Energy expenditure through physical activity in a population of community-dwelling Brazilian elderly: cross-sectional evidences from the Bambuí Cohort Study of Aging

Gasto energético com atividades físicas em idosos brasileiros residentes em comunidade: evidências do Estudo de Coorte de Idosos de Bambuí

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Abstract

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Av. Augusto de Lima 1715, 6º andar, Belo Horizonte, MG 30190-002, Brasil. jucianyr@cpqrr.fiocruz.br The aim of this study was to estimate physical activity energy expenditure among older adults. The study comprised 1,585 residents in Bambuí, Minas Gerais State, Brazil, aged ≥ 60 years (91% of the town's total elderly), and examined the frequency and duration of 23 types of physical activity among them. Median energy expenditure was 975 MET.min/week (1,195.8 among men and 803.1 among women), declining significantly with age in both sexes. The prevalence of sedentary lifestyles (< 450 MET.min/week) was 31.2%. Unhurried walking accounted for about 1/3 of total energy expenditure. Multivariate analysis based on ordinal logistic regression showed inverse associations between energy expenditure and age and hospitalizations in both sexes. Among men, inverse associations were observed with smoking, number of chronic diseases and number of medical appointments. These results emphasize the need for effective strategies to increase physical activity in older elderly, and underscore the high prevalence of walking in this

Motor Activity; Energy Metabolism; Aged; Cohort Studies

Introduction

Regular physical activity is the factor that most consistently predicts healthy aging, as observed in several cohort studies of older adults ^{1,2,3}. Physical activities reduce cardiovascular disease rate independently of arterial pressure and other risk factors ^{4,5}. Among older adults, physical activity also leads to a reduction in incapacitating conditions, as well as falls and fractures ^{6,7}. Increased physical activity among the population can mean improved quality of life and lower health care costs for this population group ⁸.

In order to estimate physical activity in a population (its prevalence and the factors associated with low levels), the instruments used should offer good validity and reproducibility, in addition to easy applicability, particularly in population-based epidemiologic studies ⁹. The doubly-labeled water method, best practice for evaluating physical activity ¹⁰, is rarely used in population studies due to its high cost and operational difficulties ^{9,11}.

The literature shows a wide diversity of methods used to measure physical activity ⁹. The questionnaire is the instrument most commonly used in epidemiologic studies. The interview may comprise a single question or a question for each activity domain ^{12,13,14,15,16} or even several questions to secure more detailed information on the activity's frequency, intensity and duration ^{17,18}.

Evaluation of physical activity using a single question or one question for each activity domain offers reasonable validity 19,20, and is a strategy widely used in Brazil 13,16,21 and in other countries ^{22,23}. More recently it has become possible to estimate weekly or daily energy expenditure 24, using instruments such as the International Physical Activity Questionnaire (IPAQ) 25,26,27,28,29,30, and others 31,32,33,34. However, to our knowledge, there are no Brazilian population-based studies estimating energy expenditure from physical activity among the elderly.

The aim of this study was to estimate energy expenditure in physical activities and the associated factors with such expenditure among baseline participants in the elderly cohort of Bambuí, Minas Gerais State, Brazil.

Methods

Bambuí cohort of older adults

The Bambuí cohort study of older adults was conducted in the town of Bambuí (population approximately 15,000), in Minas Gerais State, Brazil. The study design and procedures have been described in previous publications 35,36. The baseline population comprised all the town's residents aged 60 years or over on January 1st, 1997, as identified by a complete census performed by the research team. Of the 1,742 elderly residents, 1,606 (92.2%) participated in the baseline interviews conducted from February to May 1997. All baseline participants were included for this study.

The study was approved by the Ethics Research Committee of the Oswaldo Cruz Foundation (Fundação Oswaldo Cruz), Rio de Janeiro. All participants signed a declaration of free, informed consent.

