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

Effect of High-intensity Interval Training Session Timing on Inflammatory Biomarkers in Overweight and Obese Individuals with Different Chronotypes

Ayyappan Jayavel[1], Meera Shivasekar[2]*, Vinodhini V.M[2]

[1]SRM College of Physiotherapy, Faculty of Medicine & Health Sciences, SRM Institute of Science and Technology, SRM Nagar, Kattankulathur, 603203, Kancheepuram, Chennai, TN, India

[2]Department of Biochemistry, SRM Medical College Hospital and Research Center, Faculty of Medicine & Health Sciences, SRM Institute of Science and Technology, SRM Nagar, Kattankulathur, 603203, Kancheepuram, Chennai, TN, India

Corresponding Author: Dr. Meera Shivasekar, E-mail: meeras@srmist.edu.in

ARTICLE INFO

Article history
Received: August 21, 2023
Accepted: October 25, 2023
Published: October 31, 2023
Volume: 11 Issue: 4

Conflicts of interest: None.
Funding: None

ABSTRACT

Background: Global attention on obesity prevention emphasizes cost-effective strategies, particularly physical activity. Evaluating chronobiological influences is crucial for effective treatment plans. Noncommunicable disease management requires a nuanced approach to address this pervasive health concern**. Objective:** This study aims to determine the effect of chronotype-based high-intensity interval training (HIIT) on inflammatory biomarkers among overweight and obese individuals. **Methods:** This experimental study, involving 58 obese adults, utilized pretest and post-test evaluations. Participants, aged 31.65 ± 9.1 years with a mean BMI of 29.04 ± 4.04, underwent 12 weeks of cycling HIIT (10 sec on/10 sec off) thrice weekly. Pre- and post-training venous blood samples were collected, centrifuged, and stored at -80°C. Outcome measures included metabolic, inflammatory biomarkers, and anthropometric data. **Result:** The findings of this study demonstrate that there is a significant difference in metabolic variables such as fasting sugar, fasting insulin, 3.7%, 27% (p=0.00), and IL6 5.7% (p=0.00) levels between the chronotype-based exercise session group (CBES) and non chronotype based exercise session group (NCBES). Anthropometric and other inflammatory variables, such as Tumor necrosis factor alpha (TNF -alpha) and high sensitive C-reactive protein (hs CRP), showed no significant differences between groups. **Conclusion:** The study concluded that chronotype-based HIIT is effective on metabolic markers but not on inflammatory markers in obese individuals.

Key words: High-intensity Interval Training, Chronotypes, Insulin, Body Mass Index Inflammation, Obesity

INTRODUCTION

According to the World Health Organization (WHO), obesity is a significant public health concern obesity is a significant public health concern.(WHO, 2021). The rise in obesity rates among individuals of all age groups has been linked to the emergence of several chronic conditions, such as metabolic syndrome, type 2 diabetes, cardiovascular ailments, cancer, and arthritis(Piché et al., 2020). Recent empirical studies have emphasized the necessity of integrating physical exercise into all approaches aimed at preventing obesity and sustaining weight loss over an extended period (van Baak et al., 2021).

The association between obesity and the presence of CRP, as well as other inflammatory biomarkers, such as IL-6 and TNF-alpha, has been established in previous research (Fahed et al., 2022). Elevated amounts of C-reactive protein (CRP) and certain cytokines in the bloodstream are indicative of low-grade chronic inflammation (Fahed et al., 2022). There is a correlation between low-grade systemic inflammation and metabolic syndrome, type 2 diabetes, and atherosclerosis. There is evidence linking metabolic syndrome, type 2 diabetes, and atherosclerosis to low-grade systemic inflammation (Fahed et al., 2022). This connection is significant, as obesity is known to contribute to chronic low-grade inflammation, ultimately resulting in the development of type 2 diabetes and coronary heart disease (Fahed et al., 2022). There should be a direct relationship between these biomarkers and the classification of obesity (Mayoral et al., 2020).

The implementation of the WHO's physical exercise guidelines has proven to be difficult for global populations (WHO, 2022). This difficulty arises from various factors, including time constraints, lack of motivation, and soreness (Bull et al., 2020; Daskapan et al., 2006; F. Adeniyi et al., 2016; Kadariya & Aro, 2018). The available research unequivocally demonstrates that a dearth of physical activity plays a significant role in the development and progression of noncommunicable diseases (Kadariya & Aro, 2018). In contrast, individuals of South Asian ancestry are subject to

Published by Australian International Academic Centre PTY.LTD.

http://dx.doi.org/10.7575/aiac.ijkss.v.11n.4p.17

distinct perceptions of the obstacles they face, including familial obligations, demanding job schedules, and familial discouragement(Keating et al., 2017).

High-intensity interval training (HIIT) is a time-efficient approach that can be utilized in the treatment of obesity and its associated outcomes. HIIT is a form of exercise that entails short but vigorous bursts of physical activity executed at intensities ranging from 80% to 100% of an individual's maximal oxygen intake (VO2 Max). These intense intervals alternate with periods of low-intensity active recovery. HIIT can be carried out using a range of methods, such as body blast exercises, cycling, and other modes. The findings of a systematic review and meta-analysis suggest that HIIT can be considered comparable to moderate-intensity continuous exercise in terms of its effects (Nguyen et al., 2016). HIIT is gaining increasing popularity and recognition due to its several advantages, such as time efficiency (Maillard et al., 2018), making exercise enjoyable (Vella et al., 2017), and promising effects on adiposity (Maillard et al., 2018). However, the findings of the preceding systematic review and meta-analysis provide inconclusive evidence concerning the effects of HIIT on obesity and its associated factors, including chronic low-grade inflammation in individuals with obesity. There are consistent effects observed in anthropometric parameters (Khalafi & Symonds, 2020) and endothelial dysfunction(Ramírez-Vélez et al., 2019; Tsai et al., 2016).

