: Towards a Socially Responsible Debate on Embryo Editing." Palgrave Communications 5 (1). https://doi.org/10.1057/s41599-019-0319-5.

Mulvihill, J. J., B. Capps, Y. Joly, T. Lysaght, H. A. E. Zwart, R. Chadwick, and The International Human Genome Organisation (HUGO) Committee of Ethics, Law and Society. 2017. "Ethical Issues of CRISPR Technology and Gene Editing through the Lens of Solidarity." British Medical Bulletin 122 (1): 17-29. https://doi.org/10.1093/bmb/ldx002.

Munn Z, Peters MDJ, Stern C, Tufanaru C, McArthur A, Aromataris E. 2018. “Systematic review or scoping review? Guidance for authors when choosing between a systematic or scoping review approach”. BMC Med Res Methodol.18(1):143. doi: 10.1186/s12874-018-0611-x. PMID: 30453902; PMCID: PMC6245623.

Mussolino C, Alzubi J, Pennucci V, Turchiano G, Cathomen T. 2017. “Genome and Epigenome Editing to Treat Disorders of the Hematopoietic System.” Hum Gene Ther. 28(11):1105-1115. doi: 10.1089/hum.2017.149. Epub 2017 Aug 14. PMID: 28806883.

National Academies of Sciences Engineering and Medicine (U.S.). 2017. Human Genome Editing: Science, Ethics, and Governance. The National Academies Press, Washington, DC

National Health and Medical Research Council. Appraising and selecting studies. In: National Health and Medical Research Council editors, How to review the evidence: systematic identification and review of the scientific literature. Canberra: Commonwealth of Australia; 2000. pp. 23-25

National Health and Medical Research Council. Aetiology and risk factors. In: National Health and Medical Research Council editors, How to review the evidence: systematic identification and review of the scientific literature Canberra: Commonwealth of Australia;2000. pp. 71-77

Nelson CE, Wu Y, Gemberling MP et al. 2019. “Long-term evaluation of AAV-CRISPR genome editing for Duchenne muscular dystrophy.” Nat Med427–432.

Nerlich B, Hellsten I. 2004. “Genomics: shifts in metaphorical landscape between 2000 and 2003.” New Genet Soc. 23(3):255-68.

Neuhaus CP. 2018. “Ethical issues when modelling brain disorders innon-human primates”. J Med Ethics.44(5):323-7.

Niu Y, Shen B, Cui Y, Chen Y, Wang J, Wang L, Kang Y, Zhao X, Si W, Li W, Xiang AP, Zhou J, Guo X, Bi Y, Si C, Hu B, Dong G, Wang H, Zhou Z, Li T, Tan T, Pu X, Wang F, Ji S, Zhou Q, Huang X, Ji W, Sha J. 2014. “Generation of gene-modified cynomolgus monkey via Cas9/RNA-mediated gene targeting in one-cell embryos”. Cell. 156(4):836-43. doi: 10.1016/j.cell.2014.01.027. Epub 2014 Jan 30. PMID: 24486104.

Nolan M, Beran T, Hecker KG. 2012. “Surveys assessing students’ attitudes toward statistics: A systematic review of validity and reliability.” SERJ. 11(2):103-123

Novoselova TA, Meuwissen MPM, Huirne RBM. 2007. “Adoption of GM technology in livestock production chains: an integrating framework.” Trends in Food Science & Technology.18(4):175-88.

Nuffield Council on Bioethics. 2016. Genome Editing: an ethical review.

Nordberg, A., T. Minssen, S. Holm, M. Horst, K. Mortensen, and B. L. Moller. 2018. "Cutting Edges and Weaving Threads in the Gene Editing (Я)Evolution: Reconciling Scientific Progress with Legal, Ethical, and Social Concerns." Journal of Law Biosciences 5 (1): 35-83. https://doi.org/10.1093/jlb/lsx043.

Ohl F, Meijboom F. 2015. “Ethical issues associated with the use of animal experimentation in behavioral neuroscience research”. Curr Top Behav Neurosci.19:3-15.

Olsson IAS, Silva SPD, Townend D, Sandøe P. 2016. “Protecting Animals and Enabling Research in the European Union: An Overview of Development and Implementation of Directive 2010/63/EU.” ILAR J.1;57(3):347-357

Olsson IAS, Sandøe P. 2010. “What’s wrong with my monkey?” Ethical perspectives on germline transgenesis in marmosets.” Transgenic Res 19, 181–186.

Oluka OC, Nie S, Sun Y. 2014. “Quality assessment of TBP-based questionnaires: A systematic review. PLoS One.” 9(4):e94419

On Human Gene Editing: International Summit Statement [Internet]. 2015; Dec. 3

Owen, R., P. Macnaghten, and J. Stilgoe. 2012. "Responsible Research and Innovation: From Science in Society to Science for Society, with Society." Science and Public Policy 39 (6): 751-60. https://doi.org/10.1093/scipol/scs093.

Owen, R., von Schomberg, R and Macnaghten, P. 2021. “An unfinished journey? Reflections on a decade of responsible research and innovation”. Journal of Responsible Innovation, 8:2, 217-233,DOI:10.1080/23299460.2021.1948789

Oye et al. 2014. Regulating gene drives.Science 345,626-628. DOI:10.1126/science.1254287

Pace R, Pluye P, Bartlett G, Macaulay AC, Salsberg J, Jagosh J, et al. 2012. “Testing the reliability and efficiency of the pilot Mixed Methods Appraisal Tool (MMAT) for systematic mixed studies review”. Int J Nurs Stud. 49(1):47-53.

Parsa-Parsi RW. 2017. “The Revised Declaration of Geneva: A Modern-Day Physician’s Pledge”. JAMA. 318(20):1971–1972. doi:10.1001/jama.2017.16230

Patton, M. Q. 2015. Qualitative research & evaluation methods : integrating theory and practice. California: SAGE publication, Inc. – Fourth Edition

Petticrew M, Roberts H. 2005. Systematic Reviews in the Social Sciences – A Practical guide. 1st ed. Oxford: Blackwell Publishing

Peters, H. 2008. “Scientists as public experts”. In: Bucchi, M & Trench, B editors. Handbook of Public Communication of Science and Technolog. Routledge: USA and Canada; pp. 131-146

Pereira ÂG, Völker T. 2020. Engaging With Citizens. In: Sucha V, Sinkiewicz M editors. Science for Policy Handbook. Elsevier: European Union;. pp. 78-95

Pereira M, Samorinha C, Alves E, Machado H, Amorim M, Silva S. 2015. “Patients' views on the embryo storage time limits”. Reprod Biomed Online, 31(2):232-8. doi: 10.1016/j.rbmo.2015.04.015. Epub 2015 May 8. PMID: 26096027.

Piasecki J, Waligora M, Dranseika V. 2018. “Google Search as an Additional Source in Systematic Reviews.” Sci Eng Ethics 24(2):809-810. doi: 10.1007/s11948-017-0010-4. Epub 2017 Dec 16. PMID: 29249022; PMCID: PMC5876410.

Polcz, S., and A. Lewis. 2016. "CRISPR-Cas9 and the Non-Germline Non-Controversy." Journal of Law and the Biosciences 3 (2): 413-25. https://doi.org/10.1093/jlb/lsw016.

Prescott MJ, Lidster K. 2017. “Improving quality of science through better animal welfare: the NC3Rs strategy”. Lab Anim (NY). 46(4):152-6.

Ramos, P. A. D. F. 2021. "AUDIT - Audio-Dictate Transcribing Automated Method." OSF. October 27. https://doi.org/10.17605/OSF.IO/74QSZ.

Ramos, P.D., Marques, F., Lopes, M.M., Santos, J.B., Tavares, F., Almeida, M. S. & Olsson, I.A.S. 2022. BioLab – uma proposta para o desenvolvimento de projetos em ambiente multidisciplinar. In Cadernos de Inovação pedagógica nº2, Unidades curriculares INOVPED da Universidade do Porto, Transversalidade e Criatividade, UPorto Press, Universidade do Porto, 1ª edição, pp: 125-139

[b]Ramos, P.D., Marques, F., Lopes, M.M., Santos, J.B., Tavares, F., Almeida, M. S. & Olsson, I.A.S. 2022. Biolaboratory – multidisciplinary experimentation project: an open biolab prototype. In Mertik, M., Lopes, M. M., Beloff, L., Evers, L. (Eds.) Hybrid Labs on Transdisciplinarity: Knowledge Through Experience [Elektronski vir]: best practices book (IO4) and Conference Proceedings, AMEU – ECM, Alma Mater Press (https://press.almamater.si/index.php/amp/catalog/book/48), pp. 81-84

Reardon, S. 2016. “First CRISPR clinical trial gets green light from US panel”. Nature. https://doi.org/10.1038/nature.2016.20137

Reardon, S. 2015. “Gene-editing record smashed in pigs”. Nature. https://doi.org/10.1038/nature.2015.18525

Regalado, A. 2018. "EXCLUSIVE: Chinese scientists are creating CRISPR babies." MIT Techology Review, November 25

Regalado, A. 2014. “Who owns the biggest biotech discovery of the century?”. MIT Technology Review, December 4

Regalado, A 2019. “The next trick for CRISPR is gene-editing pain away”. Biotechnology, August 22

Rehmann-Sutter, C. 2009. “Bioethical Decisions and the Public Sphere: A Cross-Cultural Perspective”. In: Nerlich, B, Elliott, R. and Larson, B. … Communicating Biological Sciences: ethical and metaphorical dimensions. Aldershot, pp:75-91

Reincke, C. M., A. L. Bredenoord, and M. H. van Mil. 2020. "From Deficit to Dialogue in Science Communication: The Dialogue Communication Model Requires Additional Roles from Scientists." EMBO Reports 21 (9): e51278. https://doi.org/10.15252/embr.202051278.

REPORT FROM THE COMMISSION TO THE EUROPEAN PARLIAMENT AND THE COUNCIL. 2019 report on the statistics on the use of animals for scientific purposes in the Member States of the European Union in 2015-2017. 2020 Brussels, Feb 5

Rip, A. 2018. Futures of Science and Technology in Society. Wiesbaden: Springer

Rocha, L. F. M., L. A. M. Braga, and F. B. Mota. 2020. "Gene Editing for Treatment and Prevention of Human Diseases: A Global Survey of Gene Editing-Related Researchers." Human Gene Therapy 31 (15-16): 852-62. https://doi.org/10.1089/hum.2020.136.

Roemer M. 1993. “National Health systems throughout the world.” Annu. Rev. Publ. Health14:335-53

Roever L, Zoccai GB. 2015. “Critical Appraisal of Systematic Reviews and Meta-analyses”. Evidence Based Medicine and Practice 1: e106. Doi: 10.4172/2471-9919.1000e106

Rosemann, A., A. Balen, B. Nerlich, C. Hauskeller, M. Sleeboom-Faulkner, S. Hartley, X. Zhang, and N. Lee. 2019. "Heritable Genome Editing in a Global Context: National and International Policy Challenges." Hastings Center Report 49 (3): 30-42. https://doi.org/10.1002/hast.1006.

Rotolo, Daniele and Hicks, Diana and Martin, Benjamin R.. 2015. “What Is an Emerging Technology?” Research Policy, 44(10): 1827-1843, Available at SSRN: https://ssrn.com/abstract=2564094 or http://dx.doi.org/10.2139/ssrn.2564094

Rouet P, Smih F, Jasin M. 1994. "Introduction of double-strand breaks into the genome of mouse cells by expression of a rare-cutting endonuclease”. Mol Cell Biol. 14(12):8096-106. doi: 10.1128/mcb.14.12.8096-8106.1994. PMID: 7969147; PMCID: PMC359348.

Royal Swedish Academy of Sciences. The Nobel Prize in Chemistry 2020 (Press release, 7 October 2020)

RRI Tools Coordination Team and Everis. 2016. Training Module: The RRI Toolkit – Hands-on Responsible Research and Innovation. D3.2 part of / D4.1 part of, July 26.

Rufo, F, and Ficorilli, A. 2019. “From Asilomar to Genome Editing: Research Ethics and Models of Decision”. NanoEthics 13 (3):223-232.

Ruszanov, A. and Haese, N. 2018. Deliverable D4.5. “Future of RRI”. Fostering a Transition towards Responsible Research and Innovation Systems (FoTRRIS), March 28

Sander, J. D., and J. K. Joung. 2014. "CRISPR-Cas Systems for Editing, Regulating and Targeting Genomes." Nature Biotechnology 32 (4): 347-55. https://doi.org/10.1038/nbt.2842.

Sapranauskas R, Gasiunas G, Fremaux C, Barrangou R, Horvath P, Siksnys V. 2011. “The Streptococcus thermophilus CRISPR/Cas system provides immunity in Escherichia coli”. Nucleic Acids Res. 39(21):9275-82. doi: 10.1093/nar/gkr606. Epub 2011 Aug 3. PMID: 21813460; PMCID: PMC3241640.

Sawai, T., Hatta, T., and Fujita, M. 2017. “Public attitudes in Japan towards human–animal chimeric embryo research using human induced pluripotent stem cells.” Regenerative Medicine 12:3, 233-248. https://doi.org/10.2217/rme-2016-0171

Scheifele, L. Z., & Burkett, T. 2016. “The first three years of a community lab: lessons learned and ways forward.” Journal of Microbiology & Biology Education, 17(1), 81-85. https://doi.org/10.1128/jmbe.v17i1.1013

Scheufele, D., Krause, N., Freiling, I., and Brossard, D. “What we know about effective public engagement on CRISPR and beyond”. PNAS, 118 (22) e2004835117. https://doi.org/10.1073/pnas.2004835117

Schuppli CA, Weary DM. 2010. “Attitudes towards the use of genetically modified animals in research.” Public Underst Sci. 19(6):686-97.

Sekine R, Kawata T, Muramoto T. 2018. “CRISPR/Cas9 mediated targeting of multiple genes in Dictyostelium”. Sci Rep. 8(1):8471. doi: 10.1038/s41598-018-26756-z. PMID: 29855514; PMCID: PMC5981456.

Scheifele LZ, Burkett T. 2016. “The First Three Years of a Community Lab: Lessons Learned and Ways Forward”. J Microbiol Biol Educ;17(1):81-5. doi: 10.1128/jmbe.v17i1.1013. PMID: 27047597; PMCID: PMC4798823

Schlosser, R. 2007. “Appraising the Quality of Systematic Reviews”. Focus, TECHNICAL BRIEF NO. 17

Scudellari, M. 2019. “Hijacking evolution”. Springer Nature. 571

Shelley-Egan, C., M. D. Gjefsen, and R. Nydal. 2020. "Consolidating RRI and Open Science: Understanding the Potential for Transformative Change." Life Sciences, Society and Policy 16 (7): https://doi.org/10.1186/s40504-020-00103-5.

Siccama, C. J., and Stacy Penna. 2008. "Enhancing Validity of a Qualitative Dissertation Research Study by Using Nvivo." Qualitative Research Journal 8 (2): 91-103. https://doi.org/10.3316/qrj0802091.

Singh K, Evens H, Nair N, Rincon MY, Sarcar S, Samara-Kuko E, et al. 2018. “Efficient In Vivo Liver-Directed Gene Editing Using CRISPR/Cas9.” Mol Ther.26(5):1241-54.

Sinkiewicz, M. 2020. “From a Policy Problem to a Research Question: Getting It Right Together”. In: Sucha V, Sinkiewicz M editors. Science for Policy Handbook. Elsevier: European Union; 2020. pp. 53-61

So, D., R. Sladek, and Y. Joly. 2021. "Assessing Public Opinions on the Likelihood and Permissibility of Gene Editing through Construal Level Theory." New Genetics and Society: 1-25. https://doi.org/10.1080/14636778.2020.1868985.

Stadtmauer EA, Fraietta JA, Davis MM, Cohen AD, Weber KL, Lancaster E, Mangan PA, Kulikovskaya I, Gupta M, Chen F, Tian L, Gonzalez VE, Xu J, Jung IY, Melenhorst JJ, Plesa G, Shea J, Matlawski T, Cervini A, Gaymon AL, Desjardins S, Lamontagne A, Salas-Mckee J, Fesnak A, Siegel DL, Levine BL, Jadlowsky JK, Young RM, Chew A, Hwang WT, Hexner EO, Carreno BM, Nobles CL, Bushman FD, Parker KR, Qi Y, Satpathy AT, Chang HY, Zhao Y, Lacey SF, June CH. 2020. “CRISPR-engineered T cells in patients with refractory cancer.” Science.28;367(6481):eaba7365.

