THE EFFECT OF ACUTE EXERCISE ON SERUM BRAIN-DERIVED NEUROTROPHIC FACTOR LEVELS IN PATIENTS WITH CHRONIC HEART FAILURE

TRABAJO FIN DE MASTER

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El efecto del ejercicio agudo sobre los niveles séricos del factor neurotrófico derivado del cerebro en pacientes con insuficiencia cardíaca crónica

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INTRODUCTION

Chronic Heart Failure (CHF) is a complex and potentially life-threatening condition, often a consequence of untreated or poorly managed heart disease. CHF significantly impacts patient quality of life and adds to morbidity and mortality rates worldwide. Recent estimates suggest that while the number of new cases of CHF has plateaued or even decreased in developed countries, more people are living with CHF today due to the ageing population and improved survival for patients affected by cardiovascular disease. In fact, according to the most recent data, over 64 million individuals worldwide are affected by CHF (Groenewegen et al., 2020; Savarese et al., 2023).

Accordingly, there arises a pressing need for cost-effective treatment and management strategies. In this sense, current guidelines emphasize the non-pharmacological approach and underscore lifestyle change as an integral part of disease management (Piepoli et al., 2010; Visseren et al., 2021). Exercise-based cardiac rehabilitation is one such change. Physical activity in general, but especially aerobic exercise, is associated with reductions in all-cause and cardiovascular mortality in the general population (Myers et al., 2015; Nocon et al., 2008). In patients already diagnosed with heart conditions, including heart failure, exercise reduces hospitalization rates, improves symptom management, and increases exercise tolerance (Sanchis-Gomar et al., 2022).

Understanding the mechanisms behind exercise-induced protection may reinforce the therapeutic potential of exercise training for patients with CHF. It may also aid in tailoring training programs to individuals (Sanchis-Gomar et al., 2022). In the last two decades, it has been suggested that the beneficial effects of exercise, at least to a degree, may be attributed to the release of certain molecules known as "myokines." These myokines, including peptides and metabolites, are secreted by bodily tissues in response to exercise. Upon their release into circulation, myokines orchestrate a myriad of immediate molecular events that further contribute to the well-known long-term adaptations to exercise (Severinsen & Pedersen, 2020).

Among these myokines, one that stands out is the brain-derived neurotrophic factor (BDNF). BDNF, a neurotrophin primarily found in the hippocampus, has been extensively studied for its role in synaptic plasticity (Edelmann et al., 2014). In the brain, BDNF exerts its influence by binding to the tropomyosin receptor kinase B (TrkB) (Yoshii & Constantine-Paton, 2010). A significant body of research underscores the pivotal role of the BDNF/TrkB signalling in neural development, memory consolidation, and neurogenesis (Park & Poo, 2013). Notably, numerous studies have reported alterations in BDNF expression in various neurological conditions, including Alzheimer's (Holsinger et al., 2000), Parkinson's (Jin, 2020), and psychiatric disorders (Cattaneo et al., 2016).

However, in recent years, the involvement of BDNF in a pleiotropy of physiological processes, including cardiovascular function (Kermani & Hempstead, 2019; Pius-Sadowska & Machaliński, 2017), has been proposed. Evidence suggests the implication of BDNF/TrkB signalling in enhanced cardiac contractility and neovascularization following injury (Feng et al., 2015; Kermani & Hempstead, 2019). Moreover, overexpression of BDNF has resulted in increased capillary density, suggesting a potential role as a pro-angiogenic factor (Donovan et al., 2000; Hong et al., 2014).

Within the circulatory system, BDNF is highly expressed by platelets, which may explain the rapid release of BDNF into serum following platelet aggregation (Chacón-Fernández et al., 2016; Matthews et al., 2009). Additionally, endothelial cells in the vasculature and skeletal muscle also express BDNF and TrkB. In fact, BDNF concentration rises in response to muscle contraction, although muscle-derived BDNF does appear to function in an autocrine or paracrine fashion only (Matthews et al., 2009). Physical activity, particularly aerobic exercise, has been shown to increase circulating BDNF (Knaepen et al., 2010). This exercise-induced increase in

BDNF has been demonstrated in healthy young (Dinoff et al., 2017) and older adults (Coelho et al., 2013). More importantly, even a single session of aerobic exercise was able to raise BDNF concentrations in patients with Alzheimer's (Coelho et al., 2014), Parkinson's (Azevedo et al., 2022), and obesity (Inoue et al., 2020).