Chronotypes, also known as circadian rhythms, refer to the human body's physiological variations that occur over a 24-hour period and involve changes in molecular markers (Ammar et al., 2017). The diurnal variations of individuals can be categorized according to their diurnal extremes as morningness (M), eveningness (E), or neither (N) (Souissi et al., 2004) metabolism (Aloui et al., 2017) psychological function (Trine & Morgan, 1995) hormonal variations (Aloui et al., 2017) and biochemical responses(Ammar et al., 2017; Hammouda et al., 2013) in exercise training, and the same will continuously change based on the chronobiological rhythm (Mancilla et al., 2020). The metabolic processes occurring at the substrate level were found to be significantly impacted by the rhythm of chronobiological activity, with a particular emphasis on the secretion of insulin (Saad et al., 2012), carbohydrate removal (DeFronzo & Tripathy, 2009) and oxidative stress (van Moorsel et al., 2016). Therefore, temporal variability throughout the day is an unavoidable aspect that should be considered when prescribing exercise. The investigation of the association between chronotypes and exercise benefits has been limited despite the significance of chronotypes in relation to exercise outcomes (Shiotani et al., 2009).

The study conducted by Blankenship et al (2021) examined many research works that investigated the impacts of exercise conducted at different time intervals throughout the day (Blankenship et al., 2021). The findings of this review produced a combination of outcomes. Additionally, the results indicate that engaging in exercise sessions during the early part of the day resulted in a more significant weight reduction when compared to exercise sessions conducted later in the day. A study conducted by Fillon, et al (2019), proposed that engaging in physical activity in the morning, namely in close proximity to lunchtime, could potentially lead to a decrease in subsequent energy consumption. The study suggested that exercising in the morning, particularly close to lunch, might reduce subsequent energy intake (Fillon et al., 2019). Saviki et al (2019) emphasized the significance of considering circadian cycles in the scheduling of physical activity to maximize the physiological advantages of exercise and mitigate metabolic disorders (Savikj et al., 2019). Despite substantial research that has been undertaken to examine the impact of HIIT on inflammation and cardiometabolic outcomes in individuals with obesity, recent higher-quality evidence has uncovered discrepancies in the findings, indicating the need for further rigorous studies to substantiate the observed effects (Martland et al., 2020). Therefore, this study aims to examine the impact of HIIT on inflammatory biomarkers across overweight and obese individuals exhibiting varying chronotypes.

METHODS

Participants and Study Design

The study employed pre- and post-test experimental designs. The participants in this study were recruited via electronic mail, and individuals with preexisting conditions, such as coronary artery disease, type 2 diabetes, peripheral arterial disease, or hypertension, were deemed ineligible for inclusion. The study protocol received approval from the Institutional Ethical Committee of SRM Medical College IEC (1761/IEC/2019), Chennai. This study involved 60 overweight and obese persons, both male and female, who met the predetermined inclusion criteria. The participants had an average age of 31.6±9.11 years. The sample size was determined using G*Power software 3.1 to detect a moderate effect size pertaining to the alterations in metabolic and inflammatory biomarkers induced by HIIT. With a power of 80%, it was determined that a total of 60 samples (34 males and 26 females) would be necessary to detect an effect size of 0.80, assuming an average body mass index (BMI) of 29. Prior to participating in the study, all participants had an assessment to identify their chronotypes. The chronotypes were identified by assessing the scores obtained from the Olov Ostberg morningness (M) and eveningness (E) questionnaires (MEQ). The subjects were categorized into three groups based on their questionnaire scores: morning type, evening type, and neither type. These groups were then further divided into a CBES and NCBES group.

Procedure

The study adopted a CBES in which participants classified as morningness (M type) were assigned to undergo HIIT in the morning (8am–10am), while participants classified as eveningness were assigned to get HIIT in the evening (4pm–6pm) (A. Souissi et al., 2020).

The participants in the NCBES group were assigned to undertake HIIT either in the evening, if they identified as morningness subjects, or in the morning, if they identified as eveningness subjects.

Intervention Protocol

The participants in the HIIT group utilized a cycle ergometer, mainly the Spin Bike XB-5816 Energie Fitness model from India. They performed a total of 60 repetitions of HIIT exercises, maintaining a 1:1 work-to-rest ratio. Each repetition consisted of 10 seconds of cycling at high intensity, followed by 10 seconds of active recovery. This exercise routine was conducted for a duration of 20 min, three 3 days per week, for a period of 12 weeks (Kong et al., 2016). An individual with specialized training observed the stopwatch while it was employed to track the duration of the interval cycles and auditory cues. The initial resistance during the exercise phase was set at 1.0 kg. Following the completion of two consecutive sessions at this workload, the resistance was systematically increased by increments of 0.5 kg. Heart rate (measured using the Omron HEM 6161) and ratings of perceived effort (assessed using the Borg scale) were recorded before and immediately after each 10-second cycling exercise, with measurements taken every five intervals (Shim et al., 2014).

