International Journal of Kinesiology & Sports Science
ISSN: 2202-946X
www.ijkss.aiac.org.au

Chronotype-based High-intensity Interval Training: Effects on Cardiac Biomarkers and Oxidative Stress in Obese Adults

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ARTICLE INFO

Article history
Received: October 21, 2024
Revised: December 22, 2024
Accepted: January 02, 2025
Published: January 30, 2025
Volume: 13 Issue: 1

Conflicts of interest: None.
Funding: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

ABSTRACT

Background: Obesity increases the risk of chronic diseases like diabetes, heart disease, hypertension, and cancer due to inflammation, insulin resistance, and elevated homocysteine(HCY) levels. Regular physical activity improves cardiovascular health, with High-Intensity Interval Training (HIIT) emerging as an effective intervention. However, individual factors like chronotype influence responses to HIIT. This study investigates the impact of HIIT on obesity-related outcomes and explores how chronotype modulates these effects, addressing the need for personalized exercise strategies. **Methods:** This study employed a pre-test post-test experimental design involving 60 male and female volunteers. Participants were divided into chronotype-based exercise schedules (CBES) and non-chronotype-based exercise schedules (NCBES) groups. Exclusion criteria included coronary artery disease, type 2 diabetes, peripheral arterial disease, or hypertension. HIIT was conducted using a cycle ergometer for 20 minutes, three times a week, over 12 weeks. Anthropometric measurements and biochemical assays, including lipid profiles and cardiac markers, were conducted pre-and post-intervention. **Results:** Homocysteine levels significantly decreased from 20.45 ± 5.62 μmol/L to 18.22 ± 5.07 μmol/L ($p < 0.001$). Participants experienced significant reductions in weight (average loss of 3.87 kg) and BMI (average decrease of 1.38 units). Lipid profiles showed improvements, with notable reductions in total cholesterol, triglycerides, LDL, and VLDL cholesterol ($p < 0.001$). MDA levels, a marker of oxidative stress, increased significantly from 181.83 ± 6.57 nmol/mL to 250.68 ± 31.52 nmol/mL ($p < 0.001$). Both CBES and NCBES groups exhibited positive changes, although no significant differences were observed between them. **Conclusion:** HIIT effectively improves cardiovascular biomarkers and reduces body weight and BMI in overweight and obese individuals, regardless of the exercise program. However, it also increases oxidative stress markers, highlighting the need for balanced exercise protocols. The impact of chronotype on exercise outcomes warrants further investigation

Key words: High-intensity Interval Training, Homocysteine, Malondialdehyde, Body Composition Obesity

INTRODUCTION

Health problems associated with obesity have surged in recent years. Obesity raises the risk of acquiring chronic non-communicable diseases such type 2 diabetes mellitus, coronary heart diseases, hypertension, and cancer (Gadde et al., 2018). Obesity causes white adipose tissue to release various substances that assist in maintaining metabolic balance. Chronic low-grade inflammation and insulin resistance have a crucial role in all non-communicable diseases. The secretions consist of leptin, adiponectin, resistin, tumor necrosis factor- (TNF-), and interleukin-6 (IL-6) which help regulate metabolic balance (Shoelson et al., 2007). Obesity, homocysteine, and Malondialdehyde are also significantly related, in addition to the above adipokines. A recent meta-analysis found a strong link between obesity and homocysteine levels (R. Wang et al., 2021) Elevated homocysteine levels have a minimal effect as an independent indicator of the likelihood of ischemic heart disease and stroke in individuals without preexisting health conditions (Homocysteine Studies Collaboration, 2002)

Homocysteine is an established risk factor for coronary vascular diseases. Recent studies indicate that homocysteine has a crucial role in vascular disease. A recent study has

Published by Australian International Academic Centre PTY.LTD.

http://dx.doi.org/10.7575/aiac.ijkss.v.13n.1p.18

shown that hyperhomocysteinemia is a substantial and independent risk factor for cardiovascular disease (CVD) (Konukoğlu et al., 2003). High levels of homocysteine in the blood are linked to heightened vascular and platelet harm(Wald et al., 1998). Elevated levels of homocysteine were identified as the primary factor behind changes in lipid metabolism and accumulation of lipids in tissues, resulting in obesity(R. Wang et al., 2021). Homocysteine-induced endoplasmic reticulum stress disrupts cholesterol and triglyceride production pathways, causing aberrant lipid metabolism. Obesity is considered a chronic inflammatory disorder. Elevated levels of inflammatory markers such as CRP and fibrinogen were associated with high HCY levels (R. Wang et al., 2021). Oxidative stress has been linked in clinical research to atherosclerosis and its associated risk factors. These studies have primarily concentrated on small, specifically chosen patient cohorts in the later phases of the illness (Keaney et al., 2003).