Although the literature on CHF is still scarce, correlational studies have put forward the notion that BDNF may be used as a biomarker to assess the severity and progression of CHF (Fukushima et al., 2015; Nakano et al., 2020). Moreover, BDNF levels were lower in patients with CHF compared to healthy subjects and independently predicted future cardiovascular events (Kadowaki et al., 2016). In individuals with underlying heart disease, lower BDNF levels were associated with all-cause mortality and future coronary events (Jiang et al., 2011).

However, the potential of exercise to restore BDNF levels and produce clinical benefits as observed in other pathologies, remains unexplored in the context of CHF. In a study by Nakano et al. (2020), serum BDNF levels were measured in patients during cardiopulmonary exercise testing (CPET) but only at rest, and the immediate effect of exercise on BDNF levels was not examined. Thus, to the best of our knowledge, no prior research has investigated the acute effect of exercise on serum BDNF concentrations in patients with CHF. The potential implications of these findings could significantly contribute to our understanding of the role of exercise in managing CHF.

It is also worth noting that the change in BDNF levels following exposure to exercise stimuli depends on the individual patient characteristics and variables of the exercise prescription (Piepmeier & Etnier, 2015). Research to date has indicated that BDNF content tends to be lower in females, which may be attributed to hormonal variances (Chan & Ye, 2017). Along the same line of evidence, in numerous exercise studies, it has been observed that males typically exhibit greater acute increases in BDNF compared to females (Dinoff et al., 2017). In patients with CHF, serum BDNF levels drop even further as the disease progresses, revealing a strong correlation with compromised exercise capacity and skeletal muscle function (Nakano et al., 2020). Moreover, it was previously suggested that BDNF release following an exercise bout is intensity-dependent (Ferris et al., 2007). In this sense, lactate, the metabolite of anaerobic metabolism, was recognized as a trigger of cellular pathways, culminating in increased BDNF expression (Hayek et al., 2019). Hence, higher-intensity exercise, which strains the glycolytic energy system, is associated with larger increases in BDNF levels (Reycraft et al., 2020; Saucedo Marquez et al., 2015).

Therefore, the objectives of the present study were threefold. On the one hand, we sought to examine BDNF levels in patients with CHF to further contrast these levels against those reported in the literature in healthy individuals and other clinical populations. At the same time, we investigated the immediate impact of aerobic exercise on serum BDNF levels in male and female patients with CHF. Finally, we examined the correlations between individual patient characteristics (such as age, sex, and left ventricular ejection fraction [LVEF]), CPET variables (including duration and peak workload), and the post-exercise increase in serum BDNF concentrations. We speculated that baseline BDNF levels in patients with CHF would be lower compared to those in healthy individuals but similar to levels found in other pathological conditions. For our next objective, we hypothesized that exercise would increase BDNF levels in CHF patients. We anticipated slight variations, favouring male subjects and those with less deteriorated left ventricular function. Additionally, we expected individuals with higher lactate concentrations post-exercise to experience a more significant rise in serum BDNF.

METHOD

Participants

Participants were recruited from the Cardiology Department's database at General University Hospital of Alicante Dr. Balmis. Eligible individuals were subsequently contacted via telephone calls, and their participation was entirely voluntary. The inclusion criteria were as follows: (a) male and post-menopausal female patients diagnosed with CHF, with stable symptoms and medical treatment for at least the past three months, regardless of the left ventricular ejection fraction (LVEF) classification; (b) aged 18 years or older; (c) diagnosis of CHF of ischemic or non-ischemic origin; (d) New York Heart Association functional class II-III; (e) no physical limitations hindering exercise. Exclusion criteria included (a) unstable angina pectoris; (b) myocardial infarction within the last six months; (c) severe or acute exacerbation of chronic obstructive pulmonary disease; (d) presence of complex ventricular arrhythmias. Experienced cardiologists meticulously verified both inclusion and exclusion criteria.

