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Acute and Chronic Serum FGF21 and Irisin Responses to 12 Weeks of High-Intensity Interval Training in Overweight Young Males: A Preliminary Study on The Browning Process

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Abstract

This study aimed to examine changes in body composition and serum fibroblast growth factor 21 (FGF21) and irisin concentrations following 12 weeks of high-intensity interval training (HIIT) in young men with overweight and to characterize their responses to individual exercise sessions during the initial (week 1) and final (week 12) phases of the intervention. This exploratory, single-arm, pre–post study recruited sedentary men aged 18–25 years with a body mass index (BMI) of 25.0–29.9 kg/m². Fifteen participants initiated a 12-week HIIT intervention consisting of three sessions per week. Seven participants completed the intervention, and six were included in the final analysis after one participant was excluded post hoc because of anomalous myokine measurements. Blood samples were collected before exercise, immediately after exercise, and at 1 and 3 hours postexercise during weeks 1 and 12. Serum FGF21 and irisin concentrations were measured using enzyme-linked immunosorbent assays (ELISAs), while body composition was assessed using bioelectrical impedance analysis. After 12 weeks of HIIT, body fat percentage decreased in all six analyzed participants, from 27.3 ± 4.0% to 25.3 ± 5.0% ($p = 0.031$), whereas body mass remained relatively unchanged (79.08 ± 8.14 kg vs. 78.67 ± 8.80 kg; $p = 0.719$). Changes in absolute fat mass were not statistically significant. During the initial exercise session, neither serum FGF21 nor irisin concentrations increased significantly. At week 12, preexercise serum FGF21 concentrations were lower than those recorded at baseline in week 1, whereas concentrations measured 3 hours postexercise reached the highest observed values across the study. However, these differences were not statistically significant ($p = 0.144$ for the within-session comparison at week 12). Serum irisin concentrations exhibited no statistically significant changes during either assessment phase. Overall, 12 weeks of HIIT was associated with a significant reduction in body fat percentage without a corresponding significant change in body mass. However, no statistically significant changes in circulating FGF21 or irisin concentrations were detected. Given the small sample size, substantial participant attrition, and absence of a control group, these preliminary findings should be interpreted cautiously and do not establish whether HIIT induces adipose tissue browning.

INTRODUCTION

Overweight and obesity are major contributors to cardiometabolic diseases, and their prevalence continues to increase in Indonesia and worldwide (Kementerian Kesehatan Republik Indonesia, 2019). Excess adiposity results from a sustained positive energy balance; however, changes in body weight alone do not adequately reflect the metabolic adaptations associated with exercise (Gaesser & Angadi, 2021; GBD 2015 Obesity Collaborators, 2017; World Health Organization, n.d.). Regular exercise may reduce fat mass while preserving or increasing fat-free mass, whereas compensatory changes in appetite and nonexercise physical activity may attenuate weight loss (Batitucci et al., 2022; Melanson et al., 2013). These observations have stimulated growing interest in the physiological mechanisms through which exercise influences adipose tissue metabolism beyond the immediate energy expenditure associated with individual exercise sessions.

One proposed mechanism involves the browning of white adipose tissue (WAT), a process characterized by the emergence of thermogenically active beige adipocytes within white adipose tissue depots (Machado et al., 2022). Experimental studies have demonstrated that beige adipocytes can express uncoupling protein 1 (UCP1), which facilitates mitochondrial proton leak and contributes to nonshivering thermogenesis (Fernández-Verdejo et al., 2019; Mu et al., 2021). Fibroblast growth factor 21 (FGF21) and irisin have been implicated in the regulation of pathways associated with this process. FGF21 is a metabolic regulatory hormone involved in energy homeostasis and the modulation of mitochondrial oxidative metabolism (Chau et al., 2010). Irisin, a peptide hormone generated through proteolytic cleavage of fibronectin type III domain-containing protein 5 (FNDC5), was initially reported to promote a brown adipocyte-like phenotype in white adipose tissue (Boström et al., 2012). Accordingly, exercise-induced communication between skeletal muscle and adipose tissue provides a plausible biological framework for investigating the potential involvement of these circulating factors in metabolic adaptations to physical activity (Leal et al., 2018).

