



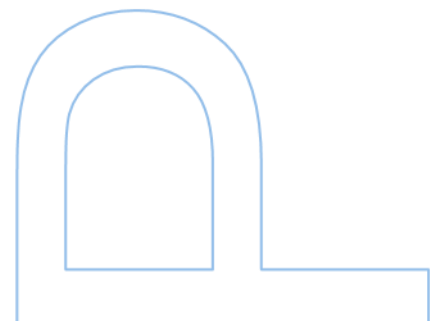
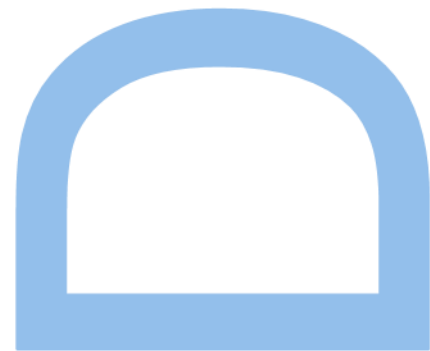
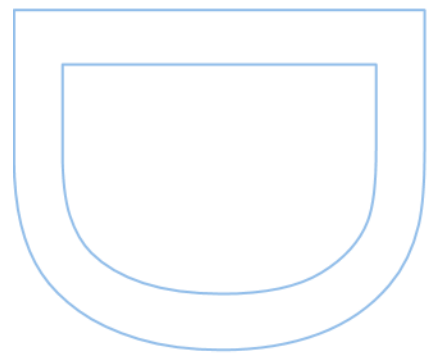
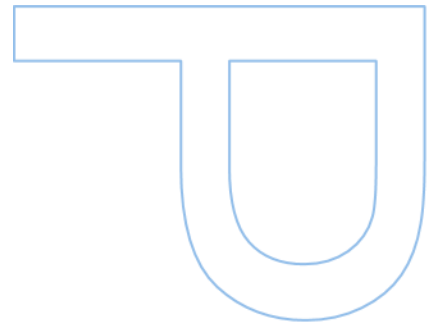
Tuberculosis in Asian elephant from One Health perspective

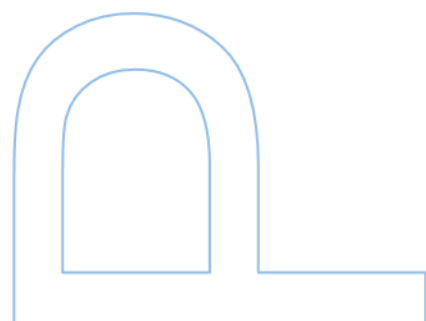
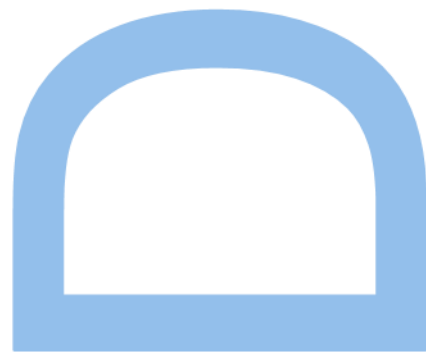
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Foreword

In compliance with no. 2 of article 4 of the General Regulation of Third Cycles of the University of Porto and with article 31 of the Decree-Law no. 74/2006 of 24 March, with the alteration introduced by the Decree-Law no. 230/2009, of 14 September, the results of already published works have been used and included in some of the chapters of this dissertation. As this work was completed in collaboration with other authors, the candidate would like to clarify that for this work, other authors also participated in obtaining, analyzing, and discussing the results, as well as in the published writings.

The Faculty of Sciences of the University of Porto was the home institution of the candidate, and the work was supervised by Professor Paulo C. Alves, and co-supervised by Prof. Christian Gortazar, and Dr. Dibesh Karmacharya, in close cooperation with Prof. José de la Fuente and Dr. João Queiros.

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Summary

Tuberculosis (TB) is one of the most significant and important human, livestock and wildlife disease that is posing some very difficult conservation challenges, especially in the captive Asian elephants of Nepal. Several studies have shown that TB is affecting captive elephants in Nepal and increasing risk of transmission/spillover to humans and other animals. In this work, I looked into the epidemiology of *Mycobacterium tuberculosis* complex (MTB) in a multi-host/species dynamic, primarily focusing on the captive Asian elephants of Nepal. The main objective of my doctoral thesis was to detect and characterize MTB complex in humans and animals (domesticated and wild) known to interact or come in close contact with captive elephants that are found in Chitwan National Park (CNP), a protected area with 932 km², situated in the sub-tropical Terai lowlands of south-central Nepal.

The CNP host 155 captive elephants, out of which 99 were included in this study, where I conducted a cross-sectional analysis consisting of risk assessment (survey questionnaire based, n=223 mahouts) and b laboratory assessment (immunological, proteomics, molecular diagnostics, and genomics), for detection of MTB complex and characterization of the pathogen in various species. I collected swabs (n=90), blood (n=99) and non-invasive (fecal, n=95) samples from elephants (n=99), humans (n=220) and environment (n=162) of wild herbivores and livestock. I collaborated with veterinarians and wildlife biologists to design the study and collect samples from the field (2018-2021).

The Sero-prevalence of MTB complex in captive elephants was 20.2%. Hand feeding to the elephant and not using personal protective equipment (PPE), as well as haphazard disposal of elephant waste were identified as most significant risky human behavior from pathogen spillover/transmission perspective. The bio-security measures were very poor in all the resting sites of captive elephants in the Chitwan National Park area.

By characterizing differentially represented proteins in response to TB in eight Asian elephants, with (n=4) and without (n=4) antibodies, I was able to identify some significantly dysregulated proteins (n=26) in response to tuberculosis. Of them, 10 (38%) were identified as immunoglobulin and 16 (62%) as non-immunoglobulin proteins. Proteomics showed Ig overrepresented in TB+ elephants, probably due to exposure to different *Mycobacterium spp.* An underrepresented Ig protein (G3UIN4) in TB+ and overrepresented in TB-animals might be a predictive biomarker for *Mycobacterium spp.*, playing a protective response role against Mycobacteria infections. While TB+ elephants

showed Ig proteins cross-reactive to various *Mycobacterium spp.*, the results suggested that TB-elephants developed a protective antibody response against mycobacterial proteins involved in disease-associated mechanisms.

The isolated *M. tuberculosis* of two deceased Asian elephants and one human sample were sequenced to generate MTB whole genome data. Three different lineages were detected: lineage 1 and lineage 2 in elephants and lineage 4 in humans. The phylogenomic analysis of the elephant-derived isolates clustered together with human-derived isolates sampled in Nepal. The human-derived isolate clustered together with other published human isolates from Argentina, Russia, and the United Kingdom.

The analyzed of gut microbiomes of various species, including elephants, were used to investigate transmission dynamics. The study focused on analyzing the diversity of gut microbiota in humans and mammalian herbivores, both domestic and wild, living in and around Chitwan National Park.

The results showed that Firmicutes and Bacteroidota were the most common phyla in all host categories, with Firmicutes being the most prevalent in humans. *Mycobacterium* genus was found in fecal microbiota of domestic and wild herbivorous mammals. The ungulates with the highest frequency of *Mycobacterium spp.* were spotted deer, followed by wild boar, rhino, captive elephants, and sheep. These findings indicate the ubiquitous presence of *Mycobacterium spp.* in the environment, and the likely inter and intra-species transmission possibilities. Humans (mahouts) have significantly lower alpha diversity and very different beta diversity compared to herbivorous host species, despite their close interactions. Captive elephants have the highest phylogenetic gut microbiome diversity; however, their alpha diversity is very similar to that of livestock and wild ungulates - Proteobacteria, Actinobacteriota and Verrucomicrobiota were some of the most common microbiota found between them. In contrast, the beta gut microbiome diversity was significantly different between captive elephants and livestock, wild ungulates, and their mahouts.

This work supports the hypothesis that MTB complex significantly impacts both human and animal health, and pathogen reservoir and spillover/transmission occur at a multi-species level. Therefore, the holistic One Health perspective is the most effective way to understand disease dynamics for implementation of effective control measures. By integrating epidemiological studies with omics technologies, I found that the health of captive elephants was closely interlinked and interconnected with humans (mahouts), but also with surrounding domestic and wild animal species. Taking these results into account, it were formulated several recommendations on how to mitigate and prevent MTB in captive elephants and other species in contact with them.

Keywords

Elephant, *Elephas maximus*, *Mycobacterium tuberculosis*, Tuberculosis, *MTB* complex, One Health, Zoonosis, Biosecurity, Mahout, Proteomics, Biomarkers, Genomics, Microbiome, human-wildlife coexistence, Chitwan National Park, Nepal

Sumário

A tuberculose (TB) é uma das doenças que ocorre em seres humanos bem como em animais domésticos e selvagens, apresentando desafios significativos para a conservação, especialmente em elefantes asiáticos no Nepal. Vários estudos demonstram que a tuberculose está afetando elefantes em cativeiro no Nepal e aumentando o risco de transmissão para os humanos e outros animais. Neste trabalho, foi investigada a epidemiologia do complexo *Mycobacterium tuberculosis* (MTB) numa perspectiva multi-hospedeiros, focando principalmente nos elefantes asiáticos cativos do Nepal. O principal objetivo desta tese de doutoramento foi detectar e caracterizar o complexo MTB em seres humanos e animais (domésticos e selvagens) conhecidos por interagir ou entrar em contato próximo com elefantes cativos que se encontram no Parque Nacional de Chitwan (CNP), uma área protegida com 932 km², situado nas terras baixas subtropicais de Terai, no centro-sul do Nepal. O CNP acolhe 111 elefantes em cativeiro, dos quais 99 foram incluídos neste estudo, onde foi realizada: a) avaliação de risco (baseada em inquéritos efectuados a tratadores e pessoas que contactam com elefantes, e; b) avaliação laboratorial (imunológica, proteómica, diagnóstico molecular e análise genómica) para detecção do complexo MTB e caracterização do patógeno em várias espécies. Foram recolhidas amostras com zaragatoas (n= 90), sangue (n=99) e não invasivas (fezes n=95) em elefantes (n=99), humanos (n=220), bem como em amostras ambientais (n=162), nomeadamente excrementos de herbívoros domésticos e selvagens. A recolha das amostras foi efectuada em colaboração com veterinários e biólogos da vida selvagem e decorreu entre 2018 e 2021.

A seroprevalência do complexo MTB em elefantes cativos foi de 20,2%. A alimentação manual do elefante e o não uso de equipamento de proteção individual, bem como a deficiente eliminação de resíduos e fezes dos elefantes, foram identificados como comportamentos humanos de risco mais significativo do ponto de vista da transmissão de patógenos. As medidas de biossegurança eram muito precárias em todos os locais de descanso de elefantes cativos na área do Parque Nacional de Chitwan.

Ao caracterizar proteínas representadas diferencialmente em resposta à tuberculose em oito elefantes asiáticos, com (n=4) e sem (n=4) anticorpos, foi possível identificar algumas proteínas significativamente desreguladas (n=26) em resposta à tuberculose. Destas, 10 (38%) foram identificadas como imunoglobulinas (Ig) e 16 (62%) como proteínas não imunoglobulinas. A proteómica mostrou Ig super-representada em elefantes TB+, provavelmente devido à exposição a diferentes *Mycobacterium spp.* A observação de proteína Ig sub-representada (G3UIN4) em animais TB+ e super-

representada em animais TB- pode potencialmente ser vista como biomarcador preditivo para *Mycobacterium spp.*, desempenhando um papel de resposta protetora contra infecções por *Mycobacteria*. Enquanto os elefantes TB+ mostraram proteínas Ig com reação cruzada a vários *Mycobacterium spp.*, os resultados sugeriram que os elefantes TB- desenvolveram uma resposta protetora de anticorpos contra proteínas micobacterianas envolvidas em mecanismos associados à doença.

O isolamento de *M. tuberculosis* em dois elefantes asiáticos infetados por tuberculose e uma amostra humana permitiu a sequenciação do genoma completo de três micobactérias. A análise destes genomas permitiu detectar três linhagens diferentes: a linhagem 1 e 2 em elefantes e a linhagem 4 em humanos. A análise filogenómica dos isolados derivados de elefantes demonstrou o seu agrupamento com isolados derivados de humanos detetados no Nepal. O genoma de MTB de humano demonstrou um agrupamento com outros isolados humanos, previamente detetados na Argentina, Rússia e Reino Unido.

A análise do microbioma intestinal das amostras fecais de várias espécies que interagem com os elefantes revelou uma dinâmica de transmissão inter e intra-espécies. Foi constatada a presença de bactérias pertencentes ao género *Mycobacterium* no microbioma fecal de mamíferos herbívoros domésticos e selvagens. Os ungulados com maior frequência de *Mycobacterium spp.* foram os veados-malhados, seguidos por javalis, rinocerontes, elefantes cativos e ovelhas. Estes resultados sugerem a presença ubíqua de *Mycobacterium spp.* no ambiente e a possibilidade de dinâmicas de transmissão inter e intra-espécies. Os seres humanos (tratadores de elefantes) têm uma diversidade alfa significativamente menor e uma diversidade beta muito diferente das espécies hospedeiras herbívoras, apesar de seu contato próximo com os elefantes. Os elefantes cativos têm a maior diversidade filogenética do microbioma intestinal, enquanto em termos de riqueza e uniformidade de espécies bacterianas, a sua diversidade alfa está num nível semelhante ao dos ungulados domésticos e selvagens. Em contraste, a diversidade beta mostrou que o microbioma intestinal de elefantes cativos era significativamente diferente dos ungulados selvagens.

Este trabalho suporta a hipótese de que o complexo MTB afeta significativamente a saúde humana e animal, e o reservatório de patógenos e a transmissão ocorrem a nível multiespecífico. Desta forma, a abordagem holística *Uma Só Saúde* é a maneira mais eficaz de entender a dinâmica das doenças bem como de permitir uma eficaz implementação de medidas de controle. Ao integrar estudos epidemiológicos com tecnologias inovadoras como a proteómica e a genómica, é realçado que a saúde dos elefantes cativos está intimamente e interrelacionada com os humanos (tratadores), mas

também com as espécies de animais domésticos e selvagens circundantes. Tendo em conta estes resultados, foram formuladas várias recomendações sobre como mitigar e prevenir a MTB em elefantes cativos e outras espécies em contacto com eles.

Palavras-chave

Elefante, *Elephas maximus*, *Mycobacterium tuberculosis*, Tuberculose, complexo MTB, Uma Só Saúde, Zoonose, Biossegurança, Tratador de elefantes, Proteómica, Biomarcadores, Genómica, Microbioma, coexistência humanos-vida selvagem, Parque Nacional de Chitwan, Nepal

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List of Abbreviation

AOR	Adjusted Odds Ratios
APOs	Apolipoproteins
BCG	Bacille Calmette-Guerin
BP	Biological Process
CC	Subcellular Localization
CITES	Convention on International Trade in Endangered Species of Wild Fauna and Flora
CNP	Chitwan National Park
DNPWC	Department of National Parks and Wildlife Conservation
DOI	Digital Object Identifier
DPP	Dual Path Platform
ELISA	Enzyme-linked immunosorbent assay
ER	Endoplasmic Reticulum
FDR	False Discovery Rate
GENETUP	German Nepal Tuberculosis Project
GO	Gene Ontology
IDA	Information-Dependent Acquisition
IGRA	Interferon gamma release assay
IRC	Institutional Review Committee
ISBN	International Standard Book Number
MAPIA	Multiantigen print immunoassay
MDR	Multi-drug resistant
MF	Molecular Function
MTC	<i>Mycobacterium Tuberculosis</i> Complex
NGS	Next Generation Sequencing
NTNC	National Trust for Nature Conservation
OR	Odds Ratio
PA	Protected Areas
PCR	Polymerase Chain Reaction
PPE	Personal Protective Equipment
PRISMA	Preferred Reporting Items for Systematic Reviews and Meta-Analyses
qPCR	Quantitative real-time Polymerase chain reaction
RBNP	Royal Bardia National Park
RCTs	Randomized Controlled Trials

SRA	Sequence Read Archive
TB	Tuberculosis
WGS	Whole Genome Sequencing
WHO	World Health Organization

Chapter 1: General Introduction

1.1 Background

This thesis focuses on tuberculosis (TB) of captive Asian elephants in Nepal along with the in-contact human population, livestock, and wildlife. Many infectious diseases affect not just wildlife but also humans and domestic animals (Hanisch-Kirkbride et al., 2013). Tuberculosis is one of the main globally important human diseases (World Health Organization, 2021), one of the livestock diseases with a higher socio-economic impact (Caminiti, 2019) and is becoming one of the top challenges for wildlife conservation (Thapa et al., 2017). In Nepal, captive elephants are used in eco-tourism, breeding programs, as well as wildlife and forest management, and thus interact closely with humans and wild populations (Mikota et al. 2006). The World Health Organization (2020) estimates that around one hundred and seventeen thousand people are infected with *Mycobacterium tuberculosis* in Nepal (Ministry of Health and Population, 2020). At the same time, TB has been detected in Nepal's captive elephant population posing a risk of transmission to humans and to wildlife populations (Paudel et al., 2014).

Tuberculosis affects humans, domestic animals, and wildlife populations (Michalak et al., 1998; Montali et al., 2001). Among the members of the *Mycobacterium tuberculosis* complex (MTC) that cause TB in mammals, *M. tuberculosis* is the main causative agent of human and elephant TB. In addition, cases of *M. caprae* and *M. bovis*, also members of the MTC, were recently reported in a captive Asian elephant (*Elephas maximus*) and free-ranging African savanna elephants (*Loxodonta africana*), respectively (Miller et al., 2021; Suga et al., 2021).

1.2 Asian elephant populations in Nepal

1.2.1 Wild elephants

Resident wild Asian elephants in Nepal are grouped into four isolated populations: Eastern, Central, Western and Far-Western (Pradhan et al., 2011). Presently, the number of resident wild Asian elephants in Nepal is estimated to be between 109 and 142 animals (DNPWC, 2012; Pradhan et al., 2011). They occur in four isolated populations (Eastern, Central, Western, and far-Western) in the Terai belt: the eastern population (10-15 animals) that migrates from West Bengal, India to the eastern districts during July to October; the central population (25 animals) mostly confined to Parsa Wildlife Reserve; the western population, (45-50 animals) residing in Royal Bardia National Park (RBNP), and; the far western population (2-18 animals) in the Churia foot hills. The far-western population moves occasionally to India via the Mahakali River and sometimes uses Royal Suklaphanta Wildlife

Reserve as a dispersal area (WWF Nepal Program, 2003). In central Nepal, it is estimated that a population of 25 to 30 elephants are largely resident within the Parsa Wildlife Reserve and Chitwan National Park (DNPWC/MoFSC/GoN, 2008; Note & Cordingley, 2011). Occasionally these elephants move towards Mahhotarri district in the east. The elephant population is estimated to occur over a forest area of 3227 km². Forest habitats and corridors are mainly intact except for a few settlements in the Bara, Rautahat and Mahhotarri districts (DNPWC/MoFSC/GoN, 2008). The area inhabited by elephants is spread over 135 Village Development Committees (VDCs) in 19 districts (17 in lowland Terai and two in the hills) of Nepal, covering about 10,982 km² of forest area (DNPWC/MoFSC/GoN, 2008). In addition, there have been several incursions of over 100 elephants from the Indian state of West Bengal into Eastern Nepal (Pradhan et al., 2011).

1.2.2 Captive elephants

At present, it is estimated that there are 208 captive elephants in Nepal, out of which 94 government-owned elephants are in various protected areas (PAs) in the Terai, namely Koshi Tappu Wildlife Reserve, Parsa Wildlife Reserve, Chitwan National Park, Bardia National Park, and Suklaphanta Wildlife Reserve. The National Trust for Nature Conservation (NTNC) owns 8 elephants in their field station in Chitwan, Bardia and Central Zoo, Kathmandu. Most private-owned elephants are in and around Chitwan National Park and are mainly used for tourism purposes, which include forest excursions in buffer zones of parks and reserves. Government-owned elephants are mainly used for patrolling and research purposes (Shrestha & Shrestha, 2021). These elephants are used to help with conservation policies and for capturing and counting other wild animals (EleAid, 2018).

During the pandemic time, the health and well-being of captive elephants were found to be degrading as they struggled for food and movement.

There were also reports of elephants being smuggled to India from Sauraha illegally. Elephants, which were old and diseased, can be taken to India and sold. Those elephants, which are sold in India, are used in various temples and religious functions including weddings. Despite of Nepal and India being signatories for the “Global convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES), the rules relating to cross-border sale of live elephants are not followed. Elephant welfare right campaigners have been opposing elephant rides for tourists. They are against to hold captive elephants for riding or other activities (Diwakar, 2021). Furthermore, Nepal government doesn't have any official records of the number of captive elephants in Nepal. The initial number of estimated elephants was estimated to decrease to almost 80 percent by the year 2020, meaning that around a fifth of elephants had presumably been sold to Indian traders (Diwakar, 2021).

1.2.3 Elephant tuberculosis

Elephant TB is a chronic disease that affects captive elephants worldwide. In Nepal, TB in captive elephants was first identified in Chitwan National Park (CNP) in 2002 (DNPWC, 2011a). In the period of seven years (2002-2009) altogether seven captive elephants died due to TB (DNPWC, 2011b). To address this situation, the Government of Nepal started surveillance on elephant TB in 2006 and a treatment in 2008. From 2002-2009 a total of 11 captive elephants tested TB positive in Nepal (Szydlowski, 2022). During the study period of 2019-2021 three elephants died of TB. The estimated annual mortality rate of elephants is around 1%.



Figure 1.1 Elephants patrolling with Mahout in the jungle

1.3 Conceptual framework

The factors such as host TB pathogen and its spillover play a vital role in TB transmission among wild animals, humans, and livestock. Since the captive elephants are in very close contact with humans, namely to the mahouts, the chance of zoonotic disease spill-over between the species is high. Moreover, wild elephants come in contact with captive elephants during the mating season paving ways for disease spill-over between captive and wild populations (Paudel et al., 2014). Finally, the contact of wild and domestic elephants with

livestock and wildlife also might disseminate TB. The disease prevalence and persistence in wild elephants can be fatal (Mikota et al., 2015). The conceptual framework for this study is explained in the figure below (Figure 1.2).



Figure 1.2 Conceptual Framework of this PhD thesis focusing in elephant Tuberculosis.

The risk of TB transmission increases as the human population living in the buffer zones usually comes in close contact with wild and domestic animals. There are evidences that veterinarians, animal keepers, and abattoir workers might be a major risk of TB transmission from infected animals (Une & Mori, 2007). Particularly animal handlers who work on a very close proximity with captive elephants have greater risk of *M. tuberculosis* infection (Michalak

et al., 1998). Livestock are often taken to buffer zone areas for grazing, and they might be at risk of disease spillover from the wild. In turn, contracted diseases can be transmitted from livestock to humans (The Weston A. Price Foundation, 2017). Furthermore, elephants are not the only wildlife species at risk, with others such as *Rhinocerotidae* (rhinoceroses) susceptible to the *M. tuberculosis* complex. The wild animals which are under the risk of TB pose a greater risk of transmission because of the fact that they can either act as reservoir hosts or spill-over host (El Idrissi & Parker, 2012).

The characterization of the host response to *M. tuberculosis* infection and TB using omics technologies such as proteomics, genomics and metagenomics as documented in this thesis might contribute to the design and implementation of interventions for reducing the risks of pathogen transmission and TB control.

1.4 Hypothesis

Risk factors and exposure to mycobacteria act as epidemiological links between captive Asian elephants, humans, domestic and wildlife cases of TB in Nepal.

1.5 Objectives

This PhD thesis aims to increase the knowledge on the presence and spill over dynamics of TB in captive elephants and their interactions with other species, primarily humans and selected wildlife and livestock. It provides new information on the effects of TB infection status on elephant immune response and serum biomarkers, on whole genome sequences of *M. tuberculosis* strains from elephants and humans, and on the fecal microbiota of elephants, humans, other livestock species and wildlife. The main objective is to contribute to a holistic One Health perspective to improve the understanding of elephant TB and setting it as an example for similar studies in other pathogens, animal species, and in the environment. This study was conducted in the Chitwan National Park, which is the first national park of Nepal, established in 1973. The park covers an area of 932 km² situated in the sub-tropical Terai lowlands of south-central Nepal. This national park is popular as one of the best wildlife viewing national parks in Asia.

1.5.1 General objective

The general objective of this PhD thesis is to characterize the epidemiology of *Mycobacterium tuberculosis* complex in a multi-host system in Nepal, with a focus on the captive Asian elephants.

1.5.2 Specific objectives

To address the general goal, seven specific objectives were implemented:

1. To assess the prevalence of *Mycobacterium tuberculosis* complex in domestic Asian elephants.
2. To assess the links between captive elephants, humans, and relevant domestic and wild host species in Nepal.
3. To assess risk factors for captive Asian elephants testing positive to antibodies against the *Mycobacterium tuberculosis* complex.
4. To characterize and assess the evolutionary history of *Mycobacterium tuberculosis* complex strains involved in the infection of the most relevant wild and domestic hosts.
5. To characterize the immunological response in *Mycobacterium tuberculosis* complex infected vs. not infected Asian elephants through serum proteomics.
6. To characterize the gut microbiome and inter species relationships including presence of *Mycobacterium tuberculosis* complex species.
7. To identify the options available for eventual interventions to contribute to reducing the TB burden in Asian elephants.

1.6 Thesis Structure

The results of this doctoral thesis are presented in seven chapters, including a review paper serving as introduction and four original papers, which are published or submitted to international indexed journals (table 1.1). In addition, it is presented a general discussion and the conclusions.

Table 1.1 Papers included in the PhD thesis.

Chapter	Title	Journal
1	Part of Introduction Understanding <i>Mycobacterium tuberculosis</i> in elephants through a One Health perspective: A systematic review	<i>Understanding Mycobacterium tuberculosis complex in elephants through a One Health perspective: a systematic review. BMC Vet Res. 2022 Jul</i>
2	Risks associated with human-elephant interactions. Human risk factors associated with the presence of antibodies against <i>Mycobacterium tuberculosis</i> complex in captive Asian elephants from Chitwan valley, Nepal	Preventative Veterinary Medicine, under review
3	Serum biomarkers of tuberculosis in elephants Differentially represented proteins in response to tuberculosis identified by quantitative serum proteomics in Asian elephants	Differentially Represented Proteins in Response to Infection with <i>Mycobacterium tuberculosis</i> Identified by Quantitative Serum Proteomics in Asian Elephants. Pathogens. 2022 Sep 3
4	Genomic signature of <i>M. tuberculosis</i> transmission Phylogenomic analysis support <i>Mycobacterium tuberculosis</i> transmission between humans and elephants	Phylogenomic analysis supports <i>Mycobacterium tuberculosis</i> transmission between humans and elephants. <i>Frontiers in Veterinary Science</i> , 10
5	Gut Microbiome Gut microbiota diversity across human, elephant, livestock, and wild herbivore assemblages in Chitwan National Park: implications for conservation medicine	Hindawi under review

In Chapter 1, which is part of the General introduction, in addition to the overall background on the thesis topic and the objectives, a systematic review on *Mycobacterium tuberculosis* in elephants through a One Health perspective is presented. The revision of 27 papers allowed to give an overview on the prevalence of TB reported in elephant populations, which ranges from 0 to 23.33% and high prevalence's are reported for elephants that are in close proximity to infected humans. This paper was published in the journal BMC Veterinary Research

In Chapter 2, the risk of TB infections associated with human-elephant interactions was evaluated. A total of 99 captive Asian elephants (*Elephas maximus*) from Chitwan valley was tested for antibodies against the *M. tuberculosis* complex (MTC) and 223 mahouts were interviewed to investigate associations between human risk factors and MTC antibody detection. The main result is the prevalence of antibodies against MTC in elephants reaching a level of 20.2%. This paper was submitted to the journal Preventative Veterinary Medicine, and it's currently under revision.

In Chapter 3, a proteomics approach was used to assess the putative proteins that might be associated with TB. Blood samples were collected from four adult animals with and without TB reactive antibodies. Proteomics analysis identified 26 significantly dysregulated proteins in response to TB. Of them, 10 (38%) were identified as immunoglobulin and 16 (62%) as non-immunoglobulin proteins. The results provided new information on the antibody response to mycobacterial infection and biomarkers associated with TB and protective response to mycobacteria in Asian elephants. These results were published in the journal Pathogens.

The whole genome of four *M. tuberculosis* found in humans and captive elephants from Nepal were sequenced and analyzed and presented in Chapter 4. The whole genome sequences of *M. tuberculosis* strains were isolated from two deceased Asian elephants and one infected human in Chitwan, Nepal. The phylogenomic trees allocated the *M. tuberculosis* strains in three different lineages known as lineage 1 and lineage 2 (elephants DG and PK respectively), and lineage 4 (humans). These results were published in Frontiers in Veterinary Science.

In Chapter 5, we studied the gut microbiome communities in captive elephants, humans, livestock and wild ungulates, across humans and mammalian herbivores, that live alongside each other in a highly populated area in and around Chitwan National Park (Southern Nepal). I also investigated gut microbiota composition in mahouts (i.e., elephant riders and trainers or keepers), who are in close contact not only with captive elephants but also with their own livestock and wild herbivores inhabiting the forests next to their villages. The results have provided valuable data on gut microbiome communities of herbivores inhabiting Chitwan National Park, along with those of people, livestock and captive elephants that constantly

interact - both directly and indirectly - with each other. The paper was submitted to the journal of Hindawi and is under review.

In the General Discussion Chapter 6, the integration of all results presented in the different chapters are integrated and discussed, and major inferences on TB transmission among captive elephants, humans, livestock, and wildlife are performed. In addition, conclusions and further research are presented.

1.7 References

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1.8 Understanding *Mycobacterium tuberculosis* in elephants through a One Health perspective: a systematic review *

1.8.1 Abstract:

Background: *Mycobacterium tuberculosis* complex (MTC) that causes the chronic infectious disease- tuberculosis (TB), often presents with a complicated epidemiological pattern where the transmission chain may include humans, domestic animals, and wildlife, including elephants. TB has been reported globally in both captive and wild elephants. The One Health perspective might be the most effective way of understanding the shared MTC infection dynamics in captive and wild animals like Asian elephants. This systematic review accumulates evidence on occurrence, transmission pathways, and preventive measures of TB in elephants from a One Health perspective.

Results: The prevalence of TB reported in elephant populations ranges from 0% to 23.33% and high prevalence is reported for elephants that are in close proximity to infected humans. The risk of elephant to human infection transmission increased significantly with exposure duration and contact with infected elephants. Some studies described the plausible TB transmission to captive elephants from other animals (wild and domestic), suggesting inter- and intra-species transmission. The results of this systematic review based on 27 relevant published works, suggest three overarching interrelated transmission pathways for *M. tuberculosis* infections in Asian elephants- i) humans and elephants, ii) other animals (wild or domestic) and elephants and iii) unclear sources of infection.

Conclusions: The progress made with new TB diagnostic tools provides multiple methods to choose from. However, lack of harmonization of TB testing in elephants and their human contacts remains a challenge to prevent TB in those animals. Routine TB screening among elephants and caretakers by setting up an occupational health program for early diagnosis of infection through combined efforts of public health, veterinary medicine, and occupational health experts is suggested. This implies the need for a One Health perspective to elephant TB control. This review reveals the need for more research on *Mycobacterium tuberculosis* complex transmission pathways at the human-animal interface.

* Rajbhandari, R.M. *et al.* (2022) 'Understanding *Mycobacterium tuberculosis* complex in elephants through a One Health perspective: a systematic review', *BMC Veterinary Research*, 18(1). doi:10.1186/s12917-022-03356-8.

Keywords: *Elephas maximus*, *Loxodonta africana*, *Mycobacterium tuberculosis*, Transmission, Prevention, Zoonosis.

1.8.2 Background

Tuberculosis (TB) affects humans, domestic animals, and wildlife populations (Michalak et al., 1998; Montali et al., 2001). Among the members of the *Mycobacterium tuberculosis* complex (MTC) that cause TB in mammals, *M. tuberculosis* is the main causative agent of human and elephant TB. In addition, cases of *M. caprae* and *M. bovis*, also members of the MTC, were recently reported in a captive Asian elephant (*Elephas maximus*) and free-ranging African savanna elephants (*Loxodonta africana*), respectively (M. A. Miller et al., 2021; Suga et al., 2021).

Human cases of TB are reported almost in all countries of the world (Lewerin et al., 2005). Generally, the human TB burden is high in the African and Asian nations, especially in low-income countries (Mikota, 2020). Amid the 20 high TB burden countries by the estimated absolute number of TB human cases, 14 countries (8 Asian and 6 African) are within the distribution range of wild elephants. The wild elephants are distributed in the African continent south of the Sahara, and south of the Himalayas throughout Southeast Asia and into China north to the Yangtze River (*Basic Facts About Elephants - Global Sanctuary For Elephants*). The global distribution of wild or free-ranging elephants includes 27 countries, 13 in Asia and 14 in Africa (*Basic Facts About Elephants - Global Sanctuary For Elephants*, n.d.). Three elephant species are recognized, the Asian elephant, the African savanna elephant, and the African forest elephant (*Loxodonta cyclotis*) (Tollis et al., 2021). Among the Asian countries, India has the greatest number of elephants and the highest number of human TB cases (Chandranaiik et al., 2017). Among the African nations, Botswana has the greatest number of wild elephants and South Africa has the highest number of human TB cases (Rosen et al., 2018). Countries that have a high prevalence of TB in humans such as Nepal and Thailand also have TB in elephants (Angkawanish et al., 2010; Paudel et al., 2014). There are also around 15,000 to 20,000 captive elephants spread worldwide in zoos, circuses and others private owners (*Elephants in Captivity*, n.d.).

TB in elephants was recognized more than 2000 years ago (Iyer, 1937). The first case of TB in elephants was reported in London Zoo, 1875 (Mikota et al., 2000). Sporadic cases of TB in captive Asian elephants were reported in the early 20th century. It was only in the mid-20th century, that the first case in an African elephant was reported. After the initiation of systematic surveillance in 1998 in the U.S, the number of TB cases in elephants is rising (Mikota, 2020). However, there is a lack of a robust surveillance system to report TB cases and deaths linked to TB in elephants. The main causative agent for TB in elephants is *M. tuberculosis* (Lewerin et al., 2005). Captive elephants are more prone to TB as they are often in close and frequent contact with potentially infected human beings. It is presumed that there are possibilities of *M.*

tuberculosis transmission between humans and elephants or between wild and domestic elephants (Zachariah et al., 2017). The direction of transmission (elephant to elephant, elephant to human or human to elephant) has not been determined in most cases. Likewise, it has been suggested that cattle with close contact with humans could get infected with *M. tuberculosis* and eventually transmit it to elephants via contaminating the same grazing field as used by elephants (Zachariah et al., 2017). However, to date there is no documented transmission of *M. tuberculosis* between elephants and livestock. There are limited reports on TB in free-ranging elephants (Chandranaiik et al., 2017; M. A. Miller et al., 2019; Obanda et al., 2013; Perera et al., 2014; Zachariah et al., 2017).

Given that TB is present in both captive (Amy Zlot et al., 2016; Michalak et al., 1998; Montali et al., 2001; Murphree et al., 2011) and free-ranging wildlife (Montali et al., 2001), it represents a considerable zoonotic risk. Some evidence suggests infection of *M. tuberculosis* in a variety of domestic animals such as dogs (Parsons et al., 2012) and cattle (Ameni et al., 2011), as well as in non-domestic animal species including elephants (Che-Amat, 2018; Lecu & Ball, 2011; LoBue et al., 2010; Martin, 1994). In some species such as cattle and goats (Bezoz et al., 2015), *M. tuberculosis* infections are self-limiting and persistence of the pathogen in the population does not occur without repeated exposure to human cases (Ameni et al., 2013). However, the dynamics of *M. tuberculosis* transmission among wild animal species remains uncertain (Ghielmetti et al., 2017) as no evidence of *M. tuberculosis* infection in wildlife outside zoos could be observed (Oh et al., 2002). A holistic understanding is needed on transmission pathways of zoonosis and reverse zoonosis of *M. tuberculosis* and other members of MTC in different species, especially in elephants. Thus, the present study aims to accumulate evidence on the occurrence, transmission pathways, and preventive measures of TB in elephants.

1.8.3 Results

1.8.3.1 Study Selection and Study Characteristics

From the 122 articles selected for the abstract review 54 articles passed the filters. After the full-text reviews, 27 articles were selected for the study with focus on studies on prevalence of TB in elephants, dynamics of MTC transmission, and preventive measures of TB. A total of 27 articles described the possible chain of transmission and major preventive measures. Among those articles, there were 14 epidemiological, one clinical research, four outbreak investigations and two review articles. A total of 14 studies assessed the prevalence of TB in elephants, 12 being epidemiological studies and one clinical research. None of the articles

with outbreak investigations and review articles had sufficient evidence to determine the prevalence of TB in elephants.

1.8.3.2 Main findings: Assessment and Prevalence

The seroprevalence of TB in elephants varied from 0% to 23.33%, however, there is a variation of seroprevalence between wild and captive elephants, African and Asian elephants. The TB seroprevalence among captive Asian elephant's ranges from 15.2% to 23.33% (Abraham, D. (Research Associate et al., 2019; Ong et al., 2013; Thapa et al., 2021; Verma-Kumar et al., 2012; YAKUBU, 2015), while in captive African elephants is approximately 17% (Rosen et al., 2018)). The point prevalence of *M. tuberculosis* infection in Asian elephants was 5.1% for the time period of 1997 to 2011, while it remained 0 in African Elephants for the same time period (Feldman et al., 2013). There are also sporadic reported TB cases in wild Asian elephants as well as wild African elephants. However, TB prevalence in wild elephants has not been adequately studied to fully understand its dynamics and transmission pathways. The evidence indicates wild elephants can maintain human TB in the wild and that the infection can be fatal (Chandranaiik et al., 2017; Miller et al., 2019).

We identified an increased risk of TB in elephants with the growth in exposure to potentially TB infected humans or animals including other elephants, and vice versa. The studies used a wide variety of diagnostic measures throughout the years. Culture of the trunk-wash sample was preferred as a diagnostic tool before more rapid methods of TB diagnosis became available. We found the application of more than one diagnostic method to determine the prevalence of *M. tuberculosis* infection in elephants, which signals the advancements made over the years in the diagnostic technologies. The gradual innovation of TB diagnostic tools has provided multiple available methods for diagnosis. For instance, elephant TB Stat-Pak has been replaced by Dual Path Platform (DPP) TB test (Paudel et al., 2018) (Table 1.2 and Table 1.3).

Table 1.2 Main findings and seroprevalence and *M. tuberculosis* infection prevalence of included studies using serology and culture methods, respectively.

S. N	Authors	Research Design	Type of diagnosis	Diagnostic method	Country	Elephant species	Source	Prevalence (%)	Main findings
1	Mikota et al. (2000)	Cross sectional study	Culture	Culture	USA	Asian Elephant	Captive and Free ranging	3.3%	Further research is essential to validate other diagnostic tests and treatment protocols.
2	Abraham (2009) (Abraham, D.)	Cross-sectional study	Serology	Elephant TB Stat-Pak	India	Asian Elephants	Captive	15.2%	The rapid serum test showed that not less than 15.2% of the population has seropositivity to <i>M.bovis</i>
3	Murphree et al. (2011)	Cohort study and onsite assessment	Culture	Culture of environmental samples and trunk wash.	USA	Asian and African elephant	Refuge	All were Negative except for specimens taken in December 2008 from 1 elephant living in the quarantine area	The risk for conversion was increased for elephant caregivers and administrative employees working in the barn housing
4	Verma et al. (2012)	Cross-sectional study design	Culture & Serology	Culture, ELISA, Immuno blot analysis	India	Asian elephant	Captive	15.9%	High prevalence of asymptomatic <i>M. tuberculosis</i> infection in Asian elephants in a captive Indian setting.
5	Feldman et al. (2013)	Prevalence study	Culture	Culture of trunk wash sample (n1 = 684 Asian, and n2=459 African)	USA	Asian elephant	Captive	5.1%	The incidence of tuberculosis differed significantly between Asian and African elephants. Accurate and species-specific knowledge of prevalence and incidence will inform our efforts to mitigate occupational risks associated with captive elephants in the USA.
6	Ong et al. (2013)	A cross-sectional study	Serology	Elephant TB Stat-Pak assay/ TB antigen rapid test, trunk wash samples	Malaysia	Asian elephant	Captive	20.4%	There was evidence of active and latent TB in the elephants and the high seroprevalence in the elephants and their handlers suggests frequent, close contact, two-way transmission

				QuantIFERON-TB Gold ELISA					between animals and humans within confined workplaces.
7	Yakubu (2015)	Cross sectional study	Molecular	PCR	Malaysia	Asian elephant	Captive	23.33%	Risk of infection to be significantly associated with facility staff, workers older than 30 years of age, mahouts, and foreigners.
8	Mikota et al. (2015)	prospective study	Serology & Culture	Elephant TB Stat-Pak assay, Culture	Nepal	Asian Elephant	Captive	0%	Culture and serological results were variable and required careful interpretation to develop criteria to assess TB risk.
9	Yakubu et al. (2016)	Cohort and Cross-sectional study	Serology	STAT-PAK and DPP VetTB Assays, QuantIFERON-TB Gold In-tube test (QFT) were used.	Malaysia	Asian elephant	Captive	23.3 %	Univariate analysis showed that elephants with assigned mahouts have significantly higher risk of TB OR=3.8 The risk of seroconversion was significantly higher among elephants with assigned mahouts [OR=4.9]
10	Magnuson et al. (2017)	Clinical research	Culture and Molecular	Culture, PCR	USA	Asian elephant	Captive	8.4%	Molecular test results can be used to support current diagnostic procedures applied by veterinarians for treatment decisions to prevent the spread of tuberculosis in elephants.
11	Rosen et al. (2018)	Cross sectional study	Serology	Elephant TB Stat-Pak and DPP VetTB Assay	Zimbabwe	African elephant	Captive	17.1%	Strong correlations were noted between contact with wild elephants and facilities (p=0.86)
12	Jeewan et al (2021)	Cross sectional study	Serology	Elephant TB Stat-Pak	Nepal	Asian elephant	Captive	21.56%	The occurrence of TB seropositive cases in other more remote national parks suggest TB may be wide spread among the captive elephant population of Nepal

PCR Polymerase chain reaction, ELISA Enzyme-linked immunosorbent assay, TB Tuberculosis

Table 1.3 Main findings and seroprevalence and *M. tuberculosis* infection prevalence of included studies using other test

S. N	Authors	Research Design	Diagnostic methods	Elephant species	Source	County	Prevalence (%)	Main findings
1.	Larsen et al. (2000)	Cross-sectional observational study design	ELISA for Screening	Asian and African elephant	Captive	USA	14.89 %	Multiple-antigen ELISA would be a valuable screening test for detecting <i>M. tuberculosis</i> infection in elephant herds.
2.	Zachariah et al. (2017)	Surveillance	PCR, postmortem examination	Asian elephant	Free ranging	India	3.4 %	Exposure of bulls to humans infected with TB during conflict activities could be the possible explanation regarding the transmission of disease. All 3 animals were emaciated and considered TB to be the cause of death.

1.8.3.3 Transmission pathways and preventive measures

We analyzed a total of 27 studies to assess the transmission pathways and preventive measures. The studies were carried out in diverse elephant populations including captive and wild Asian and African elephants. We identified more studies among captive elephants than among wild ones. These studies were conducted across different continents - America, Africa, Asia, and Europe. Three overarching interrelated themes for transmission pathways were identified: between humans and elephants, other animals to elephants, and unclear source of infection. Several studies suggested transmission between humans and elephants (Amy Zlot et al., 2016; Lassausaie et al., 2015; Michalak et al., 1998; M. A. Miller et al., 2019; Murphree et al., 2011; Oh et al., 2002; Ong et al., 2013; Stephens et al., 2013). Few of the studies described the plausible transmission from other animals (wild and domestic) to elephants (Greenwald et al., 2009; Lassausaie et al., 2015; Lewerin et al., 2005; Stephens et al., 2013).

Several studies lacked information on the source of infection (Chandranaik et al., 2017; Obanda et al., 2013; Paudel et al., 2014).

Studies of TB in captive elephants from Thailand, Malaysia, Switzerland, USA, Nepal, India, and some African countries revealed possible transmission of *M. tuberculosis* between elephants and humans (Angkawanish et al., 2010; Michalak et al., 1998; Murphree et al., 2011; YAKUBU, 2015; Paudel et al., 2018; Rosen et al., 2018;). One of the studies carried out in Southern India among wild elephants indicated that TB might be spilling over from humans (reverse zoonosis) and emerging in wild elephants (Zachariah et al., 2017). Similarly, evidence from Kenya and India showed wild elephants could harbor *M. tuberculosis* and revealed that the infection could be fatal. However, the transmission sources of infection were unclear regarding if the infections originated from humans or other animals (Obanda et al., 2013; Chandranaik et al., 2017;). Likewise, studies in Sweden, Australia, and the U.S. indicated possible transmission from elephants to elephants as well as to other captive animals, especially in zoos (; Lewerin et al., 2005; Stephens et al., 2013; Amy Zlot et al., 2016). Hence, it signifies the need for a One Health perspective to combat TB due to probable transmission pathways between humans, elephants, and other animals. These pathways could be partly mediated by environmental matrices such as shared water or feed (Allen et al., 2021).

We identified possible preventive measures for *M. tuberculosis* transmission and infection control measures. The possible chain of transmission could be interrupted through routine TB screening among elephant handlers, occupational health programs, early diagnosis and treatment of infections among the elephant handlers, isolation of infected elephants and improving TB screening methods for elephants especially among the captive elephants (Maslow & Mikota, 2015; Paudel et al., n.d.). Additionally, it is essential to perform screening of newly acquired elephants, isolation of infected elephants and early treatment of confirmed cases in captive elephants. Likewise, it is important to ensure TB screening of captive elephants before releasing them into the wild (Obanda et al., 2013; Zachariah et al., 2017) . Similarly, minimizing shared feed with other wildlife could reduce the exposure. For early detection and efficient treatment, accurate antibody tests such as the DPP Vet TB test are already available (Ong et al., 2013). However, better ways to identify culture-positive elephants are still needed given the limitations of trunk wash. Thus, there is a need to develop a blood antigen test that can identify culture-positive elephants with improved sensitivity (Songthammanuphap et al., 2020). One measure regarding diagnosis in elephants is to use a combination of diagnostic approaches as single diagnostic measures cannot always identify TB (Ong et al., 2013; YAKUBU, 2015) (Table 1.4).

Table 1.4 *Mycobacterium tuberculosis* transmission pathways and preventive measures

S.N	Study references	Elephant species	Source	Country	Disease transmission	Preventive measures
1.	Zachariah et al. (2017)	Asian elephant	Free ranging	India	Between human and elephant	Continued Surveillance among elephant population.
2.	Obanda et al. (2013)	African elephant	Free ranging	Kenya	Source of infection is unclear	Domesticated elephants into the wild require efficient screening for TB, the status of TB in wild elephants should be assessed.
3.	Angkawanish et al. (2010)	Asian elephant	Captive	Thailand	Between human and elephant	Early diagnosis of infection is necessary. Combination of diagnostic approaches is essential.
4.	Yakubu (2015)	Asian elephant	Captive	Malaysia	Between human and elephant	The need for control strategies such as screening and newly acquired elephants, isolation of infected elephants and early treatment of confirmed cases.
5.	Ong et al. (2013)	Asian elephant	Captive	Malaysia	Between human and elephant	Elephant handlers need to be aware of the risk of TB acquisition from infected animals and be educated concerning infection control measures.
6.	Murphree et al. (2011)	Asian and African elephant	Refuge	USA	Between human and elephant	Increased knowledge about MTB infection in elephants, improved infection control practices, and specific occupational health programs.
7.	Feldman et al. (2013)	Asian and African elephant	Captive	USA	The difference in species susceptibility	Mandatory annual tuberculosis screening for all the elephants.

8.	Rosen et al. (2018)	African elephant	Captive	Zimbabwe	Source of infection is unclear	Routine TB testing of elephant handlers and regular serological screening of elephants are recommended as preventive measures.
9.	Michalak et al. (1998)	Asian or African elephants	Free-ranging	USA	Between human and elephant	Veterinary practices should be initiated to reduce the risks of exposure to animals infected with MTB.
10.	Chandranaiik et al. (2017)	Asian elephant	Free-ranging	India	Source of infection is unclear	Wild elephants can harbor MTB that can become fatal. Need to assess the status of TB among wild animals and to examine whether wildlife can be a potential reservoir of the disease.
11.	Ghielmetti et al. (2017)	Asian elephant	Captive	Switzerland	Between human and elephant	Different transmission chains or prolonged infection over time.
12.	Magnuson et al. (2017)	Asian elephant	Captive	USA	Between human and elephant	Molecular test results can be used to support current diagnostic procedures applied by veterinarians for treatment decisions to prevent the spread of tuberculosis in elephants.
13.	Rosen et al. (2018)	African elephant	Captive	Five countries of Africa	Between human and elephant	Minimizing exposure through shared feed with other wildlife, routine TB testing of elephant handlers, and regular serological screening of elephants are recommended as preventive measures.

14.	Simpson et al. (2017)	Asian and African elephant	Captive	USA	Between human and elephant	Infection control protocols and careful monitoring of the treatment of captive elephants with tuberculosis are warranted.
15.	Yakubu et al. (2016)	Asian elephant	Captive	Malaysia	Between human and elephant	The need for TB screening of newly acquired elephants, isolating seropositive elephants and performing further diagnostic tests to determine their infection status, and screening elephant handlers for TB, pre- and post-employment.
16.	Amy Zlot et al. (2016)	Asian elephant	Captive	USA	Between human and elephant	Improved TB screening methods for elephants are needed to prevent exposure of human contacts.
17.	Lassausaie et al. (2015)	Asian elephant	Captive	Laos	Between human and elephant	Medical monitoring of people working or living with elephants should thus be implemented.
18.	Paudel et al. (2014)	Asian elephant	Captive	Nepal	Between human and elephant	Regular TB screening of elephant handlers to safeguard human health and help prevent transmission of TB from humans to elephants.
19.	Lewerin et al. (2005))	Asian elephant	Captive	Sweden	Between Elephants and Captive Animals	Elephants and giraffe were found to have been infected by four different strains of MTB in a large Swedish zoo.
20.	Larsen et al. (2000)	Asian and African elephant	Captive	USA	TB infected elephants are the potential source of infection	Multiple-antigen ELISA would be a valuable screening test for detecting MTB infection in elephant herds.

21.	Paudel et al. (2018)	Asian elephant	Captive	Nepal	Source of infection is unclear	Regular TB screening of elephant handlers to safeguard human health and help prevent transmission of TB from humans to elephants.
22.	Stephens et al. (2013)	Asian elephant	Captive	Australia	Elephant to Humans and other animals	The mechanism for transmission from elephants in the Australian zoo require further investigation.
23.	Montali et al. (2001)	Asian Elephant	Captive and Free-ranging	USA	Transmissions of MTB between animals and humans are uncommon	Programs for tuberculosis prevention in animal handlers.
24.	Mikota & Maslow (2011)	Asian Elephant	Captive	USA	Between human and elephant	More epidemiological investigation in the transmission.
25.	Greenwald et al. (2009)	Asian and African elephant	Captive	Europe and the USA	Elephant to Humans and other animals	Rapid and accurate antibody tests to identify infected elephants will likely allow earlier and more efficient treatment, thus limiting the transmission of infection to other susceptible animals and to humans.
26.	Mikota et al. (2000)	Asian Elephant	Captive and Free ranging	USA	Elephant and human	Further research is essential to validate other diagnostic test and treatment protocols
27.	Jeewan et al. (2021)	Asian elephant	Captive	Nepal	Between elephant to other hosts, including humans	Include blood parameters in future TB surveillance studies.

Abbreviations: MTB, *M. tuberculosis*; ELISA, enzyme-linked immunosorbent assay; TB, tuberculosis

1.9 Discussion

This review revealed that TB in elephants is widespread across the globe. In general, the rates of *M. tuberculosis* infection are higher in Asian elephants compared to African elephants (Orloski, n.d.). Confirmed *M. tuberculosis* infections are reported both in wild and captive elephants across different countries, although being more frequent in captive elephants. The same *M. tuberculosis* strain in an elephant and a handler has been reported in only one case (Michalak et al., 1998). However, most human cases have been diagnosed by tests such as the intradermal tuberculin test, Quantiferon, chest X-ray, among others, which do not identify the mycobacteria strain involved (Acharjya, 2012; Babayigit et al., 2014; Herrera Diaz et al., 2020; Nayak & *Rapid Communication on Updated Guidance on the Management of Tuberculosis in Children and Adolescents*, n.d.; WHO, 2021). Therefore, multidisciplinary interventions with inter sectoral coordination must be implemented to combat TB in elephants and humans.

Even though the cases of direct *M. tuberculosis* transmission are apparently rare, there are odds of transmission when infected elephants are at an advanced stage of this disease. Due to paucity of research in wild elephants, the occurrence of *M. tuberculosis* was mostly observed in captive elephants.

For instance, *M. tuberculosis* isolates were extracted from two elephants of Chitwan National Park and one elephant of Koshi Tappu Wildlife Reserve of Nepal. These elephants were in contact with domestic and wild animals like rhinos and different deer species along with their handlers (Paudel et al., 2014). Additionally, in a sero-surveillance program of captive elephants in Nepal, out of 153 elephants, 21.56% were serologically tested positive (Thapa et al., 2021). There are evidences that human handlers might act as a source of infection for the animals (Miller et al., n.d.). Likewise, at a zoo in the US, *M. tuberculosis* was diagnosed in an elephant, a rhino and three mountain goats (Oh et al., 2002). Seven out of 24 keepers in the south-east zoo in the U.S. tested positive in intradermal tuberculin tests and were assumed to be infected by airborne transmission from an affected white rhinoceros. In this regard, a white rhinoceros tested positive for *M. bovis* that later spread to colobus monkeys (Dalovisio et al., 1992). In a separate study, *M. tuberculosis* strains isolated from captive elephants in Thailand seemed to have originated from humans (Angkawanish et al., 2010). The transmission of *M. tuberculosis* from elephant-to-elephant and elephant to other animals is also possible (Dalovisio et al., 1992; Michalak et al., 1998; Paudel et al., n.d.; Suga et al., 2021). Yet, a clear transmission pathway and the source of *M. tuberculosis* infection between animals has not been established (Stephens et al., 2013; Maslow & Mikota, 2015; Backues & Wiedner, 2019).

Many countries are dependent on elephant-related industries such as agriculture and tourism leading to more interaction between humans and elephants (Miller et al., 2019). The tourism activities lead to increased interaction between captive elephants, humans, and wild elephants. TB in wild elephants is an emerging challenge. There are few studies reporting TB among wild elephants (Dalovisio et al., 1992; Zachariah et al., 2017; Miller et al., 2019; Tollis et al., 2021). Still, more investigation is needed to promote further epidemiological studies among wild elephants (Babayigit et al., 2014) (Paudel et al., n.d.). The surveillance and epidemiological studies on wild elephants are more complicated as it is difficult to track the exact location, number, and duration of potential exposure (Miller et al., 2019). There are multiple diagnostic and screening tools available to assist in the diagnosis of MTC in elephants. Even so, confirmation of the true diagnosis of the clinical disease remains challenging (Kay et al., 2011; Backues & Wiedner, 2019).

In the absence of highly sensitive diagnostic assays; a combination of routine medical examination could be recommended (Backues & Wiedner, 2019). Standard tests such as VetMAX™ MTBC qPCR Kit (Goosen et al., 2020), and serological tests using Elephant TB STAT-PAK,® DPP VetTB® Assay, MAPIA (multi-antigen print immunoassay) (Paudel et al., 2018), and interferon gamma release assay (IGRA) (Songthammanuphap et al., 2020) are used for detection of MTC in different parts of the world based on accessibility.

Often, TB is transmitted from droplet nuclei (i.e., respiratory secretions). *M. tuberculosis* has been isolated from respiratory secretions, trunk washes, feces, and vaginal discharges in elephants (Montali et al., 2001). TB can also be transmitted from elephants to humans (Amy Zlot et al., 2016; Murphree et al., 2011). Transmission of elephant *M. tuberculosis* to humans is more likely an occupational health concern rather than a general public health concern (Michalak et al., 1998; Murphree et al., 2011; Paudel et al., n.d.). However, with the increased use of elephants in the entertainment and tourism sector, it is likely to increase *M. tuberculosis* transmission beyond the occupational health concern.

Therefore, the One Health perspective consisting of activities like adopting infection control measures in the captive environment, routine TB screening among elephant handlers, occupation health programs, and establishing a mechanism for early diagnosis of infection among elephants is recommended. Additionally, it is essential to ensure screening of TB before the release of captive elephants into the wild as well as before adapting wild elephants into the captive environment (Obanda et al., 2013). Furthermore, elephants with active TB should be segregated and treated, which will aid in the prevention of *M. tuberculosis* transmission between species and will contribute to the conservation of elephants (Paudel et al., n.d.). Although caution is needed considering antibiotic resistance.

The review is limited to the systematic analysis of the literature and has not executed a meta-analysis. The findings of the study are based on a relatively limited number of studies; thus it is challenging to fit into the PRISMA format. There is a high variation in study design, sample size, screening, and diagnostic tools used in the included studies. Thus, it is difficult to compare the best method because of the variety of diagnostic tests employed. Nevertheless, this review highlights the need to carry out more epidemiological research in both wild and captive elephant populations to determine the exact prevalence, chain of transmission, and other related factors. There is a huge gap in the evidence on the dynamics of *M. tuberculosis* transmission between other animals and elephants, reinforcing the need to promote research to develop innovative and robust screening and diagnostic tools for the early diagnosis of *M. tuberculosis* and other members of the *M. tuberculosis* complex in elephants. This would help to reduce the risk of TB in elephants as well as in the in-contact human population and further contribute to prevent possible transmission to other animal populations.

1.10 Conclusion

M. tuberculosis infection has affected the elephant population, and in particular, Asian elephants. Recent studies suggest human-elephant and elephant-human *M. tuberculosis* transmission. It is a public health concern that needs a One Health perspective with a combined effort of biologists, public health and occupational health experts, and veterinarians to reduce the occurrence of zoonosis and reverse zoonosis of *M. tuberculosis*.

1.11 Methods

1.11.1 Overview

This is a systematic review grounded on the preferred reporting items for systematic reviews and meta-analyses (PRISMA) guidelines (Moher et al., 2009). The keywords like *Mycobacterium tuberculosis*, prevalence, transmission, prevention, and elephants were scanned in Google Scholar, PubMed, Science Direct, and Web of Science. The articles were explored without the time constraints and the articles were sorted for relevance with the keywords.

1.11.2 Literature search

A total of 2480 articles were obtained during the first search. Then, the identified articles were saved in Zotero (a reference management software) (*Digital Scholar*, n.d.). Duplicate articles were sought using the title, DOI, and ISBN fields in Zotero. If these fields match (or are absent), years of publication (if they are within a year of each other) and author/creator lists (if at least one author last name plus first initial matches) were explored to determine the replicas. The

identical articles were managed by merging them, rather than deleting one of the duplicates. Furthermore, the titles of the articles were assessed and the articles that were not relevant to keywords were excluded. A total of 122 articles were selected for the abstract review. A report of all the articles with titles and abstracts were generated in a file. The abstracts of the articles were reviewed, and 54 articles were selected for full-text review based on various parameters (Table 1.5). After the full-text review, 27 articles were selected for the study, focusing on studies about the prevalence of TB in elephants, dynamics of *M. tuberculosis* transmission, and preventive measures of TB. The selected articles were entered in MS-excel version 13.0 including the major outcomes of the studies. Then, the findings and major outcomes were analyzed and interpreted for the result of the study (Figure 1.3). Furthermore, additional literature like reports on the global distribution of elephants, country-wise elephant population, distribution of TB in humans, and high TB burden countries were also reviewed.

Table 1.5 Inclusion and exclusion criteria peer-reviewed literature

Parameter	Inclusion	Exclusion
Study design/ type	• Meta-analysis or systematic review • Randomized controlled trails • Non-randomized, prospective comparative studies • Prospective observational studies (e.g., cohort studies) • Retrospective observational studies (e.g., case-control studies) • Cross-sectional studies • Case studies • Outbreak Investigation • Clinical studies • Short communication	• Narrative review • Non-pertinent publication types (e.g., expert opinions, letters) • Editorials, comments, conference, • Abstract/poster, news, consensus document, chapter
Study Quality	• Number of subjects (no minimum) • Study duration (no minimum)	• Insufficient methodological quality (both inherent methodology as well as an insufficient description)
Study Population	• Wild and captive elephants • Contacts of wild and captive elephants (Human and other animals)	• Other animals (wild and domestic animals) which are not the contacts of elephants

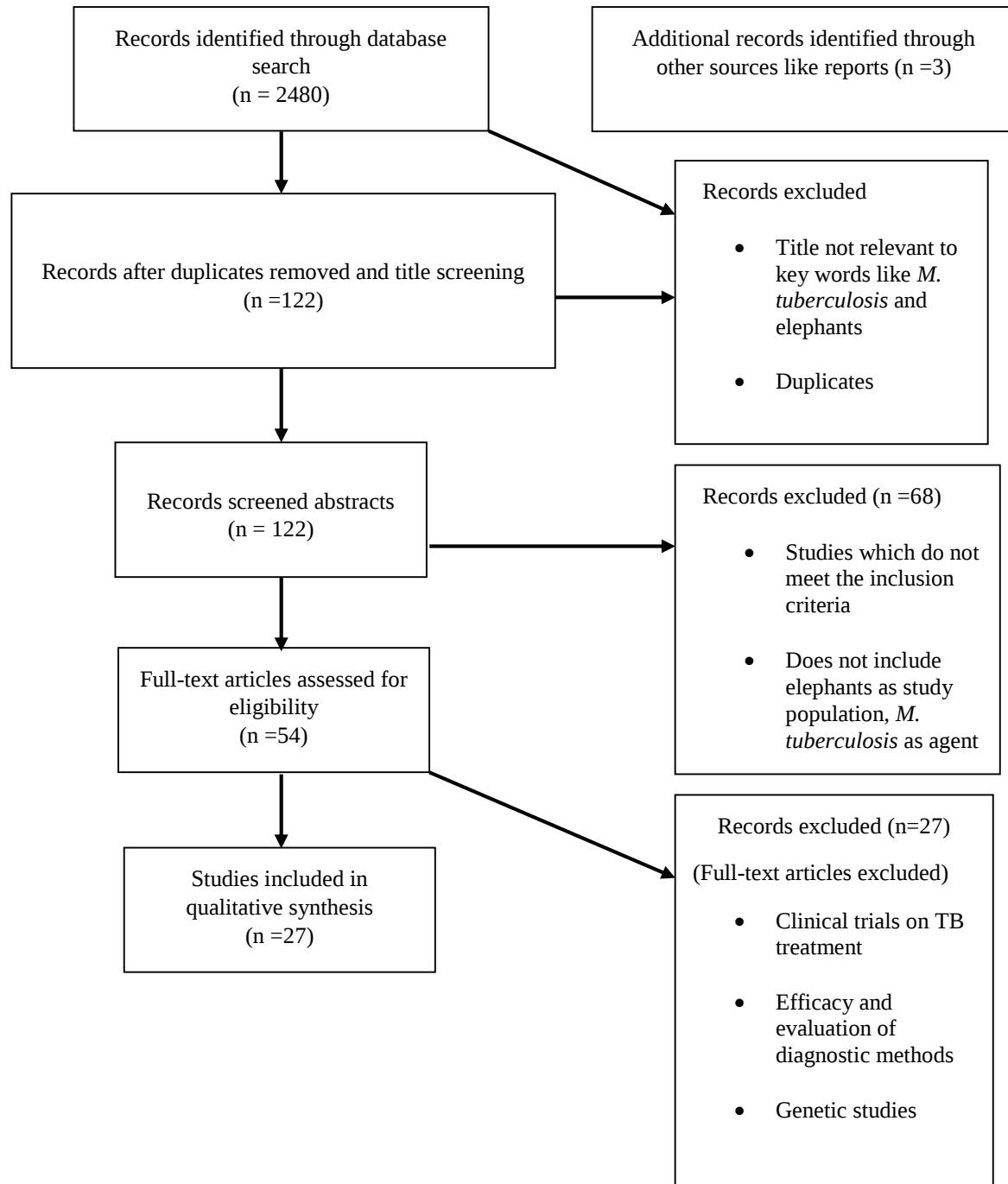


Figure 1.3 Flowchart of the systematic review

1.11.3 Article Inclusion and Exclusion Criteria

The inclusion and exclusion criteria were established on different parameters like study design/type, quality of the study, and the study population. The studies like meta-analysis/systematic review, randomized controlled trials (RCTs), prospective studies, cohort studies, cross-sectional studies, case studies, outbreak investigation, and clinical studies were involved in the review. On the other hand, narrative reviews, non-pertinent publications, opinions, news articles, abstracts were omitted in the review. Similarly, studies with any time duration or number of the study population were also embraced for the review. Contrarily, research with insufficient methodological quality as well as the insufficient description was barred from the review. Likewise, research carried out in wild and captive elephants including their contacts were encompassed in the review. Studies on TB in other animals were excluded from the review. The details of the inclusion and exclusion criteria are listed in Table 4.

