FACULDADE DE MEDICINA
UNIVERSIDADE DO PORTO

MESTRADO INTEGRADO EM MEDICINA

2015/2016

Carolina Germana Jardim Fernandes Rodrigues da Silva

Lipodystrophy and cardiovascular risk in HIV-infected patients under antiretroviral therapy – comparing SCORE, Framingham, ASCVD and metabolic syndrome

março, 2016

Carolina Germana Jardim Fernandes Rodrigues da Silva

Lipodystrophy and cardiovascular risk in HIV-infected patients under antiretroviral therapy – comparing SCORE, Framingham, ASCVD and metabolic syndrome

Mestrado Integrado em Medicina

Área: Endocrinologia

Tipologia: Dissertação

Trabalho efetuado sob a Orientação de:

Professora Doutora Paula Freitas

Trabalho organizado de acordo com as normas da revista:

BMC Infectious Diseases

março, 2016

FMUP

U.PORTO
FMUP FACULDADE DE MEDICINA UNIVERSIDADE DO PORTO
UNIDADE CURRICULAR
PROJETO DE OPÇÃO
DISSERTAÇÃO / MONOGRAFIA

Projeto de Opção do 6º ano - DECLARAÇÃO DE INTEGRIDADE

Eu, Carolina Germana Jardim Fernandes Rodrigues da Silva, abaixo assinado, nº mecanográfico 201002332, estudante do 6º ano do Ciclo de Estudos Integrado em Medicina, na Faculdade de Medicina da Universidade do Porto, declaro ter atuado com absoluta integridade na elaboração deste projeto de opção.

Neste sentido, confirmo que **NÃO** incorri em plágio (ato pelo qual um indivíduo, mesmo por omissão, assume a autoria de um determinado trabalho intelectual, ou partes dele). Mais declaro que todas as frases que retirei de trabalhos anteriores pertencentes a outros autores, foram referenciadas, ou redigidas com novas palavras, tendo colocado, neste caso, a citação da fonte bibliográfica.

Faculdade de Medicina da Universidade do Porto, 23/03/2016

Assinatura conforme cartão de identificação:

Carolina Germana

U.PORTO
FMUP FACULDADE DE MEDICINA UNIVERSIDADE DO PORTO
UNIDADE CURRICULAR PROJETO DE OPÇÃO DISSERTAÇÃO / MONOGRAFIA

Projecto de Opção do 6º ano – DECLARAÇÃO DE REPRODUÇÃO

NOME

Carolina Gennana Jardim Fernandes Rodrigues da Silva

NÚMERO DE ESTUDANTE	DATA DE CONCLUSÃO
201002331	

DESIGNAÇÃO DA ÁREA DO PROJECTO

Endocrinologia

TÍTULO DISSERTAÇÃO/~~MONOGRAFIA~~ (riscar o que não interessa)

Lipodystrophy and cardiovascular risk in HIV-infected patients under antiretroviral therapy - comparing SCORE, Framingham, ASCVD and metabolic syndrome

ORIENTADOR

Paula Isabel Marques Simões Freitas

COORIENTADOR (se aplicável)

ASSINALE APENAS UMA DAS OPÇÕES:

É AUTORIZADA A REPRODUÇÃO INTEGRAL DESTE TRABALHO APENAS PARA EFEITOS DE INVESTIGAÇÃO, MEDIANTE DECLARAÇÃO ESCRITA DO INTERESSADO, QUE A TAL SE COMPROMETE.	☒
É AUTORIZADA A REPRODUÇÃO PARCIAL DESTE TRABALHO (INDICAR, CASO TAL SEJA NECESSÁRIO, Nº MÁXIMO DE PÁGINAS, ILUSTRAÇÕES, GRÁFICOS, ETC.) APENAS PARA EFEITOS DE INVESTIGAÇÃO, MEDIANTE DECLARAÇÃO ESCRITA DO INTERESSADO, QUE A TAL SE COMPROMETE.	☐
DE ACORDO COM A LEGISLAÇÃO EM VIGOR, (INDICAR, CASO TAL SEJA NECESSÁRIO, Nº MÁXIMO DE PÁGINAS, ILUSTRAÇÕES, GRÁFICOS, ETC.) NÃO É PERMITIDA A REPRODUÇÃO DE QUALQUER PARTE DESTE TRABALHO.	☐

Faculdade de Medicina da Universidade do Porto, 23/03/2016

Assinatura conforme cartão de identificação: Carolina Gennana

Para os meus pais,

avós,

André

e Laura

Title: Lipodystrophy and cardiovascular risk in HIV-infected patients under antiretroviral therapy – comparing SCORE, Framingham, ASCVD and metabolic syndrome

Short title: Lipodystrophy in HIV-infected patients – comparing cardiovascular disease risk scores and metabolic syndrome

Carolina Germana Silva[1*], Ana Rita Gomes[2], Francisco Almeida[3], António Sarmento[4], Ana Cristina Santos[5], Davide Cavalho[6], Paula Freitas[7]

[1]* Corresponding author: Medical Student. University of Porto Medical School. Alameda Prof. Hernâni Monteiro 4200-319 Porto, Portugal. mimed10016@med.up.pt

[2] Medical Student. University of Porto Medical School. Alameda Prof. Hernâni Monteiro 4200-319 Porto, Portugal. mimed10099@med.up.pt

[3] Infectious Diseases Department, São João Hospital, University of Porto Medical School, Porto, Portugal. franciscomlrmalmeida@med.up.pt

[4] Infectious Diseases Department, São João Hospital, University of Porto Medical School, Porto, Portugal. asarment@med.up.pt

[5] Clinical Epidemiology, Predictive Medicine and Public Health, University of Porto Medical School, and EPIUnit – Epidemiological Research Unit, Institute of Public Health University of Porto, Porto, Portugal. acsantos@med.up.pt

[6] Endocrinology, Diabetes and Metabolism Department, São João Hospital, University of Porto Medical School, i3S – Instituto de Investigação e Inovação em Saúde, University of Porto, Porto, Portugal. carvdav@med.up.pt

[7] Endocrinology, Diabetes and Metabolism Department, São João Hospital, University of Porto Medical School, i3S – Instituto de Investigação e Inovação em Saúde, University of Porto, Porto, Portugal. pimsfreitas@med.up.pt

Abstract

Background: Both HIV infection and combined antiretroviral therapy (cART) are associated with metabolic and fat distribution changes, as well as cardiovascular disease. **Methods**: A cross-sectional study was performed in 572 HIV1-infected Caucasian adults on cART, who were grouped according to four different groups of fat distribution. Cardiovascular disease risk was measured using Framingham-CHD, SCORE and ASCVD scoring systems and the agreement between them was evaluated. The concordance between metabolic syndrome and cardiovascular disease risk scores was assessed. **Results**: Among 572 HIV-infected patients, 70% were male, 31.29% had isolated central fat accumulation, 26.40% had isolated lipoatrophy, 26.05% registered mixed forms of lipodystrophy, and 16.26% of the patients had no lipodystrophy. The prevalence of metabolic syndrome in our sample was 49.22%. Approximately 70% of patients with mixed forms of lipodystrophy (*vs* 30% of non-lipodystrophic patients) had this pathology. Independently of which scoring system was used, we found a high prevalence of high to very high categories of cardiovascular risk. Patients with mixed forms of lipodystrophy had higher cardiovascular risk, according to the Framingham risk score. Agreement between cardiovascular disease risk scores and between these and metabolic syndrome was not very good (kappa value < 0.50; $p<0.001$). However, when comparing lower and higher risk categories, a high proportion of the sample agreed in such conditions, which was being more evident when comparing the FHS to the ASCVD risk score. **Conclusions**: Patients had a high percentage of 10-year cardiovascular disease risk, independent of which scoring system was used, or the group of fat distribution. Patients with mixed forms of lipodystrophy, and also those with no lipodystrophy are at higher and lower risk of having cardiovascular disease, respectively.