Nevertheless, evidence supporting exercise-induced adipose tissue browning in humans remains inconsistent. Studies directly examining adipose tissue characteristics and brown adipose tissue activity have not consistently demonstrated increased thermogenic activity or beige adipocyte recruitment following exercise interventions (Martinez-Tellez et al., 2022; Tsiloulis et al., 2018; Vidal & Stanford, 2020). Methodological considerations further complicate the interpretation of circulating biomarkers. FGF21 concentrations exhibit substantial diurnal variation and may reflect secretion from multiple tissues, including the liver and skeletal muscle, rather than adipose tissue-specific activity (Cuevas-Ramos et al., 2012; Khalafi et al., 2021; Kim et al., 2013; Yu et al., 2011). Similarly, the analytical validity of commercially available irisin immunoassays has been questioned because of potential antibody cross-reactivity, limited assay specificity, and discrepancies between immunoassay-based measurements and mass spectrometry-based quantification (Albrecht et al., 2015; Jedrychowski et al., 2015). Consequently, circulating FGF21 and irisin concentrations should be regarded as indirect biomarkers of metabolic regulation rather than definitive indicators of adipose tissue browning.

Previous investigations have reported both acute exercise-induced responses and longer-term training-associated changes in circulating FGF21 and irisin concentrations. However,

findings have varied considerably according to participant characteristics, exercise intensity, training modality, intervention duration, and blood sampling schedules (Azali Alamdari & Khalafi, 2018; He et al., 2018; Huh et al., 2012; Jandova et al., 2021; Khalafi et al., 2021; Liu et al., 2024; Nygaard et al., 2015; Pekkala et al., 2013; Tofighi et al., 2017; Tsuchiya et al., 2014). These inconsistencies suggest that the temporal dynamics of circulating biomarker responses warrant further investigation. Repeated blood sampling at predetermined intervals following exercise may provide additional information regarding transient hormonal responses that would otherwise remain undetected when assessments are restricted to resting concentrations. Therefore, the present study employed an identical blood sampling schedule during the initial and final weeks of a 12-week high-intensity interval training (HIIT) intervention to characterize acute postexercise responses and potential training-associated changes in circulating FGF21 and irisin concentrations.

Building on previous experimental research conducted by our group, which demonstrated increased FGF21 secretion following high-intensity intermittent training in obese rats (Kartinah et al., 2018), the present study investigated whether comparable circulating responses could be observed in humans. Specifically, this exploratory study aimed to assess changes in body composition and characterize serum FGF21 and irisin responses in young men with overweight who participated in a 12-week HIIT intervention. Blood samples were collected before exercise, immediately after exercise, and at 1 and 3 hours postexercise during weeks 1 and 12. This repeated-measures approach was designed to examine the temporal patterns of circulating FGF21 and irisin responses to individual exercise sessions at the beginning and end of the training intervention. Although these biomarkers have been associated with pathways implicated in adipose tissue browning, the study did not directly assess beige adipocyte formation, UCP1 expression, or adipose tissue thermogenic activity. Therefore, the findings were intended to provide preliminary evidence regarding exercise-associated changes in circulating metabolic biomarkers rather than direct confirmation of adipose tissue browning in humans.

METHOD

Study design and setting

This study employed a single-arm, nonrandomized, pre–post design with repeated measurements of acute exercise responses. All participants underwent the same 12-week high-intensity interval training (HIIT) intervention. Serum fibroblast growth factor 21 (FGF21) and irisin concentrations were designated as the prespecified primary outcomes, while body composition parameters were assessed as secondary outcomes. Participant screening, body composition assessments, blood sampling, and supervised exercise sessions during weeks 1 and 12 were conducted at the Sports and Exercise Studies Cluster, Indonesian Medical Education and Research Institute (IMERI), Faculty of Medicine, Universitas Indonesia. Biochemical analyses were performed at the Department of Biochemistry and Molecular Biology, Faculty of Medicine, Universitas Indonesia.

