UNIVERSIDADE FEDERAL DE PERNAMBUCO

CENTRO ACADÊMICO DA VITÓRIA

CURSO DE BACHARELADO EM EDUCAÇÃO FÍSICA

DEYVISON GUILHERME MARTINS SILVA

# IMPACTOS DO EXERCÍCIO AERÓBIO SOBRE O BALANÇO OXIDATIVO NO CÓRTEX PRÉ-FRONTAL: UMA REVISÃO SISTEMÁTICA DE MODELOS EXPERIMENTAIS

VITÓRIA DE SANTO ANTÃO

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TCC apresentado ao Curso de Bacharelado em Educação Física da Universidade Federal de Pernambuco, Centro Acadêmico da Vitória, como requisito para a obtenção do título de bacharel em Educação Física.

**Orientador(a):** Cláudia Jacques Lagranha
**Coorientador(a):** Matheus Santos de Sousa Fernandes

**VITÓRIA DE SANTO ANTÃO**

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# IMPACTOS DO EXERCÍCIO AERÓBIO SOBRE O BALANÇO OXIDATIVO NO CÓRTEX PRÉ-FRONTAL: UMA REVISÃO SISTEMÁTICA DE MODELOS EXPERIMENTAIS

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Aprovado em 25/09/2023.

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Profº. Me. Allifer Rosendo Pereira
Centro Universitário Brasileiro

*Um papel em branco, sem linhas, pode tornar incerta a escrita de uma história qualquer, mas para a origem de histórias predestinadas como a nossa não se faz necessário sequer papel, que dirá linhas.*

*À minha esposa.*

## RESUMO

A inatividade física tem alcançado enormes proporções, o que aumenta o risco de doenças crônicas, sendo o córtex pré-frontal (CPF) no sistema nervoso central (SNC) é um dos afetados por este problema. Como o SNC tem uma grande suscetibilidade ao dano oxidativo, nos questionamos se o exercício físico aeróbico (EA) poderia beneficiar o CPF. Portanto, nosso objetivo foi investigar os impactos do EA sobre o balanço oxidativo no CPF de roedores. Para tanto, realizamos uma revisão sistemática utilizando as bases de dados Pubmed/Medline, SCOPUS e BVS (Virtual Health Library). Utilizando os descritores “physical activity”, “exercise”, “exercise training”, “aerobic exercise”, “prefrontal cortex” and “oxidative stress”, 129 manuscritos foram selecionados, dos quais, após os critérios de inclusão e exlusão, 10 manuscritos foram utilizados nesta revisão. Natação e, principalmente, corrida em esteira foram utilizados como protocolo de treinamento. A duração das sessões de exercício variou de 30-120 minutos, a intensidade variou de 50-70% da capacidade máxima de corrida e a frequência semanal de 2-5 vezes. A duração total dos programas variou de 1 sessão a 16 semanas, sendo 8 semanas a mais utilizada. Resultados conflitantes foram observados para medidas de biomarcadores de estresse oxidativo e para medidas antioxidantes. Verificamos que a maioria dos estudos não mostraram prejuízo para o balanço oxidativo e que protocolos com duração de 8 semanas ou mais melhoraram antioxidante no CPF. Adicionalmente, são necessários mais estudos com protocolos de treinamento mais homogêneos para avaliar os efeitos de diferentes intensidades e durações nos níveis de estresse oxidativo no CPF.



Palavras-chave: Exercício Físico; Sistema Nervoso Central; Córtex Pré-frontal; Metabolismo Oxidativo.

**ABSTRACT**

Physical inactivity has reached enormous proportions, which increases the risk of chronic diseases, with the prefrontal cortex (PFC) in the central nervous system (CNS) being one of those affected by this problem. As the CNS is highly susceptible to oxidative damage, we wondered whether aerobic physical exercise (AE) could benefit the PFC. Therefore, our objective was to investigate the impacts of EA on the oxidative stress in the PFC of rodents. Therefore, we carried out a systematic review using the Pubmed/Medline, SCOPUS and VHL (Virtual Health Library) databases. Using the descriptors “physical activity”, “exercise”, “exercise training”, “aerobic exercise”, “prefrontal cortex” and “oxidative stress”, 129 manuscripts were selected, of which, after the inclusion and exclusion criteria, 10 manuscripts were used in this review. Swimming and, mainly, treadmill running were used as training protocols. The duration of the exercise sessions ranged from 30-120 minutes, the intensity ranged from 50-70% of maximum running capacity and the weekly frequency ranged from 2-5 times. The total duration of the programs varied from 1 session to 16 weeks, with 8 weeks being the most used. Conflicting results were observed for measures of oxidative stress biomarkers and for antioxidant measures. We found that most studies showed no harm to oxidative balance and that protocols lasting 8 weeks or more improved antioxidant activity in the PFC. Additionally, more studies are needed with more homogeneous training protocols, to evaluate the effects of different intensities and durations on the levels of oxidative stress on PFC.