Key-words: *HIV, antiretroviral therapy, lipodystrophy, cardiovascular risk, metabolic syndrome*

Background

HIV infection was a prevalent cause of premature death. At present, the emergence of cART increased life expectancy and reduced morbidity and mortality in these patients, turning HIV into a manageable chronic disease [1-2]. cART's side effects include, among other: metabolic changes, such as lipid disorders; insulin resistance; changes in glucose metabolism, and; hypertension. Changes in fat distribution and metabolic syndrome (MS) are clearly associated with HIV patients [3-4]. Cardiovascular disease (CVD) is also a major concern for the long-term use of cART. Additionally, the virus itself triggers many pathways that end up promoting metabolic disorders, immune processes and chronic inflammation. All this culminates in metabolic and organic diseases, including endothelial dysfunction, which accelerates the atherosclerotic process, and therefore increases the risk of cardiovascular disease [5-6]. As it is an active endocrine organ, the adipose tissue may also allow metabolic and inflammatory processes to take place, leading to the emergence of atherosclerosis as well [7]. On top of this, risk factors such as obesity, smoking, hypertension, dyslipidemia and diabetes could already have been established before HIV was diagnosed, or they may arise independently of the infection and the use of medication. These risk factors are highly prevalent in patients with HIV, making this population vulnerable to CVDs. [8-11]. Factors such as sedentarism, diet, and drug addiction may boost this problematic as well. Despite involving multifactorial mechanisms, the traditional risk factors are significant determinants of CVD risk in HIV-infected patients. The DAD study found a growing relative CHD risk with increased duration of cART, due in part to traditional risk factors. Nevertheless, the absolute CHD rates were still low [12]. CVD assessment is therefore of great importance for these patients.

Study aims

The primary goal of this study was to compare different CVD risk scores and predictors in HIV-infected patients under cART, according to different fat distribution groups.

Methods

Patient selection and study design

A cross-sectional study was performed in 572 HIV-infected Caucasian adults, 402 men, and 170 women, between 30 and 79 years old, who were, at the moment of the first visit, on cART. HIV-1 patients were included from the Endocrinology Outpatient Department of São João Hospital, who had been referred from the Infectious Diseases Clinic.

The study agreement was approved by the Ethics Committee for Health of Hospital São João and every patient provided written informed consent.

Clinical assessment

For each patient, the following data was collected, using a standardized protocol: age and gender; duration of HIV infection; cART in years; CD4+ cell count, and; viral load. Further date was collected for: alcohol; smoking (active, past, never); personal history of CVD; family history of CVD; clinical history of hypertension; diabetes; use of antidiabetics; insulin, and; anti-hypertensive drugs. Resting blood pressure, after 5 minutes seated was measured, with the elbow flexed at the heart, using a standard aneroid sphygmomanometer, with the cuff on the upper right arm. Two blood pressure readings were taken, and the mean of the two readings was calculated. Data was also collected for lipid assessment: total cholesterol (TC); low-density cholesterol (LDL); high-density cholesterol (HDL); triglycerides (TG), and; use of lipid-lowering drugs (statins, fibrates). In addition height (measured in standing position using a wall stadiometer - Holtain Limited Crymych, Dyfed®), weight (measured using TANITA - Tanita®, model TBF 300), body mass index (BMI) [calculated as weight divided by

height squared (kg/m2)], and waist and hip circumference were also evaluated. Clinical lipodystrophy (CL) and central fat accumulation or abdominal prominence (AP), were defined as has been previously described [13-14]. Central fat accumulation or abdominal prominence was determined by using the latest International Diabetes Federation (IDF) criteria [15] for the metabolic syndrome, based on a waist circumference of ≥94 cm for men, and 80 cm for women. Four different groups were characterized: Group 1 - no lipodystrophy (no clinical lipoatrophy nor abdominal prominence); group 2 - isolated central fat accumulation (no clinical lipoatrophy and with abdominal prominence); group 3 - isolated lipoatrophy (with clinical lipoatrophy and without abdominal prominence), and; group 4 - mixed forms of lipodystrophy (with clinical lipoatrophy and with abdominal prominence). All clinical assessments were performed by the same practitioner (PF).

Laboratory analysis

A venous blood sample was taken after a 12-hour overnight fast and all the samples were analyzed at the central laboratory of our hospital. Plasma glucose, total cholesterol, HDL cholesterol, and triglycerides were determined, using automatic standard routine enzymatic methods. The CD4+ cell count was determined by flow cytometry, and plasma RNA-VIH was measured by a quantitative reverse transcriptase polymerase chain reaction, which had a lower limit of detection of 50 copies/mL.

Diabetes definition

Diabetes was defined if two consecutive measurements of fasting plasma glucose were ≥126 mg/dL and if 2h plasma glucose was ≥200 mg/dL during the oral glucose tolerance test (OGTT).

Cardiovascular risk evaluation

A 10-year cardiovascular risk score was performed, using the following CVD risk scores, after excluding all of those with missing essential data fom risk score calculation (*Table 1):*

- The Framingham Heart Study risk prediction score for coronary heart disease (FHS-CHD) [16] (n=527). Personal history of CVD and diabetes was included for the very high risk category.

- The European SCORE [17] (n=353) - a web-based tool was used, using the low-risk chart, according to Portugal's guidelines [18]; patients were excluded if they were under 40, or over 65 years old, with CT > 305 mg/dL, or SBP > 180 mmHg. Personal history of CVD and diabetes with other CVD risk factors was included for the very high risk category (130/353).

- The 10-year ASCVD score – from the American College of Cardiology and the American Heart Association (ACC/AHA) [19] (n=281), which is a web-based tool [20]; patients were excluded if they were under 40 or above 79 years old, with a history of CVD, LDL $\geq$ 190 mg/dL, CT < 130 mg/dL, and CT > 320 mg/dL. 94 of the excluded patients had personal CVD history and/or an LDL $\geq$ 190 mg/dL.

Metabolic syndrome

We used the latest International Diabetes Federation (IDF) definition for the diagnosis of Metabolic Syndrome [15], which has been described above. At least three of the five criteria must be present to designate metabolic syndrome (central obesity, hypertension,

elevated fasting glucose, low high-density lipoprotein or hypertriglyceridemia) (*Table 1).*

Statistical analysis

Data were presented as mean and standard deviation (SD) for quantitative variables when normally distributed. When the variable distribution was different from the normal, this was expressed as the median and respective interquartile range. For the comparison between quantitative variables and the four groups of fat distribution an analysis of variance, or the Kruskall-Wallis test was used, as appropriate. Categorical variables were described as counts and proportions, and were compared using the chi-square or Fisher's exact test. Agreement between cardiovascular risk scores was assessed by observed agreement, using Cohen's kappa (κ) statistics. The level of agreement was considered poor if $\kappa = 0.20$, fair if $\kappa = 0.21$-0.40, moderate if $\kappa = 0.41$-0.60, substantial if $\kappa = 0.61$-0.80, and very good if $\kappa > 0.80$. Statistical analysis was performed using the SPSS version 21.0 software (SPSS Inc., Chicago, Illinois, USA). All probabilities were two tailed and p values <0.05 were regarded as significant.

Results

We evaluated 572 HIV-infected patients (402 men and 170 women) on cART. We observed that 31.3% had isolated central fat accumulation, 26.4% demonstrated isolated lipoatrophy, 26.1% mixed forms of lipodystrophy, and 16.3% of the patients had no lipodystrophy (Group 1). *Table 2* describes the sample, according to the four groups of fat distribution. The whole sample analyzed is smaller in some characteristics due to missing data.

Patient characteristics

The mean age of our sample varied from 43.8 to 52.7 years, with non-lipodystrophic patients being younger. Those with mixed forms of lipodystrophy were older. Our patients were predominantly male (70.3%) and the discrepancy between genders was higher among those with no lipodystrophy and isolated lipoatrophy. Almost half of patients with isolated central fat accumulation were females.

Patients with no lipodystrophy and isolated central fat accumulation had the lowest HIV infection, as well as cART duration, CD4+ cell count and viral load suppression. Those with isolated lipoatrophy and mixed forms of lipodystrophy had been HIV positive on cART for more than 8 years, with higher CD4+ cell count, and a viral load suppression of above 86%. No differences were found between groups on IP/NNRTI intake, whereas NRTI intake was highest in patients with isolated lipoatrophy.

