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Suppression of tumorigeniciy 2 after exercise: a systematic review

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Suppression of tumorigenicity 2 after exercise: a systematic review

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TÍTULO DISSERTAÇÃO/MONOGRAFIA (riscar o que não interessa)

Suppression of tumorigenicity 2 after exercise: a systematic review

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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.	X

Faculdade de Medicina da Universidade do Porto, 01/04/2021

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Suppression of tumorigenicity 2 after exercise: a systematic review

Running title: ST-2 after exercise

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ABSTRACT

Exercise is a pivotal physiological activity, associated with benefits. Whilst the importance of physical activity is consensual along different steps of the cardiovascular (CV) continuum, there has been interest in assessing the CV adaptations to vigorous exercise. Indeed, exercise can be associated with increases in cardiac biomarkers, though the scope of this observation remains elusive. Interleukin 1 receptor related protein, Suppression of tumorigenicity 2 (ST2) is a biomarker related to the pathophysiology of fibrosis, having shown promise in the study of heart failure. Knowledge of ST2 kinetics could improve understanding of the mechanistic pathways related to CV adaptations to exercise. To assess the current state-of-the-art concerning ST2 levels after exercise in healthy individuals. A systematic review was carried out on three databases (Pubmed, ISI Web of Science and Scopus), up to October 2020, using the queries "ST2" or "ST-2" + "exercise" or "running". A total of six studies were included in the review, encompassing 349 subjects (73% male gender) in which ST2 was assessed. Most studies reported increases in ST2 levels after exercise. Three studies, encompassing a total of 219 individuals, described a cut-off level of 35 ng/dL for ST2. In these, 92.7% of subjects had ST2 levels above this cut-off after exercise (running in all studies). Most studies report increased levels of ST2 after exercise, with an important number of individuals exceeding the 35 ng/dL threshold. Given the small number of individuals represented and the lack of imaging data and long-term follow-up, further prospective larger studies should target this.

INTRODUCTION

Physical activity (PA) is one of the pillars of a healthy lifestyle [1-3]. Exercise is associated with a myriad of beneficial effects, ranging from reductions in the prevalence of several cardiovascular diseases and the incidence of different cancers, to improvements in quality of life and an overall increased longevity [3-5]. Given this background, promotion of regular moderate PA has gained a spotlight among contemporary preventive approaches [1, 6].

Exercise can affect several distinct systems, exerting its effects by a plethora of mechanisms ranging from improvements in the cardiovascular and respiratory systems, to musculoskeletal and metabolic adaptations [7-10]. Whilst the positive effects of regular moderate PA are currently consensual over a wide range of settings, ranging from healthy individuals to those with different pathologies [11-14], the overall effects of prolonged strenuous exercise have become a source of interest [15-17]. Indeed, different reports have shown potentially deleterious cardiovascular effects of prolonged strenuous exercise, though the specific mechanisms underlying these as well as their relative impact remain to be further ascertained [16, 18-21].

Biomarkers of cardiac injury and strain, such as troponins and natriuretic peptides, play a paramount role in the contemporary diagnosis and management of several cardiovascular pathologies [22-24]. Interestingly, exercise such as marathon running and triathlon can be associated with increases in the levels of different cardiac biomarkers [25-27]. Whilst this, the full scope of cardiac biomarker elevations after exercise remains elusive [25, 28, 29]. Given the current number of individuals undergoing intense exercise, further knowledge on the cardiovascular adaptations as well as on the mechanistic pathways associated is of importance, as to further refine this relationship [24, 28].

Over the years, several novel biomarkers have been under study in the context of cardiovascular disease [22]. Among these, suppression of tumorigenicity 2 protein (ST2), a member of the interleukin (IL)-1 receptor family, which can be expressed in several cells in different settings, has been the focus of interest [30-34]. This biomarker can be expressed by both cardiac and extracardiac sites, playing a role in the pathophysiological mechanisms of myocardial fibrosis and remodeling [32, 34-36]. Notably, some studies have shown the potential of ST2 in heart failure, further reinforcing its clinical importance [37-39]. Given this background, the profile of ST2 kinetics among athletes could further improve the current knowledge underlying cardiac adaptations (and potential maladaptations) in the face of exercise training [24, 40, 41].

This review aimed at addressing the impact of exercise in ST2 levels among healthy individuals, and to present a pragmatic appraisal of the current literature on this field. This is, to the best of our knowledge, the first systematic review on this specific topic and could thus further improve current knowledge on the kinetics of different cardiac biomarkers after exercise, a topic of considerable translational relevance.

METHODS

Search Strategy

The study started with a search on three databases, Medline (PubMed), ISI Web of Science and Scopus, using the queries "ST2" or "ST-2" + "exercise" or "running" (Figure 1).

The search took place between September and October 2020, and no articles were excluded based on publication date. The aim of the search was to identify studies evaluating levels of ST2 before and after exercise. The query resulted in 105 articles on the PubMed database, 97 on ISI Web of Science and 68 on Scopus. No additional studies were found after searching the references of previous review articles.

Inclusion Criteria

Only human studies were included. It was mandatory for the studies to evaluate the levels of the selected biomarker (ST2) before and after exercise (regardless of type, intensity, or duration). Though assessing levels of this biomarker after a pre-specified period of training could be of interest, the aim of this review was to assess ST2 variation after an acute bout of exercise.

To be included in the quantitative synthesis, the study had to report the cut-off used to define the upper reference limit (normal range) used.

Exclusion criteria

This review aimed at presenting data concerning healthy (or presumably healthy) subjects. As such, articles in which individuals were selected because they had a specific pathology were excluded. Additionally, case reports as well as articles written in

languages other than English and studies containing less than ten subjects were also excluded.

Summary Measure

The main summary measure in the quantitative synthesis was the number of individuals exceeding the reference levels (as defined in the different studies) after exercise.

The number of participants in some studies was calculated from the published value corresponding to the percentage.