Study variables

Physical activity

Estimation of energy expenditure was based on 23 closed and two open questions on physical activities performed in the previous 90 days. The questions included the type and frequency of activity, and the average time (in minutes) spent on each activity. The following activities were considered: walking normally without hurrying (walking, 2.5mph), climbing stairs at normal speed (up stairs, using or climbing up ladder), climbing stairs quickly or carrying a load (carrying load upstairs, general), sweeping or mopping the floor (mopping and scrubbing floors), cleaning windows (cleaning - wash windows), swimming (swimming, leisurely, not lap swimming, general), dancing (dancing, general), rhythmic dancing (dancing, general), riding a bicycle (bicycling, < 10mph, leisure, to work or for pleasure), wall painting (home repair, painting), shuttlecock (volleyball), tennis (tennis, general), volleyball, basketball (basketball, non-game, general), soccer (soccer, casual, general), walking quickly (walking, 3.5mph, level brisk, firm surface, walking for exercise), going to a gym or doing exercises at home (gymnastics, general), running or jogging (jogging, general), digging to plant garden or vegetable patch (lawn and garden: digging, spading, filling garden, composting), sawing wood (forestry, sawing by hand), cycling quickly (bicycling, general), riding a horse at a gallop or trot (horse racing, galloping; horse racing, trotting), and riding a bicycle uphill (bicycling, mountain). The open questions included activities not covered in the list above, and the related responses were carefully coded by a physiotherapist (J.R.O.R.). Those activities included picking coffee, milking, brush-cutting, using a wheelchair, pruning trees, picking beans, carrying firewood, walking slowly with a stick, well-cleaning with a hoe, putting up a fence, cutting firewood, and stonemasonry or carpentry.

The intensity of each activity was coded according to the Compendium of Physical Activities, which permits international comparisons 37, and only activities of moderate to vigorous intensity were considered. Levels of physical activity were calculated on the basis of estimated oxygen consumption for each activity, which allowed energy expenditure to be quantified in metabolic equivalents (METs). Estimation of energy expenditure included only physical activities where each instance lasted at least ten minutes, given that shorter durations of physical activity yield less significant benefits 6,38,39. Energy expenditure was calculated using the following formula: MET (intensity of the activity) x time (duration of the activity in minutes) x frequency (number of times per week the activity was performed) 38. Sedentary individuals were considered to be those whose energy expenditure was less than 450 MET.min/week, which corresponds to at least 150 minutes per week of moderately to vigorously intense physical activity, in line with recommendations for older adults 6,40.

• Independent variables

The independent variables of the study included socio-demographic characteristics (age, schooling and marital status), health-related habits (tobacco and alcohol use), number of chronic diseases and use of health services (number of medical appointments and hospitalizations in the previous 12 months). Tobacco users were considered those who reported having smoked at least 100 cigarettes in their lives, and who continued smoking. Alcohol use was defined as intake of more than two measures per day in the previous 12 months. The number of chronic diseases was based on the history of medical diagnosis for arterial hypertension, cholesterol, angina, myocardial infarction, arthritis or rheumatism, diabetes and Chagas disease. Further details can be found in other publications 35,36.

Data analysis

Unadjusted data analysis was based on the Student t-test, Kruskal-Wallis test and Pearson chi-square test, respectively, for comparisons among means, medians and frequencies. Multivariate analysis was based on prevalence ratios (PRs) and confidence intervals of 95% estimated by logistic regression model in order to examine the strength of the associations between the independent variables and energy expenditure expressed in tertiles. The parallel regression assumption for the PRs was examined by the Wald test. For that assumption to hold, the PR estimated by comparing the lower tertile with the upper two tertiles will be the same as when the lower two tertiles are compared with the upper tertile 41; schooling among women was the only variable that did not fulfill that assumption, when separate PRs were calculated for each energy expenditure tertile. The analyses were stratified by sex and it was performed using the Stata statistics package, version 11.0 (Stata Corp., College Station, USA).

Results

Of the 1,606 baseline participants in the cohort, complete information on all the variables considered in this study was available for 1,585 (98.7%), who were included in this analysis. Of those participants, 633 (39.9%) were men and 952 (60.1%), women

Table 1 shows the distribution of the sociodemographic characteristics, health-related behavior, number of chronic diseases and use of health services among the study participants. Participants' mean age was 69.3 years (standard deviation – SD = 7.4), and they had predominantly little schooling (65.4% had completed less than four years' schooling). A spouse was reported by 49.3%. Prevalence of current tobacco use was 18.8% and excessive alcohol consumption was

reported by 2%, while 74.2% had a medical diagnosis of at least one chronic disease and, in the previous 12 months, 80.4% had visited a doctor and 22.4% had been hospitalized on at least one occasion. Important differences were observed between the sexes: significantly more men reported a spouse, current tobacco use and excessive consumption of alcohol. Women reported more diagnoses of chronic diseases, medical appointments and hospitalizations.