Patients with no lipodystrophy had less than 16%, 10%, and 8% of lipid-lowering, anti-hypertensive, and oral antidiabetic drug use, respectively. Few patients were hypertensive, and they had lower TG, glucose and HbA1c medians, a higher HDL mean, normal BMI, and two times less the proportion of metabolic syndrome than those

with isolated central fat accumulation and mixed forms of lipodystrophy. On the other hand, patients with no lipodystrophy were the group that had the highest proportion of male individuals (>78%), and the highest level of active smokers (>50%), aside with Group 3 (isolated lipoatrophy). Conversely, patients with mixed forms of lipodystrophy had a higher burden of traditional CVD risk factors, except for active smokers (<30%). These were older, and used more lipid lowering, oral antidiabetic and antihypertensive drugs. Almost 50% of them were hypertensive. Anthropometric measures were the highest in this group and the isolated central fat accumulation group, with an overweight BMI mean. Glucose and HbA1c were also highest in this group, and metabolic syndrome was more than two times higher than that of non-lipodystrophic patients. When comparing the isolated central fat accumulation group with the isolated lipoatrophy group, active smoking was two times higher in the latter, and hypertension was more prevalent in the isolated central fat accumulation group. BMI, waist and hip circumference, as well as TC, LDL, and HDL were lower in those with isolated lipoatrophy, and higher in those with isolated central fat accumulation. However, TG median was highest in patients with isolated lipoatrophy. Metabolic syndrome prevalence of those with isolated central fat accumulation is similar to that of mixed forms of lipodystrophy group. Those with isolated lipoatrophy had much lower MS prevalence, with non-lipodystrophic patients having the lowest.

Seven to eight patients with no lipodystrophy and isolated lipoatrophy had a personal history of CVD, whereas 20 to 22 of those with isolated central fat accumulation and mixed forms of lipodystrophy had already experienced a CVD event.

With regard to CDC classification, C category was present in 42.9% of our sample, with A category in 55.2%. Around 40% of patients had alcohol intake and a familiar history

of CVD. Diabetes was present in 19.6% to 29.7% of our sample, being lower in non-lipodystrophic patients, and higher in mixed forms of the lipodystrophy group. However, no significant difference was found between groups with respect to these last mentioned variables.

Cardiovascular disease risk scores

Our sample has a high prevalence of high to very high risk categories, independent of which scoring system was used. FHS and ASCVD categorize patients as high/very high risk more frequently than the SCORE system does (>50% *vs.* approximately 40%, respectively) (*Table 3).*

This high prevalence of higher risk categories remains, despite which group we are observing. Nonetheless, a lower proportion of these categories exist in patients with no lipodystrophy, whereas the highest is found in patients with mixed forms of lipodystrophy. In every group, the SCORE equation classifies most individuals as being of low/moderate CVD risk. On the other hand, ASCVD is the system which classified most patients in the high to very high risk categories (except in mixed forms of lipodystrophy group, where the FHS scoring system classified most individuals in these higher categories – approximately 70%). However, statistically significant differences between groups were only found when using FHS ($p<0.001$) (*Table 2*). Patients with mixed forms of lipodystrophy are at higher CHD risk, according to FHS. On the other hand, patients with no lipodystrophy were mostly categorized as having low/moderate CHD.

Agreement of metabolic syndrome and CVD risk scores (*Table 4*)

Metabolic syndrome was compared to very high risk categories in FHS, SCORE and ASCVD. Agreement between MS and CVD risk scores was fair, with similar kappa values between each pair (0.342-0.385, $p<0.001$). 58.9%, 58.0%, and 72.4% individuals had metabolic syndrome and a "very high" risk category within FHS, SCORE or ASCVD, respectively.

Agreement of cardiovascular disease risk scores

For the whole sample, the overall observed agreement between scores was not very good (range: 0.095-0.475). The best kappa coefficient was observed in the comparison between ASCVD *vs*. FHS *(Table 5)*. Observing risk categories individually, the "very high risk" category was the one with the best agreement, regardless of the scoring system used. Most patients classified as being "moderate" FHS, are also "moderate" SCORE or ASCVD. However, 73.3% of "high risk" FHS were classified as being "low"/"moderate" SCORE, and 50% as "moderate" ASCVD. "Very high" risk category classified by FHS agreed in 71.6% of cases, in the high/very high SCORE categories and with high risk category in 95.6% using ASCVD. Finally, between SCORE and ASCVD, 53.9% of moderate SCORE are classified as being high/very high ASCVD. However, when considering the "very high" SCORE, 76.4% are also "very high" ASCVD.

Agreement between CVD risk scores in different fat distribution groups prevailed about the same as the whole sample. The agreement was poor ($k<0.20$) in FHS *vs.* SCORE and fair (0.21-0.40) in FHS *vs.* ASCVD and SCORE *vs.* ASCVD. The isolated central fat accumulation group in SCORE *vs.* ASCVD was the only one in which the agreement was substantial ($k=0.487$; $p<0.001$) (Data not shown).

Discussion

Our sample had a relatively high duration of HIV infection; all of whom were on cART, being predominantly male. A high prevalence of familiar history of CVD was present, and our mean age is considered as a CVD risk factor. Lipodystrophy was present in the majority of patients, and metabolic syndrome was diagnosed in almost half of our sample. We may already expect that there will be a high CVD risk.

Furthermore, a high burden of traditional risk factors was found: 24.25% were diabetic, 35.26% were hypertensive, and 28 to 57% were active smokers. TC and LDL means, and TG median were all above optimal. In addition, BMI mean was high in some of the described groups.

When comparing results to similar studies, samples with HIV and cART duration, as well as being predominantly male were considered. Only a few previous studies had such a high prevalence of metabolic syndrome, hypertension, or diabetes. This may be explained by the fact that this sample was obtained from an Endocrinology Department database, and therefore presented such results. However, 40.8% of an Italian study [21], and 36.8% of the DAD study [22] had lipodystrophy.

The high smoking rates present in our study, as well as in other studies are of great importance, as it is a major CVD risk factor. Our results were not consistent with the DAD study's finding, in that smoking rates decreased with cART duration [22]. Patients with isolated lipoatrophy had the highest cART duration and also, the highest prevalence of active smoking. Nevertheless, it is interesting to observe that patients with a higher proportion of active smoking are those that have fewer remaining traditional risk factors. These results may suggest that an effort is being made to alert patients with

higher burden of other CVD risk factors to stop smoking. In fact, a higher percentage of former smokers exist for patients with isolated central fat accumulation and mixed forms of lipodystrophy. On the other hand, approximately 50% of the individuals of the latter groups had never smoked at all.

Our study is one of the first to evaluate whether there is a higher CVD risk among different classes of lipodystrophy. Taking into consideration the higher burden of CVD risk factors in patients with isolated central fat accumulation and mixed forms of lipodystrophy, these are expected to have higher CVD risk. These two groups differ essentially for HIV and cART duration, being higher in the second group. Isolated central fat accumulation may already be present before HIV diagnosis, and may influence metabolic parameters by itself, without much HIV and cART influence. It may explain why, despite lower HIV and cART duration, there is a similar burden of risk factors in this last group of individuals when compared to patients with mixed forms of lipodystrophy. Patients with isolated lipoatrophy have higher HIV and cART duration, but fewer CVD risk factors. The type of fat distribution is probably associated with certain risk parameters.

As expected, a high percentage of our sample is subject to a high risk of CVD. High to very high risk categories were found in an elevated proportion of the sample, independently of which score system was used,

Most similar studies had a much lower incidence of higher risk categories, except for Begovac, J., et al. [23], which presented 58.3% of individuals having >10% risk, using FH-CVD. This last study also had a high prevalence of traditional risk factors. In a Swiss study [24] with more than 8.000 HIV-infected individuals, 2.757 were male, aged

over 40. For this subset of patients, CVD risk factors were highly prevalent and 38.5% had higher FH-CHD categories.

Differences were found between groups when using the FHS, but not with SCORE or ASCVD. Framingham high/very high risk category proportion was almost two times higher in patients with mixed forms of lipodystrophy *vs.* non-lipodystrophic patients (67.6% *vs.* 41.8%, respectively). These results were already expected, when analyzing each group's characteristics. However, groups with isolated central fat accumulation have approximately 20% less individuals than the former group, and the prevalence of higher categories is similar to that of the isolated lipoatrophy group (50.3 *vs* 49.3, respectively). The isolated lipoatrophy group has a higher mean age, a greater percentage of male individuals, higher HIV and cART duration, and CD4 cell count, a higher percentage of undetectable viral load, twice as many active smokers, higher TG median and fibrate use, and insulin intake, and lower HDL. This counteracts with a higher prevalence of personal CVD disease, hypertension, and metabolic syndrome in patients with isolated central fat accumulation. Statin, oral antidiabetic and anti-hypertensive use was also highest in this group, as well as all anthropometric measures, glucose median, and TC and LDL means. These two groups are at similar CHD risk, despite different contributing factors.