Keywords: Physical Exercise; Central Nervous System; Prefrontal cortex; Oxidative Metabolism.

**SUMÁRIO**

**ARTIGO**

**O PRESENTE TRABALHO ESTÁ APRESENTADO NO FORMATO DE ARTIGO REQUERIDO PELA REVISTA "FREE RADICAL RESEARCH", CUJAS NORMAS E COMPROVANTE DE SUBMISSÃO SE ENCONTRAM EM ANEXO.**

# Impacts of Aerobic Exercise on the Oxidative Stress in the Prefrontal Cortex: A Systematic Review

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**Running Head:** Exercise and oxidative stress on PFC

**Abstract**

Physical inactivity has reached enormous proportions, which increases the risk of chronic diseases, with the prefrontal cortex (PFC) in the central nervous system (CNS) being one of those affected by this problem. As the CNS is highly susceptible to oxidative damage, we wondered whether aerobic physical exercise (AE) could benefit the PFC. Therefore, our objective was to investigate the impacts of EA on the oxidative stress in the PFC of rodents. Therefore, we carried out a systematic review using the Pubmed/Medline, SCOPUS and VHL (Virtual Health Library) databases. Using the descriptors "physical activity", "exercise", "exercise training", "aerobic exercise", "prefrontal cortex" and "oxidative stress", 129 manuscripts were selected, of which, after the inclusion and exclusion criteria, 10 manuscripts were used in this review. Swimming and, mainly, treadmill running were used as training protocols. The duration of the exercise sessions ranged from 30-120 minutes, the intensity ranged from 50-70% of maximum running capacity and the weekly frequency ranged from 2-5 times. The total duration of the programs varied from 1 session to 16 weeks, with 8 weeks being the most used. Conflicting results were observed for measures of oxidative stress biomarkers and for antioxidant measures. We found that most studies showed no harm to oxidative balance and that protocols lasting 8 weeks or more improved antioxidant activity in the PFC. Additionally, more studies are needed with more homogeneous training protocols, to evaluate the effects of different intensities and durations on the levels of oxidative stress on PFC.

**Key words:** Physical Exercise; Central Nervous System; Prefrontal cortex; Oxidative Stress

## Introduction

According to data from the Global Physical Activity Status Report (2022), by 2030 approximately 500 million people will be affected by chronic diseases. This document predicts an annual cost of 27 billion dollars for the treatment of chronic diseases if there are no changes in the prevalence of inactivity (1). In this sense, a prospective cohort study for 12.4 years with a sample of 334.161 participants identified that physically inactive people had an increased risk for early mortality, mainly due to cardiovascular diseases (2). Additionally, a recent study showed that people with lower levels of activity are more susceptible to acquiring post-acute sequelae of SARS-CoV-2 infection, including dyspnea, insomnia, fatigue, and severe skeletal muscle pain (3).

On the other hand, prospective studies have shown that a higher amount of physical activity/exercise, mainly aerobic during the week, provides benefits in several body systems (4-6). Among them is the Central Nervous System (CNS), which has a high dependence on oxygen, lipid content, and low antioxidant capacity (7); it's highly benefited from the positive effects of exercise, mainly due to the increased in the release of neurotrophic factors including Brain-derived Neurotrophic Factor (BDNF), Insulin Growth Factor 1 (IGF-1) (8), decrease the production and release of pro-inflammatory cytokines implicated in neurodegeneration process (6, 9) and, the adaptations in oxidative metabolism (e.g.: function and mitochondrial biogenesis) (10, 11) to combat oxidative stress (OS) - understood as a chronic imbalance between the production and removal of pro-oxidant agents (12).

In this sense, the prefrontal cortex (PFC) stands out in the central nervous system as a tissue that might be impacted by physical exercise and whose interaction can be mediated by the modulation of the reduction/oxidation (REDOX) balance (13). Associated with the control of executive functions, the PFC is involved in the selection of stimuli, working memory, changing rules, decision making (14, 15) and processing stimuli based on previous experiences, producing motor action (14); in addition to being associated with psychological disorders influenced by OS (16).

Aerobic exercise (AE) can be an essential intervention in the face of positive adaptations against the oxidative balance on the CNS and PFC since data in the literature showed that AE is an excellent tool to achieve adaptations in the oxidative

metabolism of the CNS. However, a consensus on the best aerobic training design to achieve such effects needs to be improved since there are conflicting results in the literature regarding exercise intensity, duration, and model, as well as methods to measure OS. Therefore, the present review proposes to highlight this need for investigating the impacts of aerobic physical exercise on the oxidative stress in the PFC of rodents.