1.11.4 Search Strategy with Databases

The search was performed until 2021 and included only the articles that were published in English. The newspaper articles, blog posts, conference abstracts, narrative reports, editorials, and field visit reports were excluded for the study.

1.11.5 Data Extraction

Two authors screened the search results. The following information was extracted from each paper: publication year, country, type of elephants, species of elephants, study design, sample size, the sample used for diagnosis, diagnosis methods, prevalence, mortality, and major outcome. The affiliation of authors, methods of diagnostic tools used, and type of elephant included in the study, and investigated research methods were noted to sort out the research. After the full article review, based on the above-mentioned information, authors reached the consensus to include the article for review.

1.11.6 Risk of Bias and Article Quality

The sources of bias were assessed for the articles that met the inclusion criteria. The studies with screening and diagnostic tests for TB in elephants were included in the review. So, the common sources of bias in diagnostic accuracies like partial verification bias, clinical review bias, and observer or instrument variation bias were assessed in the articles. The authors ensured the methodological issues including content and methods, which included consistency assessment of twenty-seven studies between the authors. Besides, studies were assessed for a sufficient description of the methodology used. The studies with an insufficient description of the methods were not included in the study. Furthermore, the authors re-

checked for the missing information such as study population, design, and outcome of the research.

1.11.7 Abbreviations

M. tuberculosis: *Mycobacterium tuberculosis*; MTC: *Mycobacterium tuberculosis complex*; PRISMA: Preferred reporting items for systematic reviews and meta-analyses; ELISA; Enzyme-linked immunosorbent assay; MAPIA: Multiantigen print immunoassay ; DPP :Dual path platform ; qPCR : Quantitative real-time Polymerase chain reaction ; IGRA :Interferon gamma release assay

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Chapter 2: Human risk factors associated with the presence of antibodies against *Mycobacterium tuberculosis* complex in captive Asian elephants from Chitwan valley, Nepal*

2.1 Abstract:

Tuberculosis (TB) caused by *Mycobacterium tuberculosis* in elephants and humans is an increasing public health concern in Nepal. We tested 99 captive Asian elephants (*Elephas maximus*) from Chitwan valley for antibodies against the *M. tuberculosis* complex (MTC) and interviewed their 223 mahouts to investigate putative associations between human risk factors and MTC antibody detection. Bivariate analysis and multivariate logistic regression were used to assess the links between mahout socio-demographic characteristics, elephant husbandry, interaction with other animals, and TB antibody detection in elephants. The apparent prevalence of antibodies against MTC in elephants was 20.2% (95%CI: 13-29%). Regarding the social-demographic aspects of the mahouts, no factor appeared as a relevant risk for finding TB in the elephants they cared for. The most relevant results were obtained through questions on elephant husbandry. Identified risks included elephant ownership (more TB in government owned elephants), hand-feeding the elephant (as opposed to using a bucket or the floor), not having special personal protective equipment at work and disposing of elephant waste in pits (as opposed to other means). Contact with other animals and inter-species contacts also revealed risk factors. Elephants cared for by mahouts with livestock present in their household or nearby their household were at higher TB risk. Also, elephants cared for by mahouts reporting having found a dead animal and collecting it to eat or share were at higher risk. Elephants belonging to mahouts reporting that wild elephants came to interact with the captive ones were at lower TB risk. This study highlights the importance of a well-coordinated One Health program involving both the animal and human health sectors to deal, limit and prevent the spread of MTC in multi-host communities where TB remains at high prevalence, particularly in humans.

Keywords: Biosecurity measures; Cross-sectional survey; Interview; Mahout; Prevention; Zoonosis.

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2.2 Introduction

Tuberculosis (TB) in mammals, including humans, is caused by members of the *Mycobacterium tuberculosis* complex (MTC) (Michel, 2014). Among the members of the MTC, *M. bovis* is the primary species responsible for TB in domestic and wild mammals (De La Rua-Domenech, 2006), while *M. tuberculosis* is the main causative agent of disease in humans and elephants (Sternberg Lewerin et al., 2005). Captive elephants are prone to TB because they are often in very close and frequent contact with potentially infected humans. Captive elephants often have direct or indirect contact with their wild counterparts and with other wild and domestic animals. This puts the elephants at risk when serving as bridge hosts for transmitting the infection to other wild animals and livestock and back to humans (Zachariah et al., 2017). In Nepal, in a sero-surveillance program of captive elephants, out of 153 elephants 21.56% tested positive for serum antibodies against MTC (Thapa et al., 2021).

Human TB is reported almost in all countries of the world and is one of the top ten causes of death and the leading cause of death among adults in Nepal (National Tuberculosis Control Center, 2018). According to the 2020 national TB prevalence survey, 69,000 (41,000–103,000) people developed clinical TB in Nepal in 2018 (Population, n.d.). Regarding elephant TB, the first case was reported in the mid-20th century in an African elephant (*Loxodonta africana*). After the initiation of systematic surveillance in 1998, the number of TB cases in elephants increased (Mikota, 2020). Tuberculosis has a very complex epidemiological pattern where, depending on the MTC agent involved and the local host community composition, the transmission chain can include humans, domestic animals, and wildlife (Bengis et al., 2004). Transmission is not only direct but there is also indirect transmission through contamination of the environment, water, and feed (Murphree et al., 2011). Because contacts between captive and wild elephants are common in several areas of Nepal, the risk of spreading TB to wild populations could be significant (Martin, 1994).

Nepal is home to Asian elephants (*Elephas maximus*) that are still used in the timber industry (Pradhan et al., 2011; Shrestha & Shrestha, 2021), for military and national park security, and for tourism (Bale, 2016; Szydlowski, 2022). At present, there are altogether 208 captive elephants in Nepal out of which 94 government elephants are in various parks in the Terai, namely Koshi Tappu Wildlife Reserve, Parsa Wildlife Reserve, Chitwan National Park, Bardia National Park and Suklaphanta Wildlife Reserve (Pradhan et al., 2011). The ownership of an elephant is also shared between several people, and some owners may employ more than one mahout to take care of and ride their elephants. As a result, there are often two or more persons working every day in close contact with each animal. Given the proximity between mahouts and elephants, there is a concern that infection transmission may take place in both

directions (Mikota et al., 2015). Elephant TB can be accurately diagnosed by serum antibody detection (DNPWC, 2011; Greenwald et al., 2009; Lyashchenko et al., 2012; Paudel et al., 2018). Although there are studies on prevalence of TB in animals in Nepal (Paudel et al., 2014; Thapa et al., 2015; 2016 2021) and literature on human TB (Population, n.d.), the risk factors for elephant TB regarding mahout socio-demographic characteristics, elephant husbandry, and interaction with other animals, has not been previously assessed. Thus, the purpose of this study was to investigate the association between potential human risk factors through interviewing 223 mahouts and elephant caretakers and serum antibody detection against the MTC in captive elephants from the Chitwan valley.

2.3 Material and Methods

2.3.1 Study design

A cross-sectional study was conducted in Chitwan National Park established in 1973 and granted the status of a World Heritage Site in 1984. It covers an area of 952.63 km² (367.81 sq mi) and is in the subtropical Inner Terai low lands of south-central Nepal in the districts of Nawalpur, Parsa, Chitwan and Makwanpur. In altitude it ranges from about 100 m (330 ft) in the river valleys to 815 m (2,674 ft) in the Churia Hills (Shrestha & Shrestha, 2021). The National Park is home to 111 captive elephants out of which 99 elephants were included in this study. A field survey was organized to blood-sample the elephants and interview the mahouts in their villages and in logging camps around the National Park. A multidisciplinary team of veterinarians and medical doctors coordinated by Intrepid Nepal (Kathmandu) collected the data.

2.3.2 Consents and ethics clearance

Written informed consent was obtained from all recruited participants before the interview. Ethical approval for the study was permitted from Nepal Ethical Review Board, Nepal Health Research Council (IRC num: 312/2018). The permit was obtained for information and sample collection in both elephants and elephant handlers (mahouts).

2.3.3 Rapid Diagnostic Test

The veterinary team performed a clinical examination of the 99 elephants located in the National Park and collected 5 ml of blood per case. Fresh serum samples were aliquoted, transported on ice to the lab and stored at -20°C until antibody testing. Trunk washes could not be performed in the field because of difficulties in implementing the method on untrained elephants. Sera were subjected to antibody testing using an animal-side rapid test for TB ("DPP® VETTB Assay for Elephants– Chembio Diagnostics, Inc., NY, USA"; Sensitivity 100%

and Specificity 100%; performed as recommended by the manufacturer. Results were interpreted visually as per the Chembio product information. The rapid test kit gives results on presence and absence of antibodies against TB and results can be stated as antibody reactive or non-reactive/seropositive or seronegative. For verification and validation purposes, the results were recorded independently by two observers, along with visual evidence (photographic evidence).

2.3.4 Study population and interviews

A total of 223 mahouts linked to 99 elephants were included in our study. We used a questionnaire in Nepali language addressing mahout socio-demographic characteristics, elephant husbandry, and interactions with other animals (Supplementary file #1).

2.3.5 Statistical methods

Data analyses in this study comprise descriptive analyses, univariate analyses and logistic regression analyses. Pearson's chi-squared test was used to determine characteristics and behaviors of mahouts regarding elephant TB status. A two-tailed p-value of <0.05 was considered to be statistically significant. The variables that were statistically significant in chi-square tests were then fitted into logistic regression models. Initially, explanatory determinants were included in the model one at a time to examine their univariate relationship with the outcome and variables that were significant in the univariate analysis were fitted in the multivariate analysis. Multivariate logistic regression models were then used to identify the most important determinants for each outcome. Adjusted odds ratios (AORs) at 95% confidence intervals (95% CI) were used to depict the independent relationship between predictors and dependent variables. Multicollinearity was checked among the variables, and there was no significant collinearity (variance inflation factor <1.14). Multivariate analysis with all independent variables entered at the same time was completed to adjust for the effect of confounders and adjusted OR and 95% CI were computed. The Hosmer–Lemeshow test was performed to test the goodness-of-fit of the multivariate logistic regression model, and the model was found to have a good fit ($p>0.05$). All statistical analyses for this study were conducted using R program for Statistical Analysis (The R Foundation, 2018).

2.4 Results

Twenty of 99 elephants tested positive for MTC antibodies (20.2%; 95%CI: 13-29%). Mahouts were interviewed regarding three main aspects: social-demographic factors, elephant husbandry, and inter-species contacts. Most of the mahouts were below 40 years old (66.8%), belonging to the Tharu caste (70%) and married (90%). Nearly 24% of the mahouts had received formal education, 60% had received informal education, and 16% were illiterate (Table 2.1). Nearly 40% of the mahouts knew about human TB signs and symptoms like cough >2 weeks, hemoptysis, and fever. 91% also knew that TB could occur in elephants. However, while assessing knowledge about TB signs in animals, 64% responded that they only heard about it (Chapter 2 Annex, Table S1). Among mahouts or their family members, 7.2% had a history of TB among which 87.5% received treatment. Out of 14 mahout family members who had TB, nine (64%) had no interaction with elephants, 21.4% had close interaction, and 7.1 % each had moderate and limited interaction with elephants. Most of the mahouts had no history of TB while three (1.3%) had TB in the past (Chapter 2 Annex, Table S2). Most of the mahouts (60.5%) had been taking care of the elephant for > 5 years (Chapter 2 Annex, Table S3). Regarding the social-demographic aspects of the mahouts, no factor appeared as a relevant risk for finding TB in the elephants they cared for. This included factors a priori regarded as TB-risks for humans was age of mahouts (Table 2.2).

Table 2.1 Interviewee profiles regarding age, caste, education, marital status, occupation, household size and room sharing.

Characteristics	Number (N=223)	Percent
Age of mahouts		
Below 30 years	79	35.4
30-39 years	70	31.4
40 and above	74	33.2
Median age (Min.-Max.)	33.0 (18-99)	
Mean age± Std.dev	35.1±10.85	
Caste		
Tharu	156	70.0
Other castes	67	30.0
Education of mahouts		
Illiterate	37	16.6
Informal education	132	59.2
Formal education	54	24.2
Marital status		
Married	201	90.1
Divorced/Permanently Separated	1	.4
Never married	21	9.4
Main occupation		
Mahout	121	54.3
Fanit	31	13.9
Pachuwa	13	5.8
In charge	58	26.0
Monthly family income		
20,000 and below	124	55.6
21,000-40,000	74	33.2
40,000 and above	25	11.2
Household size		
1-4 member	49	22.0
5 member	42	18.8
6 and above member	132	59.2
Average Household Size	4.6 person	
Number of room shared		
Two rooms	55	24.7
Three rooms	53	23.8
Four rooms	66	29.6
Five and more rooms	49	22.0

Table 2.2 Association between socio-demographic characteristics and Mahouts who handle TB +ve elephants (Source: Interviews carried out in 2019)

Characteristics	Mahouts who handle TB + Ve elephants			P-value
	Yes (n=41,18%)	No (n=182,82%)	Total (N=223)	
Age				
Below 30 years	13 (16.5)	66 (83.5)	79 (35.4)	0.047
30-39 years	8 (11.4)	62 (88.6)	70 (31.4)	
40 years and above	20 (27)	54 (73)	74 (33.2)	
Province				
Province 3	22 (17.5)	104 (82.5)	126 (56.5)	0.684
Other provinces	19 (19.6)	78 (80.4)	97 (43.5)	
Caste				
Tharu	29 (18.6)	127 (81.4)	156 (70)	0.904
Other castes	12 (17.9)	55 (82.1)	67 (30)	
Marital Status				
Married	36 (17.9)	165 (82.1)	201 (90.1)	0.567
Never married	5 (22.7)	17 (77.3)	22 (9.9)	
Education				
Illiterate	8 (21.6)	29 (78.4)	37 (16.6)	0.578
Literate	33 (17.7)	153 (82.3)	186 (83.4)	
Occupation				
Maute	18 (14.9)	103 (85.1)	121 (54.3)	0.141
Others	23 (22.5)	79 (77.5)	102 (45.7)	
Monthly income				
20,000 and below	25 (20.2)	99 (79.8)	124 (55.6)	0.444
Above 20,000	16 (16.2)	83 (83.8)	99 (44.4)	
Family members				
Less than 6 members	8 (16.3)	41 (83.7)	49 (22)	0.674
6 and above members	33 (19)	141 (81)	174 (78)	
Number of rooms				
2-3 rooms	14 (25.5)	41 (74.5)	55 (24.7)	0.119
More than three rooms	27 (16.1)	141 (83.9)	168 (75.3)	
TB in Family				
No	36 (17.4)	171 (82.6)	207 (92.8)	0.182
Yes	5 (31.2)	11 (68.8)	16 (7.2)	
History of TB				
No	40 (18.2)	180 (81.8)	220 (98.7)	0.458
Yes	1 (33.3)	2 (66.7)	3 (1.3)	
Share rooms with others				
No	6 (13)	40 (87)	46 (20.6)	0.294
Yes	35 (19.8)	142 (80.2)	177 (79.4)	
Knowledge on symptoms of TB				
No	1 (16.7)	5 (83.3)	6 (2.7)	1
Yes	40 (18.4)	177 (81.6)	217 (97.3)	

The most relevant results were collected through questions on elephant husbandry. More than 95% of the mahouts spend ≥ 5 hours per day taking care of the elephant(s) and 89% bathed their elephant(s) daily (Chapter 2 Annex, Table S3). The majority of the mahouts (93%) had close interaction with elephants and 88% of mahouts feed the elephants with their hands

(Chapter 2 Annex, Table S4). More than 90% of the mahouts cleaned the elephant's enclosure daily but only 19.7% used disinfectant for cleaning. About half of the mahouts used a pit to dispose elephant waste, nearly 43% disposed of waste in open places and the remaining ones utilized waste by composting and making biogas (Chapter 2 Annex, Table S5). Only one fourth of mahouts (26.5%) used personal protective equipment (PPE), mostly purpose-use shoes followed by gloves, clothes, and masks and very few used aprons (Chapter 2 Annex, Table S6). Most of the mahout had heard about TB in elephants and 20% of mahout had diagnosed any elephants with TB (Chapter 2 Annex, Table S7). When elephants get sick, about 78% of mahout visit veterinary care for elephants (Chapter 2 Annex, Table S8).

Identified risk factors included elephant ownership (antibody positivity higher in government owned elephants; $P=0.002$), hand-feeding the elephant (as opposed to using a bucket or the floor; $P=0.006$), not having special PPE at work ($P=0.007$) and disposing of elephant waste in pits (as opposed to other means; $P=0.009$) (Table 2.3 and Figure 2.1). Close interaction with the elephants, bathing elephants and participating in elephant trunk washes were not identified as risks (Table 2.3).



Figure 2.1 Captive Elephants and its relations with Mahouts.

Table 2.3 Association between elephant husbandry by mahout and TB in elephants (Source: Interviews carried out in 2019).

Characteristics	Mahouts who handle TB +Ve elephants			P-value
	Yes	No	Total	
	(n=41,18%)	(n=182, 82%)	(N=223)	
Type of elephant				
Government/NTNC	34(24.8)	103(75.2)	137(61.4%)	0.002
Private	7(8.1)	79(91.9%)	86(38.6%)	
Number of Elephants				
One	37 (19.7)	151 (80.3)	188 (84.3)	0.247
More than one	4 (11.4)	31 (88.6)	35 (15.7)	
Bathing of elephant				
Daily	38 (19.2)	160 (80.8)	198 (88.8)	0.584
Not Daily	3 (12)	22 (88)	25 (11.2)	
Duration of taking care of elephant				
<5 years	14 (15.9)	74 (84.1)	88 (39.5)	0.441
>5 years	27 (20)	108 (80)	135 (60.5)	
Feeding of elephant				
By using your hand	41 (20.8)	156 (79.2)	197 (88.3)	0.006
Using Bucket/floor	0 (0)	26 (100)	26 (11.7)	
Close Interaction with elephants				
Yes	39 (18.8)	169 (81.2)	208 (93.3)	1
No	2 (13.3)	13 (86.7)	15 (6.7)	
Cleaning of elephant enclosures				
Daily	38 (18.1)	172 (81.9)	210 (94.2)	0.711
Not Daily	3 (23.1)	10 (76.9)	13 (5.8)	
Cleaning of elephants' houses				
Clean floor with water	0 (0)	9 (100)	9 (4)	0.217
Remove dung	41 (19.2)	173 (80.8)	214 (96)	
Use disinfectant to clean				
No	37 (20.7)	142 (79.3)	179 (80.3)	0.076
Yes	4 (9.1)	40 (90.9)	44 (19.7)	
Have any special protective equipment (example: shoes, masks, gloves)				
No	37 (22.6)	127 (77.4)	164 (73.5)	0.007
Yes	4 (6.8)	55 (93.2)	59 (26.5)	
Dispose of elephant waste				
Throw in pit	30 (24.6)	92 (75.4)	122 (54.7)	0.009
Open Place/Composting/Bio gas	11 (10.9)	90 (89.1)	101 (45.3)	
Participate in elephant trunk washes				
No	1 (10)	9 (90)	10 (4.5)	0.693
Yes	40 (18.8)	173 (81.2)	213 (95.5)	
Receive regular health checkup/ veterinary care				
No	2 (20)	8 (80)	10 (4.5)	1
Yes	39 (18.3)	174 (81.7)	213 (95.5)	
Heard about TB in Elephants				
No	5 (15.2)	28 (84.8)	33 (14.8)	0.603
Yes	36 (18.9)	154 (81.1)	190 (85.2)	

Contact with other animals and inter-species contacts also revealed interesting risk factors (Table 2.4). Eighteen percent of the mahouts responded that wild animals lived as a pet in or near their dwelling, 31% responded they had eaten food after an animal had touched or damaged it, and 40% responded that they consume unpasteurized dairy products from their

animals. Almost two thirds (59%) responded that animals enter the dwelling where they live and 13.5% collected dead animals to eat or share (Chapter 2 Annex, Table S9). About 24% responded having been scratched or bitten by an animal (Chapter 2 Annex, Table S10). Elephants cared for by mahouts with livestock present in their household or nearby their household were at higher risk for TB antibody seropositivity ($P<0.001$). Also, elephants cared for by mahouts reporting having found a dead animal and collecting it to eat or share ($P=0.005$), or by mahouts reporting having been scratched or bitten by an animal ($P=0.026$), were at a higher risk. Most of the captive elephants (67.7%) had interactions with wild elephants. 2.5% of the captive elephants had an interaction with the wild ones daily, 14.6% twice a month, 35% biannually, and 18% sporadically. The places for interaction were *Hattisar*/elephant breeding center (35.8%), hotel/safari (33.8%), and jungle (30.5%). Elephants belonging to mahouts reporting that wild elephants came to interact with the captive ones were at lower risk of TB antibody seropositivity ($P=0.033$). Figure 2.2 shows the wild and domestic ungulates most frequently in contact with captive elephants.

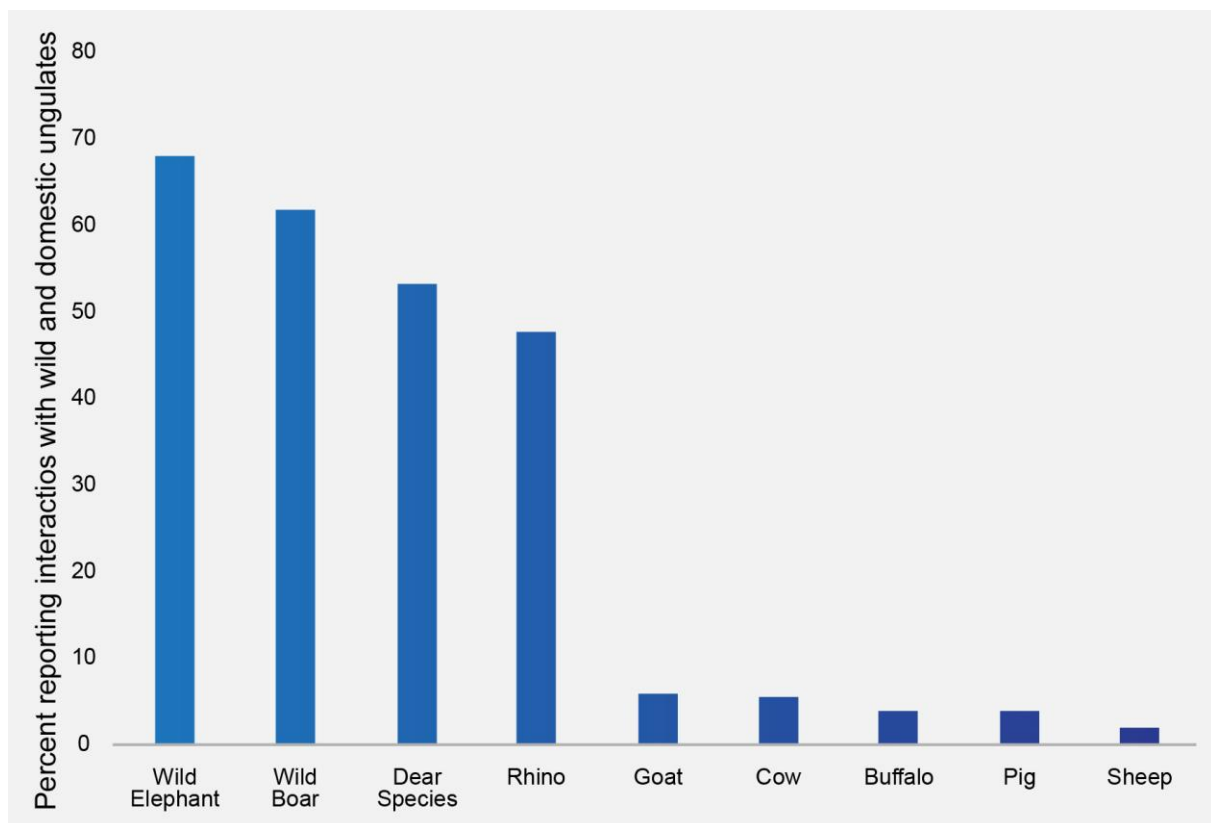


Figure 2.2. Percentage of interviewees responding “yes” to questions on captive Asian elephants’ contacts with wild and domestic ungulates in Chitwan, Nepal

Table 2.4 Association between interaction with other animals and TB among elephants (Source: Interviews carried out in 2019)

Characteristics	Mahouts who handle TB +Ve elephants			P-value
	Yes (n=41,18%)	No (n=182,82%)	Total (N=223)	
Interaction of captive elephants				
Rhino				
No	21 (17.9)	96 (82.1)	117 (52.5)	0.86
Yes	20 (18.9)	86 (81.1)	106 (47.5)	
Tiger				
No	33 (18.5)	145 (81.5)	178 (79.8)	0.906
Yes	8 (17.8)	37 (82.2)	45 (20.2)	
Wild boar				
No	17 (19.8)	69 (80.2)	86 (38.6)	0.673
Yes	24 (17.5)	113 (82.5)	137 (61.4)	
Deer species				
No	16 (15.2)	89 (84.8)	105 (47.1)	0.252
Yes	25 (21.2)	93 (78.8)	118 (52.9)	
Any wild animals				
No	32 (18.5)	141 (81.5)	173 (77.6)	0.936
Yes	9 (18)	41 (82)	50 (22.4)	
Cow				
No	40 (19)	171 (81)	211 (94.6)	0.7
Yes	1 (8.3)	11 (91.7)	12 (5.4)	
Sheep				
No	41 (18.7)	178 (81.3)	219 (98.2)	1
Yes	0 (0)	4 (100)	4 (1.8)	
Buffalo				
No	39 (18.2)	175 (81.8)	214 (96)	0.672
Yes	2 (22.2)	7 (77.8)	9 (4)	
Pig				
No	40 (18.6)	175 (81.4)	215 (96.4)	1
Yes	1 (12.5)	7 (87.5)	8 (3.6)	
Goat				
No	38 (18.1)	172 (81.9)	210 (94.2)	0.711
Yes	3 (23.1)	10 (76.9)	13 (5.8)	
Livestock				
No	40 (18.2)	180 (81.8)	220 (98.7)	0.458
Yes	1 (33.3)	2 (66.7)	3 (1.3)	
Wild elephant come to interact with Captive elephant				
No	19 (26.4)	53 (73.6)	72 (32.3)	0.033
Yes	22 (14.6)	129 (85.4)	151 (67.7)	
Interaction of mahout				
Cow				
No	36 (17.9)	165 (82.1)	201 (90.1)	0.567*
Yes	5 (22.7)	17 (77.3)	22 (9.9)	
Sheep				
No	39 (18.2)	175 (81.8)	214 (96)	0.672
Yes	2 (22.2)	7 (77.8)	9 (4)	
Buffalo				
No	40 (19)	171 (81)	211 (94.6)	0.7
Yes	1 (8.3)	11 (91.7)	12 (5.4)	
Pig				
No	38 (18.3)	170 (81.7)	208 (93.3)	0.742
Yes	3 (20)	12 (80)	15 (6.7)	

Goat				
No	34 (17.1)	165 (82.9)	199 (89.2)	0.165
Yes	7 (29.2)	17 (70.8)	24 (10.8)	
Livestock				
No	39 (18)	178 (82)	217 (97.3)	0.305
Yes	2 (33.3)	4 (66.7)	6 (2.7)	
Livestock animals present in your household or nearby your household				
No	11 (9.8)	101 (90.2)	112 (50.2)	< 0.001
Yes	30 (27)	81 (73)	111 (49.8)	
Mahout have any interaction with wild animal				
No	1 (14.3)	6 (85.7)	7 (3.1)	< 0.001
Yes	40 (18.5)	176 (81.5)	216 (96.9)	
Any wild animals lived as a pet in or near your dwelling				
No	35 (19.2)	147 (80.8)	182 (81.6)	0.492
Yes	6 (14.6)	35 (85.4)	41 (18.4)	
Eaten food after an animal has touched or damaged it				
No	25 (16.2)	129 (83.8)	154 (69.1)	0.215
Yes	16 (23.2)	53 (76.8)	69 (30.9)	
Consume any dairy products from your animals not been pasteurized				
No	20 (14.8)	115 (85.2)	135 (60.5)	0.088
Yes	21 (23.9)	67 (76.1)	88 (39.5)	
Found a dead animal and collected it to eat or share				
No	30 (15.5)	163 (84.5)	193 (86.5)	0.005
Yes	11 (36.7)	19 (63.3)	30 (13.5)	
Scratched or bitten by an animal				
No	26 (15.2)	145 (84.8)	171 (76.7)	0.026
Yes	15 (28.8)	37 (71.2)	52 (23.3)	

Factors significantly associated with the risk of an elephant testing seropositive for TB in the multivariate logistic regression are presented in (Table 2.5) along with the odds ratios and their 95% confidence intervals. These included type of elephants (odds of being TB antibody seropositive 7.74 times higher in government/NTNC owned than in private owned ones (OR=7.74; 95% CI: 2.8-21.44), interaction of wild elephants with captive elephants (protective, OR=0.36; 95% CI: 0.15-0.86), use of PPE at work (protective, OR=0.25; 95% CI: 0.08-0.81), disposing of elephant waste in pits (OR=4.11; 95% CI: 1.7-9.94), having livestock animals present in household (OR =3.94; 95% CI: 1.62-9.57), and reporting to have found a dead animal and collected it to eat and share (OR= 3.29; 95% CI: 1.1-9.8). The Hosmer–Lemeshow test showed a Chi-squared value of 7.25 and p-value of 0.50, showing that the model provided a good fit ($p>0.05$).

Table 2.5 Multivariate logistic analysis of socio-economic, animal husbandry and cross-species interaction factors associated with the risk of captive elephants testing positive for tuberculosis (COR crude odds ratio; AOR adjusted odds ratio) (Source: Interviews carried out in 2019).

Variables	COR (95%CI)	P-value	AOR (95%CI)	P-value
Type of elephant		< 0.001		< 0.001
Private	Ref		Ref	
Government/NTNC	3.73 (1.57, 8.85)		7.74 (2.8, 21.44)	
Wild elephant come to interact with Captive elephant		0.002		0.006
No	Ref		Ref	
Yes	0.48 (0.24, 0.95)		0.36 (0.15, 0.86)	
Have any special protective equipment worn at work		0.021		0.001
No	Ref		Ref	
Yes	0.25 (0.08, 0.73)		0.25 (0.08, 0.81)	
Dispose of elephant waste		0.002		< 0.001
Open Place/Composting/Bio gas	Ref		Ref	
Throw in pit	2.67 (1.26, 5.65)		4.11 (1.7, 9.94)	
Livestock animals present in your household or nearby your household		0.002		0.001
No	Ref		Ref	
Yes	3.4 (1.61, 7.2)		3.94 (1.62, 9.57)	
Found a dead animal and collected it to eat or share		0.033		0.035
No	Ref		Ref	
Yes	3.15 (1.36, 7.28)		3.29 (1.1, 9.8)	
Scratched or bitten by an animal		0.01		0.224
No	Ref		Ref	
Yes	2.26 (1.09, 4.7)		1.77 (0.71, 4.43)	

2.5 Discussion

This study assessed social-demographic aspects, elephant husbandry, and contacts with other animals as risk factors for antibodies against MTC in captive Asian elephants from Chitwan valley. Risks were assessed through detailed interviews of 223 elephant caretakers or mahouts. While no social-demographic factor appeared as a relevant risk for finding seropositivity to antibodies against MTC in the elephants they cared for, we found several risks related to elephant husbandry and to cross-species contacts.

The prevalence of serum antibodies against MTC in the captive elephant population was 20.2%. Serum antibodies reflect present or past exposure to members of the MTC or to cross-reacting organisms. While this is not equivalent to actual infection, we assumed antibody detection as a proxy for MTC exposure (Thomas et al., 2021). The recorded apparent prevalence is similar to others reported for elephants in Nepal (Population, n.d.). There have been cases of elephant mortality in Nepal reflecting the translation of seroprevalence to actual disease (Mandal, 2020; Mikota & Maslow, 2011). Close contacts between humans and

elephants in countries with a high human TB burden may increase the risk of infection in elephants through reverse zoonotic transmission (Michel et al., 2013). In our study, 16 mahouts reported having a history of TB. However, the risk may also be present in the other direction, meaning elephant-to-human or captive to-wild-elephant transmission.

The elephant-to-human TB transmission reported previously has demonstrated that mahouts are inherently at risk of exposure to zoonotic diseases due to their close contact with elephants and other animals (Murphree et al., 2011). They are likely to encounter animals infected with zoonotic pathogens and to engage in high-risk interactions with them (Caron et al., 2015; Thapa et al., 2017). The finding that having livestock in mahouts' households implies a higher risk of their elephants testing TB positive deserves further research, since livestock infections are likely due to *M. bovis* or other MTC members rather than *M. tuberculosis*. The study has also identified that elephants, which are government owned, have higher odds of being TB positive. This could be due to larger numbers of government-owned elephants living together, or other unknown ownership-specific risks.

Regarding the captive-to-wild elephant transmission, most elephants (68%) included in this study had known contacts with wild elephants. The reason may be that domesticated females are temporarily released into the forest during the breeding season to be approached by wild tusked males. This traditional method of elephant breeding is by far the most successful one in Nepal and remains crucial for the survival of the population (Pradhan et al., 2011; Michel et al., 2013). Interestingly, interactions with wild elephants were found to be of low risk, suggesting that wild elephants do not represent a significant TB source for captive ones. However, there is a risk of transmission from captive to wild elephants, e.g., by sharing water or other resources. (Rosen et al., 2018) Hence, captive elephants might represent a bridge host (Caron et al., 2015) potentially enabling transmission between wildlife, domestic animals, and human beings (Thapa et al., 2017b; Rajbhandari et al., 2022)

Our results suggest that certain preventive or biosecurity measures could eventually contribute to reduce the risk of TB in captive elephants. These include avoiding hand feeding the elephants, using PPEs at work, and properly disposing of elephant waste. Hand feeding is common and might be difficult to avoid, although it could be explored if using facemasks and disposable gloves during feed preparation reduces the risks. Clearly, PPE usage by mahouts is insufficient and requires further attention (Ocepek et al., 2005; Zumpe et al., 1980). Regarding waste disposal, it may be that the dung pits, where food remains and other waste also accumulate, attract rhesus macaques (*Macaca mulatta*) and other wildlife. This could eventually represent a (still unexplored) source of infection for the nearby elephants and their caretakers (Zumpe et al., 1980). By contrast, it was relevant not to find risks related to water

such as bathing elephants and participating in elephant trunk washes. Both potential risk activities are possibly not identified as a risk because they take place in a vast majority of cases, the only minor difference being the frequency in the case of bathing (daily for 89%). This means that risks not identified as such in the present survey should not be neglected.

In the Chitwan valley, Nepal, it is likely that TB transmission occurs from humans to elephants and vice versa, and it may also involve other wild or domestic animal species. Therefore, the Government of Nepal has set up an elephant TB control and management action plan (DNPWC, 2011), medical monitoring of people working or living with elephants should thus be implemented. Mahouts need to be aware of the risk of TB acquisition from infected animals and be informed regarding infection control measures. As elephants become infected by humans with active TB, there should be workplace policies (e.g., pre-employment screening and annual chest X-rays) to ensure that elephant handlers are TB-free. Periodic occupational hazards and training programs need to be organized for wildlife employees. They should be provided with PPE such as face masks and disposable gloves and aprons, especially when dealing with suspected elephant TB. A well-coordinated One Health perspective involving both the animal and human health sectors should be undertaken to develop a comprehensive prevention strategy to protect the elephants and their caregivers from TB.

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Chapter 3: Differentially represented proteins in response to infection with *Mycobacterium tuberculosis* identified by quantitative serum proteomics in Asian elephants*

3.1 Abstract

Tuberculosis is a major global concern. Tuberculosis in wildlife is a risk for zoonotic transmission and becoming one of the challenges for conservation globally. In elephants, the number of cases is likely rising. The aim of this study was to identify proteins related to tuberculosis infection in elephants, which could then be used for the development of diagnostic tools and/or vaccines. A serum proteomics approach was used to characterize differentially represented proteins in response to *Mycobacterium tuberculosis* in Asian elephants (*Elephas maximus*). Blood samples were collected from eight elephants, four of which were antibody positive for tuberculosis and four were antibody negative. Proteomics analysis identified 26 significantly dysregulated proteins in response to tuberculosis. Of these, 10 (38%) were identified as immunoglobulin and 16 (62%) as non-immunoglobulin proteins. The results provided new information on the antibody response to mycobacterial infection and biomarkers associated with tuberculosis and protective response to mycobacteria in Asian elephants. Protective mechanisms included defense against infection (Alpha-1-B glycoprotein A1BG, Serpin family A member 1 SERPINA1, Transthyretin TTR), neuroprotection (TTR) and reduced risks of inflammation, infections, and cancer (SERPINA1, Keratin 10 KRT10). Using a translational biotechnology approach, the results provided information for the identification of candidate diagnostic, prognostic and protective anti-gens for monitoring and control of tuberculosis in Asian elephants.

Keywords: Elephant; Tuberculosis; Proteomics; Biomarkers; Vaccine; Diagnostic

* Villar M, Rajbhandari RM, Artigas-Jerónimo S, Contreras M, Sadaula A, Karmacharya D, Alves PC, Gortázar C, de la Fuente J. Differentially Represented Proteins in Response to Infection with *Mycobacterium tuberculosis* Identified by Quantitative Serum Proteomics in Asian Elephants. *Pathogens*. 2022 Sep 3;11(9):1010. doi: 10.3390/pathogens11091010. PMID: 36145440; PMCID: PMC9505326.

3.2 Introduction

Tuberculosis (TB), a disease caused by organisms belonging to the *Mycobacterium tuberculosis* complex (MTBC) remains a major global concern for public health and economy. With an estimated 10 million cases worldwide and 1.6 million deaths in humans in the year 2021, it is the second infectious disease with global impact after COVID-19 (WHO, 2021). Tuberculosis in humans and animals is caused by a group of closely related acid-fast bacilli of the MTBC (Brites & Gagneux, 2015; Malone & Gordon, 2017). The MTBC includes of *M. tuberculosis*, *M. bovis*, *M. bovis* (*bacillus Calmette- Guerin*), *M. africanum*, *M. caprae*, *M. microti*, *M. canettii* and *M. pinnipedii* (Talip et al., 2013). In elephants, TB was probably present more than 2000 years ago (Iyer, 1937) with first case of TB in elephants reported in London Zoo in 1875 (Mikota et al., 2000). The main causative agent for TB in elephants is *M. tuberculosis* (Lewerin et al., 2005) with rare cases of other MBTC organisms such as *M. bovis* (Mikota, 2020). The sporadic cases of TB in captive Asian elephants (*Elaphas maximus*) were reported in the early 20th century but only in the mid-20th century the first case in an African elephant (*Loxodonta africana*) was reported (Mikota, 2020). After the initiation of systematic surveillance in 1998 in the U.S.A., the number of TB cases in elephants is rising (Mikota, 2020).

Nepal is home to Asian elephants that are still used in the timber industry for military and national park security and for tourism (Pradhan et al., 2011; Bale, 2016; Shrestha & Shrestha, 2021; Szydlowski, 2022). At present, there are altogether 208 captive elephants in Nepal out of which 94 government-own elephants are in various parks in the Terai, namely Koshi Tappu Wildlife Reserve, Parsa Wildlife Reserve, Chitwan National Park, Bardia National Park and Suklaphanta Wildlife Reserve (Pradhan et al., 2011). In Nepal, TB caused by different members of the MTBC has been reported in different animal species including *M. tuberculosis* from Asian elephants (Paudel et al., 2014; Thapa et al., 2015, 2016).

Tuberculosis in wildlife is becoming one of the challenges for conservation globally. The disease spillover and transmission on the human-livestock-wildlife interface affects a wide range of species (Ellwanger & Chies, 2021). Close contact between elephants and humans, especially between captive elephants and their mahouts, increases the risk of disease transmission. Furthermore, studies that have been carried out in Nepal have shown that there might be an interspecies transmission of TB among human and animal species with a recent study on bovine TB revealing a high risk of zoonotic bovine TB at the cattle-human interface (Gompo et al., 2020; Miller et al., 2021).

Regarding the diagnosis of TB in elephants, a wide variety of diagnostic measures have been used throughout the years. Culture of the trunk-wash sample was preferred as a diagnostic

tool before more rapid methods became available. Elephant TB can be accurately diagnosed by serum antibody detection (Greenwald et al., 2009; Lyashchenko et al., 2012; Mikota et al., n.d.; Paudel et al., 2018). In the absence of highly sensitive diagnostic assays, a routine medical examination is recommended (Backues & Wiedner, 2019). Standard tests such as VetMAX MTBC qPCR Kit (Goosen et al., 2020) and serological tests using Elephant TB STAT-PAK, DPP VetTB Assay, MAPIA (multi-antigen print immunoassay) (Paudel et al., 2018), and interferon gamma release assay (IGRA) (Songthammanuphap et al., 2020) are used for detection of MTBC in different parts of the world depending on sample accessibility.

The main challenge to further advances in TB diagnosis and control is the identification of biomolecules for developing diagnostic tools and vaccines. Few studies have used proteomics approaches for the characterization of protein profiles in elephantids (Cappellini et al., 2012; Pohlentz et al., 2005; Yazhini et al., 2021). However, the study reported here is the first characterization of elephant serum proteome in response to TB. These results advance our understanding of elephant immune response to *M. tuberculosis* and provide potential biomarkers for diagnosis and control of TB in this endangered species.

3.3 Material and Methods

3.3.1 Serum samples from TB-positive (TB+) and TB-negative (TB-) elephants

The study was conducted in Chitwan National Park established in 1973 in Nepal. It covers an area of 952.63 km² and is in the subtropical Inner Terai lowlands of south-central Nepal in the districts of Nawalpur, Parsa, Chitwan and Makwanpur. In altitude it ranges from about 100 m in the river valleys to 815 m in the Churia Hills (Shrestha & Shrestha, 2021). The National Park is home to 111 captive elephants. Of them, serum samples were collected from 8 adult animals with (E1, E4, E44, E66; N = 4) and without (E9, E43, E70, E71; N = 4) tuberculosis reactive anti-bodies and used for the proteomics analysis. Trained veterinarians collected blood samples from an ear vein into a serum gel tube, using the Vacutainer (R) blood collection system with butterfly needle. Serum samples were aliquoted and stored at -20 °C until analysis. Ethical approval for the study was obtained from Nepal Ethical Review Board, Nepal Health Research Council (IRC number 312/2018).

3.3.2 Diagnostic of tuberculosis

Sera were subjected to antibody testing using an animal-side rapid test for TB (DPP VetTB Assay, Chembio Diagnostic Systems Inc., Medford, USA) performed as recommended by the manufacturer. Results showed the presence of TB reactive or non-reactive antibodies. Two samples from TB+ animals E1 and E44 were further confirmed as infected with *M. tuberculosis* by cell culture.

3.3.3 Serum proteomics

The serum proteomics methodology has been previously reported (Villar et al., 2021) and adapted to this study. Protein concentration in serum samples was determined using the BCA Protein Assay with BSA (Sigma-Aldrich, St. Louis, MO, USA) as standard. Protein serum samples (150 µg per sample) were trypsin digested using the FASP Protein Digestion Kit (Expedeon Ltd., UK) and sequencing grade trypsin (Promega, Madison, WI, USA) following manufacturer's recommendations. The resulting tryptic peptides were desalted onto OMIX Pipette tips C18 (Agilent Technologies, Santa Clara, CA, USA), dried down and stored at -20 °C until mass spectrometry analysis. The desalted protein digests were resuspended in 2% acetonitrile, 5% acetic acid in water and analyzed by reverse phase liquid chromatography coupled to mass spectrometry (RP-LC-MS/MS) using an EkspertTMnanoLC 415 system coupled online with a 6600 TripleTOF® mass spectrometer (AB SCIEX; Framingham, US) through Information-Dependent Acquisition (IDA) followed by Sequential Windowed data independent Acquisition of the Total High-resolution Mass Spectra (SWATH-MS). The peptides were concentrated in a 0.1×20 mm C18 RP precolumn (Thermo Scientific) with a flow rate of 2µl/min during 10 min in solvent A. Then, peptides were separated in a 0.075×250 mm C18 RP column (New Objective, Woburn, MA, USA) with a flow rate of 300 nl/min. Elution were done in a 120-min gradient from 5% B to 30% B followed by 15-min gradient from 30% B to 60% B (Solvent A: 0,1% formic acid in water, solvent B: 0,1% formic acid in acetonitrile) and directly injected into the mass spectrometer for analysis. For IDA experiments, the mass spectrometer was set to scanning full spectra from 350 m/z to 1400 m/z (250 ms accumulation time) followed by up to 50 MS/MS scans (100-1500 m/z). Candidate ions with a charge state between +2 and +5 and counts per second above a minimum threshold of 100 were isolated for fragmentation. One MS/MS spectra was collected for 100 ms, before add-ing those precursor ions to the exclusion list for 15s (mass spectrometer operated by Analyst® TF 1.6, ABSciex®). Dynamic background subtraction was turned off. Data were acquired in high sensitivity mode with rolling collision energy on and a collision energy spread of 5. An equal amount of the four samples for each control and infected groups joined together as a representative mixed sample, which were used for the generation of the reference spectral ion library as part of SWATH-MS analysis. A total amount of 4 µg was injected by triplicate. For SWATH quantitative analysis, 8 independent samples (2 technical replicates from each of the 4 biological replicates for TB- and TB+ groups) (6 µg each) were subjected to the cyclic data independent acquisition (DIA) of mass spectra using the SWATH variable windows calculator (V 1.0, AB SCIEX) and the SWATH acquisition method editor (AB SCIEX), like established methods (Yazhini et al., 2021). A set of 50 overlapping windows was constructed (containing 1 m/z for the window overlap), covering the precursor mass range of 400–1250 m/z. For these

experiments, a 50 ms survey scan (350–1400 m/z) was acquired at the beginning of each cycle, and SWATH-MS/MS spectra were collected from 100–1500 m/z for 70 ms at high sensitivity mode, resulting in a cycle time of 3.6 s. Collision energy for each window was determined according to the calculation for a charge +2 ion-centered upon the window with a collision energy spread of 15. To create a spectral library of all the detectable peptides in the samples, the IDA MS raw files were combined and subjected to database searches in unison using ProteinPilot software v. 5.0.1 (AB SCIEX) with the Paragon algorithm. Spectra identification was performed by searching against the Uniprot Elephantidae database (28,137 entries in April 2022), with the following parameters: iodoacetamide cysteine alkylation, trypsin digestion, identification focus on biological modification and thorough ID as search effort. The detected protein threshold was set at 0.05. To assess the quality of identifications, an independent False Discovery Rate (FDR) analysis with the target-decoy approach provided by Protein Pilot™ was performed. Positive identifications were considered when identified proteins reached a 1% global FDR. The mass spectrometry proteomics data have been deposited in the Proteome change Consortium via the PRIDE partner repository with the dataset identifier PXD033830 and 10.6019/PXD033830.

3.3.4 Quality control of proteomics data

As previously validated (Villar et al., 2021), quality of proteomics data was controlled at multiple levels. First, a Zebrafish intestine digest was used for the evaluation of instrument performance. Buffer A samples were run as blanks every two injections to prevent carry-over. Two technical replicates were injected for each sample.

3.3.5 Data analysis

As previously reported for SWATH processing (Villar et al., 2021), up to 10 peptides with 7 transitions per protein were automatically selected by the SWATH Acquisition MicroApp 2.0 in the PeakView 2.2 software with the following parameters: 15 ppm ion library tolerance, 5 min XIC extraction window, 0.01 Da XIC width, and considering only peptides with at least 99% confidence and excluding those which were shared or contained modifications. However, to ensure reliable quantitation, only proteins with 3 or more peptides available for quantitation were selected for XIC peak area extraction and exported for analysis in the MarkerView 1.3 software (AB SCIEX). Global normalization according to the Total Area Sums of all detected proteins in the samples was conducted (Data S1). The Student's t-test ($p < 0.05$) was used to perform two-sample comparisons between the averaged area sums of all the transitions derived for each protein across the eight replicate runs for each group under comparison, to identify proteins that were significantly differentially represented between TB- and TB+ elephants (Data S1). Protein log fold-change relative intensity was compared between TB-

and TB+ elephants by Welch's unpaired t-test ($p < 0.05$; <https://www.graphpad.com/quickcalcs/ttest1/?Format=C>) (Shen et al., 2020; Villar et al., 2021). Data were separately analyzed for overrepresented and underrepresented proteins. For differentially represented immunoglobulin (Ig) proteins (Igs), an analytical algorithm was developed using Protein BLAST sequence alignment against non-redundant protein database (nr) using compositional matrix adjustment (Rédei, 2008) and Paratome (Kunik et al., 2012)) and used for the identification of Mycobacterium antigen binding regions and candidate biomarkers of protective response to mycobacteria (Data S2). Phylogenetic analysis was conducted for *M. tuberculosis* type I site-specific deoxyribonuclease, HsdR family (sequence ID: SGD45129.1, length: 1040 amino acids) using BLAST Tree Viewer, Fast Minimum Evolution, Max Seq Difference = 0.85, Distance Grishin Protein. For non-Ig proteins, Gene Ontology (GO) annotations for molecular function (MF), biological process (BP), and subcellular localization (CC) were done in human homolog proteins using Uniprot (<https://www.uniprot.org>) and the most significant pathways were identified by Reactome (<https://reactome.org/PathwayBrowser/>; (Fabregat et al., 2018; Jassal et al., 2020) Additional files 3 and 4). COBAL (T) (<https://www.ncbi.nlm.nih.gov/tools/cobalt/cobalt.cgi?CMD=Web>) was used for protein sequence alignment between identified mycobacterial proteins with predicted antigen binding regions to Ig proteins and *M. tuberculosis* P9WNK5|ESXB_MYCTU ESAT-6-like protein EsxB identified as highly reactive to antibodies in infected elephants (Lyashchenko et al., 2018).

3.4 Results

3.4.1 Identification of differentially represented elephant serum proteins in response to TB by quantitative serum proteomics.

In our study, sera were collected from 8 Asian elephants, four of which were seronegative for TB, and four of which were seropositive for TB and used individually for SWATH-MS proteomics analysis for the identification and quantitation of differentially represented proteins (Figure 3.1A). Proteomics analysis identified 26 significantly dysregulated proteins in TB+ elephants when compared to TB- animals (Figure 3.1B, Data S1). Of them, 10 (38%) were identified as Ig proteins (9 overrepresented and one underrepresented in TB+ elephants) and 16 (62%) as non-Ig proteins (12 overrepresented and 4 underrepresented in TB+ elephants) (Figure 3.1B). Data analysis was then focused on overrepresented and underrepresented Ig and non-Ig proteins for the identification of candidate protective or disease associated biomarkers (Figures 3.1A and 3.1C).

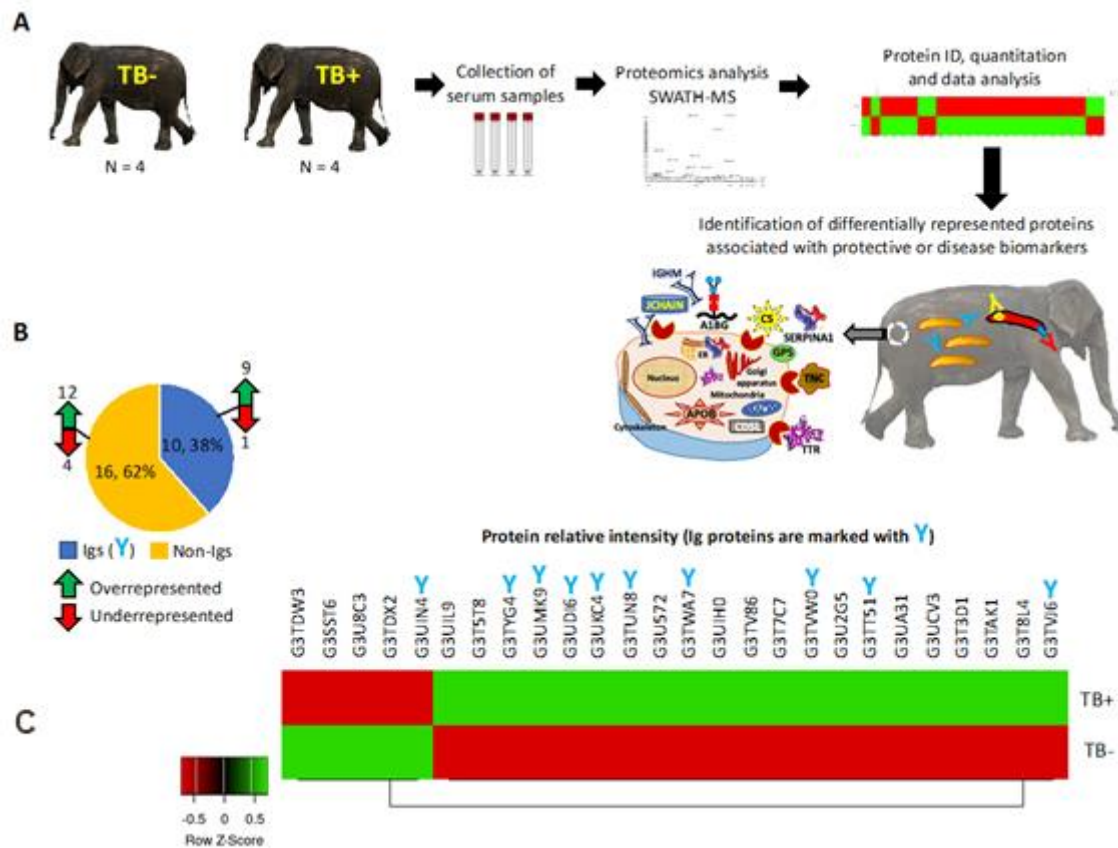


Figure 3.1. Identification of serum differentially represented proteins in response to TB in Asian elephants. (A) In our study, sera were collected from TB- and TB+ elephants. Sera were subjected to SWATH-MS quantitative proteomics to characterize serum differentially protein profiles. The proteomics results were then focused on overrepresented and underrepresented Ig and non-Ig proteins and translated into the identification candidate protective or disease-associated biomarkers. (B) Number and heatmap (Z-scored relative intensity original value) of overrepresented and underrepresented Ig and non-Ig proteins in TB+ elephants when compared to TB- animals.

3.4.2 Characterization of Ig protein profiles and correlation with mycobacterial infection or host protective capacity

Changes in Ig protein mass spectra relative intensity in individual samples were compared between TB- and TB+ elephants. The results were significantly different between groups for both overrepresented and underrepresented proteins thus further supporting proteomics results ($p < 0.05$; Figure. 3.2).

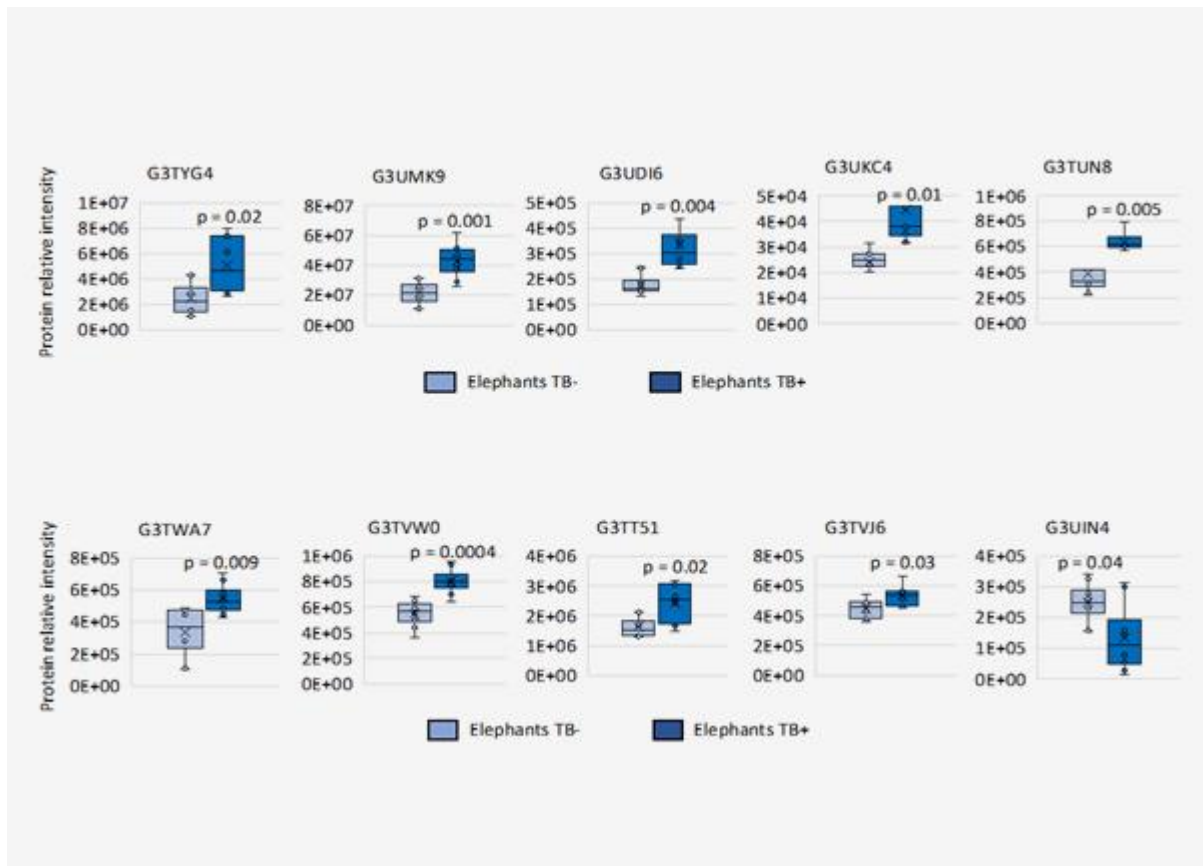


Figure 3.2. Serum Ig proteins differentially represented in TB- and TB+ elephants. Changes in Ig protein mass spectra relative intensity in individual samples of all TB- and TB+ animals. Relative intensity was compared between TB- and TB+ cohorts by Welch's unpaired t-test ($p < 0.05$; $n = 4$ biological replicates).

An analytical workflow was developed to characterize the Ig-like domain containing proteins identified as significantly dysregulated in TB+ elephants when compared to TB- animals (Figure 3.3, Data S2). The use of Paratome algorithm (<http://www.ofranlab.org>) allows the identification of antigen binding regions in identified Ig light and heavy chain variable regions (Shen et al., 2020).

The predictive models for Igs overrepresented in TB+ elephants showed an association with exposure to different *Mycobacterium spp.* (Figure 3.3). As demonstrated by an example analysis using the identified *M. tuberculosis* type I site-specific deoxyribonuclease HsdR family protein (SGD45129.1; Data S2), these mycobacteria are phylogenetically closely related for these proteins (Figure 3.3). Only one Ig protein was identified as underrepresented in TB+ elephants and thus overrepresented in TB- animals (G3UIN4; Data S2) with predicted *Mycobacterium sp.* reactive epitopes associated with biomarkers of protective response to mycobacteria (Figure 3.3).

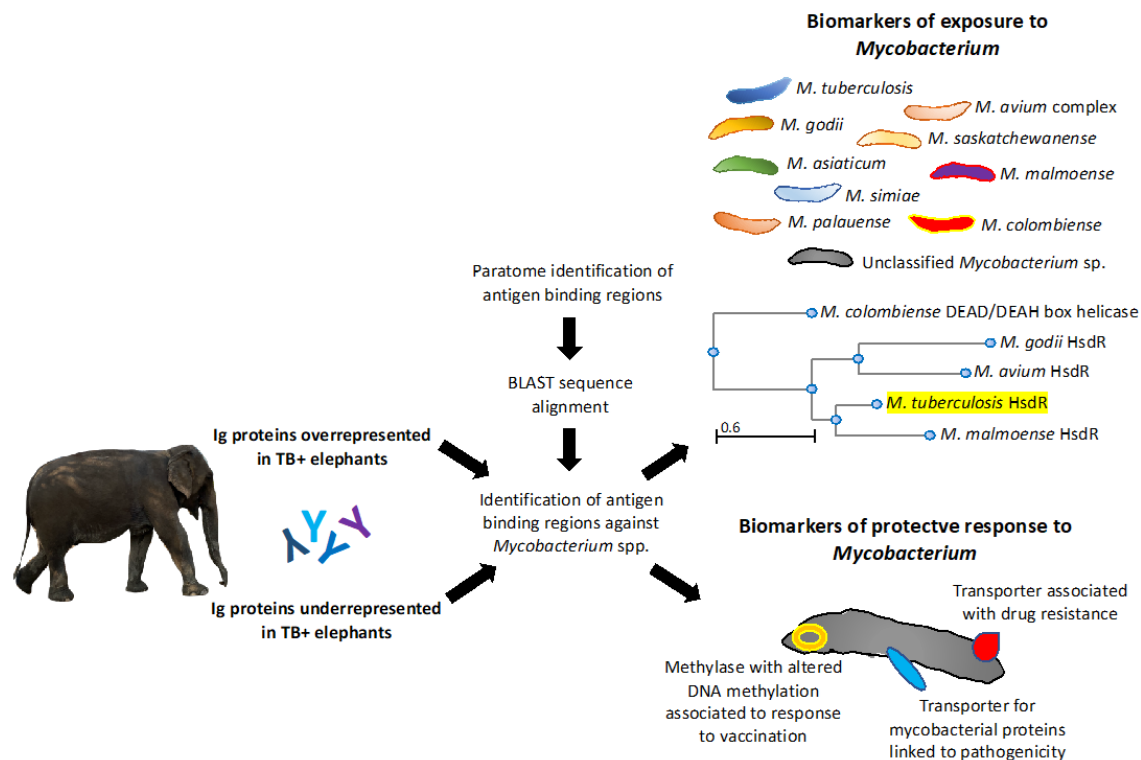


Figure 3.3: Predicted biomarkers associated with differentially represented Ig proteins. Analytical workflow based on Paratome and protein BLAST sequence alignment identified biomarkers associated with exposure to *Mycobacterium* spp. or protective response to mycobacteria in Ig proteins overrepresented and underrepresented in TB+ elephants, respectively. Phylogenetic analysis was conducted for *M. tuberculosis* type I site-specific deoxyribonuclease, HsdR family (SGD45129.1) input sequence highlighted in yellow using BLAST Tree Viewer, Fast Minimum Evolution, Max Seq Difference = 0.85, Distance Grishin Protein

3.4.3 Characterization of non-Ig protein profiles and correlation with TB

The analysis of non-Ig protein profiles in response to TB in elephants resulted in the identification of proteins in multiple biological processes and pathways associated with pathogenic and protective mechanisms (Figure 3.4; Chapter 3 Annex, Additional files 3 and 4). Most of the dysregulated proteins were secreted but some were also located in the cytoplasm, endoplasmic reticulum (ER) and cytoskeleton of multiple cell types (Figure 3.4; Data S3).