Oxidative stress (OS) is an imbalance between oxidants and antioxidants in the body, resulting in excessive formation of reactive oxygen species (ROS). Oxidative stress is increasingly acknowledged as playing a critical role in the development of several diseases. (Alfeel et al., n.d.). Ongoing research aims to clarify the relationship between hyperhomocysteinemia and oxidative stress. Endothelial dysfunction and the advancement of atherothrombotic vascular disease (Welch et al., 1997). Obese people had a 65% increase in plasma malondialdehyde (MDA) levels and a 53% decrease in vitamin E concentration compared to non-obese individuals. Oxidative stress is recognized as a crucial role in the onset of several diseases in non-obese individuals (Vincent & Taylor, 2006).

MDA causes oxidative damage and impairs glucose metabolism in obese persons. Obesity-induced hyperglycemia may lead to a sustained rise in MDA generation. MDA at 18 promotes oxidative harm and obstructs glucose metabolism in obese people. Obesity-induced chronic hyperglycemia can lead to a persistent rise in MDA production. Iron enhances the production of highly reactive oxygen species by MDA (ROS), initiating a loop of ROS synthesis and oxidative stress that disrupts regular glucose metabolism in obese individuals. Iron increases MDA's capacity to produce reactive oxygen species (ROS), initiating a cycle of ROS generation and oxidative stress that interferes with normal glucose metabolism in obese people (Vincent & Taylor, 2006). Hcy can enhance the production and function of enzymes involved in generating reactive oxygen species (ROS), such as NADPH oxidase and xanthine oxidase. This can result in increased oxidative stress in cells and tissues (Perna et al., 2003). Variations in the hCLOCK gene can result in variations in the daily patterns of homocysteine levels. Individuals with specific hCLOCK genotypes may experience elevated homocysteine levels during the evening, potentially impacting the management of cardiovascular risk (Paul et al., 2014).

Regular physical activity offers health benefits, but vigorous or prolonged exercise results in a dramatic increase in ROS production, as seen by elevated markers of oxidative damage in the blood and skeletal muscles (Powers et al., 2020). Moreover, the reduction in cardiovascular risk factors from exercising shows a clear dose-response relationship, with greater levels of exercise intensity and duration leading to more substantial health benefits(Powers et al., 2020). Exercise can significantly trigger oxidative stress and affect the circadian clock. When performed at the right intensity and duration, it can lead to beneficial health outcomes(McClean & Davison, 2022). The adaptability of exercise is correlated with the degree of reactive oxygen and nitrogen species (RONS) produced, which seems to be influenced by the intensity of the exercise session or training stimulus. High-intensity exercise leads to higher levels of RONS than low to moderate-intensity aerobic activity below 50% (McClean & Davison, 2022). A three-week regimen of high-intensity interval training (HIIT) boosts plasma antioxidant levels in individuals (McClean & Davison, 2022).

Engaging in regular exercise results in a range of health benefits, including improvements in cardiovascular health and metabolic functions. The study by Atakan et al. (2021) provided evidence-based effects of high-intensity interval training (HIIT) and moderate-intensity continuous training (MICT) on these health outcomes(Atakan et al., 2021). Steele et al. (2021) conducted a systematic review and meta-analysis comparing the effectiveness of slow and steady versus hard and fast training approaches, highlighting the nuanced benefits of both methods (Steele et al., 2021). Su et al. (2019) evaluated the effects of HIIT and MICT on cardiovascular risk factors and found significant improvements in markers of cardiovascular health following both types of exercise (Su et al., 2019). Sawyer et al. (2016) hypothesized and confirmed that HIIT induces greater improvements in cardiovascular fitness and metabolic health compared to MICT (Sawyer et al., 2016). Fisher et al. (2015) compared the effects of six weeks of HIIT versus MICT on inflammatory markers and observed that HIIT had a more pronounced impact on reducing inflammation(alabaf yousefi et al., 2021) (Fisher et al., 2015). These findings collectively underscore the potential of HIIT as a more efficient exercise modality for improving various health outcomes compared to traditional moderate-intensity training.