Study design

The study protocol received approval from the Ethics Committee of the hospital (Ref: 2022-140). Patients meeting the inclusion criteria were instructed to avoid strenuous physical activity in the 24 hours preceding the intervention. The intervention, which included a blood draw and CPET, was scheduled for the afternoon (between 4 and 7 pm). On the day of the CPET, patients received detailed information about the study and were invited to sign the informed consent form.

Prior to exercise, patients' weight and height were measured to calculate their body mass index (BMI). Subsequently, all patients underwent a symptom-limited CPET on a cycle ergometer. Blood samples were collected from the antecubital vein before and after the exercise test to quantify serum BDNF concentrations, while capillary blood samples were obtained from the earlobe to measure blood lactate concentration.

Cardiopulmonary exercise test

All patients performed a symptom-limited CPET on a cycle ergometer (SanaBike 500 easy, Truchtelfinger, Germany) under medical supervision. The CPET consisted of the following phases: (a) a 5-minute baseline period in the seated position on the cycle ergometer; (b) a 3-minute warm-up at 10 W; (c) an incremental ramp test with a personalized slope aimed to induce exhaustion in 8-12 minutes (Neder et al., 1999), with a pedalling frequency set between 60 and 70 rpm; (d) a 5-minute cool-down at 10 W.

Systolic and diastolic blood pressures were monitored during exercise, with readings taken every three minutes. Respiratory gas exchange and heart rate (HR) were continuously monitored during the test using Metalyzer 3B (CORTEX Biophysik GmbH, Leipzig, Germany) and a 12-lead electrocardiogram (Norav PC-ECG 1200, Mainz-Kastel, Germany), respectively. Ventilatory data were averaged every 10 seconds, and peak oxygen uptake (VO_2 peak) was defined as the highest mean VO_2 value reached at the end of the incremental phase. The first ventilatory threshold was established according to the V-slope method (Wasserman et al., 1994). To confirm the maximal effort during the exercise test and ensure the VO_2 peak represented VO_2 max, the criterion of the end-incremental phase peak respiratory exchange rate (RER peak) being greater than 1.10 was utilized (Mezzani et al., 2009). Peripheral and capillary blood samples were collected during both the baseline and cool-down periods.

Blood samples and analyses

A qualified nurse drew peripheral blood samples via an indwelling catheter located in the antecubital vein before and after CPET. A blood sample was collected in a dry tube with gelose, kept at room temperature for 30 minutes, and centrifuged at 1000 x g for 15 minutes. The serum

samples were harvested, aliquoted, and stored at −20 ºC for subsequent analysis of all samples altogether. The levels of serum BDNF were measured by ELISA testing according to the manufacturer's protocol.

Sample size determination and statistical analysis

G*Power was used to determine the required number of participants (Faul et al., 2009), with input variables derived from prior research (Ferris et al., 2007). The sample size was computed using a two-tailed paired *t*-test, with a significance level set at 5%, a target power of 80%, and an anticipated effect size (*d*) of 0.89. The calculation indicated a need for 12 participants, and ultimately, 12 individuals were recruited.

The Shapiro-Wilk test and box plots were utilized to assess the normality of the continuous variables. Variables with a normal distribution were expressed as mean ± standard deviation (*SD*). Conversely, variables that did not meet the assumption of normality were represented as median (25^{th} and 75^{th} percentiles). Additionally, categorical variables were reported as frequency (percentage).

Paired *t*-tests were employed to perform statistical within-group comparisons (i.e., before and after the CPET) for lactate and BDNF concentrations. The mean change, along with its 95% confidence interval (*CI*), was calculated to assess the magnitude of change in the selected outcomes. The Mann-Whitney U test was applied to determine if changes in BDNF differed based on sex (i.e., males vs. females) or the attainment of VO_2 max during CPET (i.e., confirmed vs. non-confirmed). Finally, Spearman´s rho correlation analyses were conducted to examine the relationship between changes in serum BDNF and participant characteristics (i.e., age, weight, BMI, and LVEF), as well as other analysed variables (i.e., workload peak, CPET duration, HR rest, HR peak, RER peak, VO_2 rest, VO_2 peak, baseline lactate, peak lactate, and change in lactate). The correlation strength was interpreted as negligible (0.00 – 0.09), weak (0.10 – 0.39), moderate (0.40 – 0.69), strong (0.70 – 0.89), and very strong (> 0.90) (Schober et al., 2018). Statistical significance was set at $p \leq .05$.