Measurement

Anthropometric measurement

The participants were instructed to wear lightweight attire and refrain from wearing footwear throughout the measurement of their weight, which was conducted using a standardized weighing scale (Activex, Pune, India). The heights of the participants were measured with a wall-mounted stadiometer, with measurements recorded to the closest millimeter. The WHO's Asian classification of BMI was determined by multiplying the body weight and the square of height (Pacific, 2000).

Biochemistry assays: Following a 12-hour fasting period, 5 mL of venous blood was collected from each participant using a vacutainer. The collected blood samples then underwent centrifugation to extract the serum, which was subsequently stored in a deep freezer at a temperature of -80°C. The parameters were retrieved from the samples using appropriate procedures. Fasting blood sugar levels were measured soon after the samples were collected from the subjects using the hexokinase method (Westwood et al., 1986). Fasting insulin was assayed using ECLIA techniques (Cassidy et al., 2012). The homeostasis model assessment of insulin resistance (HOMA-IR) was calculated using the formula (fasting insulin (micro U/L) x fasting glucose (nmol/L)/22.5) (Salgado et al., 2010). The lipid profile which includes measurements of total cholesterol, triglyceride, high-density lipoprotein [HDL], low-density lipoprotein [LDL], and very low-density lipoprotein [VLDL]) was determined using the cholesterol oxidase technique (Malik & Pundir, 2002).

Inflammatory biomarkers

The measurement of inflammatory biomarkers, including IL-6, TNF-alpha, and hs-CRP, was conducted. The high-sensitivity C-reactive protein (hs-CRP) was measured using the enzyme-linked immunosorbent assay (ELISA) technique. The levels of IL-6 and TNF-alpha were quantified using an ELISA kit (EliKine, Abbkine, China).

Statistical Analysis

The Kolmogorov–Smirnov test was employed to assess the normality of the data and to examine the equality of variances. Descriptive statistics were calculated for all variables included in the study. The student's t-test was employed to do a between-group analysis of the intervention. The tests were conducted with a significance threshold of 0.05 using the PSPP statistical program.

RESULTS

All the participants successfully completed the protocol requirements, except for two individuals who could not do so due to employment relocations. A total of 58 participants, consisting of 32 males and 26 females, successfully concluded a 12-week exercise program. The participants had an average age of 31.6±9.11 years (Table 1). The Kolmogorov–Smirnov test demonstrated the presence of baseline similarity between anthropometric and inflammatory indicators.

Anthropometric Values

After engaging in HIIT, notable enhancements were observed in anthropometric measurements, specifically body

Table 1. Demographic data

S.No	Variable name	n	Mean	SD
1	Age	58	31.65	9.11
2	Gender	58	Male : 32 (55.2%) Female : 26 (44.8%)	
3	Height	58	167.20	8.69
4	Weight	58	81.19	12.60
5	BMI	58	29.04	4.04
6	Over weight & Obesity	58	Overweight : 36 (62.1%) Obesity : 22 (37.9%)	

n: number of samples, SD: Standard deviation

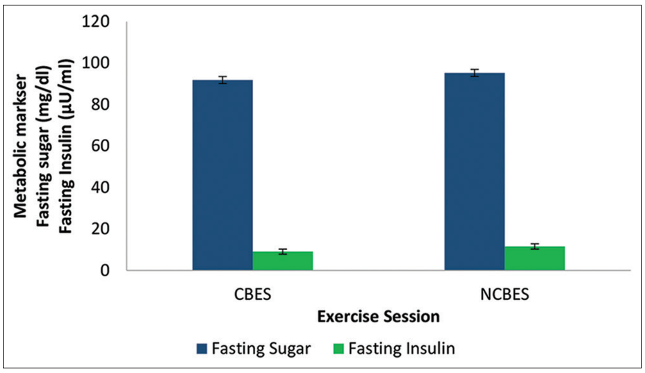

Figure 1. Comparison of Metabolic marker between CBES and NCBES Groups
**CBES: Chronotype based exercise session, NCBES: Non chronotype based exercise session

weight, BMI, and body circumference, within the experimental group. Among CBES participants (N=32), weight decreased insignificantly from 82.31 kg to 77.48 kg (mean difference: 0.24 kg, 95% CI: -6.23 to 6.71, p=0.940, Cohen's d=0.02). Waist and hip circumferences decreased modestly, and BMI showed a reduction from 30.03 to 28.28 (mean difference: 1.35, 95% CI: -0.72 to 3.39, p=0.198, Cohen's d=0.34).

After the HIIT, it was observed that fasting sugar levels decreased significantly from 101.5 mg/dL to 91.79 mg/dL (mean difference: -3.41 mg/dL, 95% CI: -6.73 to -0.07321, p=0.045, Cohen's d=0.54). Fasting insulin levels also dropped significantly from 15.17 µU/mL to 9.06 µU/mL (mean difference: -2.48 µU/mL, 95% CI: -3.54 to -1.41, p=0.000, Cohen's d=1.24). The HOMA-IR decreased from 3.79 to 2.07 (mean difference: -0.66, 95% CI: -0.94 to -0.37, p=0.000, Cohen's d=1.23) (Figure 1).