Agreement between CVD risk scores was not very good in any of the risk scoring systems that were compared (FHS/SCORE, FHS/ASCVD, ASCVD/SCORE). A critical analysis has to be made when looking at kappa coefficients, as this agreement indicator is based on all the categories taken together. When observing each category individually, there is a high percentage of very high risk category agreement, independent of the scoring system being compared. It is important to identify

individuals with high to very high CVD risk, and therefore a close analysis of these results is of great importance. When comparing lower and higher risk categories, we observed that a high proportion of the sample agreed in such conditions. This finding was more evident when comparing the FHS to the ASCVD risk score. However, many individuals categorized with higher FHS were also categorized with lower SCORE, and ASCVD categories. FHS may overestimate CHD risk. When comparing SCORE to ASCVD, more than half of patients with a moderate SCORE are attributed a high/very high risk category with ASCVD. These results for "underestimation" for the SCORE risk were already expected, as this predicts fatal outcomes better.

The DAD score, which is an equation designed specifically for HIV-infected patients [25], has been compared to other CVD risk scores. FHS-CHD also overestimates the risk when compared to this last mentioned scoring system [26-27]. On the other hand, the DAD study [22] found that FH may slightly underestimate MI risk on patients on cART. When ASCVD is compared to the DAD score, a better concordance is obtained than that of FHS or SCORE [23-26]. In fact, a recent long cohort study has suggested that ASCVD may be the better myocardial infarction risk predictor for the HIV population, even when compared to the DAD score (unpublished observation; Heidi M. Crane et al). However, differences of the outcomes predicted by each scoring system have to be taken into consideration every time we draw a conclusion on this matter. In our sample, no conclusion can be taken regarding which scoring system is the best CVD predictor, as we have no information about the outcome.

The majority of patients with MS are in the high/very high risk category, independent of the compared equation. However, concordance was not very high. We cannot know whether MS may, or may not be comparable to cardiovascular disease scoring systems,

however we can be more aware when MS is diagnosed, and a closer watch of these patients is needed. The high prevalence of higher CVD risk categories in our sample may be explained by the high prevalence of metabolic syndrome. De Socio et al. [21] reported that a clear correlation exists between estimated FHS and observed MS, and that lipodystrophy and a CD4 cell count >500 cells/m^3 are associated with the coexistence of a high/very high FHS and the presence of this syndrome.

It is hard to compare studies, owing to the different methodological designs, different age means, or other population characteristics that may importantly influence the results.

The aim of this study was never to determine conclusions that are representative sample of HIV population, which it is not, but rather to compare results between different fat distribution groups. Cardiovascular risk factors have cumulative effects throughout a lifetime, and we measured risk factors at one point in time.

This study was performed in a department highly experienced in the assessment of metabolic and body fat abnormalities in HIV-infected patients. Clinical assessment was always executed by the same practitioner (PF). To our knowledge, this is one of the first studies to evaluate whether any differences exist in cardiovascular risk factors and 10-year risk, with regards to four different groups of fat distribution. We point out that patients with mixed forms of lipodystrophy are at higher 10-year CVD risk, and that those with isolated central fat accumulation have important modifiable CVD risk factors, which require a close watch with regards to CVD risk. Our study also suggests that independent of the scoring system used, at least the majority of the high to very high risk categorized individuals are identified.

Conclusions

There is a high percentage of 10-year cardiovascular disease risk in this sample, independent of the scoring system, or of fat body distribution. HIV-infected patients with mixed forms of lipodystrophy deserve special attention, as they are at higher CVD risk. Measures need to be applied to reduce CVD risk factors, as most of them are modifiable or treatable, and apparently they are the major influence for CVD risk in these individuals.

Declarations

List of abbreviations

HIV – Human Immunodeficiency Virus; cART – combined antiretroviral therapy; CHD – Coronary Heart Disease; SCORE – Systematic Coronary Risk Evaluation; ASCVD – Arteriosclerotic cardiovascular disease; FHS – Framingham Score; MS – Metabolic Syndrome; CVD – Cardiovascular Disease; DAD – Data Collection on Adverse events of anti-HIV Drugs; TC – Total cholesterol; LDL –Low-density lipoprotein; HDL – High-density lipoprotein; TG – Triglycerides; BMI – Body Mass Index; CL – Clinical lipodystrophy; AP – Abdominal prominence; IDF – International Diabetes Federation; OGTT – Oral glucose tolerance test; FHS-CHD – Framingham coronary heart disease risk score; SBP – Systolic Blood Pressure; ACC/AHA – American College of Cardiology/American Heart Association; SD – standard deviation; k – Cohen's kappa statistics; IP – Protease Inhibitors; NNRTI – Non-Nucleoside Reverse Transcriptase Inhibitors; NRTI – Nucleoside Reverse Transcriptase Inhibitors; HbA1c – Glycated hemoglobin; CDC – Centers for Disease Control and Prevention; DBP – Diastolic Blood Pressure

Ethics approval and consent to participate

The Health Ethics Committee (CES) from Hospital São João, Oporto, approved the draft report, and it raised no objection to the project's execution.

Competing interests

Nothing to declare.

Authors' contributions

CGS and PF conceived the study, participated in its design, in the acquisition of data, and they drafted the manuscript. DC conceived the study, participated in its design and drafted the manuscript; ARG, FA, and AS participated in the acquisition of data; ACS performed the statistical analysis. ARG, ACS, and PF critically reviewed the manuscript. All authors read and approved the final manuscript.

Acknowledgements

We thank Mark Crathorne for the English proof reading.

Authors' information

[1] Student at University of Porto Medical School, Alameda Hernâni Monteiro, 4200 Porto, Portugal.

[2] Student at University of Porto Medical School, Alameda Hernâni Monteiro, 4200 Porto, Portugal.

[3] MD. Department of Infectious Disease, São João Hospital, University of Porto Medical School, Porto, Portugal.

[4] MD, PhD. Department of Infectious Disease, São João Hospital, University of Porto Medical School, Porto, Portugal.

[5] MPH, PhD. Clinical Epidemiology, Predictive Medicine and Public Health, University of Porto Medical School, and EPIUnit – Epidemiology Research Unit, University of Porto Institute of Public Health, Porto, Portugal.

[6] MD, PhD. Endocrinology Department, São João Hospital and University of Porto Medical School, Alameda Hernâni Monteiro, 4200 Porto, Portugal.

[7] MD, PhD. Endocrinology Department, São João Hospital and University of Porto Medical School, Alameda Hernâni Monteiro, 4200 Porto, Portugal.

Table 1: Variables and outcomes of different CVD risk scores and MS

	FHS-CHD	SCORE	ASCVD	MS (at least 3 of the 5 criteria)
Age	30-74	40-65	40-79	
Gender	√	√	√	
Ethnics			√	
Total cholesterol (TC)	√	√	√	
High-density lipoprotein cholesterol (HDL)	√		√	
Low-density lipoprotein cholesterol (LDL)				√
Hypertriglyceridemia				√
Systolic blood pressure	√	√	√	
Dyastolic blood pressure	√	√	√	
Hypertension				√
Hypertension treatment	√		√	
Smoking	√	√	√	
Diabetes	√	~ high/very high risk	√	
Glucose				√
BMI		√		
Central obesity				√

Low risk	0-1 CVD risk factors	<1%	<5%	
Intermediate risk	<10%	≥1 but <5%	≥5 but <7,5%	
High risk	10-20%	≥5% but <10%	≥7.5% but <10%	
Very high risk	>20%	≥10%	≥10%	
Coronary heart disease (angina pectoris, myocardial infarction, death due to CHD)	√			
CV fatal events (aortic aneurysm, myocardial infarction, stroke)		√		
Atherosclerotic CVD (nonfatal myocardial infarction, fatal coronary heart disease and nonfatal or fatal stroke)			√	

Table 2: Sample characteristics according to the presence or absence of clinical lipodystrophy and/or abdominal prominence