REFERENCES

Azevedo, L. V. dos S., Pereira, J. R., Silva Santos, R. M., Rocha, N. P., Teixeira, A. L., Christo, P. P., Santos, V. R., & Scalzo, P. L. (2022). Acute exercise increases BDNF serum levels in patients with Parkinson's disease regardless of depression or fatigue. *European Journal of Sport Science*, *22*(8), 1296–1303. https://doi.org/10.1080/17461391.2021.1922505

Cattaneo, A., Cattane, N., Begni, V., Pariante, C. M., & Riva, M. A. (2016). The human BDNF gene: peripheral gene expression and protein levels as biomarkers for psychiatric disorders. *Translational Psychiatry 2016 6:11*, *6*(11), e958–e958. https://doi.org/10.1038/tp.2016.214

Chacón-Fernández, P., Säuberli, K., Colzani, M., Moreau, T., Ghevaert, C., & Barde, Y.-A. (2016). Brain-derived Neurotrophic Factor in Megakaryocytes. *Journal of Biological Chemistry*, *291*(19), 9872–9881. https://doi.org/10.1074/jbc.M116.720029

Chan, C. B., & Ye, K. (2017). Sex differences in brain-derived neurotrophic factor signaling and functions. *Journal of Neuroscience Research*, *95*(1–2), 328–335. https://doi.org/https://doi.org/10.1002/jnr.23863

Coelho, F. G. de M., Gobbi, S., Andreatto, C. A. A., Corazza, D. I., Pedroso, R. V., & Santos-Galduróz, R. F. (2013). Physical exercise modulates peripheral levels of brain-derived neurotrophic factor (BDNF): A systematic review of experimental studies in the elderly. *Archives of Gerontology and Geriatrics*, *56*(1), 10–15. https://doi.org/https://doi.org/10.1016/j.archger.2012.06.003

Coelho, F. G. de M., Vital, T. M., Stein, A. M., Arantes, F. J., Rueda, A. V., Camarini, R., Teodorov, E., & Santos-Galduróz, R. F. (2014). Acute Aerobic Exercise Increases Brain-Derived Neurotrophic Factor Levels in Elderly with Alzheimer's Disease. *Journal of Alzheimer's Disease*, *39*, 401–408. https://doi.org/10.3233/JAD-131073

Dinoff, A., Herrmann, N., Swardfager, W., & Lanctôt, K. L. (2017). The effect of acute exercise on blood concentrations of brain-derived neurotrophic factor in healthy adults: a meta-analysis. *European Journal of Neuroscience*, *46*(1), 1635–1646. https://doi.org/https://doi.org/10.1111/ejn.13603

Donovan, M. J., Lin, I. M., Ringstedt, T., Kraemer, R., Hahn, R., Wang, S., Ibañez, C. F., Rafii, S., & Hempstead, B. L. (2000). *BDNF is an endothelial cell survival factor*.

Edelmann, E., Leßmann, V., & Brigadski, T. (2014). Pre- and postsynaptic twists in BDNF secretion and action in synaptic plasticity. *Neuropharmacology*, *76*, 610–627. https://doi.org/https://doi.org/10.1016/j.neuropharm.2013.05.043

Faul, F., Erdfelder, E., Buchner, A., & Lang, A.-G. (2009). Statistical power analyses using G*Power 3.1: Tests for correlation and regression analyses. *Behavior Research Methods*, *41*(4), 1149–1160. https://doi.org/10.3758/BRM.41.4.1149

Feng, N., Huke, S., Zhua, G., Tocchetti, C. G., Shi, S., Aiba, T., Kaludercice, N., Hooverf, D. B., Beckg, S. E., Mankowskig, J. L., Tomaselli, G. F., Bersh, D. M., Kassa, D. A., & Paoloccia, N. (2015). Constitutive BDNF/TrkB signaling is required for normal cardiac contraction and relaxation. *Proceedings of the National Academy of Sciences of the United States of America*, *112*(6), 1880–1885. https://doi.org/10.1073/pnas.1417949112