Quality assessment of studies and data extraction

Study quality and eligibility were individually assessed by three investigators. The investigators individually assessed if studies addressed ST2 levels after vigorous exercise, and if all inclusion/exclusion criteria were met. Primarily it was done through title and abstract analysis, and then, if abstracts were deemed acceptable, through full-text assessment. Additionally, the investigators specifically sought whether the articles described the number of individuals under study. Data extraction (see Tables 1 and 2) was individually done from the data published in the articles, and then compared by the investigators. Different opinions regarding the relevance of articles were solved by consensus between the authors.

Global article quality assessment was carried out according to the method used by Haffar and colleagues [42].

RESULTS

From title and abstract analysis, ten articles were included, and this set of articles was analyzed by the authors [24, 29, 43-50]. Two studies were subsequently excluded, as they lacked ST2 assessment both before and after exercise [47, 50]. Two other studies were excluded as they did not assess ST2 levels after an acute bout of exercise, analyzing its levels after a period of training [44, 46]. A flowchart depicting the literature search method, as well as the resulting number of articles selected, is displayed in figure 1.

A total of six articles, published between 2016 and 2020 and encompassing a total of 349 subjects in which ST2 was assessed were included [24, 29, 43, 45, 48, 49]. Table 1 presents the main characteristics of the different studies. The most common type of exercise was running, and most subjects (73%) were male. Most studies reported on increases in ST2 levels after exercise, as reported in Table 2.

Table 3 shows the number of participants with ST2 levels above the reference level (as defined in the different studies) after exercise (up to three hours after). As shown, after exercise (running in all studies in the quantitative synthesis) 92.7% exceeded this cut-off, whereas at baseline a total of 49.8% of individuals had ST2 levels above the pre-specified threshold.

The results of global article quality assessment carried out according to the method used by Haffar and colleagues [42] are presented in Supplementary File 2.

DISCUSSION

In the present report, a systematic review was undertaken to assess the current state-of-the-art concerning ST2 kinetics after exercise, a pivotal physiologic activity, among healthy individuals. Most studies reported on increases in the levels of ST2 after exercise (running in most cases), as shown in Table 2.

Physical exercise and resistance training can be associated with changes in different cardiac biomarkers, such as troponin [10, 16, 25, 51]. Since the first descriptions of exercise-induced cardiac troponin increase over thirty years ago, several underlying mechanisms have been proposed, ranging from reversible cell injury (with ensuing increased membrane permeability) to myocardial necrosis [10, 52]. Importantly, though the high range of individuals presenting elevated cardiac troponin levels (namely when assessed by high-sensitivity assays) after intense exercise as well as the kinetics of this biomarker in this setting would argue, in most individuals, in favour of a reversible phenomenon, the specific pathophysiological mechanisms underlying this observation as well as its long-term implications are still not fully specified [10, 53, 54].

Some studies have suggested that the accumulation of multiple acute bouts of high-intensity exercise during prolonged training could induce detrimental cardiac adaptations [25, 55]. Interestingly, exercise can have a distinct hemodynamic impact in the left and right ventricles, and some studies report that athletes have a higher rate of fibrosis [21, 55, 56]. However, different studies reporting increases in cardiac troponin after exercise have not shown evidence of fibrosis, when assessed by cardiac magnetic resonance imaging (MRI) [57, 58]. Interestingly, increases in galectin-3 (a marker of fibrosis) levels after exercise in the absence of myocardial fibrosis by cardiac MRI has also been reported, and postulated to be of extracardiac origin [59]. Given the role of fibrosis as a

fundamental component of the adverse structural remodeling of the myocardium in the failing heart, these complex data have garnered interest [16]. Whilst this, as recently expertly reviewed by Malek *et al.*, the significance of these findings remains to be further characterized [20]. Moreover, recent data derived from the UK Biobank cohort do not report on an increased risk of cardiovascular disease among healthy individuals undergoing vigorous exercise [60]. Additionally, a different study, also based on the UK Biobank cohort, did not show an increased risk of ventricular arrythmia among individuals performing vigorous PA [61]. As such, the overall effect of intense PA on the cardiovascular system remains a topic under active investigation.

ST2 is a promising and comprehensive biomarker, with an important role in several diseases [34, 36, 38]. ST2 has two isoforms, a transmembrane (ST2L) and a circulating (or soluble, sST2), which interact with IL-33 having protean effects, namely in terms of fibrosis and the inflammatory response [31, 32, 62]. sST2 can bind to IL-33 and limit the interaction between IL-33 and ST2L, thus hindering its beneficial effects [31, 39]. Studies have illustrated the potential of sST2 levels in terms of prognostication, namely in the setting of heart failure [33, 37, 62, 63]. Though sST2 levels vary in accordance with several factors (such as patient gender and extra-cardiac conditions), levels above 35 ng/mL have been associated with a worse prognosis in this context [31, 36, 64-66]. Given this background, assessing the ST2 response to acute exercise could provide further insights the cardiac (and overall cardiovascular) effects of exercise.

Most studies in this review reported increases in ST2 levels after exercise [24, 29, 43, 45, 48]. Increases in ST2 levels were related, in some studies, to exercise duration as well as type of PA [24, 29, 45]. In addition, when assessing studies which provided data on the number of individuals exceeding the cut-off of 35 ng/mL, a total of 92.7% of individuals exceeded this level [24, 29, 45]. Interestingly, while Le Goff *et al.* reported that ST2

levels continued to increase three hours after exercise, two studies which assessed levels further after exercise showed that these returned to baseline 48 hours and 48 hours to seven days after (respectively) [24, 43, 45]. On the other hand, some studies highlighted that an important number of subjects already displayed ST2 levels above the 35 ng/mL cut-off at baseline [24, 29, 45].