Dysregulated proteins in TB+ elephants were mostly associated with higher risks of disorders/diseases such as glycogen storage disease type Ia (Biotinidase, BTD), venous thrombosis and hypercoagulability (Joining chain of multimeric IgA and IgM, JCHAIN; Immunoglobulin heavy constant mu, IGHM; Christmas factor IX, F9; Glycoprotein V platelet, GP5; Complement C5, C5), postprandial hypertriglyceridemia, heart disease, cardiovascular events and mortality (Apolipoproteins C-III, APOC3; D, APOD; B, APOB), solid malignancies (Tenascin C, TNC), pathogenic bacteria induced pneumonia (CD5 molecule like, CDL5), and

inflammation and atherosclerosis (Glycosyl-phosphatidylinositol-specific phospholipase D, GPLD1) (Figure 3.4; Chapter 3 Annex, Additional files 3 and 4).

However, the non-Ig proteins identified as overrepresented in TB- elephants and thus underrepresented in TB+ animals were associated with possible protective mechanisms such as defense against infection and reduce risk of inflammation (Figure 3.4; Chapter 3 Annex, Additional files 3 and 4).

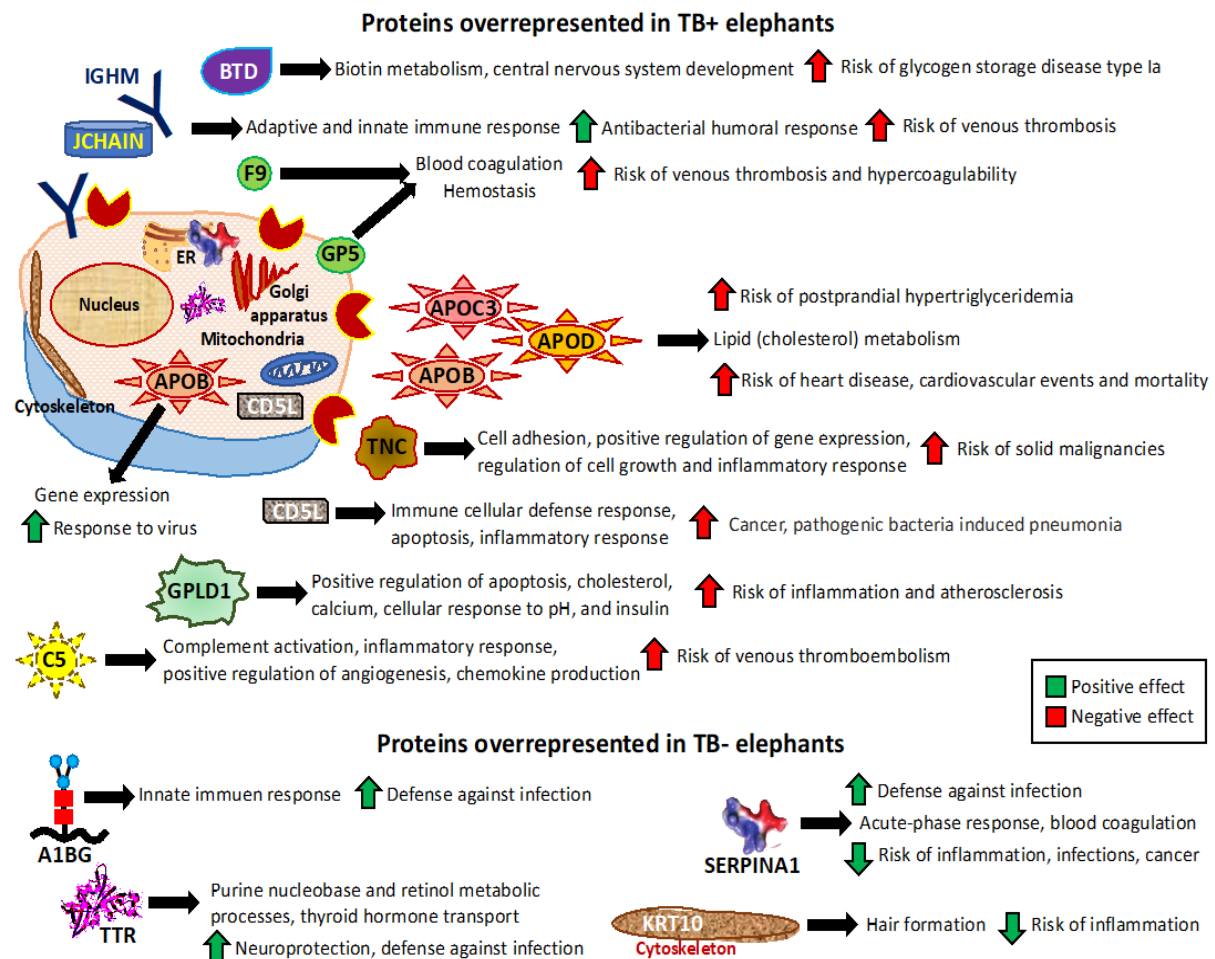


Figure 3.4. Predicted correlates with protective or disease-associated pathways in non-Ig differentially represented proteins. Analytical workflow based on Uniprot GO annotations and Reactome identified biomarkers of protective or disease-associated pathways in response to TB in elephants.

3.5 Implications for translational biotechnology

The results presented here provided information for the identification of candidate diagnostic, prognostic and protective antigens for TB in Asian elephants.

Candidate diagnostic antigens. Mycobacterial proteins reactive to Ig proteins in TB+ elephants may constitute candidate diagnostic antigens. For example, the highly represented Ig protein, G3UDI6 (TB+/TB- ratio = 1.9; Chapter 3 Annex, Additional files 1 and 2), reacted to

Mycobacterium sp. DUF3298 domain-containing protein (TAM68061.1), FkbM family methyltransferase (WP_020822873.1) and hypothetical proteins OEM_30260 and MOTT12_03025 (AGP64561.1 and ARR78689.1) (Data S2). Therefore, these antigens may be used for the diagnosis of TB in Asian elephants. Furthermore, the combination of identified reactive epitopes may be used for designing a diagnostic chimeric antigen (ISSANYG-GGGGS-WLRYYS DRNWN R-C-KLH) with putative higher capacity to diagnose TB+ elephants (Figure 3.5). The proposed candidate diagnostic chimeric antigen protein BLAST sequence alignment against mycobacteria nr database showed 31.6 to 78.9% sequence identity to environmental mycobacteria (Figure 3.5). Mycobacteria with highest sequence identity in the *M. avium* complex also contain pathogenic bacteria found in elephants (Lyashchenko et al., 2018). These results support the possibility of using this chimeric antigen combined with other tests for the diagnosis of TB in elephants.

Candidate prognostic antigens. Identified non-Ig proteins in Asian elephants may constitute TB disease prognostic antigens. For example, TNC (TB+/TB- ratio = 2.7) to KRT10 (TB+/TB- ratio = 0.8) serum protein levels (TNC/KRT10 ratio = 0.7) (Data S1) may be used to evaluate the risks of developing solid malignancies in infected animals (Figure 3.5). With results for individual animals included in this study (Data S1), the TNC/KRT10 ratio was lower in TB- (0.1-0.3) than in TB+ elephants (0.5-1.1) with animal No. 66 showing a predicted high risk for developing solid malignancies (proposed threshold at TNC/KRT10 ratio = 1) (Figure 3.5).

Candidate protective antigens. The identified underrepresented Ig protein in TB+ elephants and thus overrepresented in TB- animals (G3UIN4; Data S2) reacted to *Mycobacterium sp.* epitopes with predicted protective capacity (Figure 3.3). Therefore, the identification and combination of reactive epitopes in mycobacterial RND family transporter membrane protein and protein translocase subunit SecD (Data S2) may be used to design a new candidate protective chimeric antigen with predicted reactive immunological quantum or protective epitopes (QSLFDATDKID-FDSS-KNDEFFYL-GGGGS-LIYLASK-C-KLH) for TB control in Asian elephants (Figure 3.5).

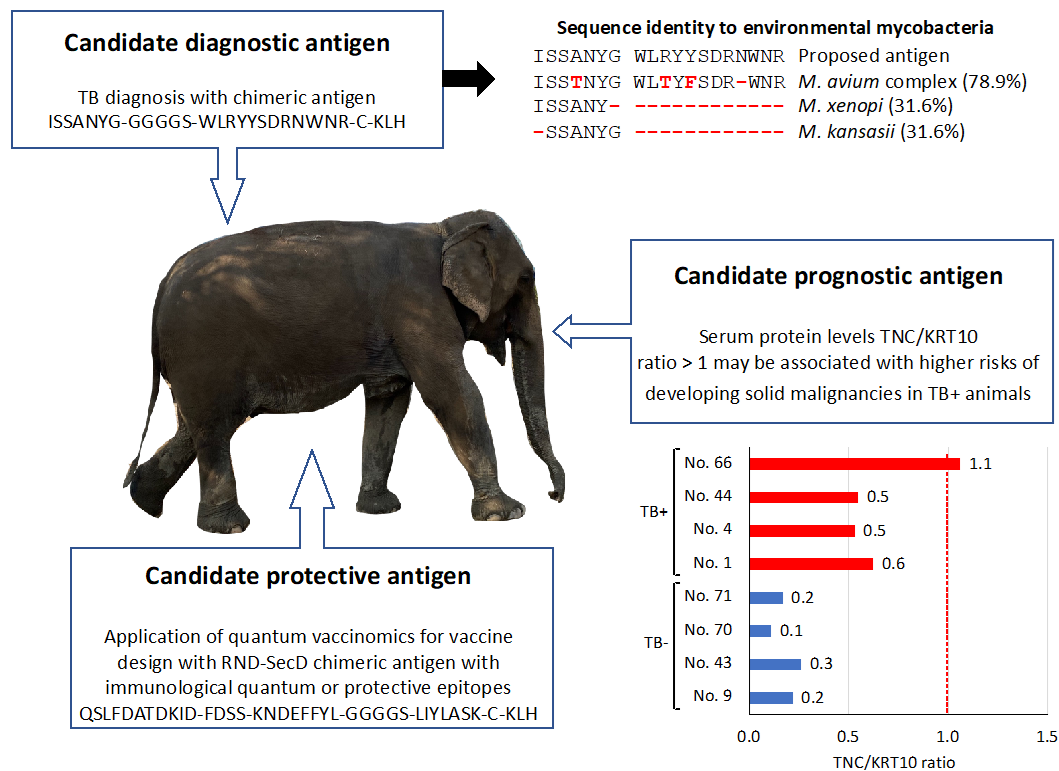


Figure 3.5. Translational biotechnology approaches applied to the differentially represented serum proteins identified in TB+ and TB- Asian elephants. For candidate diagnostic antigen, sequence alignment was conducted against environmental mycobacteria using BLAST (percent sequence identity with non-identical amino acids in red are shown). For candidate prognostic antigens, results are shown for TNC/KRT10 ratio (average of two technical replicates) in individual animals included in this study (Additional file 1). Elephant No. 66 showed a predicted high risk for developing solid malignancies (red line for proposed threshold at TNC/KRT10 ratio = 1). In candidate diagnostic and protective antigens, GGGGS and C-KLH are link peptide sequences not derived from mycobacterial proteins. The results of the study provided information for the design of candidate diagnostic, prognostic, and vaccine antigens.

3.6 Discussion

The Igs overrepresented in TB+ elephants were associated with exposure to different *Mycobacterium* spp. including *M. tuberculosis* (Ig reactive to HsdR and DinB family protein; MBZ4317686.1), *Mycobacterium intracellulare* or *Mycobacterium avium* complex (FkbM family methyltransferase; WP_020822873.1), *Mycobacterium asiaticum* (DUF2617 family protein; WP_065128554.1) and *Mycobacterium colombiense* (hypothetical protein; WP_076051897.1) (Data S2) that have been previously identified in elephants (Ong et al., 2013; Yong et al., 2011; Zachariah et al., 2017).

Other *Mycobacterium* proteins such as ESAT-6-like protein EsxB (CFP10/ESAT-6) reported as highly reactive to antibodies in infected elephants (Lyashchenko et al., 2018) were not identified as predicted antigen binding regions in our study. Nevertheless, cross-reactive epitopes between CFP10/ESAT-6 and identified mycobacterial proteins with predicted antigen

binding regions to Ig proteins may be present (Data S5). These animals are exposed to MTBC and environmental mycobacteria such as *M. simiae* and *M. avium* complex identified here and therefore antibodies induced by bacterial cross-reactive anti-gens may be present (Primm et al., 2004).

For the only identified Ig protein in TB+ overrepresented in TB- animals (G3UIN4; Data S2), predicted *Mycobacterium spp.* reactive epitopes were associated with biomarkers of protective response to mycobacteria associated with (a) resistance-nodulation-division (RND) family transporter (WP_067937080.1) membrane protein associated with drug resistance (Nikaido, 2018; Rossi et al., 2006), (b) restriction endonuclease/methylase (TSD87553.1) involved in anti-mycobacterial activity in response to vaccination that correlates with altered DNA methylation (Verma et al., 2017), and protein translocase subunit Sec D (MBV9350256.1), a transmembrane transport cell inner membrane protein involved in secretion of mycobacterial proteins linked to pathogenicity (Lycklama & Driessen, 2012; Majlessi et al., 2015) (Data S2).

These results provided new information on the antibody response to mycobacterial infection. Asian elephants. While TB+ elephants showed Ig proteins cross-reactive to various *Mycobacterium spp.*, the results suggested that TB- elephants developed a protective antibody response against mycobacterial proteins involved in disease-associated mechanisms.

Dysregulated non-Ig proteins in TB+ elephants were mostly associated with higher risks of disorders/diseases (Aran et al., 2018; Fatima et al., 2017; Gao et al., 2019; O'Brien et al., 1999; Wolf et al., 2003). The most represented biological processes and pathways were related to cardiovascular disorders/diseases with 9 out of 12 of the overrepresented proteins in TB+ elephants Figure 3.4). Cardiac disorders are typical in TB (Huaman et al., 2015). Elevated levels of vitamin K-dependent protein F9 have been associated with an increased risk of venous thrombosis (Chandler, 2019). The GP5-mediated adhesion of platelets to injured vascular surfaces in the arterial circulation is a critical initiating event in hemostasis and Apolipoproteins (APOs) may increase the risk of post-prandial hypertriglyceridemia associated with cardiovascular disease (Guan et al., 2021). Elevated plasma concentration of complement factor C5, which stimulates the locomotion of polymorphonuclear leukocytes toward sites of inflammation is associated with risk of venous thromboembolism (Skjeflo et al., 2021). Although the association between cardiovascular disorders and TB has not been documented in elephants, as in other species, these results suggest a higher risk for these disorders in TB+ Asian elephants (Szaluś-Jordanow et al., 2016; Basham et al., 2020; Huaman et al., 2020; López-López et al., 2021;).

Possible protective mechanisms associated with non-Ig proteins overrepresented in TB-elephants included defense against infection (Alpha-1-B glycoprotein A1BG, Serpin family A member 1 SERPINA1, Transthyretin TTR), neuroprotection (TTR) and reduced risks inflammationn infections, and cancer (SERPINA1, Keratin 10 KRT10) (Reichelt et al., 2004; Mattii et al., 2018; Hsiao et al., 2019; Liz et al., 2020; Miravittles et al., 2022).

The application of translational biotechnology approaches would advance the control of infectious diseases as described in a previous study on tick-borne disease (de la Fuente, 2021). A candidate di-agnostic chimeric antigen was designed based on mycobacterial proteins reactive to Ig proteins in TB+ elephants while candidate prognostic antigens were proposed using identified non-Ig pproteinsin Asian elephants (Figure 3.5). However, infection with other pathogens may also affect serum levels of these proteins and thus the proposed candidate diagnostic antigen requires additional experiments for validation in TB versus other common infections in elephants. Although elephants appear to show a genetic-based lower than expected incidence of cancer when compared to humans (Abegglen et al., 2015; Seluanov et al., 2018), the association of solid malignancies with TB may constitute a risk factor to be evaluated using the prognostic antigens proposed here. The application of quantum vaccinomics approach (Contreras et al., 2022; de la Fuente & Contreras, 2022) guided the design of a candidate protective antigen for vaccination against TB in Asian elephants (Figure 3.5). Based on the challenge that represents a better understanding of the protective mechanisms against TB (Davenne & McShane, 2016), the proposed candidate protective chimeric antigen is only speculative and requires validation in vaccination studies.

In conclusion, the results of the Asian elephant serum proteome in response to TB advanced our understanding of elephant immune response to *M. tuberculosis*. Future studies should further explore the design, production and essay of identified candidate diagnostic, prognostic and protective antigens for TB monitoring and control in Asian elephants.

Supplementary Material: The following supporting information can be downloaded at: www.mdpi.com/xxx/s1, Data S1. Serum proteomics analysis; Data S2. Analysis of immunoglobulin proteins differentially represented in TB+ elephants when compared to TB-animals; Data S3. Gene Ontology (GO) annotations for non-immunoglobulin proteins; Data S4. Most significant pathways identified by Reactome analysis of non-immunoglobulin proteins; Data S5. Protein sequence alignment between *M. tuberculosis* P9WNK5[ESXB_MYCTU ESAT-6-like protein EsxB and identified mycobacterial proteins with predicted antigen binding regions to immunoglobulin proteins.

Data Availability Statement: The mass spectrometry proteomics data have been deposited in the ProteomeXchange Consortium via the PRIDE partner repository with the dataset identifier PXD033830 and 10.6019/PXD033830.

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Chapter 4: Phylogenomic analysis supports *Mycobacterium tuberculosis* transmission between humans and elephants*

4.1 Abstract

Tuberculosis is an infectious disease caused by a group of acid-fast bacilli known as *Mycobacterium tuberculosis* complex (MTC), which has a major impact on humans. Transmission of MTC across the human-animal interface has been demonstrated by several studies. However, the reverse zoonotic transmission from humans to animals (zooanthroponosis) has often been neglected. In this study, we used Nanopore MinION and Illumina MiSeq approaches to sequence the whole genome of *M. tuberculosis* strains isolated from two deceased Asian elephants (*Elephas maximus*) and one human in Chitwan, Nepal. The evolutionary relationships and drug resistance capacity of these strains were assessed using the whole genome data generated by the stand-alone tool Tb-Profiler. Phylogenomic trees were also constructed using a non-synonymous SNP alignment of 2596 bp, including 94 whole genome sequences representative of the previously described *M. tuberculosis* lineages from elephants worldwide (lineages 1 and 4) and from humans in Nepal (lineages 1, 2 and 3). The new genomes achieved an average coverage of 99.6%, with an average mean depth of 55.67x. These *M. tuberculosis* strains belong to lineage 1 (elephant DG), lineage 2 (elephant PK) and lineage 4 (human), and none of them showed to have drug-resistant variants. The elephant-derived isolates were evolutionarily closely related to human-derived isolates previously described in Nepal, both in lineage 1 and 2, providing additional support for zooanthroponosis or bidirectional transmission between humans and elephants. The human-derived isolate clustered together with other published human isolates from Argentina, Russia and the UK in the lineage 4 clade. This complex multi-pathogen, multi-host system is challenging and highlights the need for a One Health perspective to tuberculosis prevention and control at human-animal interface, particularly in regions where human tuberculosis is highly endemic.

Keywords: *Elephas maximus*; zooanthroponosis; Whole-genome sequencing; One Health.

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Phylogenomic analysis supports *Mycobacterium tuberculosis* transmission between humans and elephants. *Frontiers in Veterinary Science*, 10

4.2 Introduction

Tuberculosis (TB) is a significant global burden and is widely reported to be a major public health and economic problem, costing the world \$617 billion between 2000 and 2015 and projected to cost \$1 trillion between 2015 and 2030 (Burki, 2018). It is the second leading cause of death after COVID-19, with an estimated 10 million cases worldwide and 1.5 million deaths in 2020 (WHO, 2021). Drug-resistant TB is also a major threat to global disease control (Chiang & Starke, 2018). It is a major contributor to global antimicrobial resistance (WHO, 2022). In 2021, 450,000 cases of rifampicin-resistant and multidrug-resistant (MDR/MDR-TB) TB were forecasted worldwide, of which 191,000 died (Wandwalo, 2020). In Nepal, the number of cases of drug-resistant TB is estimated to be around 1,500 per year (National Tuberculosis Centre, 2019).

Human and animal tuberculosis is caused by a group of closely related acid-fast bacilli known as the *Mycobacterium tuberculosis* complex (MTC) (Brites & Gagneux, 2015; Malone & Gordon, 2017). The MTC includes several species of mycobacteria, namely *M. tuberculosis*, *M. bovis* (Bacillus Calmette-Guerin), *M. africanum*, *M. caprae*, *M. microti*, *M. canettii* and *M. pinnipedii* (Talip et al., 2013). *M. tuberculosis* is the human-adapted variant and is the major cause of human TB (Talip et al., 2013). However, most publications on emerging infectious agents such as MTC often focus on their zoonotic origin, but comparatively fewer reports are published on possible zoonoanthroposis and the human origin of infectious diseases affecting domestic or wild animals (Messenger et al., 2014). Zoonoanthroposis, is an important ongoing debate regarding the transmission of pathogens from humans to animals. Evidence from a global survey showed that humans are capable of transmitting at least 21 bacterial, 12 viral and 7 fungal pathogens to animals (Messenger et al., 2014). Although there is evidence that pathogens can be transmitted from humans to animals, most reported cases involve captive or domestic animals (Miller & Olea-Popelka, 2013). Influenza is one of the best known examples of bidirectional transmission between humans and domestic pigs (Chastagner et al., 2019). The emergence and re-emergence of these pathogens has important implications for human and animal health (Epstein & Price, 2009). This risk appears to be even greater when bidirectional transmission between humans and animals is frequent, as in the case of TB in elephants (Michalak et al., 1998).

Tuberculosis in elephants has been reported worldwide, although most reported cases involve captive elephants with *M. tuberculosis*, the ethological agent of human TB (Angkawanish et al., 2010; Dalovisio et al., 1992; Mikota et al., 2001; Paudel et al., 2014; Payeur et al., 2002; Zachariah et al., 2017). In countries such as Nepal, where there is a high prevalence of active *M. tuberculosis* infection in humans, 117,000 people living with TB (Ministry of Health and

Population, 2020), and a large elephant population (more than 200 captive and 200-250 wild elephants (Shrestha & Shrestha, 2021), human wildlife interactions might be particularly relevant. Elephant-human interactions are particularly high in regions where captive elephants are used for ecotourism and patrolling protected areas, such as Chitwan National Park. This poses a serious risk of zoonanthroponosis and disease to both captive and wild elephant populations, as well as a risk to humans given the possibility of bidirectional transmission (Pradhan et al., 2011; Paudel et al., 2014;). A recent study in four national parks in Nepal showed a high seroprevalence (21.56%) of TB in captive elephants, with most cases detected in Chitwan National Park (Thapa et al., 2021). In addition, some evidence of zoonanthroponosis has been reported from previous studies in Nepal that also isolated *M. tuberculosis* from diseased elephants using multi-locus variable number of tandem repeats (MLVA) (Paudel et al., 2014, 2021), large sequence polymorphism (Paudel et al., 2019) or whole genome data (Paudel et al., 2021). However, the latter study lacks a phylogenetic analysis of the two *M. tuberculosis* genomes (Martinez et al., 2016) and their integration with previously published data (Paudel et al., 2021).

In this study, a whole genome sequencing approach was used to genotype *M. tuberculosis* isolates from a human and two deceased elephants in Chitwan, Nepal. These new data, combined with genomic data from elephants and human isolates available in the literature, allowed a comprehensive evolutionary study of *M. tuberculosis* in Nepal. These results contribute to the understanding of the possible bidirectional transmission of this pathogen at human-elephant interface.

4.3 Methods and Material

4.3.1 Study area

Chitwan District is located in the southwestern part of Bagmati Province in Nepal. It covers an area of 2,238.39 km² and had a human population of 719,859 in 2022. Nepal's first national park, Chitwan National Park (CNP), is located in this district (Figure 4.1) and was established in 1973 (Chitwan District Information : Chitwan Tourism, 2013). The park covers an area of 932 km² and is located in the subtropical Terai lowlands of south-central Nepal. It lies in a river valley basin or dun, along the floodplains of the Rapti, Reu and Narayani rivers. The Chitwan valley consists of tropical and subtropical forests. Sal forest covers 70% of the park. Sal leaves are used locally for plates in festivals and religious offerings, and are also known for their Ayurvedic use in the treatment of wounds, coughs and other ailments (Bodhare, 2022). Grasslands cover 20 per cent of the park. There are more than 50 different species of grasses, including elephant grass (*Saccharum* spp), known for its immense height. The

climate is mainly dominated by the summer monsoon, and the valley experiences three distinct seasons each year: winter, summer, and monsoon.

Chitwan National Park is regarded as one of the best national parks for wildlife viewing in Asia (CNP, 2015). It is home to 68 species of mammals, more than 576 species of birds, 49 species of reptiles and amphibians, 120 species of fish and several species of invertebrates that contribute significantly to ecosystem processes in the region. The reserve is home to many of Nepal's charismatic wildlife species, including the one-horned rhinoceros (*Rhinoceros unicornis*), Bengal tiger (*Panthera tigris*), leopard (*Panthera pardus*) and Asian elephant (*Elephas maximus*), among others (CNP, 2015; Park, 2022). Of the various recreational activities available in the park, elephant safaris are the most popular, especially in and around the buffer zones. Elephants are also used by public entities for transporting and patrolling the park. Buffer zones in the CNP were established in 1996 with the aim of involving people in the management of park resources for the conservation of biodiversity and the livelihoods of buffer zone communities (Bhusal, 2012). The buffer zone covers an area of 750 km² and is home to approximately 45,516 houses and 2,60,352 people (Budhathoki, 2004). Buffer zones are primarily designed to create human-wildlife coexistence by providing an ecological and socio-economic buffer for communities, and in these areas, where wildlife and humans share the same landscape in close proximity, their interaction is very obvious (Heinen & Mehta, 2000).

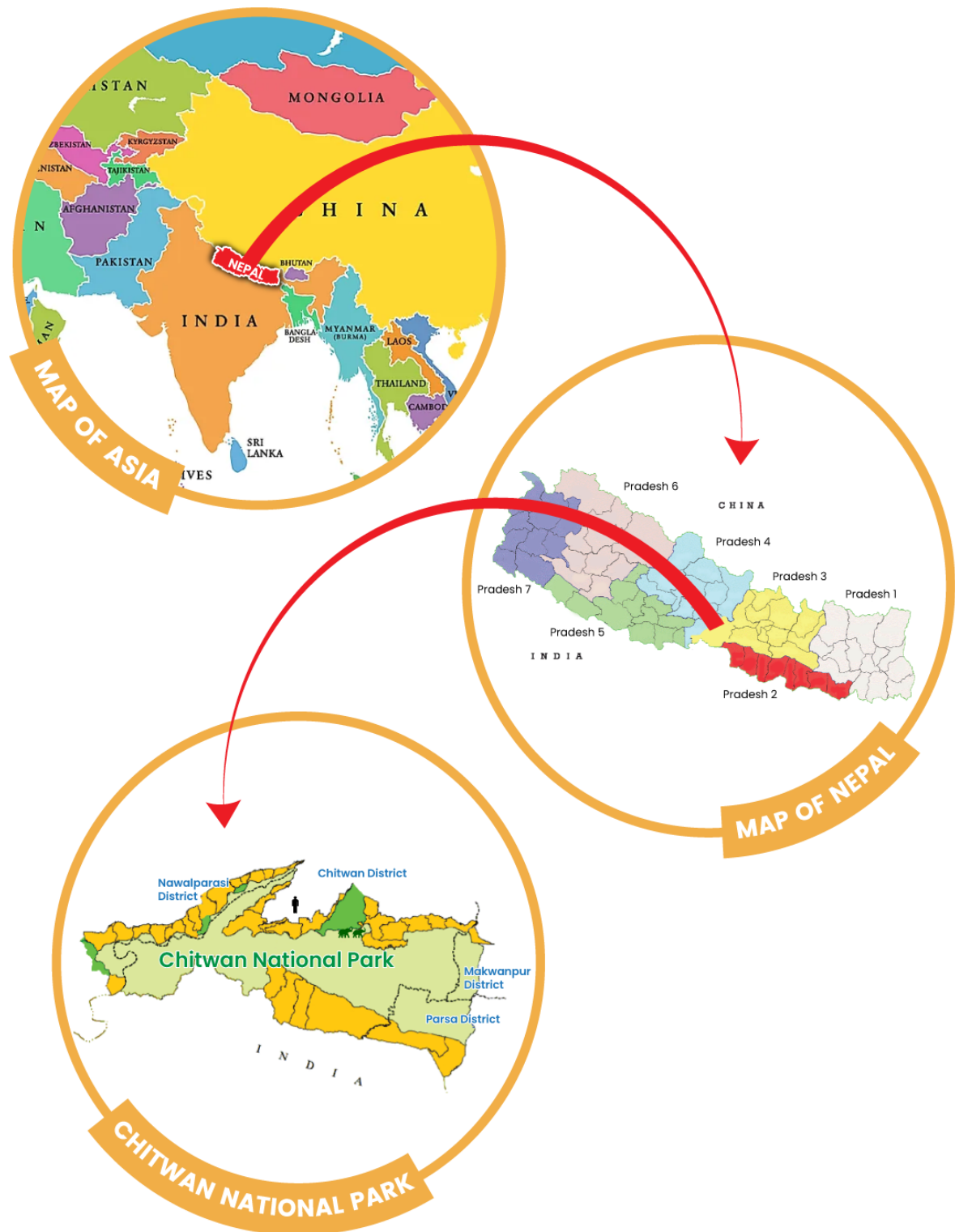


Figure 4.1 Geographical map of Nepal showing our study area, Chitwan National Park in south-central Nepal, and the distribution of the wild elephant population throughout Nepal.

4.3.2 Sample collection

Samples were collected from two deceased adult (>30 years) elephants in Sauraha (Elephant PK and Elephant DG), Chitwan, owned by the government, and used for tourism, grass collection and patrolling. Sauraha is a village at the eastern entrance to Chitwan National Park, near the East Rapti River. These animals had died in captivity and at necropsy showed granulomatous lesions in the lungs compatible with tuberculosis. Tissue samples from lung biopsies (with lesions) were taken from both elephants. The human sputum sample (No. 52G111) collected in Chitwan as part of a previous study (Ministry of Health and Population, 2020) was also genotyped, although it was not closely related to the diseased elephants. It was included to understand the evolutionary relationship with the *M. tuberculosis* strains isolated from elephants, as it was collected from the same region.

4.3.3 Molecular confirmation of *M. tuberculosis* infection

The elephant lung tissue samples (PK and DG) and human sputum samples (52G111) were sent to GENETUP (German Nepal Tuberculosis Project microbiology lab) for culture confirmation of *M. tuberculosis* as described in (Nazarova & Russell, 2017). The heat-killed cultures were then resuspended in 100µl of PBS for DNA extraction. The DNA extraction was performed using a DNA-SorB kit according to the manufacturer's instructions (Sacace Biotechnologies, Italy). Briefly, 300 µl of the lysis solution was mixed with 100 µl of the heat lysed *M. tuberculosis* PBS solution and incubated at 60°C for 5 minutes. The samples were centrifuged at 15000g for 5 minutes to separate the cell debris and DNA in the supernatant. The supernatant was then mixed with 20 µl of the sorbent to bind the DNA to the sorbent. The sorbent was washed twice, dried at 65 °C for 5 minutes and eluted with 25 µl of DNA eluent. Elution was further enhanced by incubating the tubes with DNA eluent at 65 °C for 5 minutes and periodically vortexing. The extracted DNA was then quantified using Qubit (Thermo Fisher Scientific, USA) and *M. tuberculosis* infection was diagnosed using the specific real-time PCR (Sacace Biotechnologies Srl - Via Scalabrini, n.d.).

4.3.4 Whole-genome sequencing of elephant and human *M. tuberculosis* isolates

Two complementary whole genome sequencing approaches were used in this study: Illumina short reads and MinION long reads. In the case of Illumina short reads, genomic DNA from the heat-killed cultures of *M. tuberculosis* was purified using AMPure beads (Beckman Coulter, USA) (1:1), quantified using Qubit (Thermo Fisher Scientific, USA) and normalised to 0.2 ng/µl. The normalised samples were tagged and indexed using the Nextera Indexing Kit (Illumina, USA). The indexed library was then purified using (0.8X) AMPure beads and quantified using HS kit (Thermo Fisher Scientific, USA). Each library was analysed using a bioanalyzer and normalised to a final concentration of 2 nM. The three normalised libraries were then pooled

in equal volumes of 5 µl to give a final pooled library of 2 nM. The pooled library was spiked with 5% of 2nM PhiX and denatured with NaOH. The denatured library was diluted to 20 pM, then 10 pM using hybridisation buffer, and the final 10 pM library was loaded onto the Illumina MiSeq platform using the MiSeq Reagent Kit V2 300-cycles (Illumina, USA), with an estimated coverage of approximately 100x per sample.

For long-read sequencing in Nanopore MinION, normalised genomic DNA (0.2 ng/µl) from the heat-killed cultures of *M. tuberculosis* isolated from the two elephants was also used for long-read sequencing in Nanopore MinION, using the Nextera XT Library Preparation Kit (Illumina, USA). The 20 µl of tagged genomic DNA was end-repaired using Ultra II End Prep Buffer and Ultra II End Prep Enzyme (NEB, UK) according to the manufacturer's instructions. The end-repaired DNA was then subjected to Nanopore DNA preparation using Ligation sequencing kit (Oxford Nanopore, UK) with PCR barcoding expansion kit (Oxford Nanopore, UK). The final library was purified using 0.4X AMPure beads and Long Fragment Buffer (LFB) to obtain the eluate in 15 µl elution buffer. The adapter-ligated library was quantified using the Qubit Fluorometer and approximately 15 fm of the total library was finally loaded onto the MinION for sequencing using 15 µl of sequencing buffer and 10 µl loading beads.

4.3.5 Lineage and Multi Drug resistance status determination

Tb-Profiler version 4.1.0, a stand-alone tool (Phelan et al., 2019), was used for lineage identification and drug resistance assessment of *M. tuberculosis* isolates from whole genome sequences. The tool accepts raw data from both MinION Nanopore and Illumina MiSeq sequences as well as BAM and FASTA files. It is a command-line tool that can use whole-genome data to predict lineages and identify 21 types of drug resistance (based on small variants and large deletions associated with drug resistance).

4.3.6 Processing and analysis of Illumina and Nanopore sequence data

Quality control and filtering of Illumina reads was performed using Fastp v0.20 (Chen et al., 2018), while NanoFilt v2.6.0 (De Coster et al., 2018) was used to filter Nanopore reads. After quality filtering, the Illumina and Nanopore reads were mapped separately to the *M. tuberculosis* reference genome (GenBank accession NC_000962.3) using BWA v0.7.17 (Li, 2013). SAMtools v1.15 (Danecek et al., 2021) was used to convert the SAM files of the mapped reads from both Illumina and Nanopore to BAM files, and to sort and index the BAM file reads using the view, sort and index commands. SAMtools merge was then used to merge the index and sorted BAM files into a single BAM file containing both long and short reads for each sample (Danecek et al., 2021).

4.3.7 Phylogenomic analysis

To perform a comparative phylogenomic analysis of *M. tuberculosis* isolates, we selected a sequence dataset comprising 94 full-length *M. tuberculosis* whole genomes, including data from human isolates in Nepal (n=8), and a genome of *M. africanum* to use as an outgroup. These genomes were from 33 different countries and the four different geographical lineages previously described from elephants worldwide (lineages 1 and 4) and from humans in Nepal (lineages 1, 2 and 3), and they were retrieved from the NCBI Sequence Read Archive (SRA) database (Table S1, Supplementary Information). The prefetch command in the SRA toolkit v2.11.0 (Download: Software: Sequence Read Archive: NCBI/NLM/NIH.) was used to download all whole-genome sequence files of our dataset in SRA format, which were later converted to FASTQ format using the fastq-dump command. Fastp v0.20 was used for quality control and read filtering (Chen et al., 2018). Read mapping to the reference genome (GenBank accession: NC_000962.3) was performed using BWA v0.7.17 (Li, 2013). SAMtools v1.15 was used to convert the SAM files of the mapped reads to BAM files, and then to sort and index the BAM file reads (Danecek et al., 2021). The SAMtools mpileup command generated a text pileup output, summarizing the base calls of the aligned reads to the reference sequence. The consensus call was performed using the call command in BCFtools v1.10 (Danecek et al., 2021), which produced a VCF file that was converted to a FASTQ file using the vcf2fq function in the vcfutils.pl script (Danecek et al., 2021). Seqtk v1.3 (GitHub - Lh3/Seqtk: Toolkit for Processing Sequences in FASTA/Q Formats, n.d.) was used to convert the FASTQ files to FASTA files using the seq command with the masking of bases with quality less than 20.

Variant calling for all the downloaded *M. tuberculosis* sequences and three *M. tuberculosis* isolates from this study was performed using PhaME v1.0.4 (Shakya et al., 2020), with the H37Rv genome (GenBank accession: NC_000962.3) as the reference. The PhaME analysis tool was used to remove repeats from each genome in the dataset, and genome alignment to the reference genome was performed using the nucmer2 tool from MUMmer v3.0, as described in the pipeline (Shakya et al., 2020). The output genome alignments were filtered, and SNP tables were generated for each alignment using the delta-filter and show-snps utilities, respectively, both in MUMmer v3.0. The PhaME pipeline then collated the alignment files to produce a core genome alignment and output a multiple alignment FASTA file (~200 Kbp) with the variant sites. This was further filtered with SNP-sites to remove non-informative synonymous sites, resulting in a final non-synonymous SNP alignment dataset of 2596 base pairs.

Using the non-synonymous SNP alignment data, phylogenomic analysis was performed using maximum likelihood and Bayesian inference. For maximum likelihood, the best substitution models and likelihood trees were evaluated using IQTREE v1.6.11 (Minh et al., 2020) with 500 bootstrap replicates and default parameters. Similarly, for Bayesian inference, the best nucleotide substitution model, TVM, was selected based on the Bayesian Information Criterion score in jModeltest2 v2.1.8 (Darriba et al., 2012; Guindon & Gascuel, 2003) , followed by tree reconstruction in MrBayes v3.2.7(Ronquist et al., 2012) with 2,000,000 iterations, discarding 25% as burn-in.

4.4 Results

4.4.1 Molecular screening results

The two heat-killed culture samples from elephants yielded 1.07 ng/μl and 0.35 ng/μl DNA for DG and PK, respectively, allowing confirmation of *M. tuberculosis* by real-time PCR. The *M. tuberculosis* isolated from the human sample 52G11 was already a confirmed cultured *M. tuberculosis* with a DNA concentration of 2.34 ng/μl.

4.4.2 TB-Profiler results

Lineage analysis using TB-Profiler showed that the three isolates belonged to three different *M. tuberculosis* lineages (Table 4.1). The elephant isolates DG and PK belong to lineage/sub-lineage 1.2.2.2 and 2.2, respectively, whereas the human isolate 52G111 belongs to lineage/sub-lineage 4.1.2.1. None of the three isolates had drug-resistant variants.

Table 4.1 *M. tuberculosis* lineages, sub-lineages, and drug resistance type of the two elephant and one human isolate collected and sequenced in this study and identified using TB-Profiler v4.1.0.

Sample	host	Lineage	Sub lineage	Drug Resistance type
52G111	Human	Lineage 4	Lineage 4.1.2.1	Sensitive
PK	Elephant	Lineage 2	Lineage 2.2	Sensitive
DG	Elephant	Lineage 1	Lineage 1.2.2.2	Sensitive

4.4.3 Whole genome and phylogenomic analysis

The mean coverage of the three *M. tuberculosis* genomes was 99.6%, while the mean depth was 55.67x, with the isolate DG having the lowest mean depth of 39x (Table4. 2). The PhaME pipeline generated a core genome alignment of ~200 Kbp and a non-synonymous SNP alignment of 2,596 bp size, which included the elephant and human isolates and the genomes

downloaded from the NCBI Sequence Read Archive database. The SNP alignment was used for phylogenomic analysis (Table 4. 2).

Table 4.2 Size, coverage, and mean depth of the analysed *M. tuberculosis* genomes after merging of the Illumina short reads and Nanopore long reads.

Sample	Size (bp)	Genome coverage (%)	Mean depth (x)
52G111	4,411,532	99.59	60.13
DG	4,411,532	99.69	39.29
PK	4,411,532	99.51	67.58

The topology of both maximum-likelihood and Bayesian inference trees were similar, as shown in (Figure 4.2). The phylogenomic analysis of *M. tuberculosis* sequences revealed that the two elephant-derived isolates, DG and PK, clustered into clades of two different lineages, lineage 1 and lineage 2, respectively. Similarly, the human-derived isolate, 52G111, clustered within into lineage 4 clade. This result is consistent with the lineages/sub-lineages defined by TB-Profiler. Sample DG is evolutionary closely related to two other published genomes of elephant-derived isolates sampled in Nepal (SRR14514371 and SRR14514372). These three elephant-derived sequences clustered together, in the same sub-clade, with published human-derived isolates sampled in Nepal (ERR1213884) and India (SRR5341274). Similarly, sample PK clustered closely in the same clade with published human-derived isolates sampled in Nepal (ERR553171 and ERR551520). The human sample 52G111 clustered in a clade together with other published human-derived samples from Argentina (ERR757160), Russia (ERR067670) and the UK (ERR047885). The topology of the tree also showed that previously published *M. tuberculosis* isolates derived from captive Asian elephants in the USA and Switzerland belonged to lineage 4 and clustered in a clade with human-derived isolates from nearby geographical regions. Similarly, an isolate originating from wild African elephants (SRR6487127) clustered very closely with a human-derived isolate (SRR1140950), both sampled in South Africa.

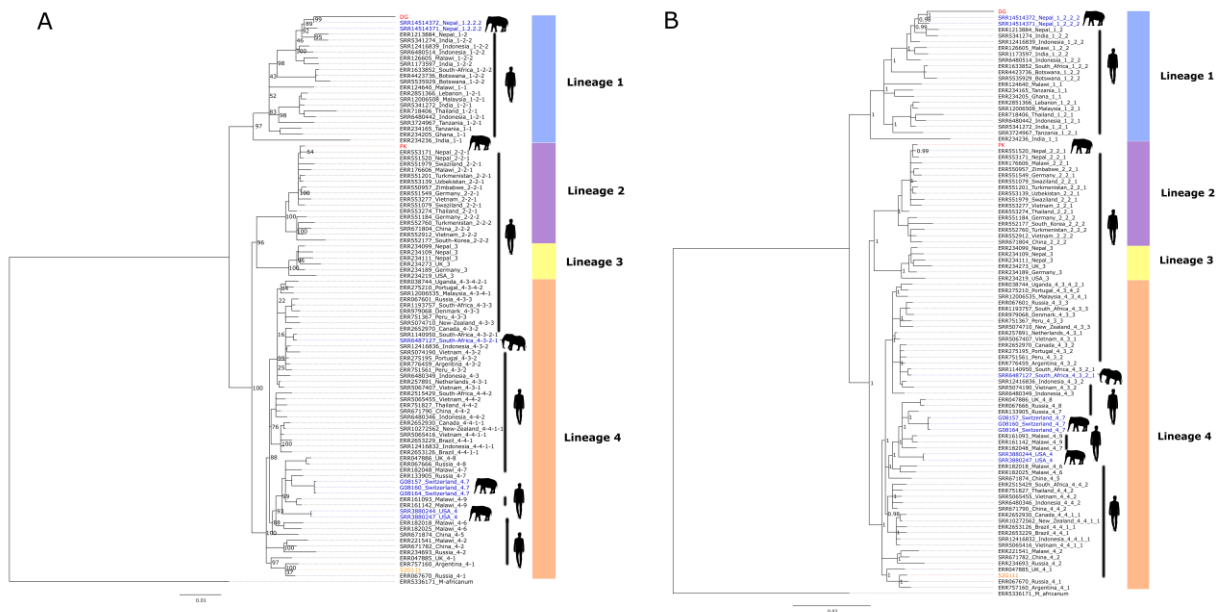


Figure 4.2 Phylogenomic trees constructed using the 2596 bp non-synonymous SNP alignment comprising the three new *M. tuberculosis* genomes described in this study and the 94 genome sequences retrieved from GenBank: (A) IQTREE maximum likelihood tree, the values in the nodes are 500 bootstrap scores; (B) MrBayes Bayesian inference tree, the values in the nodes are posterior probability scores. The elephant (PK, DG) and human (52G111) samples generated in this study are shown in red and orange font, respectively, while the published elephant and human derived samples are shown in blue and black font, respectively.

4.5 Discussion

In this study, we characterised elephant-derived *M. tuberculosis* isolates from two diseased animals (PK and DG), and a human-derived *M. tuberculosis* isolate (52G111) using a whole-genome sequencing approach combining Illumina short reads and Nanopore long reads, resulting in an average genome coverage of 99.6% with an average depth of 55.67x. Phylogenomic analyses were used to explore possible spillover of *M. tuberculosis* between humans and elephants. We also used an online-based TB profiling tool (TB-Profiler) to further identify lineages/sublineages of the sequenced isolates and to assess drug-resistant variants.

The two elephant-derived *M. tuberculosis* isolates appear to be of human origin based on both the TB-profiler result and the phylogenomic trees. These strains are evolutionarily closely related to previous human isolates sampled in Nepal (Figure 4.1), suggesting a bidirectional transmission from humans to elephants. This may be due to the high prevalence of TB in humans and the continuous exposure of elephants to their handlers, the mahouts, rather than to the sporadic contact with tourists (Szydlowski, 2022). Despite living in the same locality, the two elephant-derived *M. tuberculosis* isolates clustered in different clades of TB lineages, suggesting a complex and diverse *M. tuberculosis* transmission dynamics in the region. There must be multiple sources of infection and cross-species (human-mediated) transmission other

than direct elephant-to-elephant transmission. This conclusion could be drawn from the fact that lineage 2 is the second most prevalent lineage in Nepal according to human samples from 2009 to 2010, while lineage 1 is the least prevalent according to a study conducted in 2018 (Ministry of Health and Population, 2020). The possibility of bidirectional interspecies transmission of *M. tuberculosis* between humans and elephants has also been supported by previous reports (Abraham & Pillai, 2016; Malla et al., 2012; Ong et al., 2013). Our results reinforce previous evidence suggesting a close association between *M. tuberculosis* isolates from elephants and humans in Nepal using *M. tuberculosis* spoligotyping (Paudel et al., 2014, 2019), large sequence polymorphism (Paudel et al., 2019) or whole genome data (Paudel et al., 2021).

Tuberculosis is known as a re-emerging disease, but recently it has been considered zoonosis (Shimshony, 2008). The observed cases of zoonosis have implications for global and national elephant conservation. Human tuberculosis already contributes to a huge annual burden of morbidity and mortality, and cases are concentrated in countries such as Nepal where poverty and high population density overlap. Multi-drug resistant *M. tuberculosis* is an additional concern, and the global burden of zoonotic TB is increasing due to the uncontrolled use of antibiotics in animals (Acharya & Wilson, 2019). Zoonotic TB is severely under-reported due to diagnostic challenges and inadequate public health surveillance (Kock et al., 2021). However, this study attempts to fill this gap in Nepal. Furthermore, the risk factors prevalent in South Asian countries, including Nepal, such as high human-animal density, close and frequent contact with infected animals, inadequate disease control measures, consumption of unpasteurised milk and milk products (Bapat et al., 2017), and use of elephants in tourism (Szydlowski, 2021) explain the possibility of zoonotic TB cases in humans as well as zoonosis in animals. Nevertheless, a serious risk factor analysis should be undertaken to better understand the complex and dynamic transmission of TB in Nepal.

In conclusion, the prospect of an elephant potentially acting as a carrier/reservoir for the transmission of drug-resistant *M. tuberculosis* as a result of human-animal interactions is a serious concern that may pose future risks not only to humans but also to TB control in livestock and wildlife (Botelho et al., 2014). A One Health perspective is fundamental to understanding the spillover or transmission dynamics of infectious diseases such as TB. Therefore, regular screening and detection of these infectious diseases in elephants, other wild and domestic animals, human populations, and the environment is essential. The Food and Agriculture Organization of the United Nations (FAO) has already identified in its 'Roadmap for Zoonotic (FAO) a wide range of actions involving a wide range of stakeholders to reduce the risk of transmission, thereby bringing economic benefits and improvements in

animal welfare. The One Health Strategic Framework 2019 has already been approved in Nepal (Ministry of Agriculture and Livestock Development, 2022). In Nepal, implementation of the One Health framework appears to be possible only with a focus on zoonoses and zoonanthroponosis. Therefore, it should be monitored through surveillance systems and better screening and diagnostic strategies using information on circulating *M. tuberculosis* genotypes as identified in this study. However, to limit transmission of the pathogen, oral BCG administration provides significant protection against human (Brewer, 2000) and animal (Buddle et al., 2018) TB in addition to surveillance prevention strategies. In the context of Nepal, regular *M. tuberculosis* screening of all captive elephants and people in close contact with them, especially mahouts, should be promoted to control and prevent the spread of TB. This will also help prevent the spread of TB to other endangered species in the forest (Paudel & Sreevatsan, 2020). In addition, good practices and guidelines for tourism-related activities involving direct human-animal interactions, such as mahout-elephant interaction, should be implemented as a preventive measure.

4.6 Data Availability

The raw FASTQ data of this study has been submitted to NCBI SRA database under BioProject ID: PRJNA884899.

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Chapter 5: Gut microbiota diversity across human, elephant, livestock, and wild herbivore assemblages in Chitwan National Park: implications for conservation medicine*

5.1 Abstract

Gut microbiome composition, which is known to influence overall health and well-being of mammalian species, is mainly shaped by host phylogenetic placement, diet, and environmental factors. The gut microbial community of various hosts in a given area might be shared and inter-connected, including disease-causing bacteria. Therefore, monitoring the gut microbiome composition of wildlife and humans, especially in areas that are densely populated by both, is of paramount importance to prevent zoonotic diseases, thus protecting and strengthening public health. We studied gut microbiota diversity across humans and mammalian herbivores, both domestic and wild, living in and around Chitwan National Park (southern Nepal). Gut microbiota composition was characterized in mahouts (captive elephant trainers and handlers), their domestic livestock and wild herbivores inhabiting the surrounding forests. Firmicutes and Bacteroidota were the most predominant phyla in all host categories. In humans, the relative abundance of Firmicutes was higher, the overall microbiota alpha diversity lower and beta diversity higher compared to other host species. Domestic livestock and wild herbivores displayed similar alpha and beta diversity primarily due to the presence of Proteobacteria, Actinobacteriota and Verrucomicrobiota. The captive elephants had a higher alpha diversity and a significant beta diversity compared to other mammals. Our results indicate the taxonomic affiliation and diet niche are the main drivers of gut microbiome composition. The findings of this study shed light on microbiota sharing and interlinking in a given environment, thereby highlighting the importance of the One Health perspective to better our understanding of health in coexisting species.

Keywords: Microbiome, human-wildlife interface, One Health, Chitwan National Park, zoonosis, Nepal

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5.2 Introduction

The community of microorganisms (bacteria, viruses, archaea and protists) living in a shared environment is known as microbiota. This concept is tightly associated with the entire collection of genomic elements yielded by its members and known as microbiome (D'Argenio & Salvatore, 2015). These microbial communities are crucial in controlling host physiology as well as the development of its immune system and functions such as digestion- playing an important role in detoxification reactions (Hooper et al., 2012). In adult humans, there are more than five million different microbiota associated genes found in the digestive tract (Methé et al., 2012). Alterations in the composition of the gut microbiome are associated with health conditions such as obesity (Turnbaugh et al., 2009; Rosenbaum et al., 2015), diabetes (Jergens et al., 2019; Kieler et al., 2019), kidney diseases (Lau et al., 2018), skin (Penders et al., 2007) and immune-mediated disorders (Silverman et al., 2019), and allergies (Hua et al., 2016). On the other hand, a healthy gut flora is largely responsible for the overall good health of the associated host (von Martels et al., 2017).

Gut microbiome studies have traditionally been based on stool samples and conventional culture-based techniques. However, a major drawback of this approach is pathogens often representing the main target of such studies are undetected, especially if they are represented by taxa new to science or belong to a certain geographic area (Salmonová & Bunešová, 2017). Pathogen detectability with conventional techniques is highly reliant on culture conditions and test sensitivity, while methods based on PCR and Sanger sequencing work satisfactorily only if the primers used amplify pathogen template DNA. The advent of Next Generation Sequencing (NGS) techniques revolutionized microbiome studies allowing, among other things, to overcome these shortcomings, which not only translates into identifying unknown etiological agents but also into developing new tests for pathogen detection (Lamoureux et al., 2022). In particular, the shotgun metagenomic approach represents a powerful tool to disentangle complex patterns of pathogen transmission among ectoparasites, animal reservoirs, and humans (Takhampunya et al., 2019). On the contrary, by using culture-based techniques scientists are able to isolate only 10% to 25% of the overall microbiota (Anguita-Maeso et al., 2020). However, the vast majority of NGS studies on gut microbiome are based on 16S rRNA metabarcoding, which is used to identify the bacterial taxa and estimate their abundance (Jandhyala et al., 2015).

A considerable portion of the investigations addressing the role of microbiome on animal health are focused on the implications on humans (Bahrdorff et al., 2016). Most of these studies, based on NGS approaches, showed that habitat degradation poses risk to animal species, not only because of reduced resources but also through microbiome depletion. Such

changes in the microbial community are known to affect host fitness, often triggering a cascading effect on species interactions and thereby, impacting on their conservation and management (Bahrndorff et al., 2016). On the other hand, social interactions are also an important driver of gut microbiome composition, as evidenced in wild (e.g., yellow baboon, *Papio cynocephalus*: Tung et al., 2015) or feral (e.g., Welsh mountain ponies, *Equus caballus*: Antwis et al., 2018) animal populations as well as in humans (Dill-McFarland et al., 2019).

Untangling multiple factors that influence gut microbiome composition is crucial to understanding the ultimate effect on animal and human health. In particular, the gut microbiota is one of the key drivers behind the biochemical pathways associated with diet and, as such, it is tightly associated with metabolic disease (Rowland et al., 2018). Therefore, the characterization of the bacterial components - the largest one in the gut microbiome - and that of the interplay among multiple bacterial taxa is a fundamental prerequisite to understand the role played by other microbial components, that also exert a crucial impact on host well-being (e.g. Gao et al., 2021), and hence disentangle the complex mechanisms underlying a healthy and well-functioning gut microbiome. Likewise, understanding the reciprocal interactions between the gut microbiome of animal and human hosts sharing the same environment is topical to predict and prevent the emergence and spread of zoonotic diseases (Trinh et al., 2018) like the COVID-19 pandemic (Dhar & Mohanty, 2020) that has threatened public health and well-being at a global scale over the last two and a half years.

In this study, we used 16S rRNA-based metabarcoding to characterize the bacterial component of the gut microbiome across various host categories, namely humans, livestock, captive elephants and wild herbivores, in and around Chitwan National Park (southern Nepal) to estimate its diversity and abundance as a prerequisite to evaluate its implications in the framework of the One Health strategy (Mackenzie et al., 2013). We also evaluated the possible occurrence of *Mycobacterium*, a taxon responsible for serious diseases in mammals and humans (e.g. Namasivayam et al., 2020), as a constitutive member of the gut microbial flora in any of the herbivore host species surveyed in this study.

5.3 Material and Methods

5.3.1 Study area

Established in 1973 as the first national park in Nepal, Chitwan National Park (CNP) is a World heritage site hosting an astonishingly diverse flora and fauna (Chitwan District Information: Chitwan Tourism 2013). CNP stretches over an area of 932 km² and is situated in the Terai lowlands across the flood plains of Rapti, Reu and Narayani rivers (Figure 5.1). The climate of this area is primarily tropical and subtropical, dominated by the summer monsoon. Sal tree

(*Shorea robusta*) forests cover most (70%) of the park, while patches of grasslands (20%) are scattered across it. The park hosts more than 50 different grass species, including the elephant grass (*Saccharum* spp.) (Chitwan, 2022). The periphery of the park, designated as “buffer zone”, separates human settlements from the forest and is used for popular eco-tourism activities like elephant rides. CNP is home to around 68 species of mammals, more than 576 species of birds, 49 species of reptiles and amphibians, 120 species of fishes and numerous invertebrates (DNPWC, 2015). The greater one-horned rhino (*Rhinoceros unicornis*), the Bengal tiger (*Panthera tigris*), the Asiatic elephant (*Elephas maximus*) and the Gharial crocodile (*Gavialis gangeticus*) are some of the iconic flagship species found in CNP. Other charismatic species inhabiting the protected area are carnivores like the leopard (*Panthera pardus*), the dhole (*Cuon alpinus*), the sloth bear (*Melursus ursinus*), the Bengal fox (*Vulpes bengalensis*) and the striped hyena (*Hyaena hyaena*), as well as medium to large-sized herbivores such as the barking deer (*Muntiacus vaginalis*), the hog deer (*Axis porcinus*), the spotted deer (*Axis axis*), the sambar deer (*Rusa unicolor*), the gaur (*Bos gaurus*) and the wild boar (*Sus scrofa*). Elephant safari in the park was first allowed in 1996 to promote eco-tourism and support the livelihoods of 45,516 living around the park (CNP, 2015). Additionally, the buffer area also provides forest resources such as fuel wood, fodder and grass to the surrounding human communities, thus promoting human-wildlife co-existence (Budhathoki, 2004; Bhusal, 2012; Heinen & Mehta, 2000). This, however, entails thinner human-wildlife interface and increased possibility of contact.

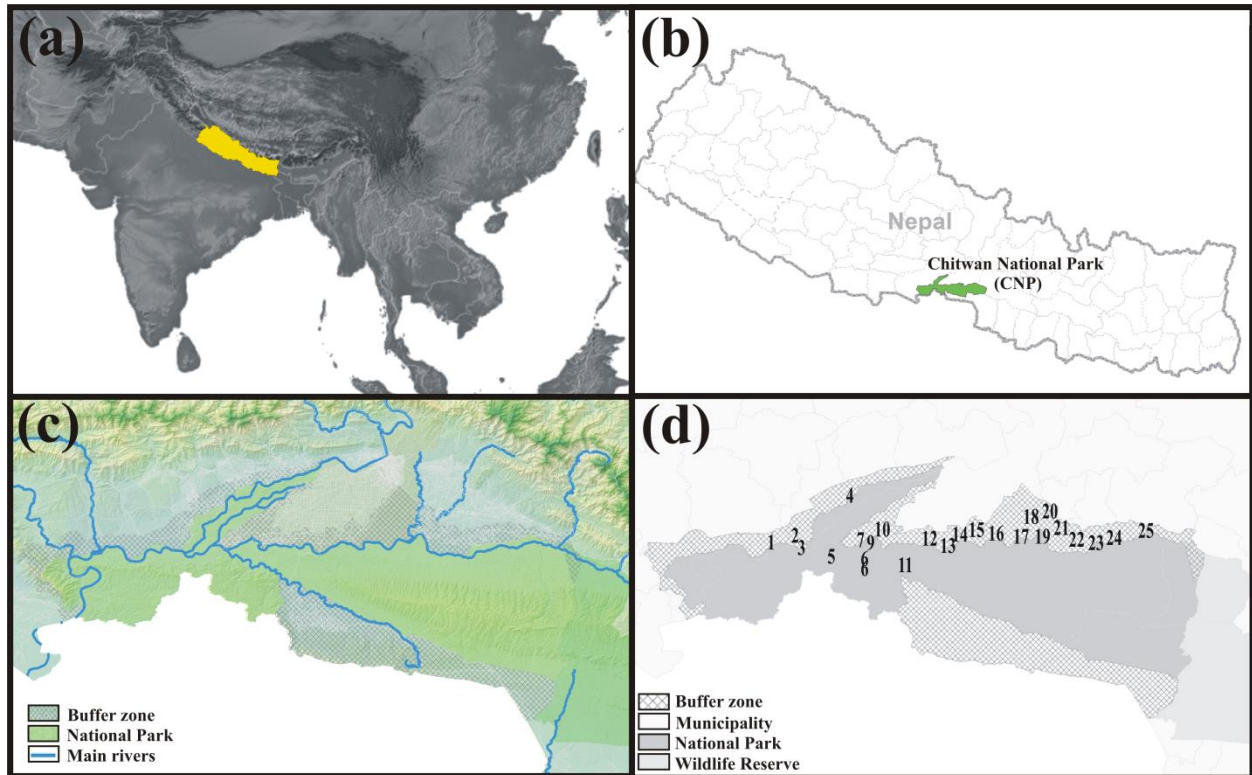


Figure 5.1 Study area and sampling locations (a) Map of southern Asia Position evidencing the location of Nepal (yellow shape). (b) Location of Chitwan National Park (green shape) in Nepal. (c) Physical map of Chitwan National Park and its buffer zone evidencing the main rivers. (d) Sampling locations within the park and the buffer zone (see Table S1 for further details).