Chronotypes represent individual variations in circadian rhythms, affecting optimum timing for sleep and everyday activities, such as eating. Individuals with late chronotypes exhibit inferior dietary quality, elevated evening caloric consumption, and heightened susceptibility to obesity and associated health complications (Beaulieu et al., 2024). Additionally, the impact of chronotypes on exercise outcomes is found, and individual preferences for activity times significantly affect physiological responses to training. The morning chronotypes may experience enhanced performance and adherence when exercising in the morning, whereas evening chronotypes showed better outcomes when training later in the day (Bruggisser et al., 2023). The research by Galen et al. (2020) further supports this by indicating that the timing of exercise sessions relative to an individual's chronotype can optimize fitness gains and overall well-being(Galan-Lopez & Casuso, 2023). Roenneberg et al. (2019) highlighted the importance of aligning exercise regimens with biological rhythms, suggesting that personalized training schedules based on chronotype can lead to more effective health interventions and improved long-term adherence to physical activity routines (Roenneberg et al., 2019). This study hypothesized that high-intensity interval training has an effect on cardiovascular biomarkers in overweight and obese adults with varying chronotypes.

METHODS

Participants and Study Design

Sixty male and female volunteers participated in this study, which employed a pretest-posttest experimental design. Based on computer-generated random numbers, the sample was assigned to the CBES and NCBES groups. Volunteers for this study were recruited via email, and those with coronary artery disease, type 2 diabetes, peripheral arterial disease, or hypertension were excluded. This study was conducted in the Physiotherapy Department of the SRM Medical College Hospital and Research Center. This study protocol was approved by the Institutional Ethical Committee of SRM Medical College IEC (1761/IEC/2019), Chennai. The sample size was determined using G Power software 3.1 to detect a moderate effect size of high-intensity interval training-induced changes in metabolic and inflammatory biomarkers with 80% power, and we determined that 60 samples (34 males and 26 females) would detect an effect size of 0.80 with an average BMI of 29 (Ouerghi et al., 2022). The chronotype of an individual was identified using Horne & Ostberg morningness and eveningness questionnaire (Adan & Almirall, 1991). The CEBS group exercise at times aligned with their individual chronotypes. The chronotype was scheduled for morning exercise (5PM to 8PM), and evening chronotypes exercised in the evening (5 PM to 8 PM). In contrast, the NCBES group followed the exercise schedule with a session conducted mid-day 10 AM to 12 PM) regardless of participant's chronotype.

Exercise Protocol

The CBES and NCBES group used a cycle ergometer (Spin Bike XB-5816 Energie Fitness, India) to complete 60 reps of high-intensity interval training with 1: 1 ratio (10 seconds cycling and 10 seconds active recovery) for 20 min, 3 days a week for 12 weeks (Kong, Sun, Liu, & Shi, 2016) An experienced physiotherapist monitored the stopwatch as it was utilized to track the interval between HIIT and active recovery. Every time, the physiotherapist used verbal cues to remind the timeline of the intervention. The initial resistance at the beginning of the workout phase was 1.0 kg. After completing two consecutive sessions at this effort, the resistance was increased in increments of 0.5 kg. Heart rate and ratings of perceived exertion were measured using the Omron HEM 6161 and Borg scale, respectively, before and after every five intervals of the 10-second cycling activity. During the intervention phase, daily physical activity was monitored with the Google Fit mobile app, while food intake was recorded using a 24-hour dietary recall (Shim, Oh, & Kim, 2014).

Test Protocol

The baseline data of anthropometric and blood samples were taken before the exercise training, and the post-test was measured after 24 hours of the last exercise session. The participants were requested to wear light clothing and no shoes when their weight was assessed using a standard weighing scale (Activex, Pune, India). The participants' heights were measured accurately using a wall-mounted stadiometer, with the readings taken and recorded to the nearest 1 millimeter for precision. Body mass index (BMI) Asian classification was computed by multiplying body weight by squared height.