The current data, although available for only a relatively limited number of individuals, concurs as to the impact of exercise in ST2 levels, with increases after PA. This could reflect the hemodynamic stress associated with exercise (particularly in the case of intense exercise) [29, 41, 67]. Also, the notion that among this population baseline levels could exceed current cut-offs should also be mentioned, as exercise history could thus be important when interpreting these results [29]. Interestingly, prior data suggests ST2 increases in relation to diastolic load, and with a predominantly extra-myocardial origin [24, 29, 35]. As such, the mechanisms underlying the modulators of both baseline and post-exercise ST2 levels among athletes, as well as the biological significance of these adaptations and their potential clinical relevance, should be the focus of further larger long-term studies.

STUDY LIMITATIONS

There are several limitations which should be acknowledged when interpreting the present data. Firstly, the heterogeneous nature of the current data (in terms of different study designs, namely encompassing the population under study, type of exercise and timing of samples) as well as the potential for confounding factors (such as pathological issues, particularly cardiovascular disease) should be considered. Furthermore, and as highlighted in the Discussion, only a relatively small number of individuals were represented, and most of these were male. Given potential differences in ST2 physiology and in the response to exercise training in terms of gender, this point should be pondered with [31, 62, 68].

Secondly, data pertaining to imaging analysis (by means of cardiac MRI) were also not present.

Finally, studies included were of short duration, and as such the long-term significance of ST2 level variation in this setting should be the focus of further studies.

CONCLUSION

Most studies assessing ST2 levels after exercise training among healthy individuals showed increases in this biomarker. Though data were available for only a relatively small number of individuals, 92.7% had ST2 levels above the cut-off of 35 ng/mL after exercise, pointing to a rather generalized phenomenon. Baseline levels of ST2 were also above this threshold in some individuals. No studies presented cardiac magnetic resonance imaging data or assessed clinical outcomes.

Further studies are needed to assess the clinical significance, if any, as well as the long-term prognosis associated with ST2 kinetics in the setting of intense exercise. Imaging studies could play an important role in unravelling the mechanisms underlying exercise-induced ST2 level changes.

REFERENCES

1. Piepoli MF, Hoes AW, Agewall S, et al. 2016 European Guidelines on cardiovascular disease prevention in clinical practice: The Sixth Joint Task Force of the European Society of Cardiology and Other Societies on Cardiovascular Disease Prevention in Clinical Practice. Eur Heart J. 2016;37(29):2315-81.

2. Ross R, Blair SN, Arena R, et al. Importance of Assessing Cardiorespiratory Fitness in Clinical Practice: A Case for Fitness as a Clinical Vital Sign: A Scientific Statement From the American Heart Association. Circulation. 2016;134(24):653-99.

3. Sharma S, Merghani A, Mont L. Exercise and the heart: the good, the bad, and the ugly. Eur Heart J. 2015;36(23):1445-53.

4. Patel AV, Friedenreich CM, Moore SC, et al. American College of Sports Medicine Roundtable Report on Physical Activity, Sedentary Behavior, and Cancer Prevention and Control. Med Sci Sports Exerc. 2019;51(11):2391-402.

5. Warburton DE, Nicol CW, Bredin SS. Health benefits of physical activity: the evidence. CMAJ. 2006;174(6):801-9.

6. Virani SS, Alonso A, Benjamin EJ, et al. Heart Disease and Stroke Statistics-2020 Update: A Report From the American Heart Association. Circulation. 2020;141(9):139-596.

7. Fontes-Carvalho R, Azevedo AI, Sampaio F, et al. The Effect of Exercise Training on Diastolic and Systolic Function After Acute Myocardial Infarction: A Randomized Study. Medicine (Baltimore). 2015;94(36):1450-6.

8. Gevaert AB, Adams V, Bahls M, et al. Towards a personalised approach in exercise-based cardiovascular rehabilitation: How can translational research help? A 'call to action' from the Section on Secondary Prevention and Cardiac Rehabilitation of the

European Association of Preventive Cardiology. Eur J Prev Cardiol. 2020;27(13):1369-85.

9. Nieman DC, Wentz LM. The compelling link between physical activity and the body's defense system. J Sport Health Sci. 2019;8(3):201-17.

10. Vilela EM, Bastos JCC, Rodrigues RP, et al. High-sensitivity troponin after running – a systematic review. The Netherlands Journal of Medicina. 2014;72(1):5-9.

11. Campbell KL, Winters-Stone KM, Wiskemann J, et al. Exercise Guidelines for Cancer Survivors: Consensus Statement from International Multidisciplinary Roundtable. Med Sci Sports Exerc. 2019;51(11):2375-90.

12. Pelliccia A, Sharma S, Gati S, et al. 2020 ESC Guidelines on sports cardiology and exercise in patients with cardiovascular disease. Eur Heart J. 2020;0(0):1-80.

13. Vilela EM, Ladeiras Lopes R, Torres S, et al. Differential Impact of a Cardiac Rehabilitation Program on Functional Parameters in Elderly versus Non-Elderly Myocardial Infarction Survivors. Cardiology. 2020;145(2):98-105.

14. Vilela EM, Ladeiras-Lopes R, Ruivo C, et al. Different outcomes of a cardiac rehabilitation programme in functional parameters among myocardial infarction survivors according to ejection fraction. Neth Heart J. 2019;27(7-8):347-53.

15. Aengevaeren VL, Mosterd A, Sharma S, et al. Exercise and Coronary Atherosclerosis: Observations, Explanations, Relevance, and Clinical Management. Circulation. 2020;141(16):1338-50.

16. Franklin BA, Thompson PD, Al-Zaiti SS, et al. Exercise-Related Acute Cardiovascular Events and Potential Deleterious Adaptations Following Long-Term Exercise Training: Placing the Risks Into Perspective-An Update: A Scientific Statement From the American Heart Association. Circulation. 2020;141(13):705-36.

17. O'Keefe JH, Lavie CJ. Run for your life...at a comfortable speed and not too far. Heart Month. 2012;0(0):1-4.

18. Biscaglia S, Campo G, Sorbets E, et al. Relationship between physical activity and long-term outcomes in patients with stable coronary artery disease. Eur J Prev Cardiol. 2020;27(4):426-36.