Stockemer D. 2019. A short introduction to survey research. In: Stockerner D editors. Quantitative methods for the social sciences: A practical introduction with examples in SPSS and Stata. Ottawa: Springer. pp.23-35

Stockemer, D. 2019. Conducting a survey. In: Stockerner D editors. Quantitative methods for the social sciences: A practical introduction with examples in SPSS and Stata. Ottawa: Springer pp.57-72

Störmer, E., Bontoux, L., Krzysztofowicz, M., Florescu, E., Bock, A. and Scapolo, F. 2020. “Foresight – Using Science and Evidence to Anticipate and Shape the Future”. In: Sucha V, Sinkiewicz M editors. Science for Policy Handbook. Elsevier: European Union; pp. 129-142

Strecht, M and Quintanilha, A. 2016. “Of Responsible Research—Exploring the Science-Society Dialogue in Undergraduate Training Within the Life Sciences”. *Biochemistry and Molecular Biology Education,* 45(1): 46-52. DOI 10.1002/bmb.20988

Sucha, V. and Dewar, M. 2020. “Institutional Framework for the Science–Policy Interaction”. In: Sucha V, Sinkiewicz M editors. Science for Policy Handbook. Elsevier: European Union; pp. 21-30

Tassone, V and Eppink, H. 2016. “Deliverable 2.3. The EnRRICH tool for educators: (Re-) Designing curricula in higher education from a “Responsible Research and Innovation” perspective”. Wageningen University

Taylor, I. 2021. “CRISPR: A guide to the health revolution that will define the 21st century”. Science Focus, November 10

The Danish Council on Ethics. 2016. “Statement from the Danish Council on Ethics on genetic modification of future humans: In response to advances in the CRISPR technology”,

The Netherlands Commission on Genetic Modification COGEM. CRISPR & Animals: Implications of genome editing for policy and society. 2018 Policy Report CGM/180501-01.

The Nuremberg Code (1947) .1996. BMJ; 313 :1448doi:10.1136/bmj.313.7070.1448

Thomas, K.R., and Capecchi, M. 1987. “Site-directed mutagenesis by gene targeting in mouse embryo-derived stem cells”. Cell. 51(3): 503-12. https://doi.org/ 10.1016/0092-8674(87)90646-5.

Timmermans J, Blok V, Braun R, Wesselink R, Nielsen R. 2020. “Social labs as an inclusive methodology to implement and study social change: the case of responsible research and innovation”. Journal of Responsible Innovation 7(3): 410-426

Topp, L., Mair, D., Smillie, L. and Cairney, P. 2020. “Skills for Co-creation”. In: Sucha V, Sinkiewicz M editors. Science for Policy Handbook. Elsevier: European Union; pp. 33-42

Ueland O, Gunnlaugsdottir H, Holm F, Kalogeras N, Leino O, Luteijn JM, et al. 2012. “State of the art in benefit-risk analysis: consumer perception.” Food Chem Toxicol. 50(1):67-76.

UN Educational, Scientific and Cultural Organisation (UNESCO), Universal Declaration on the Human Genome and Human Rights. 1997.

United States. 1978. “The belmont report: ethical principles and guidelines for the protection of human subjects of research”. The National Commission for the Protection of Human Subjects of Biomedical and Behavioral Research

United Nations. 2015. Transforming our World: The 2030 Agenda for Sustainable Development. A/RES/70/1

United Nations Human Rights. 2019. “Empowerment, Inclusion, Equality”. Office of the High Commissioner.

Urnov FD. 2018. “Genome Editing B.C. (Before CRISPR): Lasting Lessons from the "Old Testament".” CRISPR J.1(1):34-46.

Urnov FD, Miller JC, Lee YL, Beausejour CM, Rock JM, Augustus S, Jamieson AC, Porteus MH, Gregory PD, Holmes MC. 2005. “Highly efficient endogenous human gene correction using designed zinc-finger nucleases”. Nature.435(7042):646-51. doi: 10.1038/nature03556. PMID: 15806097.

VanderStoep, S.W., and D.D. Johnston, D.D. 2009. Research methods for everyday life: blending qualitative and quantitative approaches. San Francisco: Jossey-Bass

von Schomberg, R. 2013. "A vision of Responsible Research and Innovation". In Responsible Innovation: Managing the Responsible emergence of science and innovation in society, edited by R. Owen, J. Bessant and M. Heintz, 51-74. London: Wiley

von Schomberg, R. 2011. “Prospects for technology assessment in a framework of responsible research and innovation”. In: M. Dusseldorp, R. Beecroft editors. Springer: Technikfolgen abschätzen lehren, DOI 10.1007/978-3-531-93468-6_2

Vienna Declaration and Programme of Action. 1993. World Conference of Human Rights, 25 June.

Wahlberg, A., D. Dong, P. Song, and Z. Jianfeng. 2021. “The Platforming of Human Embryo Editing: Prospecting ‘Disease Free’ Futures.” *New Genetics and Society* 40 (4): 367-383. https://doi.org/10.1080/14636778.2021.1997578

Watanabe D, Saito Y, Tsuda M, Ohsawa R. 2020. “Increased awareness and decreased acceptance of genome-editing technology: The impact of the Chinese twin babies.” PLOS ONE 15(9): e0238128. https://doi.org/10.1371/journal.pone.0238128

WHO Expert Advisory Committee on Developing Global Standards for Governance and Oversight of Human Genome Editing. 2021. Human Genome Editing: position paper. Geneva: World Health Organization

Wieser, B. 2019. “Teaching the Social Construction of Technology: Time to Revise or Time to Forget?” Proceedings of the STS Conference Graz, pp:507-524. DOI: 10.3217/978-3-85125-668-0-28

Wirz, C. D., D. A. Scheufele, and D. Brossard. "Societal Debates About Emerging Genetic Technologies: Toward a Science of Public Engagement." Environmental Communication 14 (7): 859-64. https://doi.org/10.1080/17524032.2020.1811478.

World Medical Association Declaration of Helsinki: ethical principles for medical research involving human subjects. JAMA. 2013 Nov 27;310(20):2191-4. doi: 10.1001/jama.2013.281053.

Xie F, Ye L, Chang JC, Beyer AI, Wang J, Muench MO, et al. 2014. “Seamless gene correction of beta-thalassemia mutations in patient-specific iPSCs using CRISPR/Cas9 and piggyBac.” Genome Res.;24(9):1526-33.

Yan, S., Tu, Z., Li, S., Li, X. 2018. “Use of CRISPR/Cas9 to model brain diseases”. Progress in Neuro-Psychopharmacology and Biological Psychiatry. 81: 488-492. https://doi.org/10.1016/j.pnpbp.2017.04.003.

Yang S, Chang R, Yang H, Zhao T, Hong Y, Kong HE, et al. 2017. “CRISPR/Cas9-mediated gene editing ameliorates neurotoxicity in mouse model of Huntington's disease.” J Clin Invest. 2017;127(7):2719-24.

Young CS, Mokhonova E, Quinonez M, Pyle AD, Spencer MJ. 2017. “Creation of a Novel Humanized Dystrophic Mouse Model of Duchenne Muscular Dystrophy and Application of a CRISPR/Cas9 Gene Editing Therapy.” J Neuromuscul Dis. 4(2):139-45.

Yunes, M. C., D. L. Teixeira, M. A. G. von Keyserlingk, and M. J. Hotzel. 2019. "Is Gene Editing an Acceptable Alternative to Castration in Pigs?". PLoS One 14 (6): e0218176. https://doi.org/10.1371/journal.pone.0218176.

Zhang Z, Zhang Y, Gao F, Han S, Cheah KS, Tse HF, et al. 2017. “CRISPR/Cas9 Genome-Editing System in Human Stem Cells: Current Status and Future Prospects.” Mol Ther Nucleic Acids.9:230-41.

Zhong A, Darren B, Loiseau B, He LQB, Chang T, Hill J, Dimaras H. 2021. “Ethical, social, and cultural issues related to clinical genetic testing and counseling in low- and middle-income countries: a systematic review.” Genet Med. 23(12):2270-2280. doi: 10.1038/s41436-018-0090-9. Epub 2018 Aug 3. PMID: 30072741.

Appendices

Appendix I. Conceptual matrix for interview guide

Manuscript 1

Conceptual matrix

Concept		**Themes**	**Facilitation**	**Expectations**
	Soft dimensions			
Technical challenges		1. Scenarios: Future (5 years) + backcast	<u>1.Hypothesizing and backcasting</u> **Aim: To address the interviewee's perspective on the technical limitations impact in current and future and how that might modulate genome editing potential** 1.1. Ask how the interviewee sees the potential of genome editing according to its tendency to consider a hype or not. ○ Where would you think genome editing and particularly CRISPR/Cas9 will be in 5 years? ○ Which reasons lead you to think that way? ○ Where do you think there's still work to be done?	The interviewee will be exploring current technical limitations that are significant and might have an influence in the future for the development of genome editing on potential applications. The parallel between the "actual" and "next 5 years" will recall the interviewee for an observation on the possibility to infer about the promise of

			1.2. Ask the interviewee to choose the technical challenges potentially more relevant for genome editing success in the future (exercise) ○ Which technical limitations/challenges would you consider the most relevant now and in that same future? And why?	genome editing and the struggles still being faced. The interviewee will identify and weigh his/her own challenges within the laboratory and be able to choose and justify (perhaps the 2 most relevant) the ones that might play a role on genome editing progression towards success in the future.

Feasible applications	Germline and somatic gene editing Technical and ethical issues Human and animal perspective	1. Future (revisit)	2. Redefining genome editing in the context of feasible applications **Aim: To allow the interviewee to infer on the meaning of challenges and concerns faced within somatic and germline cell editing research** 2.1. Ask the interviewee to elaborate on the application of genome editing in somatic and germline cells in a rather ethical perspective ○ You mentioned some applications where technical challenges might play a role in the implementation of genome editing. Are those the only ones? ○ Literature mentions and makes a distinction on applications of genome editing between germline and somatic cell lines. Do you agree with such distinction? And why?	The interviewee will be focused on the challenges of germline and somatic cell editing and on how that is linked and can be translated in not just technical but rather ethical concerns. A wider point-of-view will be explored within human research but focus will be aimed for thoughts on genome editing in animal research settings.

			○ So, I've a list of cases where some hypothetical applications are represented: Cases: • **Somatic cell genome editing (Clinical trials): *In vivo* genome editing to target cancer and genetic diseases** • **Germline cell genome editing: Birth of an *in vitro* genome-edited embryo** • **Gene drive: A new gene-drive specie avoids insecticide use** • **Crops resistant to biotic/abiotic stress: Commercialization of brand new gene-edited species resistant to pathogens** • **Xenotransplantation: Transplantation of a gene-edited organ into a human's body** • **Modelling brain disorders: Gene-edited non-human primate to model a human disorder affecting a cognitive function** • **Large animals genome editing :Gene-edited horses with enhanced speed and muscles** ○ From those applications I've listed and showed, how would you rank them in terms of the more closest/realistic to happen in the near future? Why do you think that? There's no right or wrong answer, you just have to justify.	Elaboration of speculative scenarios within the ethically more relevant issues will be prompted by means of the interviewee to discuss on their importance for genome editing progression into applications in the future.

			2.2. Ask the interviewee to speculate about the most significant ethical concerns for genome editing progression towards the future ○ Now, let's think that such technical challenges have been overcome. Let's now focus on the ones that are most desirable and/or valuable in the future and rank them accordingly. Why do you think that? ○ Are you aware about how your choices are leveled with existing concerns? How would you interpret that? ○ Which of those concerns might be more significant in the following years of genome editing implementation and debate? In case the interviewee doesn't mention ethical concerns about somatic and germline cell editing, ask follow-up questions such as: - Do you recognize other reviewed concerns in regards to somatic and germline editing?	The interviewee will focus more immediately on the technical challenges to be overcome in the actual scenario in the first ranking exercise. If the interviewee explores the ethical issues associated with it, these considerations will be taken to 2.2 and furthermore to 3. parts. Upon redirection of the cases to a future perspective and how they might be seen as desirable or not, the

			- Are you familiarized with them? In what sense? If still not identifying ethical concerns, a more direct approached might be seek: - Apart from technical concerns, do you also recognize ethical ones when dealing with somatic and/or germline cell editing?	interviewee will be closer of the ethical issues that can lead to a change on his/her own ranking process. This will introduce the ethics sub dimension and allow for a new conversation in an ethical perspective. This will also allow for understanding on the interviewee perspective of the current concerns in regards to ethical issues on genome editing and take him/her for the next approach in 3.

Genome editing and animal research	3. Ethics on gene-edited animal research modelling	3. - 3Rs and boundaries between human and animal	3. Establishing a realistic picture of the potential and limitations of genome editing in the context of ethical consequences on animal applications **Aim: To allow the interviewee to explore the ethical implications of editing animals on a more immediate (welfare and animal number) and on a more distant (transgressing species borders) perspective.** 3.1. Ask the interviewee to recall on the exercise of the cases regarding applications to further discuss animal welfare issues related with gene-edited animals in research ○ I would like to focus a little more on the exercise you executed earlier with particular emphasis in animal applications. What do you expect to happen in relation to animal number with such genome editing strategies use? Will that illustrate a correlation with scientific impact? ○ Do you only see that increase on currently used animals in research (such as rodents)? ○ Apart from animal number, do you also recognize an alteration on the number of animal models to be used for research purpose?	The interviewee will be able to take on the exercise in 2. and explore genome editing in animal research in regards to animal welfare by envisioning of 3Rs policy (particularly Refinement and Reduction). The interviewee will also be able to infer on the rise of new potential species that might be used in research and the meaning of such events. Thoughts on the impact of new animal species in disease modelling and a translation for biomedical research in the present

			○ What impact can this increase in animal number and species have in the future? If the interviewee doesn't recognize an increase in animal number, point right away for the number of species increase question. If the interviewee doesn't recognize an increase in species number, try an approach using animal welfare as a basis whether by asking: - What do you think genome editing in animal research will mean in terms of animal welfare (more vague) - Which of the 3Rs will be affected by genome editing practices? (more specific) 3.2. Upon recalling of the same exercise and the previous considerations in 3.1. ask the interviewee to elaborate on the ethical issues that can be posed in terms of the boundaries between animal and human. ○ Let's assume once more the examples of modification of species like the cases of	and future will be raised and discussed. The interviewee is expected to recognize a potential increase in animal number or at least, in species number in the future through genome editing applications. If such doesn't happen, a more direct approach whether in a vaguer or more specific way will allow the interviewee to dissert on the animal welfare implications of genome editing and how the 3Rs policy can be influenced by such practices. - In a replacement argument, alternative strategies such as the use of iPSCs, organoids, biobanks of human tissues, "brain on a chip" derived from iPSCs - In a refinement argument, problems at the level of infrastructures facilities and cost can be raised - In a reduction argument, no increase of animal number will be associated perhaps to

			pigs, non-human primates and let's include rodents as well. Do you consider that the boundaries between human and animal can be surpassed? If so, what consequences might that have for the animals, humans and the environment? In case the interviewee doesn't see a problem on this, the question should be focused on the consequences of high level of modification of species: ○ Given that germline editing approach is a strategy for the creation of new species, particularly for disease modelling, what does might represent for animal descendants in terms of welfare, human health and environmental safety?	breeding (hard in larger animals) or to cell grafting strategies The interviewee will elaborate on the possibility for multiplex modification of animals using perhaps the "the pig case" but also by exploring germline editing of animals in research whether with low size (zebrafish, rodents) or with high size (non-human primates) animals. Thoughts on creation of new animal species inexistent in nature and introduction of human disease phenotypes in certain animals will be raised and discussed. Several questions within fairness, human-animal satisfaction, sake of research and impact in the environment can also be raised. In case the interviewee doesn't raise concerns, the questions should be directed with the aim to make him/her speak freely on animal welfare,

			A more broad question that I would like to ask as last: - Which other criteria should be regarded for a successful questioning on the level of ethical implications of genome editing in animals?	human health and environmental consequences upon new species creation with human genes in modified animals and their offspring. The interviewee will share his/her own thoughts on the ethical questions that are still necessary to be asked and provide arguments for such questioning in a quick manner.
			What other questions would you have liked to have answered?	The interviewee will offer some feedback for further interviews by sharing his/her own point-of-view.

Appendix II. Interview guide for interviews with researchers
Manuscript 1

Interview Guide

Facilitation
1.Hypothesizing and backcasting 1.1 ○ **Where would you think genome editing and particularly CRISPR/Cas9 will be in 5 years? Which reasons lead you to think that way?** ○ **Given what you mentioned, where do you think there's still work to be done?** 1.2 ○ **Which technical limitations/challenges would you consider the most relevant now and in that same future? And why?** 2. Redefining genome editing in the context of feasible applications 2.1. ○ **You mentioned some applications where technical challenges might play a role in the implementation of genome editing. Are there any other technical challenges associated to other applications?**

- **Literature mentions and makes a distinction on applications of genome editing between germline and somatic cell lines. What do you think it justifies such distinction?**

- **So, I've a list of cases where some hypothetical applications are represented:**

- Somatic cell genome editing (Clinical trials): *In vivo* genome editing to target cancer and genetic diseases
- Germline cell genome editing: Birth of an *in vitro* genome-edited human embryo
- Gene drive: A new gene-drive insect species avoids insecticide use in agriculture
- Crops resistant to biotic/abiotic stress: Commercialization of brand new gene-edited species resistant to pathogens
- Xenotransplantation: Transplantation of a gene-edited organ into a human's body
- Modelling brain disorders: Gene-edited non-human primate to model a human disorder affecting a cognitive function
- Large animals genome editing : Gene-edited horses with enhanced speed and muscles

- **From those applications I've listed and showed, how would you rank them in terms of the more closest/realistic to happen in the near future? Why do you think that? There's no right or wrong answer, you just have to justify.**

2.2.