5.3.2 Sample collection, DNA extraction and species identification

A total of 176 fecal samples (Chapter 5 Annex, Table S1) from humans ($n = 16$), domestic livestock ($n = 15$, of which three buffaloes *Bubalus bubalis*, one cow *Bos taurus*, six goats *Capra hircus* and five sheep *Ovis aries*), elephant ($n = 40$), of which one wild and 39 captive individuals), and wild herbivores ($n = 105$) were collected in this study. We selected mahouts, an ethnic group of people who are captive elephant riders, trainers or keepers, as human subjects in this study in view of their tight contact primarily with the pachyderms but also with their own livestock. Fecal samples were collected for humans and captive elephants, while fresh fecal samples were collected noninvasively for livestock and wild animals.

Fecal samples (or pellets) of wild herbivores were collected non-invasively from CNP, while those of domestic livestock and captive elephants were collected from its buffer zone- Sauraha region of Chitwan (Figure 5.1), immediately after defecation. All the fecal samples were collected in a 4 ml tube containing 2 ml DET buffer. The samples in DET buffer were transferred in cold chain to the National Trust for Nature Conservation (NTNC) laboratory (Sauraha, Chitwan) before final transfer to the Intrepid Nepal Pvt. Ltd. laboratory in Kathmandu.

Each sample suspension (600 µl) was mixed to an equal volume of 95% ethanol and centrifuged at 13,000 rpm for 1 minute. The supernatant was discarded and the precipitate was suspended in 600 µl of PBS prior to DNA extraction with the QIAmp fast DNA stool minikit (Qiagen, Hilden, Germany) following manufacturer's manual. DNA was eluted in 60 µl ATE buffer.

Species identity of wild herbivore fecal samples was assessed by sequencing a ~412 bp barcoding region of cytochrome-b (cyt-b) gene using universal primers mcb398-f and mcb869-r as described by Verma and Singh (2003), and blast searching against the NCBI GenBank database with 97% nucleotide identity threshold.

5.3.3 Gut microbiota metabarcoding analysis

The amplification of 16S rRNA of bacteria was performed using universal primers 341F and 805R (Iribos et al., 2018) targeting the V3-V4 hypervariable regions. We performed the PCR in a volume of 2.5 µl (Minich et al., 2018) using 1.25 µl of 2X Kapa HiFi Hotstart Ready Mix (Roche, Wilmington, MA, USA) 0.5 µl of each 1 pmol Illumina forward (16s_F) and (16s_R) reverse primers and 0.25 µl of template DNA. The thermal conditions were: initial denaturation at 95°C for 3 min, 35 cycles at 98°C for 30 s, 65°C for 30 s and 72 °C for 30 s, and final extension at 72°C for 10 min. The reaction was performed in triplicate for each sample and the three products of the second PCR were pooled in a single volume of 7.5 µl for each sample. Aliquots of 1.5 µl each were run on a 1.5% (w/v) agarose gel stained with Gel Red Nucleic Acid Gel Stain (Biotium, Fremont, CA) in TAE buffer and visualized through a digital imaging system expecting an approximately 600 bp-long band. The remaining 5 µl of PCR products yielding successful amplification were cleaned-up by 9 µl of Ampure XP beads (Agencourt, Beckman Coulter Inc., La Brea, CA, United States). The required product was separated from the beads using a DynaMag™ 96 Side Magnet (ThermoFisher Scientific, Waltham, MA, USA), washed with 70% ethanol (Emsure, Merck, Germany) and eluted in 15 µl of nuclease-free water.

Subsequently, 3 µl of purified product were used for the indexing PCR along with 1.5 µl of Nextera XT Index 1 Primer (N7XX), 1.5 µl of Nextera XT Index 2 Primer (S5XX) for the Nextera XT index (FC-131-1001 or FC-131-1002), 7.5 µl of 2X KAPA HiFi Hotstart Ready Mix and 1.5 µl of nuclease-free water in a final reaction volume of 15 µl. PCR conditions were: 95 °C for 3 min, 8 cycles at 95 °C for 30 s, 55 °C for 30 s and 72 °C for 5 min, and final extension at 72 °C for 5 min.

The prepared library was quantified using the KAPA Library Quantification kit by adding 6 µl of the KAPA SYBR FAST qPCR master mix with Primer Premix to 4 µl of diluted library DNA (100,000-fold). This 10 µl reaction volume was processed with the following PCR conditions:

initial denaturation at 95°C for 5 min, 35 cycles of denaturation at 95°C and annealing/extension/data acquisition at 60°C for 45 s.

The quantified library was normalized to 4 nM using nuclease-free water and pooled in equal volume to obtain a pooled 4 nM library that was later spiked with 4 nM PhiX and denatured using 0.2 N NaOH. The denatured library was neutralized using Hyb buffer to result into a 10 pM of final library which was loaded using MiSeq Reagent kit v2 (600cycle) (Illumina, USA) with a 2 x 250 bp paired-end configuration for sequencing.

5.3.4 Sequence data processing and taxonomic analysis

We first checked the quality of the raw paired-end reads using FastQC v0.11.9 (Price et al., 2009), then filtered and trimmed short-length (< 235 bp) and low-quality (q-score < 30) bases with TRIMMOMATIC v0.39 (Bolger et al., 2014). We processed the filtered reads using the QIIME2 v2021.11.0 (Caporaso et al., 2010) pipeline. We performed de-noising of paired end reads by trimming, merging, and removing chimeric sequences using the DADA2 plugin in QIIME2. We clustered Operational Taxonomic Units (OTUs) at 100% sequence similarity (amplicon sequence variants) using DADA2 and classified their taxonomy based on SKLEARN using the QIIME2 pre-trained naïve Bayes classifier trained on the Silva 16S rRNA database v138. We aligned the OTUs representative sequences using MAFFT v7.490 (Kato et al., 2002) and reconstructed a maximum-likelihood phylogenetic tree using FastTree v2.1.10 (Price et al., 2009).

5.3.5 Alpha and Beta diversity analysis

We calculated microbial diversity within (alpha diversity) and between (beta diversity) different host categories using QIIME2 based on core-diversity plugins. We performed these diversity analyses after applying rarefaction with even sub-sampling of 10,000 sequences per sample. We categorized the samples mainly based on their host species group i.e., humans, captive elephants, domestic livestock, and wild herbivores. The rarefied abundance data were analyzed with Kruskal-Wallis pairwise test to evaluate the alpha-significance of the diversity across groups. For this purpose, we assessed the alpha diversity using metrics of Faith's phylogenetic diversity (PD) (Faith, 1992) and Shannon Diversity indices (Shannon, 1948). We visualized the alpha diversity matrices using R package ggplot2 v3.34 (Wickham, 2016).

As far as the beta diversity is concerned, we calculated pairwise permutational analysis of variance (PERMANOVA) statistics by running 999 permutations based on Bray-Curtis dissimilarity (Beals, 1984; Bray & Curtis, 1957) and Weighted UniFrac dissimilarities (Lozupone & Knight, 2005) in QIIME2 that we also used to generate principal coordinate analysis (PCoA) plots using Emperor plugin.

5.3.6 *In-silico* prediction of functional composition

We used the in-silico method implemented in PICRUST2 (Phylogenetic Investigation of Communities by Reconstruction of Unobserved States) (Douglas et al., 2020) to predict the metagenomic functional composition of the gut microbiome in humans, livestock, captive elephants and wild herbivores. STAMP v2.1.3 graphical software package (Statistical analysis of taxonomic and functional profiles) (Parks et al., 2014) was used to compare the predicted pathways between different pairs of experimental groups by means of a Welch's two-tailed t-test.

5.4 Results

5.4.1 Host species

The cyt-b barcoding allowed to identify the species in all non-invasive samples (n = 105), which were assigned to spotted deer (n = 58), sambar deer (n = 1), hog deer (n = 2), greater one-horned rhino (n = 28) and wild boar (n = 16).

5.4.2 Gut microbiota composition

Overall, Firmicutes (44.95%) and Bacteroidota (36.73%) were the most abundant phyla across all host categories. We also found Proteobacteria (5.56%), Actinobacteriota (3.26%), Verrucomicrobiota (3.13%), Planctomycetota (1.42%) and Patescibacteria (1.20%) (Figure 5.2). When the different hosts were considered separately, we found that Firmicutes and Bacteroidota were invariably the most represented phyla, especially in humans (73.34% and 19.06%, respectively). We observed substantial taxonomic differences across hosts starting from the third most abundant phylum onwards.

While Fusobacteriota (3.66%) and Proteobacteria (1.86%) were the other major phyla in humans, Verrucomicrobiota (12.28%) and Planctomycetota (2.31%) ranked in the same position in domestic livestock and Fibrobacterota (3.17%) in captive elephants. When examining the wild herbivores, we found that the other major phyla were Actinobacteriota (10.28%) and Proteobacteria (7.06%) in the hog deer; Fusobacteriota (3.15%) and Patescibacteria (2.59%) in the sambar deer; Proteobacteria (7.91%), Actinobacteriota (7.43%), Verrucomicrobiota (2.63%) and Patescibacteria (2.49%) in the spotted deer; Actinobacteriota (6.46%) and Proteobacteria (5.08%) in the wild boar; Proteobacteria (5.66%) and Planctomycetota (2.94%) in the greater one-horned rhino (Figure 5.2). Information on less abundant phyla as well as lower taxonomic categories are provided in supplementary (Chapter 5 Annex, Table S2 and Supplementary file), along with the number of reads per taxa and host

categories, as well as in Figures S1-S2 (Chapter 5 Annex, Supplementary file); the number of shared genera is shown in Figure S3 (Chapter 5 Annex, Supplementary file).

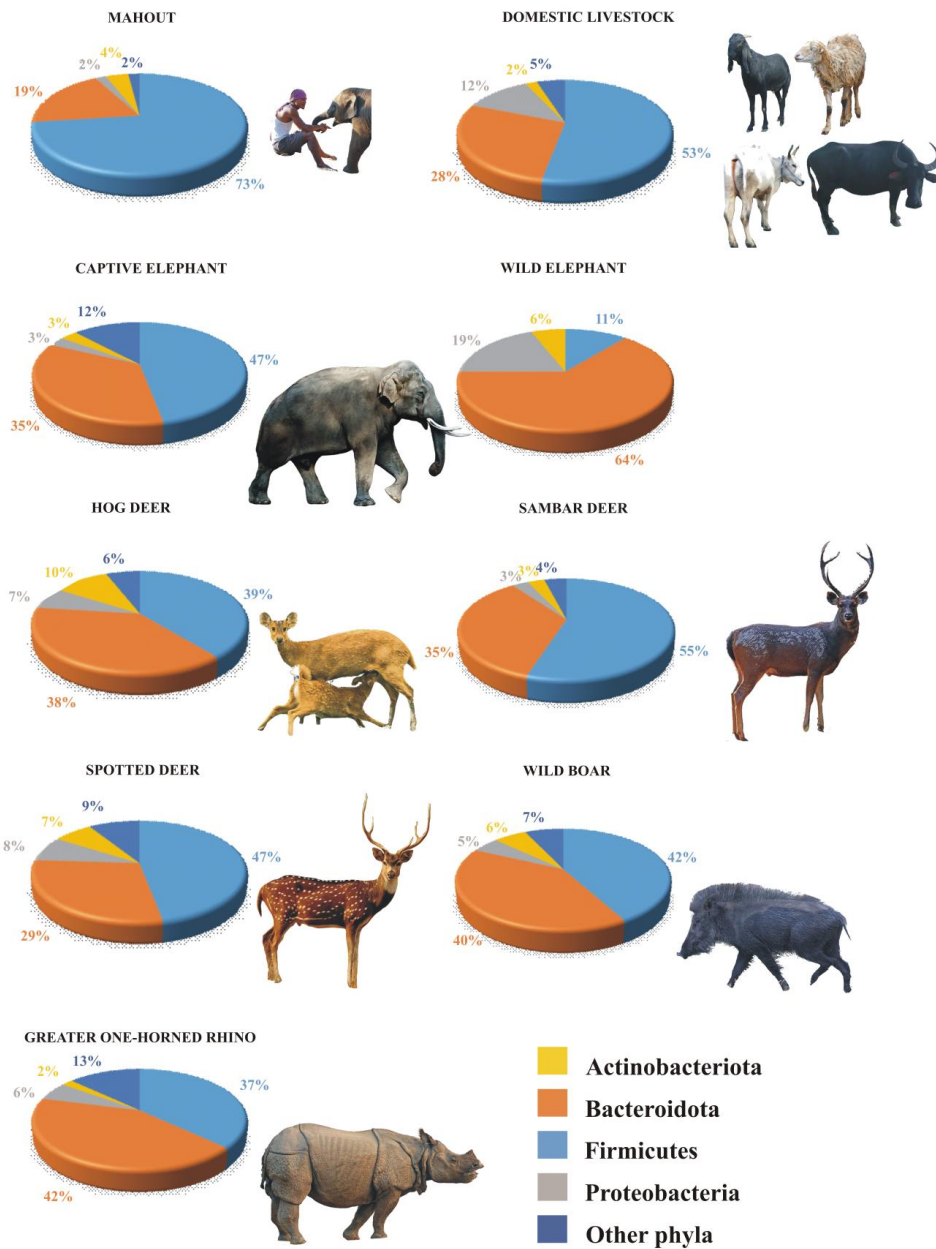


Figure 5.2 Phyla composition of gut microbiota across humans, livestock, captive elephants, and wild herbivores.

We also detected the presence of *Mycobacterium* in the fecal microbiota of some domestic and wild herbivores such as spotted deer (n = 18; 31%), wild boar (n = 16; 6.3%), rhino (n = 1; 3.6%), captive elephant (n = 2; 5.1%) and sheep (n = 1; 20%).

5.4.3 Alpha diversity of gut microbial communities across different host species

PD average values (Table 5.1) ranged from 23 in humans to 69 in captive animals, with intermediate but still comparatively high values in domestic livestock (60) and wild herbivores (58) (Figure 5.3). The PD did not differ significantly between domestic livestock and wild herbivores (Kruskal-Wallis: $H = 0.019$, $p = 0.890$), but it did across all other pairs: domestic livestock vs captive elephant (Kruskal-Wallis: $H = 7.762$, $p = 0.005$), captive elephant vs. human (Kruskal-Wallis: $H = 31.189$, $p = 2.34E-08$), captive elephant vs. wild herbivores (Kruskal-Wallis: $H = 21.749$, $p = 4.59E-06$); domestic livestock vs. human (Kruskal-Wallis: $H = 21$, $p = 4.59E-06$); human vs. wild herbivores (Kruskal-Wallis: $H = 36.350$, $p = 1.65E-09$).

Table 5.1 Faith's phylogenetic diversity (PD) scores with corresponding p -values to determine alpha diversity.

Group 1	Group 2	H	p -value
Captive elephants (n = 37)	Domestic livestock (n = 14)	7.762	0.005
Captive elephants (n = 37)	Humans (n = 15)	31.189	2.34E-08
Captive elephants (n = 37)	Wild herbivores (n = 100)	21.749	3.11E-06
Domestic livestock (n = 14)	Humans (n = 15)	21	4.59E-06
Domestic livestock (n = 14)	Wild herbivores (n = 100)	0.019	0.890
Humans (n = 15)	Wild herbivores (n = 100)	36.350	1.65E-09

The average value of Shannon diversity (Table 5.2) ranged from 4.47 in humans to 7.99 in captive elephants, while it was 7.83 in domestic livestock and 7.74 in wild herbivores (Figure 5.3). No significant differences in Shannon diversity were observed between captive elephants and domestic livestock ($H = 2.506$; $p = 0.113$) as well as captive elephants vs. wild herbivores ($H = 0.912$; $p = 0.339$) nor domestic livestock vs. wild herbivores ($H = 0.171$; $p = 0.679$). Conversely, significant values were obtained when comparing human vs. captive elephants ($H = 29.85$; $p = 4.67E-08$), human vs. domestic livestock ($H = 21$; $p = 4.59E-06$) and human vs wild herbivores ($H = 37.56$; $p = 8.86E-10$).

Table 5.2 Shannon Diversity Index scores with corresponding p-values to determine alpha diversity.

Group 1	Group 2	H	p-value
Captive elephants (n = 37)	Domestic livestock (n = 14)	2.505	0.113
Captive elephants (n = 37)	Humans (n = 15)	29.850	4.67E-08
Captive elephants (n = 37)	Wild herbivores (n = 100)	0.912	0.339
Domestic livestock (n = 14)	Humans (n = 15)	21	4.59E-06
Domestic livestock (n = 14)	Wild herbivores (n = 100)	0.171	0.678
Humans (n = 15)	Wild herbivores (n = 100)	37.561	8.86E-10

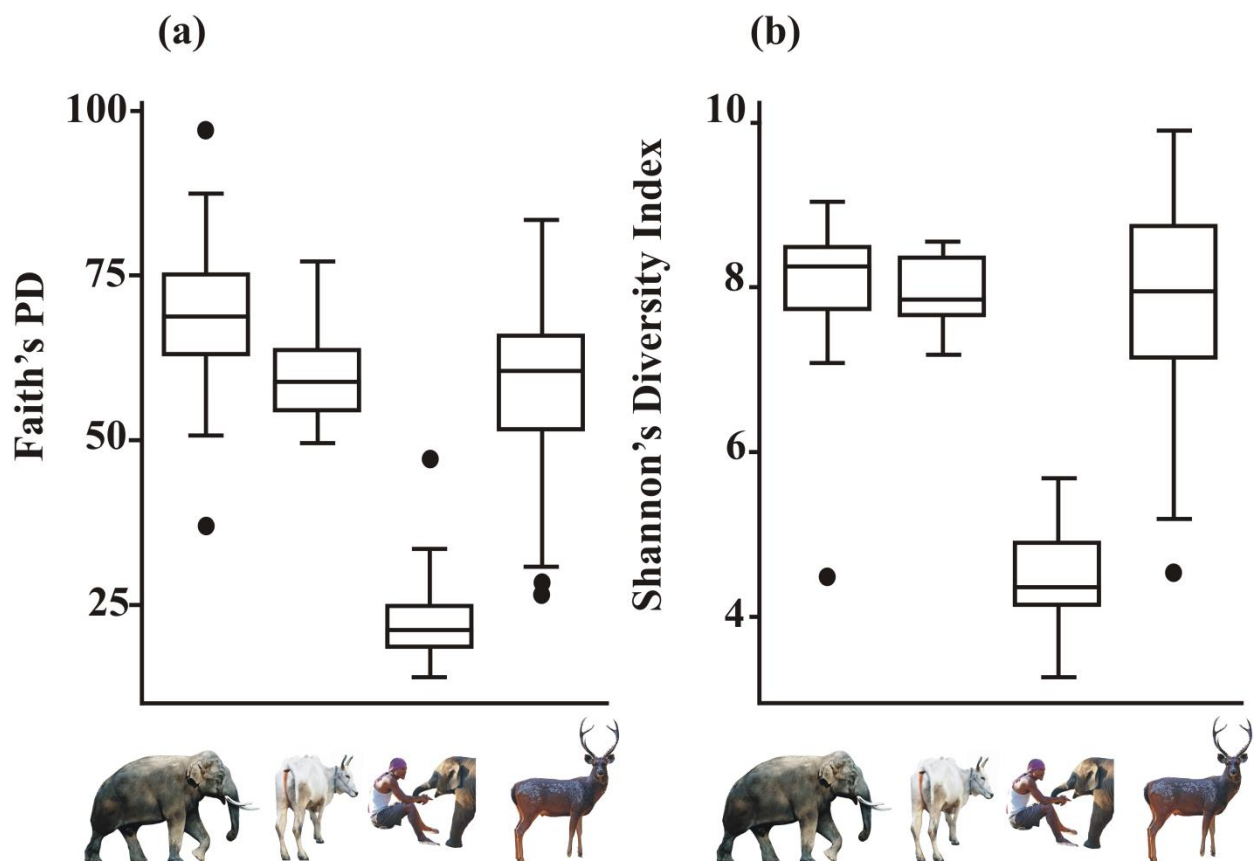


Figure 5.3 Alpha diversity based on Faith's phylogenetic diversity (a) and Shannon's diversity metrics (b) across different host categories: captive elephants, domestic livestock, humans, and wild herbivores (left to right).

5.4.4 Beta diversity of gut microbial communities across different host species

We found significant differences in Bray-Curtis beta diversity between all categorized host species groups (PERMANOVA; Pseudo-F = 10.47; $p = 0.001$) as well as Weighted Unifrac beta diversity (PERMANOVA; Pseudo-F = 16.18; $p = 0.001$) (Figure 5.4). The pairwise PERMANOVA beta diversity differences across all group combination using both Bray-Curtis and Weighted Unifrac distances were significant (Table 5.3 and Table 5.4).

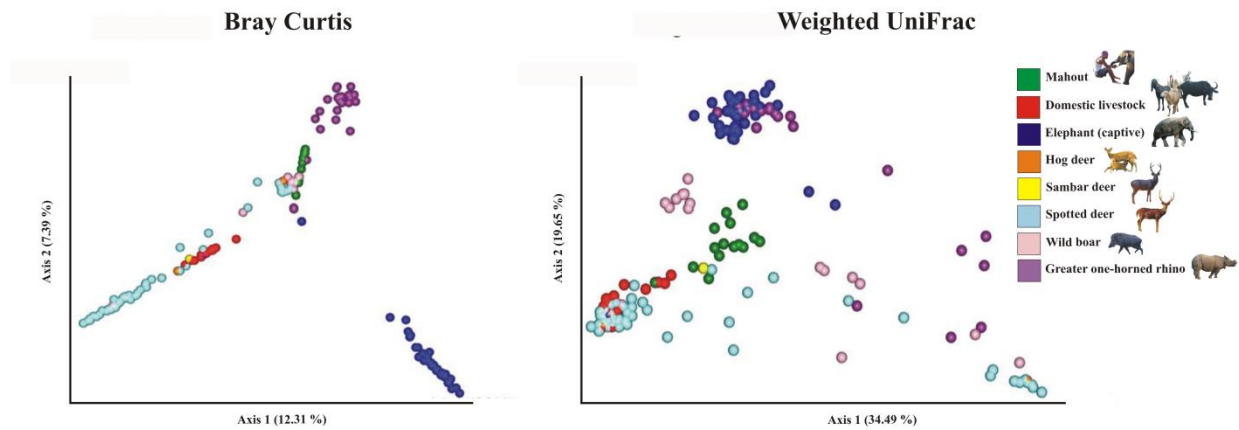


Figure 5.4 Beta diversity based on Bray Curtis and Weighted UniFrac distance metrics.

Table 5.3 Bray Curtis dissimilarity values for comparisons between captive elephants, domestic livestock, humans and wild herbivores.

Group 1	Group 2	pseudo-F	p-value
Captive elephants	Domestic livestock	10.643	0.001
Captive elephants	Humans	14.096	0.001
Captive elephants	Wild herbivores	17.607	0.001
Domestic livestock	Humans	7.462	0.001
Domestic livestock	Wild herbivores	3.632	0.001
Humans	Wild herbivores	8.646	0.001

Table 5.4 Weighted UniFrac distance values for comparisons between captive elephants, domestic livestock, humans and wild herbivores.

Group 1	Group 2	pseudo-F	p-value
Captive elephants	Domestic livestock	36.309	0.001
Captive elephants	Humans	32.994	0.001
Captive elephants	Wild herbivores	19.119	0.001
Domestic livestock	Humans	32.025	0.001
Domestic livestock	Wild herbivores	6.245	0.003
Humans	Wild herbivores	11.201	0.001

5.4.5 *In-silico* functional composition

When examining the predictions of the metagenomic functional compositions Figure S4-S9 (Chapter 5 Annex, Supplementary file), we found that a total of 201, 186, 289, 166, 263 and 248 KEGG (Kyoto Encyclopedia of Genes and Genomes) pathways were significantly enriched (Welch's t-test; $p < 0.05$) when comparing humans vs. captive elephants, humans vs. domestic livestock, humans vs. wild herbivores, captive elephants vs. domestic livestock, captive elephants vs. wild herbivores and domestic livestock vs. wild herbivores, respectively.

Overall, 50, 55 and 54 of these pathways were significantly higher, (with 151, 131 and 235 being significantly lower: Welch's t-test; $p < 0.05$) in humans than in captive elephants, domestic livestock and wild herbivores, respectively, while 103 and 82 were significantly higher (with 160 and 166 being significantly lower: Welch's t-test; $p < 0.05$) in captive elephants than in domestic livestock and wild herbivores, respectively. Finally, 61 pathways were significantly higher (with 187 being significantly lower: Welch's t-test; $p < 0.05$) in domestic livestock than in wild herbivores. Pathways "sucrose degradation IV (sucrose phosphorylase)" and "sucrose degradation III (sucrose invertase)", two of the main pathways related to sucrose metabolism, stood out among those significantly enriched in humans when these were compared to the other groups. On the other hand, "pyrimidine deoxyribonucleotides de novo biosynthesis" and "pyrimidine deoxyribonucleotides biosynthesis from CTP", related to the synthesis of nucleosides that are not only a fundamental part of all nucleic acids but are also involved in carbohydrate and lipid metabolism, appeared to have a higher mean proportion in captive elephants than in all other groups, while "L-tyrosine degradation", related to the metabolism of a proteinogenic amino acid and also involved in signal transduction as key component of neuromediators, was significantly enriched when compared to humans and captive elephants but not to wild herbivores. When looking at domestic livestock, "super-pathway of menaquinol-8 biosynthesis" and "pyruvate fermentation to acetate and lactate II" stood out as significantly enriched when compared to humans along with captive elephants and captive elephants along with wild herbivores, respectively. Finally, "super-pathway of heme biosynthesis from glutamate", related to the metabolism of an iron-containing prosthetic group yield by many essential proteins including cytochromes and globins included, "TCA cycle VIII (Helicobacter)", associated with the anaerobic metabolism of *Helicobacter pylori*, and "pyruvate fermentation to butanoate", which is associated with the acidogenic growth of some bacteria, were significantly enriched in wild herbivores when these were compared to humans and wild livestock, with "super-pathway of (R,R)-butanediol biosynthesis", an anaerobic process with unclear functions, showing a higher mean proportion than in both humans and captive elephants. Overall, 142 pathways were shared between humans and all the other host categories, 38 between humans and captive elephants plus wild herbivores, and 21 between humans and domestic livestock plus wild herbivores, respectively. On the other hand, 88 pathways were exclusively shared between humans and wild herbivores as opposed to the 38 and 21 found only in the pair humans-captive elephants and humans-domestic livestock, respectively (Figure 5.5).

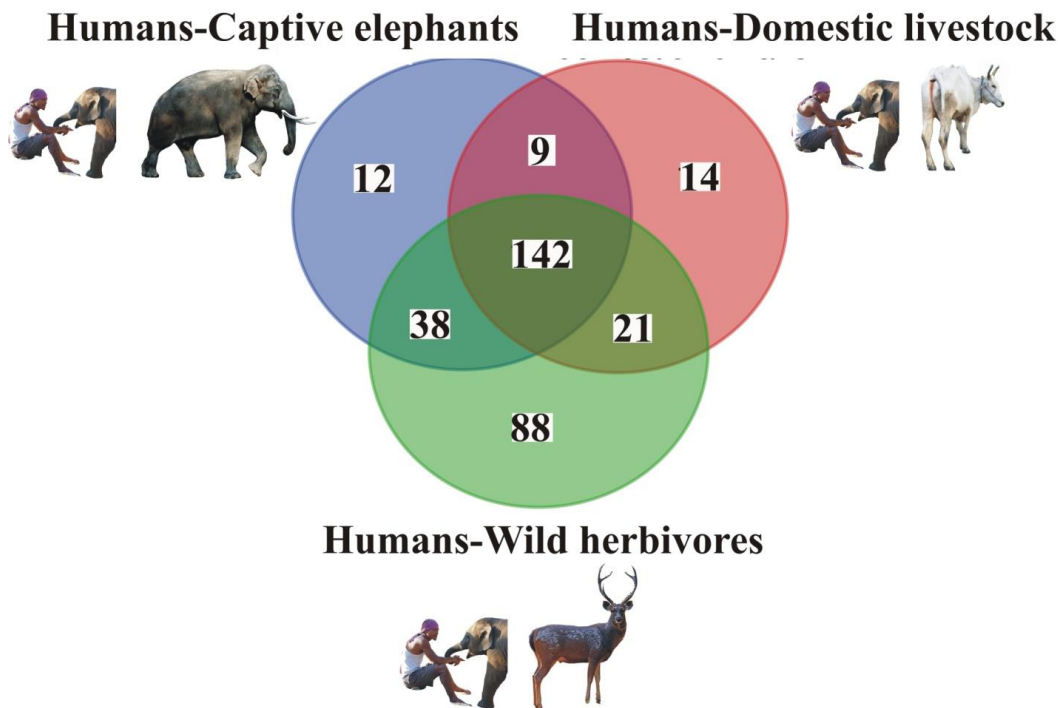


Figure 5.5 Predicted pathway abundances between domestic livestock and wild herbivores in gut microbiome. KEGG database functional categories are reported next to the histograms (left panel: mean $\pm$ SD) and q-values (right panel: 95 % confidence intervals).

5.5 Discussion

Gut microbiome makeup is widely recognized as a key driver of human and animal health. The recent advent of NGS has offered a suite of increasingly powerful and affordable tools to get insights into the abundance and structure of such microbial communities, delivering massive amount of information of evolutionary, conservation and management interest. Importantly, these data, based on the One Health perspective, are key for protecting and strengthening public health. In our study, we characterized the gut microbiome in humans and other mammals (both captive/domesticated and wild) in a human-wildlife interface ecosystem and looked into their interactions at the microbiome level.

We found that humans had a significantly lower alpha diversity and different beta diversity than the other host groups surveyed in this study despite close proximity and interactions. The gut microbiota of mahouts was dominated by Firmicutes, which are known to play a major role in human health (Rinninella et al., 2019; Rizzardi et al., 2021). Many bacteria belonging to this phylum helps breaking down carbohydrates, such as those contained in dietary fiber and resistant starch, which could not be otherwise digested by the host enzymes (Kaur et al., 2019). In contrast, all the other herbivorous mammals shared similar proportions of Firmicutes and Bacteroidota, which were invariably the most represented phyla. Likewise, both alpha diversity metrics showed that the gut microbiota community of domestic livestock and wild

herbivores was comparable both in terms of composition and relative abundance. This outcome is plausible in that all the members of these two host categories belong to former order Ungulata and to the extant order Artiodactyla, with the only exception of the greater one-horned rhino (which instead belongs to order Perissodactyla), and have largely overlapping dietary niches. Moreover, livestock and wild herbivores often co-occur and compete for the same trophic resources in CNP buffer zone. On the other hand, the beta diversity differed significantly between livestock and wild herbivores. This results could be explained in view of the overrepresentation of some host species, namely wild boar, greater one-horned rhino and especially spotted deer. The latter, other than being by far the most abundant wild herbivore of the Indian subcontinent and representing the major prey species for the resident large carnivore guild (Lamichhane et al., 2015), has a broad ecological niche, which could reflect its high microbiome beta diversity.

We also detected *Mycobacterium* in 20% of the spotted deer surveyed in this study along with few of other herbivorous mammals including captive elephants (Rajbhandari et al., 2022). Since mycobacteria are commonly found in soils as well as ground and surface water bodies (Falkinham, 2009), this finding may be indicative of some forms of environmental contamination of collected fecal samples. Considering that some pathogenic bacteria causing tuberculosis in both humans and animals belong to this bacterial genus, this finding should spur further investigations for public health reasons as well as to understand the role of these bacteria in the normal microbial flora of these mammal hosts.

We observed that captive elephants displayed a significantly higher phylogenetic diversity of bacterial taxa in their gut microbiome, while species richness and evenness quantified as alpha diversity was comparable to that of livestock and wild herbivores (Figure 5.3). On the contrary, the beta diversity of captive elephants was significantly different from that of all the other host categories, their mahouts included (Figure 5.4). Elephants, although sharing their diet with other herbivores, are phylogenetically distinct (Proboscidea). Other resident megafauna, the greater one-horned rhino, shared several similarities with the elephants (Figure 5.4).

When the taxonomic levels below the phylum were considered, a marked difference in microbiome composition emerged between captive and wild elephants, with the former showing, along with humans, the highest proportion of Lachnospirales and the latter yielding much higher proportion of Caulobacteriales and Sphingobacteriales than any other host category Figure S2 (Chapter 5 Annex, Supplementary file). Captive elephants are given variety of food throughout their lifetime, which makes it difficult to assess which diet regime corresponds to their healthier gut microbiome community. Addressing this specific question in

the near future is certainly among the research priorities of the studies that can be carried out in Chitwan (Moustafa et al., 2021; Gautum & Koju, 2022).

In spite of a constitutive proportion of common genera (the highest share of mahouts being with their pachyderms and not with their livestock: Figure S3 (Chapter 5 Annex, Supplementary file) - the assemblages of humans, livestock, captive elephants, and wild herbivores inhabiting CNP and its buffer zones still retain well-differentiated gut microbiome communities despite their close interactions. Livestock and captive elephants spend their entire lifetime in and around human settlements and are mostly dependent on humans for food and shelter. It is worth mentioning that contrasting evidence do occur, as evidenced by the substantial share of bacteria (including potentially pathogenic taxa) and their resistance genes between veterinary students and the livestock they had been in close contact with over just three months (Sun et al., 2020). Conversely, our results aligned with conventional wisdom postulating that microbiome is primarily shaped by the host taxonomic position and only secondarily by their diet niches and environment (Fu et al., 2021; Kartzinel et al., 2019; Rojas et al., 2021). Noteworthy, the marked difference in bacterial composition of gut microbiome across host categories is also corroborated by their in-silico functional composition Figure S4-S9 (Chapter 5 Annex, Supplementary file), with actually humans sharing more KEGG pathways with wild herbivores rather than captive elephants and domestic livestock Figure S10 (Chapter 5, Annex Supplementary file). It could be argued that the best approach would have been sampling fecal samples directly from the rectus of wild herbivores to prevent any possible environmental or cross-contamination, but this was not feasible due to the strong restrictions in conducting research on wildlife. However, previous research on tiger gut microbiome based on non-invasive samples had evidenced a markedly distinct composition when compared with local soil microbiome profile (Karmacharya et al., 2019) and, therefore, we have reasons to believe that gut microbiome profiling based on non-invasive samples is reasonably accurate.

5.6 Conclusions

Our research revealed the bacterial composition and diversity of the gut microbiome of various host species in a popular national park in Asia with high human and animal densities, thus providing information on microbiome sharing and interactions within an ecosystem with increasing contacts and fast thinning interface between people and wildlife.

5.7 Data Availability

The 16S sequence raw data generated in the present study were deposited in read archive (SRA) of the NCBI database under accession number PRJNA880466, while the cyt-b partial sequence were deposited in GenBank under accession numbers OQ127588-OQ127636 and OP314810-OP314869. All other related data are included in the manuscript and supplementary data.

5.8 References

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Chapter 6: General Discussion, Conclusions, Recommendations and Further Research

6.1 Synthesis of main findings: Infectious disease spill-over dynamics from One Health perspective- MTB in captive elephants of Nepal as a disease model

6.1.1 MTB in Captive elephants of Nepal- a quintessential disease model from One health perspective

Elephants in Nepal that have been held captive have been serving as one of the means of livelihood for people living in the vicinity of one of the national parks of Nepal - Chitwan National Park. Captive elephants have been used extensively to ferry tourists (for elephant back ride) inside the buffer zone area of the park, which is often seen as one of the most popular touristic activities. These captive elephants are heavy investments for owners- private or government, and therefore, are tamed by dedicated elephant caretakers, also known as mahouts. In addition to humans, these captive elephants also encounter other domesticated and wild animals. Tuberculosis (TB) is a prevalent disease among humans, with Nepal having one of the highest infection prevalence in the world. Tuberculosis is also emerging as one of the biggest health concerns among captive elephants. Apart from tuberculosis, there are several other health problems and diseases associated with elephants such as; elephant endotheliotropic herpesvirus (EEHV), skin problems, obesity, joint and foot problems (Mager Stellman, 1998) and some of the elephants in Nepal even have diseases related to their eyes. Many wildlife diseases affect not just wildlife but also human and domestic animals (Hanisch-Kirkbride et al., 2013).

In this thesis, a systematic review done in (Chapter 1) provides evidence to support that the existence of TB has been reported globally in both captive and wild elephants. The prevalence of TB reported in elephant population ranges from 0% to 23%, and higher prevalence is observed for elephants that are in contact with humans that have been infected with TB. The risk of elephant to human infection transmission increased significantly with duration of exposure and contact with infected elephants. Some studies have described possible TB transmission to captive elephants from other animals (wild and domestic), suggesting inter- and intra-species transmission (Sternberg et al., 2005; Thapa et al., 2021). Thus, in order to control and minimize the risk of transmission there must be regular TB screening, treatment, and eventually even a vaccination program, at least for in-contact humans. However, lack of

harmonization of TB testing in elephants and their human contacts remains a challenge for prevention and control of TB. The results obtained from this thesis, support the need of a routine and comprehensive TB screening program for early diagnosis of infection through combined efforts of public health, veterinary medicine, and occupational health experts. This implies the need for implementing One Health perspective to elephant TB control. Several newly published articles had been reviewed in (Chapter 1), providing new insights regarding the necessity of future research on elephant TB, under the One Health perspective.

6.1.2 Risk assessment of *Mycobacterium tuberculosis* pathogen- its reservoir and spillover/transmission dynamics

In (Chapter 2), I investigated the socio-demographic aspects of elephant husbandry, and its contact with other animals regarding TB infection risks. A total of 99 captive Asian elephants were evaluated for antibodies against *M. tuberculosis* complex (MTC), and 223 mahouts were interviewed to investigate putative associations between human risk factors and MTC antibody detection. The interview revealed several risks associated with elephant husbandry and cross-species contacts. The prevalence of serum antibodies against MTC in captive elephant population was found to be 20%.

The social-demographic aspects of the mahouts were not identified as a relevant risk factor for TB in the elephants. The most relevant results were obtained through structured questions on elephant husbandry. Identified risks included elephant ownership (more evidence of TB in among government-owned elephants), hand-feeding the elephant (as opposed to using a bucket or the floor), and not having special Personal Protective Equipment at work and disposing of elephant waste in pits (as opposed to other means). Contact with other animals and inter-species contacts also revealed other risk factors too. Captive elephants cared by mahouts with livestock present in their household, or nearby their household, were at higher TB risk. In addition, captive elephants cared by mahouts reporting having found a dead animal and collecting it to eat or share among the mahouts were at a higher risk (Chapter 2)

Close contact between humans and elephants in countries with a high human TB burden may increase risk of infection in elephants through reverse zoonotic transmission (Michel et al., 2013). In our study, 16 mahouts family having a history of TB. The high prevalence of TB in both humans and elephants could mean that there might be bi-directional infections. The results of (Chapter 2) suggest that MTC transmission risk in the study area is mostly from humans to elephants rather than from elephants to humans. Whole Genome Sequencing of infected TB elephant further confirmed this evidence of human TB cross infection to elephants (Chapter 4). This highlights the importance of a well-coordinated One Health program

involving both animal and human health sectors to prevent the spread of MTC in multi-host communities where TB remains at high prevalence, particularly in humans.

The prevalence of antibodies against MTC in elephants was found to be (20%). This estimate was within the range defined in the systematic review paper (Chapter I), where the TB prevalence in elephant populations ranged from 0% to 23%. Recent research carried out on 97 elephants at Sauraha (Chitwan) found prevalence of elephant tuberculosis to be 27% (Manuel et al., 2022). This result is slightly higher when compared to the prevalence of the present work, but this can be due to the use of different diagnostic methods. Close contact between humans and elephants in countries with a high human TB burden may increase risk of infection in elephants through reverse zoonotic transmission (Michel et al., 2013). However, the risk may also be present in the other direction, meaning elephant-to-human or captive-to-wild-elephant transmission.

The mahouts were found to be involved in high-risk behaviors, such as disposing elephant waste (43%) in open areas and using elephant dung as briquettes for cooking purposes. Personal Protective Equipments (PPEs) were found to be used by a limited number of mahouts (26%). Other risky practices, such as direct hand-feeding animals without proper safety measures were commonly observed. Preventative measures such as biosecurity practices could reduce the risk of TB infections in both humans and animals.

The contact between captive (68%) and wild elephants was found to be high. Wild animals, including deer and rhinos, come to drink and share the food and water along with the livestock and captive elephants. These situations pose a risk of transmission among livestock, captive to wild elephants, e.g., by sharing water or other resources. Hence, captive elephants might represent a bridge host, potentially enabling transmission between wildlife, domestic animals, and human beings (Caron et al., 2015).

Approximately 59% of the mahouts responded that animals enter the dwelling where they live and (13.5%) of the mahouts collect dead animals to eat or share. Eating dead animals, meeting, and sharing commonly consumed resources with livestock keep mahout at high risk because dead animals are potential carriers of TB host.

6.1.3 Tuberculosis Diagnostics

The prevalence of TB was calculated using test DPP® VETTB Assay for Elephants USA - Chembio Diagnostics DPP, for estimating the risk assessment. However, different ELISA test antigens like MPB83, ESAT-6, and the protein complex P22 were performed, with anti-elephant secondary antibodies produced in mice and rabbits. Unfortunately, this study failed

to set up an accurate ELISA test to refine TB diagnosis in elephants. This brings attention to the need of exploring alternative accurate diagnosis methods for assessing the prevalence of TB. This research complements the diagnostics capacity by performing proteomic analyses in serum samples from TB positive and negative elephant samples.

In (Chapter 3), I investigated the proteomics of TB pathogenesis, with the main objective to identify and quantify of differentially represented proteins in serum samples of TB+ and TB- elephant. Eight adult elephants' serum samples of TB reactive antibodies and non-reactive sera, - TB-positive (TB+) and TB-negative (TB-) -, were used for proteomics analysis. Protein concentration in serum samples was determined using the BCA Protein Assay with BSA (Sigma-Aldrich, St. Louis, MO, USA) and mass spectrometry for proteomics analysis was conducted. The generated data were deposited in the Proteome Xchange Consortium. Phylogenetic analysis was conducted for *M. tuberculosis* type I site-specific deoxyribonuclease, HsdR family (sequence ID: SGD45129.1, length: 1040 amino acids) using BLAST Tree Viewer. For molecular function (MF), biological process (BP), and subcellular localization (CC) were done in human homolog proteins using uniprot. COBALT was used for protein sequence alignment between identified mycobacterial proteins with predicted antigen binding regions to Ig proteins and *M. tuberculosis* P9WNK5|ESXB_MYCTU ESAT-6-like protein EsxB identified as highly reactive to antibodies in infected elephants.

Proteomics analysis identified 26 significantly dysregulated proteins in TB+ elephants (positive for presence of TB) when compared to TB- (negative for presence of TB) animals. Among these, 10 (38%) were identified as Ig proteins (9 over-represented and one under-represented in TB+ elephants) and 16 (62%) as non-Ig proteins (12 over-represented and 4 under-represented in TB+ elephants). The results were significantly different between groups for both over-represented and under-represented proteins, thus further supporting proteomics results. The predictive models for Igs over-represented in TB+ elephants showed an association with exposure to different *Mycobacterium spp.* The results provided new information regarding antibody response to mycobacterial infection and biomarkers associated with tuberculosis and protective response to mycobacteria among Asian elephants.

The deoxyribonuclease HsdR family protein (SGD45129.1), was phylogenetically closely related to mycobacteria. The only identified under-represented Ig protein in TB+ elephants and thus over-represented in TB- animals (G3UIN4;), predicted *Mycobacterium spp.* reactive epitopes were associated with biomarkers of protective response to mycobacteria. The protective mechanisms included defense against infection (Alpha-1-B glycoprotein A1BG, Serpin family A member 1 SERPINA1, Transthyretin TTR), neuroprotection (TTR) and

reduced risks of inflammation, infections, and cancer (SERPINA1, Keratin 10 KRT10) [55-59]. Mycobacterial proteins reactive to Ig proteins in TB+ elephants seemed to constitute candidate diagnostic antigens, and highly represented Ig protein, G3UDI6 (TB+/TB- ratio = 1.9). A recent work reported that potential diagnostic biomarkers for pulmonary tuberculosis in humans such as CXCL1, MMP8, IL-10 and TNF-alpha are not elevated in sera from *M. tuberculosis* infected elephants (Laura I et al., 2007). These results are in agreement with our proteomics study in which these proteins were not identified as differentially represented in response to infection.

The combination of identified reactive epitopes suggests potential for designing a chimeric diagnostic antigen (ISSANYG-GGGGS- WLRYYSDRNWNR- C-KLH). This was identified as under-represented Ig protein in TB+ elephants and thus over-represented in TB- animals (G3UIN4) reacted to *Mycobacterium spp.* epitopes with predicted protective capacity. Therefore, the application of quantum vaccinomics approach with reactive epitopes in mycobacterial RND family transporter membrane protein and protein translocase subunit SecD is Potential to design a new candidate protective chimeric antigen with predicted reactive immunological quantum or protective epitopes (QSLFDATDKID-FDSS- KNDEFFYL-GGGGS-LIYLASK-C-KLH) for TB control in Asian elephants).

The results of the Asian elephant serum proteome in response to TB advanced our understanding of elephant immune response to *M. tuberculosis*. Future studies should further explore the design, production, and assay of identified candidate diagnostic, prognostic and protective antigens for TB monitoring and control in Asian elephants.

As mentioned above, the lack of harmonization of TB testing in elephants and their human contacts remains a challenge to prevent TB in animals. Furthermore, to explore the possible potential antigen for TB detections, the study looked for the Comparative Immunological response- MTB infected vs. not infected proteomics aspect of TB pathogenesis. The predictive models for Ig over-represented in TB+ elephants showed an association with exposure to different *Mycobacterium spp.* For the only identified under-represented Ig protein in TB+ elephants and thus over-represented in TB- animals (G3UIN4;) predicted *Mycobacterium sp.* reactive epitopes were associated with biomarkers of protective response to mycobacteria. These results provided information on the antibody response to mycobacterial infection in Asian elephants. While TB+ elephants showed Ig proteins cross-reactive to various *Mycobacterium spp.*, the results suggested that TB- elephants developed a protective antibody response against mycobacterial proteins involved in disease-associated mechanisms.

The above finding will contribute to and enhance accurate diagnosis of TB in elephants. As already mentioned, due to lack of accurate diagnostic in (Chapters 1 and 2)the estimated

prevalence might show some discrepancies. In a recent work, urine samples were used for identification of biomarkers for tuberculosis for TB detection among elephants and protein analysis (Manuel et al., 2022). Biomarker urines were experimented for diagnosis of TB and this is easy to collect for carrying out further research.

We expect that markers identified through proteomics in (Chapter 3) will provide new avenues for TB testing in elephants. However, these tools need further verification maximizing the sample numbers, and particularly, number of samples from post-mortem pathology- and culture-confirmed positive and negative individuals.

The evidence generated through risk assessment (Chapter 2) for mahout provides a new direction to control TB and requirement of accurate diagnostic tools. Most of the serological diagnostics methods only detect TB in the late stage of infection, thus we need to develop an early diagnostic method, similar to GeneXpert test (Cepheid, 2011) for human TB or the AI-based method (Qin et al., 2021) for TB detection in elephants.

6.1.4 Whole Genome Sequencing and cross-species transmission

In (Chapter 4), I investigated the dynamic of potential *M. tuberculosis* transmission between elephants and humans in Nepal using phylogenomic analysis of MTB sequences. In this study, a WGS- Whole Genomic Sequencing- approach was applied to genotype *M. tuberculosis* isolates from culture-positive *M. tuberculosis* in three TB infected samples: one human and two from captive elephants. The comprehensive phylogenomic analysis of *M. tuberculosis* suggests possible bidirectional transmission of this pathogen at interface between humans and elephants. The *M. tuberculosis* culture-positive DNA from elephant (PK, DG) and human sputum samples (52G111) were extracted using a DNA-SorB kit. The extracted DNA was then quantified using Qubit (Thermo Fisher Scientific, USA) and diagnosis of *M. tuberculosis* using specific real-time PCR (Sacace Biotechnologies, 2018). *M. tuberculosis* strains were isolated from elephants and humans and whole genome sequencing was done using Nanopore MinION and Illumina MiSeq approach. Tb-Profiler version 4.1.0, a standalone tool (Phelan et al., 2019) was used to detect lineage and drug resistance of *M. tuberculosis* isolates from whole-genome sequences through the SNP alignment data. The phylogenetic analysis was performed with maximum likelihood and Bayesian inference.

The three isolated samples cluster in three different *M. tuberculosis* lineages. The elephant isolates DG and PK cluster to Lineage 1.2.2.2 and Lineage 2.2, respectively, while the human isolates the 52G111 cluster to Lineage 4.1.2.1. The average coverage of all was 99.6% and the genome was fully covered. The DG isolate clustered closely with two other published sequences of elephant-derived isolates sampled in Nepal. These three elephant-derived

sequences clustered together with published human-derived isolates sampled in Nepal and India. Similarly, the sample PK isolate clustered closely with published human-derived isolates sampled in Nepal. The sample 52G111 clustered together with other published human-derived samples from Argentina, Russia, and the UK. The topology of the tree also showed that previously published sequences derived from captive Asiatic elephants in USA and Switzerland belonged to lineage 4 among human-derived isolates in nearby geographical regions. Similarly, an isolate originating from wild African elephants also clustered very closely with an isolate derived from humans, both of which were sampled in South Africa.

In our study, 16 mahouts family having a history of TB. The high prevalence of TB in both humans and elephants could mean that there might be bi-directional infection possibilities. The prevalence of serum antibodies against MTC in the captive elephant population was found to be (20 %) (Chapter 2). Close contact between humans and elephants in countries with a high human TB burden may increase the risk of infection in elephants through reverse zoonotic transmission (Michel et al., 2013). This was verified when it was investigated through MTB spillover between species- whole genome sequence of *M. tuberculosis* transmission between elephants and humans in Nepal using phylogenomic analysis of MTB sequences. The phylogenomic analysis of MTB sequences isolated from two infected elephants clustered into two different lineages, lineage 1 and lineage 2 respectively, whereas human-derived isolates clustered within lineage 4. This is the first study that attempted to investigate using a comprehensive genomics approach to explore possible spillovers of MTB between captive Asiatic elephants and humans. This result suggests that TB infection in elephants might be transmitted by humans, which is supported by the confirmed interaction, sharing, and by living together of humans and captive elephants (Chapter 2). However, this information is still limited because only two elephant MTB isolates were sequenced. In the future, there is a need to analyze more samples to foster the observed evidence.

There are two recently published articles that reported WGS of MTB in captive elephant from Nepal and USA (Brenner et al., 2021; Paudel et al., 2021). However, in our study, in addition to the WGS we also used WGS data from our study and other published study to construct a higher resolution phylogenetic tree of *Mycobacterium tuberculosis* detected in elephants (Chapter 4). This allowed us to characterize the lineage of MTB in elephant and deduce the bi-directional transmission of MTB between human and elephant.

6.1.5 Gut microbiota diversity across human, elephant, livestock, and wild herbivore

Gut microbiota is one of the key factors that shapes the biochemical profile of the diet and thus it impacts the host's health and disease (Rowland et al., 2018). Considering the significant role that gut microbiome plays in immunological functions in animals, (Chapter 5) explore a

comparative gut microbiota analysis of shared dwelling of captive elephants and humans in the same area, Chitwan National Park. Additionally, analyses of gut microbiota of some other co-existing animal species (such as domestic livestock, wild ungulates, and wild elephants) were conducted to understand the microbiome dynamics.

A total of 176 fecal samples were analyzed, originated from humans, domestic livestock, captive elephants, wild ungulates, and a wild elephant. Among the collected fecal samples of wild ungulates, spotted deer, sambar deer, hog deer, rhinos, and wild boars had been included based on species identification by DNA barcoding technique. The gut microbiota metabarcoding analysis was done using the Illumina MiSeq sequencing platform. During analysis, sequence data was processed in the QIIME2 pipeline v2021.11.0 and taxonomy based on SKLEARN. Alpha and Beta diversity analysis result was calculated using QIIME2-based core-diversity plugins. For alpha-significance Kruskal-Wallis pairwise test was performed to assess alpha diversity metrics of Faith's phylogenetic diversity (PD) (Faith, 1992) and Shannon Diversity indices (Shannon, n.d.) was performed. The beta diversity calculated pairwise permutational analysis of variance (PERMANOVA) statistics and principal coordinate analysis (PCoA) plots were generated using the Emperor plugin in Qiime2.

The estimates relative abundance of various bacterial taxa among the host species, namely Firmicutes, Bacteroidota, Proteobacteria, Actinobacteriota, Verrucomicrobiota, Planctomycetota and Patescibacteria as major groups among all the host species. Regarding the microbiota of mahouts, it was mainly dominated by Firmicutes, which plays a major role in human health. Many bacteria belonging to this phylum helps in breaking down carbohydrates in the gut which cannot be digested by the body's enzymes, such as dietary fiber and resistant starch (Kaur et al., 2019). Firmicutes Bacterial presence were dominated in elephants, domestic livestock, hog deer, sambar deer, spotted deer, wild boars followed by Bacteroidota in all species whereas in Rhino, Bacteroidota was highest and followed by Firmicutes. The study detected the presence of bacteria belonging to *Mycobacterium* genus in the fecal microbiota of some domestic as well as wild herbivorous mammals. The ungulate has the highest frequency of *Mycobacterium spp*, spotted deer (n=18; 31%) followed by wild boar (n=16; 6.3%), rhino (n=1; 3.6%), captive elephant (n=2; 5.1%) and sheep (n=1; 20%).

Despite higher exposure (animal, human and environment), gut microbiome of humans (mahouts) have significantly lower Alpha diversity and very discerning Beta diversity compared to other host species in the study area. Based on Faith's Phylogenetic Diversity (PD) analysis, the Alpha diversity of gut microbial communities across different host species had a wide PD range (23-69), with human having relatively smaller value (PD= 23) compared to captive elephant (PD=69), domestic livestock (PD= 60) and wild ungulates (PD=58).

It was observed that captive elephants have significantly highest phylogenetic diversity of microbiome species in their gut community, while in terms of bacterial species richness and evenness, their alpha diversity is of similar level to that of livestock and wild ungulates. Regarding the Shannon index, we observed no significant diversity between Captive elephants vs Domestic livestock, wild ungulates. Whereas in humans we observed with all three groups differed significantly i.e., Human vs Captive elephants, Human vs Domestic livestock, Human vs Wild ungulates. In contrast to this, Bray-Curtis beta diversity showed that the gut microbiome of captive elephants was significantly different from livestock, and wild ungulates as well.

During risk assessment (Chapter 2) it was identified that humans, elephants, wild animals, and livestock sharing each other's food and water in their daily activities are the risk factors for TB transmission to each other (Chapter 5). While investigating the genus level, in the gut microbiome, the presence of bacteria belonging to *Mycobacterium* genus in the fecal microbiota of some domestic as well as wild herbivorous mammals was detected. This might indicate the transmission of pathogen reverse-zoonosis or bidirectional transmission dynamics between humans and animals.

6.1.6 MTB and One Health disease dynamics

TB in captive elephants and their surrounding species, including humans, has been investigated to understand the disease dynamics (reservoir, spillover, and transmission) of TB in captive elephants from a One Health perspective.

Mahouts are TB hosts for elephants and through elephants to other wild animals (Chapter 2). This study highlights the importance of a well-coordinated One Health program involving both animal and human health sectors to deal with, limit, and prevent the spread of MTC in multi-host communities where TB remains at high prevalence, particularly in humans.

The results of Asian elephant serum proteome in response to TB advanced our understanding of elephant immune response to *M. tuberculosis* (Chapter 3). This is one of the better potential diagnostic tools to harmonize all diagnostic in One Health disease surveillance aspect. This will further help early diagnosis of TB among Asian elephants, prevent the spread of TB, and help for conservation and protection from disease aspect to high-risk species.

The study (Chapter 4) attempts to investigate a comprehensive genomics approach to explore possible spillovers of MTB between captive Asiatic elephants and humans. Tuberculosis is known as a re-emerging disease, considered as a reverse zoonosis and possible bidirectional transmission of this pathogen at the interface between humans and elephants. Such observed cases of reverse zoonosis produce a huge effect in global and national arena of elephant conservation. Hence, the findings indicate the necessity of regular screening and detection of such infectious diseases among elephant and human population as crucial in the One Health perspective.

6.1.7 Overview of recent scientific information on elephant TB

Diagnosing MTC infection remains a challenge in humans and in animals, including wildlife (Thomas et al., 2021; Sharma et al., 2022). Recent advances in elephant TB diagnosis include the parallel use of several ELISAs in Asian elephants (Angkwanish et al., 2021), IGRA testing in Asian elephants as a promising complement to serology (Songthammanuphap et al., 2020), and direct PCR-based methods for antigen detection which are reported to perform well in African elephants (Goosen et al., 2020). In captive elephants, i.e., in zoos or in the case of work elephants, regular whole herd serological testing is an advisable preventive measure (Miller et al. 2018). The chimeric antigen ESAT6/CFP10 is immunodominant in both asymptomatic and symptomatic phases, making it particularly useful in the detection of infection (Ishikawa et al., 2022). We used this antigen in this PhD as it is a component of the rapid test DPP test.

Beyond infection diagnosis, using biomarkers in urine or in serum, or using serum proteomics for identification of new diagnostic tools, is an ongoing quest. Recently, (Coughlin et al., 2022) showed that diagnostic biomarkers for pulmonary tuberculosis in humans were not elevated in *M. tuberculosis* culture-positive Asian elephants. In this PhD, using proteomics, I found that the results of the Asian elephant serum proteome in response to TB advanced our understanding of elephant immune response to *M. tuberculosis* (Villar et al., 2022).

Elephants develop *M. tuberculosis* infections far more often than *M. bovis* or *M. caprae* infections. Occasionally, even mixed infections with more than one *M. tuberculosis* lineage can be found (Paudel et al., 2019). This might be a sign of high infection pressure, i.e., a high exposure risk in some elephant populations. In the common case of *M. tuberculosis* infections, most of this risk is likely due to close contact with infected humans (Chapter 2). However, occasional infections with *M. bovis* in African elephants (Miller et al., 2021) or *M. caprae* in Asian ones (Yoshida et al., 2018) have been reported. These cases are more likely linked to infected wild or domestic animals, but are uncommon and, indeed, TB is sometimes diagnosed in susceptible *M. bovis* hosts but not in sympatric elephants (Lekko et al., 2021). The dominance of *M. tuberculosis* among elephants was also confirmed in this PhD (Rajbhandari et al 2023).

Regarding risk factors for elephant TB, a study in Thailand found that seropositivity was linked with older age of the elephants, and that elephants working 7 h per day and the ones in good condition were less likely to test positive in TB serology. There were also protective effects of large camp sizes (31-50 elephants), as compared to elephants in smaller camp sizes, as well as site differences (Angkwanish et al., 2021). In this work, I found that government-owned elephants have higher levels of TB seroprevalence when compared to private-owned

elephants. Mahouts (elephant caretaker) reporting interactions with wild animals, consuming unpasteurized dairy products from their animals, or collecting dead animals to eat or share were associated with higher risk of TB antibody seropositivity in the elephants. (Schmitt et al 2023)

Finally, further research is needed regarding TB control in elephants and at the interface. (Kilburn et al., 2023) reported on the pharmacokinetics of levofloxacin in elephants. This fluoroquinolone is commonly used as part of multiple antibiotic regimens for the treatment of TB. Other recent results suggest that antigen Ag85B has the potential to be a serological marker for the prediction of disease onset and for the evaluation of treatment effectiveness in elephants (Ishikawa et al., 2022)

6.2 Future Perspectives

This study provides information on *M. tuberculosis* complex and its disease dynamics and transmission from one host to another. Also, this thesis brings new insights in bidirectional transmission of this pathogen at the interface between humans and captive elephants. The risk behavior of elephant caretakers and their environmental sharing might enhance TB transmission to wild animals and to the environment. This brings into question the need to proceed with an assessment of TB prevalence not only in captive and wild elephants, but also in humans, livestock, and wild animals. In addition, further studies on TB genome sequencing is highly recommended in order to perform phylogenomic studies for understanding directionality of the infections, among other questions.

Moreover, future research should not be limited just with in humans and elephants and some particular locations. The research areas and its scope should be made broad with comprehensive study of One Health disease dynamics, including other zoonotic diseases. The pieces of literature reviewed during the study have already indicated that there have been cases of TB in rhinos, deers, wild boars, macaques and similar livestock like sheeps, goats, pigs, and water buffaloes in Nepal. There are evidences indicating that water sources are source of pathogens, thus the role of water in TB infection transmission should be further explored.

The application of omics technologies should be used for further advancement in identification of biomolecules with a function in host-pathogen interactions. These biomolecules will provide candidate antigens for development of diagnostics, diagnosis, and control interventions to reduce risks of TB transmission and infection in humans and animals.

6.3 Conclusions

1. Captive Asian elephants from Chitwan national park (CNP) in Nepal are exposed to infection by members of the *Mycobacterium tuberculosis* complex. Using animal side lateral flow tests, we found a 20.2% apparent prevalence of antibodies in blood serum. Two of the 99 study elephants died during this survey, and *M. tuberculosis* was confirmed in one of them.
2. Risk factors for tuberculosis (presence of antibodies) in captive elephants from CNP Risk assessment of MTB transmission revealed several factors associated with elephant husbandry and cross-species contacts that increase the risk of TB infection. These include government-owned elephants, hand-feeding elephants, lack of personal protective equipment, and improper disposal of elephant waste. Close contact between captive and wild elephants, as well as contact with other animals, also poses a risk of transmission.
3. Serum proteomics identified potential biomarkers and antigens that could contribute to the diagnosis, prognosis, and prevention of tuberculosis in elephants. We revealed 26 significantly dysregulated proteins in elephants exposed to MTC as compared to uninfected controls. Among the dysregulated proteins, 38% were identified as immunoglobulins, with 9 overrepresented and 1 underrepresented in TB+ elephants. Dysregulated non-Ig proteins in exposed elephants were associated with cardiovascular diseases, glycogen storage disease, solid malignancies, and inflammation. Overrepresented non-Ig proteins in uninfected control elephants were associated with defense against infection and reduced risk of inflammation.
4. The characterization of isolates from elephants and humans in Nepal using whole-genome sequencing and phylogenomic analysis revealed that the three isolates belonged to different *M. tuberculosis* lineages. The elephant-derived isolates, despite being from the same locality, belonged to different lineages, indicating complex transmission dynamics.
5. The two elephant-derived isolates of *Mycobacterium tuberculosis* clustered with published human-derived isolates from Nepal, suggesting bidirectional transmission between humans and elephants.
6. Firmicutes and Bacteroidota were the dominant phyla of the gut microbiota across human and animal hosts in CNP. Fusobacteriota and Proteobacteria were major phyla in humans, while Verrucomicrobiota and Planctomycetota were prominent in domestic livestock, and Fibrobacterota in captive elephants. Members of the genus *Mycobacterium* were detected in the fecal microbiota of several domestic and wild herbivores.