19. Lechner K, Spanier B, Lechner B, et al. Your athlete-patient has a high coronary artery calcification score-'Heart of Stone'. What should you advise? Is exercise safe? Br J Sports Med. 2020;0(0):1-2.

20. Malek LA, Bucciarelli-Ducci C. Myocardial fibrosis in athletes-Current perspective. Clin Cardiol. 2020;43(8):882-8.

21. van de Schoor FR, Aengevaeren VL, Hopman MT, et al. Myocardial Fibrosis in Athletes. Mayo Clin Proc. 2016;91(11):1617-31.

22. Dhingra R, Vasan RS. Biomarkers in cardiovascular disease: Statistical assessment and section on key novel heart failure biomarkers. Trends Cardiovasc Med. 2017;27(2):123-33.

23. Thygesen K, Alpert JS, Jaffe AS, et al. Fourth Universal Definition of Myocardial Infarction (2018). J Am Coll Cardiol. 2018;72(18):2231-64.

24. Roca E, Nescolarde L, Lupon J, et al. The Dynamics of Cardiovascular Biomarkers in non-Elite Marathon Runners. J Cardiovasc Transl Res. 2017;10(2):206-8.

25. Eijsvogels TM, Fernandez AB, Thompson PD. Are There Deleterious Cardiac Effects of Acute and Chronic Endurance Exercise? Physiol Rev. 2016;96(1):99-125.

26. Tulloh L, Robinson D, Patel A, et al. Raised troponin T and echocardiographic abnormalities after prolonged strenuous exercise--the Australian Ironman Triathlon. Br J Sports Med. 2006;40(7):605-9.

27. Vilela EM, Bettencourt-Silva R, Nunes JP, et al. BNP and NT-proBNP elevation after running--a systematic review. Acta Cardiol. 2015;70(5):501-9.

28. Knechtle B, Nikolaidis PT. Physiology and Pathophysiology in Ultra-Marathon Running. Front Physiol. 2018;9(634):1-33.

29. Aengevaeren VL, RRJ VANK, Hopman MTE, et al. Exercise-induced Changes in Soluble ST2 Concentrations in Marathon Runners. Med Sci Sports Exerc. 2019;51(3):405-10.

30. Griesenauer B, Paczesny S. The ST2/IL-33 Axis in Immune Cells during Inflammatory Diseases. Front Immunol. 2017;8:475.

31. Nah EH, Cho S, Kim S, et al. Reference interval and the role of soluble suppression of tumorigenicity 2 (sST2) in subclinical cardiac dysfunction at health checkups. J Clin Lab Anal. 2020;34(11):e23461.

32. Pascual-Figal DA, Januzzi JL. The biology of ST2: the International ST2 Consensus Panel. Am J Cardiol. 2015;115(7 Suppl):3B-7B.

33. van Vark LC, Lesman-Leegte I, Baart SJ, et al. Prognostic Value of Serial ST2 Measurements in Patients With Acute Heart Failure. J Am Coll Cardiol. 2017;70(19):2378-88.

34. Homsak E, Gruson D. Soluble ST2: A complex and diverse role in several diseases. Clin Chim Acta. 2020;507:75-87.

35. Bartunek J, Delrue L, Van Durme F, et al. Nonmyocardial production of ST2 protein in human hypertrophy and failure is related to diastolic load. J Am Coll Cardiol. 2008;52(25):2166-74.

36. Suthahar N, Meems LMG, Ho JE, et al. Sex-related differences in contemporary biomarkers for heart failure: a review. Eur J Heart Fail. 2020;22(5):775-88.

37. Aimo A, Januzzi JL, Jr., Bayes-Genis A, et al. Clinical and Prognostic Significance of sST2 in Heart Failure: JACC Review Topic of the Week. J Am Coll Cardiol. 2019;74(17):2193-203.

38. Aimo A, Vergaro G, Passino C, et al. Prognostic Value of Soluble Suppression of Tumorigenicity-2 in Chronic Heart Failure: A Meta-Analysis. JACC Heart Fail. 2017;5(4):280-6.

39. Bayes-Genis A, de Antonio M, Vila J, et al. Head-to-head comparison of 2 myocardial fibrosis biomarkers for long-term heart failure risk stratification: ST2 versus galectin-3. J Am Coll Cardiol. 2014;63(2):158-66.

40. Pearson MJ, King N, Smart NA. Effect of exercise therapy on established and emerging circulating biomarkers in patients with heart failure: a systematic review and meta-analysis. Open Heart. 2018;5(2):e000819.

41. Le Goff C, Farre Segura J, Dufour P, et al. Intense sport practices and cardiac biomarkers. Clin Biochem. 2020;79:1-8.

42. Haffar S, Shalimar, Kaur RJ, et al. Acute liver failure caused by hepatitis E virus genotype 3 and 4: A systematic review and pooled analysis. Liver Int. 2018;38(11):1965-73.

43. Bekos C, Zimmermann M, Unger L, et al. Non-professional marathon running: RAGE axis and ST2 family changes in relation to open-window effect, inflammation and renal function. Sci Rep. 2016;6:32315.

44. Galliera E, Lombardi G, Marazzi MG, et al. Acute exercise in elite rugby players increases the circulating level of the cardiovascular biomarker GDF-15. Scand J Clin Lab Invest. 2014;74(6):492-9.

45. Le Goff C, Kaux JF, Farre Segura J, et al. Evolution of the slopes of ST2 and galectin-3 during marathon and ultratrail running compared to a control group. Clin Chem Lab Med. 2020;58(2):314-21.

46. Sponder M, Lichtenauer M, Wernly B, et al. Serum heart-type fatty acid-binding protein decreases and soluble isoform of suppression of tumorigenicity 2 increases significantly by long-term physical activity. J Investig Med. 2019;67(5):833-40.

47. Agoston-Coldea L, Lupu S, Hicea S, et al. Serum levels of the soluble IL-1 receptor family member ST2 and right ventricular dysfunction. Biomarkers Medicine. 2014;8(1):95-106.