- **Now, let's think that such technical challenges have been overcome. Let's now focus on the ones that are most**

desirable and/or valuable in the future and rank them accordingly. Why do you think that?

- **Do you recognize other reviewed concerns in regards to somatic and germline editing where there's a debate going on? How would you interpret them?**
- **Which of those concerns might be more significant in the following years of genome editing implementation and debate?**

- What do you understand about the ethical concerns you mentioned?
- Are you familiarized with some reviewed concerns? In what sense?

- Apart from technical concerns, do you also recognize ethical ones when dealing with somatic and/or germline cell editing?

3. Establishing a realistic picture of the potential and limitations of genome editing in the context of ethical consequences on animal applications

3.1.

- **I would like to focus a little more on the exercise you executed earlier with particular emphasis in animal applications. What do you expect to happen in relation to animal number with such genome editing strategies use? Will that illustrate a correlation with scientific impact?**
- **Do you only see that increase on currently used animals in research (such as rodents or zebrafish)?**

- **Apart from animal number, do you also recognize an alteration on the number of animal models to be used for research purpose?**
- **What impact can this increase in animal number and species have in the future?**

- What do you think genome editing imposition in animal research will mean in terms of animal welfare?
- How will the 3Rs be affected by genome editing practices?

3.2.

- **Let's assume once more the examples of modification of species like the cases of pigs, non-human primates and let's also include rodents and zebrafish. What do you have to say about the boundaries between human and animal? Can they ever be surpassed?**

- **(We've seen some literature where groups use NHP models to study regulation of sympathetic nervous activity using CRISPR. Others used mutant Rhesus with Rett syndrome ending with autism-like behavior…)**
- **(We are also aware of studies where iPSCs cells from post-synaptic neurons have been gene-edited and disease modelling of neurodegenerative disorders like AD, PD and neuropsychiatric illnesses etc)**
  - **If so, what consequences might that have for the animals, humans and lately, in the environment?**

- Given that germline editing approach is a strategy for the creation of new species, particularly for disease modelling, what does might represent for animal descendants in terms of welfare, human health and environmental safety?

A more broad question that I would like to ask as last:

- **Which other criteria should be regarded for a successful questioning on the level of ethical implications of genome editing in animals?**
- **What other concerns about genome editing do you have in general, despite from animal-related ones?**

- **What other questions would you have liked to have answered?**

Appendix III. Codebook from content analysis of interviews with researchers
Manuscript 1

Codebook – 1st and 2nd order codes

1st order coding categories	Description	Color
Technical challenges	Technical challenges	
and Technical status	Status of the technology so far (what is being implemented in terms of techniques	
Technical-ethical (safety)	Safety issues associated with implementation of CRISPR-Cas9 technology posed by technical limitations and its repercussions in human health, animal welfare and environment balance	
Feasible Applications	Genome editing technologies implementation among current and future applications	
Ethical concerns	Ethical concerns	
and Regulation	Genome editing and CRISPR-Cas9 regulation among different countries and entities	
Consequences for animal research	Impact of genome editing strategies imposition for animals used in research, namely animal welfare and other aspects of the 3Rs	

Chimeras and transgressing species borders	Consequences of mixing animal species genomes, through KIs for the creation of chimeras (animal-animal) and surpassing animal-human boundaries	
Other (Scientific potential)	Scientific potential perspective and impact of genome editing and CRISPR-Cas9 technologies in the future	
Other (Impact on research quality)	How different variables in regards to IP, licenses, funding, reproducibility might impact on the quality of research not only but also for genome editing	

List of 2nd order codes following sub-coding of 1st order codes identified in the first codebook

1st order coding categories	2nd order coding categories	Description	Color
Technical challenges	Efficiency	Success of on-target editing in cells	
	Off-target effects	Undesired targeting effect upon editing	
	Efficacy	Easiness and speed of genome editing	

	Delivery	Ability to deliver editing tools on cells, tissue, organ, etc	
	Fidelity	Precision and specificity of nuclease and DNA template integration upon genome editing	
	Genome-related	Targeting or editing influenced by regulation and interaction in the genome and epigenome	
	Other (tech)	Other technicalities related or not with genome editing	
Ethical concerns	Cultural differences	Points-of-view defined by culture, religion and traditions differences among people, countries, etc	
	Germline vs somatic	Editing consequences between germline and somatic cell lines	
	Lack of knowledge - Unknown effects	Unknown impact of lack of knowledge in editing	

	Lack of regulation	Absence or deficitary regulation imposed regarding Genome editing	
	Overriding biology	How technology can take over evolution and species biology	
	Public attitude - Engagement	Attitude from the public towards genome editing and their engagement in debate	
	Purpose	Meaning or benefit of the technology	
	Social justice	Impact of genome editing in socio-economic status	
	Other		
Consequences for animal research	Costs	Cost of genome editing for animal research or farming in both economic and ecological scenarios	

	Decision on animal waste vs knowledge	Balance between editing of animals for increasing knowledge and their waste as a resource
	Diversity of animal models	Impact of genome editing on animal models diversity
	Quality - specificity of animal models	Importance of the quality or specificity of a given animal model
	Reduction	Alteration on the number of animals towards 3R policy
	Refinement	Impact of genome editing in animal welfare/health
	Replacement	Substitution of animals or animal models for other in vitro and high technology models
	Reproducibility	Reliability of animal models and their translational significance and relevance

	Time	Time-wise revolution of genome editing in animal models generation both in KOs and KIs approaches	

Appendix IV. Audio-dictate transcribing automated tool (AUDIT).
Presented at cool tools for science event

Abstract

AUDIT: Audio-Dictate Transcribing Automated Tool

Common methods of transcription involve recurring to professionally trained transcribers who are able to accurately transcribe *ad verbatim* from audio but this process is usually time-consuming and their hiring even when online is costly [1,2,3]. Other very accurate software tools that convert speech into text like Dragon NaturallySpeaking also require an expensive license and are time-dependent on one's transcribing abilities and experience [4]. Importantly, new automated methods which require audio deposit at a website started to emerge as time-wise option for journalists and social scientists [3,5]. However, when we deal with confidential or sensitive audio that needs protection like in social science research, sharing it with hired transcribers or depositing it at a website to be either transcribed by a human or a non-human automated tool are not options, and an alternative automated method is therefore needed [5]. The method devised here requires the use of a free software program and a valid active license of Microsoft Office that researchers often have access to. It works by joining a method of audio conversion in the computer audio settings with a tool that converts speech into text automatically. This way, any regular researcher is able to safely convert any saved audio file into text without additional costs. This method, nevertheless, doesn't excuse the presence of a human individual to insert manual punctuation and perform word correction. Accuracy of the transcription output also improves with the sound quality of the audio file and the clearness of the speaker's accent and voice. This Cool tool represents an advance in social science methodology overcoming a frequent

time-limiting step by enhancing the speed of transcription while simultaneously reducing the costs of the process and preserving quality control and confidentiality of the audio data. A demonstration of the tool's features will be performed at the event.

Doi:

References

[1] Fagan J. What is the going rate for transcription services? Prices for transcription. 2019 Jan 1 [cited 1 Feb 2021]. In: Advice [Internet]. https://www.tptranscription.co.uk/what-is-the-going-rate-for-transcription-services-prices-for-transcription/

[2] Available from: https://www.opaltranscriptionservices.com/transcription-rates/

[3] LaForme R. The best automatic transcription tools for journalists. 2017 Nov 20 [cited 1 Feb 2021]. In: Poynter [Internet]. https://www.poynter.org/tech-tools/2017/the-best-automatic-transcription-tools-for-journalists/

[4] Available from: https://www.nuance.com/dragon.html

[5] Taster. Disrupting transcription – How automation is transforming a foundational research method. 2019 Sep 17 [cited 1 Feb 2021]. In: LSE blogs [Internet]. London: Impact of social sciences. Available from: https://blogs.lse.ac.uk/impactofsocialsciences/2019/09/17/disrupting-transcription-how-technology-is-transforming-a-foundational-research-method/

Appendix V. Informed consent used for interviews with researchers
Manuscript 1

INFORMED CONSENT

Please read the following information carefully, If you think that something is not correct or is unclear, do not hesitate to ask for more information. If you agree, please sign this document.

Study title Redacted for blinding

Context: This study is part of the Redacted for blinding project Redacted for blinding and a Ph D project in Biomedical Sciences at the under the supervision of Redacted for blinding , Redacted for blinding Redacted for blinding and Redacted for blinding . The regulation of the Redacted for blinding Ethics Committee will be followed.

Outline of the study: This study will involve interviews with audio recording of experts on genome editing from a scientific, ethical and/or regulatory perspective. Experts will be recruited through the professional network of the research team. The interview method will consist in a set of questions to be asked to the interviewee where the participant will share thoughts, opinions, points-of-view and arguments on the topic mentioned and some exercises might also be proposed. All the information will be recorded for later data transcription and further analysis. The time for recording can vary between 1h to 2h in total and will only consist in audio. Interviews will be done face-to-face or over Skype/telephone. Audio recordings will be destroyed as soon as interview analysis is completed**.**

Conditions and funding: This study is funded by Horizon 2020 under the Marie Skłodowska-Curie Actions program (MSCA) which refers to a European Training Network (ETN) with the acronym Redacted for blinding Every participant will participate voluntarily in the interviews and receive no monetary compensation. The study is approved Redacted for blinding

Confidentiality and anonymity: All the data collected will be kept under confidentiality for exclusive purpose of the present study and will only be used within the project to which the study is associated with. Audio recordings will not contain any reference to interviewee identity, their transcripts are stored without association to interviewee identity and no database containing interviewee identity is created. Personal data will not

become public. Participants will be invited directly through contact information available whether by e-mail, phone call or in person, if necessary. No data will be shared with persons outside the team of researchers named under the heading Context above.

Name and contact details of the researcher to whom consent is given: Redacted for blinding

Signature: …

~~~~~~

I declare that I have read and understood this document, as well as the additional information that were given by the person signing above, and that I consider them sufficient. I have been guaranteed the possibility to withdraw from the study at any time, without any consequences for me. In this way, I accept to participate in the study and allow the research use of the information that I am providing voluntarily under the conditions specified in this document.

I understand that I may be quoted in research reports resulting from this study and I agree to be quoted directly if my name is not published and a made-up name (pseudonym) is used.

Name: _____

Signature: Date: __ /__ /_____

THIS DOCUMENT CONSISTING IN ONE (1) PAGE IS ISSUED IN TWO COPIES, ONE FOR THE RESEARCHER AND ONE FOR THE CONSENTING PARTICIPANT.
~~~~~~

Appendix VI. Figures S1 and S2 showing results from ranking cases with researchers
Manuscript 1

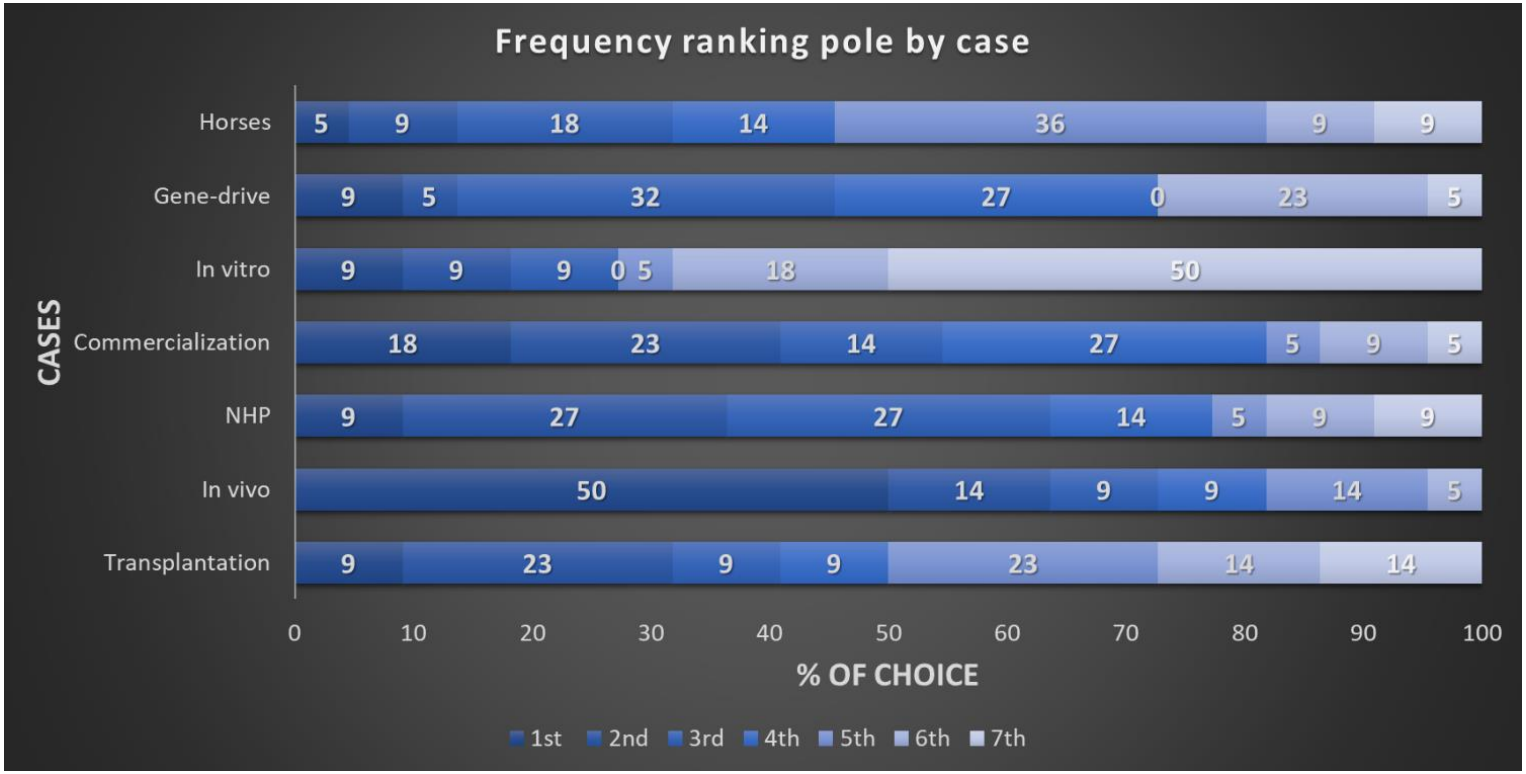


Figure S1. Relative frequency of interviewees that chose a certain ranking position for a given ranking case under a realistic perspective. Each number in the graph represents the % of interviewees that choose a ranking position from 1st to 7th place for each of the 7 ranking cases. The colour gradient of the figure goes from the 1st to the 7th place. Therefore, the darkest orange indicates the 1st position and the most realistic according with interviewees perspective and the 7th lightest orange position represents the least realistic in interviewees' view.

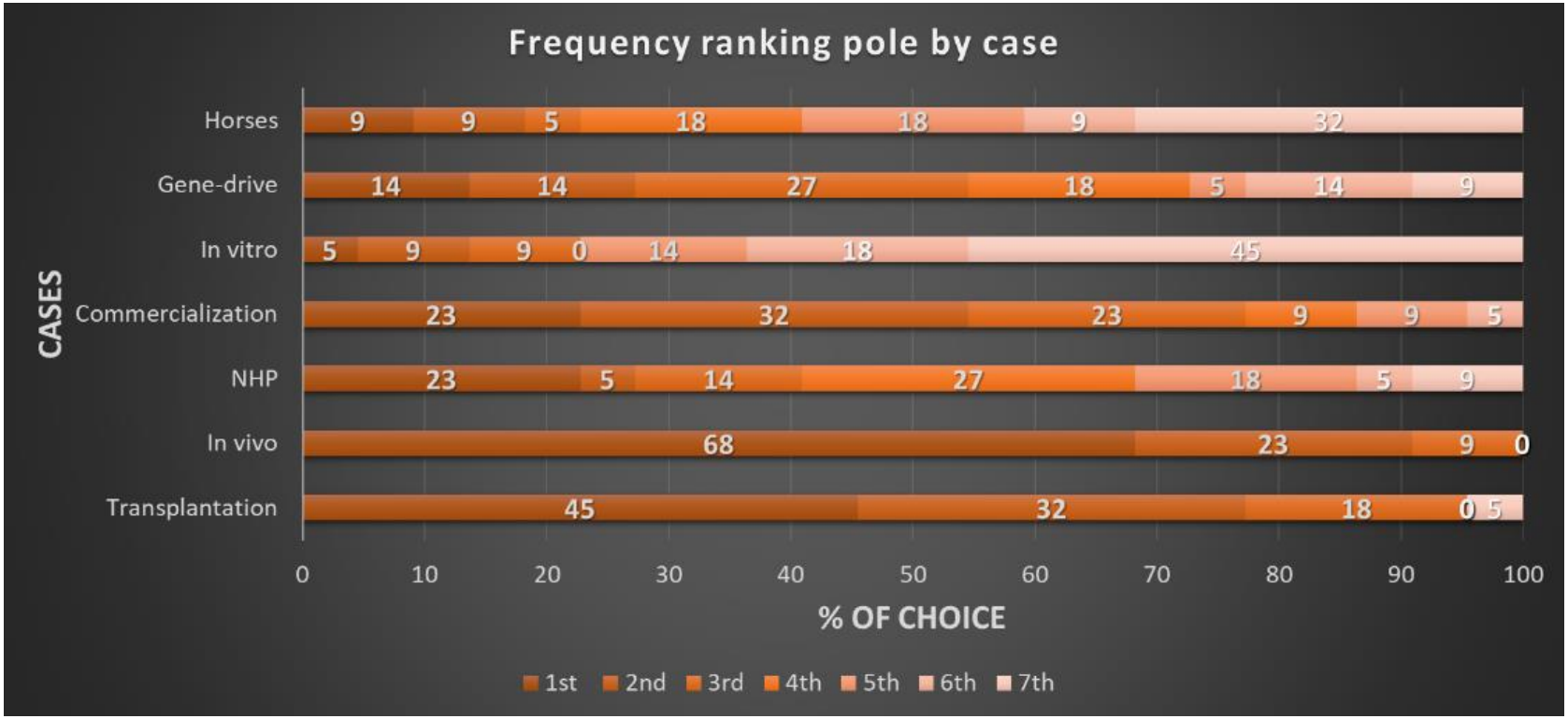


Figure S2. Relative frequency of interviewees that chose a certain ranking position for a given ranking case under a desirable perspective. Each number in the graph represents the % of interviewees that choose a ranking position from 1st to 7th place for each of the 7 ranking cases. The colour gradient of the figure goes from the 1st to the 7th place. Therefore, the darkest orange indicates the 1st position and the most desirable according with interviewees perspective and the 7th lightest orange position represents the least desirable in interviewees' view.