## METHODS

The present systematic review was based on the PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) guidelines (17).

### *Information sources*

The data search was performed, with no publishing time limit, for manuscripts published until February 2023. The PubMed (Medline), Scopus and BVS (Virtual Health Library) databases were used to track manuscripts published in English. In addition, the search was systematized using Boolean operators (AND and OR) with the following search equation: *(((((physical activity) OR (exercise)) OR (exercise training)) OR (aerobic exercise)) AND (prefrontal cortex)) AND (oxidative stress).*

### *Study Selection and Eligibility*

Eligibility criteria were previously used to minimize the risk of bias. The inclusion and exclusion criteria followed the PICOS (**P**opulation / **I**ntervention/ **C**ontrol/ **O**utcomes/ **S**tudy) **(Table 1)** (18). Manuscripts that did not meet the following eligibility criteria were excluded **(a)** Studies that used only rodents (mice and rats); **(b)** Studies that intervened with only the aerobic exercise; **(c)** Manuscripts without limitation of publication date and time; **(d)** Studies that analyzed the oxidative balance components (oxidative stress biomarkers and antioxidant defenses); **(e)** Studies with the other organisms; as well as **(1)** reviews, (2) letter for editor, (3) duplicates, and presence of data used; (4) full text not available; (5) texts not written in English; (6) did not use

aerobic exercise; (7) did not evaluate parameters of oxidative balance in the prefrontal cortex; (8) non-comparative study were excluded.

**Table 1.** PICOS strategy

| | **Inclusion Criteria** | **Exclusion Criteria** |
|---|---|---|
| **P**opulation | Rodents | Humans and other organisms |
| **I**ntervention | Aerobic Exercise | No Aerobic Exercise |
| **C**ontrol | No Aerobic Exercise | - |
| **O**utcomes | Oxidative Balance - Carbonyls, Thiobarbituric Acid Reactive Species, Reactive Oxygen Species, Hydrogen peroxide, and Superanion. For antioxidant outcomes were collected including the activity of Superoxide dismutase , Catalase, Glutathione-S-Transferase, Glutathione peroxidase and Glutathione reductase enzymes was measured, in addition to the content of total thiols, reduced glutathione and ferric reducing antioxidant power. | No Oxidative Balance |
| **S**tudy Design | Animals Studies | Reviews; Case report; Letter to editor; comments, etc.. |

**Methodological Quality Assessment**

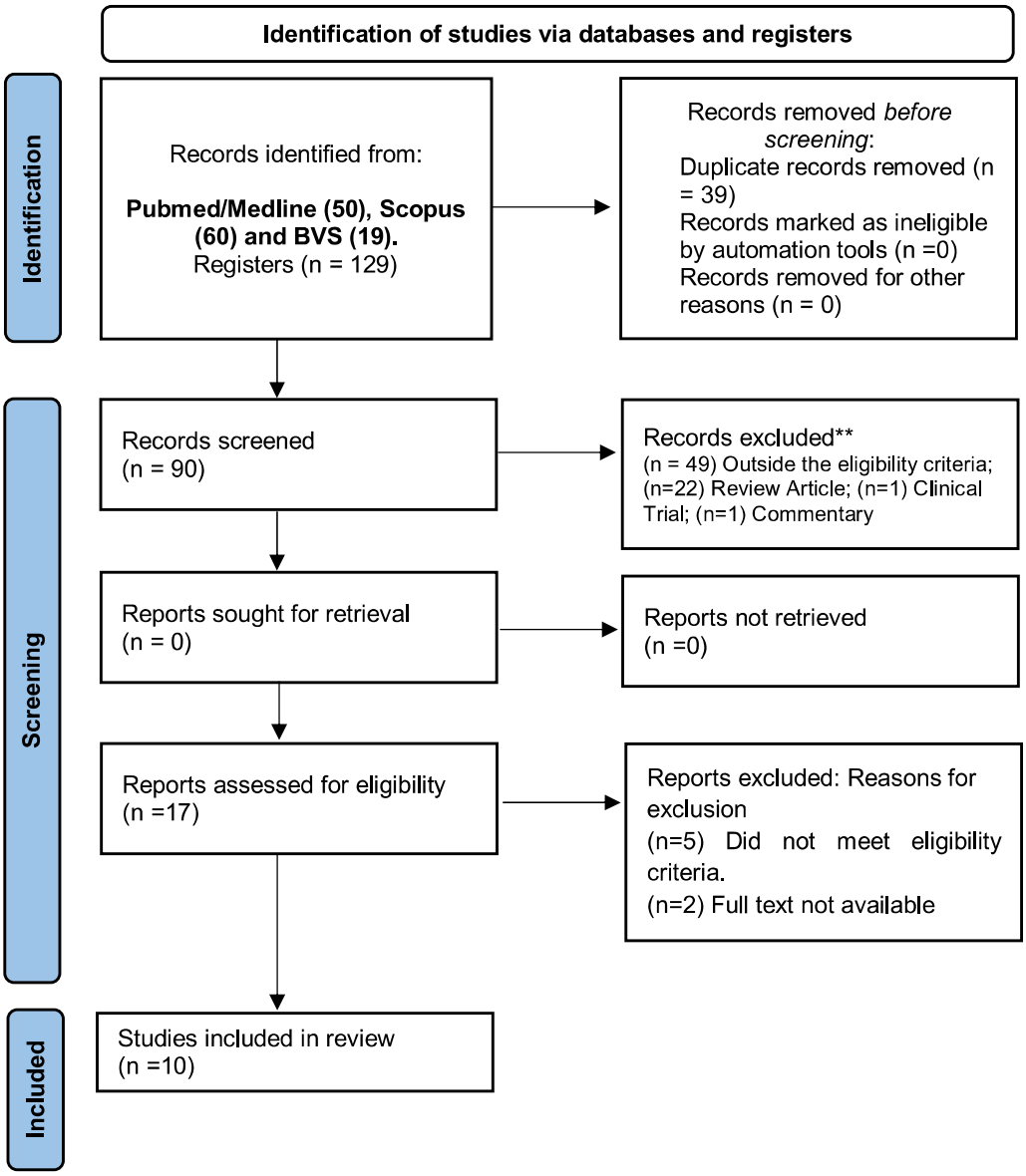
The SYRCLE's strategy was used to assess the methodological quality of the animal studies. The tool consisted of ten questions that evaluate methodological criteria: Q1- Was the allocation sequence adequately generated and applied? Q2- Were the groups similar at baseline, or were they adjusted for confounders in the analysis? Q3- Was the allocation to the different groups adequately concealed? Q4- Were the animals randomly housed during the experiment? Q5- Were the caregivers and/or investigators blinded from knowledge of which intervention each animal received during the investigation? Q6- Were animals selected at random for outcome assessment? Q7- Was the outcome assessor-blinded? Q8- Were incomplete outcome data adequately