	No lipodystrophy	Isolated central fat accumulation	Isolated lipoatrophy	Mixed forms of lipodystrophy	p
n (%)	93 (16.3)	179 (31.3)	151 (26.4)	149 (26.0)	
Age [years, mean (sd)]	43.8 (11.7)	47.8 (12.1)	44.7 (9.6)	52.7 (10.9)	<0.001
Gender [n,(%)]					<0.001
Male	73 (78.5)	99 (55.3)	135 (89.4)	95 (63.8)	
Female	20 (21.5)	80 (44.7)	16 (10.6)	54 (36.2)	
Duration of HIV infection [years, median (IQR)]	7.0 (6.0)	6.0 (7.0)	10.0 (7.0)	9.0 (6.0)	<0.001
cART [years, median (IQR)]	5.0 (8.0)	4.0 (7.0)	9.0 (6.0)	8.0 (6.0)	<0.001
CD4 cell count [cells/mm³, median (IQR)]	401.0 (325.0)	460.0 (332.0)	509.0 (357.0)	557.0 (404.0)	0.002
HIV RNA [n (%)]					0.003
≥ 50 copies/mL (n=75)	15 (23.4)	32 (27.4)	14 (11.1)	14 (13.3)	
	49 (76.6)	85 (72.6)	112 (88.9)	91 (86.7)	

<50 copies/mL					
CDC [n (%)] A B C 24 (39.3)	 32 (52.5) 5 (8.2)	 58 (56.9) 1 (1.0) 43 (42.2)	 67 (55.8) 0 (0.0) 53 (44.2)	 54 (54.5) 1 (1.0) 44 (44.4)	0.061
ART [n (%)] IP NNRTI NRTI	 49 (58.3) 35 (41.5) 78 (91.8)	 87 (54.0) 64 (40.5) 147 (91.3)	 74 (51.0) 73 (50.7) 144 (99.3)	 68 (48.2) 70 (50.0) 134 (94.4)	 0.484 0.180 0.004
Alcohol (n,%)	37 (41.1)	67 (40.6)	69 (47.9)	60 (40.8)	0.531
Smoking history [n (%)] Never Active Former	 29 (32.6) 47 (52.8) 13 (14.6)	 86 (52.1) 47 (28.5) 32 (19.4)	 42 (29.3) 81 (56.3) 21 (14.6)	 67 (46.2) 41 (28.3) 37 (25.5)	<0.001
Personal history of CVD [n (%)]	7 (7.6)	20 (11.6)	8 (5.4)	22 (14.8)	0.043
Familiar history of CVD [n (%)]	35 (40.2)	68 (41.7)	63 (43.4)	68 (46.9)	0.737

Statins [n (%)]	13 (14.1)	50 (27.9)	33 (22.0)	51 (34.2)	0.003
Fibrates [n (%)]	14 (15.2)	34 (19.0)	45 (30.0)	48 (32.2)	0.002
Oral antidiabetic [n (%)]	7 (7.7)	29 (16.2)	16 (10.7)	32 (21.5)	0.010
Insulin [n (%)]	8 (8.7)	4 (2.2)	13 (8.7)	7 (4.7)	0.039
SBP [mmHg, median (IQR)]	120.0 (24.0)	126.0 (30.0)	115.5 (29.0)	130.0 (20.0)	<0.001
DBP [mmHg, median (IQR)]	70.0 (17.0)	80.0 (14.0)	71.5 (20.0)	80.0 (15.0)	<0.001
Hypertension [n (%)]	22 (23.7)	65 (36.3)	42 (28.2)	72 (48.3)	<0.001
Anti-hypertensive therapy [n (%)]	9 (9.8)	37 (20.7)	21 (14.0)	46 (31.1)	<0.001
Weight [kg, mean (sd)]	64.0 (10.1)	80.9 (16.1)	61.6 (9.9)	73.9 (12.9)	<0.001
BMI [(kg/m2), mean (sd)]	22.9 (2.7)	29.7 (4.7)	21.8 (2.9)	27.2 (3.7)	<0.001
Waist circumference [cm, mean, (sd)]	83.4 (7.9)	102.8 (11.3)	83.0 (7.1)	98.8 (8.3)	<0.001
Hip circumference [cm, mean, (sd)]	92.6 (4.8)	104.0 (9.0)	88.8 (5.4)	96.3 (7.5)	<0.001
Waist/hip circumference ratio	0.91 (0.07)	0.99 (0.08)	0.94 (0.06)	1.03 (0.07)	<0.001

Total cholesterol (TC) [mg/dL, mean (sd)]	213.5 (57.9)	225.4 (56.1)	208.3 (54.4)	222.4 (59.2)	0.032
LDL-cholesterol (LDL) [mg/dL, mean (sd)]	131.7 (47.5)	137.3 (44.6)	120.9 (42.6)	135.3 (49.4)	0.009
HDL-cholesterol (HDL) [mg/dL, mean (sd)]	48.4 (13.8)	48.8 (14.0)	44.3 (13.7)	46.4 (12.6)	0.016
Triglycerides (TG) [mg/dL, median (IQR)]	157.0 (133.0)	170.0 (136.0)	204.0 (225.0)	198.0 (165.0)	0.009
Glucose 0' [mg/dL, median (IQR)]	90.0 (21.0)	97.0 (28.0)	94.0 (27.0)	100.0 (37.0)	<0.001
HbA1C [%, median (IQR)]	5.15 (1.0)	5.40 (1.0)	5.30 (1.0)	5.60 (1.0)	<0.001
Diabetes [n (%)]	18 (19.6)	42 (23.5)	34 (22.7)	44 (29.7)	0.286
Metabolic syndrome [n (%)]	20 (29.0)	63 (65.6)	46 (35.7)	60 (66.7)	<0.001
Framingham [n (%)] Low risk (0-1					<0.001

CVD risk factors)	8 (9.3)	8 (5.0)	1 (0.7)	2 (1.4)	
Moderate risk (≥2 CVD risk factors and a 10-year CVD risk <10%)	42 (48.8)	71 (44.7)	70 (50.0)	44 (31.0)	
High risk (≥2 CVD risk factors and a 10-year CVD risk 10-20%)	18 (20.9)	34 (21.4)	33 (23.6)	30 (21.1)	
Very high risk (CHD or CHD equivalent or ≥2 CVD risk factors and a 10-year CVD risk >20%)	18 (20.9)	46 (28.9)	36 (25.7)	66 (46.5)	
SCORE [n (%)]					0.194
Low risk (10-year CVD risk <1%)	9 (17.6)	31 (29.5)	22 (24.7)	19 (17.6)	
Moderate risk (10-year CVD risk ≥1 but <5%)	24 (47.1)	27 (25.7)	34 (38.2)	41 (38.0)	
High risk (diabetes					

without any other CVD risk factor or 10-year CVD risk ≥5 but <10%)	3 (5.9)	6 (5.7)	2 (2.2)	4 (3.7)	
Very high risk (previous CVD or diabetes with other CVD risk factors or 10-year CVD risk ≥10)	15 (29.4)	41 (39.0)	31 (34.8)	44 (40.7)	
ASCVD score [n (%)]					0.244
Low risk (10-year CVD risk <5%)	9 (25.7)	27 (35.5)	19 (24.7)	18 (19.4)	
Moderate risk (10-year CVD risk ≥5 but <7.5%)	9 (25.7)	10 (13.2)	20 (26.0)	16 (17.2)	
High risk (10-year CVD risk ≥7.5% but <10%)	4 (11.4)	6 (7.9)	6 (7.8)	8 (8.6)	
Very High risk (10-year CVD risk	13 (37.1)	33 (43.4)	32 (41.6)	51 (54.8)	

≥ 10%)					

CL – clinical lipodystrophy; AP – abdominal prominence; IP – Protease Inhibitors; NNRTI – Non-Nucleoside Reverse Transcriptase Inhibitors; NRTI – Nucleoside Reverse Transcriptase Inhibitors; SBD – systolic blood pressure; DBP – diastolic blood pressure; BMI – body mass index; TC – total cholesterol; LDL – low-density lipoproteinHbA1c – glycated hemoglobin; CVD – cardiovascular disease; ASCVD – atherosclerotic cardiovascular disease

Table 3: Proportions of CVD risk classifications in different CVD risk scores for the whole sample.

	FHS	SCORE	ASCVD
Low (n,%)	3.6 (19)	22.9 (81)	26.0 (73)
Intermediate (n,%)	43.1 (227)	35.7 (126)	19.6 (55)
High (n,%)	21.8 (115)	4.2 (15)	8.5 (24)
Very high (n,%)	31.5 (166)	37.1 (131)	45.9 (129)
Total (n,%)	527 (100)	353 (100)	281 (100)

Table 4: Agreement of low/moderate/high and very high risk categories within MS.