Before and after HIIT, the CBES and NCEBS exhibited notable differences in their levels of inflammatory markers. Specifically, there was a significant decrease in markers, such as hs-CRP (34%, p=0.000), IL6 (63%, p=0.000), and TNF-alpha (12%, p=0.000). However, when comparing these markers between the two groups, the differences in hs-CRP (2.1%, p=0.784) and TNF-alpha (3.5%, p=0.145) were not statistically significant. On the contrary, IL6 exhibited a statistically significant difference of 5.7%, as illustrated in Table 2, Figure 2. This finding substantiates the presence of a statistically significant difference.

Conversely, the CBES group showed smaller changes in inflammatory biomarkers. Hs-CRP slightly increased from 6.5 mg/L to 4.26 mg/L (mean difference: 0.09 mg/L, 95% CI: -0.57 to 0.75, p=0.784, Cohen's d=0.07). Interleukin-6 (IL-6) increased from 53.32 pg/mL to 87.22 pg/mL (mean difference: 4.98 pg/mL, 95% CI: 1.71 to 8.25, p=0.003, Cohen's d=0.81). Tumor necrosis factor-alpha (TNF-alpha) decreased from 191.01 pg/mL to 168.50 pg/mL (mean difference: -5.75 pg/mL, 95% CI: -13.55 to 2.05, p=0.145, Cohen's d=0.39) (Table 3). Regarding lipid profiles, CBES participants (N=32) displayed a decrease in total cholesterol from 189.94 mg/dL to 184.68 mg/dL (mean difference: -7.82 mg/dL, 95% CI: -19.32 to 3.67, p=0.178, Cohen's d=0.36). HDL levels remained nearly unchanged. Triglycerides decreased slightly, from 140.22 mg/dL to 132.71 mg/dL (mean difference: 0.98 mg/dL, 95% CI: -25.45 to 27.43, p=0.940, Cohen's d=0.02). LDL levels exhibited minimal changes. VLDL levels showed slight decreases from 25.33 mg/dL to 19.71 mg/dL (mean difference: -1.78 mg/dL, 95% CI: -6.8 to 3.2, p=0.482, Cohen's d=0.19).

DISCUSSION

The primary objective of this research endeavor was to assess the influence of CBES on inflammatory biomarkers in obese individuals. The key finding of this investigation revealed that while chronotype-based HIIT had a discernible impact on metabolic variables, it did not produce any significant effects on the selected inflammatory biomarkers when contrasted with non-chronotype-based HIIT.

Weight, BMI, and body circumference exhibited improvements in both the CBES and NCEBS groups before and after the implementation of HIIT. Notably, there was no significant disparity between the CBES and NCEBS

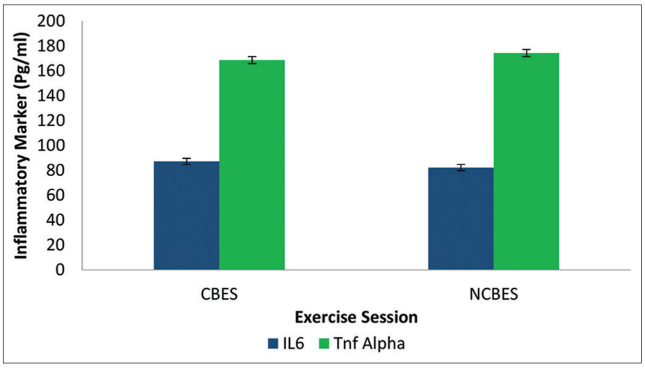

Figure 2. Comparison of Inflammatory marker between CBES and NCBES Groups

**CBES: Chronotype based exercise session, NCBES: Non chronotype based exercise session

Table 2. Anthropometric measures, metabolic and inflammatory biomarkers are compared between CBES and NCBES

Variables	CBES Baseline (N=32)	CBES 12 week N=26	NCBES Baseline N=32	NCBES At 12 weeks N=26	Mean Difference at 12 week	95% CI	P value	Cohen d
Weight	82.31±13.69	77.48±12.9	79.8±11.2	77.23±11.26	0.24±3.23	-6.23 to 6.71	0.940	0.02
Waist Circumference	38.15±3.05	35.44±2.63	37.91±2.74	34.59±3.97	0.84±0.87	-0.90 to 2.59	0.336	0.26
Hip Circumference	45.43±3.89	42.56±3.98	44.92±3,59	41.65±4.35	0.91±1.09	-1.28 to 3.11	0.407	0.22
BMI CBES	30.03±4.16	28.28±3.99	27.8±3.6	26.95±3.73	1.35±1.02	-0.72 to 3.39	0.198	0.34
Fasting Sugar	101.5±4.77	91.79±4.42	98.77±2.85	95.19±8.04	-3.41±1.67	-6.73 to -0.07	0.045	0.54
Fasting Insulin	15.17±2.55	9.06±1.97	14.05±2.3	11.55±2.06	- 2.48±0.53	-3.54 to -1.41	0.000	1.24
HOMA IR	3.79±0.61	2.07±0.46	3.29±0.78	2.73±0.62	-0.66±0.14	-0.94 to -0.37	0.000	1.23
hs CRP CBES	6.5±1.37	4.26±1.36	6.24±1.21	4.17±1.09	0.09±0.33	-0.57 to 0.75	0.784	0.07
IL 6 CBES	53.32±5.87	87.22±6.24	61.55±8.7	82.24±6.11	4.98±1.63	1.71 to 8.25	0.003	0.81
TNF Alpha	191.01±8.38	168.50±15.09	188.11±8.18	174.25±14.33	-5.75±3.8	-13.55 to 2.05	0.145	0.39

*CBES: Chronotype based exercise session, NCBES: Non chronotype based exercise session, CI Confidence interval.