Heart rate and ratings of perceived exertion were measured using the Omron HEM 6161 and Borg scale, respectively, before and after every five intervals of the 10-second cycling activity. During the intervention phase, daily physical activity was monitored with the Google Fit mobile app, while food intake was recorded using a 24-hour dietary recall (Shim, Oh, & Kim, 2014). The biochemical samples were processed using specific enzymatic method, which involves biochemical reactions catalyzed by enzymes to quantify the concentration of these markers (Alam et al., 2019; Bevan et al., 2003; Malik & Pundir, 2002a)

Lipid Profile

After 12 hours fast, 5 mL of venous blood was drawn from each participant in a vacutainer, centrifuged to separate the serum, and kept in deep freezer at -80 degrees Celsius. Using proper methodologies, the following parameters were extracted from the samples. Lipid profiles (Total cholesterol, Triglyceride, HDL, LDL, VLDL) were assayed using the cholesterol oxidase technique with measure the level of mg/dl (Malik & Pundir, 2002b).

Cardiac Marker

Cardiac markers such as homocysteine and MDA (Malondialdehyde) were measure with enzymatic methods using the unit of μmol/L(Alam et al., 2019; Bevan et al., 2003)

Statistical Analysis

The Kolmogorov – Smirnov test was used to determine data normality and variance equality. Descriptive statistics were computed for all study variables. The student 't' test was used to find out the between group analysis of the intervention. All the tests were performed at a 0.05 level of significance using PSPP statistical software

RESULT

A total of 59 participants were included in the study for final analysis. One person quit the study due to translocation of their job. Their average age was 31.49± 9.12 years. Participants were within a healthy weight range, with an average pre-intervention body mass index (BMI) of 29.17 (SD 4.13). Their average height was 167.20 cm (SD 8.62) and their average pre-intervention weight was 81.54 kg (SD 12.78) (Table 1).

Table 1. Demographic data

	N	Mean & SD
AGE	59.00	31.49±9.12
Height	59.00	167.20±8.62
Pre weight	59.00	81.54±12.78
Pre BMI	59.00	29.17±4.13

BMI Body Mass Index

Homocysteine levels were not reduced from a mean of 24.25 ± 14.1 µmol/L pre-intervention to 24.19± 14.41 µmol/L post-intervention ($t = 0.91$, $p < 0.941$) (Table 2). Weight and BMI also decreased significantly, with participants losing an average of 3.87 kg and 1.38 BMI units, respectively ($t = 15.36$ and 15.6, respectively, $p < 0.001$ for both). Lipid profiles showed positive changes as well, with total cholesterol decreasing from 193.09 ± 22.23 mg/dL to 188.19 ± 21.71 mg/dL ($t = 6.32$, $p < 0.001$), triglycerides from 139.57 ± 50.39 mg/dL to 132.28 ± 48.58 mg/dL ($t = 6.34$, $p < 0.001$), LDL cholesterol from 122.97 ± 22.02 mg/dL to 117.59 ± 21.45 mg/dL ($t = 10.34$, $p < 0.001$), and VLDL cholesterol from 26.35 ± 10.60 mg/dL to 20.50 ± 9.42 mg/dL ($t = 10.20$, $p < 0.001$). Notably, these decreases occurred without affecting HDL cholesterol levels (48.07 ± 9.8 mg/dL pre-intervention vs. 47.69 ± 7.71 mg/dL post-intervention, $p = 0.614$). MDA levels, a marker of oxidative stress, increased significantly from 181.83 ± 6.57 nmol/mL to 250.68 ± 31.52 nmol/mL ($t = 15.732$, $p < 0.001$).

Both CBES and NCBES groups showed positive changes in some cardiac markers after the intervention, although differences emerged between them. Homocysteine levels remained largely unchanged in both groups (CBES: 17.96 ± 5.11 µmol/L; NCBES: 18.58 ± 5.10 µmol/L), with no statistically significant difference observed ($p = 0.644$) (Table 3). Both groups experienced favorable changes in lipid profiles, with the CBES group exhibiting statistically significant reductions in total cholesterol (TCL) to 183.44 ± 3.60 mg/dL compared to 194.66 ± 4.28 mg/dL in the NCBES group ($p = 0.049$). LDL cholesterol also decreased significantly in the CBES group to 116.56 ± 3.33 mg/dL compared to 118.98 ± 4.86 mg/dL in the NCBES group, although this difference did not reach statistical significance ($p = 0.672$). While both groups saw slight decreases in triglyceride (TGL) levels, no significant difference was detected between them ($p = 0.738$). Notably, HDL cholesterol remained stable in both groups ($p = 0.770$).