	With MS	Without MS	Kappa coefficient	p
FHS			0.385	<0.001
Low/moderate/high	76 (41.1)	148 (79.6)		
Very high	109 (58.9)	37 (20.4)		
SCORE			0.343	<0.001
Low/moderate/high	60 (42.0)	86 (77.5)		
Very high	83 (58.0)	25 (22.5)		
ASCVD			0.342	<0.001
Low/moderate/high	29 (27.6)	63 (61.8)		
Very high	76 (72.4)	39 (38.2)		

FHS – Framingham Score; MS – Metabolic syndrome

Table 5: Agreement proportions (n, %) for the whole sample

					Kappa coefficient
			FHS		
SCORE	Low risk	Moderate risk	High risk	Very high risk	
Low risk	1 (50.0)	67 (61.5)	12 (11.9)	0 (0.0)	
Moderate risk	1 (50.0)	26 (23.9)	62 (61.4)	37 (28.5)	
High risk	0 (0.0)	4 (3.7)	3 (3.0)	8 (6.2)	
Very high risk	0 (0.0)	12 (11.0)	24 (23.8)	85 (65.4)	0.095
			FHS		
ASCVD		Moderate risk	High risk	Very high risk	
Moderate risk		15 (93.8)	39 (50.0)	1 (0.9)	
High risk		1 (6.3)	19 (24.4)	4 (3.5)	
Very high risk		0 (0.0)	20 (25.6)	108 (95.6)	0.475
			SCORE		
ASCVD	Low risk	Moderate risk	High risk	Very high risk	
Low risk	53 (84.1)	14 (12.5)	0 (0.0)	6 (8.3)	
Moderate risk	8 (12.7)	38 (33.9)	0 (0.0)	7 (9.7)	
High risk	2 (3.2)	18 (16.1)	0 (0.0)	4 (5.6)	
Very high risk	0 (0.0)	42 (37.5)	7 (100.0)	55 (76.4)	0.408

References

[1] Bhaskaran, K., et al., *Changes in the risk of death after HIV seroconversion compared with mortality in the general population.* Jama, 2008. **300**(1): p. 51-9

[2] *Life expectancy of individuals on combination antiretroviral therapy in high-income countries: a collaborative analysis of 14 cohort studies.* Lancet, 2008. **372**(9635): p. 293-9.

[3] Grinspoon, S. and Carr, A. *Cardiovascular risk and body-fat abnormalities in HIV-infected adults.* N Engl J Med, 2005. **352**(1): p. 48-62.

[4] Worm, S.W., et al., *High prevalence of the metabolic syndrome in HIV-infected patients: impact of different definitions of the metabolic syndrome.* Aids, 2010. **24**(3): p. 427-35.

[5] Grunfeld, C., et al., *Preclinical atherosclerosis due to HIV infection: carotid intima-medial thickness measurements from the FRAM study.* Aids, 2009. **23**(14): p. 1841-9.

[6] Francisci, D., et al., *HIV type 1 infection, and not short-term HAART, induces endothelial dysfunction.* Ibid.(5): p. 589-96.

[7] Freitas, P., et al., *Carotid intima media thickness is associated with body fat abnormalities in HIV-infected patients.* BMC Infect Dis, 2014. **14**: p. 348.

[8] Friis-Moller, N., et al., *Cardiovascular disease risk factors in HIV patients--association with antiretroviral therapy. Results from the DAD study.* Aids, 2003. **17**(8): p. 1179-93.

[9] Kaplan, R. C., et al., *Ten-year predicted coronary heart disease risk in HIV-infected men and women.* Clin Infect Dis, 2007. **45**(8): p. 1074-81.

[10] Brown, T. T., et al., *Antiretroviral therapy and the prevalence and incidence of diabetes mellitus in the multicenter AIDS cohort study.* Arch Intern Med, 2005. **165**(10): p. 1179-84.

[11] Mdodo, R., et al., *Cigarette smoking prevalence among adults with HIV compared with the general adult population in the United States: cross-sectional surveys.* Ann Intern Med, 2015. **162**(5): p. 335-44.

[12] Friis-Moller, N., et al., *Combination antiretroviral therapy and the risk of myocardial infarction.* N Engl J Med, 2003. **349**(21): p. 1993-2003.

[13] Freitas, P., et al., *Fat mass ratio: an objective tool to define lipodystrophy in hiv-infected patients under antiretroviral therapy.* J Clin Densitom, 2010. **13**(2): p. 197-203.

[14] Freitas, P., et al., *Assessment of body fat composition disturbances by bioimpedance analysis in HIV-infected adults.* J Endocrinol Invest, 2011. **34**(10): p. e321-9.

[15] Alberti, K. G., et al., *Harmonizing the metabolic syndrome: a joint interim statement of the International Diabetes Federation Task Force on Epidemiology and Prevention; National Heart, Lung, and Blood Institute; American Heart Association; World Heart Federation; International Atherosclerosis Society; and International Association for the Study of Obesity.* Circulation, 2009. **120**(16): p. 1640-5.

[16] Wilson P.W., D'Agostino R.B., Levy D., Belanger A.M., Silbershatz H., Kannel W.B. *Prediction of coronary heart disease using risk factor categories*. Circulation 1998; 97: 1837

[17] Conroy, R. M., et al., *Estimation of ten-year risk of fatal cardiovascular disease in Europe: the SCORE project.* Eur Heart J, 2003. **24**(11): p. 987-1003.

[18] https://escol.escardio.org/heartscore/calc.aspx?model=europelow ; Accessed 2 March 2016

[19] Goff, D. C., Jr., et al., *2013 ACC/AHA guideline on the assessment of cardiovascular risk: a report of the American College of Cardiology/American Heart Association Task Force on Practice Guidelines.* J Am Coll Cardiol, 2014. **63**(25 Pt B): p. 2935-59.

[20] http://tools.acc.org/ASCVD-Risk-Estimator/ ; Accessed 2 March 2016

[21] De Socio, G.V., et al., *Identifying HIV patients with an unfavorable cardiovascular risk profile in the clinical practice: results from the SIMONE study*. J Infect, 2008. **57**(1): p. 33-40.

[22] Law, M. G., et al., *The use of the Framingham equation to predict myocardial infarctions in HIV-infected patients: comparison with observed events in the D:A:D Study*. HIV Med, 2006. **7**(4): p. 218-30.

[23] Begovac, J., et al., *Comparison of four international cardiovascular disease prediction models and the prevalence of eligibility for lipid lowering therapy in HIV infected patients on antiretroviral therapy.* Croat Med J, 2015. **56**(1): p. 14-23.

[24] Glass, T. R., et al., *Prevalence of risk factors for cardiovascular disease in HIV-infected patients over time: the Swiss HIV Cohort Study*. HIV Med, 2006. **7**(6): p. 404-10.

[25] Friis-Moller, N., et al., *Predicting the risk of cardiovascular disease in HIV-infected patients: the data collection on adverse effects of anti-HIV drugs study.* Eur J Cardiovasc Prev Rehabil, 2010. **17**(5): p. 491-501.

[26] Krikke, M., et al., *Cardiovascular risk prediction in HIV-infected patients: comparing the Framingham, atherosclerotic cardiovascular disease risk score (ASCVD), Systematic Coronary Risk Evaluation for the Netherlands (SCORE-NL) and Data Collection on Adverse Events of Anti-HIV Drugs (D:A:D) risk prediction models.* HIV Med, 2015.

[27] Edwards-Jackson, N., et al., *Cardiovascular risk assessment in persons with HIV infection in the developing world: comparing three risk equations in a cohort of HIV-infected Thais.* HIV Med, 2011. **12**(8): p. 510-5.

Agradecimentos

A todos os que tornaram possível a realização deste trabalho:

Um especial agradecimento à minha Orientadora, Professora Doutora Paula Freitas, pelo apoio, disponibilidade, orientação e carinho prestado ao longo destes últimos meses.

À Professora Ana Cristina Santos, pelo apoio e conhecimento partilhados. Pela paciência e dedicação ao trabalho.

À minha colega Rita Gomes, pela sua incessante prestabilidade, não só na leitura crítica do artigo como também em todas as questões burocráticas necessárias para a sua entrega atempada.