Ferris, L. E. E. T., Williams, J. S., & Shen, C.-L. (2007). The Effect of Acute Exercise on Serum Brain-Derived Neurotrophic Factor Levels and Cognitive Function. *Medicine & Science in Sports & Exercise*, *39*(4). https://journals.lww.com/acsm-msse/fulltext/2007/04000/the_effect_of_acute_exercise_on_serum.20.aspx

Fukushima, A., Kinugawa, S., Homma, T., Masaki, Y., Furihata, T., Yokota, T., Matsushima, S., Takada, S., Kadoguchi, T., Oba, K., Okita, K., & Tsutsui, H. (2015). Serum Brain-Derived Neurotropic Factor Level Predicts Adverse Clinical Outcomes in Patients With Heart Failure. *Journal of Cardiac Failure*, *21*(4), 300–306. https://doi.org/10.1016/j.cardfail.2015.01.003

Groenewegen, A., Rutten, F. H., Mosterd, A., & Hoes, A. W. (2020). Epidemiology of heart failure. *European Journal of Heart Failure*, *22*(8), 1342–1356. https://doi.org/https://doi.org/10.1002/ejhf.1858

Hayek, L. El, Khalifeh, M., Zibara, V., Rawad, X., Assaad, A., Emmanuel, N., Karnib, N., El-Ghandour, R., Nasrallah, X. P., Bilen, M., Ibrahim, P., Younes, J., Haidar, E. A., Barmo, N., Jabre, V., Stephan, J. S., Sama, X., & Sleiman, F. (2019). *Lactate Mediates the Effects of Exercise on Learning and Memory through SIRT1-Dependent Activation of Hippocampal Brain-Derived Neurotrophic Factor (BDNF)*. https://doi.org/10.1523/JNEUROSCI.1661-18.2019

Holsinger, R. M. D., Schnarr, J., Henry, P., Castelo, V. T., & Fahnestock, M. (2000). Quantitation of BDNF mRNA in human parietal cortex by competitive reverse transcription-polymerase chain reaction: decreased levels in Alzheimer's disease. *Molecular Brain Research*, *76*(2), 347–354. https://doi.org/https://doi.org/10.1016/S0169-328X(00)00023-1

Hong, J.-H., Park, H.-M., Byun, K. H., Lee, B. H., Kang, W. C., & Jeong, G.-B. (2014). BDNF expression of macrophages and angiogenesis after myocardial infarction. *International Journal of Cardiology*, *176*(3), 1405–1408. https://doi.org/10.1016/j.ijcard.2014.08.019

Inoue, D. S., Monteiro, P. A., Gerosa-Neto, J., Santana, P. R., Peres, F. P., Edwards, K. M., & Lira, F. S. (2020). Acute increases in brain-derived neurotrophic factor following high or moderate-intensity exercise is accompanied with better cognition performance in obese adults. *Scientific Reports*, *10*(1), 13493. https://doi.org/10.1038/s41598-020-70326-1

Jiang, H., Liu, Y., Zhang, Y., & Chen, Z. Y. (2011). Association of plasma brain-derived neurotrophic factor and cardiovascular risk factors and prognosis in angina pectoris. *Biochemical and Biophysical Research Communications*, *415*(1), 99–103. https://doi.org/10.1016/j.bbrc.2011.10.020

Jin, W. (2020). Regulation of BDNF-TrkB Signaling and Potential Therapeutic Strategies for Parkinson's Disease. *Journal of Clinical Medicine*, *9*(1). https://doi.org/10.3390/JCM9010257

Kadowaki, S., Shishido, T., Honda, Y., Narumi, T., Otaki, Y., Kinoshita, D., Nishiyama, S., Takahashi, H., Arimoto, T., Miyamoto, T., Watanabe, T., & Kubota, I. (2016). Additive clinical value of serum brain-derived neurotrophic factor for prediction of chronic heart failure outcome. *Heart and Vessels*, *31*(4), 535–544. https://doi.org/10.1007/s00380-015-0628-6