48. Ho JE, Shi L, Day SM, et al. Biomarkers of cardiovascular stress and fibrosis in preclinical hypertrophic cardiomyopathy. Open Heart. 2017;4(2):615-23.

49. Sanz-de la Garza M, Iannino N, Finnerty V, et al. Cardiopulmonary, biomarkers, and vascular responses to acute hypoxia following cardiac transplantation. Clin Transplant. 2018;32(9):e13352.

50. Zheng YG, Yang T, He JG, et al. Plasma soluble ST2 levels correlate with disease severity and predict clinical worsening in patients with pulmonary arterial hypertension. Clin Cardiol. 2014;37(6):365-70.

51. Shave R, Baggish A, George K, et al. Exercise-induced cardiac troponin elevation: evidence, mechanisms, and implications. J Am Coll Cardiol. 2010;56(3):169-76.

52. Airaksinen KEJ. Cardiac Troponin Release After Endurance Exercise: Still Much to Learn. J Am Heart Assoc. 2020;9(4):1-3.

53. Aengevaeren VL, Hopman MTE, Thompson PD, et al. Exercise-Induced Cardiac Troponin I Increase and Incident Mortality and Cardiovascular Events. Circulation. 2019;140(10):804-14.

54. Gresslien T, Agewall S. Troponin and exercise. Int J Cardiol. 2016;221:609-21.

55. La Gerche A, Burns AT, Mooney DJ, et al. Exercise-induced right ventricular dysfunction and structural remodelling in endurance athletes. Eur Heart J. 2012;33(8):998-1006.

56. Breuckmann F, Mohlenkamp S, Nassenstein K, et al. Myocardial Late Gadolinium Enhancement: Prevalence, Pattern, and Prognostic Relevance in Marathon Runners. Radiology. 2009;251(1):50-7.

57. Hanssen H, Keithahn A, Hertel G, et al. Magnetic resonance imaging of myocardial injury and ventricular torsion after marathon running. Clin Sci (Lond). 2011;120(4):143-52.

58. Trivax JE, Franklin BA, Goldstein JA, et al. Acute cardiac effects of marathon running. J Appl Physiol (1985). 2010;108(5):1148-53.

59. Hattasch R, Spethmann S, de Boer RA, et al. Galectin-3 increase in endurance athletes. Eur J Prev Cardiol. 2014;21(10):1192-9.

60. Ramakrishnan R, Doherty A, Smith-Byrne K, et al. Accelerometer measured physical activity and the incidence of cardiovascular disease: Evidence from the UK Biobank cohort study. PLoS Med. 2021;18(1):e1003487.

61. Elliott AD, Linz D, Mishima R, et al. Association between physical activity and risk of incident arrhythmias in 402 406 individuals: evidence from the UK Biobank cohort. Eur Heart J. 2020;41(15):1479-86.

62. Sharim J, Daniels LB. Soluble ST2 and Soluble Markers of Fibrosis: Emerging Roles for Prognosis and Guiding Therapy. Curr Cardiol Rep. 2020;22(6):41.

63. Wu AH, Wians F, Jaffe A. Biological variation of galectin-3 and soluble ST2 for chronic heart failure: implication on interpretation of test results. Am Heart J. 2013;165(6):995-9.

64. Mueller T, Dieplinger B. Soluble ST2 and galectin-3: what we knowand don't know analytically. eJIFCC. 2016;3(3):224-37.

65. Nishimura M, Brann A, Chang KW, et al. The Confounding Effects of Non-cardiac Pathologies on the Interpretation of Cardiac Biomarkers. Curr Heart Fail Rep. 2018;15(4):239-49.

66. Watson CJ, Tea I, O'Connell E, et al. Comparison of longitudinal change in sST2 vs BNP to predict major adverse cardiovascular events in asymptomatic patients in the community. J Cell Mol Med. 2020;24(11):6495-9.

67. Mirna M, Lichtenauer M, Wernly B, et al. Novel cardiovascular biomarkers in patients with cardiovascular diseases undergoing intensive physical exercise. Panminerva Med. 2020;62(3):135-42.

68. Witvrouwen I, Van Craenenbroeck EM, Abreu A, et al. Exercise training in women with cardiovascular disease: Differential response and barriers - review and perspective. Eur J Prev Cardiol. 2019:2047487319838221.