addressed? Q9- Are reports of the study free of selective outcome reporting? Q10- Was the analysis free of other problems that could result in a high risk of bias? Questions were answered with 'Yes,' 'No,' or 'Not clear.' When the answer was 'yes,' a score was given; when the answer was 'no' or 'not clear,' no score was given. The overall scores for each article were calculated as a score of 0-10 points, with the quality of each study being classified as high (8-10), moderate (5-7), or low (<5). The two reviewers independently reviewed all included studies. Discrepancies between raters were resolved by consensus or by a third party (CJL) when necessary. The quality outcomes are described in **Table 2**.

***Search Strategy and Selection Process*:**

The articles retrieved through the cited databases were filtered based on the already defined inclusion and exclusion criteria. A PRISMA flow chart was created to describe identifying, sorting, and including selected texts (**Figure 1**). Using the descriptors, 129 articles were retrieved across the three databases. Of these, 39 duplicate articles were excluded, leaving 90 articles. Then, the researchers screened the texts based on their titles and abstracts to check their compatibility with the selection criteria and search strategy. This resulted in the exclusion of 73 articles (49 outside the inclusion criteria, 22 reviews, one trial, and one comment). There were 17 texts left for reading, of which five were excluded (two unavailable full text and five outside the criteria). Ten articles, therefore, were included and thoroughly analyzed for this systematic review.

**Figure 1.** PRISMA flow chart




## Data items

To answer the hypothesis of this systematic review, different data were extracted. Initially, we collected the following information: author and year, species, sex, and age. In addition, data were collected on the structure of aerobic exercise protocol, including the type of aerobic exercise, familiarization strategy, duration, frequency, volume, and intensity **(Table 3)**. Next, data on the prefrontal cortex, oxidative stress biomarkers

were evaluated, such as Carbonyls, Thiobarbituric Acid Reactive Species (TBARS), Reactive Oxygen Species (ROS levels), Hydrogen peroxide (H2O2), and Superoxide (O2•). Antioxidant outcomes were collected, including the activity of Superoxide dismutase (SOD), Catalase (CAT), Glutathione-S-Transferase (GST), Glutathione peroxidase (GPx), and Glutathione reductase (GR) enzymes were measured, in addition to the content of total thiols, reduced glutathione (GSH) and ferric reducing antioxidant power (FRAP) **(Table 4)**.