Both groups demonstrated statistically significant reductions in weight and BMI post-intervention. The CBES group experienced slightly greater reductions in weight (76.81 ± 12.97 kg vs. 78.83 ± 11.34 kg in NCBES) and BMI (27.97 ± 4.07 vs. 27.54 ± 0.77 in NCBES), but these differences did not reach statistical significance ($p = 0.536$ and $p = 0.684$, respectively). MDA, a marker of oxidative stress, increased significantly in both groups following the intervention, with no significant difference observed between them ($p = 0.563$).

Table 2. Comparison of cardiac markers between before and after high intensity interval training.

Variable Name	Mean	T value	*P*-value
PRE Homocystinine	20.45±5.62	10.49	0.000
Post Homocystinine	18.22±5.07		
Pre test MDA	181.83±6.57	15.732	0.000
Post test MDA	250.68±31.52		
Pre weight	81.54±12.78	15.36	0.000
Post Weight	77.67±12.24		
Pre BMI	29.17±4.13	15.6	0.000
Post BMI	27.79±3.96		
Pre TCL	193.09±22.23	6.32	0.000
Post TCL	188.19±21.71		
Pre HDL	48.07±9.8	0.507	0.614
Post HDL	47.69±7.71		
Pre TGL	139.57±50.39	6.34	0.000
Post TGL	132.28±48.58		
Pre LDL	122.97±22.02	10.34	0.000
Post LDL	117.59±21.45		
Pre VLDL	26.35±10.60	10.20	0.000
Post VLDL	20.50±9.42		

Malondialdehyde , TCL- Total Cholestrol, HDL- High Density lipoprotein, LDL- Low Density Lipoprotein,
VLDL - Very-low-density lipoprotein

DISCUSSION

The purpose of this was to investigate the effect of chronotype-based exercise session of HIIT on cardiac biomarkers and oxidative stress among obese individuals. Fifty-nine subjects with an average age of 31±49 participated in this study. There was a significant decrease in all parameters compared to prior high-intensity interval training. Improvements were observed in all measures, including cardiovascular risk markers, anthropometric measurements, lipid profile, and oxidative stress. When comparing the results of exercise training based on chronotype with exercise training not based on chronotype, no significant differences were noticed except total cholesterol.

The primary discovery of this study shows a considerable decrease in homocysteine levels following the exercise training. We notice a significant difference in homocysteine levels before and after exercising. This may be caused by changes in the blood lipid profile and insulin sensitivity. (e Silva & da Mota, 2014). And also this findings were consistent with previous research, such as Konig et al.'s conclusion on intensity and volume.(König et al., 2003).

Comparing exercise training based on chronotype with exercise training not based on chronotype showed no difference among the groups. Existing literature indicates a significant influence of chronotype on homocysteine levels (Lavie & Lavie, 2004). This study is the first to compare the effects of high-intensity interval training (HIIT) based on chronotype and non-chronotype on homocysteine levels in obese individuals. Our results indicate that there was no significant

Table 3. Comparison between cardiac markers between CBES group and NCBES group

Exercise session	Mean	Mean Difference	95% CI	*P*-value
Post Homocystinine				
CBES (n=34)	17.96±5.11	0.62±1.3	-3.3 to 2.06	0.644
NCBES (n=25)	18.58±5.10			
Post test MDA				
CBES (n=34)	252.74±31.53	4.8±8.3	-11.8 to 21.58	0.563
NCBES (n=25)	247.88±31.94			
Post Weight				
CBES (n=34)	76.81±12.97	2.02±3.2	-8.51 to 4.47	0.536
NCBES (n=25)	78.83±11.34			
Post BMI				
CBES (n=34)	27.97±4.07	0.42±1.02	-1.6 to 2.53	0.684
NCBES (n=25)	27.54±0.77			
Post TCL				
CBES (n=34)	183.44±3.60	11.2±5.57	-22.44 to 0.009	0.049
NCBES (n=25)	194.66±4.28			
Post HDL				
CBES (n=34)	47.95±1.42	0.60±1.9	-3.3 to 4.6	0.770
NCBES (n=25)	47.35±1.41			
Post TGL				
CBES (n=34)	130.44±8.04	4.3±12.8	-30.64 to 21.9	0.738
NCBES (n=25)	134.77±10.34			
Post LDL				
CBES (n=34)	116.56±3.33	2.4±5.6	-13.81 to 8.9	0.672
NCBES (n=25)	118.98±4.86			
Post VLDL				
CBES (n=34)	19.31±1.53	2.8±2.4	-7.7 to 2.14	0.261
NCBES (n=25)	22.12±2			

Malondialdehyde , TCL- Total Cholestrol, HDL- High Density lipoprotein, LDL- Low Density Lipoprotein, VLDL - Very-low-density lipoprotein, CBES- Chronotype Based Exercise Session, NCBES – Non Chronotype Based Exercise Session

difference discovered between exercise based on chronotype and non-chronotype. When comparing the results of exercise training based on chronotype with exercise training not based on chronotype, no significant differences were noticed except total cholesterol(König et al., 2003; Lavie & Lavie, 2004). In future research could consider these factors for optimal results of homocysteine.