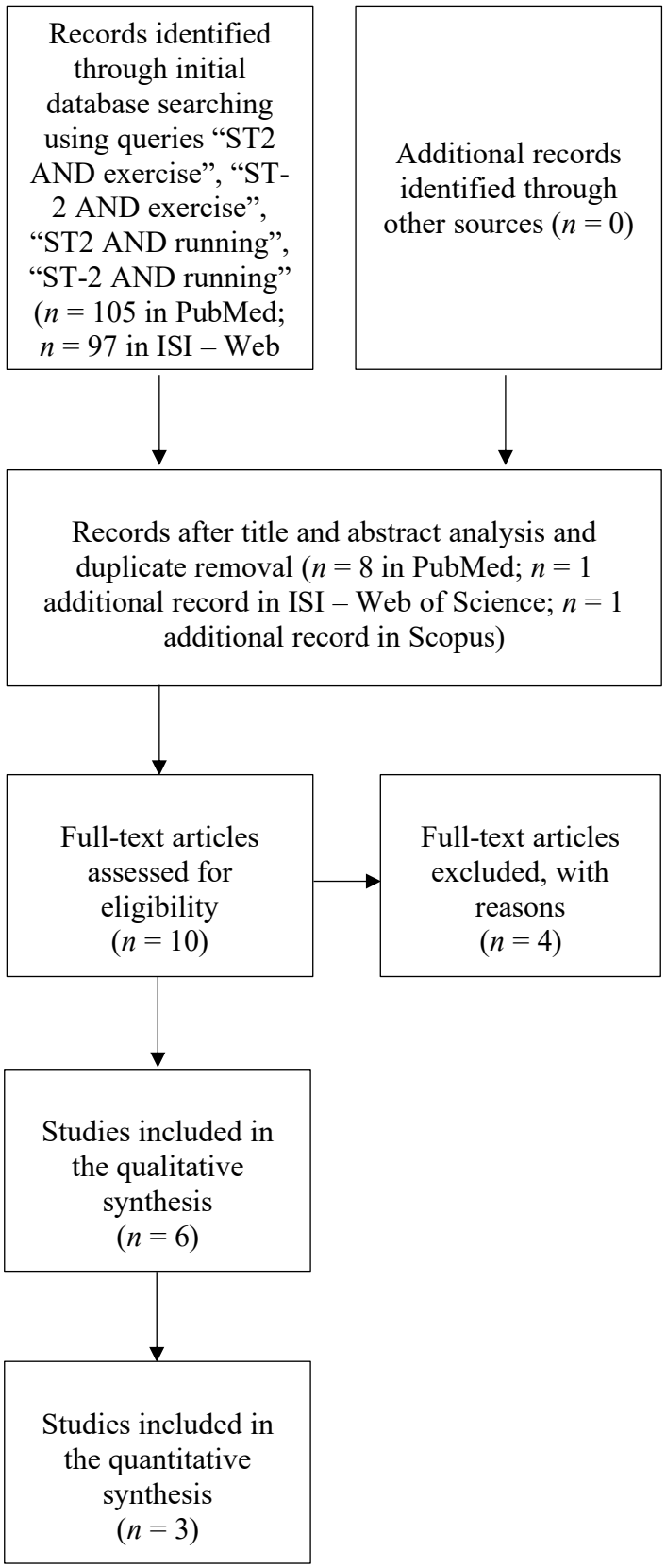

Figure 1. Flowchart showing the literature search method.

Legend: *n* = number

Table 1. Overview of the selected studies.

Study (year)	***n***	**Male/female ratio**	**Mean age (years)**	**Type of exercise**	**Biomarkers assessed**	**Time of sample collection**
Bekos *et al.* (2016) [43]	34; 36	24/10; 19/17	36.8; 36.9	Running different lengths: marathon, half-marathon	ST2 CRP CK, CK-MB CK18 GGT HMGB1 IL-1RA IL-33 Lactate Na, creatinine RAGE RBC, WBC, platelets	Before, immediately after and 2-7 days after the race
Roca *et al.* (2017) [24]	79	57/22	39	Running (marathon)	ST2 CK hsTnT NT-proBNP	Before, after (1-2 hours) and 48 hours after the race
Ho *et al.* (2017) [48]	41	17/24	17	Maximal exercise test (treadmill)	ST2 BAP CITP Gal-3 hsTnI Periostin PICP	Baseline, just after and 4 hours after exercise

					NT-proBNP	
Sanz-de la Garza *et al.* (2018) [49]	17	13/4	50.0	Maximal exercise test (bicycle ergometer)	ST2 hs-IL-6 VEGF	Baseline and just after exercise
Aengevaeren *et al.* (2019) [29]	82	65/17	48	Running (marathon)	ST2 hematocrit*, Hg* hsTnI	Before and after the race
Le Goff *et al.* (2020) [45]	14; 19; 27	60/0	37.7; 42.0; 45.3	Running different lengths: 1h (control), *circa* 4h (marathon, 42.195km), *circa* 8h (ultra-marathon, 67km)	ST2 CK-MB hs-CRP Gal-3 hematocrit, Hg hsTnT NT-proBNP	Before, just after and 3 hours after the race

Legend: *BAP* = bone alkaline phosphatase; *CITP* = C-terminal telopeptide of type I collagen; *CRP* = C-reactive protein; hs-*CRP* = high-sensitivity C-reactive protein; *CK* = creatine kinase; *CK18* = cytokeratin 18; *CK-MB* = creatine kinase isoform MB; *HMGB1* = high mobility group box 1; *Hg* = hemoglobin; *Gal-3* = galectin-3; *GGT* = gama-glutamil transferase; *IL-1RA* = interleukin-1 receptor antagonist*; hs-IL6* = high-sensitivity interleukin-6; *IL-33* = interleukin-33*; n* = number of individuals; *Na* = sodium; *NT-proBNP* = N-terminal pro-hormone of brain natriuretic peptide; *PICP* = C-terminal propeptide of procollagen type I; *RAGE* = receptor of advanced glycation endproducts; *RBC* = red blood cells; ST2 = suppression of tumorigenicity 2 protein; *hsTnI* = high-sensitivity troponin I; *hsTnT* = high-sensitivity troponin T; *VEGF* = vascular endothelial growth factor; *WBC* = white blood cells; * = determined in a subset of individuals

Table 2. Major findings concerning ST2, in the six articles included in the systematic review.

Study (year)	**Major findings**
Bekos *et al.* (2016) [43]	ST2 increased significantly after a marathon, returning to baseline levels after 2 to 7 days of recovery.
Roca *et al.* (2017) [24]	ST2 levels above the cut-off (35 ng/dL) in 48.7% of individuals at baseline. ST2 increased significantly immediately after a marathon, returning to baseline levels 48 hours post-race. ST2 elevations directly related to race time.
Ho *et al.* (2017) [48]	ST2 levels increased 4 hours after exercise.
Sanz-de la Garza *et al.* (2018) [49]	No difference in ST2 levels after exercise.
Aengevaeren *et al.* (2019) [29]	ST2 levels above the cut-off (35 ng/dL) in 48% of individuals at baseline. ST2 levels were higher in faster runners.
Le Goff *et al.* (2019) [45]	ST2 levels above the cut-off (35 ng/dL) at baseline in 71.4% of controls, 63.2% of marathon runners and 40.7% of ultra-marathon runners. ST2 levels increased significantly immediately after running and continued to increase at the three-hour mark.

Table 3. Number of participants with ST2 levels above the reference level (as defined in the different studies) after exercise (up to three hours).