7. The gut microbiota of captive elephants, domestic livestock, and wild herbivores showed similarities in composition and abundance. Humans had the lowest alpha diversity values, while captive elephants had the highest values. No significant differences in alpha diversity were observed between domestic livestock and wild herbivores, but significant differences were found in all other pair comparisons. Significant differences in beta diversity were observed between all host species groups.
8. This Thesis emphasizes the need for surveillance, screening, and diagnostic strategies to control and prevent the spread of tuberculosis, especially in captive elephants and those in close contact with them.
9. Based on these findings, it is recommended to implement a routine and comprehensive TB screening program for elephants, involving collaboration between public health, veterinary medicine, and occupational health experts. The development of accurate diagnostic methods, based on proteomic and genomic analysis, should be prioritized to improve early detection and control of TB in captive elephants. Additionally, efforts should be made to minimize the risk of TB transmission from humans to elephants through improved hygiene practices and biosecurity measures.
10. This thesis supports that the *M. tuberculosis* complex impacts both human and animal health significantly. The holistic One Health perspective can be deemed to be best for understanding disease dynamics. It is probably the best way to get a comprehensive view on all aspects of disease dynamics, pathogen reservoirs, spillover risk factors, and transmission mechanisms. It is hereby established using epidemiological studies in combination with omics technologies that captive elephants are closely interlinked and interconnected with not only humans (mahouts) but also surrounding domestic and wild animal species. Understanding pathogens and affected hosts is a starting point in designing effective preventive and control interventions.

6.4 References

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Chapter 1: Annex

Annex 1: Sample Collection and Elephant Description

Tuberculosis in Asian Elephant: A one Health Perspective

SN	Site Name	GPS		Animals ID	Sex	Age class	Health Status	Morphometric Measurements (cm)							Morphometric Measurements (cm)				
								WE(cm)	WH(cm)	N(cm)	LB(cm)	LF(cm)	C	HB(cm)	He(cm)	Ha	LE(cm)	T(cm)	G(cm)
1	NTNC,CNP(Hattisar)	N27.57305°	E084.50158°	CE01	M	A	Healthy	79 CM	80 cm	230cm	170cm	180cm	Grey	370cm	277cm	Less Hairs	88cm	165cm	440cm
2	NTNC,CNP(Hattisar)	N27.57305°	E084.50158°	CE02	M	A	Healthy	69cm	64cm	220cm	160cm	160cm	Grey	320cm	258cm	Less Hairs	80cm	100cm	370cm
3	NTNC,CNP(Hattisar)	N27.57305°	E084.50158°	CE03	M	A	Healthy	60cm	60cm	210cm	173cm	176cm	Grey	355cm	274cm	Less Hairs	80cm	170cm	375cm
4	NTNC,CNP(Hattisar)	N27.57305°	E084.50158°	CE04	F	A	Healthy	71cm	80cm	214cm	164cm	168cm	Grey	320cm	263cm	Less Hairs	79cm	146cm	360cm
5	NTNC,CNP(Hattisar)	N27.57305°	E084.50158°	CE05	F	A	Healthy	70cm	78cm	223cm	155cm	164cm	Grey	303cm	238cm	Less Hairs	78cm	138cm	340cm
6	NTNC,CNP(Hattisar)	N27.57305°	E084.50158°	CE06	M	A	Healthy	68cm	75cm	225cm	160cm	183cm	Grey	294cm	278cm	Less Hairs	74cm	143cm	355cm
7	NTNC,CNP(Hattisar)	N27.57305°	E084.50158°	CE07	M	A	Healthy	61cm	73cm	177cm	150cm	163cm	Grey	290cm	242cm	Less Hairs	67cm	146cm	345cm
8	NTNC,CNP(Hattisar)	N27.57305°	E084.50158°	CE08	M	A	Healthy	68cm	80cm	210cm	170cm	175cm	Grey	320cm	275cm	Less Hairs	80cm	144cm	370cm
9	NTNC,CNP(Hattisar)	N27.57305°	E084.50158°	CE09	M	A	Healthy	52cm	65cm	178cm	145cm	148cm	Grey	259cm	205cm	Less Hairs	60cm	136cm	310cm
10	NTNC,CNP(Hattisar)	N27.57305°	E084.50158°	CE10	M	A	Healthy	60cm	70cm	210cm	195cm	200cm	Grey	318cm	260cm	Less Hairs	90cm	160cm	390cm

11	NTNC,CNP(Hattisar)	N27.57305°	E084.50158°	CE11	M	A	Healthy	74cm	80cm	183cm	173cm	198cm	Grey	344cm	290cm	Less Hairs	91cm	54cm	360cm
12	EBC,Khorsar	N27.58192°	E084.46268°	CE12	F	A	Healthy	59cm	89cm	160cm	170cm	180cm	Grey	350cm	235cm	Less Hairs	63cm	130cm	430cm
13	EBC,Khorsar	N27.58192°	E084.46268°	CE13	F	A	Healthy	51cm	90cm	170cm	155cm	170cm	Grey	314cm	252cm	Less Hairs	84cm	135cm	432cm
14	EBC,Khorsar	N27.58192°	E084.46268°	CE14	F	A	Healthy	66cm	92cm	183cm	163cm	165cm	Grey	355cm	225cm	Less Hairs	74cm	92cm	410cm
15	EBC,Khorsar	N27.58192°	E084.46268°	CE15	F	A	Healthy	50cm	70cm	155cm	160cm	160cm	Grey	370cm	220cm	Less Hairs	65cm	120cm	520cm
16	EBC,Khorsar	N27.58192°	E084.46268°	CE16	F	A	Healthy	60cm	105cm	172cm	155cm	155cm	Grey	260cm	218cm	Less Hairs	58cm	140cm	305cm
17	EBC,Khorsar	N27.58192°	E084.46268°	CE17	F	A	Healthy	70cm	90cm	180cm	145cm	170cm	Grey	340cm	245cm	Less Hairs	80cm	140cm	420cm
18	EBC,Khorsar	N27.58192°	E084.46268°	CE18	F	A	Healthy	70cm	95cm	150cm	170cm	160cm	Grey	310cm	260cm	Less Hairs	70cm	120cm	365cm
19	EBC,Khorsar	N27.58192°	E084.46268°	CE19	F	A	Healthy	65cm	80cm	160cm	160cm	170cm	Grey	295cm	235cm	Less Hairs	70cm	145cm	390cm
20	EBC,Khorsar	N27.58192°	E084.46268°	CE20	F	A	Healthy	53cm	80cm	226cm	178cm	182cm	Grey	273cm	280cm	Less Hairs	83cm	130cm	480cm
21	EBC,Khorsar	N27.58192°	E084.46268°	CE21	M	A	Healthy	55cm	75cm	112cm	128cm	130cm	Grey	216cm	197cm	Less Hairs	35cm	120cm	315cm
22	NTNC,CNP	N27.57354°	E084.49707°	CE23	F	A	Healthy	75cm	85cm	205cm	170cm	180cm	Grey	330cm	250cm	Less Hairs	85cm	145cm	385cm
23	NTNC,CNP	N27.57354°	E084.49707°	CE24	F	A	Healthy	73cm	84cm	210cm	180cm	175cm	Grey	328cm	240cm	Less Hairs	90cm	120cm	384cm
24	NTNC,CNP	N27.57354°	E084.49707°	CE25	F	A	Healthy	73cm	83cm	205cm	176cm	174cm	Grey	331cm	255cm	Less Hairs	96cm	142cm	386cm
25	NTNC,CNP	N27.57354°	E084.49707°	CE26	F	A	Healthy	72cm	72cm	205cm	175cm	174cm	Grey	330cm	245cm	Less Hairs	90cm	142cm	385cm

26	NTNC,CNP	N27.57354°	E084.49707°	CE27	F	A	Healthy	74cm	84cm	200cm	175cm	173cm	Grey	330cm	252cm	Less Hairs	86cm	147cm	384cm
27	Kashara	N27.55254°	E084.33529°	CE28	M	A	Healthy	55cm	80cm	180cm	180cm	185cm	Grey	270cm	220cm	Less Hairs	70cm	130cm	355cm
28	Kashara	N27.55254°	E084.33529°	CE29	M	A	Healthy	65cm	75cm	205cm	175cm	180cm	Grey	280cm	250cm	Less Hairs	70cm	135cm	340cm
29	Kashara	N27.55254°	E084.33529°	CE30	F	A	Healthy	60cm	80cm	180cm	180cm	195cm	Grey	325cm	250cm	Less Hairs	80cm	160cm	400cm
30	Kashara	N27.55254°	E084.33529°	CE31	F	A	Healthy	55cm	70cm	185cm	180cm	190cm	Grey	290cm	240cm	Less Hairs	70cm	100cm	380cm
31	Kashara	N27.55254°	E084.33529°	CE32	F	A	Healthy	75cm	87cm	190cm	180cm	180cm	Grey	350cm	260cm	Less Hairs	80cm	135cm	370cm
32	Kashara	N27.55254°	E084.33529°	CE33	F	A	Healthy	75cm	70cm	210cm	170cm	180cm	Grey	345cm	260cm	Less Hairs	70cm	175cm	390cm
33	Bagai Post Madi	N27.41941°	E084.45629°	CE34	M	A	Healthy	60cm	75cm	158cm	150cm	170cm	Grey	300cm	236cm	Less Hairs	69cm	148cm	361cm
34	Bagai Post Madi	N27.41941°	E084.45629°	CE35	F	A	Healthy	70cm	88cm	189cm	160cm	170cm	Grey	295cm	254cm	Less Hairs	81cm	167cm	390cm
35	Khorsar	N27.58192°	E084.46268°	CE36	F	A	Healthy	70cm	90cm	180cm	165cm	180cm	Grey	280cm	215cm	Less Hairs	80cm	145cm	330cm
36	Amritya Post	N27.56092°	E084.57591°	CE37	F	A	Healthy	85cm	87cm	210cm	175cm	165cm	Grey	360cm	271cm	Less Hairs	70cm	160cm	415cm
37	Amritya Post	N27.56092°	E084.57591°	CE38	M	Sub-A	Healthy	50cm	65cm	190cm	150cm	150cm	Grey	360cm	220cm	Less Hairs	57cm	130cm	315cm
38	Pyaridhap Post	N27.57385°	E084.65109°	CE39	F	A	Healthy	70cm	90cm	190cm	175cm	180cm	Grey	310cm	244cm	Less Hairs	80cm	150cm	365cm
39	Pyaridhap Post	N27.57385°	E084.65109°	CE40	F	A	Healthy	65cm	95cm	210cm	195cm	185cm	Grey	315cm	254cm	Less Hairs	80cm	156cm	399cm
40	Janakpur Post	N27.55896°	E084.55832°	CE41	M	A	Healthy	57cm	50cm	179cm	143cm	148cm	Grey	263cm	207cm	Less Hairs	65cm	139cm	321cm

41	Janakpur Post	N27.55896°	E084.55832°	CE42	M	A	Healthy	55cm	58cm	146cm	140cm	142cm	Grey	228cm	210cm	Less Hairs	54cm	136cm	305cm
42				CE43									Grey			Less Hairs			
43	Belsar Post	N27.57266°	E084.40656°	CE44	F	A	Healthy	60cm	70cm	180cm	145cm	160cm	Grey	310cm	255cm	Less Hairs	75cm	150cm	370cm
44	Belsar Post	N27.57266°	E084.40656°	CE45	M	A	Healthy	50cm	70cm	140cm	140cm	150cm	Grey	250cm	220cm	Less Hairs	66cm	130cm	300cm
45	Safari Narayani Hotel	N27.57073°	E084.36718°	CE46	F	A	Healthy	65cm	110cm	190cm	175cm	185cm	Grey	330cm	265cm	Less Hairs	82cm	160cm	410cm
46	Safari Narayani Hotel	N27.57073°	E084.36718°	CE47	F	A	Healthy	75cm	97cm	195cm	168cm	175cm	Grey	340cm	270cm	Less Hairs	80cm	168cm	345cm
47	Safari Narayani Hotel	N27.57073°	E084.36718°	CE48	F	A	Healthy	70cm	95cm	180cm	158c	168cm	Grey	315cm	262cm	Less Hairs	75cm	130cm	395cm
48	Safari Narayani Hotel	N27.57073°	E084.36718°	CE49	F	A	Healthy	70cm	95cm	210cm	165cm	190cm	Grey	340cm	275cm	Less Hairs	75cm	140cm	440cm
49	Kashara Resort	N27.56620°	E084.35123°	CE50	F	A	Healthy	50cm	75cm	200cm	180cm	180cm	Grey	330cm	255cm	Less Hairs	75cm	172cm	370cm
50	Kashara Resort	N27.56620°	E084.35123°	CE51	F	A	Healthy	50cm	85cm	150cm	180cm	175cm	Grey	290cm	250cm	Less Hairs	67cm	130cm	340cm
51	Jungle Villa Resort	N27.55926°	E084.33662°	CE52	F	A	Healthy	65cm	95cm	220cm	165cm	185cm	Grey	325cm	265cm	Less Hairs	75cm	150cm	415cm
52	Jungle Villa Resort	N27.55926°	E084.33662°	CE53	F	A	Healthy	60cm	96cm	190cm	155cm	170cm	Grey	315cm	245cm	Less Hairs	75cm	120cm	410cm
53	Tiger Land Safari	N27.56740°	E084.30211°	CE54	F	A	Healthy	55cm	75cm	220cm	165cm	175cm	Grey	320cm	260cm	Less Hairs	85cm	170cm	365cm
54	Tiger Land Safari	N27.56740°	E084.30211°	CE55	F	A	Healthy	60cm	75cm	165cm	155cm	170cm	Grey	280cm	2258c	Less Hairs	80cm	140cm	355cm
55	Sarang Wildlife Sanctuary	N27.57590°	E084.21676°	CE56	F	A	Healthy	65cm	87cm	180cm	165cm	170cm	Grey	300cm	255cm	Less Hairs	81cm	160cm	355cm

56	Sarang Wildlife Sanctuary	N27.57590°	E084.21676°	CE57	F	A	Healthy	65cm	90cm	190cm	170cm	165cm	Grey	290cm	255cm	Less Hairs	76cm	156cm	390cm
57	Sarang Wildlife Sanctuary	N27.57590°	E084.21676°	CE58	F	A	Healthy	65cm	80cm	180cm	170cm	170cm	Grey	280cm	260cm	Less Hairs	70cm	155cm	355cm
58	CG	N27.57197°	E084.20541°	CE59	F	A	Healthy	55cm	80cm	200cm	170cm	176cm	Grey	340cm	280cm	Less Hairs	85cm	140cm	410cm
59	CG	N27.57197°	E084.20541°	CE60	F	A	Healthy	75cm	90cm	190cm	165cm	175cm	Grey	230cm	255cm	Less Hairs	80cm	150cm	430cm
60	Bamboo Cottage	N27.56951°	E084.20112°	CE61	F	A	Healthy	80cm	95cm	200cm	175cm	170cm	Grey	235cm	275cm	Less Hairs	90cm	160cm	390cm
61	Barahi Resort	N27.56548°	E084.19284°	CE62	F	A	Healthy	70cm	90cm	210cm	175cm	190cm	Grey	330cm	285cm	Less Hairs	81cm	160cm	435cm
62	Barahi Resort	N27.56548°	E084.19284°	CE63	F	A	Healthy	55cm	70cm	140cm	135cm	135cm	Grey	240cm	190cm	Less Hairs	50cm	130cm	285cm
63	Barahi Resort	N27.56548°	E084.19284°	CE64	F	A	Healthy	75cm	70cm	210cm	175cm	180cm	Grey	320cm	265cm	Less Hairs	75cm	140cm	395cm
64	Barahi Resort	N27.56548°	E084.19284°	CE65	F	A	Healthy	55cm	75cm	175cm	160cm	180cm	Grey	260cm	225cm	Less Hairs	75cm	130cm	335cm
65	Divya Puri	N27.65383°	E084.21435°	CE66	M	A	Healthy	75cm	85cm	215cm	170cm	180cm	Grey	330cm	260cm	Less Hairs	80cm	150cm	430cm
66	Divya Puri	N27.65383°	E084.21435°	CE67	M								Grey			Less Hairs			
67	Island	N27.63155°	E084.17107°	CE68	F	A	Healthy	70cm	90cm	190cm	180cm	195cm	Grey	340cm	275cm	Less Hairs	75cm	170cm	415cm
68	Island	N27.63155°	E084.17107°	CE69	F	A	Healthy	80cm	80cm	160cm	150cm	155cm	Grey	300cm	235cm	Less Hairs	70cm	135cm	355cm
69	Island	N27.63155°	E084.17107°	CE70	F	A	Healthy	80cm	75cm	190cm	180cm	185cm	Grey	320cm	265cm	Less Hairs	75cm	150cm	365cm
70	Tiger Tops(Tharu Lodge)	N27.57044°	E084.10016°	CE71	F	A	Healthy	75cm	75cm	180cm	160cm	180cm	Grey	330	260	Less Hairs	80cm	160	370

71	Tiger Tops(Tharu Lodge)	N27.57044°	E084.10016°	CE72	F	A	Healthy	70cm	85cm	175cm	170cm	175cm	Grey	330	250	Less Hairs	75cm	140	380
72	Tiger Tops(Tharu Lodge)	N27.57044°	E084.10016°	CE73	F	A	Healthy	75cm	70cm	200cm	175cm	180cm	Grey	280	250	Less Hairs	85cm	145	360
73	Tiger Tops(Tharu Lodge)	N27.57044°	E084.10016°	CE74	F	A	Healthy	55cm	90cm	200cm	160cm	180cm	Grey	320	265	Less Hairs	75cm	140	390
74	Tiger Tops(Tharu Lodge)	N27.57044°	E084.10016°	CE75	F	A	Healthy	60cm	80cm	155cm	170cm	170cm	Grey	290	230	Less Hairs	70cm	130	360
75	Tiger Tops(Tharu Lodge)	N27.57044°	E084.10016°	CE76	F	A	Healthy	80cm	85cm	180cm	165cm	165cm	Grey	300	240	Less Hairs	70cm	140	360
76	Tiger Tops(Tharu Lodge)	N27.57044°	E084.10016°	CE77	F	A	Healthy	70cm	90cm	200cm	165cm	175cm	Grey	310	245	Less Hairs	80cm	145	375
77	Tiger Tops(Tharu Lodge)	N27.57044°	E084.10016°	CE78	F	A	Healthy	70cm	80cm	190cm	180cm	175cm	Grey	300	265	Less Hairs	90cm	150	340
78	Tiger Tops(Tharu Lodge)	N27.57044°	E084.10016°	CE79	F	A	Healthy	60cm	75cm	200cm	180cm	180cm	Grey	320	260	Less Hairs	85cm	160	370
79	Tiger Tops(Tharu Lodge)	N27.57044°	E084.10016°	CE80	F	A	Healthy	75cm	85cm	195cm	175cm	170cm	Grey	340	255	Less Hairs	95cm	165	410
80	Sukhibar Post	N27.53625°	E084.24999°	CE81	F	A	Healthy	75cm	70cm	185cm	160cm	170cm	Grey	300	255	Less Hairs	65cm	150	330
81	Sukhibar Post	N27.53625°	E084.24999°	CE82	M	A	Healthy	74cm	71cm	175cm	143cm	160cm	Grey	260	225	Less Hairs	64cm	149	340
82	Tiger Tops(Kor Jungle)	N27.54272°	E084.18855°	CE83	F	A	Healthy	70cm	90cm	170cm	165cm	165cm	Grey	310	250	Less Hairs	70cm	160	360
83	Tiger Tops(Kor Jungle)	N27.54272°	E084.18855°	CE84	F	A	Healthy	70cm	90cm	180cm	180cm	180cm	Grey	330	255	Less Hairs	85cm	160	390
84	Tiger Tops(Kor Jungle)	N27.54272°	E084.18855°	CE85	F	A	Healthy	80cm	85cm	175cm	170cm	170cm	Grey	330	260	Less Hairs	77cm	150	370
85	Tiger Tops(Kor Jungle)	N27.54272°	E084.18855°	CE86	F	A	Healthy	80cm	90cm	190cm	180cm	190cm	Grey	340	275	Less Hairs	70cm	150	442

86	Khoriyamuni Post	N27.54853°	E084.14133°	CE87	F	A	Healthy	75cm	80cm	200cm	185cm	175cm	Grey	345	265	Less Hairs	80cm	160	375
87	Khoriyamuni Post	N27.54853°	E084.14133°	CE88	M	A	Healthy	70cm	88cm	185cm	183cm	180cm	Grey	350	270	Less Hairs	65cm	145	380
88	Sauraha	N27.57869°	E084.49517°	CE89	F	A	Healthy	80cm	95cm	190cm	180cm	190cm	Grey	320	260	Less Hairs	75cm	170	410
89	Sauraha	N27.57869°	E084.49517°	CE90	F	A	Healthy	70cm	95cm	180cm	165cm	170cm	Grey	300	260	Less Hairs	85cm	140	400
90	Sauraha	N27.57869°	E084.49517°	CE91	F	A	Healthy	71cm	80cm	180cm	150cm	166cm	Grey	300	246	Less Hairs	72cm	150	360
91	Sauraha	N27.57869°	E084.49517°	CE92	F	A	Healthy	82cm	95cm	190cm	180cm	180cm	Grey	320	255	Less Hairs	85cm	130	410
92	Sauraha(Bagmara)	N27.59361°	E084.48826°	CE93	F	A	Healthy	80cm	70cm	170cm	170cm	180cm	Grey	290	255	Less Hairs	70cm	160	380
93	Sauraha(Bagmara)	N27.59361°	E084.48826°	CE94	F	A	Healthy	70cm	80cm	160cm	160c	170cm	Grey	300	260	Less Hairs	80cm	150	375
94	Sauraha(Bagmara)	N27.59361°	E084.48826°	CE95	F	A	Healthy	75cm	70cm	170cm	160c	170cm	Grey	310	220	Less Hairs	80cm	140	415
95	Sauraha(Bagmara)	N27.59361°	E084.48826°	CE96	F	A	Healthy	80cm	80cm	190cm	170cm	170cm	Grey	290	230	Less Hairs	70cm	150	400
96	Sauraha	N27.57896°	E084.49537°	CE97	F	A	Healthy	70cm	75cm	180cm	170cm	170cm	Grey	300	240	Less Hairs	60cm	140	380
97	Sauraha	N27.57896°	E084.49537°	CE98	F	A	Healthy	60cm	80cm	190cm	185cm	190cm	Grey	310	240	Less Hairs	70cm	160	370
98	Sauraha	N27.57658°	E084.49726°	CE99	F	A	Healthy	75cm	70cm	195cm	175cm	180cm	Grey	320	245	Less Hairs	80cm	150	385
99	Sauraha	N27.57685°	E084.49928°	CE100	F	A	Healthy	65cm	80cm	200cm	160cm	170cm	Grey	350	265	Less Hairs	85cm	160	430

[Note : A: Adult ,ES:Ear sharpe, WE:Width of one Ear, WH: Width of Head, N:Length of Nose, LB:Length of leg Back, LF:Length of leg fornt, C: Coloration(Colour),HB: Head body, He:Height, Ha: Hairer,Less hair,LE: Length of Ear, T: Length of Tail, G: Girth .

Chapter 2: Annex Supplementary Information

2.1 Interview form in English and Nepali

QUESTIONNAIRE

SOCIO-DEMOGRAPHIC DETAILS

Q. N.	Questions and Filters	Coding Categories	Skip to
1.	How old are you?	Age: (Write the completed years)	
2.	Where do you live?	Village/Town/City _____ Province/State _____ Latitude _____ Longitude _____	
3.	What is your caste? (Specify Ethnic Group/Caste)	Ethnicity/Caste _____ Code No: _____ (Specify)	
4.	What is your educational status? Code: (Circle '00' if illiterate, '19' for the literate without attending the school, and write exact number of the completed grade)	Illiterate..... 00 Literate19 SLC Passed.....13 Grade: (Write the completed grade) Intermediate passed..... 14 Graduated and above.....15 	
5.	What is your present marital status?	Married1 Divorced/Permanently Separated.....2 Widow3 Never married4 Code No:	
6.	What is your monthly family income?	<10,000.....1 10,000-20,000.....2 21,000-30,000.....3 31,000-40,000.....4 40,000-50,000.....5 >50,000.....6 Code No:	
7.	What is your main occupation?	_____	

8.	How many members are there in your family who share a house or room with you currently?	_____	
9.	What are the signs and symptoms seen in a person before being diagnosed with TB?	_____ _____ _____	
10.	Is anyone in your family suffering from Tuberculosis?	Yes.....1 No.....2 Code No:	→ Q.11
10.1.	If yes, is he/she getting treatment? If yes, for how long have they been receiving treatment?	Yes.....1 No.....2 Code No: Duration of treatment: _____	
10.2	What level of interaction does your family member with TB have with the elephant(s)? <u>Close</u> = daily direct interaction with chance of contact with body fluids such as saliva, urine, feces, blood <u>Moderate</u> = Occasional direct interactions with chance of contact with body fluids <u>Limited</u> = Rare direct interactions with chance of contact with body fluids	Close interaction1 Moderate interaction.....2 Limited interaction3 No interaction4 Code No:	
11.	Do you have a history of Tuberculosis?	Yes.....1 No.....2 Code No:	
11.1.	If yes, did you take Anti-TB drugs?	Yes.....1 No.....2 Code No:	→ Q.12
11.2.	For how many months did you take your medicine?	_____	
12.	Do you share a room with your friends?	Yes.....1 No.....2 Code No:	→ Q.13

12.1	If yes, how many friends live together in that room?	Two.....1 Three.....2 Four.....3 Five.....4 More than Five.....5 Others.....96 Code No:	
13.	Do you hear or see that any of your friends is suffering from Tuberculosis or taking Anti-TB drugs?	Yes.....1 No.....2 Code No:	 Q.14
13.1	If yes how long have they been taking Anti-TB drugs?		
14.	Do you know what animals can be infected with tuberculosis?	Cattle1 Buffaloes2 Elephants3 Goats4 None5 Other96 Code No:	
15.	What are the signs and symptoms of tuberculosis in animals?		

MALARIA

Q. N.	Questions and Filters	Coding Categories	Skip to
16	Have you ever been diagnosed with Malaria?	Yes.....1 No.....2 Code No:	→ Q.17
16.1	If yes, did you take any anti-malaria drugs?	Yes.....1 No.....2 Code No:	
16.2	For how many months did you take your medicine?	_____	

RISK FACTORS

i) Elephant

Q. N.	Questions and Filters	Coding Categories	Skip to
17.	How many elephants do you take care of?	One.....1 Two-Three.....2 Four-Five.....3 More than Five.....4 Other _____ _____ 96 Code No:	
18.	In a day, how much time do you spend taking care of the elephant(s)?	<1 hrs.....1 1-2 hrs.....2 3-5 hrs.....3 >5 hrs.....4 Others (specify) _____ 96 Code No:	
19.	How often do you bath the elephant?	Daily.....1 Once a week.....2 Twice a week.....3 Thrice a week.....4 Other _____ 96 Code No:	

20.	How long have you been taking care of elephant(s)? Select one option.	<1 year.....1 1-2 year.....2 3-4 year.....3 5 years.....4 >5 years.....5 Code No:																			
21.	How do you feed the elephant?	By using your hand.....1 By using a bucket.....2 Other _____ 96 Code No:																			
22.	How closely do you interact with the elephant? <u>Close</u> = daily direct interaction with chance of contact with body fluids such as saliva, urine, feces, blood <u>Moderate</u> = Occasional direct interactions with chance of contact with body fluids <u>Limited</u> = Rare direct interactions with	Close interaction1 Moderate interaction2 Limited interaction3 Other _____ 96 Code No:																			
23.	Do Wild elephant come to interact with Captive elephant?	Yes.....1 No.....2 Code No:	→ Q.24																		
23.1	If yes, how often does the interaction occurs?	Once a week.....1 Twice a month.....2 Once in six month.....3 Other _____ 96 6																			
23.2	Where does the interaction mostly occur?	 _____																			
24.	Do other wild animals come to interact with the captive elephant?	Yes.....1 No.....2 Code No:	→ Q.25																		
	Wild animals	Number of Interaction with captive elephants <table border="1"> <tr> <td>Week</td> <td>Last 15 days</td> <td>Months</td> </tr> <tr> <td>Rhino</td> <td></td> <td></td> </tr> <tr> <td>Tiger</td> <td></td> <td></td> </tr> <tr> <td>Wild Boar</td> <td></td> <td></td> </tr> <tr> <td>Deer</td> <td></td> <td></td> </tr> <tr> <td>Others</td> <td></td> <td></td> </tr> </table>	Week	Last 15 days	Months	Rhino			Tiger			Wild Boar			Deer			Others			Type of Interaction (direct, indirect, shared resources)
Week	Last 15 days	Months																			
Rhino																					
Tiger																					
Wild Boar																					
Deer																					
Others																					

25.	How often are the elephant's enclosures cleaned?	Select one option: Daily.....1 Weekly.....2 Monthly.....3 As needed.....4 Never.....5 Code No:	
26.	How do you clean the elephant's houses?	Remove dung.....1 Clean floor with water.....2 Use Disinfectants.....3 Code No:	
27.	Do you use disinfectant to clean? What is the name of the disinfectant?	Yes.....1 No.....2 Code No: Name: _____	→ Q. 28
27.1	If yes, do you always use disinfectants to clean the following:	Select all that apply: Animal enclosures.....1 Food bins.....2 Counter tops.....3 Slaughtering/butchering equipment.....4 Hands.....5 Special protective equipment.....6 Floors.....7	
27.2	How frequently do you use the disinfectant?	Select one option: Daily.....1 Weekly.....2 Monthly.....3 As needed.....4 Never.....5 Code No:	
28.	What are the things you do after working/ cleaning in farm?	Change clothing1 Clean hand and leg with regular water...2 Clean hand and leg with soap water.....3 Use disinfectants.....4 Don't do anything.....5 Code No:	
29.	Do you have any special protective equipment (example: shoes, masks, gloves) only worn at work?	Yes.....1 No.....2 Code:	→ Q.30

29.1	If yes, which protective equipment do you use?	Select all that apply. Shoes.....1 Mask.....2 Clothes.....3 Gloves.....4 Gown/Apron.....5 Code No: ☐	
30.	How do you dispose of elephant waste?	Throw in pit.....1 Bio gas.....2 Direct disposal in farmland.....3 Composting.....4 Others.....5 Code No: ☐	
31.	Do you participate in elephant trunk washes?	Yes.....1 No.....2 Code No: ☐ ☐	
32.	Does the elephant(s) receive regular (annual or more frequently) health checkup/veterinary care?	Yes.....1 No.....2 Code No: ☐	
33.	Has an animal health official ever inspected your animals?	Yes.....1 No.....2 Code No: ☐	
34.	What do you do when an animal gets sick?	Select all that apply. Kill the animal and dispose of the carcass.1 Kill the animal and sell it.....2 Sell the live animal for regular price.....3 Sell the live animal for discounted price...4 Nothing different.....5 Get veterinary care.....6 Report to authorities.....7 Other _____ 96 Code No: ☐ ☐	
34.1	Whether an animal is sick or healthy are any supplements or medicines given the elephant?	Yes.....1 No.....2 Code No: ☐ ☐	
35.	Have you heard about Tuberculosis in Elephants?	Yes.....1 No.....2 Code No: ☐	

36.	Have the elephants here, been diagnosed with Tuberculosis?	Yes.....1 No.....2 Code No:	 Q.37
36.1	If yes, has the elephant received treatment?	Yes.....1 No.....2 Code No:	
36.2	What do you think was the source of infection of TB to the elephant?	_____ _____	
36.3	Does the TB infected elephant reside in the same shelter as your cattle?	Yes1 No2 Code No:	
36.4	If No, where do they reside and how far between elephants and cattle?	_____ -	

ii) Cattle

Q. N.	Questions and Filters	Coding Categories	Skip to
37.	Do you have any livestock animals present in your household?	Yes.....1 No.....2 Code No:	 Q.38
37.1.	Name of the livestock that you own; (Multiple answers)	Sheep.....1 Cow.....2 Buffalo.....3 Goat.....4 Other _____ 96 Code No:	
37.2.	How are animals reared in your farm?	Stall fed.....1 Free range.....2 Both.....3 Code No:	
37.3	What are the sources of water for animals in your farm?	Tap water.....1 Well water.....2 Pond water.....3 River water.....4 Code No:	

38.	Do you share water source with animals (elephant/livestock) for washing?	Yes.....1 No.....2 Code No: ☐	
39.	Have you seen animal feces in or near food before you have eaten it?	Yes.....1 No.....2 Code No: ☐	
40.	Have you slaughtered an animal?	Yes.....1 No.....2 Code No: ☐	
40.1	What animal was slaughtered and when?	_____ -	
41.	Have you eaten an animal that you knew was not well or was sick?	Yes.....1 No.....2 Code No: ☐	
41.1	What was animal sick with, and when was it eaten?	_____	
42.	Is there a designated area for rubbish, including animal waste from slaughter/butcher and animal excrement?	Yes.....1 No.....2 Code No: ☐	
42.1	How often are the animal enclosures cleaned?	Select one option: Daily.....1 Weekly.....2 Monthly.....3 As needed.....4 Never.....5 Code No: ☐	
43.	Do you use disinfectant to clean?	Yes.....1 No.....2 Code No: ☐	→ Q.44.1
43.1	If yes, do you always use disinfectants to clean the following:	Select all that apply: Animal enclosures.....1 Food bins.....2 Counter tops.....3 Slaughtering/butchering equipment.....4 Hands.....5 Special protective equipment.....6 Floors.....7 Code No: ☐	

44.	Does your livestock receive regular (annual or more frequently) health checkup/veterinary care?	Yes.....1 No.....2 Code No:																																						
45.	Has the animal health official ever inspected your livestock?	Yes.....1 No.....2 Code No:																																						
46.	What do you do when an animal gets sick?	Select all that apply. Kill the animal and dispose of the carcass.1 Kill the animal and sell it.....2 Sell the live animal for discounted price...3 Nothing different.....4 Get veterinary care.....5 Report to authorities.....6 Other _____ 96 Code No:																																						
47.	Do captive elephant come to interact with cattle?	Yes.....1 No.....2 Code No:	→ Q.48																																					
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Goat																																								
Others																																								
48.	Do wild elephant come to interact with cattle?	Yes.....1 No.....2 Code No:	→																																					
48.1	<table border="1"> <thead> <tr> <th rowspan="2">Livestock</th> <th colspan="3">Number of Interaction with Wild Elephant</th> <th rowspan="2">Type of Interaction (direct, indirect, shared resources)</th> </tr> <tr> <th>Week</th> <th>Last 15 days</th> <th>Month</th> </tr> </thead> <tbody> <tr><td>Cow</td><td></td><td></td><td></td><td></td></tr> <tr><td>Sheep</td><td></td><td></td><td></td><td></td></tr> <tr><td>Wild Buffalo</td><td></td><td></td><td></td><td></td></tr> <tr><td>Pig</td><td></td><td></td><td></td><td></td></tr> <tr><td>Goat</td><td></td><td></td><td></td><td></td></tr> <tr><td>Others</td><td></td><td></td><td></td><td></td></tr> </tbody> </table>	Livestock	Number of Interaction with Wild Elephant			Type of Interaction (direct, indirect, shared resources)	Week	Last 15 days	Month	Cow					Sheep					Wild Buffalo					Pig					Goat					Others					
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Wild Buffalo																																								
Pig																																								
Goat																																								
Others																																								

49.	Do the livestock interact with you?	Yes 1 No 2	→
		Code No:	

Livestock	Number of Interaction with Maute			Type of Interaction
	Week	Last 15 days	Months	
Cow				
Sheep				
Wild Buffalo				
Pig				
Goat				
Others				

iii) Wildlife

Q. N.	Questions and Filters	Coding Categories	Skip to
48.	Do you (mahout) have any interaction with wild animals?	Yes 1 No 2	→ Q. 49
		Code No:	
48.1	If yes, what are the major wild animals which you interact with?	- _____ -	
48.2	How often do these interactions occur?	Once a week.....1 Twice a month.....2 Once in six months.....3 Other _____ 9 6	
		Code No:	
49.	Have any wild animals lived as a pet in or near your dwelling?	Yes.....1 No.....2	
		Code No:	
50.	Have you eaten food after an animal has touched or damaged it?	Yes.....1 No.....2	
		Code No:	
51.	Do you consume any dairy products from your animals which has not been pasteurized?	Yes.....1 No2	
		Code No:	

52.	Do any animals come inside the dwelling where you live?	Yes.....1 No.....2 Code No: ☐	
53.	Have you found a dead animal and collected it to eat or share?	Yes.....1 No.....2 Code No: ☐	
54.	Have you been scratched or bitten by an animal?	Yes.....1 No.....2 Code No: ☐ When: _____	
55.	The last time you were scratched, bitten, or cut yourself while butchering or slaughtering, what did you do? [multiple responses allowed]	Let someone else take over.....1 Wash wound with soap and water.....2 Rinse wound with water.....3 Bandage wound.....4 Visit Doctor.....5 Nothing- kept working.....6 Never butcher or slaughter.....7 N/A.....8 Code No: ☐	

2.2 Supplementary file #2:

Supplementary file #2: Tables with detailed responses

Table S1: Interviewee knowledge on signs of human and animal tuberculosis.

Characteristics	Number (N=223)	Percent
Knowledge about sign and symptoms of TB on human		
Cough>2 weeks	87	39.0
Hemoptysis	87	39.0
Chest pain	61	27.4
Fever	88	39.5
Body weight loss	27	12.1
Night sweat	5	2.2
Fatigue	17	7.6
Pneumonia	2	.9
Wound	1	.4
Don't know	52	23.3
Know about TB infection on animals		
Cattle	13	5.8
Buffalo	16	7.2
Elephant	203	91.0
Goats	7	3.1
None	11	4.9
Others	8	3.6
Knowledge about sign and symptoms of TB on animals		
Cough>2 weeks	22	9.9
Hemoptysis	12	5.4
Chest pain	12	5.4
Fever	16	7.2
Body weight loss	10	4.5
Fatigue	12	5.4
Wound	36	16.1
Black out	4	1.8
Only heard	142	63.7

Table S2: Tuberculosis history of family member/mahouts and related risk behavior.

Characteristics	Number (N=223)	Percent
History of TB in your family		
Yes	16	7.2
No	207	92.8
If yes, is he/she getting treatment (n=16)		
Yes	14	87.5
No	2	12.5
Level of interaction of your family member with TB have with the elephant (n=14)		
Close interaction	3	21.4
Moderate interaction	1	7.1
Limited interaction	1	7.1
No interaction	9	64.3
History of Tuberculosis		
Yes	3	1.3
No	220	98.7
If yes, taken Anti-TB drugs (n=3)		
Yes	2	66.7
No	1	33.3

Table S3: Care of elephant by timing, day frequency and duration.

Characteristics	Number (N=223)	Percent
Time spends taking care of the elephant(s) per day		
<5 hrs	10	4.5
≥5 hrs	213	95.5
Bathing of the elephant		
Daily	198	88.8
Once a week	13	5.8
Twice a week	2	.9
Thrice a week	2	.9
Less often	8	3.6
Duration of you been taking care of elephant(s)		
<1 year	15	6.7
1-2 year	34	15.2
3-4 year	22	9.9
5 years	17	7.6
>5 years	135	60.5

Table S4: Interaction with elephants.

Characteristics	Number (N=223)	Percent
Type of interaction with the elephant		
Close interaction	208	93.3
Moderate interaction	8	3.6
Limited interaction	4	1.8
Other	3	1.3
Feeding of elephant		
By using your hand	197	88.3
By using a bucket	10	4.5
On floor	16	7.2
Interaction of Wild elephant with Captive elephant		
Yes	151	67.7
No	72	32.3
If yes, how often does the interaction occurs (n=151)		
Frequently in a week	49	32.5
Twice a month	22	14.6
Once in six month	53	35.1
Seasonable	27	17.9
Place for interaction		
Hattisar	54	35.8
Hotel/Safari	51	33.8
Post /Jungle	46	30.5

Table S5: Elephant enclosure cleaning and disinfection.

Characteristics	Number (N=223)	Percent
Cleaning of elephant's enclosures		
Daily	210	94.2
Weekly	7	3.1
As needed	5	2.2
Never	1	.4
Cleaning of the elephant's houses		
Remove dung	214	96.0
Clean floor with water	9	4.0
Use of disinfection to clean elephants' enclosures		
Yes	44	19.7
No	179	80.3
Name of the disinfectant (n=44)		
Korsylon	44	100.0
Lime	35	79.5
Phonel	3	6.8
Potash	3	6.8
Hands	1	2.3
Floors	19	43.2
Frequency of using disinfectant (n=44)		
Weekly	5	11.4
Monthly	8	18.2
As needed	31	70.5
Dispose of elephant waste		
Throw in pit	122	54.7
Bio gas	3	1.3
Composting	3	1.3
Open Place	95	42.6
Cleanliness behavior after working farm		
Change clothing	67	30.0
Clean hand and leg with regular water	135	60.5
Clean hand and leg with soap water	166	74.4
Use disinfectants	14	6.3
Don't do anything	2	.9

Table S6: Use of protective equipment during elephant interaction.

Characteristics	Number (N=223)	Percent
Use of any special protective equipment (example: shoes, masks, gloves) only worn at work		
Yes	59	26.5
No	164	73.5
Protective equipment used		
Shoes	54	91.5
Mask	12	20.3
Clothes	17	28.8
Glove	31	52.5
Apron	2	3.4

Table S7: Knowledge on tuberculosis in elephants.

Characteristics	Number (N=223)	Percent
Heard about Tuberculosis in Elephants		
Yes	190	85.2
No	33	14.8
Diagnosed any elephants with TB		
Yes	45	20.2
No	178	79.8
If yes, elephant received treatment (n=45)		
Yes	42	93.3
No	3	6.7
Source of infection of TB to the elephant (n=45)		
Weakness	3	6.7
No idea	1	2.2
Diet, Food	8	17.8
Over Worked	1	2.2
Water	1	2.2
Air	1	2.2
Decrease in food Supply	4	8.9
Dirty water	1	2.2
Livestock, Animals	1	2.2
Self	1	2.2
Human Transmission	5	11.1
Don't know	18	40.0
TB infected elephant reside in the same shelter as your cattle (n=45)		
Yes	10	22.2
No	35	77.8
If no, where do they reside and how far between elephants and cattle (n=35)		
Up to 50 meters	12	34.3
More than 50 meters	23	65.7

Table S8: Caring for sick elephants.

Characteristics	Number (N=223)	Percent
Care of sick animals		
Kill the animal and dispose of the Carcass	1	.4
Kill the animal and sell it	4	1.8
Sell the live animal for regular price	1	.4
Sell the live animal for discounted price	3	1.3
Nothing different	2	.9
Get veterinary care	173	77.6
Report to authorities	118	52.9
Other	5	2.2
Any supplements or medicines given to the sick elephant		
Yes	184	82.5
No	39	17.5

Table S9: Behavior of mahouts with wild animals.

Characteristics	Number (N=223)	Percent
Any wild animals lived as a pet in or near your dwelling		
Yes	41	18.4
No	182	81.6
Eaten food after an animal has touched or damaged it		
Yes	69	30.9
No	154	69.1
Consume any dairy products from your animals which has not been pasteurized		
Yes	88	39.5
No	135	60.5
Animals come inside the dwelling where you live		
Yes	132	59.2
No	91	40.8
Found a dead animal and collected it to eat or share		
Yes	30	13.5
No	193	86.5

Table S10: Wild animal attacks and first aid measures.

Characteristics	Number (N=223)	Percent
Have you been scratched or bitten by an animal		
Yes	52	23.3
No	171	76.7
If yes, when you been scratched or bitten by an animal (n=52)		
Within one year	24	47.1
Before one year	27	52.9
First aid measure taken after being scratched or bitten by wild animal (n=52)		
Wash wound with soap and water	14	27.5
Rinse wound with water	11	21.6
Bandage wound	7	13.7
Visit Doctor	14	27.5
Nothing- kept working	7	13.7
N/A	7	13.7

Table S11: Livestock enclosure cleaning and use of disinfection.

Characteristics	Number (N=111)	Percent
Share water source with animals (elephant/livestock) for washing		
Yes	69	62.2
No	42	37.8
Frequency of cleaning animal enclosures		
Daily	101	91.0
Weekly	7	6.3
Monthly	3	2.7
Seen animal feces in or near food before you have eaten it		
Yes	52	46.8
No	59	53.2
Use disinfectant to clean		
Yes	8	7.2
No	103	92.8
Use disinfection to clean enclosures of livestock animal (n=8)		
Animal enclosures	6	75.0
Food bins	1	12.5
Slaughtering/butchering equipment	1	12.5
Hands	3	37.5
Floors	3	37.5

Table S12: Health care for livestock.

Characteristics	Number (N=111)	Percent
Received livestock receive regular health checkup/veterinary care		
Yes	78	70.3
No	33	29.7
Animal health official ever inspected your livestock		
Yes	74	66.7
No	37	33.3
Care of sick livestock animal		
Kill the animal and dispose of the carcass	1	.9
Kill the animal and sell it	3	2.7
Sell the live animal for regular price	1	.9
Sell the live animal for discounted price	6	5.4
Nothing different	6	5.4
Get veterinary care	102	91.9
Report to authorities	2	1.8

Chapter 3: Annex

3.1 Supplementary Information

Additional file 1. Serum Proteomics analysis

Additional file 2. Analysis of immunoglobulin proteins differentially represented in TB+ elephants when compared to TB- animals.

Additional file 3. Gene Ontology (GO) annotations for non-immunoglobulin proteins.

Additional file 4. Most significant pathways identified by Reactome analysis of nonimmunoglobulin proteins.

Additional file 5. Protein sequence alignment between *M. tuberculosis*

P9WNK5|ESXB_MYCTU ESAT-6-like protein EsxB and identified mycobacterial proteins with predicted antigen binding regions to immunoglobulin proteins.

All above datas are available in the given link below

Link <https://pubmed.ncbi.nlm.nih.gov/36145440/>

Chapter 4: Annex Supplementary information

Table S1: Detailed information about the 94 whole genomes retrieved from GenBank and used in this study.

S. N.	Accession	Country	Region	Year	Lineage	Sub-lineage	Reference	Host	Sequence ID
1	ERR124640	Malawi	Malawi	NA	1	1.1	Guerra-Assunção_eLife_2015	Human	ERR124640_Malawi_1-1
2	ERR234236	India	South Asia	NA	1	1.1	Comas_NatGenet_2013	Human	ERR234236_India_1-1
3	ERR234165	Tanzania	East Africa	NA	1	1.1	Comas_NatGenet_2013	Human	ERR234165_Tanzania_1-1
4	ERR234205	Ghana	East Africa	NA	1	1.1	Comas_NatGenet_2013	Human	ERR234205_Ghana_1-1
5	ERR2851366	Lebanon	Eurasia	2016	1	1.2.1	NA	Human	ERR2851366_Lebanon_1-2-1
6	ERR718406	Thailand	South East Asia	2003	1	1.2.1	NA	Human	ERR718406_Thailand_1-2-1
7	SRR12006508	Malaysia	South East Asia	2016	1	1.2.1	NA	Human	SRR12006508_Malaysia_1-2-1
8	SRR3724967	Tanzania	Africa	2013	1	1.2.1	NA	Human	SRR3724967_Tanzania_1-2-1
9	SRR5341272	India	South Asia	2015	1	1.2.1	NA	Human	SRR5341272_India_1-2-1
10	SRR6480442	Indonesia	South East Asia	2014	1	1.2.1	NA	Human	SRR6480442_Indonesia_1-2-1
11	ERR126605	Malawi	Africa	2007	1	1.2.2	NA	Human	ERR126605_Malawi_1-2-2
12	ERR1633852	South Africa	Africa	2010	1	1.2.2	NA	Human	ERR1633852_South Africa_1-2-2
13	ERR4423736	Botswana	Africa	2013	1	1.2.2	NA	Human	ERR4423736_Botswana_1-2-2
14	SRR1173597	India	South Asia	2001	1	1.2.2	NA	Human	SRR1173597_India_1-2-2
15	SRR12416839	Indonesia	South East Asia	2017	1	1.2.2	NA	Human	SRR12416839_Indonesia_1-2-2
16	SRR5341274	India	South Asia	2015	1	1.2.2	NA	Human	SRR5341274_India_1-2-2
17	SRR5535929	Botswana	Africa	2013	1	1.2.2	NA	Human	SRR5535929_Botswana_1-2-2
18	SRR6480514	Indonesia	South East Asia	2015	1	1.2.2	NA	Human	SRR6480514_Indonesia_1-2-2
19	ERR176606	Malawi	Malawi	NA	2	2.2.1	Guerra-Assunção	Human	ERR176606_Malawi_2-2-1

							_eLife_2015		
20	ERR550957	Zimbabwe	Eastern Africa	NA	2	2.2.1	Merker_NatGenet_2015	Human	ERR550957_Zimbabwe_2-2-1
21	ERR553277	Vietnam	Vietnam	NA	2	2.2.1	Merker_NatGenet_2015	Human	ERR553277_Vietnam_2-2-1
22	ERR553139	Uzbekistan	Central Asia	NA	2	2.2.1	Merker_NatGenet_2015	Human	ERR553139_Uzbekistan_2-2-1
23	ERR551201	Turkmenistan	Central Asia	NA	2	2.2.1	Merker_NatGenet_2015	Human	ERR551201_Turkmenistan_2-2-1
24	ERR553274	Thailand	South-Eastern Asia	NA	2	2.2.1	Merker_NatGenet_2015	Human	ERR553274_Thailand_2-2-1
25	ERR551079	Swaziland	Southern Africa	NA	2	2.2.1	Merker_NatGenet_2015	Human	ERR551079_Swaziland_2-2-1
26	ERR551979	Swaziland	Southern Africa	NA	2	2.2.1	Merker_NatGenet_2015	Human	ERR551979_Swaziland_2-2-1
27	ERR551549	Germany	Northern Europe	NA	2	2.2.1	Merker_NatGenet_2015	Human	ERR551549_Germany_2-2-1
28	ERR552912	Vietnam	Vietnam	NA	2	2.2.2	Merker_NatGenet_2015	Human	ERR552912_Vietnam_2-2-2
29	ERR552760	Turkmenistan	Central Asia	NA	2	2.2.2	Merker_NatGenet_2015	Human	ERR552760_Turkmenistan_2-2-2
30	ERR552177	South Korea	Eastern Asia	NA	2	2.2.2	Merker_NatGenet_2015	Human	ERR552177_South Korea_2-2-2
31	ERR551184	Germany	Northern Europe	NA	2	2.2.2	Merker_NatGenet_2015	Human	ERR551184_Germany_2-2-2
32	SRR671804	China	China	NA	2	2.2.2	Zhang_NatGenet_2013	Human	SRR671804_China_2-2-2
33	ERR234189	Germany	Europe	NA	3	3	Comas et al. 2013	Human	ERR234189_Germany_3
34	ERR234219	USA	NA	NA	3	3	Comas et al. 2013	Human	ERR234219_USA_3
35	ERR234273	UK	Northern Europe	NA	3	3	Comas et al. 2013	Human	ERR234273_UK_3
36	ERR047885	UK	Northern Europe	NA	4	4.1	Casali_NatGenet_2014	Human	ERR047885_UK_4-1

37	ERR067670	Russia	Russia	NA	4	4.1	Casali_NatGenet_2014	Human	ERR067670_Russia_4-1
38	ERR757160	Argentina	Argentina	NA	4	4.1	Eldholm_NatComms_2015	Human	ERR757160_Argentina_4-1
39	ERR234693	Russia	Russia	NA	4	4.2	Casali_NatGenet_2014	Human	ERR234693_Russia_4-2
40	ERR221541	Malawi	Malawi	NA	4	4.2	Guerra-Assunção_eLife_2015	Human	ERR221541_Malawi_4-2
41	SRR671782	China	China	NA	4	4.2	Zhang_NatGenet_2013	Human	SRR671782_China_4-2
42	SRR6480349	Indonesia	South east Asia	2014	4	4.3	NA	Human	SRR6480349_Indonesia_4-3
43	SRR671874	China	China	NA	4	4.5	Zhang_NatGenet_2013	Human	SRR671874_China_4-5
44	ERR182018	Malawi	Malawi	NA	4	4.6	Guerra-Assunção_eLife_2015	Human	ERR182018_Malawi_4-6
45	ERR182025	Malawi	Malawi	NA	4	4.6	Guerra-Assunção_eLife_2015	Human	ERR182025_Malawi_4-6
46	ERR133905	Russia	Russia	NA	4	4.7	Casali_NatGenet_2014	Human	ERR133905_Russia_4-7
47	ERR182048	Malawi	Malawi	NA	4	4.7	Guerra-Assunção_eLife_2015	Human	ERR182048_Malawi_4-7
48	ERR047886	UK	Northern Europe	NA	4	4.8	Casali_NatGenet_2014	Human	ERR047886_UK_4-8
49	ERR067666	Russia	Russia	NA	4	4.8	Casali_NatGenet_2014	Human	ERR067666_Russia_4-8
50	ERR161093	Malawi	Malawi	NA	4	4.9	Guerra-Assunção_eLife_2015	Human	ERR161093_Malawi_4-9
51	ERR161142	Malawi	Malawi	NA	4	4.9	Guerra-Assunção_eLife_2015	Human	ERR161142_Malawi_4-9
52	ERR257891	Netherlands	Europe	1994	4	4.3.1	NA	Human	ERR257891_Netherlands_4-3-1
53	SRR5067407	Vietnam	South east Asia	2010	4	4.3.1	NA	Human	SRR5067407_Vietnam_4-3-1

54	ERR2652970	Canada	North America	2000	4	4.3.2	NA	Human	ERR2652970_Canada_4-3-2
55	ERR275195	Portugal	Europe	2009	4	4.3.2	NA	Human	ERR275195_Portugal_4-3-2
56	ERR751561	Peru	South America	2011	4	4.3.2	NA	Human	ERR751561_Peru_4-3-2
57	ERR776459	Argentina	South America	2005	4	4.3.2	NA	Human	ERR776459_Argentina_4-3-2
58	SRR12416836	Indonesia	South east Asia	2017	4	4.3.2	NA	Human	SRR12416836_Indonesia_4-3-2
59	SRR5074190	Vietnam	South east Asia	2009	4	4.3.2	NA	Human	SRR5074190_Vietnam_4-3-2
60	SRR1140950	South Africa	Africa	2013	4	4.3.2.1	NA	Human	SRR1140950_South Africa_4-3-2-1
61	ERR067601	Russia	Eurasi a	2008	4	4.3.3	NA	Human	ERR067601_Russia_4-3-3
62	ERR1193757	South Africa	Africa	NA	4	4.3.3	NA	Human	ERR1193757_South Africa_4-3-3
63	ERR751367	Peru	South America	2009	4	4.3.3	NA	Human	ERR751367_Peru_4-3-3
64	ERR979068	Denmark	Europe	1994	4	4.3.3	NA	Human	ERR979068_Denmark_4-3-3
65	SRR5074710	New Zealand	Pacific	2007	4	4.3.3	NA	Human	SRR5074710_New Zealand_4-3-3
66	SRR12006535	Malaysia	South east Asia	2013	4	4.3.4.1	NA	Human	SRR12006535_Malaysia_4-3-4-1
67	ERR275210	Portugal	Europe	2010	4	4.3.4.2	NA	Human	ERR275210_Portugal_4-3-4-2
68	ERR038744	Uganda	Africa	2004	4	4.3.4.2.1	NA	Human	ERR038744_Uganda_4-3-4-2-1
69	ERR2653229	Brazil	South America	2010	4	4.4.1	NA	Human	ERR2653229_Brazil_4-4-1
70	ERR2652930	Canada	North America	1997	4	4.4.1.1	NA	Human	ERR2652930_Canada_4-4-1-1
71	ERR2653126	Brazil	South America	2009	4	4.4.1.1	NA	Human	ERR2653126_Brazil_4-4-1-1
72	SRR10272562	New Zealand	Pacific	2001	4	4.4.1.1	NA	Human	SRR10272562_New Zealand_4-4-1-1
73	SRR12416832	Indonesia	South east Asia	2017	4	4.4.1.1	NA	Human	SRR12416832_Indonesia_4-4-1-1

74	SRR5065416	Vietnam	South east Asia	2011	4	4.4.1.1	NA	Human	SRR5065416_Vietnam_4-4-1-1
75	ERR2515429	South Africa	Africa	NA	4	4.4.2	NA	Human	ERR2515429_South Africa_4-4-2
76	ERR751827	Thailand	South east Asia	NA	4	4.4.2	NA	Human	ERR751827_Thailand_4-4-2
77	SRR5065455	Vietnam	South east Asia	2010	4	4.4.2	NA	Human	SRR5065455_Vietnam_4-4-2
78	SRR6480346	Indonesia	South east Asia	2014	4	4.4.2	NA	Human	SRR6480346_Indonesia_4-4-2
79	SRR671790	China	East Asia	NA	4	4.4.2	NA	Human	SRR671790_China_4-4-2
80	ERR234099	Nepal	NA	NA	3	3	Comas et al. 2013	Human	ERR234099_Nepal_3
81	ERR234109	Nepal	NA	NA	3	3	Comas et al. 2013	Human	ERR234109_Nepal_3
82	ERR234111	Nepal	NA	NA	3	3	Comas et al. 2013	Human	ERR234111_Nepal_3
83	ERR1213884	Nepal	South ern Asia	NA	1	1.2	Phelan_B MCMedicine_2016	Human	ERR1213884_Nepal_1-2
84	ERR551520	Nepal	South ern Asia	NA	2	2.2.1	Merker_N atGenet_2015	Human	ERR551520_Nepal_2-2-1
85	ERR553171	Nepal	South ern Asia	NA	2	2.2.1	Merker_N atGenet_2015	Human	ERR553171_Nepal_2-2-1
86	SRR14514372	Nepal	NA	NA	1	1.2.2.2	https://journals.asm.org/doi/10.1128/MRA.00614-21	Asiatic Elephant	SRR14514372_Nepal_1-2-2-2
87	SRR14514371	Nepal	NA	NA	1	1.2.2.2	https://journals.asm.org/doi/10.1128/MRA.00614-21	Asiatic Elephant	SRR14514371_Nepal_1-2-2-2
88	SRR6487127	South Africa	NA	NA	4	4.3.2.1	https://www.frontiersin.org/articles/10.3389/fvets.2019.00018/full	African Savanna Elephant	SRR6487127_South Africa_4-3-2-1
89	G08157	Switzerland	NA	NA	4	4.7	DOI:10.1038/s41598-017-15278-9	Asiatic Elephant	G08157_Switzerland_4-7
90	G08164	Switzerland	NA	NA	4	4.7	DOI:10.1038/s41598-017-15278-9	Asiatic Elephant	G08164_Switzerland_4-7

91	G08160	Switzerland	NA	NA	4	4.7	DOI:10.1038/s41598-017-15278-9	Asiatic Elephant	G08160_Switzerland_4-7
92	SRR3880244	USA	NA	NA	4	4	http://dx.doi.org/10.3201/eid2303.160726	Asiatic Elephant	SRR3880244_USA_4
93	SRR3880247	USA	NA	NA	4	4	http://dx.doi.org/10.3201/eid2303.160726	Asiatic Elephant	SRR3880247_USA_4
94	ERR5336171	Outgroup	NA	NA	<i>M. africanum</i>	NA	NA	NA	NA

Chapter 5: Annex Supplementary Material

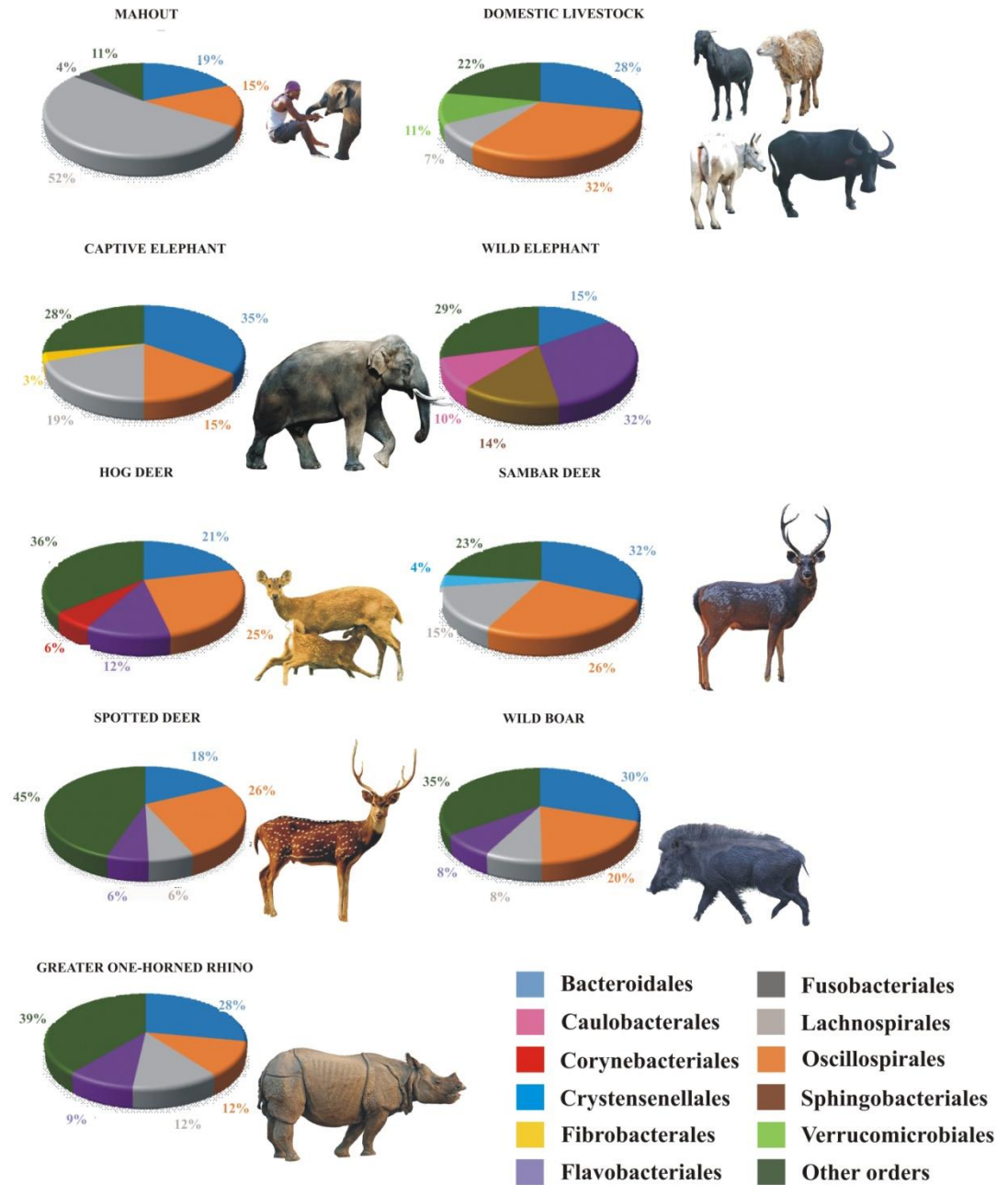


FIGURE S1 Order -level composition of gut microbiota across humans, livestock, captive elephants and wild herbivores.