Aos meus Pais e Avós:

Pelo apoio emocional incondicional e pela presença constante na minha vida ao longo destes anos. Pelo apoio financeiro que fez com este sonho se tornasse realidade. Por terem ajudado na construção da pessoa que sou hoje. Obrigada pelo carinho, conselhos e orientação. Obrigada por terem tornado possível este meu precurso pela Faculdade de Medicina da Universidade do Porto.

Ao André:

Pelo carinho, amor, companheirismo e amizade. Por ter facilitado estes anos longe de casa e ter feito com que "casa" passasse a ter um outro significado. Obrigada pela presença constante na minha vida. Foste e és o meu grande apoio nesta "cidade nova" que entretanto se tornou tão familiar.

À Mónica e à Laura:

Não foram só colegas de casa. Foram amigas que fiz para a vida. Obrigada por terem tornado melhor este meu precurso pela Faculdade. Pelas muitas conversas, desabafos, choros e risos partilhados. Obrigada por me terem ensinado a viver convosco. Foi um prazer ter-vos comigo.

Aos meus amigos mais próximos:

Aos de sempre, obrigada por terem mantido o contacto, por não deixarem apagar a amizade que nos une, apesar da distância.

A todos os que conheci devido à minha passagem pela FMUP, obrigada pelo companheirismo, diversão e amizade. Juntos tornaram esta minha "estadia", uma grande experiência de vida.

ANEXOS

1. Parecer da Comissão de Ética
2. Normas da revista BMC Infectious Diseases

Comissão de Ética para a Saúde do HSJ

Parecer

Projecto de investigação: "Lipodystrophy and cardiovascular risk in HIV-infected patients under retroviral therapy – comparing SCORE, Framingham, ASCVD and metabolic syndrome".

Promotores:
- Faculdade de Medicina da Universidade do Porto.

- Pertinência do estudo

- Trata-se de um estudo observacional transversal, a realizar no âmbito da tese de Mestrado Integrado em Medicina na Faculdade de Medicina da Universidade do Porto (FMUP), que tem como objectivos principais: avaliar a presença de síndrome metabólico e o risco cardiovascular pelos score de Framingham, SCORE e ASCVD em doentes VIH sob terapêutica antiretrovírica combinada (TARc) no total da amostra e de acordo com 4 categorias de composição corporal (1- sem lipoatrofia clínica, sem proeminência abdominal; 2- sem lipoatrofia clínica, com proeminência abdominal; 3 - com lipoatrofia clínica, sem proeminência abdominal; 4 - com lipoatrofia clínica, com proeminência abdominal); avaliar a concordância entre os scores e síndrome metabólico na categorização do risco cardiovascular
- Serão incluídos todos os indivíduos com infecção VIH seguidos na consulta de Endocrinologia do Centro Hospitalar de S. João, entre janeiro de 2004 e janeiro de 2016.
- O estudo terminaria em março de 2015 e não terá nem precisará de qualquer apoio financeiro. *Dada a submissão deste protocolo à CES em Janeiro de 2016, o prazo indicado terá que ser alargado.*
- Todos os dados a colher do processo clínico e de forma anónima [sócio-demográficos, antropométricos, clínicos, factores de risco cardiovasculares clássicos (hipertensão arterial, diabetes *mellitus*, tabagismo e dislipidemia), virológicos, analíticos e terapêuticos] são pertinentes e adequados aos objectivos do estudo.
    - O estudo é pertinente, importante e está bem fundamentado.
    - O protocolo de estudo, os critérios de inclusão e de exclusão estão suficientemente detalhados e não levantam quaisquer questões do foro ético.
    - A Investigadora Principal, Carolina Jardim Fernandes Rodrigues Silva, estudante do 6º ano do curso de Medicina da FMUP, tendo como elo de ligação (e orientadora da Tese) a Médica especialista de Endocrinologia, a Professora Paula Freitas (especialista do Serviço de Endocrinologia do Hospital de S. João EPE), dispõe das competências técnica e científica para a realização do estudo.
    - O estudo será realizado no Serviço de Endocrinologia do Hospital de S. João, EPE e dispõe da autorização para a sua realização pelo seu Director,

Professor Davide Carvalho. O serviço proponente dispõe das condições necessárias para a realização do estudo.

– **Benefício/Risco**

- Dada a natureza retrospectiva do estudo, não haverá riscos, incómodos ou benefícios para os participantes.

– **Respeito pela liberdade e autonomia do sujeito do ensaio**

- Dada a natureza retrospectiva do estudo, não há necessidade de proceder à obtenção do consentimento informado.

– **Confidencialidade dos dados**

- A confidencialidade e a privacidade dos dados são garantidas.

– **Indemnização por danos**

Não aplicável.

– **Continuação do tratamento**

Não aplicável.

- **Propriedade dos dados**

Não aplicável.

Conclusão

Em face da análise do protocolo de "Lipodystrophy and cardiovascular risk in HIV-infected patients under retroviral therapy – comparing SCORE, Framingham, ASCVD and metabolic syndrome", proponho a sua aprovação pela CES do HSJ/FMUP, depois de obtida a resposta à questão sublinhada e em itálico.

Porto, 29 de janeiro de 2016

O Relator
Prof. Manuel Vaz Silva

Submission Guidelines BMC Infectious Diseases

Research article

Criteria

Research articles should report on original primary research, but may report on systematic reviews of published research provided they adhere to the appropriate reporting guidelines which are detailed in our editorial policies. Please note that non-commissioned pooled analyses of selected published research will not be considered.

Preparing your manuscript

Title page

The title page should:

- present a title that includes, if appropriate, the study design e.g.:
- "A versus B in the treatment of C: a randomized controlled trial", "X is a risk factor for Y: a case control study", "What is the impact of factor X on subject Y: A systematic review"
- or for non-clinical or non-research studies a description of what the article reports
- list the full names, institutional addresses and email addresses for all authors
- if a collaboration group should be listed as an author, please list the Group name as an author. If you would like the names of the individual members of the Group to be searchable through their individual PubMed records, please include this information in the “Acknowledgements” section in accordance with the instructions below
- indicate the corresponding author

Preparing main manuscript text

Manuscripts must be written in concise English. For help on scientific writing, or preparing your manuscript in English, please see BioMed Central‟s Author Academy.
Quick points:

- Use double line spacing
- Include line and page numbering
- Use SI units: Please ensure that all special characters used are embedded in the text, otherwise they will be lost during conversion to PDF
- Do not use page breaks in your manuscript

Abstract

The Abstract should not exceed 350 words. Please minimize the use of abbreviations and do not cite references in the abstract. Reports of randomized controlled trials should follow the CONSORT extension for abstracts. The abstract must include the following separate sections:

- **Background:** the context and purpose of the study
- **Methods:** how the study was performed and statistical tests used
- **Results:** the main findings
- **Conclusions:** brief summary and potential implications
- **Trial registration:** If your article is a systematic review or reports the results of a health care intervention on human participants, it must be registered in an appropriate registry and the registration number and date of registration should be in stated in this section. See our editorial policies for more information on trial registration

Keywords

Three to ten keywords representing the main content of the article.

Background

The Background section should explain the background to the study, its aims, a summary of the existing literature and why this study was necessary or its contribution to the field.

Methods

The methods section should include:

- the aim, design and setting of the study
- the characteristics of participants or description of materials
- a clear description of all processes, interventions and comparisons. Generic drug names should generally be used. When proprietary brands are used in research, include the brand names in parentheses
- the type of statistical analysis used, including a power calculation if appropriate

Results

This should include the findings of the study including, if appropriate, results of statistical analysis which must be included either in the text or as tables and figures.

Discussion

This section should discuss the implications of the findings in context of existing research and highlight limitations of the study.

Conclusions

This should state clearly the main conclusions and provide an explanation of the importance and relevance of the study reported.

Declarations

List of abbreviations

If abbreviations are used in the text they should be defined in the text at first use, and a list of abbreviations should be provided.

Ethics approval and consent to participate

Manuscripts reporting studies involving human participants, human data or human tissue must:

- include a statement on ethics approval and consent (even where the need for approval was waived)
- include the name of the ethics committee that approved the study and the committee‟s
- reference number if appropriate

Studies involving animals must include a statement on ethics approval. See our editorial policies for more information.
If your manuscript does not report on or involve the use of any animal or human data or tissue,
this section is not applicable to your submission. Please state “Not applicable” in this section.