Participants

Men aged 18-25 years living in Greater Jakarta were eligible if their body mass index (BMI) was 25.0-29.9 kg/m², they were non-smokers, and they had performed less than 150

minutes of structured physical activity per week during the previous six months. Exclusion criteria included epilepsy, asthma, cardiac disease, renal or hepatic impairment, and previous fracture, tendon injury, or ligament injury that could make vigorous exercise unsafe or difficult to interpret. Written informed consent was obtained before study procedures.

Fifteen participants started the programme. Seven completed both sampling days. One completer was excluded after data collection because FGF21 and irisin concentrations were markedly different from those of the other participants at all time points in both phases and no assay problem could be identified retrospectively. This exclusion was post hoc and was not pre-specified. All analyses therefore refer to the same six participants. The consequence of this decision is addressed in the limitations.

Sample size

The approved protocol planned to recruit 15 participants, consistent with common rules of thumb for preliminary physiological studies (Hertzog, 2008; Julious, 2005). No formal power calculation was performed because an estimate of within-participant variance for the repeated post-exercise myokine response was not available for this population. Attrition during the pandemic reduced the complete analysable sample to six.

HIIT intervention

Participants were prescribed three sessions per week on alternate days for 12 weeks. Each session included 3-5 minutes of stretching, 3-5 minutes of jogging with short accelerations, 2-3 minutes of recovery, the HIIT circuit, and a 3-5 minute cool-down. The circuit comprised five bodyweight movements: a 4-m shuttle run, 4-m zigzag shuttle run, 2-m box run, star jumps, and burpees. Each movement was performed in 8-second maximal-effort bouts separated by 25 seconds of active recovery. The number of bouts per movement increased from three in week 1 to fourteen in week 12. This increased training volume while keeping the requested effort during each work bout maximal.

Table 1. Progression of session volume across the 12-week programme.

Week	Warm-up (min)	Interval repetitions	HIIT block duration	Cool-down (min)
1	14.5	3	16 min 10 s	10
2	14.5	4	18 min 55 s	10
3	14.5	5	21 min 40 s	10
4	14.5	6	24 min 25 s	10
5	14.5	7	27 min 10 s	10
6	14.5	8	29 min 55 s	10
7	14.5	9	32 min 40 s	10
8	14.5	10	35 min 25 s	10
9	14.5	11	38 min 10 s	10
10	14.5	12	40 min 55 s	10
11	14.5	13	43 min 40 s	10
12	14.5	14	46 min 25 s	10

Heart rate was monitored with a Polar H9 chest sensor. Because directly measured maximal heart rate was not available, intensity was expressed relative to age-predicted maximal heart rate (220-age), with 85% used as the lower threshold for high intensity. Sessions conducted away from IMERI were recorded by participants and reviewed weekly. The study took place during public-activity restrictions, so training was divided between on-site and remote sessions.

Body composition

Body mass, fat mass, fat-free mass, and body fat percentage were assessed by bioelectrical impedance before the first session and after the final session. Measurements were performed in the morning after an overnight fast and voiding, with participants in light clothing. Participants were asked to avoid vigorous activity for 24 hours and alcohol for 48 hours before measurement.