Kermani, P., & Hempstead, B. (2019). BDNF actions in the cardiovascular system: Roles in development, adulthood and response to injury. In *Frontiers in Physiology* (Vol. 10, Issue APR). Frontiers Media S.A. https://doi.org/10.3389/fphys.2019.00455

Knaepen, K., Goekint, M., Heyman, E. M., & Meeusen, R. (2010). Neuroplasticity — Exercise-Induced Response of Peripheral Brain-Derived Neurotrophic Factor. *Sports Medicine*, *40*(9), 765–801. https://doi.org/10.2165/11534530-000000000-00000

Matthews, V. B., Åström, M. B., Chan, M. H. S., Bruce, C. R., Krabbe, K. S., Prelovsek, O., Åkerström, T., Yfanti, C., Broholm, C., Mortensen, O. H., Penkowa, M., Hojman, P., Zankari, A., Watt, M. J., Bruunsgaard, H., Pedersen, B. K., & Febbraio, M. A. (2009). Brain-derived neurotrophic factor is produced by skeletal muscle cells in response to contraction and enhances fat oxidation via activation of AMP-activated protein kinase. *Diabetologia*, *52*(7), 1409–1418. https://doi.org/10.1007/s00125-009-1364-1

Mezzani, A., Agostoni, P., Cohen-Solal, A., Corrà, U., Jegier, A., Kouidi, E., Mazic, S., Meurin, P., Piepoli, M., Simon, A., Laethem, C. Van, & Vanhees, L. (2009). Standards for the use of cardiopulmonary exercise testing for the functional evaluation of cardiac patients: A report from the Exercise Physiology Section of the European Association for Cardiovascular Prevention and Rehabilitation. *European Journal of Preventive Cardiology*, *16*(3), 249–267. https://doi.org/10.1097/HJR.0B013E32832914C8

Myers, J., McAuley, P., Lavie, C. J., Despres, J.-P., Arena, R., & Kokkinos, P. (2015). Physical Activity and Cardiorespiratory Fitness as Major Markers of Cardiovascular Risk: Their Independent and Interwoven Importance to Health Status. *Progress in Cardiovascular Diseases*, *57*(4), 306–314. https://doi.org/https://doi.org/10.1016/j.pcad.2014.09.011

Nakano, I., Kinugawa, S., Hori, H., Fukushima, A., Yokota, T., Takada, S., Kakutani, N., Obata, Y., Yamanashi, K., & Anzai, T. (2020). Serum brain-derived neurotrophic factor levels are associated with skeletal muscle function but not with muscle mass in patients with heart failure. *International Heart Journal*, *61*(1), 96–102. https://doi.org/10.1536/ihj.19-400

Neder, J. A., Nery, L. E., Castelo, A., Andreoni, S., Lerario, M. C., Sachs, A., Silva, A. C., & Whipp, B. J. (1999). Prediction of metabolic and cardiopulmonary responses to maximum cycle ergometry: a randomised study. *The European Respiratory Journal*, *14*(6), 1304–1313. https://doi.org/10.1183/09031936.99.14613049

Nocon, M., Hiemann, T., Müller-Riemenschneider, F., Thalau, F., Roll, S., & Willich, S. N. (2008). Association of physical activity with all-cause and cardiovascular mortality: a systematic review and meta-analysis. *European Journal of Cardiovascular Prevention and Rehabilitation*, *15*(3), 239–246. https://doi.org/10.1097/HJR.0b013e3282f55e09

Park, H., & Poo, M. (2013). Neurotrophin regulation of neural circuit development and function. *Nature Reviews Neuroscience*, *14*(1), 7–23. https://doi.org/10.1038/nrn3379

Piepmeier, A. T., & Etnier, J. L. (2015). Brain-derived neurotrophic factor (BDNF) as a potential mechanism of the effects of acute exercise on cognitive performance. *Journal of Sport and Health Science*, *4*(1), 14–23. https://doi.org/https://doi.org/10.1016/j.jshs.2014.11.001