Consent for publication

If your manuscript contains any individual person‟s data in any form (including individual details, images or videos), consent to publish must be obtained from that person, or in the case of children, their parent or legal guardian. All presentations of case reports must have consent to publish. You can use your institutional consent form or our consent form if you prefer. You should not send the form to us on submission, but we may request to see a copy at any stage (including after publication).

See our editorial policies for more information on consent for publication.

If your manuscript does not contain any individual persons data, please state “Not applicable” in this section.

Availability of data and materials

For all journals, BioMed Central strongly encourages all datasets on which the conclusions of the manuscript rely to be either deposited in publicly available repositories (where available and appropriate) or presented in the main paper or additional supporting files, in machine-readable format (such as spreadsheets rather than PDFs) whenever possible. Please see the list of recommended repositories in our editorial policies.

For some journals, deposition of the data on which the conclusions of the manuscript rely is an absolute requirement. Please check the Criteria section for this article type (located at the top of this page) for journal specific policies.

For all journals, authors must include an "Availability of data and materials" section in their article detailing where the data supporting their findings can be found. If you do not wish to share your data, please state that data will not be shared, and state the reason.

For information on how to cite your data and format this section see preparing your manuscript.

Competing interests

All financial and non-financial competing interests must be declared in this section. See our editorial policies for a full explanation of competing interests. If you are unsure whether you or any of your co-authors have a competing interest please contact the editorial office.

Funding

All sources of funding for the research reported should be declared. The role of the funding body in the design of the study and collection, analysis, and interpretation of data and in writing the manuscript should be declared.

Authors' contributions

The individual contributions of authors to the manuscript should be specified in this section. Guidance and criteria for authorship can be found in our editorial policies.

Acknowledgements

Please acknowledge anyone who contributed towards the article who does not meet the criteria for authorship including anyone who provided professional writing services or materials.

Authors should obtain permission to acknowledge from all those mentioned in the Acknowledgements section.See our editorial policies for a full explanation of acknowledgements and authorship criteria. Group authorship: if you would like the names of the individual members of a collaboration Group to be searchable through their individual PubMed records, please ensure that the title of the collaboration Group is included on the title page and in the submission system and also include collaborating author names as the last paragraph of the "Acknowledgements" section. Please add authors in the format First Name, Middle initial(s) (optional), Last Name. You can add institution or country information for each author if you wish, but this should be consistent across all authors. Please note that individual names may not be present in the PubMed

record at the time a published article is initially included in PubMed as it takes PubMed additional time to code this information.

Authors' information

You may choose to use this section to include any relevant information about the author(s) that may aid the reader's interpretation of the article, and understand the standpoint of the author(s). This may include details about the authors' qualifications, current positions they hold at institutions or societies, or any other relevant background information. Please refer to authors using their initials. Note this section should not be used to describe any competing interests.

Endnotes

Endnotes should be designated within the text using a superscript lowercase letter and all notes (along with their corresponding letter) should be included in the Endnotes section. Please format this section in a paragraph rather than a list.

References

All references, including URLs, must be numbered consecutively, in square brackets, in the order in which they are cited in the text, followed by any in tables or legends. The reference numbers must be finalized and the reference list fully formatted before submission. For further information including example references please read our reference preparation guidelines.

What should be cited?

Only articles, clinical trial registration records and abstracts that have been published or are in press, or are available through public e-print/preprint servers, may be cited.

Unpublished abstracts, unpublished data and personal communications should not be included in the reference list, but may be included in the text and referred to as "unpublished observations" or "personal communications" giving the names of the involved researchers. Obtaining permission to quote personal communications and unpublished data from the cited colleagues is the responsibility of the author. Footnotes are not allowed, but endnotes are permitted. Journal abbreviations follow Index Medicus/MEDLINE.

Any in press articles cited within the references and necessary for the reviewers' assessment of the manuscript should be made available if requested by the editorial office.

How to format your references

Examples of the BioMed Central reference style are shown below. Please ensure that the reference style is followed precisely; if the references are not in the correct style, they may need to be retyped and carefully proofread.

Web links and URLs: All web links and URLs, including links to the authors' own websites, should be given a reference number and included in the reference list rather than within the text of the manuscript. They should be provided in full, including both the title of the site and the URL, as well as the date the site was accessed, in the following format: The Mouse Tumor Biology Database. http://tumor.informatics.jax.org/mtbwi/index.do. Accessed 20 May 2013. If an author or group of authors can clearly be associated with a web link, such as for weblogs, then they should be included in the reference.

Authors may wish to make use of reference management software to ensure that reference lists are correctly formatted. An example of such software is Papers, which is part of Springer Science+Business Media.

Example reference style:

Article within a journal

Smith JJ. The world of science. Am J Sci. 1999;36:234-5.

Article within a journal (no page numbers)

Rohrmann S, Overvad K, Bueno-de-Mesquita HB, Jakobsen MU, Egeberg R, Tjønneland A, et al. Meat consumption and mortality - results from the European Prospective Investigation into Cancer and Nutrition. BMC Med. 2013;11:63.

Article within a journal by DOI

Slifka MK, Whitton JL. Clinical implications of dysregulated cytokine production. Dig J Mol Med. 2000; doi:10.1007/s801090000086.

Article within a journal supplement

Frumin AM, Nussbaum J, Esposito M. Functional asplenia: demonstration of splenic activity by bone marrow scan. Blood 1979;59 Suppl 1:26-32.

Book chapter, or an article within a book

Wyllie AH, Kerr JFR, Currie AR. Cell death: the significance of apoptosis. In: Bourne GH, Danielli JF, Jeon KW, editors. International review of cytology. London: Academic; 1980. p.251-306.

OnlineFirst chapter in a series (without a volume designation but with a DOI)

Saito Y, Hyuga H. Rate equation approaches to amplification of enantiomeric excess and chiral symmetry breaking. Top Curr Chem. 2007. doi:10.1007/128_2006_108.

Complete book, authored

Blenkinsopp A, Paxton P. Symptoms in the pharmacy: a guide to the management of common illness. 3rd ed. Oxford: Blackwell Science; 1998.

Online document

Doe J. Title of subordinate document. In: The dictionary of substances and their effects. Royal Society of Chemistry. 1999. http://www.rsc.org/dose/title of subordinate document. Accessed 15 Jan 1999.

Online database

Healthwise Knowledgebase. US Pharmacopeia, Rockville. 1998. http://www.healthwise.org. Accessed 21 Sept 1998.

Supplementary material/private homepage

Doe J. Title of supplementary material. 2000. http://www.privatehomepage.com. Accessed 22 Feb 2000.

University site

Doe, J: Title of preprint. http://www.uni-heidelberg.de/mydata.html (1999). Accessed 25 Dec 1999.

FTP site

Doe, J: Trivial HTTP, RFC2169. ftp://ftp.isi.edu/in-notes/rfc2169.txt (1999). Accessed 12 Nov 1999.

Organization site

ISSN International Centre: The ISSN register. http://www.issn.org (2006). Accessed 20 Feb 2007.

Dataset with persistent identifier

Zheng L-Y, Guo X-S, He B, Sun L-J, Peng Y, Dong S-S, et al. Genome data from sweet and grain sorghum (Sorghum bicolor). GigaScience Database. 2011. http://dx.doi.org/10.5524/100012.

Tables and captions

When preparing tables, please follow the formatting instructions below.

- Tables should be numbered and cited in the text in sequence using Arabic numerals (i.e.Table 1, Table 2 etc.).
- Tables less than one A4 or Letter page in length can be placed in the appropriate location within the manuscript.
- Tables larger than one A4 or Letter page in length can be placed at the end of the document text file. Please cite and indicate where the table should appear at the relevant location in the text file so that the table can be added in the correct place during production.
- Larger datasets, or tables too wide for A4 or Letter landscape page can be uploaded as additional files. Please see [below] for more information.
- Tabular data provided as additional files can be uploaded as an Excel spreadsheet (.xls) or comma separated values (.csv). Please use the standard file extensions.
- Table titles (max 15 words) should be included above the table, and legends (max 300 words) should be included underneath the table.
- Tables should not be embedded as figures or spreadsheet files, but should be formatted using „Table object" function in your word processing program.
- Color and shading may not be used. Parts of the table can be highlighted using superscript, numbering, lettering, symbols or bold text, the meaning of which should be explained in a table legend.
- Commas should not be used to indicate numerical values.