## RESULTS

### *Search results in databases*

In an initial search, 129 manuscripts were identified [PubMed/Medline (50); Scopus (60); VHL (19)]. Then, 39 duplicates excluded using the EndNote® software. 90 manuscripts were screened and submitted to the eligibility criteria, and 73 manuscripts were excluded based on title and abstract reading. Seventeen studies remained for full-text reading. Seven studies were excluded due to the following reasons: Five not agreeing with the eligibility criteria, and two studies did not have the full text available. Finally, 10 studies in this systematic review were included **(Figure 1).**

### *Methodological Quality Assessment*

Methodological quality assessment of the included studies is shown in **Table 2**. All studies showed adequate and randomized allocation with randomly selected rodents. In addition, incomplete results were handled appropriately, free from selective results and bias. As these are studies involving intervention (aerobic exercise), it is not possible to consider the investigation and analysis of the results blind. In general, all studies presented satisfactory quality criteria.

### *Characteristics of the included studies*

A summary containing the sample and aerobic protocols description of the 10 manuscripts (19-28) included in this systematic review can be found in **Table 3**.

The age of the animals from each manuscript ranged from 28 days to 9 months of age. Training protocols varied between running on a treadmill (7 studies), swimming (2 studies) and voluntary running on a wheel (1 study). Control groups were non-exercised and/or non-exercised animals placed on the switched off treadmill for the same length of time as the protocol for trained animals. The duration of the exercise sessions varied from 30 to 120 minutes, the intensity varied from 50 to 70% of the maximum running capacity and the weekly frequency from 2 to 5 times a week, the latter being the most recurrent frequency. The total duration of the programs ranged from 1 session to 16 weeks.

The studies were relatively homogeneous regarding the evaluation of pro-oxidant and antioxidant agents. As a measure of pro-oxidant molecules, the manuscripts evaluated the levels of reactive oxygen species (ROS), Thiobarbituric Acid Reactive Species (TBARS), carbonyls, superoxide ($O_2^{\bullet}$) and hydrogen peroxide ($H_2O_2$). For antioxidant evaluation, the activity of Superoxide dismutase (SOD), Catalase (CAT), Glutathione-S-Transferase (GST), Glutathione peroxidase (GPx) and Glutathione reductase (GR) enzymes was measured, in addition to the content of total thiols, reduced glutathione (GSH) and ferric reducing antioxidant power (FRAP).

***Aerobic exercise on pro-oxidant agents in the PFC***

9 manuscripts evaluated the TBARS content, of these, 7 manuscripts showed no difference (19, 22-26, 28), 1 study showed a reduction (20) and 1 study an increase (21); 3 studies analyzed the carbonyl content, of which one study showed a reduction (20) and 2 studies showed no difference (22, 23); 4 studies evaluated the total content of ROS, without demonstrating any difference (21, 25, 27, 28) and 1 study evaluated the mitochondrial content of superoxide and hydrogen peroxide (20), both reduced in the exercised groups, except for $O_2^{\bullet}$ in the adults group **Table 4**.

***Impacts of aerobic exercise on antioxidant capacity on PFC***

5 studies evaluated SOD activity, which was: increased (20, 27), decrease (24) or no significant difference (19, 22, 26); 4 studies evaluated CAT activity, which increased (20) or no significant difference (22, 27, 28); 6 studies evaluated GPx activity,

its activity was increased (20, 27) or no significant difference (19, 21, 28); 3 studies evaluated GST activity, which demonstrated increase (22) or no significant difference (20, 27); and 2 studies evaluated the GR activity, which was increased (20) or no significant difference (27). 5 studies evaluated the GSH content, which was increased (20-22) or without significant difference (25, 28); 2 studies evaluated the total thiol content, where one study showed an increase (22) and 1 study showed no significant difference (23); 1included study evaluated the FRAP (24), which showed no difference **Table 4**.

## DISCUSSION

The aim of this systematic review was to highlight the impacts of AE on the oxidative balance in the PFC of rodents. In our results, we observed that there was great variation in relation to the AE protocols among the works that aimed to investigate its effect on the oxidative balance. Both animals (29, 30) and clinical studies (31) indicate that AE is an intervention that promotes adaptations in oxidative metabolism, although these effects are dependent on the manipulation of variations applied to the training protocol, such as type, volume, and intensity (32). Here, we found that 5 papers showed modulation in oxidative metabolism variables, while another 5 showed no change.

In physiological concentrations, ROS play an important role in physiological mechanisms such as cell differentiation and proliferation, apoptosis and REDOX signaling (33, 34). However, the deregulation of this balance can lead to the installation of OS, damaging biomolecules (such as lipids, proteins, and nucleic acids), whose repercussions are especially greater in the brain (35). Our data showed that only one included study demonstrated a reduction in the content of carbonyls, MDA, $O_2^{\bullet}$ and $H_2O_2$ content in the PFC ((19, 21, 23-25, 28), another showed an increase in MDA content (FLÔRES *et al*., 2014) and the other 8 showed no change.