**BMI: Body mass index, hs CRP: high sensitivity C- reactive protein, IL 6: interleukin 6, TNF alpha: Tumor Necrosis factor alpha,.

Table 3. Lipid profile comparison between CBES and NCBES

Variables	CBES Baseline (N=32)	CBES 12 week	NCBES Baseline (N=26)	NCBES 12 weeks	Mean Difference at 12 week	95% CI	P value	Cohen d
Total cholesterol	189.94±22.09	184.68±20.88	196.96±22.65	192.51±22.74	-7.82±5.7	-19.32 to 3.67	0.178	0.36
HDL	47.94±10.25	47.76±8.16	48.23±9.61	47.61±7.4	0.144±2.07	-4.00 to 4.29	0.945	0.02
Triglycerides CBES	140.22±49.56	132.71±47.40	138.77±53.32	131.73±51.84	0.98±13.05	-25.45 to 27.43	0.940	0.02
LDL CBES	122.91±20.70	117.68±19.34	123.04±24.55	117.46±24.55	0.22±5.76	-11.31 to 11.76	0.969	0.01
VLDL CBES	25.33±9.9	19.71±9.03	27.59±11.55	21.49±10.12	-1.78±2.5	-6.8 to 3.2	0.482	0.19

**CBES: Chronotype based exercise session, NCBES: Non chronotype based exercise session, CI: Confidence interval.
**HDL: High density lipoprotein, LDL: Low density lipoprotein, VLDL: Very low-density lipoprotein

groups regarding changes in weight reduction and body circumference, which mirrored the findings of Ryan et al.'s (2020) study. The study concluded that HIIT interventions resulted in noticeable differences between pre- and post-intervention measurements. However, when the outcomes of chronotype-based and non-chronotype-based HIIT sessions were juxtaposed, no distinctions emerged with respect to weight reduction and circumferences (Ryan et al., 2020). However, when the outcomes of chronotype-based and non-chronotype-based HIIT sessions were juxtaposed, no distinctions emerged with respect to weight reduction and circumferences. The rationale behind this observed similarity could be attributed to the notion that chronotype primarily influences wakefulness and performance but may not significantly affect changes in body composition following a period of exercise. Several research studies have also suggested that the timing of exercise does not substantially influence weight loss (Vitale & Weydahl, 2017).

Nevertheless, in our investigation, metabolic variables demonstrated a noteworthy improvement in the HOMA-IR between the CBES and NCEBS groups. This enhancement may be attributable to the preference for carbohydrate oxidation as the primary source of energy during HIIT exercises(Peake et al., 2014). Additionally, reports indicate that the timing of exercise, whether in the morning or evening, can influence glucose control in individuals with diabetes and in obese individuals with insulin resistance (Savikj et al., 2019). This improvement in glucose control may be attributed to the influence of the circadian clock on the regulation of food intake and metabolism (Wolff & Esser, 2019). which aligns with the conclusions drawn by Savikj et al (2019) regarding the potential for scheduled exercise sessions to enhance glycemic control in type 2 diabetes patients (Savikj et al., 2019).

Significant changes were observed in hs-CRP, interleukin-6 (IL-6), and tumor necrosis factor-alpha (TNF-alpha) before and after CBES and NCEBS training. Although no significant differences were noted in hs-CRP and TNF-alpha between the CBES and NCEBS groups, a substantial difference was observed in IL-6 levels. Our findings supporting the notion that HIIT exercise may have a favorable impact on inflammatory biomarkers are substantiated by previous research (Khalafi & Symonds, 2020). However, there is no distinction between the CBES and NCBES groupings. This may be because the impact of HIIT on inflammatory markers is directly proportional to exercise duration and volume, rather than exercise sessions time (Khalafi & Symonds, 2020).

However, these distinctions were not discernible between the CBES and NCEBS groups, suggesting that the influence of HIIT on inflammatory markers may be more closely tied to exercise duration and volume than to the timing of exercise. Moreover, apart from HDL, triglyceride, total cholesterol, and LDL levels witnessed substantial reductions following HIIT in both the CBES and NCEBS groups. These outcomes are in agreement with previous studies that have indicated that obese individuals may experience reductions in cholesterol levels after four weeks of HIIT (Abdelbasset et al., 2019; Minnebeck et al., 2021). Another study examining the immediate effects of HIIT on efficient triglyceride processing following a single session of HIIT reported transient decreases in plasma-free fatty acid levels (Wilhelmsen et al., 2019). However, in our study, no noteworthy disparities in lipid metrics were observed between the CBES and NCEBS groups.

This study has a number of limitations. First, the subjects utilized by each group were distinct. As a result, we were unable to compare individuals' responses to exercise. The same-subject crossover trial design could be used in the future to replicate this type of study. Another drawback is that we did not have control over our participants' behavior.