Appendix VI. Table S1 showing the number of approvers of GE among citizens in pre-CRISPR and CRISPR periods
Manuscript 2

Pre-CRISPR (1987-2012)

		Approvers (in a total of 10 respondents)						
		Genetic modification of animals			Genetic modification of humans			
Authors (Year)	**Country(ies)**	**Transplants and/or medicines**	**Meat (Pork, Sheep, Cow)**	**Milk (Cow, Sheep)**	**Somatic (disease)**	**Somatic (enhancement)**	**Germline (disease)**	**Germline (enhancement)**
Office of Technology Assessment (OTA) (1987) [34]	US		7 (farm animals)		8 ***Prevent – 8 9 - children	5	***Prevent – 8 8 (non-fatal)	4 (intelligence), 4 (physical)
Macer DRJ (1992)	Japan	-	-	-	5 7 - children	-	-	-
Macer DRJ et al (1995)	Asia/Oceania				10 (TH), 9 (NZ, AU and IS), 8 (J and RU), 7 (IN) ***Prevent	More ethical 8 (TH), 6 (IN), 3 (NZ, AU, RU and IS), 2 (J)	10 (TH), 9 (AU, IN), 8 (NZ, J, RU and IS) Non-fatal 9 (TH), 8 (AU, NZ), 7	Physical 8 (TH), 6 (IN) 4 (RU), 3 (AU and J), 2 (NZ and IS)

					8 (NZ, AU, IN, TH, RU and IS), 7 (J) Children 8 (AU, TH, IN, NZ and IS), 7 (J), 6 (RU)		(RU and IS), 6 (J, IN)	Intelligence 7 (TH and IN), 3 (AU, J and RU), 2 (NZ and IS)
Eurobarometer 35.1 (1991)	EC12	9	4	4	7	-	-	-
Eurobarometer 39.1 (1993)	EC12	-	4	4	7	-	-	-
		Approvers (in a total of 10 respondents)						
		Genetic modification of animals			**Genetic modification of humans**			
Authors (Year)	**Country(ies)**	**Transplants and/or medicines**	**Meat (Pork, Sheep, Cow)**	**Milk (Cow, Sheep)**	**Somatic (disease)**	**Somatic (enhancement)**	**Germline (disease)**	**Germline (enhancement)**
Eurobarometer 46.1 (1996)	EU15	4 - mice and pigs	-	-	-	-	-	-

Eurobarometer 58.0 (2002)	EU15	4	-	-	-	-	-	-
Eurobarometer 63.1 (2005)	EU23	-	3	-	5	-	-	-
Sato et al (2006)	Japan	-	-	-	Opinion formation increased greatly after the first gene therapy success (only 56% formed an opinion)			
Barnett et al (2007)	UK	-	-	-	Trust in government and people in charge reveals favoring to allow gene therapy Belief in public involvement, awareness, interest and levels of education don't favor to allow for gene therapy			
Eurobarometer 73.1 (2010)	EU27	6	-	-	6	5	4	-
Ng MAC et al (2000)	Japan	5 (mice) 3 (pigs)	5	4	7 ***Prevent - 6	-	7	5 3 (physical) 2 (intelligence)
Macer DRJ et al (2000)	Asia/Oceania	-	8 (TH), 7 (IN), 6 (AU and J), 5 (NZ), 4 (IS), 3 (RU)	8 (TH), 7 (IN), 4 (NZ, AU, J and IS), 2 (RU)	10 (TH), 9 (NZ, AU and IS), 8 (J and RU), 7 (IN)	More ethical 8 (TH), 6 (IN), 3 (NZ, AU, RU and IS), 2 (J)	10 (TH), 9 (AU and IN), 8 (NZ, J, RU and IS)	Physical 8 (TH), 6 (IN), 4 (RU), 3 (AU

					***Prevent 8 (NZ, AU, IN, TH, RU and IS), 7 (J)		Non-fatal 9 (TH), 8 (AU, NZ), 7 (RU and IS), 6 (J, IN)	and J), 2 (NZ, IS) Intelligence 7 (TH and IN), 3 (RU, AU and J), 2 (NZ and IS)
		Approvers (in a total of 10 respondents)						
		Genetic modification of animals			**Genetic modification of humans**			
Authors (Year)	**Country(ies)**	**Transplants and/or medicines**	**Meat (Pork, Sheep, Cow)**	**Milk (Cow, Sheep)**	**Somatic (disease)**	**Somatic (enhancement)**	**Germline (disease)**	**Germline (enhancement)**
Evans MDR et al (2005)	Australia	-	-	-	-	-	4 (Serious defect) 3 (Minor defect) 2 (Aggression and violence)	1 - cosmetic

Cook AJ et al (2004)	New Zealand	5	-	-	4	-	-	-
Human Genetics Commission (2001)	United Kingdom	-	-	-	9 8 - children	-	-	-
Sturgis P et al (2005)	United Kingdom	-	-	-	9 – cystic fibrosis, 8 – heart disease, 6 – Baldness 7 – schizophrenia 6 – less aggressive *** Prevent 7 – heart disease 2 - baldness	4 – average height 2 – height, intelligence, sexual option	6-8 – cystic fibrosis, 5-6 – heart disease 2-4 - baldness	1 – sex of unborn baby
Marteau T et al (1995)	United Kingdom	-	-	-	2 - aggressive behavior and alcoholism	1 – adults (intelligence/specific skills)	-	-

						1 – children (appearance/behavior)		
Hampel J et al (2000)	Germany	4 – lab: medical	2 – farm: agricultural	-	7	-	-	-
		Approvers (in a total of 10 respondents)						
		Genetic modification of animals			**Genetic modification of humans**			
Authors (Year)	**Country(ies)**	**Transplants and/or medicines**	**Meat (Pork, Sheep, Cow)**	**Milk (Cow, Sheep)**	**Somatic (disease)**	**Somatic (enhancement)**	**Germline (disease)**	**Germline (enhancement)**
Norton J et al (1998)	Australia	-	3 - sheep and pork	-	-	-	-	-
Magnusson MK and Hursti UKK (2002)	Sweden	-	2 - pork and salmon	-	-	-	-	-
Macer DRJ (1997)	Asia/Oceania	-	8 (TH), 7 (J and IN), 5 (AU and NZ), 4 (IS), 3 (RU)	8 (TH and IN), 4 (J, AU, NZ and IS), 2 (RU)	-	-	-	-

Macer DRJ and Ng MAC (2000)	Japan	3	5	4	-	-	-	-
Inaba M and Macer DRJ (2003)	Japan	5 - mosquitoes	5	4	-	-	-	-
Small BH, Parminter TG and Fisher MW (2005)	New Zealand	2	-	2	-	-	-	-
Nayga RM (2006)	US and South Korea	-	3 (US) 2 (South Korea)	-	-	-	-	-
Govindasamy R et al (2008)	South Korea	-	2	-	-	-	-	-
Hallman WK et al (2002)	US	8 - sheep	3	8 - sheep	-	-	-	-
		Approvers (in a total of 10 respondents)						
		Genetic modification of animals			**Genetic modification of humans**			

Authors (Year)	Country(ies)	Transplants and/or medicines	Meat (Pork, Sheep, Cow)	Milk (Cow, Sheep)	Somatic (disease)	Somatic (enhancement)	Germline (disease)	Germline (enhancement)
Hallman WK et al (2003)	US	-	3	-	-	-	-	-
Puduri V et al (2004)	US	-	3	-	-	-	-	-
Macer DRJ et al (1997)	Japan and New Zealand	Pigs 5 (J) 3 (NZ) Mice 6 (J) 5 (NZ)	-	4 (J and NZ)	-	-	-	-
Inaba M and Macer DRJ (2003)	Japan	3	-	-	-	-	-	-

CRISPR (2013-2022)

		Approvers (in a total of 10 respondents)						
		Genetic modification of animals			Genetic modification of humans			
Authors (Year)	**Country(ies)**	**Transplant/Medicines**	**Meat /Milk**	**Welfare**	**Somatic (disease)**	**Somatic (enhancement)**	**Germline (disease)**	**Germline (enhancement)**
Chikhazhe TL (2015)	New Zealand	1	1	6	2	1	-	-
McCaughey et al (2016)	Global				6 (life-threatening and debilitating)	-	6 (life-threatening and debilitating)	3
STAT and Harvard (2016)	US				-	-	3	1 (Intelligence of physical)
Funk, Kennedy, Sciupac (2016)	US				-	-	5 ***Prevent: 1-4	-
Cormick C and Mercer R (2017)	Australia	5	3	-	7 (General)			
Chen C and Liang Z (2017)	China				6	Intelligence: 2	6	-

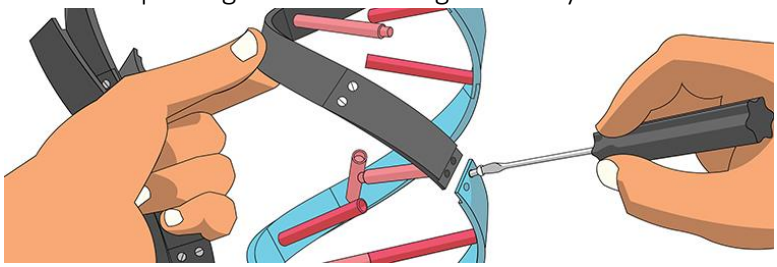
					Disease: 7-8 Non-disease (high cholesterol): 3	Skin color: 1		
Gaskell et al (2017)	Europe				8	2	6	0
Scheufele et al (2017)	US	-	-	-	6	4	6	3
Weisberg et al (2017)	US	-	-	-	Risk -7 ; No risk - 8			
Wang J-H et al (2017)	China	-	-	-	8 – adults and children	4	6	4
Hopkins and van Mill (2017)	UK	7 (mosquitoes and organs)	5 – efficiency of food 3 – profit	7 – resistant to disease 6 – invasive species 5 – control pest and hornless cows	8 - (in)curable 7 – non-life threatening 6 - Disorder not inherited	5 – prolong life 2 – cosmetic 3 - intelligence	8	-

		Approvers (in a total of 10 respondents)						
		Genetic modification of animals			Genetic modification of humans			
Authors (Year)	**Country(ies)**	**Transplant/Medicines**	**Meat /Milk**	**Welfare**	**Somatic (disease)**	**Somatic (enhancement)**	**Germline (disease)**	**Germline (enhancement)**
Hendriks S et al (2018)	The Netherlands	-	-	-	9	-	7 – Neuromuscular 3 - HIV	2
Uchiyama et al (2018)	Japan	-	-	-	The highest the awareness, the highest the support			
Lakomý M et al (2018)	Europe	-	3-6	-	8-9 (disease) ***Prevent 8-9 (Disease) 7-9 (Disabilities)	-	-	3-5
Pew Research Center (2018)	US				-	-	7 (Treat serious) ***Prevent - 6	2 (Intelligence)

Funk C and Heferon M (2018)	US	6 (transplants) 7 (mosquitoes)	4	-				
McCaughey T et al (2019)	Global				6 (life-threatening and debilitating)	-	6 (life-threatening and debilitating)	3
Critchley C et al (2019)	Australia	7	6	-	***Prevent - 8	5	***Prevent - 8	4
McConnachie E et al (2019)	US	-	6	9				
Yunes MC et al (2019)	Brazil	-	4	3				
Kohl PA et al (2019)	US			Wildlife - 1				

Appendix VII. Deliverable D19. Guideline for ethically-acceptable genome editing
Submitted as part of the IMGENE project (Ramos and Olsson 2021)

IMGENE
Improving Genome Editing Efficiency



Guideline for ethically acceptable genome editing

Deliverable (D19)

Pedro Ramos[1,2] and Anna Olsson[1,2]

[1-] Institute of Research and Innovation and Health (i3S), Institute of Biology and Molecular Science (IBMC)
[2-] Institute of Biomedical Sciences Abel Salazar (ICBAS)

3 August 2021

Contact: pedro.ramos@ibmc.up.pt

olsson@ibmc.up.pt

"This project has received funding from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No 765269."

Contents

What this white paper is … and what is not

IMGENE is a European Training Network (ETN) focusing on genome editing using CRISPR-Cas9. This report is produced within the subproject Genome editing as RRI, addressing the predicted harm-risk/benefit of genome editing as well as its public acceptance. While our qualitative research has covered more than just European perspectives, the project is EU-funded and therefore we focus the report on the European context. The present paper provides background and reflection using a combined approach, where we reflect on the ethical features identified by experts from the European Group on Ethics (EGE) [1] in relation to genome editing while at the same time we provide our own contribution from the qualitative work that we produced. This perspective results from the body of evidence that emerges from both in-depth interviews with biomedical and animal science researchers as well as from a systematic review of surveys administered to the general public on the topic of genome editing.

We will therefore not reflect on additional features identified in other body of knowledge than the EGE opinion and we will also focus only in the ethical questions of genome editing in humans and animals, which are the applications covered in our own research.

Within our problem-solution approach, we present an idea for a toolbox or toolkit for European research projects on genome editing which will allow the project to work on genome editing as Responsible Research and Innovation.

Background

Genome editing has without doubt been the biotechnological breakthrough of the second decade of the 21st century as confirmed by the Nobel Prize of 2020 that has been awarded to researchers Jennifer Doudna and Emmanuelle Charpentier for their work regarding “the genetic scissors: a tool to rewrite the code of life” whereby “using these, researchers can change the DNA of animals, plants and microorganisms with extremely high precision. This technology has had a revolutionary impact on the life sciences, is contributing to new cancer therapies and may make the dream of curing inherited diseases come true” [2]. The resurgence of Clusterly Regulated Interspaced Palindromic Repeats (CRISPR) associated with a protein with genetic scissoring activity (Cas9) - an adaptive mechanism of defense used by bacteria to defend themselves of invading viruses initially identified in the 1990s - in the academic environment has brought renewed excitement towards what was previously a laborious and complex process of precisely modifying the genome mainly achieved by its predecessors, zinc-finger nucleases (ZFNs) and transcription activator-like effector nucleases (TALENs) [3,4,5]. This has been made possible through the joint effort of many researchers whose work has allowed to adapt this mechanism to mammalian cells taking the complex and laborious out of the equation while keeping the high precision of the editing process [6,7,8]. This notable effort conjugated with its low cost led that CRISPR-Cas9 would be adopted by labs worldwide as their main genome modification technology [9,10].

The exquisite path of quick adoption of this biotechnology led to a high production of knowledge on basic research involving different organisms and promising results [11,12,13,14,15,16,17,18,19,20,21,22,23,25]. As usual with breakthroughs in the genetics area, a plethora of scientific papers were produced by scholars and brought to light old and new questions surrounding the ethical, legal, regulatory and social aspects of such a revolutionary technology that was moving in quick steps from basic to applied scenarios [26,27,28,29].

Simultaneously, statements and reports were also released following important meetings and summits addressing genome editing as an emergent biotechnology for which an ethical discussion and establishment of criteria for application were urgent [30,31,32,33]. Moreover, episodes of germline genome editing research with tripronuclear zygotes [34] and the announced birth of the first genome-edited babies [35] both in China resulted in two moratoria being requested by renowned scholars in 2015 and 2019, respectively [9,36,37]. Pressing issues like safety, informed consent from future generations, difference between disease and enhancement or between disease and prevention were evoked by scholars and several prominent documents like the Oviedo Convention [38], the Helsinki Declaration [39], the EU Charter of Human Rights and the UNESCO Declaration were revisited in this context [28,40]. In the somatic sphere, the first clinical trials using genome editing either targeting lung cancer, multiple solid tumors, melanoma and sarcoma and genetic diseases like B-thalassemia, Lieber congenital eye disease in patients were launched and preliminary results start to be known [41,42,43,44,45]. Both germline and somatic genome editing motivated statements and assessments by scholars on bioethics, ethics, sociology, philosophy and law, in particular relating to safety [9,28,46,47,48,49].