The most frequent physical activity was unhurried walking (72.4%), followed by sweeping or mopping the floor (48.8%), digging to plant a garden or vegetable patch (32.7%), walking briskly (29.5%), window cleaning (13.9%), horse-riding at a gallop or trot (7.2%) and dancing (6.1%). Prevalences of all these activities were significantly higher in men then in women, except for housework (sweeping or mopping the floor, and cleaning windows were more frequent activities among women), and dancing (where there was no significant difference) (Table 2).

Among the activities above, "walking normally without hurrying" was responsible for the greatest percentage of energy expenditure in the population studied (median = 34.8%; p25-p75: 8.5%-71.1%), corresponding to around 1/3 of total expenditure. The corresponding values for men and women were, respectively, 43.7% (p25p75: 21.4%-91.1%) and 27.3% (p25-p75: 3.1%-57.8%) (p < 0.001).

Table 3 shows physical activity energy expenditures by age and sex. Median total energy expenditure in the population studied was 975.0 METs.min/week (p25-p75: 292.5-1881.8). That value was significantly higher (p < 0.001) in men (1195.8; p25-p75: 397.5-2677.0) than in women (803.1; p25-p75: 237.2-1630.3), and the difference was observed consistently in the three age groups. By age, energy expenditures were observed to decline sharply in both sexes from the youngest to the oldest age group.

Overall prevalence of sedentarism was 31.2%. Prevalence was higher in women (34.4%) than in men (26.4%) (p = 0.001). Prevalence of sedentarism increased markedly with age (22.5%, 36.2% and 57.2% in the 60-69, 70-79 and 80 years or over age groups, respectively; p < 0.001).

Tables 4 and 5 show the results of multivariate analysis of the association between energy expenditure tertiles and socio-demographic characteristics, health-related behavior, health conditions and use of health services, by sex. Graded negative associations were observed between energy expenditure and age and number of hospitalizations in both sexes. Among men only, graded negative associations were observed between energy expenditure and current

Table 1

Distribution of socio-demographic characteristics, health-related behavior, number of chronic diseases and use of health services, by sex. The Bambuí Cohort Study of Aging, 1997.

| Variables | Total (N = 1.585) | Sex | | p-value * |
|---|-------------------|---------------|-----------------|-----------|
| | | Men (n = 633) | Women (n = 952) | |
| | % | % | % | |
| Mean age in years (standard deviation) | 69.3 (7.4) | 69.0 (7.3) | 69.5 (7.5) | 0.907 |
| Schooling (complete years) | | | | |
| 0-3 | 65.4 | 63.4 | 66.8 | 0.156 |
| ≥ 4 | 34.6 | 36.6 | 33.2 | |
| Married or consensual union | 49.3 | 73.2 | 33.5 | < 0.001 |
| Current smoker | 18.8 | 31.4 | 10.4 | < 0.001 |
| Alcohol use (more than 2 doses | 2.0 | 4.7 | 0.2 | < 0.001 |
| per day) in the previous 12 months | | | | |
| Number of chronic diseases diagnosed | | | | |
| by a doctor | | | | |
| 0 | 25.8 | 37.0 | 18.4 | < 0.001 |
| 1 | 35.0 | 34.4 | 35.4 | |
| ≥ 2 | 39.2 | 28.6 | 46.2 | |
| Number of medical appointments | | | | |
| in the previous 12 months | | | | |
| 0 | 19.6 | 28.6 | 13.7 | < 0.001 |
| 1-3 | 48.3 | 47.9 | 48.5 | |
| ≥ 4 | 32.1 | 23.5 | 37.8 | |
| Hospitalized once or more in the previous 12 months | 22.4 | 18.3 | 25.1 | 0.002 |

^{*} Student t-test for differences among means and Pearson chi-square test for differences among frequencies.

Note: all results in percentages, except where specified.