CONCLUSION

The present study concludes that chronotype-based HIIT yields favorable outcomes regarding metabolic markers among obese individuals. However, it does not appear to exert a significant influence on inflammatory markers in this population. Consequently, the timing of HIIT sessions, whether conducted in the morning or evening, may warrant consideration as a means of mitigating low-grade inflammation. To bolster these findings, future research could employ a crossover design to corroborate and extend the current results.

Ethical Committee Approval

This research works was approved by Institutional Ethical committee, SRM Medical College Hospital & Research Center.

Author Contribution

AJ and MS involved in conception, designing, data collection, data analysis, and manuscript writing, VD involved in 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.

REFERENCES

Abdelbasset, W. K., Tantawy, S. A., Kamel, D. M., Alqahtani, B. A., & Soliman, G. S. (2019). A randomized controlled trial on the effectiveness of 8-week high-intensity interval exercise on intrahepatic triglycerides, visceral lipids, and health-related quality of life in diabetic obese patients with nonalcoholic fatty liver disease. *Medicine (United States)*, *98*(12). https://doi.org/10.1097/MD.0000000000014918

Aloui, K., Abedelmalek, S., Chtourou, H., Wong, D. P., Boussetta, N., & Souissi, N. (2017). Effects of time-of-day on oxidative stress, cardiovascular parameters, biochemical markers, and hormonal response following level-1 Yo-Yo intermittent recovery test. *Physiology International*, *104*(1), 77–90.

Ammar, A., Chtourou, H., & Souissi, N. (2017). Effect of Time-of-Day on Biochemical Markers in Response to Physical Exercise. *Journal of Strength and Conditioning Research*, *31*(1), 272–282. https://doi.org/10.1519/JSC.0000000000001481

Blankenship, J. M., Rosenberg, R. C., Rynders, C. A., Melanson, E. L., Catenacci, V. A., & Creasy, S. A. (2021). Examining the Role of Exercise Timing in Weight Management: A Review. *International Journal of Sports Medicine*, *42*(11), 967–978. https://doi.org/10.1055/a-1485-1293

Bull, F. C., Al-Ansari, S. S., Biddle, S., Borodulin, K., Buman, M. P., Cardon, G., Carty, C., Chaput, J. P., Chastin, S., Chou, R., Dempsey, P. C., Dipietro, L., Ekelund, U., Firth, J., Friedenreich, C. M., Garcia, L., Gichu, M., Jago, R., Katzmarzyk, P. T.,… Willumsen, J. F. (2020). World Health Organization 2020 guidelines on physical activity and sedentary behaviour. *British Journal of Sports Medicine*, *54*(24), 1451–1462. https://doi.org/10.1136/BJSPORTS-2020-102955

Daskapan, A., Tuzun, E. H., & Eker, L. (2006). Perceived Barriers to Physical Activity in University Students. *Journal of Sports Science & Medicine*, *5*(4), 615–615.

DeFronzo, R. A., & Tripathy, D. (2009). Skeletal Muscle Insulin Resistance Is the Primary Defect in Type 2 Diabetes. *Diabetes Care*, *32*(Suppl 2), S157–S163. https://doi.org/10.2337/dc09-S302

F. Adeniyi, A., M. Anjana, R., & B. Weber, M. (2016). Global Account of Barriers and Facilitators of Physical Activity Among Patients with Diabetes Mellitus: A Narrative Review of the Literature. *Current Diabetes Reviews*, *12*(4), 440–448. https://doi.org/10.2174/1573399812666160609102956

Fahed, G., Aoun, L., Zerdan, M. B., Allam, S., Zerdan, M. B., Bouferraa, Y., & Assi, H. I. (2022). Metabolic Syndrome: Updates on Pathophysiology and Management in 2021. *International Journal of Molecular Sciences*, *23*(2). https://doi.org/10.3390/ijms23020786

Fillon, A., Mathieu, M. E., Boirie, Y., & Thivel, D. (2019). Appetite control and exercise: Does the timing of exercise play a role? *Physiology & Behavior*, *218*. https://doi.org/10.1016/j.physbeh.2019.112733ï

Hammouda, O., Chtourou, H., Chaouachi, A., Chahed, H., Bellimem, H., Chamari, K., & Souissi, N. (2013). Time-of-day effects on biochemical responses to soccer-specific endurance in elite Tunisian football players. *Journal of Sports Sciences*, *31*(9), 963–971. https://doi.org/10.1080/02640414.2012.757345

Kadariya, S., & Aro, A. R. (2018). Barriers and facilitators to physical activity among urban residents with diabetes in Nepal. *PloS One*, *13*(6). https://doi.org/10.1371/JOURNAL.PONE.0199329

Keating, S. E., Johnson, N. A., Mielke, G. I., & Coombes, J. S. (2017). A systematic review and meta-analysis of interval training versus moderate-intensity continuous training on body adiposity. *Obesity Reviews*, *18*(8), 943–964. https://doi.org/10.1111/obr.12536

Khalafi, M., & Symonds, M. E. (2020). The impact of high-intensity interval training on inflammatory markers in metabolic disorders: A meta-analysis. *Scandinavian Journal of Medicine and Science in Sports*, *30*(11), 2020–2036. https://doi.org/10.1111/sms.13754