In sum, there were many aspects raised by several of these scholars when looking closely to CRISPR-Cas9 and other subsequent genome editing technologies. While we cannot match the same level of expertise or aptitude to address these questions, we can nevertheless reflect on one of the most recent body of expertise provided by the European Group on Ethics (EGE) in Sciences and New Technologies about their opinion on ethics of genome editing and contribute with our qualitative work to help closing the gap on some of the issues reported and others equally absent from this opinion document. In addition, we provide a toolbox or toolkit that would help in equipping future researchers involved in bench work dedicated to better facilitate an ethical acceptable genome editing. The toolbox draws on the experiences we have gained during two projects, IMGENE on gene editing and NewHoRRIzon on Responsible Research and Innovation in Horizon2020, and aims to provide support for genome editing research projects working according to the principles of RRI.

Review and Reflection

Here we will cover the main conceptual considerations elaborated in the EGE opinion document covering human and animal applications of genome editing. We stress that our goal here is to bring out the most important ethical issues identified by EGE and reflect on them and relate them to current literature. Whenever relevant, we have complemented with reflections from our own research, consisting in a systematic review of studies of public opinion regarding genetic modifications and interview studies with researchers in genome editing as well as experts in policy making and ethics.

Humans

The main ethical questions covered by EGE as regards genome editing in humans are dedicated to humanness and naturalness, diversity, distinctions between therapy, prevention and enhancement and finally, safety.

The fundamental humanness concept is not seen as being challenged by genetic interventions, because the biological dimension is not at stake. From this point-of-view, neither the method of intervention nor the multiplicity of gene transfers that may come from humans or from other

organisms will alter the humanness of an individual. If we go to universal meaning of the word "humanness" we see that it refers to the "quality or state of being a human" [50]. The EGE opinion here defends that the biological status of being human is not dependent on how much or how little of not-human is in a human: *"Is this an intervention in humanness? Biologically speaking, no – not as long as only such genetic changes are mad e that lead to genes that are otherwise present in humans. Even if DNA from another organism is introduced in a human genome this does not change the humanness of that entity. There is no percentage or sharp threshold beyond which the host is no longer considered to be human" (page 25) [1].* This is similar to the fact that there are more bacterial cells (and therefore bacterial DNA) in the human body than there are human cells [51].

The EGE is less assertive when it comes to philosophical or social dimensions of humanness, naturalness and the consequences that genome editing may have for these, and rather than providing answers the document asks questions, and remarks that "it is clear that there is no one scientific, unambiguous and thus binding answer as to what the relation between the genome of a human embryo and humanness and naturalness is, and what ethical orientation this can provide. Rather, the need arises for a broad, inclusive and nuanced social debate" (pp 26-27) [1].

Here, bringing in the public perception in addition to the theoretical philosophical reflection is important, because this issue pertains to the social complexity of the world where we live in. There is an argument to be made since there are many points-of-view related with indexes like law, regulation and therefore engaging with the public opinion at an early stage rather than making the public mainly a recipient of declarations, also because some of these concepts are being challenged by important emergent biotechnologies like genome editing. It would be relevant to engage and involve members of the general public in a participatory process that would bring the concept of genetic intervention closer to reality [52]. A recent survey study found a relationship between public perception of genome editing applications and the social construction of this technology, and reflected that this has consequences for a constructive deliberation: "If members of the public think about some realistic uses of CRISPR as if they were distant thought experiments or treat farfetched science fiction as imminent threats, it clearly represents an impediment to the assessment of 'societal consensus'." [53]. The general public is oftentimes not engaged at the right time or when it is, limitations in public awareness and knowledge levels on the topic are obstacles for a relevant discussion [52]. We have seen a lot of examples along the history of genetic engineering where misinformation or lack of information among citizens is associated with the adoption of radical postures in situations where there was no previous engagement with the public [32]. We also saw what misinformation and fake news provoked within this Covid-19 pandemic [54] and a correlation between misinformation with public interest and knowledge has been established elsewhere [55].

In addition to naturalness and humanness, equality and human dignity appear as significant concepts in the discussion around genome editing in humans. For a further discussion of these, please see [29,49,56,57] which explore these issues more deeply than EGE document [1]

In particular, the case of the impact of germline interventions for future generations intersects with two other subjects exposed in the EGE opinion document: responsibility and genetic determinism. Justification for the intervention is related to the seriousness of the condition, and these are fundamental aspects to substantiate acceptability. An additional and critically important question seems to be: who defines who is responsible when the affected individual cannot provide their consent? [27]. Usually, a physician assumes responsibility for a medical

intervention even if consent cannot be obtained; physicians also swear the Hippocrates oath which stresses that they have to intervene if there is no alternative when imminent death or saving a human life is at stake and abide by other medical ethics guidelines which places the health and well-being of the patient as the primary consideration [58]. A germline intervention is not a measure to avoid imminent death, or an urgent medical intervention that will save a patient's life. It is avoiding serious disease that will be a burden for the individual and very likely to their parents and that will diminish life quality as regards to physical capacity and level of independence considered under the World Health Organization Quality of Life (WHOQOL) [59].

Genetic determinism is associated with many of the previous issues. To what extent are individuals defined by an exceptional and differentiated fingerprint that is encoded in their genetic code? The EGE stresses that while genetic fingerprint influences individuals, other factors associated with cultural and generational backgrounds (or environmental factors) also need to be taken into account: *"The values placed on this kind of diversity can echo, or depart from, those in the biological domain (contending that cultural and linguistic diversity increases the adaptation strength of human societies, with a wider range of human knowledge and – by making us aware of a variety of distinct human ways of life – of counter-factual possibilities, 'other ways of living')"* and *"Diversity therefore does not stand alone as a value, rather it is context dependent. Our relationship to diversity, the norms and obligations surrounding it, and in particular any attempt to draw conclusions about those features to be deemed worthy of protection or liable to be jettisoned, must recognize the cultural, historical, biological and ecological factors guiding those choices. These must take into account past, present and future generations in all their diversity" (page 28) [1].* We know that there is a feedback interaction between genes and social behavior [60]. A related question is how much a germline intervention influences an individual and if that affects them more than what will be his or her own living experiences. It is often argued that the paradigm of genetic determinism is outdated, as it fails to consider how epigenetics and the regulation of non-coding regions play a role in the genome as well as that environmental factors are responsible for the prevalence of diseases or traits to be expressed [29,61]. But the idea that only genetics strongly define an individual and that genetic interventions interfere with what a person can be considered to be is still present in the discussion [29,62]. This is related to the question of eugenics, which is not an aspect addressed in the EGE opinion, but which has been discussed by other authors [29,31]. Our systematic review on public opinion regarding genetic modifications (53 primary publications from 1987-2019) found almost no references to eugenics and it seems to be both rarely included in questionnaires nor mentioned by respondents when enumerating arguments to justify their answers [63 – in prep].

In regards to diversity, the document produced by the EGE has a much more determined perspective. It argues that if genome editing interventions were to take place in the germline, it would be necessary to alter the gene pool of humans subsequently over many (undefined) generations to put human diversity at stake: *"In order to significantly influence the diversity of the human gene pool, a very broad use of genome editing on embryos over many generations would be necessary" (page 29). [1]* Therefore, diversity cannot be solely seen in terms of the diversity of the gene pool as it is associated with much wider issues of culture, history, biology, ecology, language and beliefs. The associated values and norms are not explored further in the EGE opinion, but this has been extensively done in other scientific articles and other similar reports [31,57].

Therapy, prevention and enhancement

EGE opinion illustrates very coherently the difference between therapy, prevention and enhancement in its definitions and how this related with scenarios of disease and no-disease. While therapy is attributed to somatic genome editing in order to treat an existing disease, prevention and enhancement blur the line between the intervention stage – germline or somatic – and the purpose – avoiding disease and improving a 'normal feature or function' (page 29) [1]. We aim to discuss these two enumerated points-of-view.

The former definition is straightforward because therapy can only concern an individual that already carries a given disease. However, when a genetic intervention takes place at the human embryo stage to prevent the future individual to become a disease carrier, it is sometimes questioned if an interference that takes place so early in the development for a recessive allele that will certainly (with 100% genetic probability) lead to a monogenetic disease (like cystic fibrosis) [27,48] falls under the prevention or therapy definitions. Other than this, we are always speaking of likelihood and therefore we cannot assume that because an individual is a carrier of a gene that encodes for a loss-of-function, he or she will develop that same disease. Similarly, when we are talking about future generations, we are always talking about prevention of disease. Nonetheless, therapy and prevention is much more than a discussion about the definitions *per se* because factors of social, regulatory and economic order are at stake. In economic terms, there is the burden for public health sector for an individual that is free from the disease avoided through germline intervention versus one who is not and could have been [36]. Through the role that tax payers play in enabling the national health service system to ensure that every individual can have access to any treatments in a public health service [64], their opinion also seems relevant. Also, when prevention of certain severe diseases become possible by prenatal interventions and decisions, a public discussion is also needed to address issues of how future patients will be seen and treated once parents are presented with a choice of intervention when deciding about their babies [65]. Whereas a genetic disease is no one's fault, is that limited to the situation of the natural process of conception? Is that absence of guilt turning to effective guilt once parents know the likelihood of having a baby with such a handicap and are not able to intervene or decide to abstain from doing so? A public discussion is necessary for the development of a regulatory framework for germline intervention in the prevention of inherited monogenetic diseases clear [27,33]. For this discussion to be constructive, awareness and knowledge of the public about genome modification is critical. Questions such as those above can be answered emotionally, but the more informed the public is, the more prepared they will be to answer them reliably [52]. Also, specific stakeholders need to be heard due to their high stake in questions that involve them directly. This includes patient organizations and professionals that deal with these questions such as genetic counselors and physicians [66]. Qualitative and quantitative inquiries with these groups of interest about genome editing and show a need for looking at their views in a much more careful way [67,68].

Enhancement or, in EGEs' words, *"improvement of a statistically and medically 'normal' feature of function" (page 29)* [1] is complicated by the fact that the concept of 'normal' in a society is context- and culture-dependent and answers to much more than just physical parameters. Even the physical parameters themselves are complex traits, with phenotypes that are dependent of both gene pleiotropy and other factors that are not inherent to genetics [29,62,67]. Enhancement is not straightforward for most traits when compared with interventions in germline for the purposes of prevention as mentioned previously for autosomal, monogenetic and inherited diseases [48,69]. The EGE here emphasizes the role of public perception and attitudes, highlighting that enhancement is recognized to be less acceptable for *'most people'*

(page 31) and it is also pointed that dis-enhancement can be a concerned for the *'wider public' (page 32). [1]*

Distinction between therapy, prevention and enhancement is also evident in studies of public attitude to genetic modifications that we have systematically reviewed. These show high public acceptability (rates between 75-95% worldwide) of genome editing in therapeutic settings in both adults and children for serious or fatal diseases and for the prevention of developing such diseases for these individuals but low citizen acceptability if these would happen for enhancement purposes (between 10 to 40% worldwide) with only arguments about having healthier individuals as the only considerable reason to proceed with it [63 – in prep].

Prevention also overlaps with enhancement according with EGE opinion. This has to do with the dual purpose of the intervention and it is correlated with the harm-risk/benefit assessment over the social implications that it brings, particularly exacerbated when germline intervention is being considered [27,28]. Other authors also elaborated about the onus of accountability over the decision for when prevention leads to incidental enhancements, where editing the genome with one purpose might incidentally lead to an unintended consequence like in the case of upregulation of Klotho protein for neurodegenerative diseases that may lead to cognitive enhancement [69]. As we have also seen in our own work, questions on enhancement raise alarm bells in many contexts, but are not limited to germline genome editing as they may also apply to somatic genome editing technology in patients for the purpose of therapy [36,46,47,70].

Safety in the context of somatic genome editing is addressed by EGE in the context of the existent regulatory guidelines from EU regulatory bodies regarding gene therapy like Advanced Therapy Medicinal Products (ATMP) and other national ones (page 32) [1]. It emphasizes a combination of anticipated benefits evaluation together with risk assessment, in order to prepare clinical trials. This assessment task has been addressed extensively by other authors [36,46,47,70]. Some reports have also comprehensively addressed risk-benefit assessment and the difficulty in establishing a standard [32,33]. Here, safety as both inherent to technical aspects of the genome editing platforms [29,46,70] and to the effects or consequences associated with administering genome editing treatments to patients and what impacts might these bring to their health [47]. The latter is more directly related with the risk-benefit assessment that has to take place before the clinical trial phase and when deciding to opt for an *in vivo* or *ex-vivo* genome editing therapeutic procedure as well as to which cells or tissues to be applied [46,47]. The former one concerns technical issues such as on- and off-target effects, delivery and efficiency features of the technology, issues that are presently addressed at the bench rather than at the bedside stage, despite their potential impact for the tools to be translated into the clinic [29,70]. In our interviews with scientists, these technical challenges alongside with others on fidelity and efficacy on-target have been identified. Assessment and detection of OTEs, higher efficiency through improvement of homology-directed repair (HDR) mechanism, improving in vivo delivery rates to bring them closer to those of in vitro delivery, multiplexing to accelerate CRISPR-Cas9 editing and, improvement of Cas9 cutting specificity have been some of the challenges that need to be tackled to bring genome editing closer to the clinic [71 – in prep]. These are highly technical issues with direct consequences for the 'safe enough criterion', which adds significant challenges for the ethics committees that are in charge of approving these trials and which expertise they need to include.

Regulating the implementation of this technology is a topic that goes beyond the question of safety. In our own research, we are presently addressing existent European and non-European

laws and regulations in interviews with ethicists and policy makers. The current regulation and legislation that concerns human somatic genome editing is intermediate (where existent], meaning that there is no a clear guidance on how permissive or restrictive this is for gene therapy [72]. Moreover, a more consistent (or harmonized) regulatory framework among countries has been emphasized and even consensus being an ambitious an objective, there is a need for a broad engagement of several regulatory bodies between countries to establish a productive communication and at least converge on some relevant points and avoid discrepancies namely at the level of travel jurisdictions and unequal citizen access to genome editing therapies [32]. Medical tourism with promises or advertisements that may take advantage of vulnerable patients by misguiding them to a genetic treatment that may not be their best option [66]. In fact, our interviews with ethicists and policy-makers show that some of them are indeed worried about the consequences if regulation and legislation affecting genome editing does not follow a previously agreed standard between entities in different countries and continents [73 - in prep].

Another aspect of safety applies to consequences on the societal level, considering that regulations to approve therapies for patients or other non-therapeutic applications to healthy citizens might reinforce situations of inequality since this may lead to the creation of genome-edited and non-genome-edited people classes or how the deepening of the socio-economic gap between low and high-income citizens would lead to the marginalization of societal groups without access to genome-editing technology [57,74]. Safety might also be challenged by enhancement of human beings under military premises [75].

The question of safety in the context of germline genome editing is complicated by the fact that those whose safety needs to be taken into account are not limited to individuals who exist here and now since heritability comes into play. Germline genome editing has consequences for future generations, and this needs to be taken into consideration when assessing the benefit and risk as pointed by EGE: *"The proportionality of benefit in terms of preventing a serious genetically transmitted disorder has to be balanced with the risk not only of not correcting the genetic defect but also of introducing unintentional modifications that could have serious implications for the child and future generations – perhaps even more serious than the one that should be prevented". (page 34) [1].* Overall, the majority of reports from national and international entities reject germline human genome editing based on safety arguments [76]. On one hand, these include technical issues which are also identified by the respondents in our interviews with researchers: efficacy, off-targets, efficiency, and genome-related issues [71 – in prep]. While this type of safety is considered in the 'safe enough criterion' score and is more palpable, there are other safety issues related with the societal consequences of the implementation of the technology rather than the technology preparedness *per se*. These are clearly more difficult to judge when assessing risks and benefits, because they are surrounded of much more uncertainty and are often considered more subjective [77]. Rather than arguing for a moratorium or the rejection of germline genome editing, moderate authors seek instead to see the value of the benefits for the individual who is 'edited' and consider in which situations society will win more than it loses, using a utilitarian ethics argumentation. This approach may result in the approval for genome editing to prevent or avoid disease [33,48,78]. In line with this, after the International Summit on Genome Editing in 2015, 10 conditional criteria were established to justify the use of germline genome editing in humans after the technology has been established as safe enough [33,79]. One of the criteria that EGE stresses is the absence of alternative methods to germline genome editing in case where both parents are carriers of an autosomal recessive disorder: *"Another central proportionality question is posed by the*

availability of technological alternatives to genome editing for avoiding heritable disorders, such as preimplantation genetic diagnosis and donation of gametes (yet, those raise other ethical questions). Only few reproductive constellations exclude all strategies but genome editing to ensure that a child is born without a disorder. This is the case, for example, if both parents are carriers of two alleles of a recessive disorder, so that every embryo can only inherit disease-causing alleles." (page 35) [1]. Other authors and reports also recognized the use of germline genome editing for disease prevention and personalized assisted-reproductive therapies (ART) as likely to provide more benefits than risks for future individuals for this situation and in others similar like the case if one of the parents [80,81]. However, many of the entities that assume a moderate position acknowledge that at the moment, risks and benefits cannot really be measured and that there is a need for further safety investigation in the clinic [76]. As for the ones who disavow germline genome editing based on criteria other than technical risks, these base their argumentation in the social inequalities that may be emphasized in an already uneven society [33,76] as well as the need for further investigation in preclinical and animal settings to test the safety of germline genome editing before moving to the clinical application in humans [82]. Also an assessment based on respect for the moral concept of human dignity may be used to both favor and reject it, since this could be seen as a moral imperative of human dignity to avoid serious disease in the future child as well as a respect for human dignity when avoiding the modification of the human genetic code of future generations [83]. How difficult the analysis is can be seen very clearly in the report by Danish Council on Ethics on the genetic modification of human beings with a clear separation of perspectives written in the released document [84]. However, assessment of benefits and risks applies to any health technology and the emerging technology of genome editing is no different either regarding issues like the dual-use dilemma [28].