The study revealed an increase in oxidative stress following HIIT training. HIIT generates a higher amount of reactive oxygen species (ROS) than other exercise types. (Atakan et al., 2022a) HIIT has a significant impact on oxidative stress due to several variables. The determinants include intensity, duration, recovery time, nutritional state, and individual differences (Amaro-Gahete et al., 2018). Adequate intervals between activities enable the body to efficiently manage high levels of oxidative stress. In our study, the recuperation and exercise time had a 1:1 interval ratio, leading to severe oxidative stress (Amaro-Gahete et al., 2018). Moreover, acute elevation of oxidative stress (MDA levels) is a usual process in HIIT exercise and tends to decrease over time(Lu et al., 2021). The established literature demonstrating that exercise often results in decreased oxidative stress. (Atakan et al., 2022b; de Matos et al., 2018; Reljic et al., 2020). There were no significant differences in MDA levels reported between obese persons who exercised based on their chronotype and those who did not. This study did not discover any significant improvement in MDA levels between individuals depending on their chronotype and those who exercised regardless of their chronotype.

Regarding the lipid profile, a significant difference was observed before and after high-intensity interval training in both groups. Existing data indicates that HIIT shows promising improvement in lipid profile in obese individuals.(Wood et al., 2019) Only total cholesterol was significantly lowered when comparing the CBES and NCBES groups, but indicators such as HDL and LDL were not significantly reduced. We detected a modest drop in triglyceride levels when comparing these two groups. The correlation between chronotype and lipid and body composition was firmly established. (R. D. Wang, n.d.)

There is a bigger difference in body composition between pre- and post-HIIT training. Both groups experienced a significant reduction in weight and BMI when compared within the group. However, no significant differences were observed when comparing between groups. Both CBES and NCBES demonstrated no significant difference in weight loss and BMI. An existing analysis suggests that those classified as evening types may be more resistant to weight reduction compared to morning types (Yu et al., 2015).

This study has many limitations. First, the subjects utilized by each group were distinct. As a result, we were unable to compare individuals' responses to exercise. We did not control the extraneous factors like food intake, types of food and their nutrient values, which can alter the biomarkers especially homocysteine. The same-subject crossover trial design could be used in the future. And also in future research to examine the influence of individual chronotype on cardiac markers. This study shows that both CBES and NCBES HIIT programs improve cardiac biomarkers, lipid profiles, and body composition in obese adults, with no significant differences between the two approaches. While chronotype-based exercise holds promise, further research is needed to confirm its practical benefits.

CONCLUSION

This study demonstrates that high-intensity interval training (HIIT) improves cardiac biomarkers, lipid profiles, and body composition in obese adults, regardless of exercise session chronotype. Cardiovascular indicators, lipid profiles, and oxidative stress improved significantly, but except for total cholesterol, chronotype- and non-chronotype-based sessions had no significant differences. HIIT may lower homocysteine, improve lipid profiles, and enhance weight loss, although chronotype effect is unknown. Due to participant variability and uncontrolled dietary factors, crossover trials with chronotype-specific focus are advised for future research. These findings help optimize obese exercise routines, but further research is needed to demonstrate chronotype-based exercise interventions' practical benefits.

IEC Approval: This study was approved by Institutional ethical committee, SRM Medica; College & Research Institute

Data Availability

The datasets generated during this study are available from the corresponding author upon reasonable request.

COMPETING INTEREST DECLARATION

The authors declare that there are no competing interests.

AUTHOR CONTRIBUTIONS

Author 1: was involved in the conception, design, data collection, analysis, and manuscript writing.

Author 2: contributed to the conception, design, and manuscript writing.

Author 3: contributed to the conception, design, and manuscript writing.

ACKNOWLEDGEMENT

Mr. J. Thirunavukarasu and Mr. Dineshnath Phd, scholars, Department of Biochemistry, SRMMCH & RI, SRMIST, deserve special thanks for their unwavering support and assistance

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