Table 2 Distribution of main physical activities performed, by sex. The Bambuí Cohort Study of Aging, 1997.

| Variables | Total (N = 1.585) | Sex | | p-value * |
|--|-------------------|---------------|-----------------|-----------|
| | | Men (n = 633) | Women (n = 952) | |
| | % | % | % | |
| Normal (unhurried) walking | 72.4 | 81.7 | 66.1 | < 0.001 |
| Sweeping/Mopping floor | 48.8 | 27.7 | 62.8 | < 0.001 |
| Digging to plant garden or vegetable patch | 32.7 | 40.0 | 27.9 | < 0.001 |
| Walking briskly | 29.5 | 38.4 | 23.6 | < 0.001 |
| Window cleaning | 13.9 | 7.0 | 18.4 | < 0.001 |
| Horse-riding (gallop or trot) | 7.2 | 16.8 | 0.8 | < 0.001 |
| Dancing | 6.1 | 6.1 | 6.1 | 0.966 |

^{*} Pearson chi-square test.

Note: physical activities (lasting 10 minutes or more) reported by less than 5% of total participants are not shown in the table.

Table 3 Physical activity energy expenditure in metabolic equivalents (MET.min/week), by sex and age group. The Bambuí Cohort Study of Aging, 1997.

| Age group (years) | Total | Sex | | |
|-------------------|-------------------------|-------------------------|-------------------------|--|
| | | Men | Women | |
| 60-69 | 1,170.0 (475.5-2,176.2) | 1,399.3 (585.0-3,035.2) | 1,040.1 (440.7-1,828.8) | |
| 70-79 | 750.0 (270.0-1,611.8) | 1,107.2 (390.0-2,010.0) | 662.1 (195.0-1,430.5) | |
| ≥ 80 | 292.5 (0.0-1,170.0) | 548.8 (120.0-1,254.8) | 237.3 (0.0-877.5) | |
| All | 975 (292.5-1,881.8) | 1,195.8 (397.5-2,677.0) | 803.1 (237.2-1,630.3) | |

Note: results expressed in medians (25^{th} percentile – 75^{th} percentile). Kruskall-Wallis test, p < 0.001 for comparisons among age groups (total and for each sex) and between the sexes.

Table 4 Analysis of factors associated with energy expenditure, in tertiles, for men. The Bambuí Cohort Study of Aging, 1997.

| Variables | Lower tertile (n = 211) | Middle tertile (n = 213) | Upper tertile (n = 209) | PR (95%CI) * |
|--|----------------------------|-----------------------------|----------------------------|------------------|
| | % | % | % | |
| Mean age in years (standard deviation) | 70.8 (7.9) | 69.1 (7.3) | 67.0 (6.0) | 0.95 (0.93-0.97) |
| Schooling (complete years) | | | | |
| 0-3 | 64.0 | 65.3 | 60.8 | 1.00 |
| ≥ 4 | 36.0 | 34.7 | 39.2 | 0.91 (0.66-1.24) |
| Marital status | | | | |
| Married or in consensual union | 69.2 | 70.9 | 79.4 | 1.00 |
| Single, divorced or widowed | 30.8 | 29.1 | 20.6 | 0.82 (0.58-1.15) |
| Current tobacco use | | | | |
| No | 61.1 | 70.4 | 74.2 | 1.00 |
| Yes | 38.9 | 29.6 | 25.8 | 0.56 (0.40-0.78) |
| Alcohol use > 2 doses per day in the | | | | |
| previous 12 months | | | | |
| No | 96.2 | 94.4 | 95.2 | 1.00 |
| Yes | 3.8 | 5.6 | 4.8 | 1.05 (0.54-2.07) |
| Number of chronic diseases diagnosed | | | | |
| by a doctor | | | | |
| 0 | 28.9 | 32.9 | 49.3 | 1.00 |
| 1 | 33.2 | 36.6 | 33.5 | 0.67 (0.47-0.95) |
| ≥ 2 | 37.9 | 30.5 | 17.2 | 0.40 (0.27-0.60) |
| Number of medical appointments in | | | | |
| the previous 12 months | | | | |
| 0 | 22.3 | 24.4 | 39.2 | 1.00 |
| 1-3 | 44.5 | 52.6 | 46.4 | 0.85 (0.59-1.22) |
| ≥ 4 | 33.2 | 23.0 | 14.4 | 0.54 (0.34-0.85) |
| Hospitalized in the previous 12 months | | | | |
| No | 71.1 | 86.4 | 87.6 | 1.00 |
| Yes | 28.9 | 13.6 | 12.4 | 0.60 (0.39-0.91) |

^{*} PR (95%CI): prevalence ratios and 95% confidence intervals estimated by ordinal logistic regression and adjusted for the variables listed in the table; the dependent variable was the tertile of energy expenditure expressed in MET.min/week. The Wald p-test values for the models indicate that the parallel regression hypothesis for the prevalence ratios was confirmed for all variables (p = 0.339).