What becomes clear from the reflections from EGE opinion group is the reinforcement of the need for a societal engagement in a broader discussion of the ethical issues that have been identified around genome editing in order to bring this discussion beyond that which exists on a mere scholar level: *"In light of the variety of ethical challenges posed by heritable human genome editing, inclusive societal debate is necessary. A broad societal consensus is precondition for the reproductive use of human genome editing to be considered." (page 36)* [1]. If there is something where there is a real consensus among scholars is this same reflection that EGE does: the need to involve different stakeholders in society to debate and deliberate on the different applications of genome editing technology, in particular germline genome editing which remains the most challenging application in humans [32,33,57,82]. We agree with the importance of such a broad societal engagement of stakeholders and underline that for public engagement to be productive, it needs to be done at the right time and with an insightful approach. Stakeholder engagement needs to happen before – or at least as part of – the drafting of policies and recommendations [82,85]. In particular, groups with high stakes in genome editing must be involved and prioritized, and this includes patients that are prospective future parents, future parents without known-genetic diseases and healthcare professionals that deal with these patients [33,82]. On the other hand, inclusion of professionals with special expertise in the technology (scientists) and the social, ethical, policy and legal issues that may drive the implementation of (social scientists, (bio)ethicists and legal scholars, but also policy-makers and legislators) is absolutely crucial for an insightful and constructive discussion [52,82]. The critical step would be the inclusion of the general public. A couple of questions need answer first: who is the general public? How can we know, and how do we ensure, that this is more than a group of people that also belong to any of the other interest groups represented elsewhere? The next

question is about when and how to best include these people in the discussion. While it is clear that public engagement is crucial, how upstream this should happen is still a matter of discussion [52,86]. The majority of scholars who advocate societal engagement evoke the need for a deliberative democracy discussion leave the specification of such a deliberative approach undefined [32,33,82]. Moreover, in our view, the methods of public engagement when it comes to genome editing have to move from consultation to participation. We have systematically reviewed surveys administered to the general public concerning their perception of genome editing from 2013-2019 [63– in prep] and this evidences the wide application of the consultation approach. It is now time to consider more seriously methods of public deliberation already emphasized by authors such as town-hall meetings, citizen juries, global citizens' assembly, online fora and community-based participatory research and leave other recurrent methods like polling, surveys and open comments to panel experts more aside [57,78,87,88]. This way we would move to a stronger and meaningful discussion of the relevant cross-cutting issues posed by genome editing and establish a real conversation between interest groups and the general public, possibly through civil society organizations (CSOs) groups. The question of incorporating the views of vulnerable and marginalized members of society that usually escape such deliberative processes has been identified as important [56,57]. For an effective policy-making regarding genome editing implementation, especially in germline, that so many ethical questions raise, this would be one way to drive the discussion and particularly when looking at the current legislation on the permissibility of heritable genome editing for reproduction that is so diverse worldwide [89]. As such and since this goes beyond a mere ethical discussion, we recommend the work done by other authors on the matter [69,89,90] and we hope to contribute to complement their overview as soon as our analysis of interviews with ethicists, bioethicists and policy-makers are finished [73 – in prep].

Germline genome editing for enhancement purposes is not addressed by the EGE opinion group, but merits some reflection. The concept of incidental enhancement is also of relevance similar to what happens when dealing with non-therapeutic purposes and the choice that parents might have when selecting embryos in in vitro fertilization (IVF) or pre-implantation genetic diagnosis (PGD) scenarios for susceptibility diseases [48,84]. Some authors consider enhancement to be a far-off consequence of GGE [48]. Others consider this to be closer step to eugenics and designer babies [37]. There are also questions that resound from somatic genome editing that are related with inequity [82,84]. Our systematic review shows that the public clearly differentiate between therapeutic and non-therapeutic or cosmetic purposes and find the former acceptable but the latter much more questionable. The challenges in distinguishing between disease and no-disease, susceptibility and certainty of disease are important aspects that repeatedly came up in our interviews with researchers. [71 – in prep]. In the case of susceptibility, questions regarding access became exacerbated. In addition, if safety issues are surpassed: in places where IVF is allowed, how much can future individuals be altered to satisfy parent's fear of contracting a characteristic considered to not be 'normal' in the society we live in?

Animals

Humans are not the only stakeholders in the discussion around genome editing. While animals cannot speak with us humans to let us know what they think and feel; they are nevertheless definitely an interested part in the discussion. Because we cannot subject them to surveys or interviews, their involvement as a group of interest over which we need to reflect has to be considered differently.

The EGE document establishes two ways to consider animals: their use for research purposes (or instrumentalisation) and their value as subjects to be considered as a mean rather than as an end (or intrinsic value) [1]. The EGE also recognizes that using animals for research and for food are not neutral questions over which there is societal consensus [1]. The aspects of 3Rs (Replacement, Refinement, Reduction) of animal experimentation and older discussions around xenotransplantation and the use of non-human primates resurface in this document.

Animals in research (experimentation and/or instrumentalisation)

The EGE highlights the role of dedicated EU legislation: *"There are many guidelines on the use of animals in research and since 1986 a dedicated EU Directive addresses the protection of animals used for scientific purposes (revised in 2010)." (page 38)* and official statistics that concern the use of vertebrate animals in research: *"In line with this Directive, the European Commission publishes reports on statistics about the use of animals for scientific purposes in the EU." (page 39) [1].* Data is available on parameters like number of animals used, number of animals with genetic alterations produced, used, killed and discarded and how severe were these animals affected by the genetic interventions and laboratory procedures when used as models for disease. The EGE bases this mainly in the report from 2015 to 2017 about the factual number of animals used, accounting for approximately 10 million, where about 10% of them are used as genetically-altered lines to be maintained for research purposes. Additionally, half a million of genetically-altered animals that only served as quality control were discarded following breeding [1]. In the discussion of genome editing in vertebrate animals, in particular two of the parameters of the 3Rs policy are at play: reduction and refinement. One of the major features is that number of animals used decreased between 2015 and 2017, but the use of non-human primates as second and third-generation breeds has been reported for the first time in 2017 [91]. For research models, there is an ethical requirement to minimize severity and animal suffering and to ensure high quality to obtain reliable and reproducible scientific results from animal experiments [92]. Creation of better animal models of disease in order to enhance human health through the development of new therapies and medicines is identified as one of the top ethical reasons to use genome editing in animals by many authors and such a use could be used to really decrease animal suffering by avoiding undesired genetic effects in these animals [93]. The EGE supports this assessment in their own words: *"Genome editing can contribute significantly to refinement, but apparently not to reduction overall. Although the 3Rs are considered equally important, how we balance them does sometimes change with different technologies" (page 52) [1]*. Some authors even suggested that 3Rs are not enough to faithfully ethical assess animal research in terms of scientific value required, suggesting 3 other Rs to be added to equation: robustness, registration and reporting [94]. The EGE opinion document also emphasizes the insufficiency of the 3Rs naming the need to introduce a fourth R, either Responsibility or Recourse to alternatives in the context of genome editing for animal research: *"One can also recall that the addition of a fourth R for 'Responsibility' was proposed by Max Planck, in line with the concept of Responsible Research and Innovation. In the context of genome editing, one might consider a further R for 'Recourse to innovative alternative strategies', which would go beyond refinement and which would, at least in the context of NHP research, require*

investment in alternative solutions" (page 52) [1]. The relationship between 3Rs and genome editing adoption in animal research was also often referred to by researchers in our interview studies. When it comes to public perception of genome editing for animals in research, the EGE recognizes the absence of information. Studies of public attitudes to the use of animals in genome editing research regarding their morality, usefulness, risks, benefits and naturalness are primarily from before the availability of the CRISPR-Cas9 technology [95,96]. Apart from EGE, another policy report elucidated about themes like speed, enforceability, complexity, mobilization potential, naturalness and proportionality to be tackled in regards to public dialogue about genome editing in animals and pinpointed steps for effective stakeholder engagement when addressing this topic [97]. The EGE also highlights the first R, replacement, and the importance to continue to seek alternatives to animal models in biomedical research, and the possibility to use genome editing techniques on non-animal models such as organoids [1].

The regulation of animal use in research requires an assessment, typically done by a committee, before any experiment with animals is allowed to start. This procedure is important also in the discussion of genome editing in research with animals. Quality of experimental design and translational potential are two types of benefits that are recommended to researchers by AALAS-FELASA to be looked at during harm-benefit assessment (HBA) for animal experiments and similarly, number, health status of animals and type of experiments that are inflicted to them in terms of duration, intensity, phenotypic manipulation and endpoint constitute some of the harms to be taken into account [98]. To what extent the analysis should also include benefits that will only occur after research translates into a clinical application is a question over which there is no agreement [99]. Genome editing can also have a direct reflection in the number of animals being used in the short- and long-run as well as in their welfare due to the increase of knowledge versus the suffering of animals necessary for it when developing new animal models of disease [93,100]. On one hand, less animals might be needed if the models are more precise, but on the other hand, the rate of animal experimentation may increase largely as a consequence of the availability of genome editing technology to create a much larger variety of models [31]. These observations were frequent among researchers in our interview study [71 – in prep].

Another animal welfare related issue addressed by EGE specific is the possibility to produce genome-edited animals that might be able to not feel any pain, although this has not yet taken place: *"The potential to change the nature of animals, sometimes referred to as 'de-animalisation', i.e. to add or remove certain capacities from animals (such as cognitive capacities or the ability to feel pain), is of ethical concern (see also section 4.5.3. on humanisation and non-human primates). In that regard, humanisation can also be understood as a form of de-animalisation." (page 52) [1].* An ethical reflection would possibly need to take place before such tentative is made since animals are sentient beings and taking out their ability to feel pain, although mesmerizing and avoiding one of the ways to experience suffering, the intrinsic essence of the animal (telos) would simultaneously be challenged [100]. A final aspect that EGE elucidates on is on how technical challenges also inherent to genome editing in humans like off-targets might influence the welfare of animals, despite little data existent on this therefore still unaware on the real impacts it might bring – either more off-targets due to undesired effects caused by some methods used in animal research like somatic cell nuclear transfer (SNCT) or less off-targets due to careful design of animal experiments [93]. Additionally, there have been already research innovative methods with CRISPR nucleases using a different method from the microinjection for embryo generation in pseudopregnant female mice like i-GONAD [101].

Similarly, some methods to assess OTEs in mice embryos like GOTI have been developed ever since [102]. These methods avoid several steps in the generation of genetically-modified animals and therefore lead to a reduction in numbers compared with previous methods. By increasing precision, it is expected that this will lead to better experimental models and avoiding animal waste.

With the low cost, easiness and velocity for the creation of genome-edited animals irrespective of species, the prospect of providing more specific disease models by designing a genome-edited animal that is not necessarily mice with a phenotype that will inform about an individual human pathology shifts the balance to the use of diverse types of animals like pigs and primates [31]. The use of non-humane primates is perceived to be particularly ethically challenging, and the EGE provides an extensive consideration about state-of-the-art, morality and philosophical use of these animals in pages 53, 54, 55 and 56 [1]. The challenges here are inherent to the greater ethical sensitivity of research with non-human animals, which are phylogenetically closer to humans. For much of disease modelling other animals like pigs have a higher similarity in organ size and resemblance but if we go to cognitive and behavior evaluation, NHPs present a closer phylogenetic ancestry to humans and that is already obvious at the genome and social level similitude [100,103,104]. Moreover, knowledge gained from mouse models in what it constitutes brain disorders is highly challenging to translate to humans [105]. So, the closeness of non-human primates to humans, brain-wise and in the potential to yield knowledge about brain disorders also implies closeness to humans in displaying similar emotional and cognitive experiences since this is linked to how neurons communicate and are able to retrieve feelings like pain and stress [104,105]. However, it is not obvious that the greater phylogenetically proximity to humans means that research will be more harmful to NHPs than to other mammals. In an ethics analysis published in response to the first transgenic primates being born in 2009, Olsson and Sandøe (2010) argued that "The biological and social proximity of monkeys and humans may also benefit the animals by making it easier for scientists and caretakers to recognize signs of suffering and increasing the human motivation to limit it. The animal welfare and research impacts of the transition to marmoset use will depend very much on the extent to which researchers take these issues seriously and seek to minimize animal harm and optimize human benefit." [106].

The phylogenetic proximity gives rise to the question of whether genome editing of primates can make them even more similar to humans, in a way that will in itself raise ethical issues. The ability to speak a human language and communicate it in understandable means with us, humans, is certainly one such characteristic. Will genome editing be able to bring other primates closer to human language capacity? Language is learned, and we know that primates have that ability as well [103]. If genome editing would enhance this capacity seems at least partly to depend on what is addressed through the edit. If we edit a non-human primate brain in a zone that is related with the ability to keep memories like in Alzheimer's, these are linked to the hypothalamus [107] and we are not necessarily interfering in the zone of language-learning which pertains to the broca area [108]. With the present focus on using genome editing to create disease models, scenarios of such humanization in the form of these incidental cognitive enhancements (a discussion if this is an enhancement or dis-enhancement might be interesting here) seem very unlikely. Our interviews with researchers also showed that they do not consider the question of a potential crossing of species boundaries in terms of increased cognitive capacity to be likely or of significant ethical concern, although some did reflect on the consequences of cognitive enhancement for characteristics typical of humans like language in

combination of suffering that can be communicated to us in comprehensible means [71 – in prep].

A final point that we would like to stress in this reflection concerns the fact that genome editing in NHPs is not approached in terms of public perception. Whereas primate use has often been part of studies of public attitude to animal research in general, we are not aware of any study that specifically addresses NHP use in genome editing research. This is perhaps in line with the fact that these animals have become more and more protected: for example, Directive 2010/63/EU strongly limits the research for which primate species can be used, and the use of great apes even requires the activation of a "safeguard clause" (Olsson et al 2016) [109]. However, with the advent of CRISPR-CasX technologies, the current use of other smaller non-human primates in research like cynmologous monkeys and rhesus macaques [110,111,112] and the strong investment of countries like China in the research with non-human primates' [103], clearly show that primate research is not on its way out.

The EGE also addresses the question of potentially crossing species barriers in the context of genome-edited organs for transplantation purposes [1]. Interspecies chimerism is presently considered a complex ethical question in the guidance for ethics review for Horizon2020 and Horizon Europe projects. Cross-species xenotransplantation success of genetically-modified pigs' hearts to baboons has been reported recently [113] but for now, only technical issues about this specific procedure have been considered [114]. In brain chimerism, both single cell or whole cell that is introduced in an organism may constitute a chimera, although from different order and this is also equivalent to transplantation of brain organoid to which we agree that chimerism discussion falls short and brain enhancement should be considered instead [115]. Ethical considerations at the level of how patients face the transplanted organ coming from a human-animal chimera research have also been raised and contrasted with the organ shortage as well as the long waiting for transplants [116].

A final ethical consideration that needs to enter in this discussion concerns the risk of cross-species transmission of diseases [116], a question that has been highlighted in the context of the ongoing Covid-19 pandemic [117].

Xenotransplantation of genetically-modified pig hearts for humans has been approached in surveys with the general public, which overall show that this is perceived as useful and moderately acceptable in terms of morality and benefit-risk perception [63 – in prep].