Roca *et al.* (2017) [24]	69 / 79
Aengevaeren *et al.* (2019) [29]	75 / 80
Le Goff *et al.* (2020) [45]	13 / 14 (control) 19 / 19 (marathon) 27 / 27 (ultra-marathon)
Total	203 / 219 (92.7%)

ATTACHMENTS

Supplementary file 1

Authors	Did the patient(s) represent the whole case(s) of the medical center?	Was the diagnosis correctly made?	Were other important diagnosis excluded?	Were all important data cited in the report?	Was the outcome correctly ascertained?	**Global quality assessment**
Bekos et al. (2016)	No	Yes	Yes	Yes	Yes	Moderate
Roca et al. (2017)	Yes	Yes	Yes	Yes	Yes	Good
Ho et al. (2017)	Yes	Yes	Yes	Yes	Yes	Good
Sanz-de la Garza et al. (2018)	Yes	Yes	Yes	Yes	Yes	Good
Aengevaeren et al.	No	Yes	Yes	Yes	Yes	Moderate
Le Goff et al. (2020)	Yes	Yes	Yes	Yes	Yes	Good

Supplementary file 2: full search details for Medline (Pubmed) database.

The search on the Medline (Pubmed) database was done between September and October 2020, using the following queries (individually): “ST2 AND exercise”; “ST-2 AND exercise”; “ST2 AND running”; “ST-2 AND running” (see below detailed queries).

The aim of the database search was to identify studies assessing the ST2 levels after vigorous exercise, and no articles were excluded based on publication date (see main document for further inclusion and exclusion criteria). This search resulted in a total of 105 articles (query “ST2 AND exercise” 53 articles; “ST-2 AND exercise” 38 articles; “ST2 AND running” 8 articles; “ST-2 AND running” 6 articles).

The abstracts were then individually assessed by three investigators (regarding study quality and eligibility), and duplicates (same study present in more than one of the queries) were removed. After title and abstract analysis, the full-text articles were then included for analysis (please refer to the Results section and Figure 1 of the main document for specification).

User query using pre-defined search terms

ST2 AND exercise

Query translation

"st2"[All Fields] AND ("exercise"[MeSH Terms] OR "exercise"[All Fields] OR "exercises"[All Fields] OR "exercise therapy"[MeSH Terms] OR ("exercise"[All Fields] AND "therapy"[All Fields]) OR "exercise therapy"[All Fields] OR "exercise s"[All Fields] OR "exercised"[All Fields] OR "exerciser"[All Fields] OR "exercisers"[All Fields] OR "exercising"[All Fields])

Individual Translations

exercise: "exercise"[MeSH Terms] OR "exercise"[All Fields] OR "exercises"[All Fields] OR "exercise therapy"[MeSH Terms] OR ("exercise"[All Fields] AND "therapy"[All Fields]) OR "exercise therapy"[All Fields] OR "exercise's"[All Fields] OR "exercised"[All Fields] OR "exerciser"[All Fields] OR "exercisers"[All Fields] OR "exercising"[All Fields]

User query using pre-defined search terms

ST-2 AND exercise

Query translation

"st-2"[All Fields] AND ("exercise"[MeSH Terms] OR "exercise"[All Fields] OR "exercises"[All Fields] OR "exercise therapy"[MeSH Terms] OR ("exercise"[All Fields] AND "therapy"[All Fields]) OR "exercise therapy"[All Fields] OR "exercise s"[All Fields] OR "exercised"[All Fields] OR "exerciser"[All Fields] OR "exercisers"[All Fields] OR "exercising"[All Fields])

Individual Translations

exercise: "exercise"[MeSH Terms] OR "exercise"[All Fields] OR "exercises"[All Fields] OR "exercise therapy"[MeSH Terms] OR ("exercise"[All Fields] AND "therapy"[All Fields]) OR "exercise therapy"[All Fields] OR "exercise's"[All Fields] OR "exercised"[All Fields] OR "exerciser"[All Fields] OR "exercisers"[All Fields] OR "exercising"[All Fields]

User query using pre-defined search terms

ST2 AND running

Query translation

"st2"[All Fields] AND ("running"[MeSH Terms] OR "running"[All Fields] OR "runnings"[All Fields])

Individual Translations

running: "running"[MeSH Terms] OR "running"[All Fields] OR "runnings"[All Fields]

User query using pre-defined search terms

ST-2 AND running

Query translation

"st-2"[All Fields] AND ("running"[MeSH Terms] OR "running"[All Fields] OR "runnings"[All Fields])

Individual Translations

running: "running"[MeSH Terms] OR "running"[All Fields] OR "runnings"[All Fields]

AGRADECIMENTOS

No término deste trabalho, gostaria de expressar um agradecimento a todos os envolvidos, que possibilitarem a sua realização.

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Um agradecimento final à minha família, em especial, mãe, pai e avó por sempre acreditarem em mim e por todo o apoio. Ao meu João pelo companheirismo de uma vida, que sempre esteve e estará ao meu lado.

O meu sincero obrigada a todos!

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- Competing interests
- Funding
- Authors' contributions
- Acknowledgements

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Article with organization as author

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Books

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BjÃ¸rn Lomborg, ed. RethinkHIV - Smarter ways to invest in ending HIV in Sub-Saharan Africa. Cambridge: Cambridge University Press; 2012.
Meltzer PS, Kallioniemi A, Trent JM. Chromosome alterations in human solid tumors. In: Vogelstein B, Kinzler KW, eds. The genetic basis of human cancer. New York, NY: McGraw-Hill; 2002. pp 93-113.