Kong, Z., Fan, X., Sun, S., Song, L., Shi, Q., & Nie, J. (2016). Comparison of high-intensity interval training and moderate-to-vigorous continuous training for cardiometabolic health and exercise enjoyment in obese young women: A randomized controlled trial. *PLoS ONE*, *11*(7). https://doi.org/10.1371/journal.pone.0158589

Maillard, F., Pereira, B., & Boisseau, N. (2018). Effect of High-Intensity Interval Training on Total, Abdominal and Visceral Fat Mass: A Meta-Analysis. *Sports Medicine (Auckland, N.Z.)*, *48*(2), 269–288. https://doi.org/10.1007/S40279-017-0807-Y

Malik, V., & Pundir, C. S. (2002). Determination of total cholesterol in serum by cholesterol esterase and cholesterol oxidase immobilized and co-immobilized on to arylamine glass. *Biotechnology and Applied Biochemistry*, *35*(3), 191–197.

Mancilla, R., Krook, A., Schrauwen, P., & Hesselink, M. K. C. (2020). Diurnal Regulation of Peripheral Glucose Metabolism: Potential Effects of Exercise Timing. *Obesity (Silver Spring, Md.)*, *28 Suppl 1*(Suppl 1), S38–S45. https://doi.org/10.1002/oby.22811

Martland, R., Mondelli, V., Gaughran, F., & Stubbs, B. (2020). Can high-intensity interval training improve physical and mental health outcomes? A meta-review of 33 systematic reviews across the lifespan. *Journal of Sports Sciences*, *38*(4), 430–469. https://doi.org/10.1080/02640414.2019.1706829

Mayoral, L. P. C., Andrade, G. M., Mayoral, E. P. C., Huerta, T. H., Canseco, S. P., Rodal Canales, F. J., Cabrera-Fuentes, H. A., Cruz, M. M., Pérez Santiago, A. D., Alpuche, J. J., Zenteno, E., Ruíz, H. M., Cruz, R. M., Jeronimo, J. H., & Perez-Campos, E. (2020). Obesity subtypes, related biomarkers & heterogeneity. *The Indian Journal of Medical Research*, *151*(1), 11–21. https://doi.org/10.4103/ijmr.IJMR_1768_17

Minnebeck, K., Vorona, E., Zinn, S., Gellner, R., Hinder, J., Brand, S. M., Kabar, I., Alten, F., & Schmitz, B. (2021). Four weeks of high-intensity interval training (HIIT) improve the cardiometabolic risk profile of overweight patients with type 1 diabetes mellitus (T1DM). *European Journal of Sport Science*, *21*(8), 1193–1203. https://doi.org/10.1080/17461391.2020.1810782

Nguyen, X. M. T., Lane, J., Smith, B. R., Nguyen, N. T., Amini, M., Djazayery, A., Khosravi, M., Shafaatdoost, M., Booth, F. W., Roberts, C. K., Laye, M. J., F. Adeniyi, A., M. Anjana, R., B. Weber, M., Daskapan, A., Tuzun, E. H., Eker, L., Mailey, E. L., Huberty, J.,… Hu, F. B. (2016). Short-Term High-Intensity Interval Training on Body Composition and Blood Glucose in Overweight and Obese Young Women. *Journal of Diabetes Research*, *2016*, 4073618–4073618. https://doi.org/10.1155/2016/4073618

Pacific, W. H. Organization. R. O. for the W. (2000). *The Asia-Pacific perspective: Redefining obesity and its treatment*. Sydney : Health Communications Australia. http://iris.wpro.who.int/handle/10665.1/5379

Peake, J. M., Tan, S. J., Markworth, J. F., Broadbent, J. A., Skinner, T. L., & Cameron-Smith, D. (2014). Metabolic and hormonal responses to isoenergetic high-intensity interval exercise and continuous moderate-intensity exercise. *American Journal of Physiology. Endocrinology and Metabolism*, *307*(7), E539–E552. https://doi.org/10.1152/AJPENDO.00276.2014

Ramírez-Vélez, R., Hernández-Quiñones, P. A., Tordecilla-Sanders, A., Álvarez, C., Ramírez-Campillo, R., Izquierdo, M., Correa-Bautista, J. E., Garcia-Hermoso, A., & Garcia, R. G. (2019). Effectiveness of HIIT compared to moderate continuous training in improving vascular parameters in inactive adults. *Lipids in Health and Disease*, *18*(1), 42. https://doi.org/10.1186/s12944-019-0981-z

Ryan, B. J., Schleh, M. W., Ahn, C., Ludzki, A. C., Gillen, J. B., Varshney, P., Van Pelt, D. W., Pitchford, L. M., Chenevert, T. L., Gioscia-Ryan, R. A., Howton, S. M., Rode, T., Hummel, S. L., Burant, C. F., Little, J. P., & Horowitz, J. F. (2020). Moderate-Intensity Exercise and High-Intensity Interval Training Affect Insulin Sensitivity Similarly in Obese Adults. *The Journal of Clinical Endocrinology and Metabolism*, *105*(8), e2941–e2941. https://doi.org/10.1210/CLINEM/DGAA345

Saad, A., Dalla Man, C., Nandy, D. K., Levine, J. A., Bharucha, A. E., Rizza, R. A., Basu, R., Carter, R. E., Cobelli, C., Kudva, Y. C., & Basu, A. (2012). Diurnal pattern to insulin secretion and insulin action in healthy individuals. *Diabetes*, *61*(11), 2691–2700. https://doi.org/10.2337/db11-1478