Genome editing in farm animals

The theme of genome editing in farm animals is not addressed in great detail by the EGE, which frame questions about "practices of pushing farmed animals to their 'biological limit'" and how genome editing can have a positive impact in productivity and animal welfare. Several authors have addressed how the editing of certain alleles in the genome of farm animals would improve their welfare and how this would preferable to conventional breeding [118,119,120]. Alongside this, four main purposes of genome editing in animals were identified and they are: human health, animal health, improvement of productivity and animal welfare [121]. This is possibly the right time to promote a discussion of the editing of alleles and genes that will influence traits in farm animals since at the moment there are no are no genome editing breeding programs for farm animals (although there is for aquaculture) [118]. Focusing in productivity, the myostatin gene (*MSTN*) is one that can be targeted by genome editing since its knock-out is responsible for increased muscle growth in cattle and therefore increase in meat production, but it comes with disclaimers about animal welfare issues like overweight implications, breathing problems, fertility concerns and pregnancy complications [118,121]. This kind of genome editing only serves a human purpose, and the outcome is detrimental for the animal. Compared to changing traits through conventional breeding, genome editing is more precise and predictable and therefore desirable for monogenic trait [119]. This is also true when comparing the older method using embryonic stem (ES) cells with cytoplasmic injection of the CRISPR components [121]. Efficiencies of genome edited liveborns of cattle, sheep and goats achieved by cytoplasmic microinjections in zygotes were tremendously lower than when using the previously predominant method of SNCT, but the pregnancy rates achieved were higher [118,121]. Polledness, the lack of horns, is another monogenetic trait of interest for genome editing in cattle, given the animal welfare concerns related with the need to dehorn calves or deal with the problems of keeping horned cattle [119]. Interestingly, the aspect of naturalness (one of the topics that is mostly approached in public surveys) is not approached by EGE. However, the EGE emphasizes asking how justifiable it is to use genome editing even for this purpose which, in principle, is to ameliorate its wellbeing [1]. We can clearly distinguish polledness from muscle growth increase since the first one has undoubtedly a positive effect in animal welfare and the second a clear negative effect. What might be discussed as here, is instead the intention to which this is made: is it solely based on animal welfare or is it first a matter of facilitating human work by having dehorned cattle? Are we aiming for polled cattle because it allows us to keep more cattle than would otherwise be possible? [118,120]. Similar questions can be asked for the use of genome editing to improve animal health by increasing udder health in regards to clinical mastitis in cows and porcine reproductive and respiratory syndrome (PRRS) and African swine fever [118,121]. Likewise, other applications to the improvement of pig welfare like GE pigs to produce more alpha-lactalbumin for increase of survival in piglets and increase of milk lysozyme production in goats that confer higher antimicrobial resistance have been also identified previous to genome editing as potential situations where genetically-engineered animals would be better than non-use of GE [122]. Of course, the same questions about off-target effects and their consequences for animal health must be asked here as in human applications. Indeed, off-target effects have been reported for ovine and caprine CRISPR-Cas9 injections [121]. The public also gave their views on dehorning in cattle considering it useful and good for their welfare despite the majority also seeing some concerns about the naturalness of the process [63 – in prep].

Recommendations

For overall recommendations for how to handle genome editing responsibly in society, and especially in the European Union, we refer to the set of recommendations provided by the EGE. On two pages (pp 5-6), this expert group provides a list addressing overarching matters and concerns, genome editing in humans, genome editing in animals, genome editing in plants and gene drives [1].

A toolbox/toolkit for to build guidelines for an ethical acceptable genome editing

It would be beyond our competence to propose recommendations for society or for regulations, or even for how to carry out genome editing research responsibly. What we can and proposed ourselves to do in this white paper is to use our experience and training in providing guidance for researchers in how to apply Responsible Research and Innovation (RRI) principles to better address emerging technologies of this magnitude. We therefore hope to contribute to the adoption of RRI rationale to address genome editing in future research of the kind in which this document has been produced.

Our own starting point has been a revision of literature on the ethics of genome editing, and on studies of public perception. This literature, including grey literature such as reports, should constitute the basis of building a matrix for future questioning. With this in mind, we will propose a set of recommendations for researchers that wish to address genome editing technology perks and perils by providing some advices based on our experience while conducting our project.

This toolkit would, in our view, better prepare a researcher from biological sciences performing bench work to constructively reflect on the ethical questions raised by genome editing and ultimately facilitate an ethically acceptable guideline for its adoption. This would also be in line with the incorporation of a RRI approach for researchers to explore the dimensions of their research in what concerns the impact it may have in society.

We propose to put this into practice through an approach we are already familiarized due to some initiatives that we participated involving multi-stakeholders. For a typical 3-year research project, we propose a 3-session workshop that would occur along the duration of the project. Ideally, these sessions would be facilitated by a person with expertise in science in society questions, and would involve the contribution of different actors. We envision a discussion with and between researchers in a first phase, the debate about the most burning questions regarding genome editing and therefore raising the most important ethical issues that surround it and finally, the establishment of a criteria to be used for an ethical acceptable guide of implementation of genome editing practices both in research and other environments.

For the first session, we would invite a duo or a trio of important actors linked to ethics, policy-making and social sciences. This initial set of stakeholders would share with researchers their experience when addressing genome editing under their perspective and would be open to take questions from the audience. The idea would be to debit a big chunk of questions regarding issues involving applications in the field of genome editing like humans, animals, crops and gene drives. Here, a participatory and interactive session would now take place with these 3 stakeholders facilitating and engaging with researchers to better understand their most pressing

questions involving such applications. It would have space for basic research as well at this stage given the fact that many of researchers could be working in the laboratory and with little or no expertise in the translational to applications. At this point, many ethical questions would emerge from the discussions in groups between researchers that would be deployed in a series of mind-map to illustrate their workflows. Questions like: What are the ethical questions raised by germline or somatic genome editing? What are the main ethical questions involving human genome editing? Are genome-edited animals in the laboratory in a better or worse situation in regards to welfare when compared with farm animals? Do gene drives raise any ethical questions?

This would be followed by another set of questions that would evolve within the previous ones and that might be more complex.

- What distinguishes a predicted harm-risk benefit assessment between germline and somatic genome editing?
- How would somatic genome editing influence the access of the wider public to treatments?
- Should personalized medicine or socioeconomic status be the most influential parameter to distinguish genome editing treatment priorities?
- Who would be responsible by technical challenges like off-targets when translating a given genome-editing therapy to the clinic? What are the most important technical challenges and how such safety assessment be made?
- Genome-edited animals should be done to prevent diseases or increase food production primarily?

After reconvening participants and facilitators and depicting the most burning questions to be addressed in a following stage, proposals for stakeholders to approach would have to be suggested so that they could be present at the second session of the workshop. In the end of the first session and in order to prepare for the second one, recruitment of stakeholders would need to happen by the organizers of the workshop.

In the second session, stakeholders representing international and national bodies and national entities, institutional, university and research infrastructures would be present. Here, the more that would be willing to be present, the better. We can imagine some groups of interest that would be also willing to have representatives at this stage like patient organizations, animal advocacy groups and farm animal breeding associations. This would increase familiarity with the subject of the study and empathy with stakeholders from whom one would wish to collect perspectives. Maybe spend some time looking for perspectives of contrasting and similar views about genome editing. At this stage it would be necessary to adapt the discussions to the different stakeholders that could be present and present a more structured framework to enlighten about the most important questions that would result from the interaction between researchers and the groups of interest or stakeholders. Designing consensual formulations at this stage with relevant questions to be tackled would be desirable. Maybe with patients trying to establish the value of adopting germline genome editing therapies could be related with the type of disease and therefore what would be the most important criteria that would need to be present to answer such premise? With animal advocacy groups, what would be the level of modification that could be adopted in scenarios where animals suffer from a painful disease and which brings impact both to their welfare and to human consumption (if this is considered at all)? Once again, and similarly to the first session, a closure is necessary and these questions should obviously be much less and simultaneously much more meaningful, structured and

extensive than the ones from the first session. However, no answers for them are needed at this point, so that these step would have to be taken at the third session.

At the third session, specific case-studies focusing on some ethical questions from the second session and resulting of the interaction with the different stakeholders that were in the second session would yield the necessary refection to take place only among biomedical researchers. This time, no other external stakeholders would be present and researchers would have to present their own reflection to their colleagues. This presentation would have to be about a defined theme (e.g. access of genome editing technology by socio-economic disfavored groups) and guidelines to address this issue would have to be proposed along with a set of at least 3 solutions to tackle the main ethical barriers that would be framed. Stakeholders to help with these solutions should be suggested (independently if they were present in previous sessions).

Based on our experience from the NewHoRRIzon project, where we were invited to participate as representatives of the MSCA-ITN type of project, we propose the social lab methodology for the sessions above [123]. This method stimulates an active engagement of all participants, and ensures that issues are addressed in an incremental way, with contributions across different groups.

Complementary tools

There are is also another tool to add to the kit that could be a good option for researchers after attending these workshops, regardless of the wave in which they participate. This could be courses, modules and seminars in ethics in order to make researchers more familiar with ethics in research with human subjects, animal subjects, human and animal tissues, embryonic stem cells, induced-pluripotent stem cells, organoids and so forth. The would also offer the possibility learning more about regulatory and legislation frameworks at national and international level and which bodies are in charge of ethically revising research projects using genome editing in both humans, laboratory animals, farm animals and crops.

A second tool is to be involved in scientific education and public engagement initiatives, important pillars of the RRI concept. Knowing all about the ethical questions that refer to genome editing becomes irrelevant if the public is not aware, not knowledgeable or not interested about it. Oftentimes there is a disconnection between what scholars write, publish and lecture and what the public really is interested and genome editing is not an exception to this situation. From our systematic review of surveys [63 – in prep] we demonstrated the importance of awareness and knowledge for the public perception about genome editing. Sensational media reports will remain a very or even the most impactful resource to draw attention to the topic as we saw with the genome-edited babies in 2018 but an informed and engaging attitude to reach the public is strongly needed and that can only be done with some other initiatives that will rescue the public to the bright side of Science. Design thinking, bio-art initiatives and public using digital resources like video-making and comics could be ways of sharing genome editing with the public. As for public engagement, inviting civil society representatives to be part of scientific discussions at the workplace could be a way of engagement in the beginning of projects and not only considering the public as a recipient of information derived from research.

References

1. European Group on Ethics in Science and New Technologies. Opinion on ethics of Genome Editing. 2021 March
2. Royal Swedish Academy of Sciences. The Nobel Prize in Chemistry 2020 (Press release, 7 October 2020)
3. Sander S, Young K. CRISPR-Cas systems for editing, regulating and targeting genomes. Nat Biotechnol. 2014 Apr;32(4):347-55
4. Barrangou R, Horvath P. A decade of discovery: CRISPR functions and applications. Nat Microbiol. 2017;2:17092.
5. Adli M. The CRISPR tool kit for genome editing and beyond. Nat Commun. 2018;9(1):1911.
6. Lander ES. The Heroes of CRISPR. Cell. 2016;164(1-2):18-28.
7. Ledford H. The unsung heroes of CRISPR. Nature, 2016, July 21 [Cited 2021 Mar 2021]. Available from: https://www.nature.com/articles/535342a
8. Urnov FD. Genome Editing B.C. (Before CRISPR): Lasting Lessons from the "Old Testament". CRISPR J. 2018;1(1):34-46.
9. Camporesi S, Cavaliere G. Emerging ethical perspectives in the clustered regularly interspaced short palindromic repeats genome-editing debate. Per Med. 2016;13(6):575-586
10. Siwo GH. The global state of genome editing (bioRxiv preprint). 2018.
11. Long C, Annally J, Shelton J, Mireault A, Bassel-Duby R, Olson E. Prevention of muscular dystrophy in mice by CRISPR/Cas9–mediated editing of germline DNA. Science 2014 Sep 5;345(6201):1184-1188.
12. Nelson CE, Wu Y, Gemberling MP et al. Long-term evaluation of AAV-CRISPR genome editing for Duchenne muscular dystrophy. Nat Med 2019 Feb 25:427–432.
13. Kang X, He W, Huang Y, Yu Q, Chen Y, Gao X, et al. Introducing precise genetic modifications into human 3PN embryos by CRISPR/Cas-mediated genome editing. J Assist Reprod Genet. 2016;33(5):581-8.
14. Yoshimi K, Kunihiro Y, Kaneko T, Nagahora H, Voigt B, Mashimo T. ssODN-mediated knock-in with CRISPR-Cas for large genomic regions in zygotes. Nat Commun. 2016;7:10431.
15. Xie F, Ye L, Chang JC, Beyer AI, Wang J, Muench MO, et al. Seamless gene correction of beta-thalassemia mutations in patient-specific iPSCs using CRISPR/Cas9 and piggyBac. Genome Res. 2014;24(9):1526-33.
16. Lessard S, Francioli L, Alfoldi J, Tardif JC, Ellinor PT, MacArthur DG, et al. Human genetic variation alters CRISPR-Cas9 on- and off-targeting specificity at therapeutically implicated loci. Proc Natl Acad Sci U S A. 2017;114(52):E11257-E66.

17. Young CS, Mokhonova E, Quinonez M, Pyle AD, Spencer MJ. Creation of a Novel Humanized Dystrophic Mouse Model of Duchenne Muscular Dystrophy and Application of a CRISPR/Cas9 Gene Editing Therapy. J Neuromuscul Dis. 2017;4(2):139-45.

18. Zhang Z, Zhang Y, Gao F, Han S, Cheah KS, Tse HF, et al. CRISPR/Cas9 Genome-Editing System in Human Stem Cells: Current Status and Future Prospects. Mol Ther Nucleic Acids. 2017;9:230-41.

19. Ma H, Marti-Gutierrez N, Park SW, Wu J, Lee Y, Suzuki K, et al. Correction of a pathogenic gene mutation in human embryos. Nature. 2017;548(7668):413-9.

20. Yang S, Chang R, Yang H, Zhao T, Hong Y, Kong HE, et al. CRISPR/Cas9-mediated gene editing ameliorates neurotoxicity in mouse model of Huntington's disease. J Clin Invest. 2017;127(7):2719-24.

21. Singh K, Evens H, Nair N, Rincon MY, Sarcar S, Samara-Kuko E, et al. Efficient In Vivo Liver-Directed Gene Editing Using CRISPR/Cas9. Mol Ther. 2018;26(5):1241-54.

22. Bjursell M, Porritt MJ, Ericson E, Taheri-Ghahfarokhi A, Clausen M, Magnusson L, et al. Therapeutic Genome Editing With CRISPR/Cas9 in a Humanized Mouse Model Ameliorates alpha1-antitrypsin Deficiency Phenotype. EBioMedicine. 2018;29:104-11.

23. Lyu C, Shen J, Wang R, Gu H, Zhang J, Xue F, et al. Targeted genome engineering in human induced pluripotent stem cells from patients with hemophilia B using the CRISPR-Cas9 system. Stem Cell Res Ther. 2018;9(1):92.

24. Lillico SG, Proudfoot C, Carlson DF, Stverakova D, Neil C, Blain C, et al. Live pigs produced from genome edited zygotes. Sci Rep. 2013;3:2847.

25. Sun J, Carlson-Stevermer J, Das U, Shen M, Delenclos M, Snead AM, et al. CRISPR/Cas9 editing of APP C-terminus attenuates beta-cleavage and promotes alpha-cleavage. Nat Commun. 2019;10(1):53.

26. Caplan AL, Parent B, Shen M, Plunkett C. No time to waste--the ethical challenges created by CRISPR: CRISPR/Cas, being an efficient, simple, and cheap technology to edit the genome of any organism, raises many ethical and regulatory issues beyond the use to manipulate human germ line cells. EMBO Rep. 2015;16(11):1421-6.

27. Howard HC, van El CG, Forzano F, Radojkovic D, Rial-Sebbag E, de Wert G, et al. One small edit for humans, one giant edit for humankind? Points and questions to consider for a responsible way forward for gene editing in humans. Eur J Hum Genet. 2018;26(1):1-11.

28. Nordberg A, Minssen T, Holm S, Horst M, Mortensen K, Moller BL. Cutting edges and weaving threads in the gene editing (capital YA, Cyrillic)evolution: reconciling scientific progress with legal, ethical, and social concerns. J Law Biosci. 2018;5(1):35-83.

29. Brokowski C, Adli M. CRISPR Ethics: Moral Considerations for Applications of a Powerful Tool. J Mol Biol. 2019;431(1):88-101.

30. On Human Gene Editing: International Summit Statement [Internet]. 2015; Dec. 3 (Statement, 2015)

31. Nuffield Council on Bioethics. Genome Editing: an ethical review. 2016

32. National Academies of Sciences Engineering and Medicine (U.S.), Human Genome Editing: Science, Ethics, and Governance, *The National Academies Press*, Washington,DC, 2017

33. European Academies Science Advisory Council. Genome editing: scientific opportunities, public interests and policy options in the European Union. 2017 EASAC policy report 31

34. Liang P, Xu Y, Zhang X, Ding C, Huang R, Zhang Z, et al. CRISPR/Cas9-mediated gene editing in human tripronuclear zygotes. Protein Cell. 2015;6(5):363-72.

35. Regalado A. EXCLUSIVE: Chinese scientists are creating CRISPR babies (2018). MIT Techology Review. November 25, 2018

36. Polcz S, Lewis A. CRISPR-Cas9 and the non-germline non-controversy. Journal of Law and the Biosciences. 2016;3(2):413-25.

37. Lander S. Adopt a moratorium on heritable genome editing. Nature, 2019, Mar 13 [Cited 2021 Jan 22]. Available from: https://www.nature.com/articles/d41586-019-00726-5

38. Convention for the Protection of Human Rights and Dignity of the Human Being with regard to the Application of Biology and Medicine: Convention on Human Rights and Biomedicine. Council of Europe 1997

39. World Medical Association Declaration of Helsinki: ethical principles for medical research involving human subjects. *JAMA*. 2013 Nov 27;310(20):2191-4. doi: 10.1001/jama.2013.281053.