Table 5 Analysis of factors associated with energy expenditure, in tertiles, for women. The Bambuí Cohort Study of Aging, 1997.

| Variables | Lower tertile (n = 319) % | Middle tertile (n = 316) % | Upper tertile (n = 317) % | PR (95%CI) * |
|--|---------------------------------|----------------------------------|---------------------------------|--------------------------------------|
| | | | | |
| Mean age in years (standard deviation) | 72.3 (8.3) | 68.9 (6.9) | 67.3 (6.2) | 0.94 (0.92-0.95) |
| Schooling (complete years) | | | | |
| 0-3 | 69.6 | 70.9 | 59.9 | 1.00 |
| ≥ 4 | 30.4 | 29.1 | 40.1 | 0.99 (0.73-1.35) 1.35 (1.00-1.81) |
| Marital status | | | | , |
| Married or in consensual union | 27.3 | 32.3 | 41.0 | 1.00 |
| Single, divorced or widowed | 72.7 | 67.7 | 59.0 | 0.83 (0.63-1.07) |
| Current tobacco use | | | | |
| No | 90.0 | 89.2 | 89.6 | 1.00 |
| Yes | 10.0 | 10.8 | 10.2 | 0.90 (0.60-1.34) |
| Alcohol use > 2 doses per day | | | | |
| in the previous 12 months | | | | |
| No | 99.7 | 100.0 | 99.7 | 1.00 |
| Yes | 0.3 | 0.0 | 0.3 | 0.47 (0.03-8.39) |
| Number of chronic diseases | | | | |
| diagnosed by a doctor | | | | |
| 0 | 18.5 | 15.5 | 21.1 | 1.00 |
| 1 | 31.7 | 37.0 | 37.6 | 1.06 (0.74-1.52) |
| ≥ 2 | 49.8 | 47.5 | 41.3 | 0.83 (0.58-1.19) |
| Number of medical appointments | | | | |
| in the previous 12 months | | | | |
| 0 | 9.4 | 12.7 | 18.9 | 1.00 |
| 1-3 | 48.9 | 46.8 | 49.9 | 0.76 (0.52-1.13) |
| ≥ 4 | 41.7 | 40.5 | 31.2 | 0.71 (0.47-1.09) |
| Hospitalized in the previous 12 | | | | |
| months | | | | |
| No | 64.6 | 74.4 | 85.8 | 1.00 |
| Yes | 35.4 | 25.6 | 14.2 | 0.51 (0.38-0.68) |

^{*} PR (95%CI): prevalence ratios and 95% confidence intervals estimated by ordinal logistic regression and adjusted for the variables listed in the table; the dependent variable was the tertile of energy expenditure expressed in MET.min/week. The Wald p-test values for the models indicate that the parallel regression hypothesis for the prevalence ratios was confirmed for all variables (p = 0.761), except schooling (p = 0.047).

tobacco use, number of chronic diseases, and number of medical appointments in the previous 12 months. Among women, a positive association was found between energy expenditure and greater schooling.

Discussion

The results of this study show that median energy expenditure in the population studied (975.0 MET.min/week) was 2.2 times greater than the 450 MET.minutes/week stipulated as the defining cutoff for sedentarism 6,41. Relative to that cutoff point, prevalence of sedentarism among older adults in Bambuí was 31.2%.

A study using data from Risk and Protective Factors Surveillance System for Chronic Non-Communicable Diseases Through Telephone Interview (VIGITEL), conducted on a representative sample of adults residing in domiciles with fixed telephones in the capitals of 26 states and the Federal District, showed that the prevalence of leisure-time sedentarism (physical activities of light or moderate intensity for at least 30 minutes < 3 days a week) among older adults was 86.3% 21. Another study, using the baseline survey of the Bambuí cohort of older adults and using the same instrument, showed a prevalence of leisure-time sedentarism similar to that observed in the VIGITEL population (87.2%) 13. Brazilian studies of other elderly populations, using various instruments that do not estimate energy expenditure, have shown prevalence of leisuretime sedentarism between 70.9% and 77.7% 42,43 and prevalence of sedentarism in overall activities ranging from 26.1% to 69.1% ^{25,26,44}.