Piepoli, M. F., Corrà, U., Benzer, W., Bjarnason-Wehrens, B., Dendale, P., Gaita, D., McGee, H., Mendes, M., Niebauer, J., Zwisler, A. D. O., & Schmid, J. P. (2010). Secondary prevention through cardiac rehabilitation: From knowledge to implementation. A position paper from

the cardiac rehabilitation section of the European association of cardiovascular prevention and rehabilitation. In *European Journal of Cardiovascular Prevention and Rehabilitation* (Vol. 17, Issue 1, pp. 1–17). SAGE Publications Inc. https://doi.org/10.1097/HJR.0b013e3283313592

Pius-Sadowska, E., & Machaliński, B. (2017). BDNF – A key player in cardiovascular system. In *Journal of Molecular and Cellular Cardiology* (Vol. 110, pp. 54–60). Academic Press. https://doi.org/10.1016/j.yjmcc.2017.07.007

Reycraft, J. T., Islam, H., Townsend, L. K., Hayward, G. C., Hazell, T. O. M. J., & MacPherson, R. E. K. (2020). Exercise Intensity and Recovery on Circulating Brain-derived Neurotrophic Factor. *Medicine & Science in Sports & Exercise*, *52*(5). https://journals.lww.com/acsm-msse/fulltext/2020/05000/exercise_intensity_and_recovery_on_circulating.24.aspx

Sanchis-Gomar, F., Lavie, C. J., Marín, J., Perez-Quilis, C., Eijsvogels, T. M. H., O'Keefe, J. H., Perez, M. V., & Blair, S. N. (2022). Exercise effects on cardiovascular disease: From basic aspects to clinical evidence. In *Cardiovascular Research* (Vol. 118, Issue 10, pp. 2253–2266). Oxford University Press. https://doi.org/10.1093/cvr/cvab272

Saucedo Marquez, C. M., Vanaudenaerde, B., Troosters, T., & Wenderoth, N. (2015). High-intensity interval training evokes larger serum BDNF levels compared with intense continuous exercise. *Journal of Applied Physiology*, *119*(12), 1363–1373. https://doi.org/10.1152/japplphysiol.00126.2015

Savarese, G., Becher, P. M., Lund, L. H., Seferovic, P., Rosano, G. M. C., & Coats, A. J. S. (2023). Global burden of heart failure: a comprehensive and updated review of epidemiology. In *Cardiovascular research* (Vol. 118, Issue 17, pp. 3272–3287). NLM (Medline). https://doi.org/10.1093/cvr/cvac013

Schober, P., Boer, C., & Schwarte, L. A. (2018). Correlation Coefficients: Appropriate Use and Interpretation. *Anesthesia & Analgesia*, *126*(5). https://journals.lww.com/anesthesia-analgesia/fulltext/2018/05000/correlation_coefficients__appropriate_use_and.50.aspx

Severinsen, M. C. K., & Pedersen, B. K. (2020). Muscle–Organ Crosstalk: The Emerging Roles of Myokines. In *Endocrine Reviews* (Vol. 41, Issue 4, pp. 594–609). Endocrine Society. https://doi.org/10.1210/ENDREV/BNAA016

Visseren, F., Mach, F., Smulders, Y. M., Carballo, D., Koskinas, K. C., Bäck, M., Benetos, A., Biffi, A., Boavida, J. M., Capodanno, D., Cosyns, B., Crawford, C. A., Davos, C. H., Desormais, I., Di Angelantonio, E., Duran, O. H. F., Halvorsen, S., Richard Hobbs, F. D., Hollander, M., … Mullabayeva, G. (2021). 2021 ESC Guidelines on cardiovascular disease prevention in clinical practice. In *European Heart Journal* (Vol. 42, Issue 34, pp. 3227–3337). Oxford University Press. https://doi.org/10.1093/eurheartj/ehab484

Wasserman, K., Stringer, W. W., Casaburi, R., Koike, A., & Cooper, C. B. (1994). Determination of the anaerobic threshold by gas exchange: Biochemical considerations, methodology and physiological effects. *Zeitschrift Fur Kardiologie*, *83*(SUPPL. 3), 1–12.

Yoshii, A., & Constantine-Paton, M. (2010). Postsynaptic BDNF-TrkB signaling in synapse maturation, plasticity, and disease. *Developmental Neurobiology*, *70*(5), 304–322. https://doi.org/https://doi.org/10.1002/dneu.20765