Regarding the effect of AE on OS biomarkers, the available literature seems not to have consolidated yet. Peripherally, aerobic training models conducted in 60 minutes a day, 5 times a week, at 50% or 60% of the maximum running capacity and with a total duration of 4 or 8 weeks, respectively, the exercise did not change markers of lipid and protein oxidation in the heart (36) and liver (30). On the other hand, however, evidence

demonstrating that AE reverses measures of lipid and protein oxidation in the CNS (29). In a model of moderate running on a treadmill also performed at moderate intensity, Macêdo *et al*., (2017) demonstrated an exercise-induced reversal in the concentration of MDA and carbonyls in the hippocampus in 46-day-old *Wistar* rats. Thus, it is reasonable to consider that there is a tissue-specific response to the effect of exercise on pro-oxidant markers, so that the CNS is a preferentially modulable target.

About the effect of AE on the entire antioxidant system (enzymatic and non-enzymatic) there is little more evidence to support the benefit of its use. Dealing with its enzymatic portion, our data demonstrate that in 3 articles the AE increased the activity of SOD, CAT, GPx, GST, and GR (20, 22, 27); while 1 single study demonstrated a reduction in GPx and SOD activity (24) and another 5 articles did not make any difference. SOD, CAT and GPx act, in a convergent manner and through two reactions, to carry out the complete reduction of $O_2^{\bullet}$ to water (33, 37). Shi et al., (2018) and Macêdo et al., (2017), both studying the hippocampus, demonstrated, respectively, that moderate-intensity aerobic exercise was able to attenuate the cognitive dysfunction induced by a high-fat diet by increasing Manganese-dependent Superoxide Dismutase activity and total SOD and CAT activity (29, 38).

Thus, the contradictory effect of AE on SOD and GPx activity observed in article 7 of this review can be explained by the author's experimental model. MORADI-KOR *et al.*, 2020 used the wheel running model, where there is no control of two of the main parameters associated with adaptations to exercise – volume and intensity (39, 40). Therefore, the reduction in the activity of these enzymes may have been due to overtraining provided by free access to the running wheel.Regarding the non-enzymatic portion of the antioxidant system, the effect of AE was identified through the increase in the content of GSH and total thiols (20-22), or no difference (HOEPERS *et al.,* 2020; NEVES ET *al.,* 2015; SCHIMITD *et al.,* 2014). Considering GSH as the main intracellular thiol related to cellular antioxidant capacity, the ratio between its reduced and oxidized form is one of the best indicators of cellular REDOX status (41).

Corroborating the positive effect of AE that we observed on these antioxidant measures, moderate-intensity AE training programs increase the concentration of GSH, the GSH/GSSG ratio, and the number of total thiols in different tissues, such as the liver (30) and heart (36). On the other hand, that during physical effort it promotes several physiological adaptations, dependent on the correct application of variables linked to

physical training including volume, intensity, and frequency of performance (COFFEY and HAWLEY, 2007). The possible lack of difference observed for some measures of oxidative balance may be related to the heterogeneity of protocols used among the articles included in this systematic review.

**Strengths, limitations, and future directions**

Thus, the lack of methodological homogeneity employed makes it difficult to interpret the results obtained and prevents drawing incontestable conclusions. This may be due to: 1) a combination of treadmill running, wheel running and swimming; 2) training volume taken to exhaustion or not controlled; 3) wide range of intensities and intensities based on percentage of body weight or percentage of aerobic capacity (treadmill training). Even so, considering the tendency of running training on a moderate-intensity treadmill to improve antioxidant capacities vital to the maintenance of REDOX homeostasis, future work should compare the effects of this and other intensity ranges on the PFC oxidative balance in animals submitted to running on a treadmill.

## CONCLUSION

Thus, it can be concluded that aerobic exercise protocols during 8 weeks or more showed improvement in the antioxidant system in PFC from rodents. However, there was no consensus response to measures of OS biomarkers and antioxidant defenses in response to AE. Therefore, comparative works that isolate the variables volume, intensity, type of exercise and study them independently may help to better understand the special effects of oxidative metabolism in response to aerobic physical exercise.

## CONFLICT OF INTEREST STATEMENT

The authors declare no conflict of interest

## AUTHORSHIP CONTRIBUTION STATEMENT

All authors had full access to all the data in the study and take responsibility for the integrity and the accuracy of the all data analysis. **D.G.M.S:** *Conceptualization, Methodology,*

*Investigation, Data curation, Visualization, Discuss data, Writing - original draft.* **M.S.S.F & J. H. S;** *Methodology, Investigation, Data curation, Discuss data and Writing – editing & review draft.* **C.J.L.:** *Resources, Mentoring, Project administration, Funding acquisition, Writing - review & editing final version.*

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## DATA AVAILABILITY STATEMENT

The data that supports the findings of this study are available from the corresponding author, upon reasonable request.