The main physical activity of older adults in Bambui was unhurried walking, which corresponded to about 1/3 of total energy expenditure reported by that population. Other activities included housework (sweeping or mopping the floor and cleaning windows), particularly among women, in addition to digging to plant a garden or vegetable patch and horse-riding, reflecting specific characteristics to the area studied, where agriculture occupies an prominent place in the economy 35,36.

Our results show marked differences in physical activities between older men and women. Generally speaking, energy expenditure was greater among men than women in all age groups. When analyzed separately, greater prevalences were observed in men for all activities except housework. These results agree with those of other studies indicating that by and large, women are less active in most domains of physical activity, except in the domain of household activities 30,45.

Age was strongly associated with declining energy expenditure, even when adjusted for other significant factors, and that decline was observed consistently in both sexes. The gradual reduction of physical activities with age has been amply described of the literature 16,25,31, and our results agree with those observations. The number of chronic diseases showed a graded, inverse association with energy expenditure among men, reflecting the difficulty that people in worse health conditions have in practicing physical activities. The number of hospitalizations also showed an inverse association with energy expenditure among both men and women. Number of medical appointments was also associated positively associated with less energy expenditure among men. One plausible explanation for that association is the existence of incapacitating conditions that lead to greater recourse to health care.

The association between schooling and physical activity has been amply described in the literature; better levels of education area have been associated with lower prevalence of sedentarism 27,33,42. However, schooling showed a positive association with energy expenditure only among women. Tobacco use is another factor described in the literature as associated with reduced physical activities, including among older adults 27,33,42. This study found an inverse association between the condition of current smoker and energy expenditure among men, but not among women. This inconsistency is probably due to the low prevalence of tobacco use among older women.

Population-based epidemiologic studies estimating energy expenditure are rare, and older adults generally take part as one of several adult age groups 29,34. That is justified by the operational difficulties of applying such instruments in large populations. This study used an instrument containing twenty three questions on physical activities in different older age groups, in addition to questions on the frequency and duration of such activities; that was one advantage of this study. Other advantages were the large population base, the high survey response rate, the exhaustive training of the field teams, and the standardization of data collection 35,36. The main limitation was the cross-sectional nature of the study, from which it is impossible to establish time relationships among the variables or to examine how individual energy expenditure evolved over time. However, this design is appropriate for prevalence studies, as is the outcome here.

To summarize, the results of this study lead to the following conclusions: firstly, the broad predominance of walking among older adults' physical activities underlines the need to encourage this practice in health promotion policies for this population; secondly, the marked reduction in caloric expenditure with age raises the need for effective strategies to increase physical activities among the older elderly.

Resumo

O objetivo deste trabalho foi estimar o gasto energético das atividades físicas entre idosos. Participaram do estudo 1.585 residentes da cidade de Bambuí, Minas Gerais, Brasil, com ≥ 60 anos (91% da população total de idosos). Foram pesquisados 23 tipos de atividade física, sua frequência e duração. A mediana do gasto energético foi de 975,0 MET.min/semana, (1.195,8 entre homens e 803,1 entre mulheres), com acentuado declínio com a idade nos dois sexos. A prevalência do sedentarismo (< 450 MET.min/semana) foi de 31,2%. Caminhar normalmente (sem pressa) correspondeu a cerca de 1/3 do total do gasto energético estimado. Em ambos os sexos, os resultados da análise multivariada baseada na regressão logística ordinal mostraram associações inversas entre gasto energético e idade e ocorrência de hospitalizações. Entre homens, foram observadas associações inversas com tabagismo, número de doenças crônicas e consultas médicas. Esses resultados ressaltam a necessidade de estratégias efetivas para aumentar as atividades físicas de idosos mais velhos, chamando atenção para o predomínio da caminhada nesse grupo.

Atividade Motora; Metabolismo Energético; Idoso; Estudos de Coortes

Contributors

J. R. O. Ramalho participated in the data analysis, interpretation of results, and drafting the paper. M. F. Lima-Costa collaborated in the study design, data analysis, and review of the manuscript. J. O. A. Firmo contributed to the study design, and critical review of the manuscript. S. V. Peixoto was responsible for supervising the data analysis, discussion of the results, and drafting and final review of the manuscript.

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