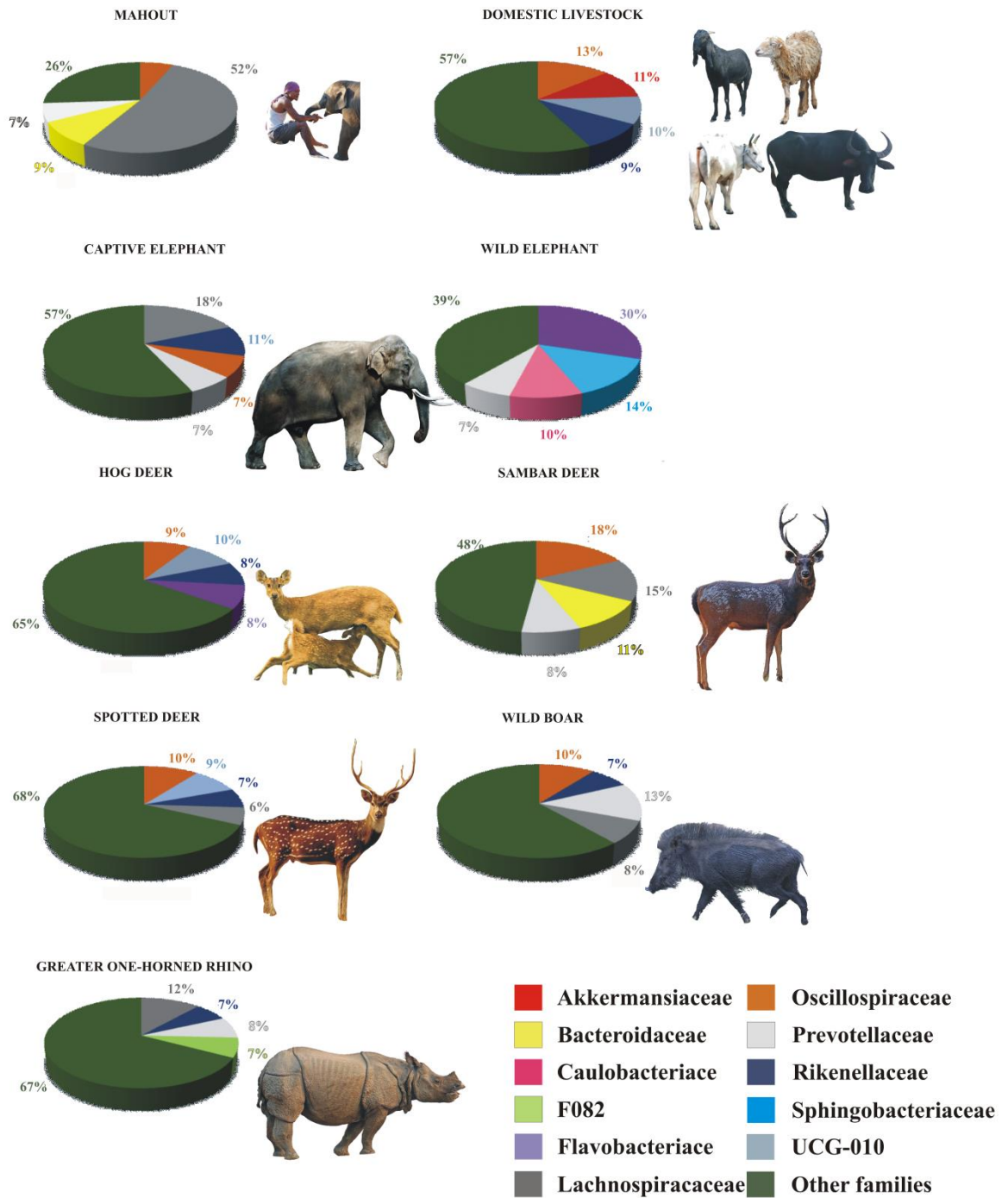


FIGURE S2 Family -level composition of gut microbiota across humans, livestock, captive elephants and various species of wild herbivores.

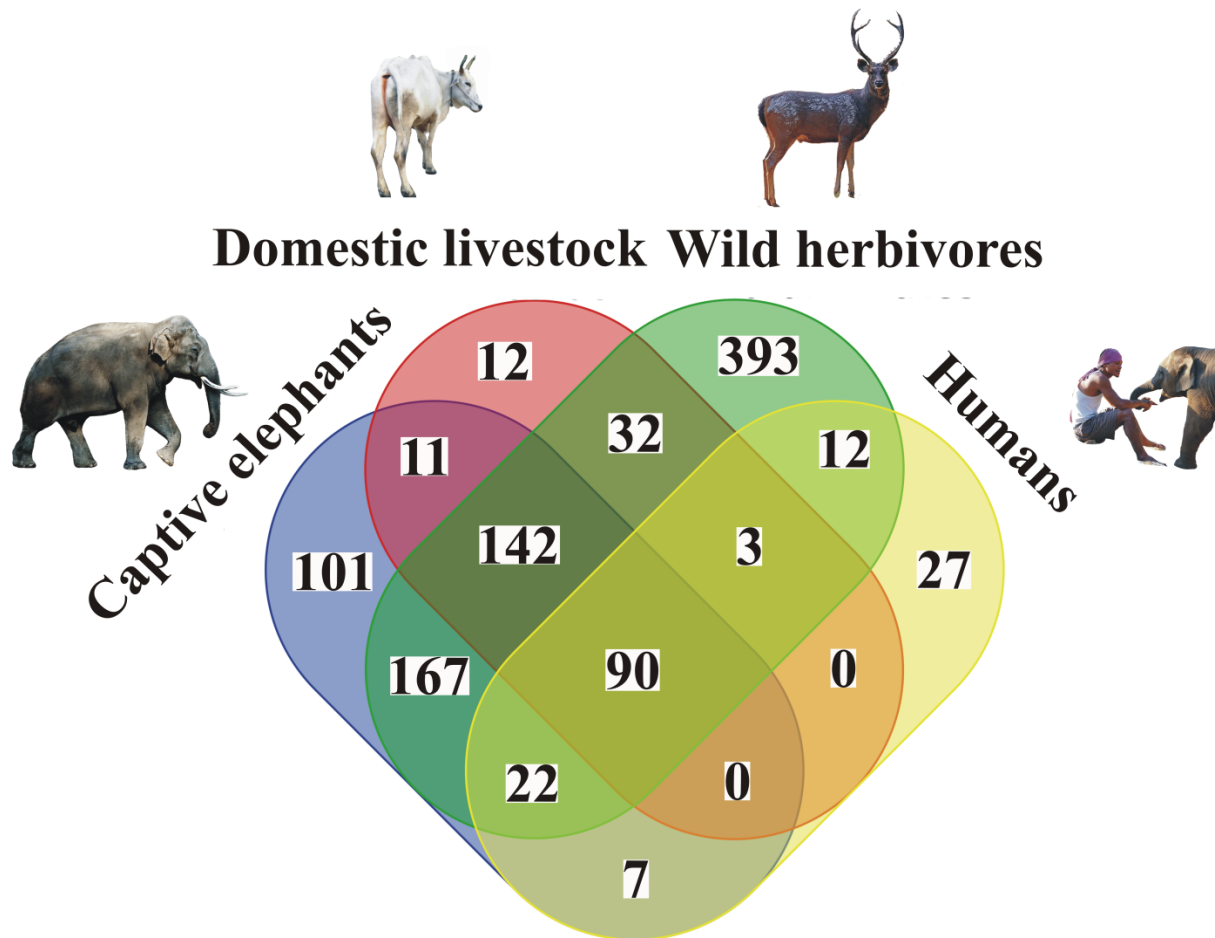
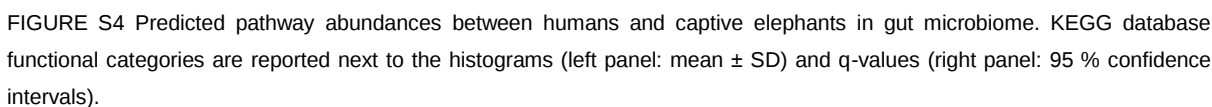


FIGURE S3 Sharing of bacterial genera across the gut microbiota of captive elephants, domestic livestock, wild herbivores and humans (*mahouts*).



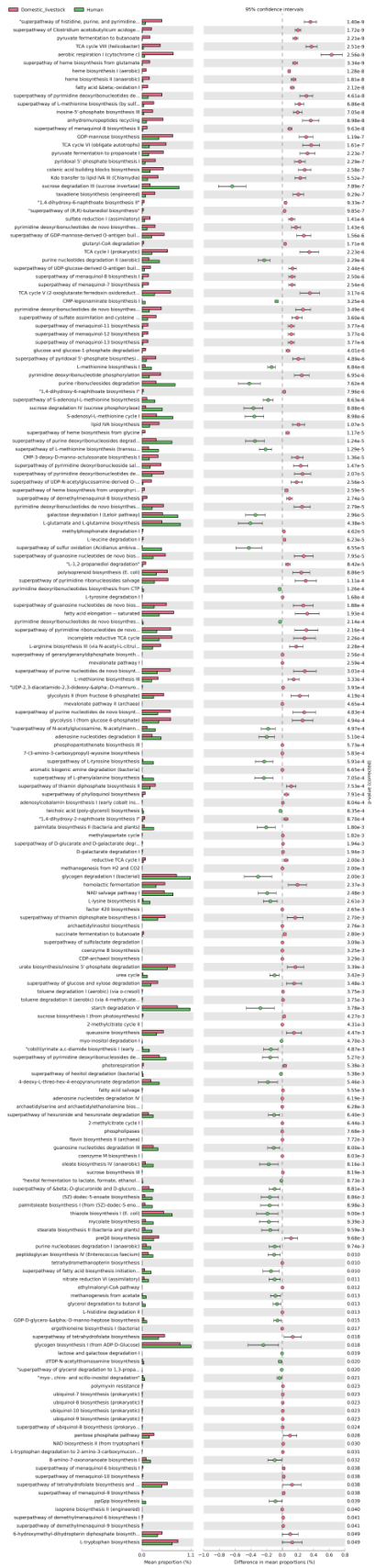


FIGURE S5 Predicted pathway abundances between humans and domestic livestock in gut microbiome. KEGG database functional categories are reported next to the histograms (left panel: mean $\pm$ SD) and q-values (right panel: 95 % confidence intervals).

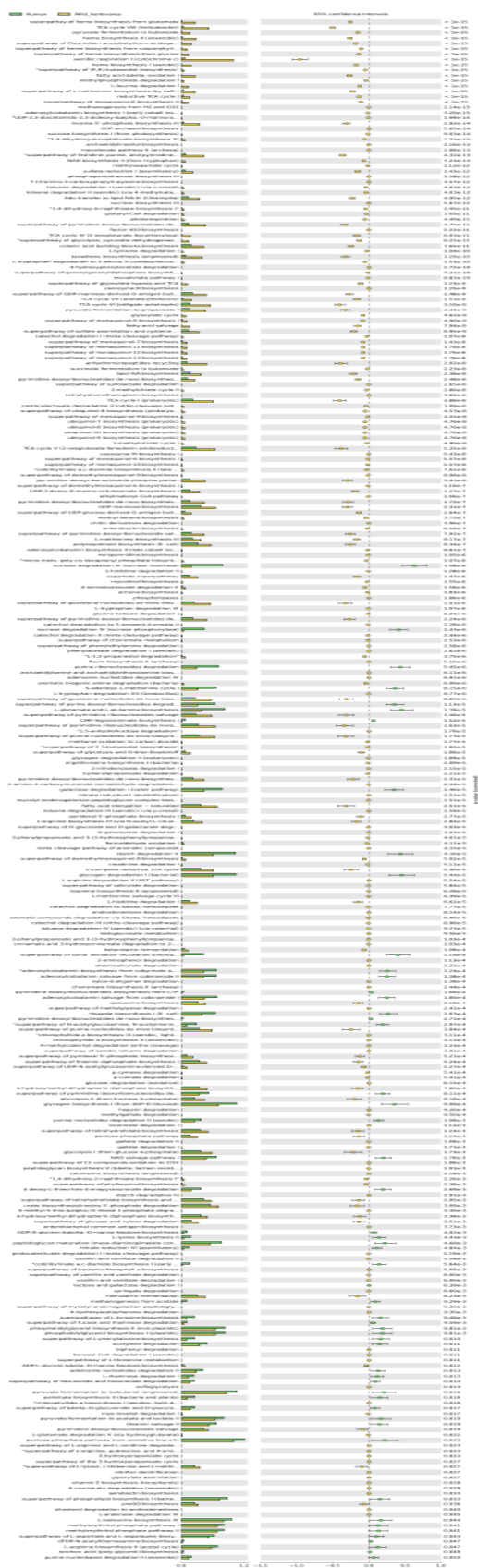


FIGURE S6 Predicted pathway abundances between humans and wild herbivores in gut microbiome. KEGG database functional categories are reported next to the histograms (left panel: mean ± SD) and q-values (right panel: 95 % confidence intervals).

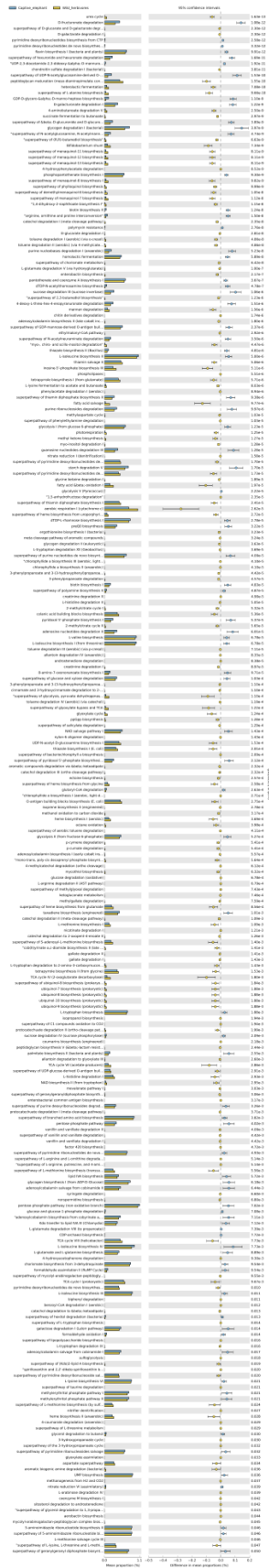


FIGURE S7 Predicted pathway abundances between captive elephants and domestic livestock in gut microbiome. KEGG database functional categories are reported next to the histograms (left panel: mean ± SD) and q-values (right panel: 95 % confidence intervals).

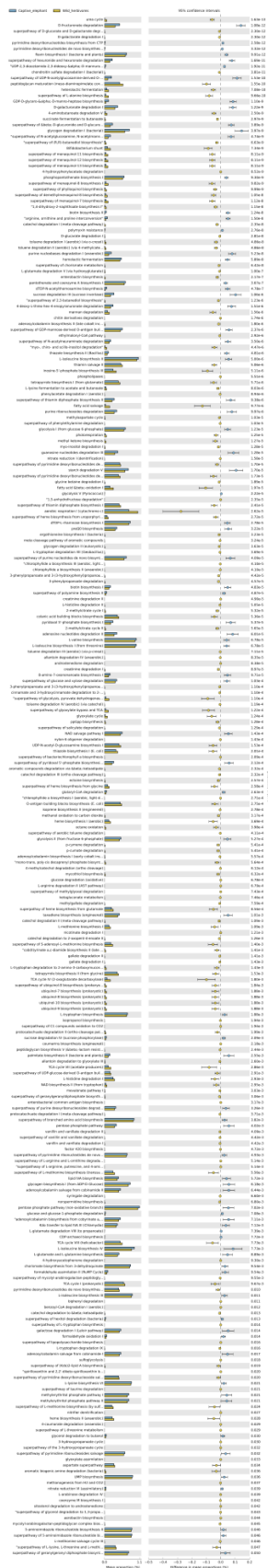


FIGURE S8 Predicted pathway abundances between captive elephants and wild herbivores in gut microbiome. KEGG database functional categories are reported next to the histograms (left panel: mean ± SD) and q-values (right panel: 95 % confidence intervals).

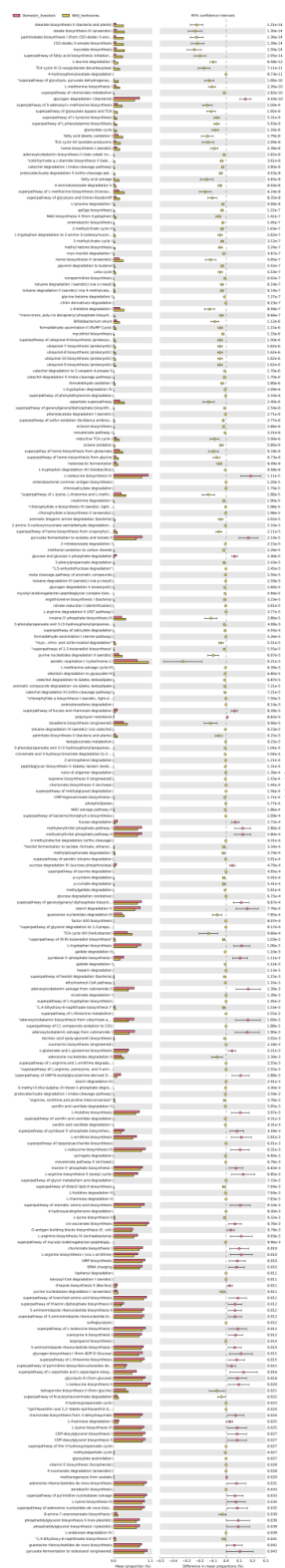


FIGURE S9 Sharing of KEGG pathways related to gut microbiota between humans and each one of the other host categories as well as among all of them.

5.1 TABLE S1

List of fecal samples used in this study with location coordinates and *cyt-b* GenBank accession number.

N	Collection Date	Latitude	Longitude	Sampling site	Map location - Fig. 1(d)	Animal ID	Species	Storage medium	Associated environment	Type	MiSeq_id	Cyt-b GenBank accession number
1	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-B7	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WKB7	OQ127 623
2	13th Feb 2019	N27.55 841°	E084.10452°	Amaltari	3	WAm-B1	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WAmB1	OQ127 625
3	9th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-B3	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WKB3	OQ127 632
4	9th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-B4	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WKB4	OP314 869
5	9th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-B5	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WKB5	OQ127 630
6	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-B6	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WKB6	OQ127 631
7	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-B8	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WKB8	OQ127 636
8	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-B9	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WKB9	OQ127 629
9	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-B10	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WKB10	OQ127 622
10	7th Feb 2019	N27.53 625°	E084.24999°	Sukhivar Post	11	WS-B1	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WSB1	OQ127 635
11	7th Feb 2019	N27.53 625°	E084.24999°	Sukhivar Post	11	WS-B2	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WSB2	OQ127 634
12	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-B1	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WTB1	OQ127 628
13	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-B2	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WTB2	OQ127 633
14	13th Feb 2019	N27.55 841°	E084.10452°	Amaltari	3	WAm-B2	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WAmB2	OQ127 627
15	13th Feb 2019	N27.55 841°	E084.10452°	Amaltari	3	WAm-B3	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WAmB3	OQ127 624
16	13th Feb 2019	N27.55 841°	E084.10452°	Amaltari	3	WAm-B4	<i>Sus scrofa</i>	Fe-DET	Jungle	Wild	WAmB4	OQ127 626
1	7th Feb 2019	N27.54 853°	E084.14133°	Khoriyami Post	5	WKh-R1	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WKR1	OQ127 621
2	7th Feb 2019	N27.54 853°	E084.14133°	Khoriyami Post	5	WKh-R2	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WKR2	OQ127 616
3	7th Feb 2019	N27.54 853°	E084.14133°	Khoriyami Post	5	WKh-R3	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WKR3	OQ127 600
4	7th Feb 2019	N27.54 853°	E084.14133°	Khoriyami Post	5	WKh-R4	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WKR4	OQ127 619
5	7th Feb 2019	N27.54 853°	E084.14133°	Khoriyami Post	5	WKh-R5	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WKR5	OQ127 620
6	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-R1	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WTR1	OQ127 598
7	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-R2	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WTR2	OQ127 597
8	7th Feb 2019	N27.53 625°	E084.24999°	Sukhivar Post	11	WS-R1	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WSR1	OQ127 617
9	7th Feb 2019	N27.53 625°	E084.24999°	Sukhivar Post	11	WS-R2	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WSR2	OQ127 599
10	7th Feb 2019	N27.56 364°	E084.04956°	Amaltari	1	WAm-R1	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR1	OQ127 618
11	7th Feb 2019	N27.56 364°	E084.04956°	Koluhawa	1	WAm-R2	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR2	OQ127 609
12	7th Feb 2019	N27.56 364°	E084.04956°	Koluhawa	1	WAm-R3	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR3	OQ127 608
13	7th Feb 2019	N27.56 364°	E084.04956°	Koluhawa	1	WAm-R4	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR4	OQ127 607
14	13th Feb 2019	N27.56 364°	E084.04956°	Koluhawa	1	WAm-R5	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR5	OQ127 606
15	13th Feb 2019	N27.56 137°	E084.10571°	Amaltari	3	WAm-R6	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR6	OQ127 605
16	13th Feb 2019	N27.56 137°	E084.10571°	Amaltari	3	WAm-R7	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR7	OQ127 604
17	13th Feb 2019	N27.56 137°	E084.10571°	Amaltari	3	WAm-R8	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR8	OP314 867
18	13th Feb 2019	N27.56 137°	E084.10571°	Amaltari	3	WAm-R9	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR9	OQ127 603
19	13th Feb 2019	N27.56 137°	E084.10571°	Amaltari	3	WAm-R10	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR10	OQ127 614
20	13th Feb 2019	N27.56 137°	E084.10571°	Amaltari	3	WAm-R11	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR11	OQ127 613
21	13th Feb 2019	N27.55 923°	E084.10547°	Amaltari	3	WAm-R12	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR12	OQ127 612

22	13th Feb 2019	N27.55 923°	E084.10547°	Amaltari	3	WAm-R13	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR1 3	OQ127 611
23	13th Feb 2019	N27.56 066°	E084.10592°	Amaltari	3	WAm-R14	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WAmR1 4	OQ127 610
24	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-R1	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WIR1	OQ127 595
25	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-R2	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WIR2	OQ127 602
26	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-R3	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WIR3	OQ127 601
27	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-R4	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WIR4	OQ127 615
28	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-R5	<i>Rhinoceros unicornis</i>	Fe-DET	Jungle	Wild	WIR5	OQ127 596
1	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D4	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD4	OP314 831
2	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D5	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD5	OP314 832
3	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D6	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD6	OP314 833
4	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D7	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD7	OP314 834
5	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D8	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD8	OP314 835
6	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D9	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD9	OP314 836
7	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D10	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD10	OP314 819
8	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D11	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD11	OP314 820
9	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D12	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD12	OP314 821
10	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D13	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD13	OP314 822
11	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D14	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD14	OP314 823
12	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D15	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD15	OP314 824
13	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D16	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD16	OP314 825
14	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D17	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD17	OP314 826
15	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D18	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD18	OP314 827
16	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D19	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD19	OP314 828
17	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D20	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD20	OP314 829
18	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-D21	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKD21	OP314 830
19	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D1A	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 1A	OP314 856
20	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D2A	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 2A	OP314 857
21	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D3	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 3	OP314 861
22	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D4A	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 4A	OP314 858
23	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D5	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 5	OP314 862
24	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D6	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 6	OP314 859
25	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D7	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 7	OQ127 588
26	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D8	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 8	OP314 860
27	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D9	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 9	OQ127 589
28	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D10	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 10	OP314 850
29	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D11	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 11	OP314 851
30	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D12	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 12	OP314 852
31	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D13	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 13	OP314 853
32	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D14	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 14	OP314 854
33	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D15	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WTSPD 15	OP314 855
34	7th Feb 2019	N27.54 853°	E084.14133°	Khoriyami Post	5	WKH-Sp-D1A	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKHSP D1A	OP314 837
35	7th Feb 2019	N27.54 853°	E084.14133°	Khoriyami Post	5	WKH-Sp-D3	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKHSP D3	OP314 838
36	7th Feb 2019	N27.54 853°	E084.14133°	Khoriyami Post	5	W-Kh-Sp-D1B	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WKHSP D1B	OQ127 591
37	7th Feb 2019	N27.53 625°	E084.24999°	Sukhivar Post	11	WS-Sp-D1	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WSSPD 1	OP314 848
38	7th Feb 2019	N27.53 625°	E084.24999°	Sukhivar Post	11	WS-Sp-D3	<i>Axis axis</i>	Fe-DET	Jungle	Wild	WSSPD 3	OP314 849

39	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D1B	Axis axis	Fe-DET	Jungle	Wild	WtSPD 1B	OQ127 590
40	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-Sp-D2	Axis axis	Fe-DET	Jungle	Wild	WISPD 2	OP314 815
41	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-Sp-D3	Axis axis	Fe-DET	Jungle	Wild	WISPD 3	OP314 816
42	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-Sp-D4	Axis axis	Fe-DET	Jungle	Wild	WISPD 4	OP314 817
43	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-Sp-D5	Axis axis	Fe-DET	Jungle	Wild	WISPD 5	OP314 818
44	18th Dec 2018	N27.57 640°	E084.21659°	Meghauli	10	WM-Sp-D1	Axis axis	Fe-DET	Jungle	Wild	WMSP D1	OP314 839
45	18th Dec 2018	N27.57 640°	E084.21659°	Meghauli	10	WM-Sp-D2A	Axis axis	Fe-DET	Jungle	Wild	WMSP D2A	OP314 840
46	18th Dec 2018	N27.57 640°	E084.21659°	Meghauli	10	WM-Sp-D3	Axis axis	Fe-DET	Jungle	Wild	WMSP D3	OP314 841
47	18th Dec 2018	N27.57 640°	E084.21659°	Meghauli	10	WM-Sp-D4	Axis axis	Fe-DET	Jungle	Wild	WMSP D4	OP314 842
48	18th Dec 2018	N27.57 640°	E084.21659°	Meghauli	10	WM-Sp-D5	Axis axis	Fe-DET	Jungle	Wild	WMSP D5	OP314 843
49	18th Dec 2018	N27.57 640°	E084.21659°	Meghauli	10	WM-Sp-D6	Axis axis	Fe-DET	Jungle	Wild	WMSP D6	OP314 844
50	13th Feb 2019	N27.56 066°	E084.10592°	Amaltari	3	WAm-Sp-D1	Axis axis	Fe-DET	Jungle	Wild	WAmSP D1	OP314 810
51	18th Dec 2018	N27.57 640°	E084.21659°	Meghauli	10	WM-Sp-D2B	Axis axis	Fe-DET	Jungle	Wild	WMSP D2B	OQ127 592
52	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D2B	Axis axis	Fe-DET	Jungle	Wild	WTSPD 2B	OQ127 594
53	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops	6	WT-Sp-D4B	Axis axis	Fe-DET	Jungle	Wild	WTSPD 4B	OQ127 593
1	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-Sa-D1	Rusa unicolor	Fe-DET	Jungle	Wild	WISAD 1	OP314 868
2	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-Sa-D2	Rusa unicolor	Fe-DET	Jungle	Wild	WISAD 2	OP314 811
3	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-Sa-D3	Rusa unicolor	Fe-DET	Jungle	Wild	WISAD 3	OP314 812
4	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-Sa-D4	Rusa unicolor	Fe-DET	Jungle	Wild	WISAD 4	OP314 813
5	14th Dec 2018	N27.57 305°	E084.50158°	Hattisar/Icharni	21	WI-Sa-D5	Rusa unicolor	Fe-DET	Jungle	Wild	WISAD 5	OP314 814
6	7th Feb 2019	N27.53 625°	E084.24999°	Sukhivar Post	11	WS-Sa-D1	Rusa unicolor	Fe-DET	Jungle	Wild	WSSAD 1	OP314 845
7	7th Feb 2019	N27.53 625°	E084.24999°	Sukhivar Post	11	WS-Sa-D2	Rusa unicolor	Fe-DET	Jungle	Wild	WSSAD 2	OP314 846
8	7th Feb 2019	N27.53 625°	E084.24999°	Sukhivar Post	11	WS-Sa-D3	Rusa unicolor	Fe-DET	Jungle	Wild	WSSAD 3	OP314 847
1	10th Feb 2019	N27.58 192°	E084.46268°	Khorsor	18	WK-R1 A	Elephas maximus	Fe-DET	Jungle	Wild	WKR1-A	OP314 865
1	12th Dec 2018	N27.57 564°	E084.45025 2°	Hattisar	17	LH-01-B1	Bubalus bubalis	Fe-DET	Human	Dome stic	LH01B1	
2	12th Dec 2018	N27.57 490°	E084.50201°	Hattisar	21	LH-02-B1	Bubalus bubalis	Fe-DET	Human	Dome stic	LH02B1	
3	12th Dec 2018	N27.57 966°	E084.50458°	Badreni	21	CE-43-01-B2	Bubalus bubalis	Fe-DET	Human	Dome stic	CE4301 B2	
1	12th Feb 2019	N27.57 222°	E084.52177°	Kumroj	22	LKu-C2	Bos taurus	Fe-DET	Human	Dome stic	LKUC2	
1	12th Dec 2018	N27.57 911°	E084.50290°	Badreni	21	CE-23-01-G1	Capra hircus	Fe-DET	Human	Dome stic	CE2301 G1	
2	12th Dec 2018	N27.57 911°	E084.50290°	Badreni	21	CE-23-01-G2	Capra hircus	Fe-DET	Human	Dome stic	CE2301 G2	
3	12th Dec 2018	N27.58 605°	E084.49068°	Badreni	20	CE-24-01-G1	Capra hircus	Fe-DET	Human	Dome stic	CE2401 G1	
4	12th Dec 2018	N27.58 605°	E084.49068°	Badreni	20	CE-24-01-G2	Capra hircus	Fe-DET	Human	Dome stic	CE2401 G2	
5	12th Dec 2018	N27.58 605°	E084.49068°	Badreni	20	CE-24-01-G3	Capra hircus	Fe-DET	Human	Dome stic	CE2401 G3	
6	12th Feb ,2019	N27.57 127°	E084.52181°	Kumroj	22	LKu-G4	Capra hircus	Fe-DET	Human	Dome stic	LKUG4	
1	18th Dec-2018	N27.58 044°	E084.47357°	Megauli	19	LM-Sh1	Ovis aries	Fe-DET	Human	Dome stic	LMSh1	OP314 866
2	18th Dec-2018	N27.58 044°	E084.47357°	Megauli	19	LM-Sh2	Ovis aries	Fe-DET	Human	Dome stic	LMSh2	
3	18th Dec-2018	N27.58 044°	E084.47357°	Megauli	19	LM-Sh3	Ovis aries	Fe-DET	Human	Dome stic	LMSh3	
4	18th Dec-2018	N27.58 044°	E084.47357°	Megauli	19	LM-Sh4	Ovis aries	Fe-DET	Human	Dome stic	LMSh4	
5	18th Dec-2018	N27.58 044°	E084.47357°	Megauli	19	LM-Sh5	Ovis aries	Fe-DET	Human	Dome stic	LMSh5	
1	10th Dec 2018	N27.56 092°	E084.57591°	Amritya Post	24	CE-38	Elephas maximus	Fe-DET	Human	Wild	CE3801	
2	10th Dec 2018	N27.57 385°	E084.65109°	Pyaridhap Post	25	CE-40	Elephas maximus	Fe-DET	Human	Wild	CE4001	
3	10th Dec 2018	N27.55 896°	E084.55832°	Janakpur Post	23	CE-41	Elephas maximus	Fe-DET	Human	Wild	CE41	
4	10th Dec 2018	N27.55 896°	E084.55832°	Janakpur Post	23	CE-42	Elephas maximus	Fe-DET	Human	Wild	CE42	
6	13th Dec 2018	N27.57 266°	E084.40656°	Belsar Post	16	CE-44	Elephas maximus	Fe-DET	Human	Wild	CE44	
7	13th Dec 2018	N27.57 266°	E084.40656°	Belsar Post	16	CE-45	Elephas maximus	Fe-DET	Human	Wild	CE45	

8	13th Dec 2018	N27.57 073°	E084.36718°	Safari Narayani Hotel	15	CE-49	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE49	
9	13th Dec 2018	N27.56 620°	E084.35123°	Kashara Resort	14	CE-50	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE50	
10	13th Dec 2018	N27.56 620°	E084.35123°	Kashara Resort	14	CE-51	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE51	
11	13th Dec 2018	N27.55 926°	E084.33662°	Jungle Villa Resort	13	CE-53	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE53	
12	18th Dec 2018	N27.56 740°	E084.30211°	Tiger Land Safari	12	CE-55	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE55	
13	18th Dec 2018	N27.57 590°	E084.21676°	Sarang Wildlife Sanctuary	10	CE-56	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE56	
14	18th Dec 2018	N27.57 590°	E084.21676°	Sarang Wildlife Sanctuary	10	CE-58	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE58	
15	18th Dec 2018	N27.57 197°	E084.20541°	CG	9	CE-60	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE60	
16	18th Dec 2018	N27.56 951°	E084.20112°	Bamboo Cottage	8	CE-61	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE61	
17	18th Dec 2018	N27.56 548°	E084.19284°	Barahi Resort	7	CE-62	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE62	
18	18th Dec 2018	N27.56 548°	E084.19284°	Barahi Resort	7	CE-63	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE63	
19	18th Dec 2018	N27.56 548°	E084.19284°	Barahi Resort	7	CE-65	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE65	
20	9th Jan 2019	N27.63 155°	E084.17107°	Island	4	CE-69	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE69	
21	9th Jan 2019	N27.63 155°	E084.17107°	Island	4	CE-70	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE70	
22	10th Jan 2019	N27.57 044°	E084.10016°	Tiger Tops(Tharu Lodge)	2	CE-71	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE71	
23	10th Jan 2019	N27.57 044°	E084.10016°	Tiger Tops(Tharu Lodge)	2	CE-72	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE72	
24	10th Jan 2019	N27.57 044°	E084.10016°	Tiger Tops(Tharu Lodge)	2	CE-73	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE73	
25	10th Jan 2019	N27.57 044°	E084.10016°	Tiger Tops(Tharu Lodge)	2	CE-77	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE77	
26	10th Jan 2019	N27.57 044°	E084.10016°	Tiger Tops(Tharu Lodge)	2	CE-78	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE78	
27	10th Jan 2019	N27.57 044°	E084.10016°	Tiger Tops(Tharu Lodge)	2	CE-79	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE79	
28	10th Jan 2019	N27.57 044°	E084.10016°	Tiger Tops(Tharu Lodge)	2	CE-80	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE80	
29	7th Feb 2019	N27.53 625°	E084.24999°	Sukhibar Post	11	CE-82	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE82	
30	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops(Kor Jungle)	6	CE-83	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE83	
31	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops(Kor Jungle)	6	CE-84	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE84	
32	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops(Kor Jungle)	6	CE-85	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE85	OP314 863
33	7th Feb 2019	N27.54 272°	E084.18855°	Tiger Tops(Kor Jungle)	6	CE-86	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE86	
34	7th Feb 2019	N27.54 853°	E084.14133°	Khoriamuni Post	5	CE-87	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE87	
35	7th Feb 2019	N27.54 853°	E084.14133°	Khoriamuni Post	5	CE-88	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE88	
36	9th Feb 2019	N27.57 869°	E084.49517°	Sauraha	21	CE-89	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE89	
37	9th Feb 2019	N27.57 869°	E084.49517°	Sauraha	21	CE-91	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE91	
38	9th Feb 2019	N27.57 869°	E084.49517°	Sauraha	21	CE-92	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE92	OP314 864
39	9th Feb 2019	N27.59 361°	E084.48826°	Sauraha(Ba gmara)	20	CE-96	<i>Elephas maximus</i>	Fe-DET	Human	Wild	CE96	
1	23th April 2021	27.5730 47°	84.501191°	NTNC, Hattisar	21	H1	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU1	
2	23th April 2021	27.5730 47°	84.501191°	NTNC, Hattisar	21	H2	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU2	
3	23th April 2021	27.5730 47°	84.501191°	NTNC, Hattisar	21	H3	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU3	
4	23th April 2021	27.5730 47°	84.501191°	NTNC, Hattisar	21	H4	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU4	
5	23th April 2021	27.5730 47°	84.501191°	NTNC, Hattisar	21	H5	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU5	

6	23th April 2021	27.5730 47°	84.501191°	NTNC, Hattisar	21	H6	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU6	
7	23th April 2021	27.5730 47°	84.501191°	NTNC, Hattisar	21	H7	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU7	
8	23th April 2021	27.5730 47°	84.501191°	NTNC, Hattisar	21	H8	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU8	
9	23th April 2021	27.5730 47°	84.501191°	NTNC, Hattisar	21	H9	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU9	
10	23th April 2021	27.5730 47°	84.501191°	NTNC, Hattisar	21	H10	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU10	
11	23th April 2021	27.5730 47°	84.501191°	NTNC, Hattisar	21	H11	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU11	
12	23th April 2021	27.5820 23°	84.463279°	EBC, Khorsar	18	H12	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU12	
13	23th April 2021	27.5820 23°	84.463279°	EBC, Khorsar	18	H13	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU13	
14	23th April 2021	27.5820 23°	84.463279°	EBC, Khorsar	18	H14	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU14	
15	23th April 2021	27.5820 23°	84.463279°	EBC, Khorsar	18	H15	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU15	
16	23th April 2021	27.5820 23°	84.463279°	EBC, Khorsar	18	H16	<i>Homo sapiens</i>	Fe-DET	Elephant/Jungle	Dome stic	HU16	

5.2 TABLE S2

Number of bacterial 16S reads retrieved across different hosts and taxonomic levels.

Phylum

index	Elephant_captive	Elephant_Wild	Domestic_livestock	Hog_deer	Sambar_deer	Spotted_deer	Wild_Boar	Rhino	Hu_mahout
d__Bacteria;p__Proteobacteria	408	502	133	1747	173	5505	1269	570	177
d__Bacteria;p__Actinobacteriota	741	26	59	2546	10	5174	1614	222	14
d__Bacteria;p__Bacteroidota	17644	1681	7428	9409	3123	19886	10010	4241	1811
d__Bacteria;p__Firmicutes	23341	289	13947	9620	4926	32955	10427	3764	6967
d__Bacteria;p__Verrucomicrobiota	1516	37	3223	504	124	1831	531	253	71
d__Bacteria;p__Fibrobacterota	1577	15	61	0	0	22	55	72	0
d__Bacteria;p__Patescibacteria	272	3	211	367	233	1733	48	184	67
d__Bacteria;p__Planctomycetota	1148	19	606	310	5	963	455	296	0
d__Bacteria;p__Synergistota	885	16	4	0	0	12	19	10	3
d__Bacteria;p__Armatimonadota	242	0	0	0	0	10	0	21	1
d__Bacteria;p__Campilobacterota	18	0	56	0	15	58	16	1	1
d__Bacteria;p__Fusobacteriota	2	0	0	0	283	1	4	2	348
d__Bacteria;p__Cyanobacteria	662	11	233	154	24	543	259	55	7
d__Bacteria;p__WPS-2	42	0	0	0	0	5	33	52	1
d__Archaea;p__Euryarchaeota	36	0	20	22	15	92	43	1	1
d__Bacteria;p__Elusimicrobiota	126	3	42	39	9	156	40	40	10
d__Archaea;p__Halobacterota	85	0	14	19	0	24	0	25	0
d__Bacteria;p__Chloroflexi	43	0	9	0	48	98	8	24	3
d__Bacteria;p__Bdellovibrionota	0	0	0	0	0	10	0	53	1
d__Bacteria;p__Spirochaetota	526	2	88	7	0	85	67	61	1
d__Bacteria;__	277	3	72	4	3	238	50	55	8
d__Bacteria;p__Myxococcota	24	0	10	0	0	9	0	19	0
d__Archaea;p__Thermoplasmatota	67	9	8	4	0	19	19	8	0
d__Bacteria;p__SAR324_clade(Marine_group_B)	19	0	0	0	0	5	0	7	0
d__Bacteria;p__Desulfobacterota	27	0	21	0	0	46	8	8	0
d__Bacteria;p__Acidobacteriota	12	0	2	1	0	18	0	7	0
d__Bacteria;p__Deinococcota	2	0	0	4	0	5	0	0	4
d__Bacteria;p__Sumerlaeota	1	0	0	0	0	2	1	4	0
Unassigned;__	30	0	4	0	0	71	1	6	4
d__Bacteria;p__Abditibacteriota	0	0	0	3	0	10	0	0	0
d__Bacteria;p__Gemmatimonadota	0	0	0	0	0	5	0	4	0
d__Eukaryota;__	5	0	1	0	0	5	1	1	0
d__Archaea;p__Aenigmarchaeota	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Deferribacterota	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Hydrogenedentes	0	0	0	0	0	1	0	0	0
d__Eukaryota;p__Ascomycota	0	0	0	0	0	1	0	0	0
d__Eukaryota;p__Arthropoda	0	0	0	0	0	1	0	0	0

Class

index	Elephant_captive	Elephant_Wild	Domestic_livestock	Hog_deer	Sambar_deer	Spotted_deer	Wild_Boar	Rhino	Hu_mahout
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria	294	486	127	1725	148	5103	443	467	11
d__Bacteria;p__Actinobacteriota;c__Actinobacteria	328	12	25	2518	10	4878	1539	155	8
d__Bacteria;p__Bacteroidota;c__Bacteroidia	17642	1681	7428	9407	3123	19874	10010	4241	1811
d__Bacteria;p__Firmicutes;c__Clostridia	21269	240	13610	9506	4856	31323	9673	3205	6859
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae	166	29	2831	286	107	1307	453	176	3
d__Bacteria;p__Fibrobacterota;c__Fibrobacteria	1577	15	61	0	0	22	55	72	0
d__Bacteria;p__Patescibacteria;c__Saccharimonadia	232	3	208	367	233	1721	48	113	62
d__Bacteria;p__Planctomycetota;c__Planctomycetes	1115	19	599	270	5	888	451	280	0
d__Bacteria;p__Synergistota;c__Synergistia	885	16	4	0	0	12	19	10	3
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria	113	16	6	22	25	400	826	102	166
d__Bacteria;p__Armatimonadota;c__uncultured	240	0	0	0	0	2	0	20	0
d__Bacteria;p__Campilobacterota;c__Campylobacteriia	18	0	56	0	15	58	16	1	1
d__Bacteria;p__Fusobacteriota;c__Fusobacteriia	2	0	0	0	283	1	4	2	348
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria	868	8	386	218	17	492	66	37	68
d__Bacteria;p__Cyanobacteria;c__Vampirivibrionia	646	6	228	154	16	467	253	38	0
d__Bacteria;p__Firmicutes;__	35	0	18	0	0	54	140	9	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia	408	14	33	28	0	277	74	41	5
d__Bacteria;p__WPS-2;c__WPS-2	42	0	0	0	0	5	33	52	1
d__Bacteria;p__Firmicutes;c__Negativicutes	404	11	68	10	0	130	76	31	21

d__Bacteria;p__Firmicutes;c__Bacilli	1617	38	247	104	70	1442	538	517	87
d__Archaea;p__Euryarchaeota;c__Methanobacteria	36	0	20	22	15	92	43	1	1
d__Bacteria;p__Elusimicrobiota;c__Elusimicrobia	61	3	39	39	9	156	40	25	10
d__Archaea;p__Halobacterota;c__Methanomicrobia	74	0	14	19	0	23	0	23	0
d__Bacteria;p__Patescibacteria;c__Gracilibacteria	37	0	1	0	0	3	0	67	0
d__Bacteria;p__Elusimicrobiota;c__Endomicrobia	62	0	3	0	0	0	0	15	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae	22	0	9	0	48	73	6	13	1
d__Bacteria;p__Bdellovibrionota;c__Oligoflexia	0	0	0	0	0	1	0	46	0
d__Bacteria;p__Spirochaetota;c__Spirochaetia	523	2	88	7	0	85	67	36	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia	15	5	5	0	8	76	6	17	7
d__Bacteria;__;__	277	3	72	4	3	238	50	55	8
d__Bacteria;p__Myxococcota;c__Myxococcia	22	0	10	0	0	2	0	7	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae	26	0	7	40	0	74	3	8	0
d__Bacteria;p__Spirochaetota;c__MVP-15	0	0	0	0	0	0	0	24	0
d__Archaea;p__Halobacterota;c__Methanosarcinia	11	0	0	0	0	1	0	2	0
d__Archaea;p__Thermoplasmata;c__Thermoplasma	67	9	8	4	0	19	19	8	0
d__Bacteria;p__Actinobacteriota;c__Acidimicrobiia	3	0	0	0	0	14	0	26	0
d__Bacteria;p__Firmicutes;c__Incertae_Sedis	0	0	4	0	0	6	0	0	0
d__Bacteria;p__Firmicutes;c__uncultured	5	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Limnochordia	6	0	0	0	0	0	0	0	0
d__Bacteria;p__Chloroflexi;c__Chloroflexia	16	0	0	0	0	16	0	8	2
d__Bacteria;p__SAR324_clade(Marine_group_B);c__SAR324_clade(Marine_group_B)	19	0	0	0	0	5	0	7	0

d__Bacteria;p__Myxococcota;c__Polyangia	1	0	0	0	0	7	0	12	0
d__Bacteria;p__Desulfobacterota;c__Desulfovibrionia	16	0	20	0	0	43	8	3	0
d__Bacteria;p__Actinobacteriota;c__Thermoleophilia	2	0	1	0	0	3	0	0	1
d__Bacteria;p__Acidobacteriota;c__Blastocatellia	6	0	1	0	0	7	0	4	0
d__Bacteria;p__Spirochaetota;c__Leptospirae	3	0	0	0	0	0	0	1	0
d__Bacteria;p__Patescibacteria;c__Parcubacteria	1	0	1	0	0	0	0	0	5
d__Bacteria;p__Deinococcota;c__Deinococci	2	0	0	4	0	5	0	0	4
d__Bacteria;p__Planctomycetota;c__vadinHA49	2	0	0	0	0	0	0	8	0
d__Bacteria;p__Sumerlaeota;c__Sumerlaeia	1	0	0	0	0	2	1	4	0
d__Bacteria;p__Bdellovibrionota;c__Bdellovibrionia	0	0	0	0	0	9	0	7	1
Unassigned;__;__	30	0	4	0	0	71	1	6	4
d__Bacteria;p__Firmicutes;c__Desulfotomaculia	2	0	0	0	0	0	0	2	0
d__Bacteria;p__Firmicutes;c__Syntrophomonadia	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Kiritimatiellae	481	0	6	0	0	30	12	40	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae	4	0	0	1	0	7	0	1	0
d__Bacteria;p__Patescibacteria;c__Microgenomatia	0	0	0	0	0	7	0	0	0
d__Bacteria;p__Abditibacteriota;c__Abditibacteria	0	0	0	3	0	10	0	0	0
d__Bacteria;p__Armatimonadota;c__Armatimonadia	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Gemmatimonadota;c__Gemmatimonadetes	0	0	0	0	0	5	0	3	0
d__Bacteria;p__Patescibacteria;c__Dojkabacteria	1	0	0	0	0	0	0	2	0
d__Bacteria;p__Bacteroidota;c__Rhodothermia	2	0	0	2	0	5	0	0	0
d__Bacteria;p__Armatimonadota;c__Fimbriimonadia	1	0	0	0	0	4	0	1	1
d__Eukaryota;__;__	5	0	1	0	0	5	1	1	0

d__Bacteria;p__Firmicutes;c__Moorellia	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Desulfobacterota;c__Desulfuromonadia	11	0	1	0	0	3	0	5	0
d__Bacteria;p__Actinobacteriota;c__MB-A2-108	0	0	0	0	0	2	1	0	0
d__Bacteria;p__Chloroflexi;c__TK10	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Elusimicrobiota;c__Rs-M47	3	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Kapabacteria	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Acidobacteriota;c__Vicnamibacteria	2	0	0	0	0	2	0	2	0
d__Bacteria;p__Patescibacteria;c__ABY1	1	0	0	0	0	0	0	1	0
d__Bacteria;p__Chloroflexi;c__KD4-96	2	0	0	0	0	3	1	0	0
d__Bacteria;p__Chloroflexi;c__JG30-KF-CM66	1	0	0	0	0	0	0	0	0
d__Archaea;p__Aenigmarchaeota;c__Deep_Sea_Euryarchaeotic_Group(DSEG)	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Verrucomicrobiota;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Chloroflexi;c__Dehalococcoidia	1	0	0	0	0	2	1	3	0
d__Bacteria;p__Deferribacterota;c__Deferribacteres	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Planctomycetota;c__OM190	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Myxococcota;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;__	3	0	0	0	0	0	1	0	0
d__Bacteria;p__Chloroflexi;c__Ktedonobacteria	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Armatimonadota;c__Chthonomonades	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;__	1	0	0	0	0	2	0	1	0
d__Bacteria;p__Acidobacteriota;c__Subgroup_5	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Acidobacteriota;c__Subgroup_18	0	0	0	0	0	1	0	0	0

d__Bacteria;p__Planctomycetota;c__Pla4_lineage	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Omnitrophia	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Hydrogenedentes;c__Hydrogenedentia	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Gemmatimonadota;c__S0134_terrestrial_group	0	0	0	0	0	0	0	1	0
d__Eukaryota;p__Ascomycota;__	0	0	0	0	0	1	0	0	0
d__Eukaryota;p__Arthropoda;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;__	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Patescibacteria;c__Berkelbacteria	0	0	1	0	0	1	0	0	0
d__Bacteria;p__Spirochaetota;c__Brachyspirae	0	0	0	0	0	0	0	0	1
d__Bacteria;p__Cyanobacteria;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Patescibacteria;c__uncultured	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Bacteroidota;c__SJA-28	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Chlamydiae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Patescibacteria;c__WWE3	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Acidobacteriota;c__Aminicenantia	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__Gitt-GS-136	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Chloroflexi;__	0	0	0	0	0	1	0	0	0

Order

index	Elephant_captive	Elephant_Wild	Domestic_livestock	Hog_deer	Sambar_deer	Spotted_deer	Wild_Boar	Rhino	Hu_mahout
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonadales	19	113	1	689	9	2828	91	101	4
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales	146	7	7	747	7	2200	1073	77	4
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales	171	840	13	2868	251	3922	1902	939	2
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales	17253	397	7404	5233	2833	12347	7574	2825	1808
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales	9346	118	1963	861	1360	4286	1965	1203	4922
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales	33	364	1	1240	37	3218	510	229	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales	108	0	13	1446	0	1933	395	5	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales	7300	62	8374	6212	2325	18007	4908	1189	1403
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Verrucomicrobiales	131	12	2757	258	85	1065	125	56	3
d__Bacteria;p__Fibrobacterota;c__Fibrobacteria;o__Fibrobacteriales	1577	15	61	0	0	22	55	72	0
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonadales	232	3	208	367	233	1721	48	113	62
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales	1280	7	307	391	301	1073	325	206	283
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Pirellulales	1101	19	593	270	0	804	438	237	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacteriales	3	258	1	438	56	452	39	103	1
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Acetobacteriales	3	0	2	90	4	369	16	3	0
d__Bacteria;p__Synergistota;c__Synergistia;o__Synergistales	885	16	4	0	0	12	19	10	3
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia	691	0	54	26	0	95	10	156	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales	17	76	8	409	15	862	81	100	3
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales	17	5	0	323	3	478	46	65	1
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales	69	0	26	23	350	174	502	17	170

d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Aeromonadales	31	0	3	0	0	7	708	0	157
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia_vadinBB60_group	560	2	671	767	91	2123	476	63	19
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Streptosporangiales	0	0	1	0	0	85	0	1	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Christensenellales	1221	51	817	421	368	2072	775	199	6
d__Bacteria;p__Armatimonadota;c__uncultured;o__uncultured	240	0	0	0	0	2	0	20	0
d__Bacteria;p__Campilobacterota;c__Campylobacteriia;o__Campylobacteriales	18	0	56	0	15	58	16	1	1
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Opitutales	26	0	73	25	22	162	319	15	0
d__Bacteria;p__Fusobacteriota;c__Fusobacteriia;o__Fusobacteriales	2	0	0	0	283	1	4	2	348
d__Bacteria;p__Bacteroidota;c__Bacteroidia;__	163	0	7	0	2	37	13	44	0
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria;o__Victivallales	832	8	384	218	17	490	66	21	68
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia_UCG-014	322	0	694	477	4	1492	375	78	47
d__Bacteria;p__Cyanobacteria;c__Vampirivibrionia;o__Gastraerophilales	646	6	228	154	12	467	253	38	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Monoglobales	89	0	279	143	41	692	119	12	7
d__Bacteria;p__Firmicutes;__;__	35	0	18	0	0	54	140	9	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales	407	14	33	28	0	277	74	41	5
d__Bacteria;p__WPS-2;c__WPS-2;o__WPS-2	42	0	0	0	0	5	33	52	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales	13	27	1	23	0	202	2	65	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales-Selenomonadales	151	4	3	0	0	7	14	19	13
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales	930	11	114	30	16	395	100	130	84
d__Bacteria;p__Firmicutes;c__Bacilli;o__Acholeplasmatales	323	14	96	25	48	270	169	84	3
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales	9	53	2	43	0	147	9	139	1
d__Archaea;p__Euryarchaeota;c__Methanobacteriia;o__Methanobacteriales	36	0	20	22	15	92	43	1	1
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodobacterales	11	37	0	39	59	213	139	62	3

d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodospirillales	177	2	99	49	5	256	58	42	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Paenibacillales	6	11	0	6	0	209	106	125	0
d__Bacteria;p__Elusimicrobiota;c__Elusimicrobia;o__Elusimicrobiales	61	3	39	39	9	156	40	21	10
d__Archaea;p__Halobacterota;c__Methanomicrobia;o__Methanomicrobiales	74	0	14	19	0	23	0	23	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Streptomycetales	2	0	0	0	0	105	5	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Izermoplasmatales	223	0	24	37	4	152	52	25	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micromonosporales	1	0	0	0	0	23	3	1	0
d__Bacteria;p__Patescibacteria;c__Gracilibacteria;o__Absconditabacteriales_(SR1)	37	0	1	0	0	2	0	67	0
d__Bacteria;p__Firmicutes;c__Clostridia;__	334	0	387	166	14	1198	198	73	0
d__Bacteria;p__Elusimicrobiota;c__Endomicrobia;o__Endomicrobiales	62	0	3	0	0	0	0	15	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__Anaerolineales	19	0	7	0	48	65	6	12	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacterales	4	0	0	3	0	62	6	90	0
d__Bacteria;p__Bdellovibrionota;c__Oligoflexia;o__O319-6G20	0	0	0	0	0	1	0	46	0
d__Bacteria;p__Spirochaetota;c__Spirochaetia;o__Spirochaetales	523	2	88	7	0	85	67	36	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Chloroplast	14	5	5	0	0	63	6	14	7
d__Bacteria;__;__;__	277	3	72	4	3	238	50	55	8
d__Bacteria;p__Firmicutes;c__Clostridia;o__Eubacteriales	43	0	11	13	2	40	5	7	0
d__Bacteria;p__Myxococcota;c__Myxococcia;o__Myxococcales	22	0	10	0	0	2	0	7	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__uncultured	1	13	0	0	0	2	0	14	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Tepidisphaerales	21	0	6	40	0	69	3	4	0
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria;o__Oligosphaerales	36	0	2	0	0	2	0	16	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__uncultured	9	0	19	6	0	53	8	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Reyranellales	0	0	0	0	0	4	0	14	0

d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Pseudonocardiales	1	0	0	2	0	16	0	0	0
d__Bacteria;p__Spirochaetota;c__MVP-15;o__MVP-15	0	0	0	0	0	0	0	24	0
d__Archaea;p__Halobacterota;c__Methanosarcinia;o__Methanosarciniales	11	0	0	0	0	1	0	2	0
d__Archaea;p__Thermoplasmatota;c__Thermoplasma;o__Methanomassiliicoccales	67	9	8	4	0	19	19	8	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Acidaminococcales	253	7	65	10	0	123	62	12	8
d__Bacteria;p__Actinobacteriota;c__Acidimicrobiia;o__Microtrichales	3	0	0	0	0	12	0	26	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Planctomycetales	7	0	4	0	0	30	4	28	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Mycoplasmatales	51	0	3	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;__	13	0	0	4	0	68	8	6	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Bacillales	33	2	1	2	2	325	92	140	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptococcales	5	0	8	0	0	13	6	2	2
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;__	7	0	3	2	0	24	7	2	0
d__Bacteria;p__Firmicutes;c__Incertae_Sedis;o__DTU014	0	0	4	0	0	6	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales	54	0	12	4	0	61	12	15	0
d__Bacteria;p__Firmicutes;c__uncultured;o__uncultured	5	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Limnochordia;o__Limnochordia	6	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Enterobacterales	35	0	0	0	11	44	29	6	1
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales	35	0	2	0	0	4	9	5	0
d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Thermomicrobiales	14	0	0	0	0	13	0	1	2
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Actinomycetales	42	0	0	0	0	4	1	0	1
d__Bacteria;p__SAR324_clade(Marine_group_B);c__SAR324_clade(Marine_group_B);o__SAR324_clade(Marine_group_B)	19	0	0	0	0	5	0	7	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Polyangiales	1	0	0	0	0	4	0	12	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Tistrellales	0	0	0	0	0	3	0	0	0

d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Micavibrionales	1	0	0	0	0	10	0	15	0
d__Bacteria;p__Desulfobacterota;c__Desulfovibrionia;o__Desulfovibrionales	16	0	20	0	0	43	8	3	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Caldicoprobacterales	0	0	0	0	0	5	1	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Isosphaerales	2	0	0	0	0	18	1	12	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Phycisphaerales	4	0	1	0	0	4	0	4	0
d__Bacteria;p__Actinobacteriota;c__Thermoleophilina;o__Solirubrobacterales	1	0	0	0	0	3	0	0	0
d__Bacteria;p__Acidobacteriota;c__Blastocatellia;o__Blastocatellales	5	0	1	0	0	7	0	4	0
d__Bacteria;p__Spirochaetota;c__Leptospirae;o__Leptospirales	3	0	0	0	0	0	0	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Bifidobacteriales	5	0	3	0	0	4	15	0	2
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Frankiales	2	0	1	0	0	15	0	5	0
d__Bacteria;p__Patescibacteria;c__Parcubacteria;o__Candidatus_Nomurabacteria	1	0	0	0	0	0	0	0	5
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Xanthomonadales	3	0	0	2	0	59	9	9	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Azospirillales	1	0	0	3	0	6	0	4	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales	23	12	3	19	10	210	23	25	7
d__Bacteria;p__Deinococcota;c__Deinococci;o__Deinococcales	2	0	0	4	0	5	0	0	4
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Pedosphaerales	4	4	1	0	0	12	3	0	0
d__Bacteria;p__Planctomycetota;c__vadinHA49;o__vadinHA49	2	0	0	0	0	0	0	8	0
d__Bacteria;p__Sumerlaeota;c__Sumerlaeia;o__Sumerlaeales	1	0	0	0	0	2	1	4	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Pseudomonadales	18	4	0	1	0	71	53	48	0
d__Bacteria;p__Bdellovibrionota;c__Bdellovibrionia;o__Bdellovibrionales	0	0	0	0	0	9	0	7	1
Unassigned;__;__;__	30	0	4	0	0	71	1	6	4
d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Kallotenuales	2	0	0	0	0	2	0	0	0
d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Chloroflexales	0	0	0	0	0	1	0	7	0

d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Gemmatales	5	0	2	0	5	36	8	3	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Elsterales	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Desulfotomaculia;o__Desulfotomaculales	2	0	0	0	0	0	0	2	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Paracaeidibacterales	0	0	1	2	0	8	0	3	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Kineospirales	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Firmicutes;c__Syntrophomonadia;o__Syntrophomonadales	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Micropepsales	0	0	0	0	0	2	0	2	0
d__Bacteria;p__Verrucomicrobiota;c__Kiritimatiella;o__WCHB1-41	481	0	6	0	0	30	12	40	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Nannocystales	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae;o__Acidobacteriales	2	0	0	1	0	4	0	0	0
d__Bacteria;p__Patescibacteria;c__Microgenomati;a;o__Candidatus_Pacebacteria	0	0	0	0	0	7	0	0	0
d__Bacteria;p__Abditibacteriota;c__Abditibacteria;o__Abditibacteriales	0	0	0	3	0	10	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Defluviicoccales	0	0	0	0	0	1	0	1	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__RF39	0	0	2	0	0	18	2	1	0
d__Bacteria;p__Armatimonadota;c__Armatimonadia;o__Armatimonadales	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Gemmatimonadota;c__Gemmatimonadetes;o__Gemmatimonadales	0	0	0	0	0	5	0	3	0
d__Bacteria;p__Patescibacteria;c__Dojkabacteria;o__Dojkabacteria	1	0	0	0	0	0	0	2	0
d__Bacteria;p__Bacteroidota;c__Rhodothermia;o__Rhodothermales	2	0	0	2	0	3	0	0	0
d__Bacteria;p__Armatimonadota;c__Fimbriimonadia;o__Fimbriimonadales	1	0	0	0	0	4	0	1	1
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae;o__Bryobacteriales	2	0	0	0	0	0	0	1	0
d__Eukaryota;__;__;__	5	0	1	0	0	5	1	1	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Staphylococcales	3	0	5	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Moorellia;o__Desulfotibacteriales	1	0	0	0	0	0	0	0	0

d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Salinisphaerales	0	0	0	0	0	2	0	3	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;__	2	0	0	0	0	9	1	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__CCD24	1	0	0	0	0	0	0	1	0
d__Bacteria;p__Elusimicrobiota;c__Elusimicrobia;o__Lineage_IV	0	0	0	0	0	0	0	4	0
d__Bacteria;p__Actinobacteriota;c__Acidimicrobiia;__	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Desulfobacterota;c__Desulfuromonadia;o__PB19	1	0	0	0	0	0	0	3	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Ectothiorhodospirales	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Cyanobacteriales	1	0	0	0	0	9	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;__	1	0	0	0	0	0	3	1	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Limnitrachales	0	0	0	0	5	0	0	1	0
d__Bacteria;p__Desulfobacterota;c__Desulfuromonadia;o__Bradymonadales	10	0	1	0	0	3	0	2	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Pseudanabaenales	0	0	0	0	3	0	0	1	0
d__Bacteria;p__Actinobacteriota;c__MB-A2-108;o__MB-A2-108	0	0	0	0	0	2	1	0	0
d__Bacteria;p__Chloroflexi;c__TK10;o__TK10	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Pasteurellales	0	0	0	0	0	0	0	0	1
d__Bacteria;p__Elusimicrobiota;c__Rs-M47;o__Rs-M47	3	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Cellvibrionales	0	0	0	0	0	3	1	5	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__S-BQ2-57_soil_group	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae;o__Acidobacteriae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Kapabacteria;o__Kapabacteriales	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Diplorickettsiales	0	0	0	0	0	1	0	1	0
d__Bacteria;p__Acidobacteriota;c__Vicinamibacteriia;o__Vicinamibacteriales	2	0	0	0	0	2	0	2	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__SBR1031	1	0	0	0	0	1	0	0	1

d__Bacteria;p__Patescibacteria;c__ABY1;o__Can didatus_Kerfeldbacteria	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria ;o__Nitriliruptorales	2	0	0	0	0	2	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__B acteroidetes_VC2.1_Bac22	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__KD4-96;o__KD4-96	2	0	0	0	0	3	1	0	0
d__Bacteria;p__Bacteroidota;c__Rhodothermia;o__ _Balneolales	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Chloroflexi;c__JG30-KF- CM66;o__JG30-KF-CM66	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__C aldilineales	2	0	2	0	0	4	0	1	0
d__Bacteria;p__Patescibacteria;c__ABY1;o__Can didatus_Falkowbacteria	1	0	0	0	0	0	0	0	0
d__Archaea;p__Aenigmarchaeota;c__Deep_Sea_ Euryarchaeotic_Group(DSEG);o__Deep_Sea_Eury archaeotic_Group(DSEG)	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia; o__OPB41	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Verrucomicrobiota;__;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteob acteria;o__Oceanospirillales	0	0	0	0	0	0	0	2	0
d__Bacteria;p__Chloroflexi;c__Dehalococcoidia;o__ _S085	1	0	0	0	0	1	1	2	0
d__Bacteria;p__Cyanobacteria;c__Vampirivibrionia ;o__Obscuribacterales	0	0	0	0	4	0	0	0	0
d__Bacteria;p__Deferribacterota;c__Deferribactere s;o__Deferribacterales	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia; o__Synechococcales	0	0	0	0	0	3	0	1	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobac teria;o__Puniceispirillales	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Planctomycetota;c__OM190;o__O M190	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Myxococcota;__;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;__;__	3	0	0	0	0	0	1	0	0
d__Bacteria;p__Chloroflexi;c__Ktedonobacteria;o__ _C0119	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Armatimonadota;c__Chthonomona detes;o__Chthonomonadales	1	0	0	0	0	0	0	0	0

d__Bacteria;p__Proteobacteria;__;__	1	0	0	0	0	2	0	1	0
d__Bacteria;p__Patescibacteria;c__Parcubacteria;o__Candidatus_Moranbacteria	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Thermoleophil ia;o__Gaiellales	1	0	1	0	0	0	0	0	1
d__Bacteria;p__Acidobacteriota;c__Subgroup_5;o__ Subgroup_5	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Acidobacteriota;c__Subgroup_18; o__Subgroup_18	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__S JA-15	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Planctomycetota;c__Pla4_lineage; o__Pla4_lineage	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Omnitrophia ;o__Omnitrophales	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteob acteria;o__Alteromonadales	0	0	0	0	4	3	0	0	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae ;o__Elev-16S-1166	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobac teria;o__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Hydrogenedentes;c__Hydrogened entia;o__Hydrogenedentiales	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__Dehalococcoidia;o__ SAR202_clade	0	0	0	0	0	1	0	1	0
d__Bacteria;p__Gemmatimonadota;c__S0134_terr estrial_group;o__S0134_terrestrial_group	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Patescibacteria;c__Gracilibacteria; o__JGI_0000069-P22	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__un cultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__ml e1-27	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaera e;o__Pla1_lineage	0	0	0	0	0	1	0	0	0
d__Eukaryota;p__Ascomycota;__;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae ;o__Solibacterales	0	0	0	0	0	1	0	0	0
d__Eukaryota;p__Arthropoda;__;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicr obiae;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;__;__	0	0	0	0	0	2	0	0	0

d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Oxyphotobacteria_Incertae_Sedis	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Patescibacteria;c__Berkelbacteria;o__Berkelbacteria	0	0	1	0	0	1	0	0	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Blf di19	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Spirochaetota;c__Brachyspirae;o__Brachyspirales	0	0	0	0	0	0	0	0	1
d__Bacteria;p__Acidobacteriota;c__Blastocatellia;o__DS-100	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__S-70	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Cyanobacteria;__;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Kiloniellales	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Patescibacteria;c__uncultured;o__uncultured	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__LD1-PB3	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Bacteroidota;c__SJA-28;o__SJA-28	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Parvibaculales	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Alicyclobacillales	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Gammaproteobacteria_Incertae_Sedis	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Verrucomicrobiota;c__Chlamydiae;o__Chlamydiales	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Patescibacteria;c__WWE3;o__WWE3	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Acidobacteriota;c__Aminicenantia;o__Aminicenantales	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__Gitt-GS-136;o__Gitt-GS-136	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Chloroflexi;__;__	0	0	0	0	0	1	0	0	0