Salgado, A. L. F. D. A., De Carvalho, L., Oliveira, A. C., Dos Santos, V. N., Vieira, J. G., & Parise, E. R. (2010). Insulin resistance index (HOMA-IR) in the differentiation of patients with non-alcoholic fatty liver disease and healthy individuals. *Arquivos de Gastroenterologia*, *47*(2), 165–169. https://doi.org/10.1590/S0004-28032010000200009

Savikj, M., Gabriel, B. M., Alm, P. S., Smith, J., Caidahl, K., Björnholm, M., Fritz, T., Krook, A., Zierath, J. R., & Wallberg-Henriksson, H. (2019). Afternoon exercise is more efficacious than morning exercise at improving blood glucose levels in individuals with type 2 diabetes: A randomised crossover trial. *Diabetologia*, *62*(2), 233–237. https://doi.org/10.1007/s00125-018-4767-z

Shim, J.-S., Oh, K., & Kim, H. C. (2014). Dietary assessment methods in epidemiologic studies. *Epidemiology and Health*, *36*, e2014009–e2014009. https://doi.org/10.4178/EPIH/E2014009

Shiotani, H., Umegaki, Y., Tanaka, M., Kimura, M., & Ando, H. (2009). Effects of aerobic exercise on the circadian rhythm of heart rate and blood pressure. *Chronobiology International*, *26*(8), 1636–1646. https://doi.org/10.3109/07420520903553443

Souissi, N., Gauthier, A., Sesboüé, B., Larue, J., & Davenne, D. (2004). Circadian rhythms in two types of anaerobic cycle leg exercise: Force-velocity and 30-s Wingate tests. *International Journal of Sports Medicine*, *25*(1), 14–19. https://doi.org/10.1055/S-2003-45226

Trine, M. R., & Morgan, W. P. (1995). Influence of time of day on psychological responses to exercise. A review. *Sports Medicine (Auckland, N.Z.)*, *20*(5), 328–337. https://doi.org/10.2165/00007256-199520050-00004

Tsai, H. H., Lin, C. P., Lin, Y. H., Hsu, C. C., & Wang, J. S. (2016). High-intensity Interval training enhances mobilization/functionality of endothelial progenitor cells and depressed shedding of vascular endothelial cells undergoing hypoxia. *European Journal of Applied Physiology*, *116*(11–12), 2375–2388. https://doi.org/10.1007/S00421-016-3490-Z

van Baak, M. A., Pramono, A., Battista, F., Beaulieu, K., Blundell, J. E., Busetto, L., Carraça, E. V., Dicker, D., Encantado, J., Ermolao, A., Farpour-Lambert, N., Woodward, E., Bellicha, A., & Oppert, J. M. (2021). Effect of different types of regular exercise on physical fitness in adults with overweight or obesity: Systematic review and meta-analyses. *Obesity Reviews : An Official Journal of the International Association for the Study of Obesity*, *22 Suppl 4*(Suppl 4). https://doi.org/10.1111/OBR.13239 van Moorsel, D., Hansen, J., Havekes, B., Scheer, F. A. J. L., Jörgensen, J. A., Hoeks, J., Schrauwen-Hinderling, V. B., Duez, H., Lefebvre, P., Schaper, N. C., Hesselink, M. K. C., Staels, B., & Schrauwen, P. (2016). Demonstration of a day-night rhythm in human skeletal muscle oxidative capacity. *Molecular Metabolism*, *5*(8), 635–645. https://doi.org/10.1016/j.molmet.2016.06.012

Vella, C. A., Taylor, K., & Drummer, D. (2017). High-intensity interval and moderate-intensity continuous training elicit similar enjoyment and adherence levels in overweight and obese adults. *European Journal of Sport Science*, *17*(9), 1203–1211. https://doi.org/10.1080/17461391.2017.1359679

Vitale, J. A., & Weydahl, A. (2017). Chronotype, Physical Activity, and Sport Performance: A Systematic Review. *Sports Medicine (Auckland, N.Z.)*, *47*(9), 1859–1868. https://doi.org/10.1007/S40279-017-0741-Z

Westwood, A., Bullock, D. G., & Whitehead, T. P. (1986). An examination of the hexokinase method for serum glucose assay using external quality assessment data. *Annals of Clinical Biochemistry*, *23*(1), 92–96. https://doi.org/10.1177/000456328602300111

WHO. (2021). Obesity and overweight. *WHO*. https://www.who.int/news-room/fact-sheets/detail/obesity-and-overweight

WHO. (2022). Physical activity. *WHO*. https://www.who.int/news-room/fact-sheets/detail/physical-activity

Wilhelmsen, A., Mallinson, J., Jones, R., Cooper, S., Taylor, T., & Tsintzas, K. (2019). Chronic effects of high-intensity interval training on postprandial lipemia in healthy men. *Journal of Applied Physiology (Bethesda, Md. : 1985)*, *127*(6), 1763–1771. https://doi.org/10.1152/japplphysiol.00131.2019

Wolff, C. A., & Esser, K. A. (2019). Exercise timing and circadian rhythms. *Current Opinion in Physiology*, *10*, 64–69. https://doi.org/10.1016/j.cophys.2019.04.020

Author Query???

AQ1: Kindly cite figure 1 in the text part