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## APÊNDICE A – TABELAS

**Table 2.** Methodological Quality Assessment

| | | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|
| Aksu et al. 2009 | Y | Y | Y | Y | N | Y | N | Y | Y | Y |
| Abhijit et al.2019 | Y | Y | Y | Y | N | Y | N | Y | Y | Y |
| Acikgoz et al.2006 | Y | Y | Y | Y | N | Y | N | Y | Y | Y |
| Fernandes et al. 2022 | Y | U | Y | Y | N | Y | N | Y | Y | Y |
| Flôres et al. 2014 | Y | Y | Y | Y | N | Y | N | Y | Y | Y |
| Hoepers et al.2020 | Y | Y | Y | Y | N | Y | N | Y | Y | Y |
| Moradi K et al.2020 | Y | Y | Y | Y | N | Y | N | Y | Y | Y |
| Neves et al.2015 | Y | Y | Y | Y | N | Y | N | Y | Y | Y |
| Souza et al.2013 | Y | Y | Y | Y | N | Y | N | Y | Y | Y |
| Schimitd et al.2020 | Y | Y | Y | Y | N | Y | N | Y | Y | Y |

**Legend:** Q1. Was the allocation sequence adequately generated and applied? Q2. Were the groups similar at baseline or were they adjusted for confounders in the analysis?; Q3. Was the allocation to the different groups adequately concealed during? Q4. Were the animals randomly housed during the experiment?; Q5. Were the caregivers and/or investigators blinded from knowledge which intervention each animal received during the experiment?; Q6. Were animals selected at random for outcome assessment? Q7. Was the outcome assessor blinded? Q8. Were incomplete outcome data adequately addressed? Q9. Are reports of the study free of selective outcome reporting?; Q10. Was the study apparently free of other problems that could result in high risk of bias? Y, Yes; N, No; U, Unclear

**Table 3.** Sample and aerobic exercise protocol characteristics

| Author, Year | Species, Sex and Age | Animals per group | Aerobic Exercise Protocol Description | | | | Intervention Time (Weeks) |
|---|---|---|---|---|---|---|---|
| | | | Type | Frequency | Volume | Intensity | |
| Aksu et al. 2009 | Sprague-Dawley rats; Male; 22 weeks old | 8 | Treadmill run | 5 days/wk | 60 min/session or until exhaustion | 10 – 20 m/min exhaustive exercise | 8 wks |
| Abhijit et al.2019 | *Wistar* rats; Male; 2 groups: 4 and, 18 months old | 9 | Swimming | - | 120 min/session | 3% body weigh | 16 wks |
| Acikgoz et al.2006 | *Wistar* rats; Male; 24 weeks old | 49 | Treadmill run | 1 day | Until exhaustion | 25 m/min, incline of 5º | 1 day |
| Fernandes et al. 2022 | Mice *C57B6*; Male; 8 weeks old | 6 | Treadmill run | 5 days/wk | 60 min/session | 60% max running capacity | 8 wks |
| Flôres et al. 2014 | *Wistar* rats; Male; 9 months old | 16-18 | Treadmill run | 5 days/wk | 30 min/session | 60 - 70% $VO_2$ max | 12 wks |
| Hoepers et al.2020 | Wild-type *C57BJ6*; Male; 28 days old | 15 | Treadmill run | 2 days/wk | 30 min/session | Low-intensity | 8 wks |
| Moradi K et al.2020 | *Wistar* rats; Female; 30 days old | 10 | Wheel Running | Free access to running wheel | Not controlled | Not controlled | 15 days |
| Neves et al.2015 | *Wistar* rats; Male; 45 days old | 8-12 | Treadmill run | 5 days/wk | 50 min/session | 50 - 70% $VO_2$ max | 8 wks |
| Souza et al.2013 | *Swiss* albino mice; Male; 2-4 months old | 10 | Swimming | 5 days/wk | 20 - 60 min/session | 0 - 3 % body weigh | 8 wks |
| Schimitd et al.2020 | *Wistar* rats; Male; 2 months old | 8-10 | Treadmill run | 5 days/wk | 30 min/session | 60 - 70% $VO_2$ max | 8 wks |