Family

index	Elephant_captive	Elephant_Wild	Domestic_livestock	Hog_deer	Sambar_deer	Spotted_deer	Wild_Boar	Rhino	Hu_mahout
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonadales;f__Sphingomonadaceae	19	113	1	689	9	2828	91	101	4
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Micrococcaceae	86	7	6	508	7	1475	860	56	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Weeksellaceae	107	36	2	827	90	1271	169	218	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__p-251-o5	1660	36	28	186	0	60	2	42	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae	9226	111	1934	815	1360	4199	1954	1165	4921
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales;f__Sphingobacteriaceae	32	364	1	1240	37	3214	510	226	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Nocardiaceae	4	0	0	1429	0	1870	380	3	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae	3721	43	3501	2189	1580	7196	2573	574	612
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Flavobacteriaceae	61	785	11	2041	161	2635	1732	626	1
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Verrucomicrobiales;f__Akermansiaceae	130	3	2757	256	85	974	125	2	3
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__uncultured	143	0	872	841	0	1604	84	0	0
d__Bacteria;p__Fibrobacterota;c__Fibrobacteria;o__Fibrobacteriales;f__Fibrobacteraceae	1577	15	61	0	0	22	55	72	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Rikenellaceae	5393	111	2431	2037	366	4743	1848	661	150
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prevotellaceae	3257	186	1693	980	731	2664	3250	757	629
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonadales;f__Saccharimonadaceae	220	3	206	365	233	1655	39	100	51
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae	1035	7	1057	677	344	2075	800	214	560
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidales_BS11_group	647	18	1	0	0	4	0	86	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcaceae	7	0	69	217	102	271	45	1	271
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Pirellulales;f__Pirellulaceae	1101	19	593	270	0	804	438	237	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Paludibacteraceae	1843	5	74	0	0	174	34	22	0

d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__UCG-010	1452	3	2544	2416	306	6230	743	204	19
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacteriales;f__Caulobacteraceae	3	258	1	438	53	446	39	102	1
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Acetobacteriales;f__Acetobacteraceae	3	0	2	90	4	369	16	3	0
d__Bacteria;p__Synergistota;c__Synergistia;o__Synergistales;f__Synergistaceae	885	16	4	0	0	12	19	10	3
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia;f__Hungateiclostridiaceae	690	0	54	26	0	95	10	156	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidaceae	262	0	954	641	1006	1522	843	2	844
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae	12	14	4	344	8	610	27	35	2
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales;f__Nocardioideaceae	14	5	0	323	3	475	46	65	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__M2PB4-65_termite_group	29	0	15	0	0	259	0	22	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;f__Clostridiaceae	68	0	26	23	305	170	501	16	170
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__[Eubacterium]_coprostanoligenes_group	849	2	983	781	75	1766	608	156	193
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Aeromonadales;f__Succinivibrionaceae	30	0	3	0	0	7	707	0	157
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidales_RF16_group	1240	15	786	10	0	442	272	87	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Promicromonosporaceae	0	0	0	22	0	102	66	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidales_UCG-001	624	11	55	0	0	2	0	154	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Dysgonomonadaceae	626	0	12	0	0	86	9	46	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Microbacteriaceae	12	0	1	43	0	299	31	14	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__F082	1051	7	121	161	0	148	30	701	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Cellulomonadaceae	6	0	0	166	0	264	56	3	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Anaerovoracaceae	1235	7	235	174	169	779	280	203	11
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia_vadinBB60_group;f__Clostridia_vadinBB60_group	560	2	671	767	91	2123	476	63	19
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Streptosporangiales;f__Thermomonosporaceae	0	0	1	0	0	77	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Christensenellales;f__Christensenellaceae	1221	51	817	421	368	2072	775	199	6

d__Bacteria;p__Armatimonadota;c__uncultured;o__uncultured;f__uncultured	240	0	0	0	0	2	0	20	0
d__Bacteria;p__Campilobacterota;c__Campylobacteria;o__Campylobacteriales;f__Campylobacteraceae	11	0	56	0	15	57	15	1	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Muribaculaceae	14	0	138	207	259	264	754	7	2
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Butyrivibrionaceae	32	0	113	75	14	288	99	4	10
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Opitutales;f__Puniceicoccaceae	26	0	72	25	22	150	319	4	0
d__Bacteria;p__Fusobacteriota;c__Fusobacteriia;o__Fusobacteriales;f__Fusobacteriaceae	2	0	0	0	283	0	4	2	348
d__Bacteria;p__Bacteroidota;c__Bacteroidia;__;	163	0	7	0	2	37	13	44	0
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria;o__Victivallales;f__vadinBE97	487	6	108	38	3	75	21	20	8
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia_UCG-014;f__Clostridia_UCG-014	322	0	694	477	4	1492	375	78	47
d__Bacteria;p__Cyanobacteria;c__Vampirivibrionia;o__Gastranaerophilales;f__Gastranaerophilales	646	6	228	154	12	467	253	38	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Monoglobales;f__Monoglobaceae	89	0	279	143	41	692	119	12	7
d__Bacteria;p__Firmicutes;__;__;__	35	0	18	0	0	54	140	9	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Eggerthellaceae	241	8	28	19	0	162	19	21	2
d__Bacteria;p__WPS-2;c__WPS-2;o__WPS-2;f__WPS-2	42	0	0	0	0	5	33	52	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidetes_BD2-2	58	3	0	0	0	1	0	50	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidales	0	0	0	0	0	43	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;__	356	5	102	37	0	101	80	120	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Spirosomaceae	2	27	0	22	0	151	0	43	0
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria;o__Victivallales;f__Victivallaceae	345	2	276	180	14	415	45	1	60
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales-Selenomonadales;f__Selenomonadaceae	151	4	3	0	0	6	13	18	4
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__p-2534-18B5_gut_group	0	0	34	0	0	16	259	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelatoclostridiaceae	546	4	65	20	4	233	24	56	65
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Xanthobacteraceae	1	0	1	9	0	50	1	8	0

d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Defluviitaleaceae	118	7	27	46	0	84	9	35	1
d__Bacteria;p__Firmicutes;c__Bacilli;o__Acholeplasmatales;f__Acholeplasmataceae	323	14	96	25	48	270	169	84	3
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae	6	53	1	43	0	146	9	125	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Tannerellaceae	8	0	6	0	418	23	96	1	115
d__Archaea;p__Euryarchaeota;c__Methanobacteria;o__Methanobacteriales;f__Methanobacteriaceae	36	0	20	22	15	92	43	1	1
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodobacterales;f__Rhodobacteraceae	11	37	0	39	59	213	139	62	3
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodospirillales;f__uncultured	177	2	99	49	2	253	58	41	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__	178	7	150	70	6	392	51	26	7
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Barnesiellaceae	5	0	64	110	35	133	9	0	14
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Crocinitomicaceae	2	19	0	0	0	14	1	92	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Paenibacillales;f__Paenibacillaceae	6	11	0	6	0	209	106	125	0
d__Bacteria;p__Elusimicrobiota;c__Elusimicrobia;o__Elusimicrobiales;f__Elusimicrobiaeae	61	3	39	39	9	156	40	21	10
d__Archaea;p__Halobacterota;c__Methanomicrobia;o__Methanomicrobiales;f__Methanocorpusculaceae	74	0	14	19	0	23	0	20	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Streptomycetales;f__Streptomycetaceae	2	0	0	0	0	105	5	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Izomoplasmatales;f__Izomoplasmataceae	56	0	24	37	4	151	52	17	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Coriobacteriales_Incertae_Sedis	117	0	1	4	0	20	1	6	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micromonosporales;f__Micromonosporaceae	1	0	0	0	0	23	3	1	0
d__Bacteria;p__Patescibacteria;c__Gracilibacteria;o__Absconditabacteriales_(SR1);f__Absconditabacteriales_(SR1)	37	0	1	0	0	2	0	67	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Bogoriellaceae	2	0	0	0	0	5	43	0	3
d__Bacteria;p__Firmicutes;c__Clostridia;f__	334	0	387	166	14	1198	198	73	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Izomoplasmatales;f__Izomoplasmataceae	166	0	0	0	0	1	0	8	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Mycobacteriaceae	2	0	1	17	0	58	2	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Beijerinckiacae	1	0	1	19	0	91	7	4	1

d__Bacteria;p__Elusimicrobiota;c__Endomicrobia;o__Endomicrobiales;f__Endomicrobiaceae	62	0	3	0	0	0	0	15	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Verrucomicrobiales;f__Verrucomicrobiaceae	1	9	0	2	0	64	0	52	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__Anaerolineales;f__Anaerolineaceae	19	0	7	0	48	65	6	12	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Devosiaceae	3	62	1	28	7	74	46	29	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacterales;f__Xiphinematobacteraceae	2	0	0	0	0	6	2	75	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__uncultured	16	6	1	0	0	16	44	11	0
d__Bacteria;p__Bdellovibrionota;c__Oligoflexia;o__0319-6G20;f__0319-6G20	0	0	0	0	0	1	0	46	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Atopobiaceae	0	0	3	5	0	68	8	0	0
d__Bacteria;p__Spirochaetota;c__Spirochaetia;o__Spirochaetales;f__Spirochaetaceae	523	2	88	7	0	85	67	36	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Chloroplast;f__Chloroplast	14	5	5	0	0	63	6	14	7
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__COB_P4-1_termite_group	0	0	4	0	0	9	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Marinifilaceae	33	0	13	23	15	44	4	28	53
d__Bacteria;__;__;__	277	3	72	4	3	238	50	55	8
d__Bacteria;p__Firmicutes;c__Clostridia;o__Eubacteriales;f__Eubacteriaceae	29	0	0	0	0	3	0	2	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacterales;f__Terrimicrobiaceae	1	0	0	0	0	36	1	13	0
d__Bacteria;p__Myxococcota;c__Myxococcia;o__Myxococcales;f__Myxococcaceae	22	0	10	0	0	2	0	6	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__uncultured;f__uncultured	1	13	0	0	0	2	0	14	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Streptosporangiales;f__Streptosporangiaceae	0	0	0	0	0	7	0	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__CAP-aah99b04	0	0	0	0	0	1	0	16	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Microscillaceae	1	0	1	0	0	8	0	17	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae	367	7	43	10	12	141	71	70	19
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__D05-2	0	0	0	4	0	13	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Tepidisphaerales;f__WD2101_soil_group	12	0	5	37	0	47	3	4	0

d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Corynebacteriaceae	99	0	12	0	0	4	5	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ethanoligenenaceae	17	0	0	0	0	2	0	5	0
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria;o__Oligosphaerales;f__Oligosphaeraeae	36	0	2	0	0	2	0	16	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Beutenbergiaceae	2	0	0	8	0	7	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Amb-16S-1323	0	0	0	0	0	0	0	12	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__uncultured;f__uncultured	9	0	19	6	0	53	8	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Reyranellales;f__Reyranellaceae	0	0	0	0	0	4	0	14	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospirales	7	0	23	4	0	35	31	5	2
d__Bacteria;p__Firmicutes;c__Clostridia;o__Eubacteriales;f__Anaerofustaceae	13	0	11	13	2	37	5	4	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Intrasporangiaceae	22	0	0	0	0	6	1	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Pseudonocardiales;f__Pseudonocardiaceae	1	0	0	2	0	16	0	0	0
d__Bacteria;p__Spirochaetota;c__MVP-15;o__MVP-15;f__MVP-15	0	0	0	0	0	0	0	24	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Micrococcales_Incertae_Sedis	1	0	0	0	0	5	6	2	0
d__Archaea;p__Halobacterota;c__Methanosarcinia;o__Methanosarcinales;f__Methanosarcinaceae	11	0	0	0	0	1	0	2	0
d__Archaea;p__Thermoplasmatota;c__Thermoplasmata;o__Methanomassiliococcales;f__Methanomethylophilaceae	67	9	8	4	0	19	19	8	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Acidaminococcales;f__Acidaminococcaceae	253	7	65	10	0	123	62	12	8
d__Bacteria;p__Actinobacteriota;c__Acidimicrobiia;o__Microtrichales;f__Microtrichaceae	1	0	0	0	0	1	0	17	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Planctomycetales;f__Rubinisphaeraceae	1	0	1	0	0	16	2	25	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Methylophilaceae	0	0	0	0	0	6	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Mycoplasmatales;f__Mycoplasmataceae	51	0	3	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcales-Tissierellales	26	0	1	0	0	8	0	0	1
d__Bacteria;p__Firmicutes;c__Bacilli;__;__	13	0	0	4	0	68	8	6	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__uncultured	9	0	1	0	0	5	5	3	0

d__Bacteria;p__Firmicutes;c__Bacilli;o__Bacillales;f__Planococcaceae	8	2	1	2	0	141	50	95	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiales_Incertae_Sedis	0	0	1	0	0	2	0	8	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Tepidisphaerales;f__Tepidisphaerae	6	0	1	3	0	21	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Marinilabiliaceae	0	0	0	0	3	2	0	23	0
d__Bacteria;p__Campilobacterota;c__Campylobacteriales;f__Arcobacteraceae	7	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptococcales;f__Peptococcaceae	5	0	8	0	0	13	6	2	2
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Sedimentibacteraceae	9	0	0	0	17	4	0	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;f__	7	0	3	2	0	24	7	2	0
d__Bacteria;p__Firmicutes;c__Incertae_Sedis;o__DTU014;f__DTU014	0	0	4	0	0	6	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Kaistiaceae	0	0	0	0	0	5	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales;f__Mitochondria	44	0	8	1	0	23	7	6	0
d__Bacteria;p__Firmicutes;c__uncultured;o__uncultured;f__uncultured	5	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales;f__uncultured	10	0	3	3	0	24	5	4	0
d__Bacteria;p__Firmicutes;c__Limnochordia;o__Limnochordia;f__Limnochordia	6	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Enterobacterales;f__Enterobacteriaceae	34	0	0	0	11	43	29	6	1
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Carnobacteriaceae	7	0	1	0	0	0	0	0	0
d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Thermomicrobiales;f__JG30-KF-CM45	14	0	0	0	0	12	0	1	2
d__Bacteria;p__Actinobacteriota;c__Coriobacterii;o__Coriobacteriales;f__	31	0	0	0	0	10	1	3	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__uncultured	7	0	1	0	0	4	1	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Dietziaceae	3	0	0	0	0	1	8	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Sanguibacteraceae	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Actinomycetales;f__Actinomycetaceae	42	0	0	0	0	4	1	0	1
d__Bacteria;p__SAR324_clade(Marine_group_B);c__SAR324_clade(Marine_group_B);o__SAR324_clade(Marine_group_B);f__SAR324_clade(Marine_group_B)	19	0	0	0	0	5	0	7	0

d__Bacteria;p__Myxococcota;c__Polyangia;o__Polyangiales;f__Polyangiaceae	0	0	0	0	0	3	0	6	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Tistrellales;f__Geminococcaceae	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__[Clostridium]__methylopentose_group	0	0	2	0	0	14	2	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__A0839	0	0	0	1	0	3	0	0	0
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonadales;__	4	0	1	2	0	30	0	2	4
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Mycovibrionales;f__uncultured	1	0	0	0	0	10	0	15	0
d__Bacteria;p__Desulfobacterota;c__Desulfovibrionia;o__Desulfovibrionales;f__Desulfovibrionaceae	16	0	20	0	0	43	8	3	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Polyangiales;f__Sandaracinaceae	0	0	0	0	0	0	0	4	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Caldicoprobacteriales;f__Caldicoprobacteraceae	0	0	0	0	0	5	1	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Saprospiraceae	1	0	1	0	0	0	0	6	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Isosphaerales;f__Isosphaeraceae	2	0	0	0	0	18	1	12	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;__	0	0	0	0	45	2	1	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Hymenobacteraceae	5	0	0	1	0	22	2	1	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Phycisphaerales;f__Phycisphaeraceae	4	0	1	0	0	4	0	4	0
d__Bacteria;p__Actinobacteriota;c__Thermoeophilii;o__Solirubrobacterales;f__Solirubrobacteraceae	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Acidobacteriota;c__Blastocatellia;o__Blastocatellales;f__Blastocatellaceae	5	0	1	0	0	7	0	4	0
d__Bacteria;p__Spirochaetota;c__Leptospirae;o__Leptospirales;f__Leptospiraceae	3	0	0	0	0	0	0	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Bifidobacteriales;f__Bifidobacteriaceae	5	0	3	0	0	4	15	0	2
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Frankiales;f__Geodermatophilaceae	1	0	0	0	0	7	0	1	0
d__Bacteria;p__Patescibacteria;c__Parcubacteria;o__Candidatus_Nomurabacteria;f__Candidatus_Nomurabacteria	1	0	0	0	0	0	0	0	5
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Xanthomonadales;f__Xanthomonadaceae	2	0	0	2	0	57	9	9	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Azospirillales;f__Azospirillaceae	0	0	0	3	0	6	0	4	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Alcaligenaceae	15	0	0	1	0	51	0	1	0

d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Cyclobacteriaceae	5	0	0	0	0	9	0	2	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Dermabacteraceae	6	0	0	0	0	0	0	0	0
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonadales;f__LWQ8	2	0	0	0	0	13	3	4	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Opitutales;f__Opitutaceae	0	0	1	0	0	12	0	11	0
d__Bacteria;p__Deinococcota;c__Deinococci;o__Deinococcales;f__Deinococcaceae	2	0	0	4	0	3	0	0	4
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Pedosphaerales;f__Pedosphaeraceae	4	4	1	0	0	12	3	0	0
d__Bacteria;p__Planctomycetota;c__vadinHA49;o__vadinHA49;f__vadinHA49	2	0	0	0	0	0	0	8	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacterales;f__Chthoniobacteraceae	1	0	0	3	0	16	3	2	0
d__Archaea;p__Halobacterota;c__Methanomicrobia;o__Methanomicrobiales;f__Methanomicrobiaceae	0	0	0	0	0	0	0	3	0
d__Bacteria;p__Sumerlaeota;c__Sumerlaeae;o__Sumerlaeales;f__Sumerlaeaceae	1	0	0	0	0	2	1	4	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Pseudomonadales;f__Moraxellaceae	12	4	0	1	0	57	48	39	0
d__Bacteria;p__Bdellovibrionota;c__Bdellovibrionia;o__Bdellovibrionales;f__Bdellovibrionaceae	0	0	0	0	0	9	0	7	1
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonadales;f__Saccharimonadales	6	0	1	0	0	23	5	7	7
Unassigned;__;__;__	30	0	4	0	0	71	1	6	4
d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Kallotenuales;f__AKIW781	2	0	0	0	0	2	0	0	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales-Selenomonadales;f__Veillonellaceae	0	0	0	0	0	0	1	1	9
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;__	8	0	5	0	0	16	0	1	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteria;o__Coriobacteriales;f__Coriobacteriaceae	0	0	0	0	0	1	0	0	3
d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Chloroflexales;f__Roseiflexaceae	0	0	0	0	0	1	0	3	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Gemmatales;f__Gemmataceae	5	0	2	0	5	36	8	3	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Frankiales;f__Nakamurellaceae	1	0	1	0	0	6	0	4	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacterales;f__Hyphomonadaceae	0	0	0	0	3	6	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Oxalobacteraceae	1	2	0	11	0	71	1	9	1

d__Bacteria;p__Actinobacteriota;c__Acidimicrobiia;o__Microtrichales;f__	0	0	0	0	0	0	0	2	0
d__Bacteria;p__Abditibacteriota;c__Abditibacteriae;o__Abditibacteriales;f__Abditibacteriaceae	0	0	0	3	0	10	0	0	0
d__Bacteria;p__Actinobacteriota;c__Acidimicrobiia;o__Microtrichales;f__Lamiaceae	0	0	0	0	0	4	0	2	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Sutterellaceae	2	0	1	4	0	13	4	0	6
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Pseudomonadales;f__Pseudomonadaceae	6	0	0	0	0	14	5	9	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Deφυluicoccales;f__uncultured	0	0	0	0	0	1	0	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales;f__SM2D12	0	0	0	0	0	6	0	2	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__RF39;f__RF39	0	0	2	0	0	18	2	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__uncultured	1	0	0	0	0	0	0	3	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Bacillales;f__Bacillaceae	17	0	0	0	2	111	26	18	0
d__Bacteria;p__Armatimonadota;c__Armatimonadota;o__Armatimonadales;f__Armatimonadaceae	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Gemmatimonadota;c__Gemmatimonadetes;o__Gemmatimonadales;f__Gemmatimonadaceae	0	0	0	0	0	5	0	3	0
d__Bacteria;p__Patescibacteria;c__Dojkabacteria;o__Dojkabacteria;f__Dojkabacteria	1	0	0	0	0	0	0	2	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Polyangiales;f__Blrii41	1	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Stappiaceae	0	0	0	4	0	2	0	0	0
d__Bacteria;p__Bacteroidota;c__Rhodothermia;o__Rhodothermales;f__Rhodothermaceae	2	0	0	2	0	3	0	0	0
d__Bacteria;p__Armatimonadota;c__Fimbriimonadota;o__Fimbriimonadales;f__Fimbriimonadaceae	1	0	0	0	0	4	0	1	1
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Fusibacteraceae	0	0	0	0	13	2	0	0	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae;o__Bryobacteriales;f__Bryobacteraceae	2	0	0	0	0	0	0	1	0
d__Eukaryota;f__	5	0	1	0	0	5	1	1	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Staphylococcales;f__Staphylococcaceae	3	0	5	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__UCG-011	2	0	0	0	0	5	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales;f__Propionibacteriaceae	3	0	0	0	0	3	0	0	1

d__Bacteria;p__Firmicutes;c__Moorellia;o__Desulfotribacteriales;f__Desulfotribacteraceae	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Salinisphaerales;f__Solimonadaceae	0	0	0	0	0	2	0	3	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;__;__	2	0	0	0	0	9	1	1	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Bacillales;__	8	0	0	0	0	73	16	27	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__CCD24;f__CCD24	1	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodospirillales;f__Rhodospirillaceae	0	0	0	0	3	3	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Planctomycetales;f__uncultured	5	0	3	0	0	11	2	1	0
d__Bacteria;p__Elusimicrobiota;c__Elusimicrobia;o__Lineage_IV;f__Lineage_IV	0	0	0	0	0	0	0	4	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__NS9_marine_group	0	0	0	0	0	0	0	2	0
d__Bacteria;p__Actinobacteriota;c__Acidimicrobiia;__;__	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Desulfobacterota;c__Desulfurimonadia;o__PB19;f__PB19	1	0	0	0	0	0	0	3	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__37-13	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__MWH-CFBk5	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Ectothiorhodospirales;f__Thiobacillaceae	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Cyanobacteriales;f__Chroococcidiopsaceae	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;__;__	1	0	0	0	0	0	3	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Cytophagaceae	0	0	0	0	0	8	0	2	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Limnotrichales;f__Limnotrichaceae	0	0	0	0	5	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Rhodocyclaceae	1	0	0	0	8	1	0	0	0
d__Bacteria;p__Desulfobacterota;c__Desulfurimonadia;o__Bradymonadales;f__Bradymonadales	10	0	1	0	0	3	0	2	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Methylobacteriaceae	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;__	3	0	2	0	0	8	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales;f__Candidatus_Hepaticola	0	0	0	0	0	1	0	0	0

d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Oceanospirillales;__	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Chloroflexi;c__Dehalococcoidia;o__S085;f__S085	1	0	0	0	0	1	1	2	0
d__Bacteria;p__Cyanobacteria;c__Vampirivibrionia;o__Obscuribacterales;f__Obscuribacteraceae	0	0	0	0	4	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prolixibacteraceae	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Enterococcaceae	0	0	0	0	0	0	3	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Aeromonadales;f__Aeromonadaceae	1	0	0	0	0	0	1	0	0
d__Bacteria;p__Deferribacterota;c__Deferribacteres;o__Deferribacterales;f__Deferribacteraceae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Thermotaleaceae	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Synechococcales;f__Cyanobiaceae	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;__	1	0	2	0	0	3	4	1	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;__;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;__	1	0	0	0	0	2	0	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Puniceispirillales;f__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Dermatophilaceae	0	0	0	0	0	3	1	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacterales;__	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Aerococcaceae	5	0	1	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;c__OM190;o__OM190;f__OM190	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Izermoplasmatales;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacterales;f__Coriobacteriales	2	0	0	0	0	0	1	0	0
d__Bacteria;p__Myxococcota;__;__;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;__;__;__	3	0	0	0	0	0	1	0	0
d__Bacteria;p__Firmicutes;c__Desulfotomaculalia;o__Desulfotomaculales;f__Desulfall-Sporotomaculum	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Chloroflexi;c__Ktedonobacteria;o__C0119;f__C0119	1	0	0	0	0	0	0	0	0

d__Bacteria;p__Patescibacteria;c__Gracilibacteria;o__JGI_0000069-P22;f__JGI_0000069-P22	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__uncultured;f__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__mle1-27;f__mle1-27	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Planctomycetales;f__Gimesiaceae	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Pla1_lineage;f__Pla1_lineage	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Streptosporangiales;f__Nocardiopsaceae	0	0	0	0	0	1	0	0	0
d__Eukaryota;p__Ascomycota;__;__;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae;o__Solibacterales;f__Solibacteraceae	0	0	0	0	0	1	0	0	0
d__Eukaryota;p__Arthropoda;__;__;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;__;__;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales;f__AB1	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacterales;f__01D2Z36	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;__;__;__	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteria;o__Cyanobacteriales;f__Phormidiaceae	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteria;o__Oxyphotobacteria_Incertae_Sedis;f__Unknown_Family	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteria;o__Cyanobacteriales;f__Coleofasciculaceae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Patescibacteria;c__Berkelbacteria;o__Berkelbacteria;f__Berkelbacteria	0	0	1	0	0	1	0	0	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Blfdi19;f__Blfdi19	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Spirochaetota;c__Brachyspirae;o__Brachyspirales;f__Brachyspiraceae	0	0	0	0	0	0	0	0	1
d__Bacteria;p__Actinobacteriota;c__Thermoeophila;o__Gaiellales;f__uncultured	0	0	0	0	0	0	0	0	1
d__Bacteria;p__Acidobacteriota;c__Blastocatella;o__DS-100;f__DS-100	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Enterobacterales;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Tepidisphaerales;f__Tepidisphaerales	2	0	0	0	0	0	0	0	0

d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__S-70;f__S-70	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Azospirillales;f__uncultured	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Eubacteriales;f__Eubacteriales	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Cyanobacteria;__;__;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Xanthomonadales;f__Rhodanobacteraceae	1	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Dermacoccaceae	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Kiloniellales;f__Fodinicurvataceae	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Patescibacteria;c__uncultured;o__uncultured;f__uncultured	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Oceanospirillales;f__Saccharospirillaceae	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__LD1-PB3;f__LD1-PB3	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Synechococcales;f__Synechococceae	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Bacteroidota;c__SJA-28;o__SJA-28;f__SJA-28	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Chitinimonadaceae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Parvibaculales;f__Parvibaculaceae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Fusobacteriota;c__Fusobacteriia;o__Fusobacteriales;f__Leptotrichiaceae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Alicyclobacillales;f__Alicyclobacillaceae	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Myxococcota;c__Myxococcia;o__Myxococcales;f__Anaeromyxobacteraceae	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Gammaproteobacteria_Incertae_Sedis;f__Unknown_Family	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Verrucomicrobiota;c__Chlamydiae;o__Chlamydiales;f__Chlamydiaceae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Acidaminobacteraceae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Patescibacteria;c__WWE3;o__WWE3;f__WWE3	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Acidobacteriota;c__Aminicenantia;o__Aminicenantales;f__Aminicenantales	0	0	0	0	0	1	0	0	0

d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Thermomicrobiales;f__AKYG1722	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__Gitt-GS-136;o__Gitt-GS-136;f__Gitt-GS-136	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Burkholderiaceae	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Amoebophilaceae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Xanthomonadales;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Enterobacterales;f__Erwiniaaceae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Frankiales;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;__;__;__	0	0	0	0	0	1	0	0	0

Genus

index	Elephant_captive	Elephant_Wild	Domestic_livestock	Hog_deer	Sambar_deer	Spotted_deer	Wild_Boar	Rhino	Hu_mahout
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonadales;f__Sphingomonadales;g__Sphingomonas	9	0	0	561	3	1789	74	11	1
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Micrococcaceae;g__Arthrobacter	1	7	1	326	7	1122	736	41	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Weeksellaceae;g__Chryseobacterium	11	4	0	816	90	1258	10	46	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__p-251-o5;g__p-251-o5	1660	36	28	186	0	60	2	42	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_AC2044_group	1069	0	11	0	0	7	1	143	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales;f__Sphingobacteriaceae;g__Sphingobacterium	17	316	1	641	37	2449	490	146	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Nocardiaceae;g__Gordonia	1	0	0	503	0	561	5	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__UCG-005	804	11	2467	1705	1224	4673	1700	110	22
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Agathobacter	66	0	0	0	0	0	2	1	1726
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonadales;f__Sphingomonadales;g__Novosphingobium	1	0	0	17	0	538	2	4	1
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Nocardiaceae;g__Rhodococcus	0	0	0	564	0	485	370	2	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Flavobacteriaceae;g__Flavobacterium	52	785	4	1996	161	2631	1731	608	1
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Micrococcaceae;g__	4	0	0	182	0	341	107	14	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Verrucomicrobiales;f__Akkermansiaceae;g__Akkermansia	130	3	2757	256	85	974	125	2	3
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Nocardiaceae;g__Williopsis	3	0	0	352	0	494	0	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__uncultured;g__uncultured	143	0	872	841	0	1604	84	0	0
d__Bacteria;p__Fibrobacterota;c__Fibrobacteria;o__Fibrobacteriales;f__Fibrobacteraceae;g__Fibrobacter	1577	15	61	0	0	22	55	56	0

d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Rikenellaceae;g__Alistipes	85	0	698	618	103	1889	155	0	148
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prevotellaceae;g__Prevotellaceae_NK3B31_group	1	0	1	0	0	4	1640	0	0
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonadales;f__Saccharimonadaceae;g__Candidatus_Saccharimonas	210	3	204	361	233	1544	36	85	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__uncultured	198	7	305	414	133	930	138	43	35
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Rikenellaceae;g__Rikenellaceae_RC9_gut_group	4763	97	1515	1161	175	2414	1567	630	2
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidales_BS11_gut_group;g__Bacteroidales_BS11_gut_group	647	18	1	0	0	4	0	86	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcaceae;g__Romboutsia	1	0	34	176	34	183	19	0	255
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prevotellaceae;g__Prevotellaceae_UCG-004	421	5	1477	973	39	1798	114	262	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Pirellulales;f__Pirellulaceae;g__p-1088-a5_gut_group	1042	19	398	251	0	501	361	134	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Dorea	7	0	75	49	16	195	11	0	83
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Paludibacteraceae;g__uncultured	1840	5	74	0	0	165	34	20	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__UCG-010;g__UCG-010	1452	3	2544	2416	306	6230	743	204	19
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prevotellaceae;g__Prevotella	815	26	35	0	287	210	432	48	607
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacteriales;f__Caulobacteraceae;g__Brevundimonas	3	258	1	426	53	390	38	90	1
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Acetobacteriales;f__Acetobacteraceae;__	0	0	0	21	0	162	3	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Nocardiaceae;g__Nocardia	0	0	0	10	0	325	5	0	0
d__Bacteria;p__Synergistota;c__Synergistia;o__Synergistales;f__Synergistaceae;g__uncultured	730	11	1	0	0	3	0	5	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia;f__Hungateiclostridiaceae;g__Saccharofermentans	454	0	11	14	0	17	2	101	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;__	438	8	515	215	97	1046	258	82	4
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidaceae;g__Bacteroides	262	0	954	641	1006	1522	843	2	844
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;__	4912	77	1532	648	995	3128	1231	644	390

d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__Allorhizobium-Neorhizobium-Pararhizobium-Rhizobium	6	14	0	317	8	374	8	27	1
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_XPB1014_group	1034	0	2	13	0	14	0	53	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales;f__Nocardioidaceae;g__Nocardioides	9	5	0	305	3	413	30	58	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonadales;f__Sphingomonadaceae;__	5	29	1	33	0	213	7	28	2
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__M2PB4-65_termite_group;g__M2PB4-65_termite_group	29	0	15	0	0	259	0	22	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;f__Clostridiaceae;__	0	0	0	16	209	71	221	1	1
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Oribacterium	295	0	0	0	0	0	22	28	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__[Eubacterium]_coprostanoligenes_group;g__[Eubacterium]_coprostanoligenes_group	849	2	983	781	75	1766	608	156	193
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Aeromonadales;f__Succinivibrionaceae;g__Succinivibrio	25	0	1	0	0	1	635	0	149
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_NK4A136_group	2	0	6	0	0	5	23	6	702
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales;f__Sphingobacteriaceae;__	0	34	0	38	0	168	10	39	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prevotellaceae;__	364	32	15	4	21	147	468	132	14
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidales_RF16_group;g__Bacteroidales_RF16_group	1240	15	786	10	0	442	272	87	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prevotellaceae;g__Prevotellaceae_UCG-003	1051	63	87	0	0	123	112	89	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Promicromonosporaceae;g__Cellulosimicrobium	0	0	0	22	0	94	66	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidales_UCG-001;g__Bacteroidales_UCG-001	624	11	55	0	0	2	0	154	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Pirellulales;f__Pirellulaceae;g__CPla-4_termite_group	42	0	190	19	0	238	74	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Dysgonomonadaceae;g__Dysgonomonas	409	0	0	0	0	4	7	12	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Microbacteriaceae;g__Microbacterium	1	0	0	43	0	259	28	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prevotellaceae;g__Alloprevotella	123	0	23	3	148	214	434	129	7
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__F082;g__F082	1051	7	121	161	0	148	30	701	0

d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscill spirales;f__Ruminococcaceae;g__Ruminococcus	414	0	334	64	36	326	309	118	84
d__Bacteria;p__Actinobacteriota;c__Actinobacteria ;o__Micrococcales;f__Cellulomonadaceae;g__Cell ulomonas	3	0	0	136	0	246	17	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Pepto streptococcales- Tissierellales;f__Anaerovoracaceae;g__Family_XII I_AD3011_group	250	3	148	131	112	363	210	59	9
d__Bacteria;p__Proteobacteria;c__Alphaproteobac teria;o__Acetobacteriales;f__Acetobacteraceae;g__ _Roseomonas	2	0	0	55	0	176	12	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscill spirales;f__Ruminococcaceae;g__Candidatus_So leiferrea	71	0	73	66	31	157	52	13	2
d__Bacteria;p__Patescibacteria;c__Saccharimona dia;o__Saccharimonadales;f__Saccharimonadace ae;g__TM7a	7	0	1	4	0	109	2	15	46
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__B acteroidales;f__Dysgonomonadaceae;g__uncultur ed	149	0	12	0	0	82	2	33	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachn spirales;f__Lachnospiraceae;g__Roseburia	11	0	26	8	52	65	126	0	448
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostr idia_vadinBB60_group;f__Clostridia_vadinBB60_g roup;g__Clostridia_vadinBB60_group	560	2	671	767	91	2123	476	63	19
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__B acteroidales;f__Rikenellaceae;g__dgA- 11_gut_group	68	6	174	256	88	383	83	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__B acteroidales;f__Prevotellaceae;g__Prevotellaceae _UCG-001	422	54	38	0	231	148	20	92	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachn spirales;f__Lachnospiraceae;g__Lachnospira	3	0	1	0	0	7	6	1	599
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachn spirales;f__Lachnospiraceae;g__Marvinbryantia	183	9	12	2	7	37	121	20	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__S phingobacteriales;f__Sphingobacteriaceae;g__Ped obacter	9	14	0	505	0	433	8	38	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscill spirales;f__Oscillospiraceae;g__UCG-002	262	0	130	65	20	232	64	27	360
d__Bacteria;p__Actinobacteriota;c__Actinobacteria ;o__Streptosporangiales;f__Thermomonosporacea e;g__Actinomadura	0	0	0	0	0	74	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Christ ensenellales;f__Christensenellaceae;g__Christens enellaceae_R-7_group	1058	41	791	402	368	1966	747	175	6
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscill spirales;f__Oscillospiraceae;g__Papillibacter	184	0	13	0	0	14	3	17	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__B acteroidales;f__Rikenellaceae;g__hoa5- 07d05_gut_group	294	0	1	0	0	5	1	15	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobac teria;o__Rhizobiales;f__Rhizobiaceae;__	1	0	1	12	0	167	16	5	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscill spirales;f__Oscillospiraceae;g__NK4A214_group	1824	24	273	137	201	813	271	319	32

d__Bacteria;p__Synergistota;c__Synergistia;o__Synergistales;f__Synergistaceae;__	119	0	0	0	0	1	0	0	0
d__Bacteria;p__Armatimonadota;c__uncultured;o__uncultured;f__uncultured;g__uncultured	240	0	0	0	0	2	0	20	0
d__Bacteria;p__Campilobacterota;c__Campylobacteria;o__Campylobacteriales;f__Campylobacteraceae;g__Campylobacter	11	0	56	0	15	57	15	1	1
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_UCG-009	439	0	0	0	0	0	0	54	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Muribaculaceae;g__Muribaculaceae	14	0	138	207	259	264	754	7	2
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Butyrivibrionaceae;g__UCG-009	28	0	111	75	4	279	90	4	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Acetivibrio	209	6	12	7	12	55	101	33	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Opitutales;f__Puniceicoccaceae;g__Cerasicoccus	6	0	51	5	9	82	300	2	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Weeksellaceae;g__uncultured	89	25	1	11	0	4	113	158	0
d__Bacteria;p__Fusobacteriota;c__Fusobacteriia;o__Fusobacteriales;f__Fusobacteriaceae;g__Fusobacterium	2	0	0	0	6	0	4	2	345
d__Bacteria;p__Bacteroidota;c__Bacteroidia;__;__	163	0	7	0	2	37	13	44	0
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria;o__Victivallales;f__vadinBE97;g__vadinBE97	487	6	108	38	3	75	21	20	8
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonadales;f__Sphingomonadaceae;g__Altererythrobacter	2	0	0	8	0	75	1	3	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Anaerovoracaceae;g__Anaerovorax	584	0	15	0	0	43	9	97	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__[Eubacterium]_hallii_group	114	6	8	0	0	39	17	23	11
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia_UCG-014;f__Clostridia_UCG-014;g__Clostridia_UCG-014	322	0	694	477	4	1492	375	78	47
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Pseudobutyrvibrio	132	0	0	0	0	3	0	13	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;f__Clostridiaceae;g__Clostridium_sensu_stricto_1	49	0	26	7	67	91	183	13	169
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcaceae;__	2	0	25	41	42	58	10	1	1
d__Bacteria;p__Cyanobacteria;c__Vampirivibrionia;o__Gastranaerophilales;f__Gastranaerophilales;g__Gastranaerophilales	646	6	228	154	12	467	253	38	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Monoglobales;f__Monoglobaceae;g__Monoglobus	89	0	279	143	41	692	119	12	7

d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__XBB1006	5	0	27	0	0	54	18	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Christensenellales;f__Christensenellaceae;g__uncultured	106	0	16	16	0	54	14	9	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Anaerovoracaceae;g__	294	4	14	1	0	82	23	34	0
d__Bacteria;p__Firmicutes;__;	35	0	18	0	0	54	140	9	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Anaerovoracaceae;g__[Eubacterium]_nodatum_group	13	0	34	8	39	152	12	1	1
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Eggerthellaceae;g__uncultured	102	5	3	0	0	2	8	4	1
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__[Ruminococcus]_gnavus_group	1	0	0	0	0	0	0	0	137
d__Bacteria;p__WPS-2;c__WPS-2;o__WPS-2;f__WPS-2;g__WPS-2	42	0	0	0	0	5	33	52	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidetes_BD2-2;g__Bacteroidetes_BD2-2	58	3	0	0	0	1	0	50	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidales;g__Bacteroidales	0	0	0	0	0	43	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__	169	0	217	96	18	353	167	34	22
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_FCS020_group	52	0	0	0	0	1	3	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__uncultured	102	0	24	15	6	122	49	9	2
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__Faecalibacterium	6	0	4	5	0	28	3	0	302
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__Oscillospiraceae	1	0	6	0	0	41	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;__;	356	5	102	37	0	101	80	120	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Spirosomaceae;g__Dyadobacter	1	8	0	20	0	135	0	20	0
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria;o__Victivallales;f__Victivallaceae;g__Victivallaceae	344	2	264	174	8	398	39	1	4
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales-Selenomonadales;f__Selenomonadaceae;g__Quinella	94	0	0	0	0	2	1	8	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__Incertae_Sedis	13	0	32	21	3	87	14	1	9
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Dysgonomonadaceae;g__Proteiniphilum	51	0	0	0	0	0	0	0	0

d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__p-2534-18B5_gut_group;g__p-2534-18B5_gut_group	0	0	34	0	0	16	259	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelatoclostridiaceae;g__UCG-004	437	4	18	3	0	67	8	30	1
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Xanthobacteraceae;g__Rhodopseudomonas	0	0	0	9	0	30	1	3	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Coprococcus	17	0	18	30	16	47	22	0	98
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Defluviitaleaceae;g__Defluviitaleaceae_UCG-011	117	7	27	46	0	84	9	35	1
d__Bacteria;p__Firmicutes;c__Bacilli;o__Acholeplasmales;f__Acholeplasmataceae;g__Anaeroplasmata	258	5	74	21	43	197	126	54	3
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_UCG-010	26	0	54	13	29	102	15	4	11
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Anaerovoracaceae;g__[Eubacterium]_brachy_group	3	0	15	25	15	82	5	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Taibaiella	1	53	0	26	0	65	8	56	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonadales;f__Sphingomonadaceae;g__Sphingobium	0	0	0	70	0	111	0	16	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;f__Clostridiaceae;g__Clostridium_sensu_stricto_3	11	0	0	0	29	5	81	2	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Tannerellaceae;g__Parabacteroides	8	0	6	0	0	1	95	0	115
d__Archaea;p__Euryarchaeota;c__Methanobacteria;o__Methanobacteriales;f__Methanobacteriaceae;g__Methanobrevibacter	27	0	18	20	8	74	42	1	1
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales;f__Nocardiodiaceae;g__Aeromicrobium	2	0	0	15	0	29	11	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__UCG-003	0	0	0	0	0	0	0	0	151
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodobacterales;f__Rhodobacteraceae;g__	1	37	0	3	36	120	103	47	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodospirillales;f__uncultured;g__uncultured	177	2	99	49	2	253	58	41	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Tannerellaceae;g__Macellibacteroides	0	0	0	0	418	22	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__[Eubacterium]_elgens_group	2	0	0	0	0	0	3	0	263
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__	178	7	150	70	6	392	51	26	7
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia;f__Hungateiclostridiaceae;g__Ruminiclostridium	132	0	7	0	0	16	3	22	0

d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonadales;f__Sphingomonadaceae;g__Sphingopyxis	1	84	0	0	6	95	6	36	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__Pygmaibacter	38	0	4	0	4	8	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Rikenellaceae;g__Mucinivorans	105	8	0	0	0	0	0	2	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodobacterales;f__Rhodobacteraceae;g__Gemmobacter	3	0	0	0	13	46	26	14	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Butyrivibrio	37	0	7	0	0	9	9	10	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Barnesiellaceae;g__uncultured	5	0	64	110	35	133	9	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__uncultured	56	0	0	0	0	3	0	31	2
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Crocinitomicaceae;g__Fluviicola	2	19	0	0	0	14	1	92	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Acetobacteriales;f__Acetobacteraceae;g__Belnapia	0	0	0	14	0	20	1	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Blautia	34	7	12	9	11	27	19	1	110
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__Oscillospira	30	0	2	2	0	17	79	1	7
d__Bacteria;p__Firmicutes;c__Bacilli;o__Paenibacillales;f__Paenibacillaceae;g__Paenibacillus	6	11	0	6	0	202	106	113	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Anaerocolumna	0	0	0	0	105	22	11	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Christensenellales;f__Christensenellaceae;g__	57	10	10	3	0	52	14	15	0
d__Bacteria;p__Elusimicrobiota;c__Elusimicrobia;o__Elusimicrobiales;f__Elusimicrobiaceae;g__Elusimicrobium	61	3	39	39	9	156	40	21	10
d__Archaea;p__Halobacterota;c__Methanomicrobia;o__Methanomicrobiales;f__Methanocorpusculaceae;g__Methanocorpusculum	74	0	14	19	0	23	0	20	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Streptomycetales;f__Streptomyetaceae;g__Streptomyces	2	0	0	0	0	98	5	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__CAG-352	68	0	3	0	0	2	2	0	36
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Micrococcaceae;g__Glutamicibacter	59	0	1	0	0	4	17	1	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Rikenellaceae;g__	65	0	15	2	0	41	41	12	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales;f__Nocardioidaceae;g__Mumia	0	0	0	3	0	26	5	0	0

d__Bacteria;p__Firmicutes;c__Bacilli;o__Izemoplasmales;f__Izemoplasmales;g__Izemoplasmales	56	0	24	37	4	151	52	17	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__[Ruminococcus]_torques_group	0	0	0	0	0	0	0	0	93
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Coriobacteriales_Incertae_Sedis;g__Phoenicibacter	99	0	0	0	0	0	0	6	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Eggerthellaceae;g__	99	3	6	5	0	55	7	10	0
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria;o__Victivallales;f__Victivallaceae;g__Victivallis	1	0	11	6	6	14	6	0	56
d__Bacteria;p__Actinobacteriota;c__Actinobacteriia;o__Micromonosporales;f__Micromonosporaceae;g__Micromonospora	1	0	0	0	0	22	3	1	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__[Eubacterium]_ruminantium_group	24	0	15	14	0	23	34	0	58
d__Bacteria;p__Patescibacteria;c__Gracilibacteria;o__Absconditabacteriales_(SR1);f__Absconditabacteriales_(SR1);g__Absconditabacteriales_(SR1)	37	0	1	0	0	2	0	67	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_UCG-006	174	0	0	0	0	1	0	30	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteriia;o__Micrococcales;f__Bogoriellaceae;g__Georgenia	2	0	0	0	0	5	43	0	3
d__Bacteria;p__Firmicutes;c__Clostridia;g__	334	0	387	166	14	1198	198	73	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales;f__Sphingobacteriaceae;g__Mucilaginibacter	2	0	0	17	0	119	0	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__[Eubacterium]_ventriosum_group	3	0	2	9	0	40	9	2	19
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcaceae;g__Terrisporobacter	4	0	5	0	20	24	13	0	1
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__Aureimonas	1	0	1	13	0	52	2	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__Negativibacillus	1	0	16	7	100	56	3	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Izemoplasmales;f__Izemoplasmataceae;g__Izemoplasmataceae	166	0	0	0	0	1	0	8	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteriia;o__Corynebacteriales;f__Mycobacteriaceae;g__Mycobacterium	2	0	1	17	0	58	2	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Beijerinckiaceae;g__Bosea	0	0	0	12	0	22	1	1	1
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_UCG-008	51	0	0	0	0	10	0	22	0
d__Bacteria;p__Elusimicrobiota;c__Endomicrobia;o__Endomicrobiales;f__Endomicrobiaceae;g__Endomicrobium	62	0	3	0	0	0	0	15	0

d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Aeromonadales;f__Succinivibrionaceae;g__Ruminobacter	4	0	2	0	0	4	64	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelatoclostridiaceae;g__Catenibacterium	0	0	0	0	0	3	1	0	51
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Verrucomicrobiales;f__Verrucomicrobiaceae;g__Verrucomicrobium	0	7	0	0	0	26	0	32	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_UCG-001	1	0	19	0	0	35	0	0	27
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Cellulomonadaceae;g__Oerskovia	0	0	0	28	0	14	39	1	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__Colidexibacter	38	0	5	0	15	26	67	1	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__Anaerolineales;f__Anaerolineaceae;g__Flexilinea	19	0	7	0	48	62	6	12	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales;f__Sphingobacteriaceae;g__Nubella	0	0	0	39	0	31	2	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_ND3007_group	57	6	0	0	0	0	1	4	2
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__Oscillibacter	7	0	39	42	17	145	77	0	33
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__Intestinimonas	0	0	2	5	0	27	2	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__FD2005	18	0	0	0	0	0	0	2	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Devosiaceae;g__Devosia	3	62	1	28	7	70	46	27	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Moryella	19	0	5	4	0	11	7	1	2
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacterales;f__Xiphinematobacteraceae;g__Candidatus_Xiphinematobacter	2	0	0	0	0	6	2	75	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__uncultured;g__uncultured	16	6	1	0	0	16	44	11	0
d__Bacteria;p__Bdellovibrionota;c__Oligoflexia;o__0319-6G20;f__0319-6G20;g__0319-6G20	0	0	0	0	0	1	0	46	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Eggerthellaceae;g__Parvibacter	1	0	7	3	0	15	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Pirellulales;f__Pirellulaceae;g__Pir4_lineage	2	0	0	0	0	7	1	37	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__Ruminococcaceae	1	0	43	2	6	40	20	0	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Atopobiaceae;g__Olsenella	0	0	1	5	0	36	7	0	0

d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prevotellaceae;g__uncultured	38	6	1	0	0	1	0	5	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacteriales;f__Caulobacteraceae;g__Caulobacter	0	0	0	6	0	16	0	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia;f__Hungateiclostridiaceae;g__	95	0	36	12	0	61	5	32	0
d__Bacteria;p__Spirochaetota;c__Spirochaetia;o__Spirochaetales;f__Spirochaetaceae;g__GWE2-31-10	11	0	43	5	0	21	1	1	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Cellulosilyticum	49	0	12	0	52	7	18	1	1
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Chloroplast;f__Chloroplast;g__Chloroplast	14	5	5	0	0	63	6	14	7
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Shuttleworthia	3	0	1	3	0	19	1	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__COB_P4-1_termite_group;g__COB_P4-1_termite_group	0	0	4	0	0	9	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_NK3A20_group	23	0	11	0	0	35	3	2	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelatoclostridiaceae;g__Erysipelatoclostridium	18	0	1	0	0	6	0	12	2
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Weeksellaceae;g__	1	7	0	0	0	4	35	6	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Verrucomicrobiales;f__Verrucomicrobiaceae;g__Prostheco bacter	0	2	0	2	0	30	0	12	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__Subdoligranulum	19	0	0	0	0	0	14	4	56
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Marinifilaceae;g__uncultured	30	0	0	0	0	0	0	28	0
d__Bacteria;__;__;__;__	277	3	72	4	3	238	50	55	8
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Anaerovoracaceae;g__uncultured	50	0	0	0	0	0	0	3	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Eubacteriales;f__Eubacteriaceae;g__Eubacterium	29	0	0	0	0	3	0	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Anaerovoracaceae;g__Family_XII_UCG-001	34	0	7	9	0	19	12	8	1
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Pirellulales;f__Pirellulaceae;g__uncultured	4	0	0	0	0	17	2	22	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prevotellaceae;g__Prevotellaceae_Ga6A1_group	22	0	16	0	5	19	24	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Anaerospore bacter	2	0	15	0	0	28	46	8	0

d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Terrimonas	1	0	1	0	0	0	0	19	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Dysgonomonadaceae;g__	16	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales;f__Sphingobacteriaceae;g__uncultured	4	0	0	0	0	9	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_UCG-004	30	0	3	0	0	5	2	6	29
d__Bacteria;p__Synergistota;c__Synergistia;o__Synergistales;f__Synergistaceae;g__Pyramidobacter	33	2	1	0	0	7	19	5	2
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__[Eubacterium]_siraenum_group	5	0	20	0	0	49	65	0	10
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Tuzzerella	2	0	4	0	0	14	5	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Acholeplasmatales;f__Acholeplasmataceae;g__NED5E9	14	0	0	0	0	0	0	10	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacteriales;f__Terrimicrobiaceae;g__Terrimicrobium	1	0	0	0	0	34	1	13	0
d__Bacteria;p__Myxococcota;c__Myxococcia;o__Myxococcales;f__Myxococcaceae;g__P3OB-42	20	0	0	0	0	1	0	2	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Coriobacteriales_Incertae_Sedis;g__uncultured	18	0	1	4	0	18	1	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__UCG-007	12	0	3	0	0	11	3	3	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__uncultured;f__uncultured;g__uncultured	1	13	0	0	0	2	0	14	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Streptosporangiales;f__Streptosporangiaceae;g__Nonomuraea	0	0	0	0	0	7	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__CAP-aah99b04;g__CAP-aah99b04	0	0	0	0	0	1	0	16	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Microscillaceae;g__Ohtaekwangia	0	0	0	0	0	4	0	14	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Beijerinckiaceae;g__Methylobacterium-Methylorubrum	0	0	0	4	0	38	1	1	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Catenisphaera	14	2	0	0	0	2	6	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelatoclostridiaceae;g__uncultured	83	0	39	14	4	139	8	9	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Pirellulales;f__Pirellulaceae;g__Pirellula	7	0	2	0	0	15	0	24	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__D05-2;g__D05-2	0	0	0	4	0	13	0	0	0

d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Fusicatenibacter	0	0	0	0	0	0	0	0	24
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__	49	0	5	0	0	20	4	10	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodobacterales;f__Rhodobacteraceae;g__Falsirhodobacter	0	0	0	11	0	19	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__uncultured	217	3	3	0	0	21	40	35	4
d__Bacteria;p__Firmicutes;c__Bacilli;o__Acholeplasmatales;f__Acholeplasmataceae;g__Acholeplasma	9	6	5	0	0	14	0	14	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Tepidisphaerales;f__WD2101_soil_group;g__WD2101_soil_group	12	0	5	37	0	47	3	4	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Corynebacteriaceae;g__Corynebacterium	99	0	12	0	0	3	5	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ethanologenaceae;g__Incertae_Sedis	12	0	0	0	0	2	0	5	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Dielma	1	0	5	5	6	29	5	1	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Rikenellaceae;g__SP3-e08	11	0	28	0	0	8	1	2	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Marinifilaceae;g__Odoribacter	2	0	11	12	15	32	3	0	50
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria;o__Oligosphaerales;f__Oligosphaeraceae;g__h-orsej-a03	27	0	0	0	0	0	0	14	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__V9D2013_group	11	0	7	0	0	16	0	5	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacterales;f__Caulobacteraceae;g__Phenylobacterium	0	0	0	6	0	30	0	2	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Pirellulales;f__Pirellulaceae;g__	1	0	0	0	0	20	0	20	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Opitutales;f__Puniceicoccaceae;g__uncultured	16	0	18	20	13	53	16	1	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__Caproiciproducens	30	0	2	2	2	25	5	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__GCA-900066575	16	0	1	0	0	2	2	0	5
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Beutenbergiaceae;g__Salan	1	0	0	8	0	6	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Amb-16S-1323;g__Amb-16S-1323	0	0	0	0	0	0	0	12	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__uncultured;f__uncultured;g__uncultured	9	0	19	6	0	53	8	0	0

d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Reyranelles;f__Reyranelaceae;g__Reyranelle	0	0	0	0	0	3	0	14	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__[Anaerotruncus]_furcosa_group	65	2	14	4	6	26	4	22	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Edaphobaculum	0	0	0	11	0	32	0	4	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Acholeplasmatales;f__Acholeplasmataceae;g__EMP-G18	39	3	17	4	5	54	5	6	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Marinifilaceae;g__Sanguibacteroides	1	0	2	11	0	11	1	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Micrococcaceae;g__Micrococcus	11	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospirales;g__Hydrogenoanaerobacterium	7	0	22	4	0	35	31	5	2
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Eggerthellaceae;g__Gordonibacter	0	0	1	2	0	8	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Hungatella	0	0	0	0	0	0	0	0	18
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__Flavonifractor	0	0	14	3	0	7	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Eubacteriales;f__Anaerofustaceae;g__Anaerofustis	13	0	11	13	2	37	5	4	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Micrococcaceae;g__Pseudarthrobacter	0	0	0	0	0	6	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Intrasporangiaceae;g__	7	0	0	0	0	4	1	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodobacterales;f__Rhodobacteraceae;g__Paracoccus	5	0	0	25	0	21	10	0	3
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Pseudonocardiales;f__Pseudonocardaceae;g__Pseudonocardia	0	0	0	2	0	10	0	0	0
d__Bacteria;p__Spirochaetota;c__MVP-15;o__MVP-15;f__MVP-15;g__MVP-15	0	0	0	0	0	0	0	24	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Micrococcales_Incertae_Sedis;g__Timonella	1	0	0	0	0	5	6	2	0
d__Archaea;p__Halobacterota;c__Methanosarciniales;o__Methanosarciniales;f__Methanosarcinaceae;g__Methanosarcina	10	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Acholeplasmatales;f__Acholeplasmataceae;g__T2WK15B57	0	0	0	0	0	5	28	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales;f__Anaerovoracaceae;g__Mogibacterium	5	0	1	0	3	30	8	1	0
d__Archaea;p__Thermoplasmatota;c__Thermoplasmatas;o__Methanomassiliicoccales;f__Methanomethylophilaceae;g__	3	0	5	0	0	4	17	0	0

d_Bacteria;p_Firmicutes;c_Negativicutes;o_Acidaminococcales;f_Acidaminococcaceae;g_Phascalarctobacterium	248	7	63	10	0	117	62	12	6
d_Bacteria;p_Bacteroidota;c_Bacteroidia;o_Flavobacteriales;f_Flavobacteriaceae;g_Confluentibacter	1	0	0	0	0	0	0	18	0
d_Bacteria;p_Actinobacteriota;c_Acidimicrobiia;o_Microtrichales;f_Microtrichaceae;g_uncultured	0	0	0	0	0	0	0	17	0
d_Bacteria;p_Plactomycetota;c_Plactomycetes;o_Plactomycetales;f_Rubinisphaeraceae;g_Plactomicrobium	1	0	0	0	0	1	0	22	0
d_Bacteria;p_Proteobacteria;c_Alphaproteobacteria;o_Rhizobiales;f_Methylophilaceae;g_Methylophila	0	0	0	0	0	5	0	0	0
d_Bacteria;p_Firmicutes;c_Clostridia;o_Lachnospirales;f_Lachnospiraceae;g_[Bacteroides]_pectinophilus_group	7	0	0	0	0	1	1	0	0
d_Bacteria;p_Firmicutes;c_Clostridia;o_Lachnospirales;f_Lachnospiraceae;g_Lachnospiraceae_UCG-002	10	0	2	0	15	23	3	3	0
d_Bacteria;p_Proteobacteria;c_Alphaproteobacteria;o_Sphingomonadales;f_Sphingomonadaceae;g_Sandaracinobacter	0	0	0	0	0	5	0	0	0
d_Bacteria;p_Firmicutes;c_Clostridia;o_Lachnospirales;f_Lachnospiraceae;g_Tyzzerella	0	0	0	0	0	7	15	0	0
d_Bacteria;p_Firmicutes;c_Bacilli;o_Mycoplasmatales;f_Mycoplasmataceae;g_Mycoplasma	51	0	3	0	0	0	0	0	0
d_Bacteria;p_Firmicutes;c_Clostridia;o_Peptostreptococcales-Tissierellales;f_Peptostreptococcales-Tissierellales;g_Alkaliphilus	0	0	0	0	0	6	0	0	0
d_Bacteria;p_Spirochaetota;c_Spirochaetia;o_Spirochaetales;f_Spirochaetaceae;g_Sphaerochaeta	142	2	1	0	0	0	16	7	0
d_Bacteria;p_Bacteroidota;c_Bacteroidia;o_Chitinophagales;f_Chitinophagaceae;g_Pseudoflavitalea	1	0	0	0	0	0	0	20	0
d_Bacteria;p_Firmicutes;c_Clostridia;o_Oscillospirales;f_Oscillospiraceae;g_Sporobacter	7	0	0	0	0	0	0	0	0
d_Bacteria;p_Spirochaetota;c_Spirochaetia;o_Spirochaetales;f_Spirochaetaceae;g_Treponema	347	0	40	2	0	63	48	28	0
d_Bacteria;p_Firmicutes;c_Clostridia;o_Clostridiales;f_Clostridiaceae;g_Clostridium_sensu_stricto_6	0	0	0	0	0	0	12	0	0
d_Bacteria;p_Actinobacteriota;c_Coriobacteriia;o_Coriobacteriales;f_Atopobiaceae;g_Atopobium	0	0	1	0	0	19	1	0	0
d_Archaea;p_Thermoplasmata;c_Thermoplasmata;o_Methanomassiliococcales;f_Methanomethylophilaceae;g_uncultured	51	5	3	4	0	15	1	7	0
d_Bacteria;p_Actinobacteriota;c_Coriobacteriia;o_Coriobacteriales;f_Eggerthellaceae;g_Enterorhabdus	20	0	7	0	0	27	4	7	1
d_Bacteria;p_Proteobacteria;c_Alphaproteobacteria;o_Acetobacteriales;f_Acetobacteraceae;g_Acetobacteraceae	0	0	0	0	0	5	0	0	0
d_Bacteria;p_Actinobacteriota;c_Actinobacteria;o_Micrococcales;f_Intrasporangiaceae;g_Ornithinimicrobium	15	0	0	0	0	1	0	0	0