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PRISMA 2009 Checklist

Section/topic	#	Checklist item	Reported on page and paragraph/ table #
TITLE			
Title	1	Identify the report as a systematic review, meta-analysis, or both.	Page 1 “Suppression of tumorigenicity 2 after exercise: a systematic review”
ABSTRACT			
Structured summary	2	Provide a structured summary including, as applicable: background; objectives; data sources; study eligibility criteria, participants, and interventions; study appraisal and synthesis methods; results; limitations; conclusions and implications of key findings; systematic review registration number.	Page 3 “Exercise is a pivotal physiological activity (...) should target this.”
INTRODUCTION			
Rationale	3	Describe the rationale for the review in the context of what is already known.	Page 5 “Notably, some studies have shown the potential of ST2 in heart failure, further reinforcing its clinical importance. Given this background, the profile of ST2 kinetics among athletes could further improve the current knowledge underlying cardiac adaptations (and potential maladaptations) in the face of exercise training.”
Objectives	4	Provide an explicit statement of questions being addressed with reference to participants, interventions, comparisons, outcomes, and study design (PICOS).	Page 5 “This review aimed at addressing the impact of exercise in ST2 levels among healthy individuals, and to present a pragmatic appraisal of the current literature on this field. This is, to the best of our knowledge, the first systematic review on this specific topic and could thus further improve current knowledge on the kinetics of different cardiac biomarkers after exercise, a topic of considerable translational relevance.”
METHODS			
Protocol and registration	5	Indicate if a review protocol exists, if and where it can be accessed (e.g., Web address), and, if available, provide registration information including registration number.	Not available
Eligibility criteria	6	Specify study characteristics (e.g., PICOS, length of follow-up) and report characteristics (e.g., years considered, language, publication status) used as criteria for eligibility, giving rationale.	Page 6 “Only human studies were included. It was mandatory for the studies to evaluate the levels of the selected biomarker (ST2) before and after exercise (regardless of type, intensity, or duration). Though assessing levels of this biomarker after a pre-specified period of training could be of interest, the aim of this

			review was to assess ST2 variation after an acute bout of exercise. To be included in the quantitative synthesis, the study had to report the cut-off used to define the upper reference limit (normal range) used."
Information sources	7	Describe all information sources (e.g., databases with dates of coverage, contact with study authors to identify additional studies) in the search and date last searched.	Page 6 "The search took place between September and October 2020, and no articles were excluded based on publication date. The aim of the search was to identify studies evaluating levels of ST2 before and after exercise. The query resulted in 105 articles on the PubMed database, 97 on ISI Web of Science and 68 on Scopus. No additional studies were found after searching the references of previous review articles."
Search	8	Present full electronic search strategy for at least one database, including any limits used, such that it could be repeated.	Page 6 "The study started with a search on three databases, Medline (PubMed), ISI Web of Science and Scopus, using the queries "ST2" or "ST-2" + "exercise" or "running"."
Study selection	9	State the process for selecting studies (i.e., screening, eligibility, included in systematic review, and, if applicable, included in the meta-analysis).	Page 22
Data collection process	10	Describe method of data extraction from reports (e.g., piloted forms, independently, in duplicate) and any processes for obtaining and confirming data from investigators.	Page 7
Data items	11	List and define all variables for which data were sought (e.g., PICOS, funding sources) and any assumptions and simplifications made.	Page 7 "The main summary measure in the quantitative synthesis was the number of individuals exceeding the reference levels (as defined in the different studies) after exercise."
Risk of bias in individual studies / Risk of bias across studies	12/ 15	Describe methods used for assessing risk of bias of individual studies (including specification of whether this was done at the study or outcome level), and how this information is to be used in any data synthesis.	Page 7 "Study quality and eligibility were individually assessed by three investigators. Different opinions regarding the relevance of articles were solved by consensus between the authors."
Summary measures	13	State the principal summary measures (e.g., risk ratio, difference in means).	Non applicable, since this review does not include a meta-analysis
Synthesis of results	14	Describe the methods of handling data and combining results of studies, if done, including measures of consistency (e.g., I^2) for each meta-analysis.	Non applicable, since this review does not include a meta-analysis
Additional analyses	16	Describe methods of additional analyses (e.g., sensitivity or subgroup analyses, meta-regression), if done, indicating which were pre-specified.	Non applicable, since this review does not include a meta-analysis
RESULTS			

Study selection	17	Give numbers of studies screened, assessed for eligibility, and included in the review, with reasons for exclusions at each stage, ideally with a flow diagram.	Page 22
Study characteristics	18	For each study, present characteristics for which data were extracted (e.g., study size, PICOS, follow-up period) and provide the citations.	Pages 23 and 24
Risk of bias within and across studies	19/ 22	Present data on risk of bias of each study and, if available, any outcome level assessment (see item 12).	Pages 25 and 26
Results of individual studies	20	For all outcomes considered (benefits or harms), present, for each study: (a) simple summary data for each intervention group (b) effect estimates and confidence intervals, ideally with a forest plot.	Non applicable, since this review does not include a meta-analysis
Synthesis of results	21	Present results of each meta-analysis done, including confidence intervals and measures of consistency.	Non applicable, since this review does not include a meta-analysis
Additional analysis	23	Give results of additional analyses, if done (e.g., sensitivity or subgroup analyses, meta-regression [see Item 16]).	Non applicable, since this review does not include a meta-analysis
DISCUSSION			
Summary of evidence	24	Summarize the main findings including the strength of evidence for each main outcome; consider their relevance to key groups (e.g., healthcare providers, users, and policy makers).	Page 11 “The current data (…) should be the focus of further larger long-term studies.”
Limitations	25	Discuss limitations at study and outcome level (e.g., risk of bias), and at review-level (e.g., incomplete retrieval of identified research, reporting bias).	Page 12 “There are several limitations (...) significance of ST2 level variation in this setting should be the focus of further studies.”
Conclusions	26	Provide a general interpretation of the results in the context of other evidence, and implications for future research.	Page 13 “Further studies are needed to assess the clinical significance, if any, as well as the long- term prognosis associated with ST2 kinetics in the setting of intense exercise. Imaging studies could play an important role in unravelling the mechanisms underlying exercise- induced ST2 level changes.”
FUNDING			
Funding	27	Describe sources of funding for the systematic review and other support (e.g., supply of data); role of funders for the systematic review.	Page 2 “Supportive foundations: none.”

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