40. Committee on Bioethics (DH-BIO). Statement on genome editing technologies. 2015 Statement of Council of Europe DH-BIO/INF 13 Final (Statement, 2015)

41. ClinicalTrials.gov [Internet]. Sichuan University; Chengdu Medgencell (MD): National Library of Medicine (US). 2016 June 8 - . Identifier NCT02793856, PD-1 Knockout Engineered T Cells for Metastatic Non-small Cell Lung Cancer; 2016 June 8 [cited 2021 Aug 3]; [about 6 screens]. Available from: https://www.clinicaltrials.gov/ct2/show/NCT02793856?term=CRISPR&cond=cancer&cntry=CN&rank=5

42. ClinicalTrials.gov [Internet]. Chinese PLA General Hospital (MD): National Library of Medicine (US). 2018 June 4 - . Identifier NCT03545815, Study of CRISPR-Cas9 Mediated PD-1 and TCR Gene-knocked Out Mesothelin-directed CAR-T Cells in Patients With Mesothelin Positive Multiple Solid Tumors.; 2018 June 4 [cited 2021 Aug 3]; [about 4 screens]. Available from: https://www.clinicaltrials.gov/ct2/show/NCT03545815?term=CRISPR&cond=cancer&cntry=CN&rank=1

43. Stadtmauer EA, Fraietta JA, Davis MM, Cohen AD, Weber KL, Lancaster E, Mangan PA, Kulikovskaya I, Gupta M, Chen F, Tian L, Gonzalez VE, Xu J, Jung IY, Melenhorst JJ, Plesa G, Shea J, Matlawski T, Cervini A, Gaymon AL, Desjardins S, Lamontagne A, Salas-

Mckee J, Fesnak A, Siegel DL, Levine BL, Jadlowsky JK, Young RM, Chew A, Hwang WT, Hexner EO, Carreno BM, Nobles CL, Bushman FD, Parker KR, Qi Y, Satpathy AT, Chang HY, Zhao Y, Lacey SF, June CH. CRISPR-engineered T cells in patients with refractory cancer. Science. 2020 Feb 28;367(6481):eaba7365.

44. Frangoul H, Altshuler D, Cappellini MD, Chen YS, Domm J, Eustace BK, Foell J, de la Fuente J, Grupp S, Handgretinger R, Ho TW, Kattamis A, Kernytsky A, Lekstrom-Himes J, Li AM, Locatelli F, Mapara MY, de Montalembert M, Rondelli D, Sharma A, Sheth S, Soni S, Steinberg MH, Wall D, Yen A, Corbacioglu S. CRISPR-Cas9 Gene Editing for Sickle Cell Disease and β-Thalassemia. N Engl J Med. 2021 Jan 21;384(3):252-260. doi: 10.1056/NEJMoa2031054. Epub 2020 Dec 5. PMID: 33283989.

45. ClinicalTrials.gov [Internet]. Editas Medicine (MD): National Library of Medicine (US). 2016 Feb 29 - . Identifier NCT03872479, Single Ascending Dose Study in Participants With LCA10; 2019 Mar 13 [cited 2021 Aug 3]; [about 5 screens]. Available from: https://www.clinicaltrials.gov/ct2/show/NCT03872479?term=NCT%2303872479&rank=1

46. Araki M, Ishii T. Providing Appropriate Risk Information on Genome Editing for Patients. Trends Biotechnol. 2016;34(2):86-90.

47. Baylis F, McLeod M. First-in-human Phase 1 CRISPR Gene Editing Cancer Trials: Are We Ready? Curr Gene Ther. 2017;17(4):309-19.

48. Gyngell C, Douglas T, Savulescu J. The Ethics of Germline Gene Editing. J Appl Philos. 2017;34(4):498-513.

49. Cribbs A, Perera S. Science and Bioethics of CRISPR-Cas9 Gene Editing: An Analysis Towards Separating Facts and Fiction. Yale Journal of Biology and Medicine 2017 Dec 19;90(4):625-634

50. https://www.definitions.net/definition/humanness (Definition of humaness)

51. Weiss RA. Open questions: knowing who's who in multicellular animals is not always as simple as we imagine. BMC Biol. 2018;16(1):115.

52. Wirz CD, Scheufele DA, Brossard D. Societal Debates About Emerging Genetic Technologies: Toward a Science of Public Engagement. Environmental Communication. 2020;14(7):859-64.

53. So D, Sladek R, Joly Y. Assessing public opinions on the likelihood and permissibility of gene editing through construal level theory. New Genetics and Society. 2021:1-25.

54. World Health Organism. Fighting misinformation in the time of COVID-19, one click at a time. 2021, Apr 27 [Cited 3 August 2021). Available at: https://www.who.int/news-room/feature-stories/detail/fighting-misinformation-in-the-time-of-covid-19-one-click-at-a-time

55. Agley J, Xiao Y. Misinformation about COVID-19: evidence for differential latent profiles and a strong association with trust in science. BMC Public Health. 2021;21(1):89.

56. Mulvihill JJ, Capps B, Joly Y, Lysaght T, Zwart HAE, Chadwick R, et al. Ethical issues of CRISPR technology and gene editing through the lens of solidarity. Br Med Bull. 2017;122(1):17-29.

57. Halpern J, O'Hara SE, Doxzen KW, Witkowsky LB, Owen AL. Societal and Ethical Impacts of Germline Genome Editing: How Can We Secure Human Rights? CRISPR J. 2019;2(5):293-8.

58. Hajar R. The Physician's Oath: Historical Perspectives. Heart Views. 2017 Oct-Dec; 18(4):154–159.

59. DIVISION OF MENTAL HEALTH AND PREVENTION OF SUBSTANCE ABUSE WORLD HEALTH ORGANIZATION 1998. WHOQOL User Manual. Rev 2012.03

60. Robinson GE, Fernald RD, Clayton DF. Genes and social behavior. Science. 2008;322(5903):896-900.

61. Nerlich B, Hellsten I. Genomics: shifts in metaphorical landscape between 2000 and 2003. New Genet Soc. 2004;23(3):255-68.

62. Comfort N. Genetic determinism redux. Nature 2018 Sep 561:461-463

63. What do people think about genetic engineering? A systematic review of studies pre- and post-CRISPR (In prep)

64. Roemer M. National Health systems throughout the world. Annu. Rev. Publ. Health 1993 14:335-53

65. Reincke CM, Bredenoord AL, van Mil MH. From deficit to dialogue in science communication: The dialogue communication model requires additional roles from scientists. EMBO Rep. 2020;21(9):e51278.

66. Meagher KM, Allyse MA, Master Z, Sharp RR. Reexamining the Ethics of Human Germline Editing in the Wake of Scandal. Mayo Clin Proc. 2020;95(2):330-8.

67. Armsby AJ, Bombard Y, Garrison NA, Halpern-Felsher BL, Ormond KE. Attitudes of Members of Genetics Professional Societies Toward Human Gene Editing. CRISPR J. 2019;2(5):331-9.

68. Hoffman-Andrews L, Mazzoni R, Pacione M, Garland-Thomson R, Ormond KE. Attitudes of people with inherited retinal conditions toward gene editing technology. Mol Genet Genomic Med. 2019;7(7):e00803.

69. Juengst ET, Henderson GE, Walker RL, Conley JM, MacKay D, Meagher KM, et al. Is Enhancement the Price of Prevention in Human Gene Editing? CRISPR J. 2018;1:351-4.

70. Abou-El-Enein M, Cathomen T, Ivics Z, June CH, Renner M, Schneider CK, et al. Human Genome Editing in the Clinic: New Challenges in Regulatory Benefit-Risk Assessment. Cell Stem Cell. 2017;21(4):427-30.

71. Ethical and technical challenges in genome editing: a qualitative study with researchers (In prep)

72. Isasi R, Kleiderman E, Knoppers BM. Editing policy to fit the genome? Science 2016 361(6271): 337-339

73. What do (bio)ethicists and policy-makers perceive from genome editing? a qualitative comprehensive analysis (In prep)

74. Hildebrandt C, Marron J. Justice in CRISPR-Cas9 research and clinical applications. AMA Journal of Ethics 2018 Sep 20(9):826-833

75. Greene M, Master Z. Ethical Issues of Using CRISPR Technologies for Research on Military Enhancement. J Bioeth Inq. 2018;15(3):327-35.

76. Brokowski C. Do CRISPR Germline Ethics Statements Cut It? CRISPR J. 2018;1:115-25.

77. Morrison M, de Saille S. CRISPR in context: towards a socially responsible debate on embryo editing. Palgrave Communications. 2019;5(1).

78. de Lecuona I, Casado M, Marfany G, Lopez Baroni M, Escarrabill M. Gene Editing in Humans: Towards a Global and Inclusive Debate for Responsible Research. Yale J Biol Med. 2017 Dec 19;90(4):673-681

79. National Academies of Sciences Engineering and Medicine (U.S.), Human Genome Editing: Science, Ethics, and Governance, Criteria for heritable germline editing Washington,DC, 2017

80. Ishii T. Germ line genome editing in clinics: the approaches, objectives and global society. Brief Funct Genomics. 2017;16(1):46-56.

81. Canadian Institutes of Health Research. Human Germline Gene Editing: Points to Consider from a Canadian Perspective. 2016 Dec

82. Matthews KRW, Iltis AS. Are we ready to genetically modify a human embryo? Or is it too late to ask? Account Res. 2019;26(4):265-70.

83. de Wert G, Heindryckx B, Pennings G, Clarke A, Eichenlaub-Ritter U, van El CG, et al. Responsible innovation in human germline gene editing. Background document to the recommendations of ESHG and ESHRE. Hum Reprod Open. 2018;2018(1):hox024.

84. Danish Council on Ethics 2016. Statement from the Danish Council on Ethics on genetic modification of future humans - In response to advances in the CRISPR technology. 2016

85. Juengst ET. Crowdsourcing the Moral Limits of Human Gene Editing? Hastings Cent Rep. 2017;47(3):15-23.

86. Tait J. Upstream engagement and the governance of Science. EMBO reports 2009 10: S18-S22

87. Jasanoff S, Hurlbut JB, Saha K. CRISPR democracy: Gene editing and the need for inclusive deliberation. Issues in Sci Technol. 2015;32(1):1-14 (Jasanoff et al, 2015)

88. Dryzek, JS, Nicol D, Niemeyer S, et al. Global citizen deliberation on genome editing. Science 2020 Sep 369(6510): 1435-1437

89. Baylis F, Darnovsky M, Hasson K, Krahn TM. Human Germ Line and Heritable Genome Editing: The Global Policy Landscape. CRISPR J. 2020;3(5):365-77.

90. Boggio A, Knoppers BM, Almqvist J, Romano CPR. The Human Right to Science and the Regulation of Human Germline Engineering. CRISPR J. 2019;2:134-42.

91. REPORT FROM THE COMMISSION TO THE EUROPEAN PARLIAMENT AND THE COUNCIL. 2019 report on the statistics on the use of animals for scientific purposes in the Member States of the European Union in 2015-2017. 2020 Brussels, Feb 5

92. Prescott MJ, Lidster K. Improving quality of science through better animal welfare: the NC3Rs strategy. Lab Anim (NY). 2017;46(4):152-6.

93. de Graeff N, Jongsma KR, Johnston J, Hartley S, Bredenoord AL. The ethics of genome editing in non-human animals: a systematic review of reasons reported in the academic literature. Philos Trans R Soc Lond B Biol Sci. 2019;374(1772):20180106.

94. Strech D, Dirnagl U. 3Rs missing: animal research without scientific value is unethical. BMJ Open Science. 2019;3(1).

95. Macnaghten P. Animals in their Nature. Sociology. 2016;38(3):533-51.

96. Ormandy EH, Schuppli CA, Weary DM. Public Attitudes toward the Use of Animals in Research: Effects of Invasiveness, Genetic Modification and Regulation. Anthrozoös. 2015;26(2):165-84.

97. The Netherlands Commission on Genetic Modification COGEM. CRISPR & Animals: Implications of genome editing for policy and society. 2018 Policy Report CGM/180501-01.

98. Laber K, Newcomer CE, Decelle T, Everitt JI, Guillen J, Bronstad A. Recommendations for Addressing Harm-Benefit Analysis and Implementation in Ethical Evaluation - Report from the AALAS-FELASA Working Group on Harm-Benefit Analysis - Part 2. Lab Anim. 2016;50(1 Suppl):21-42.

99. Grimm H, Eggel M, Deplazes-Zemp A, Biller-Andorno N. The Road to Hell Is Paved with Good Intentions: Why Harm-Benefit Analysis and Its Emphasis on Practical Benefit Jeopardizes the Credibility of Research. Animals (Basel). 2017;7(9).

100. Ohl F, Meijboom F. Ethical issues associated with the use of animal experimentation in behavioral neuroscience research. Curr Top Behav Neurosci. 2015;19:3-15.

101. Ohtsuka M, Sato M, Miura H, Takabayashi S, Matsuyama M, Koyano T, et al. i-GONAD: a robust method for in situ germline genome engineering using CRISPR nucleases. Genome Biol. 2018;19(1):25.

102. Marx V. A rocky road for the maturation of embryo-editing methods. Nat Methods. 2019;16(2):147-50.

103. Carvalho C, Gaspar A, Knight A, Vicente L. Ethical and Scientific Pitfalls Concerning Laboratory Research with Non-Human Primates, and Possible Solutions. Animals (Basel). 2018;9(1).

104. Coors ME, Glover JJ, Juengst ET, Sikela JM. The ethics of using transgenic non-human primates to study what makes us human. Nat Rev Genet. 2010;11(9):658-62.

105. Neuhaus CP. Ethical issues when modelling brain disorders innon-human primates. J Med Ethics. 2018;44(5):323-7.

106. Olsson IAS, Sandøe P. "What's wrong with my monkey?" Ethical perspectives on germline transgenesis in marmosets. Transgenic Res 19, 181–186 (2010).

107. Thompson RF, Kim JJ. Memory systems in the brain and localization of a memory. Proc Natl Acad Sci U S A. 1996 Nov 26;93(24):13438-44.

108. Nishitani N, Schürmann M, Amunts K, Hari R. Broca's region: from action to language. Physiology (Bethesda). 2005 Feb;20:60-9

109. Olsson IAS, Silva SPD, Townend D, Sandøe P. Protecting Animals and Enabling Research in the European Union: An Overview of Development and Implementation of Directive 2010/63/EU. ILAR J. 2016 May 1;57(3):347-357

110. Yan S, Tu Z, Li S, Li XJ. Use of CRISPR/Cas9 to model brain diseases. Prog Neuropsychopharmacol Biol Psychiatry. 2018;81:488-92.

111. Niu Y, Shen B, Cui Y, Chen Y, Wang J, Wang L, et al. Generation of gene-modified cynomolgus monkey via Cas9/RNA-mediated gene targeting in one-cell embryos. Cell. 2014;156(4):836-43.

112. Chen Y, Niu Y, Ji W. Genome editing in nonhuman primates: approach to generating human disease models. J Intern Med. 2016;280(3):246-51.

113. Langin M, Mayr T, Reichart B, Michel S, Buchholz S, Guethoff S, et al. Consistent success in life-supporting porcine cardiac xenotransplantation. Nature. 2018;564(7736):430-3.

114. Knosalla C. Success for cross-species heart transplants. Nature 2018 Dec 564:352-53

115. Chen HI, Wolf JA, Blue R, Song MM, Moreno JD, Ming GL, et al. Transplantation of Human Brain Organoids: Revisiting the Science and Ethics of Brain Chimeras. Cell Stem Cell. 2019;25(4):462-72.

116. Luna A. The Unknown of Human Animal Chimera Research. Intersect. 2017;10(3):1-8 (Luna, 2017)

117. Haider N, Rothman-Ostrow P, Osman AY, Arruda LB, Macfarlane-Berry L, Elton L, Thomason MJ, Yeboah-Manu D, Ansumana R, Kapata N, Mboera L, Rushton J, McHugh TD, Heymann DL, Zumla A and Kock RA. COVID-19—Zoonosis or Emerging Infectious Disease? Front. Public Health 2020 8:596944

118. Eriksson S, Jonas E, Rydhmer L, Rocklinsberg H. Invited review: Breeding and ethical perspectives on genetically modified and genome edited cattle. J Dairy Sci. 2018;101(1):1-17.

119. Mueller ML, Cole JB, Sonstegard TS, Van Eenennaam AL. Comparison of gene editing versus conventional breeding to introgress the POLLED allele into the US dairy cattle population. J Dairy Sci. 2019;102(5):4215-26.

120. Kramer K, Meijboom FLB. Using Breeding Technologies to Improve Farm Animal Welfare: What is the Ethical Relevance of Telos? Journal of Agricultural and Environmental Ethics. 2021;34(1).

121. Ishii T. Genome-edited livestock: Ethics and social acceptance. Animal Frontiers. 2017;7(2):24-32.

122. Murray JD, Maga EA. Is there a risk from not using GE animals? Transgenic Res. 2010;19(3):357-61.

123. Timmermans J, Blok V, Braun R, Wesselink R, Nielsen R. Social labs as an inclusive methodology to implement and study social change: the case of responsible research and innovation, Journal of Responsible Innovation 2020 7(3): 410-426