d__Bacteria;p__Firmicutes;c__Bacilli;__;__;__	13	0	0	4	0	68	8	6	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__uncultured;g__uncultured	9	0	1	0	0	5	5	3	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Bacillales;f__Planococcaceae;__	6	2	1	2	0	106	36	75	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiales_Incertae_Sedis;g__Bauldia	0	0	0	0	0	1	0	8	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Tepidisphaerales;f__Tepidisphaeraceae;g__Tepidisphaera	3	0	1	3	0	11	0	0	0
d__Bacteria;p__Fusobacteriota;c__Fusobacteriia;o__Fusobacteriales;f__Fusobacteriaceae;g__Cetobacterium	0	0	0	0	277	0	0	0	3
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales;f__Sphingobacteriaceae;g__Arcticibacter	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Marinilabiliaceae;g__uncultured	0	0	0	0	0	1	0	23	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Xanthobacteraceae;g__Tardiphaga	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Erysipelotrichaceae_UCG-009	3	0	3	0	0	17	1	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnoclostridium	0	0	0	0	50	1	4	2	39
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__[Ruminococcus]_gavvreauii_group	0	0	6	0	0	17	1	0	3
d__Bacteria;p__Campilobacterota;c__Campylobacteriia;o__Campylobacteriales;f__Arcobacteraceae;g__Arcobacter	7	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Frisingicoccus	4	0	3	0	0	3	13	2	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Paludibacteriaceae;g__Paludibacter	0	0	0	0	0	9	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptococcales;f__Peptococcaceae;g__uncultured	5	0	8	0	0	13	6	2	2
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales;f__Sedimentibacteraceae;g__Sedimentibacter	9	0	0	0	17	4	0	1	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__UCG-001	0	0	1	0	0	8	1	1	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospiraceae;g__Pseudoflavonifractor	1	0	1	0	0	6	0	0	1
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;__;__;__	7	0	3	2	0	24	7	2	0
d__Bacteria;p__Firmicutes;c__Incertae_Sedis;o__DTU014;f__DTU014;g__DTU014	0	0	4	0	0	6	0	0	0

d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacteriales;f__Caulobacteraceae;g__uncultured	0	0	0	0	0	6	0	3	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__Aliihoeflea	1	0	1	0	0	5	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Solobacterium	7	0	3	0	0	11	1	1	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_FE2018_group	0	0	0	0	0	6	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Kaistiaceae;g__Kaistia	0	0	0	0	0	5	0	0	0
d__Bacteria;p__Myxococcota;c__Myxococcia;o__Myxococcales;f__Myxococcaceae;g__	0	0	10	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Brezznakia	3	0	4	0	0	7	2	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Weeksellaceae;g__Cloacibacterium	0	0	0	0	0	1	9	0	0
d__Archaea;p__Euryarchaeota;c__Methanobacteria;o__Methanobacteriales;f__Methanobacteriaceae;g__Methanosphaera	0	0	1	2	7	18	1	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__Shinella	0	0	0	0	0	4	0	0	0
d__Archaea;p__Thermoplasmatota;c__Thermoplasmata;o__Methanomassiliicoccales;f__Methanomethylophilaceae;g__Candidatus_Methanomethylophilus	13	4	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales;f__Mitochondria;g__Mitochondria	44	0	8	1	0	23	7	6	0
d__Bacteria;p__Firmicutes;c__uncultured;o__uncultured;f__uncultured;g__uncultured	5	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales;f__uncultured;g__uncultured	10	0	3	3	0	24	5	4	0
d__Bacteria;p__Fibrobacterota;c__Fibrobacteria;o__Fibrobacteriales;f__Fibrobacteraceae;g__uncultured	0	0	0	0	0	0	0	16	0
d__Bacteria;p__Firmicutes;c__Limnochordia;o__Limnochordia;f__Limnochordia;g__Hydrogenispora	6	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Enterobacteriales;f__Enterobacteriaceae;g__	5	0	0	0	0	3	8	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Carnobacteriaceae;g__uncultured	5	0	1	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Beijerinckiaceae;g__uncultured	0	0	0	0	0	8	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales;f__Peptostreptococcaceae;g__Paenibacillus	0	0	5	0	0	1	0	0	6
d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Thermomicrobiales;f__JG30-KF-CM45;g__JG30-KF-CM45	14	0	0	0	0	12	0	1	2

d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;g__	31	0	0	0	0	10	1	3	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;f__Clostridiaceae;g__Sarcina	7	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelatoclostridiaceae;g__Erysipelotrichaceae_UCG-003	0	0	0	0	0	0	0	0	9
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Weeksellaceae;g__Moheibacter	1	0	1	0	0	4	0	8	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Paludibacteraceae;g__F0058	3	0	0	0	0	0	0	1	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Anaerostipes	0	0	0	0	0	0	0	0	8
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__uncultured;g__uncultured	7	0	1	0	0	4	1	1	0
d__Archaea;p__Euryarchaeota;c__Methanobacteria;o__Methanobacteriales;f__Methanobacteriaceae;g__Methanobacterium	8	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Microbacteriaceae;g__Agromyces	0	0	1	0	0	8	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales;f__Nocardiodiaceae;g__Marmoricola	1	0	0	0	0	4	0	3	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales;f__Sphingobacteriaceae;g__Olivibacter	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria;o__Oligosphaerales;f__Oligosphaeraceae;g__Z20	9	0	2	0	0	2	0	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Howardella	1	0	0	0	0	8	0	0	2
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales-Selenomonadales;f__Selenomonadaceae;g__Schwartzia	15	0	0	0	0	0	0	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Dietziaceae;g__Dietzia	3	0	0	0	0	1	8	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Barnesiellaceae;g__Barnesiella	0	0	0	0	0	0	0	0	14
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Aeromonadales;f__Succinivibrionaceae;g__uncultured	0	0	0	0	0	0	4	0	7
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__CAG-56	0	0	0	0	0	0	0	0	7
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Micrococcaceae;g__Citricoccus	3	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Sanguibacteraceae;g__Sanguibacter	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Actinomycetales;f__Actinomycetaceae;g__Arcanobacterium	17	0	0	0	0	0	0	0	0

d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Coproba cillus	1	0	1	3	0	8	1	0	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__C hitinophagales;f__Chitinophagaceae;g__unculture d	0	0	0	2	0	9	0	2	0
d__Bacteria;p__SAR324_clade(Marine_group_B); c__SAR324_clade(Marine_group_B);o__SAR324_ clade(Marine_group_B);f__SAR324_clade(Marine_ _group_B);g__SAR324_clade(Marine_group_B)	19	0	0	0	0	5	0	7	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__C ytrophagales;f__Spirosomaceae;g__Leadbetterella	1	8	0	0	0	1	0	12	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Bacillales ;f__Planococcaceae;g__Solibacillus	0	0	0	0	0	12	6	13	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobac teria;o__Rhizobiales;f__Xanthobacteraceae;__	0	0	0	0	0	9	0	2	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Fl avobacteriales;f__Flavobacteriaceae;g__unculture d	1	0	6	45	0	4	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachn ospirales;f__Lachnospiraceae;g__Lachnospiracea e_UCG-003	3	0	0	0	0	0	8	0	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Po lyangiales;f__Polyangiaceae;g__Chondromyces	0	0	0	0	0	0	0	4	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobac teria;o__Tistrellales;f__Geminicoccaceae;g__Gemi nicoccus	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscill ospirales;f__[Clostridium]_methylpentosum_group; g__[Clostridium]_methylpentosum_group	0	0	2	0	0	14	2	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobac teria;o__Rhizobiales;f__A0839;g__A0839	0	0	0	1	0	3	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteob acteria;o__Enterobacteriales;f__Enterobacteriaceae; g__Escherichia-Shigella	29	0	0	0	11	40	21	6	1
d__Bacteria;p__Patescibacteria;c__Saccharimona dia;o__Saccharimonadales;__;__	4	0	1	2	0	30	0	2	4
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscill ospirales;f__Ethanoligenaceae;g__Acetanaerob acterium	5	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria ;o__Actinomycetales;f__Actinomycetaceae;g__Fla viflexus	16	0	0	0	0	0	0	0	0
d__Bacteria;p__Myxococcota;c__Myxococcia;o__ Myxococcales;f__Myxococcaceae;g__Myxococcus	1	0	0	0	0	0	0	4	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__B acteroidales;f__Prevotellaceae;g__Paraprevotella	0	0	0	0	0	0	6	0	1
d__Bacteria;p__Firmicutes;c__Negativicutes;o__V eillonellales- Selenomonadales;f__Selenomonadaceae;g__Ana erovibrio	35	4	0	0	0	2	6	3	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipel otrichales;f__Erysipelotrichaceae;g__Holdemanella	0	0	0	0	0	0	0	0	8
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia ;o__Coriobacteriales;f__Eggerthellaceae;g__Denitr obacterium	18	0	0	0	0	0	0	0	0

d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Micavibrionales;f__uncultured;g__uncultured	1	0	0	0	0	10	0	15	0
d__Bacteria;p__Desulfobacterota;c__Desulfovibronia;o__Desulfovibrionales;f__Desulfovibrionaceae;g__	6	0	2	0	0	1	0	0	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Polyangiales;f__Sandaracinaceae;g__uncultured	0	0	0	0	0	0	0	4	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Caldicoprobacteriales;f__Caldicoprobacteraceae;g__Caldicoprobacter	0	0	0	0	0	5	1	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Saprospiraceae;g__uncultured	1	0	1	0	0	0	0	6	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Isosphaerales;f__Isosphaeraceae;g__uncultured	2	0	0	0	0	11	0	6	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Microbacteriaceae;g__	7	0	0	0	0	23	3	14	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcaceae;g__Intestinibacter	0	0	0	0	0	0	0	0	5
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Verrucomicrobiales;f__Verrucomicrobiaceae;g__Roseimicrobium	0	0	0	0	0	1	0	5	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;f__	0	0	0	0	45	2	1	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Anaerovoracaceae;g__Amnipila	0	0	1	0	0	3	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Microbacteriaceae;g__Agrococcus	1	0	0	0	0	5	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__[Eubacterium]_xylanophilum_group	0	0	0	0	0	0	3	0	3
d__Bacteria;p__Firmicutes;c__Bacilli;o__Acholeplasmatales;f__Acholeplasmataceae;g__	3	0	0	0	0	0	10	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Hymenobacteraceae;g__Adhaeribacter	0	0	0	1	0	6	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Phycisphaerales;f__Phycisphaeraceae;g__SM1A02	3	0	0	0	0	1	0	3	0
d__Bacteria;p__Actinobacteriota;c__Thermoleophilina;o__Solirubrobacterales;f__Solirubrobacteraceae;g__Patulibacter	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcales-Tissierellales;g__W5053	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Pirellulales;f__Pirellulaceae;g__Termite_planctomycete_cluster	0	0	1	0	0	2	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Promicromonosporaceae;g__Promicromonospora	0	0	0	0	0	6	0	0	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Atopobiaceae;g__	0	0	1	0	0	8	0	0	0

d__Bacteria;p__Acidobacteriota;c__Blastocatellia;o__Blastocatellales;f__Blastocatellaceae;g__Blastocatella	2	0	0	0	0	0	0	4	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Beijerinckiaceae;g__Chelatococcus	0	0	0	0	0	0	5	0	0
d__Bacteria;p__Spirochaetota;c__Leptospirae;o__Leptospirales;f__Leptospiraceae;g__Rs-H88_termite_group	3	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Aurantisolmonas	0	0	0	0	0	2	0	7	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Eggerthellaceae;g__DNF00809	1	0	4	9	0	55	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Bifidobacteriales;f__Bifidobacteriaceae;g__Bifidobacterium	1	0	0	0	0	0	10	0	2
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Tepidisphaerales;f__Tepidisphaeraceae;g__Tepidisphaeraceae	3	0	0	0	0	10	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__	1	0	0	0	0	7	0	4	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Frankiales;f__Geodermatophilaceae;g__Klenkia	0	0	0	0	0	4	0	1	0
d__Bacteria;p__Patescibacteria;c__Parcubacteria;o__Candidatus_Nomurabacteria;f__Candidatus_Nomurabacteria;g__Candidatus_Nomurabacteria	1	0	0	0	0	0	0	0	5
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Xanthomonadales;f__Xanthomonadaceae;g__Stenotrophomonas	1	0	0	2	0	40	8	9	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Erysipelothrix	3	0	1	0	0	3	6	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Azospirillales;f__Azospirillaceae;g__Azospirillum	0	0	0	3	0	3	0	3	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacteriales;f__Caulobacteraceae;g__	0	0	0	0	0	1	1	3	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Alcaligenaceae;g__	10	0	0	1	0	21	0	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Cyclobacteriaceae;g__Algoriphagus	0	0	0	0	0	6	0	2	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Dermabacteraceae;g__Brachybacterium	5	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Microbacteriaceae;g__Curtobacterium	2	0	0	0	0	1	0	0	0
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonadales;f__LWQ8;g__LWQ8	2	0	0	0	0	13	3	4	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Chitinophaga	1	0	0	0	0	1	0	3	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Beijerinckiaceae;g__	0	0	1	3	0	13	0	1	0

d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscill ospirales;f__Butyrivibrionaceae;g__	0	0	2	0	10	4	4	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobi otae;o__Opitutales;f__Opitutaceae;g__Lacunisphaera	0	0	0	0	0	5	0	3	0
d__Bacteria;p__Deinococcota;c__Deinococci;o__Deinococ cales;f__Deinococcaceae;g__Deinococcus	2	0	0	4	0	3	0	0	4
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobi otae;o__Pedosphaerales;f__Pedosphaeraceae;g__DEV114	2	4	1	0	0	5	0	0	0
d__Bacteria;p__Planctomycetota;c__vadinHA49;o__vadinHA49 ;f__vadinHA49;g__vadinHA49	2	0	0	0	0	0	0	8	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospir ales;f__Lachnospiraceae;g__Lachnospiraceae_NK4B4_group	1	0	3	0	0	2	4	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobi otae;o__Chthoniobacterales;f__Chthoniobacteraceae;g__Chthoniobacter	1	0	0	3	0	12	1	2	0
d__Archaea;p__Halobacterota;c__Methanomicrobiota;o__Methanomicr obiales;f__Methanomicrobiaceae;g__Methanomicrobium	0	0	0	0	0	0	0	3	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophag ales;f__Chitinophagaceae;g__Flavibacter	1	0	0	0	0	4	0	2	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobi otae;o__Opitutales;f__Puniceicoccaceae;g__	4	0	3	0	0	14	3	1	0
d__Bacteria;p__Sumerlaeota;c__Sumerlaeae;o__Sumerlaeal es;f__Sumerlaeaceae;g__Sumerlaea	1	0	0	0	0	2	1	4	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__P seudomonadales;f__Moraxellaceae;g__Acinetobacter	12	4	0	1	0	57	48	39	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophag ales;f__Chitinophagaceae;g__Niabella	0	0	0	0	0	4	0	4	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichal es;f__Erysipelatoclostridiaceae;g__	7	0	5	0	0	10	4	5	1
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizob iales;f__Beijerinckiaceae;g__Microvirga	0	0	0	0	0	6	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiot ae;o__Verrucomicrobiales;f__Verrucomicrobiaceae;g__uncultured	1	0	0	0	0	7	0	3	0
d__Bacteria;p__Bdellovibrionota;c__Bdellovibrionia;o__Bdellovib ionales;f__Bdellovibrionaceae;g__Bdellovibrio	0	0	0	0	0	9	0	7	1
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococ cales;f__Peptostreptococcaceae;g__Paraclostridium	0	0	0	0	0	0	2	0	2
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonad ales;f__Saccharimonadaceae;g__Saccharimonadales	6	0	1	0	0	23	5	7	7
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonad ales;f__Sphingomonadaceae;g__DSSF69	0	0	0	0	0	0	1	2	0
Unassigned;__;__;__;__	30	0	4	0	0	71	1	6	4

d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Kallotenuales;f__AKIW781;g__AKIW781	2	0	0	0	0	2	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcales-Tissierellales;g__Anaerococcus	2	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales-Selenomonadales;f__Veillonellaceae;g__Dialister	0	0	0	0	0	0	0	0	4
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia;f__Hungateiclostridiaceae;g__UCG-012	4	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__;	8	0	5	0	0	16	0	1	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Bacillales;f__Planococcaceae;g__Chungangia	0	0	0	0	0	3	1	0	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Coriobacteriaceae;g__Collinsella	0	0	0	0	0	1	0	0	3
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Opitutales;f__Opitutaceae;g__	0	0	1	0	0	5	0	0	0
d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Chloroflexales;f__Roseiflexaceae;g__uncultured	0	0	0	0	0	1	0	3	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteriia;o__Bifidobacteriales;f__Bifidobacteriaceae;g__Aeriscardovia	1	0	1	0	0	4	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Spirosomaceae;g__Persicitalea	0	0	0	0	0	0	0	3	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodobacterales;f__Rhodobacteraceae;g__Rhodobacter	0	0	0	0	0	2	0	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Rikenellaceae;g__Anaerocella	2	0	0	0	0	1	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Planctomycetales;f__Rubinisphaeraceae;g__SH-PL14	0	0	1	0	0	10	1	3	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Gemmatales;f__Gemmataceae;g__uncultured	3	0	1	0	3	9	3	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteriia;o__Frankiales;f__Nakamurellaceae;g__Nakamurella	1	0	1	0	0	6	0	4	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteriia;o__Streptosporangiales;f__Thermomonosporaceae;g__Actinocorallia	0	0	1	0	0	2	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacteriales;f__Terrimicrobiaceae;g__FukuN18_freshwater_group	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Butyricicoccaceae;g__Butyricoccus	3	0	0	0	0	3	3	0	10
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacteriales;f__Hyphomonadaceae;g__Hirschia	0	0	0	0	0	2	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Oxalobacteraceae;g__Massilia	0	0	0	10	0	47	1	5	0

d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Catenibacillus	1	0	1	0	0	1	0	0	0
d__Bacteria;p__Synergistota;c__Synergistia;o__Synergistales;f__Synergistaceae;g__Synergistes	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Weeksellaceae;g__Weeksella	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__[Eubacterium]_oxidoreducens_group	15	0	22	6	0	67	8	12	0
d__Bacteria;p__Actinobacteriota;c__Acidimicrobiia;o__Microtrichales;f__Ilumatobacteraceae;g__Ilumatobacter	1	0	0	0	0	1	0	2	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Xanthobacteraceae;g__Pseudorhodoplanes	0	0	0	0	0	4	0	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Cnuella	0	0	0	4	0	2	0	1	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Faecalitalea	1	0	1	1	0	0	0	0	3
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Opitutales;f__Opitutaceae;g__Diplosphaera	0	0	0	0	0	0	0	2	0
d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Chloroflexales;f__Chloroflexaceae;g__FFCH7168	0	0	0	0	0	0	0	4	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Frankiales;f__Geodermatophilaceae;g__Blastococcus	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Pleomorphomonadaceae;g__Chthonobacter	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Comamonadaceae;g__	2	10	0	3	2	60	12	12	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Micrococcaceae;g__Kocuria	4	0	2	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__Angelakisella	0	0	0	0	0	2	3	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Opitutales;f__Opitutaceae;g__IMCC26134	0	0	0	0	0	2	0	4	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Cellulomonadaceae;g__	1	0	0	2	0	2	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Bacillales;f__Planococcaceae;g__Lysinibacillus	0	0	0	0	0	19	4	7	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodobacterales;f__Rhodobacteraceae;g__Ketogulonicigenium	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Elsterales;f__Elsteraceae;g__Lacibacterium	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcales-Tissierellales;g__Murdochella	3	0	0	0	0	0	0	0	0

d__Bacteria;p__Firmicutes;c__Desulfotomaculia;o__Desulfotomaculales;f__Desulfotomaculales;g__Cryptanaerobacter	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__Ochrobactrum	2	0	0	0	0	3	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__Paludicola	2	0	3	0	11	4	3	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Spirosomaceae;g__uncultured	0	11	0	0	0	3	0	5	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Paenibacillales;f__Paenibacillaceae;g__Cohnella	0	0	0	0	0	5	0	9	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Paracaedibacteriales;f__Paracaedibacteraceae;g__uncultured	0	0	1	2	0	6	0	3	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Kineosporiales;f__Kineosporiaceae;g__Quadrisphaera	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Gemmatales;f__Gemmataceae;g__Fimbrioglobus	1	0	0	0	0	9	2	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Anaerovoracaceae;g__[Eubacterium]_saphenum_group	2	0	0	0	0	5	1	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonadales;f__Sphingomonadaceae;g__Sphingourantiacus	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Cellulomonadaceae;g__Actinotalea	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Spirosomaceae;g__Larkinella	0	0	0	2	0	4	0	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Actinomycetales;f__Actinomycetaceae;g__Actinomyces	2	0	0	0	0	0	0	0	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__;	1	0	0	0	0	1	0	4	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Streptosporangiales;f__Thermomonosporaceae;g__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Syntrophomonadia;o__Syntrophomonadales;f__Syntrophomonadaceae;g__Syntrophomonas	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Acetobacteriales;f__Acetobacteraceae;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Acetobacteriales;f__Acetobacteraceae;g__Roseococcus	0	0	0	0	4	3	0	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Corynebacteriaceae;g__Lawsonella	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcales-Tissierellales;g__Helcococcus	10	0	0	0	0	0	0	0	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Polyangiales;f__Polyangiaceae;g__Pajaroellobacter	0	0	0	0	0	3	0	2	0

d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacteriales;f__Hyphomonadaceae;g__Hyphomonas	0	0	0	0	3	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Micropepsales;f__Micropepsaceae;g__uncultured	0	0	0	0	0	2	0	2	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Gemmatales;f__Gemmataceae;g__Gemmatia	0	0	1	0	2	16	2	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Alcaligenaceae;g__Verticella	0	0	0	0	0	17	0	0	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Polyangiales;f__Phaselicystidaceae;g__Phaselicystis	0	0	0	0	0	1	0	1	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Opitutales;f__Opitutaceae;g__uncultured	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Promicromonosporaceae;g__Isoptericola	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Aeromonadales;f__Succinivibrionaceae;g__	1	0	0	0	0	0	4	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales;f__NS11-12_marine_group;g__NS11-12_marine_group	0	0	0	0	0	3	0	3	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__Mesorhizobium	0	0	0	0	0	1	1	1	0
d__Bacteria;p__Acidobacteriota;c__Blastocatellia;o__Blastocatellales;f__Blastocatellaceae;g__	2	0	0	0	0	5	0	0	0
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonadales;f__Saccharimonadaceae;g__TM7x	0	0	0	0	0	0	0	0	3
d__Bacteria;p__Verrucomicrobiota;c__Kiritimatiella;o__WCHB1-41;f__WCHB1-41;g__WCHB1-41	481	0	6	0	0	30	12	40	0
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonadales;f__Saccharimonadaceae;g__Saccharimonadaceae	1	0	1	0	0	1	0	0	2
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Micrococcaceae;g__Rothia	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Lachnospiraceae_UCG-007	0	0	0	0	0	1	2	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Lachnospiraceae;g__Syntrophococcus	0	0	0	0	0	6	2	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacteriales;f__Caulobacteraceae;g__Asticcacaulis	0	0	0	0	0	3	0	2	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Frankiales;f__Geodermatophilaceae;g__Geodermatophilus	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Beijerinckiaceae;g__Methylocella	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Filimonas	0	0	0	0	0	5	0	0	0

d__Bacteria;p__Actinobacteriota;c__Acidimicrobia ;o__Microtrichales;f__lamiaceae;g__lamia	0	0	0	0	0	4	0	2	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__C ytophagales;f__Cyclobacteriaceae;g__Mariniradius	1	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobac teria;o__Rhizobiales;f__Devosiaceae;g__Pelagiba cterium	0	0	0	0	0	4	0	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria ;o__Micromonosporales;f__Micromonosporaceae; __	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteob acteria;o__Xanthomonadales;f__Xanthomonadace ae;__	0	0	0	0	0	9	1	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteob acteria;o__Burkholderiales;f__Sutterellaceae;g__S utterella	1	0	0	0	0	0	4	0	6
d__Bacteria;p__Proteobacteria;c__Gammaproteob acteria;o__Pseudomonadales;f__Pseudomonadac eae;g__Pseudomonas	6	0	0	0	0	14	5	9	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobac teria;o__Defluviicooccales;f__uncultured;g__uncultu red	0	0	0	0	0	1	0	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__C hitinophagales;f__Chitinophagaceae;g__Paraseget ibacter	0	0	0	0	0	3	0	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Pepto streptococcales- Tissierellales;f__Peptostreptococcales- Tissierellales;g__Finegoldia	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobac teria;o__Rickettsiales;f__SM2D12;g__SM2D12	0	0	0	0	0	6	0	2	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__RF39;f__ RF39;g__RF39	0	0	2	0	0	18	2	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria ;o__Micrococcales;f__Cellulomonadaceae;g__Pse udactinotalea	1	0	0	0	0	2	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria ;o__Micrococcales;f__Micrococcaceae;g__Enterac tinococcus	2	0	2	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachn ospirales;f__Lachnospiraceae;g__Herbinix	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__C hitinophagales;f__uncultured;g__uncultured	1	0	0	0	0	0	0	3	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Bacillales ;f__Bacillaceae;g__Bacillus	16	0	0	0	2	88	22	17	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobac teria;o__Rhodobacterales;f__Rhodobacteraceae;g__ Rubellimicrobium	1	0	0	0	0	1	0	0	0
d__Bacteria;p__Armatimonadota;c__Armatimonadi a;o__Armatimonadales;f__Armatimonadales;g__Ar matimonadales	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Gemmatimonadota;c__Gemmatim onadetes;o__Gemmatimonadales;f__Gemmatimon adaceae;g__Gemmatimonas	0	0	0	0	0	0	0	3	0
d__Bacteria;p__Patescibacteria;c__Dojkabacteria; o__Dojkabacteria;f__Dojkabacteria;g__Dojkabacte ria	1	0	0	0	0	0	0	2	0

d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Rurimicrobium	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Beijerinckiaceae;g__Camilimonas	0	0	0	0	0	1	0	1	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Polyangiales;f__Blrii41;g__Blrii41	1	0	0	0	0	0	0	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Actinomycetales;f__Actinomycetaceae;g__uncultured	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Stappiaceae;g__Agaricola	0	0	0	4	0	2	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Isosphaerales;f__Isosphaeraceae;g__Tundrisphaera	0	0	0	0	0	3	1	0	0
d__Bacteria;p__Bacteroidota;c__Rhodothermia;o__Rhodothermales;f__Rhodothermaceae;g__Rubrivirga	2	0	0	2	0	2	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Bacillales;f__Planococcaceae;g__Planococcus	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Butyricicoccaceae;g__UCG-008	1	0	0	0	0	1	2	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Spirosomaceae;g__Emticicia	0	0	0	0	0	3	0	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Spirosomaceae;g__Spirosoma	0	0	0	0	0	2	0	1	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Atopobiaceae;g__uncultured	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Oxalobacteraceae;g__-	1	2	0	0	0	20	0	4	1
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Acetobacteriales;f__Acetobacteraceae;g__Rhodovarius	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Armatimonadota;c__Fimbriimonadia;o__Fimbriimonadales;f__Fimbriimonadaceae;g__Fimbriimonadaceae	1	0	0	0	0	4	0	1	1
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Aeromonadales;f__Succinivibrionaceae;g__Succinatimonas	0	0	0	0	0	0	0	0	1
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Bifidobacteriales;f__Bifidobacteriaceae;g__-	2	0	2	0	0	0	0	0	0
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonadales;f__Saccharimonadaceae;g__-	2	0	0	0	0	1	1	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Alcaligenaceae;g__GKS98_freshwater_group	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales;f__Nocardioideaceae;g__-	2	0	0	0	0	2	0	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales;f__Fusibacteraceae;g__Fusibacter	0	0	0	0	13	2	0	0	0

d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacterales;f__Chthoniobacteraceae;g__LD29	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae;o__Bryobacterales;f__Bryobacteraceae;g__Bryobacter	2	0	0	0	0	0	0	1	0
d__Bacteria;p__Desulfobacterota;c__Desulfovibronia;o__Desulfovibrionales;f__Desulfovibrionaceae;g__Mailhella	9	0	16	0	0	41	2	0	0
d__Eukaryota;__;__;__;__	5	0	1	0	0	5	1	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Xanthobacteraceae;g__Anacylobacter	1	0	0	0	0	2	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Flavitalea	0	0	0	0	0	4	0	1	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Staphylococcales;f__Staphylococcaceae;g__Staphylococcus	3	0	4	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__UCG-011;g__UCG-011	2	0	0	0	0	5	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacterales;f__Propionibacteriaceae;g__Propioniciclava	1	0	0	0	0	0	0	0	1
d__Bacteria;p__Firmicutes;c__Moorellia;o__Desulfotibacterales;f__Desulfotibacteraceae;g__Desulfotibacter	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__Aminobacter	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Salinisphaerales;f__Solimonadaceae;g__Hydrocarboniphaga	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Salinisphaerales;f__Solimonadaceae;g__Solimonas	0	0	0	0	0	0	0	2	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Gemmatales;f__Gemmataceae;g__Zavarzinella	0	0	0	0	0	2	1	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales;f__Peptostreptococcaceae;g__Sporacetigenium	0	0	0	0	0	1	1	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Pseudonocardiales;f__Pseudonocardiaceae;g__Actinomycetospira	0	0	0	0	0	6	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Xanthobacteraceae;g__Vairiobacter	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Rikenellaceae;g__uncultured	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__Aquamicrobium	0	0	0	0	0	0	0	0	1
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;f__Defluviitaleaceae;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;__;__;__	2	0	0	0	0	9	1	1	0

d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__NS9_marine_group;g__NS9_marine_group	0	0	0	0	0	0	0	2	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodobacterales;f__Rhodobacteraceae;g__Pseudorhodobacter	0	0	0	0	10	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Acidimicrobia;f__Acidimicrobiaceae;g__Acidimicrobium	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Bifidobacteriales;f__Bifidobacteriaceae;g__Pseudoscardovia	0	0	0	0	0	0	5	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Kineosporiales;f__Kineosporiaceae;g__Kineococcus	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Spirochaetota;c__Spirochaetia;o__Spirochaetales;f__Spirochaetaceae;g__Spirochaeta	22	0	3	0	0	1	2	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__Ensifer	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Acidimicrobia;o__Microtrichales;f__Ilumatobacteraceae;g__Ilumatobacter	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodobacterales;f__Rhodobacteraceae;g__Amaricoccus	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonadales;f__Sphingomonadaceae;g__Sphingosinicella	0	0	0	0	0	1	0	1	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Merdibacter	0	0	0	0	0	0	0	0	2
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales;f__Peptostreptococcaceae;g__Clostridioides	0	0	0	0	0	1	0	0	1
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Holdemania	1	0	0	0	0	0	0	0	1
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Actinomycetales;f__Actinomycetaceae;g__Molibiluncus	3	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Weeksellaceae;g__Bergeyella	4	0	0	0	0	0	1	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodobacterales;f__Rhodobacteraceae;g__Tabrizicola	1	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Carnobacteriaceae;g__Carnobacter	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales;f__Selenomonadaceae;g__Selenomonas	2	0	1	0	0	0	0	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia;f__Hungateiclostridiaceae;g__Hungateiclostridiaceae	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Synergistota;c__Synergistia;o__Synergistales;f__Synergistaceae;g__Fretibacterium	0	0	2	0	0	1	0	0	1
d__Bacteria;p__Desulfobacterota;c__Desulfuromonadia;o__PB19;f__PB19;g__PB19	1	0	0	0	0	0	0	3	0

d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__37-13;g__37-13	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__MWH-CFBk5;g__MWH-CFBk5	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Turicibacter	0	0	3	0	0	5	1	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Microbacteriaceae;g__Leucobacter	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Methylophilaceae;g__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Gemmatimonadota;c__Gemmatimonadetes;o__Gemmatimonadales;f__Gemmatimonadaceae;g__uncultured	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Isosphaerales;f__Isosphaeraceae;g__	0	0	0	0	0	2	0	4	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Spirosomaceae;g__Fibrisoma	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales;f__Veillonellaceae;g__Veillonella	0	0	0	0	0	0	0	0	5
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Beutenbergiaceae;g__	1	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Actinomycetales;f__Actinomycetaceae;g__Truoperella	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Synergistota;c__Synergistia;o__Synergistales;f__Synergistaceae;g__Cloacibacillus	1	3	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Ectothiorhodospirales;f__Thioalkalspiraceae;g__Thioalkalspiraceae-Sulfurivermis	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Cyanobacteriales;f__Chroococcidiopsaceae;g__uncultured	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Dermabacteraceae;g__Dermabacter	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;f__	1	0	0	0	0	0	3	1	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Opitutales;f__Opitutaceae;g__Opitutus	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Cytophagaceae;g__Cytophaga	0	0	0	0	0	1	0	2	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Microscillaceae;g__uncultured	1	0	1	0	0	3	0	3	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia;f__Hungateiclostridiaceae;g__uncultured	0	0	0	0	0	1	0	1	0

d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Limnotrichales;f__Limnotrichaceae;g__Limnotrix	0	0	0	0	5	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Rhodocyclaceae;g__Thauera	0	0	0	0	8	0	0	0	0
d__Bacteria;p__Desulfobacterota;c__Desulfuromonadia;o__Bradymonadales;f__Bradymonadales;g__Bradymonadales	10	0	1	0	0	3	0	2	0
d__Archaea;p__Halobacterota;c__Methanosarcinia;o__Methanosarciniales;f__Methanosarcinaceae;g__Methanomicrococcus	1	0	0	0	0	1	0	2	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Methylobacteriaceae;g__uncultured	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Tissierella	3	0	2	0	0	8	0	0	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Acidaminococcales;f__Acidaminococcaceae;g__Succiniclasticum	0	0	2	0	0	4	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Kineosporiales;f__Kineosporiaceae;g__Pseudokineococcus	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Atopobiaceae;g__Coriobacteriaceae_UCG-002	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales;f__Candidatus_Hepaticola;g__Candidatus_Hepaticola	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Acetobacteriales;f__Acetobacteraceae;g__Siccirubricoccus	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Intrasporangiaceae;g__Phycococcus	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Sutterellaceae;g__Parasutterella	1	0	1	4	0	13	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Flavisolibacter	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Haoranjia	0	0	0	0	0	0	0	0	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Flavobacteriaceae;g__Salinimicrobium	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Frankiales;f__Geodermatophilaceae;g__Antriccoccus	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcales-Tissierellales;g__Tissierella	4	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Lactobacillaceae;g__Lactobacillus	1	0	0	0	0	0	5	3	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Planctomycetales;f__Schlesneriaceae;g__Planctopirius	1	0	0	0	0	1	0	2	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Pseudanabaenales;f__Pseudanabaenaceae;g__Pseudanabaena_PCC-7429	0	0	0	0	3	0	0	1	0

d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcaceae;g__Proteocatella	0	0	0	0	6	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__S-BQ2-57_soil_group;f__S-BQ2-57_soil_group;g__S-BQ2-57_soil_group	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__Anaerolineales;f__Anaerolineaceae;g__uncultured	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Pirellulales;f__Pirellulaceae;g__Blastopirellula	1	0	1	0	0	3	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Planctomycetales;f__Rubinisphaeraceae;g__uncultured	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae;o__Acidobacteriae;f__Acidobacteriae;g__Paludibaculum	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Kapabacteria;o__Kapabacteriales;f__Kapabacteriales;g__Kapabacteriales	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Campilobacterota;c__Campylobacteria;o__Campylobacteriales;f__Helicobacteraceae;g__Helicobacter	0	0	0	0	0	0	1	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Diplorickettsiales;f__Diplorickettsiaceae;g__Aquicella	0	0	0	0	0	1	0	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Devosiaceae;g__	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__	0	0	0	0	0	2	0	2	0
d__Bacteria;p__Desulfobacterota;c__Desulfovibrionia;o__Desulfovibrionales;f__Desulfovibrionaceae;g__Desulfovibrio	1	0	2	0	0	1	6	1	0
d__Bacteria;p__Acidobacteriota;c__Vicinamibacteria;o__Vicinamibacteriales;f__Vicinamibacteraceae;g__Vicinamibacteraceae	2	0	0	0	0	2	0	1	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Coriobacteriales_Incertae_Sedis;g__Raoultibacter	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Frankiales;f__Frankiaceae;g__Jatrophihabans	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;f__Clostridiaceae;g__Candidatus_Arthromitus	0	0	0	0	0	1	1	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Xanthomonadales;f__Xanthomonadaceae;g__Luteimonas	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcales-Tissierellales;g__Ezakiella	0	0	0	0	0	0	0	0	1
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__SBR1031;f__A4b;g__A4b	1	0	0	0	0	0	0	0	1
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cyclophagales;f__Cyclobacteriaceae;g__	3	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales;f__Propionibacteriaceae;g__Tessaracoccus	1	0	0	0	0	0	0	0	0

d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Flavobacteriaceae;g__Salegentibacter	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Spirochaetota;c__Spirochaetia;o__Spirochaetales;f__Spirochaetaceae;g__Sediminispirochaeta	1	0	1	0	0	0	0	0	0
d__Archaea;p__Euryarchaeota;c__Methanobacteria;o__Methanobacteriales;f__Methanobacteriaceae;g__Methanobacteriaceae	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Bacteroidetes_vadinHA17;g__Bacteroidetes_vadinHA17	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcales-Tissierellales;g__uncultured	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Pedosphaerales;f__Pedosphaeraceae;g__	1	0	0	0	0	0	1	0	0
d__Bacteria;p__Patescibacteria;c__ABY1;o__Candidatus_Kerfeldbacteria;f__Candidatus_Kerfeldbacteria;g__Candidatus_Kerfeldbacteria	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales;f__Rickettsiaceae;g__uncultured	0	0	0	0	0	1	0	1	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Phycisphaerales;f__Phycisphaeraceae;g__uncultured	0	0	1	0	0	0	0	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Nitriliruptorales;f__Nitriliruptoraceae;g__Egicoccus	1	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidetes_VC2.1_Bac22;f__Bacteroidetes_VC2.1_Bac22;g__Bacteroidetes_VC2.1_Bac22	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__KD4-96;o__KD4-96;f__KD4-96;g__KD4-96	2	0	0	0	0	3	1	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cyclophagales;f__Cyclobacteriaceae;g__Lunatimonas	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cyclophagales;f__Hymenobacteraceae;g__Pontibacter	1	0	0	0	0	0	1	0	0
d__Archaea;p__Thermoplasmatota;c__Thermoplasmatas;o__Methanomassiliicoccales;f__Methanomyxosphaeraceae;g__RumEn_M2	0	0	0	0	0	0	1	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiales_Incertae_Sedis;g__Phreatobacter	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Planctomycetales;f__Rubinisphaeraceae;g__Rubinisphaera	0	0	0	0	0	1	1	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Eubacteriales;f__Garcillaceae;g__Irregularibacter	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Bacteroidota;c__Rhodothermia;o__Balneolales;f__Balneolaceae;g__CK06-06-MudMAS4B-21	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Cyanobacteriales;f__Microcystaceae;g__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;g__	1	0	0	0	0	0	0	1	0

d_Bacteria;p_Firmicutes;c_Bacilli;o_Paenibacillales;f_Paenibacillaceae;g_Ammuniphilus	0	0	0	0	0	1	0	0	0
d_Bacteria;p_Proteobacteria;c_Gammaproteobacteria;o_Burkholderiales;f_Alcaligenaceae;g_Achromobacter	0	0	0	0	0	5	0	0	0
d_Bacteria;p_Gemmatimonadota;c_Gemmatimonadetes;o_Gemmatimonadales;f_Gemmatimonadaceae;g_Roseisolibacter	0	0	0	0	0	3	0	0	0
d_Bacteria;p_Bacteroidota;c_Bacteroidia;o_Chitinophagales;f_Chitinophagaceae;g_Niastella	0	0	0	0	0	1	0	0	0
d_Bacteria;p_Proteobacteria;c_Gammaproteobacteria;o_Burkholderiales;f_Comamonadaceae;g_Comamonas	0	0	0	0	0	3	2	1	0
d_Bacteria;p_Proteobacteria;c_Alphaproteobacteria;o_Rhizobiales;f_Xanthobacteraceae;g_Bradyrhizobium	0	0	0	0	0	1	0	0	0
d_Bacteria;p_Proteobacteria;c_Gammaproteobacteria;o_Burkholderiales;f_Methylophilaceae;g_Methylophilus	0	0	0	0	0	3	0	0	0
d_Bacteria;p_Cyanobacteria;c_Cyanobacteriia;o_Cyanobacteriales;f_Nostocaceae;g_Nostoc_PCC-7107	0	0	0	0	0	1	0	0	0
d_Bacteria;p_Actinobacteriota;c_Actinobacteria;o_Streptomycetales;f_Streptomycetaceae;g_	0	0	0	0	0	1	0	0	0
d_Bacteria;p_Proteobacteria;c_Alphaproteobacteria;o_Rickettsiales;g_	0	0	1	0	0	2	0	1	0
d_Bacteria;p_Bacteroidota;c_Bacteroidia;o_Cytophagales;f_Cytophagaceae;g_Rhodocytophaga	0	0	0	0	0	3	0	0	0
d_Bacteria;p_Proteobacteria;c_Gammaproteobacteria;o_Xanthomonadales;f_Xanthomonadaceae;g_Pseudoxanthomonas	0	0	0	0	0	5	0	0	0
d_Bacteria;p_Actinobacteriota;c_Acidimicrobia;o_Microtrichales;f_Microtrichaceae;g_IMCC26207	1	0	0	0	0	1	0	0	0
d_Bacteria;p_Firmicutes;c_Clostridia;o_Oscillospirales;f_Ruminococcaceae;g_Anaerotruncus	0	0	0	0	0	0	0	0	1
d_Bacteria;p_Firmicutes;c_Bacilli;o_Erysipelotrichales;f_Erysipelatoclostridiaceae;g_Candidatus_Stoquefichus	0	0	1	0	0	0	0	0	0
d_Bacteria;p_Firmicutes;c_Clostridia;o_Lachnospirales;f_Lachnospiraceae;g_Lachnospiraceae_NC2004_group	3	0	6	0	0	5	1	0	0
d_Bacteria;p_Bacteroidota;c_Bacteroidia;o_Bacteroidales;f_Porphyromonadaceae;g_Porphyromonas	3	0	0	0	0	0	0	0	1
d_Bacteria;p_Firmicutes;c_Syntrophomonadia;o_Syntrophomonadales;f_Syntrophomonadaceae;g_	1	0	0	0	0	0	0	0	0
d_Bacteria;p_Chloroflexi;c_JG30-KF-CM66;o_JG30-KF-CM66;f_JG30-KF-CM66;g_JG30-KF-CM66	1	0	0	0	0	0	0	0	0
d_Bacteria;p_Chloroflexi;c_Anaerolineae;o_Caldilineales;f_Caldilineaceae;g_uncultured	2	0	2	0	0	4	0	1	0
d_Bacteria;p_Proteobacteria;c_Alphaproteobacteria;o_Acetobacteriales;f_Acetobacteraceae;g_Acetobacter	1	0	2	0	0	0	0	0	0

d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Actinomycetales;f__Actinomycetaceae;__	2	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Lachnospirales;__;__	2	0	2	0	0	3	2	3	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales-Selenomonadales;f__Selenomonadaceae;g__uncultured	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Patescibacteria;c__ABY1;o__Candidatus_Falkowbacteria;f__Candidatus_Falkowbacteria;g__Candidatus_Falkowbacteria	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Acidaminococcales;f__Acidaminococcaceae;__	5	0	0	0	0	2	0	0	0
d__Bacteria;p__Actinobacteriota;c__Thermoleophilina;o__Solirubrobacterales;f__67-14;g__67-14	1	0	0	0	0	0	0	0	0
d__Archaea;p__Aenigmarchaeota;c__Deep_Sea_Euryarchaeotic_Group(DSEG);o__Deep_Sea_Euryarchaeotic_Group(DSEG);f__Deep_Sea_Euryarchaeotic_Group(DSEG);g__Deep_Sea_Euryarchaeotic_Group(DSEG)	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Alcaligenaceae;g__Parapusillimonas	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Phycisphaerales;f__Phycisphaeraceae;g__AKYG587	1	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__OPB41;f__OPB41;g__OPB41	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Xanthomonadales;f__Xanthomonadaceae;g__Lysobacter	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Bifidobacteriales;f__Bifidobacteriaceae;g__Parascardovia	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridia;__;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__Erysipelotrichaceae_UCG-008	1	0	1	0	0	0	0	0	0
d__Bacteria;p__Verrucomicrobiota;__;__;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__gir-aah93h0;g__gir-aah93h0	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Patescibacteria;c__Saccharimonadia;o__Saccharimonadales;f__S32;g__TM7	0	0	0	0	0	0	1	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Oceanospirillales;__;__	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Chloroflexi;c__Dehalococcoidia;o__S085;f__S085;g__S085	1	0	0	0	0	1	1	2	0
d__Bacteria;p__Actinobacteriota;c__Acidimicrobiia;o__Microtrichales;f__Ilumatobacteraceae;g__uncultured	1	0	0	0	0	1	0	1	0

d__Bacteria;p__Cyanobacteria;c__Vampirivibrionia;o__Obscuribacterales;f__Obscuribacteraceae;g__Obscuribacteraceae	0	0	0	0	4	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prolixibacteraceae;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Pedosphaerales;f__Pedosphaeraceae;g__ADurb.Bin063-1	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcales-Tissierellales;g__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Enterococcaceae;g__Enterococcus	0	0	0	0	0	0	3	1	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;f__Clostridiaceae;g__Clostridium_sensu_stricto_10	0	0	0	0	0	0	1	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Ferruginibacter	0	0	0	0	0	0	1	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Aeromonadales;f__Aeromonadaceae;g__Aeromonas	1	0	0	0	0	0	1	0	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales-Selenomonadales;f__Selenomonadaceae;g__Mitsuokella	0	0	0	0	0	0	4	0	0
d__Bacteria;p__Deferribacterota;c__Deferribacteres;o__Deferribacterales;f__Deferribacteraceae;g__Mucispirillum	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Paenibacillales;f__Paenibacillaceae;g__	0	0	0	0	0	1	0	3	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Streptosporangiales;f__Streptosporangiaceae;g__	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Thermotaleaceae;g__	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Cyanobacteriales;f__Microcystaceae;g__Cyanothece_PCC-8801	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Synechococcales;f__Cyanobiaceae;g__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__	1	0	2	0	0	3	4	1	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;f__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__	1	0	0	0	0	2	0	1	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Corynebacteriales;f__Nocardaceae;g__	0	0	0	0	0	5	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Oxalobacteraceae;g__Duganella	0	0	0	0	0	4	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Puniceispirillales;f__uncultured;g__uncultured	0	0	0	0	0	1	0	0	0

d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales;f__Propionibacteriaceae;g__Mariniluteococcus	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Pedosphaerales;f__Pedosphaeraceae;g__DEV008	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales;f__Rickettsiaceae;g__Candidatus_Megaira	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__Tianweiania	0	0	1	2	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Dermatophilaceae;g__uncultured	0	0	0	0	0	3	1	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Synechococcales;f__Cyanobiaceae;g__Cyanobium_PCC-6307	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacteriales;__	0	0	0	0	0	3	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales;f__Propionibacteriaceae;g__Microlunatus	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Aerococcaceae;g__Aerococcus	3	0	1	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;c__OM190;o__OM190;f__OM190;g__OM190	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Izermoplasmales;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Coriobacteriia;o__Coriobacteriales;f__Coriobacteriales;g__Coriobacteriales	2	0	0	0	0	0	1	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Alcaligenaceae;g__Oligella	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Myxococcota;__;__;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Pedosphaerales;f__Pedosphaeraceae;g__Pedosphaeraceae	1	0	0	0	0	2	1	0	0
d__Bacteria;p__Planctomycetota;__;__;__	3	0	0	0	0	0	1	0	0
d__Bacteria;p__Firmicutes;c__Desulfotomaculia;o__Desulfotomaculales;f__Desulfallas-Sporotomaculum;g__Desulfallas-Sporotomaculum	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Chloroflexi;c__Ktedonobacteria;o__C0119;f__C0119;g__C0119	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Armatimonadota;c__Chthonomonadetes;o__Chthonomonadales;f__Chthonomonadaceae;g__Chthonomonas	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales;f__env.OPS_17;g__env.OPS_17	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Neisseriaceae;g__Siemoniella	1	0	0	0	0	0	0	0	0

d__Bacteria;p__Proteobacteria;__;__;__	1	0	0	0	0	2	0	1	0
d__Archaea;p__Euryarchaeota;c__Methanobacteria;o__Methanobacteriales;f__Methanobacteriaceae;__	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Patescibacteria;c__Parcubacteria;o__Candidatus_Moranbacteria;f__Candidatus_Moranbacteria;g__Candidatus_Moranbacteria	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Thermoleophilina;o__Gaiellales;__;__	1	0	1	0	0	0	0	0	0
d__Bacteria;p__Acidobacteriota;c__Subgroup_5;o__Subgroup_5;f__Subgroup_5;g__Subgroup_5	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Lentisphaeria;o__Victivallales;f__Victivallaceae;__	0	0	1	0	0	3	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Oscillospirales;g__Oscillospirales	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Acidobacteriota;c__Blastocatellia;o__Blastocatellales;f__Blastocatellaceae;g__Aridibacter	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Flavobacteriaceae;g__Aequorivita	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Pirellulales;f__Pirellulaceae;g__Roseimariima	2	0	1	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Ruminococcaceae;g__Harryflintia	0	0	0	0	0	0	1	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__ZOR0006	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales-Selenomonadales;f__Veillonellaceae;g__Megasphaera	0	0	0	0	0	0	1	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiaceae;g__Pseudocrohobactrum	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Cellvibrionales;f__Cellvibrionaceae;g__Cellvibrion	0	0	0	0	0	3	1	2	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcaceae;g__uncultured	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Sphingobacteriales;f__AKYH767;g__AKYH767	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Acidobacteriota;c__Subgroup_18;o__Subgroup_18;f__Subgroup_18;g__Subgroup_18	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__SJA-15;f__SJA-15;g__SJA-15	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacteriales;f__Hyphomonadaceae;g__SWB02	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__Anaerolineales;f__Anaerolineaceae;__	0	0	0	0	0	1	0	0	0

d__Bacteria;p__Planctomycetota;c__Pla4_lineage;o__Pla4_lineage;f__Pla4_lineage;g__Pla4_lineage	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Omnitrophia;o__Omnitrophales;f__Omnitrophaceae;g__Candidatus_Omnitrophus	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Alteromonadales;f__Alteromonadaceae;g__Rheinheimera	0	0	0	0	4	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Methylophilaceae;g__Methylotenera	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Sediminibacterium	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Paracaeidibacteriales;f__Paracaeidibacteraceae;g__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae;o__Elev-16S-1166;f__Elev-16S-1166;g__Elev-16S-1166	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Pedosphaerales;f__Pedosphaeraceae;g__SH3-11	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhodospirillales;f__Rhodospirillaceae;g__uncultured	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__uncultured;f__uncultured;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Acidobacteriota;c__Blastocatellia;o__Blastocatellales;f__Blastocatellaceae;g__Stenotrophobacter	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Tepidisphaerales;f__CPLA-3_termite_group;g__CPLA-3_termite_group	1	0	0	0	0	1	0	0	0
d__Bacteria;p__Hydrogenedentes;c__Hydrogenedentia;o__Hydrogenedentiales;f__Hydrogenedensaceae;g__Hydrogenedensaceae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Cyanobacteriales;f__Oscillatoriaceae;g__Phoridium_ETS-05	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Aeromonadales;f__Succinivibrionaceae;g__Anaerobiospirillum	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Prolixibacteraceae;g__WCHB1-32	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Veillonellales-Selenomonadales;f__Sporomusaceae;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Phycisphaerales;f__Phycisphaeraceae;g__CL500-3	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Alteromonadales;f__Alteromonadaceae;g__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Reyranellales;f__Reyranellaceae;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacteriales;f__Chthoniobacteraceae;g__Candidatus_Udaeobacter	0	0	0	0	0	1	2	0	0

d__Bacteria;p__Firmicutes;c__Clostridia;o__Oscillospirales;f__Butyricicoccaceae;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Acidobacteriota;c__Vicinamibacteriia;o__Vicinamibacteriales;f__uncultured;g__uncultured	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Chloroflexi;c__Dehalococcoidia;o__SAR202_clade;f__SAR202_clade;g__SAR202_clade	0	0	0	0	0	1	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Aquaspirillaceae;g__Microvirgula	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Gemmatimonadota;c__S0134_terrestrial_group;o__S0134_terrestrial_group;f__S0134_terrestrial_group;g__S0134_terrestrial_group	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Patescibacteria;c__Gracilibacteria;o__JGI_0000069-P22;f__JGI_0000069-P22;g__JGI_0000069-P22	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Marinifilaceae;g__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Microscillaceae;g__OLB12	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Chitinophagales;f__Chitinophagaceae;g__Lacibacter	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__Anaerolineae;o__uncultured;f__uncultured;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__mle1-27;f__mle1-27;g__mle1-27	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Planctomycetales;f__Gimesiaceae;g__uncultured	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Pla1_lineage;f__Pla1_lineage;g__Pla1_lineage	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Streptosporangiales;f__Nocardiopsaceae;g__Nocardiopsis	0	0	0	0	0	1	0	0	0
d__Eukaryota;p__Ascomycota;__;__;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Alcaligenaceae;g__Pigmentiphaga	0	0	0	0	0	7	0	0	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriiae;o__Solibacterales;f__Solibacteraceae;g__Candidatus_Solibacter	0	0	0	0	0	1	0	0	0
d__Eukaryota;p__Arthropoda;__;__;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;__;__;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rickettsiales;f__AB1;g__AB1	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Actinobacteriota;c__Thermoleophilina;o__Solirubrobacterales;f__Solirubrobacteraceae;g__Solirubrobacter	0	0	0	0	0	1	0	0	0

d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Cyanobacteriales;f__Nostocaceae;g__Sphaerospermopsis_BCCUSP55	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Chthoniobacterales;f__01D2Z36;g__01D2Z36	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Opitutales;f__Puniceicoccaceae;g__Verruc-01	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;__;__;__	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Paracaeidibacterales;f__Paracaeidibacteraceae;g__Candidatus_Captivus	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Caulobacterales;f__Hyphomonadaceae;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Cyanobacteriales;f__Phormidiaceae;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Bacillales;f__Planococcaceae;g__Psychrobacillus	0	0	0	0	0	1	3	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Oxyphotobacteria_Incertae_Sedis;f__Unknown_Family;g__Leptolyngbya_ANT.L52.2	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Alcaligenaceae;g__Bordetella	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Cyanobacteriales;f__Coleofasciculaceae;g__Microcoleus_Es-Yyy1400	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Cyanobacteriales;f__Phormidiaceae;g__Tycho nema_CCAP_1459-11B	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Patescibacteria;c__Berkelbacteria;o__Berkelbacteria;f__Berkelbacteria;g__Berkelbacteria	0	0	1	0	0	1	0	0	0
d__Bacteria;p__Myxococcota;c__Polyangia;o__Blf di19;f__Blfdi19;g__Blfdi19	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Spirosomaceae;g__Fibrella	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Spirochaetota;c__Brachyspirae;o__Brachyspirales;f__Brachyspiraceae;g__Brachyspira	0	0	0	0	0	0	0	0	1
d__Bacteria;p__Firmicutes;c__Negativicutes;o__Acidaminococcales;f__Acidaminococcaceae;g__Acidaminococcus	0	0	0	0	0	0	0	0	2
d__Bacteria;p__Actinobacteriota;c__Thermoleophil ia;o__Gaiellales;f__uncultured;g__uncultured	0	0	0	0	0	0	0	0	1
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcales-Tissierellales;g__Gallicola	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Acidobacteriota;c__Blastocatellia;o__DS-100;f__DS-100;g__DS-100	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Rhodocyclaceae;g__Azoarcus	1	0	0	0	0	0	0	0	0

d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Sphingomonadales;f__Sphingomonadaceae;g__Sphingorhabdus	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Acidobacteriota;c__Acidobacteriae;o__Acidobacteriales;f__Acidobacteriaceae_(Subgroup_1);g__Bryocella	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Enterobacterales;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__Tepidisphaerales;f__Tepidisphaerales;g__Tepidisphaerales	2	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Aerococcaceae;g__Facklamia	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Acidobacteriota;c__Blastocatellia;o__Blastocatellales;f__Blastocatellaceae;g__JGI_0001001-H03	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;c__Phycisphaerae;o__S-70;f__S-70;g__S-70	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Aerococcaceae;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Pseudonocardiales;f__Pseudonocardaceae;g__Saccharopolyspora	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Azospirillales;f__uncultured;g__uncultured	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Cyclobacteriaceae;g__uncultured	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Eubacteriales;f__Eubacteriales;g__Alkalibaculum	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Flavobacteriaceae;g__Subsaxibacter	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Flavobacteriales;f__Flavobacteriaceae;g__Capnocytophaga	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Planctomycetota;c__Planctomycetes;o__Gemmatiales;f__Gemmataceae;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Cyanobacteria;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Propionibacteriales;f__Propionibacteriaceae;__	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Xanthomonadales;f__Rhodanobacteraceae;g__Luteibacter	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Nitrilibrupatorales;f__Nitrilibrupatoraceae;g__Nitrilibrupator	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Peptostreptococcales-Tissierellales;f__Peptostreptococcales-Tissierellales;g__Guggenheimella	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Micrococcales;f__Dermacoccaceae;__	1	0	0	0	0	0	0	0	0

d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Kiloniellales;f__Fodinicurvataceae;g__uncultured	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Myxococcota;c__Myxococcia;o__Myxococcales;f__Myxococcaceae;g__uncultured	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Erysipelotrichales;f__Erysipelotrichaceae;g__[Clostridium]_innocuum_group	1	0	0	0	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Lactobacillales;f__Streptococcaceae;g__Lactococcus	1	0	0	0	0	0	1	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Rhizobiales_Incertae_Sedis;g__uncultured	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Xanthobacteraceae;g__uncultured	0	0	1	0	0	0	0	0	0
d__Bacteria;p__Patescibacteria;c__uncultured;o__uncultured;f__uncultured;g__uncultured	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Actinobacteriota;c__Acidimicrobiia;o__Microtrichales;f__Ilumatobacteraceae;g__CL500-29_marine_group	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Oceanospirillales;f__Saccharospirillaceae;g__Oceanobacter	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Hyphomicrobiaceae;g__Hyphomicrobium	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Bacteroidales;f__Paludibacteraceae;g__	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__LD1-PB3;f__LD1-PB3;g__LD1-PB3	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Synechococcales;f__Synechococcaceae;g__Synechococcus_PCC-7942	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Spirochaetota;c__Leptospirae;o__Leptospirales;f__Leptospiraceae;g__RBG-16-49-21	0	0	0	0	0	0	0	1	0
d__Bacteria;p__Bacteroidota;c__SJA-28;o__SJA-28;f__SJA-28;g__SJA-28	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Chitinimonadaceae;g__Chitinimonas	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Verrucomicrobiota;c__Verrucomicrobiae;o__Pedosphaerales;f__Pedosphaeraceae;g__RS25G	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Cyanobacteria;c__Cyanobacteriia;o__Cyanobacteriales;f__Microcystaceae;g__Synechocystis_PCC-6803	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Rhodothermia;o__Balneolales;f__Balneolaceae;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Parvibaculales;f__Parvibaculaceae;g__Parvibaculum	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Fusobacteriota;c__Fusobacteriia;o__Fusobacteriales;f__Leptotrichiaceae;g__Hypnocyclus	0	0	0	0	0	1	0	0	0

d__Bacteria;p__Chloroflexi;c__Chloroflexia;o__Thermomicrobiales;f__AKYG1722;g__AKYG1722	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;c__Gitt-GS-136;o__Gitt-GS-136;f__Gitt-GS-136;g__Gitt-GS-136	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Xanthomonadales;f__Rhodanobacteraceae;g__Rhodanobacter	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Burkholderiaceae;__	0	0	0	0	0	2	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Salinisphaerales;f__Solimonadaceae;g__Panacagrimonas	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Bacteroidia;o__Cytophagales;f__Amoebophilaceae;g__Candidatus_Amoebophilus	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Xanthomonadales;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Methylophilaceae;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Burkholderiales;f__Oxalobacteraceae;g__Noviherbaspirillum	0	0	0	1	0	0	0	0	0
d__Bacteria;p__Firmicutes;c__Clostridia;o__Clostridiales;f__Clostridiaceae;g__Clostridium_sensu_stricto_8	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Gammaproteobacteria;o__Enterobacterales;f__Erwinaceae;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Proteobacteria;c__Alphaproteobacteria;o__Rhizobiales;f__Hyphomicrobiaceae;g__uncultured	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Actinobacteriota;c__Actinobacteria;o__Frankiales;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Bacteroidota;c__Rhodothermia;o__Rhodothermales;f__Rhodothermaceae;g__Rhodothermaceae	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Chloroflexi;__;__;__	0	0	0	0	0	1	0	0	0
d__Bacteria;p__Firmicutes;c__Bacilli;o__Staphylococcales;f__Staphylococcaceae;__	0	0	1	0	0	0	0	0	0