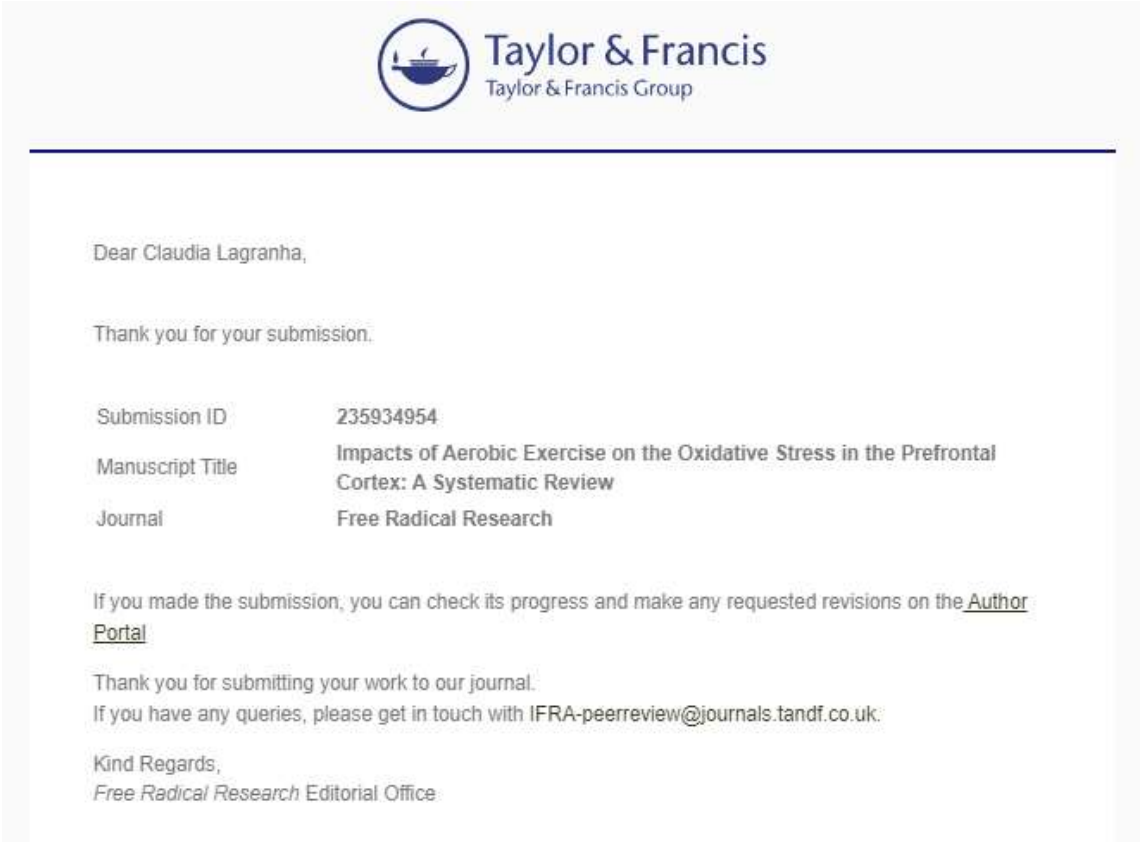
**Legend: $VO_2$, maximal oxygen consumption.**

**Table 4.** Impacts of aerobic exercise on oxidative stress and antioxidants outcomes in prefrontal cortex

| Author, Year | Oxidative Stress Biomarkers | Antioxidants Outcomes |
|---|---|---|
| Aksu et al. 2009 | = MDA (TBARS) | =GPx; =SOD |
| Abhijit et al.2020 | ↓ $O_2$-; ↓ Carbonyls; ↓ $H_2O_2$; ↓MDA (TBARS) | ↑ Catalase; ↑ GPx; =GST; ↑GR; ↑GSH; ↑SOD |
| Acikgoz et al.2006 | = MDA (TBARS) | =GPx; =SOD |
| Fernandes et al. 2022 | = Carbonyls; = MDA (TBARS); | = Catalase; ↑GST; ↑GSH; =SOD; ↑ Free Thiols |
| Flôres et al. 2014 | ↑ MDA (TBARS); = ROS levels | =GPx; ↑GSH; |
| Hoepers et al.2020 | = Carbonyls; = MDA (TBARS); | = Free Thiols |
| Moradi K et al.2020 | = MDA (TBARS) | ↓GPx; ↓SOD; = FRAP |
| Neves et al.2015 | = MDA (TBARS); = ROS levels | = GSH |
| Souza et al.2013 | = ROS levels | = Catalase; ↑ GPx; = GST; = GR; = NPSH; ↑SOD |
| Schimitd et al.2014 | = MDA (TBARS); = ROS levels | = Catalase; = GPx; = GSH; |

**Legend: FRAP:** Ferric Reducing Antioxidant Power; **GST**: Glutathione S-transferase, **GPx:** Glutathione Peroxidase; **GSH:** Reduced Glutathione, **GSSG:** Oxidized Glutathione; **H2O2:** hydrogen peroxide; **MDA:** Malondialdehyde; **ROS levels:** Reactive Oxygen Species; **SOD:** Superoxide Dismutase.

## APÊNDICE B – COMPROVANTE DE SUBMISSÃO



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