Blood sampling and biochemical analysis

Blood was collected during the first session of week 1, designated the acute phase, and the final session of week 12, designated the chronic phase. These terms are retained from the original study report and are used throughout. On each sampling day, 5 mL of venous blood was drawn immediately before exercise, immediately after exercise, and 1 and 3 hours after exercise. This schedule covered the early period in which acute FGF21 and irisin responses have been reported (He et al., 2018; Khalafi et al., 2021). Sampling sessions began within the same two-hour morning window to reduce the effect of FGF21 diurnal variation (Yu et al., 2011). Participants were asked to reproduce their previous evening meal pattern before the week-12 visit. Serum was separated after centrifugation at 3000 rpm for 10 minutes and stored at -20 °C until batch analysis.

Serum FGF21 and irisin were measured by enzyme-linked immunosorbent assay (R&D Systems). Samples from the same participant were analysed on the same plate and in the same run to minimise between-assay variation in within-participant comparisons.

Statistical analysis

Analyses were performed in SPSS version 25. Normality was assessed using the Shapiro-Wilk test. Paired t-tests were used for normally distributed paired comparisons and Wilcoxon signed-rank tests otherwise. A two-sided p value <=0.05 was considered statistically significant. Given the small sample, emphasis was placed on the size and direction of changes rather than on statistical significance alone. Mean differences and 95% confidence intervals are reported where available.

Spearman rank correlation was used descriptively to examine whether change in fat mass corresponded with change in resting FGF21 or with the week-12 post-exercise FGF21 excursion. No area-under-the-curve analysis was performed because only four samples were available over three hours and there was no time-matched non-exercise day to separate exercise-related change from the known diurnal pattern of FGF21 (Yu et al., 2011).

Ethics

The protocol was approved by the Health Research Ethics Committee, Faculty of Medicine, Universitas Indonesia-RSUPN Dr. Cipto Mangunkusumo (408/UN2.F1/ETIK/PPM.00.02/2020; amendment ND-490/UN2.F1/ETIK/PPM.00.02/2021). The study was conducted in accordance with the Declaration of Helsinki.

RESULTS AND DISCUSSION

Participant flow and intervention delivery

Forty-two men expressed interest, 31 attended eligibility assessment, and 15 started the programme. Eight discontinued: five after SARS-CoV-2 infection and three for personal reasons unrelated to the intervention. Seven completed the programme and week-12 sampling;

one was excluded post hoc as described above, leaving six participants for analysis. No musculoskeletal or cardiovascular adverse events were reported during 96 documented sessions.

Session documentation was incomplete. Of 216 prescribed sessions among the six analysed participants, 96 (44%) were logged. Two participants logged 34/36 and 28/36 sessions, whereas the other four logged 7-9 sessions. Unlogged sessions could not be distinguished from sessions that were not performed. Among logged sessions, 48 were on site and 48 were remote. Across 474 movement-level recordings, mean peak heart rate was 83% of age-predicted maximum, close to the 85% threshold, which 45% of recorded bouts exceeded.

Table 2. Baseline characteristics of analysed participants (n=6).

Variable	Mean ± SD
Age (years)	22.0 ± 1.5
Height (cm)	170.6 ± 5.2
Body mass (kg)	79.08 ± 8.14
BMI (kg/m²)	27.1 ± 1.8
Fat mass (kg)	21.75 ± 5.03
Fat-free mass (kg)	57.3 ± 4.4
Body fat (%)	27.3 ± 4.0
VO_2max (mL/kg/min; n=5)	40.7 ± 6.6

Body composition

Body mass changed little after 12 weeks (79.08±8.14 to 78.67±8.80 kg; mean change -0.42 kg, 95% CI -3.19 to 2.36; p=0.719). Fat mass decreased from 21.75±5.03 to 20.10±5.35 kg (mean change -1.65 kg, 95% CI -3.33 to 0.03; p=0.054), while fat-free mass increased from 57.33±4.39 to 58.57±5.86 kg (p=0.219). Body fat percentage decreased in all six participants, from 27.25±3.98% to 25.28±4.99% (p=0.031). BMI was unchanged (27.12±1.76 to 26.98±2.19 kg/m²; p=0.875). Changes in body mass, fat mass, and body fat percentage are shown in Figure 1.

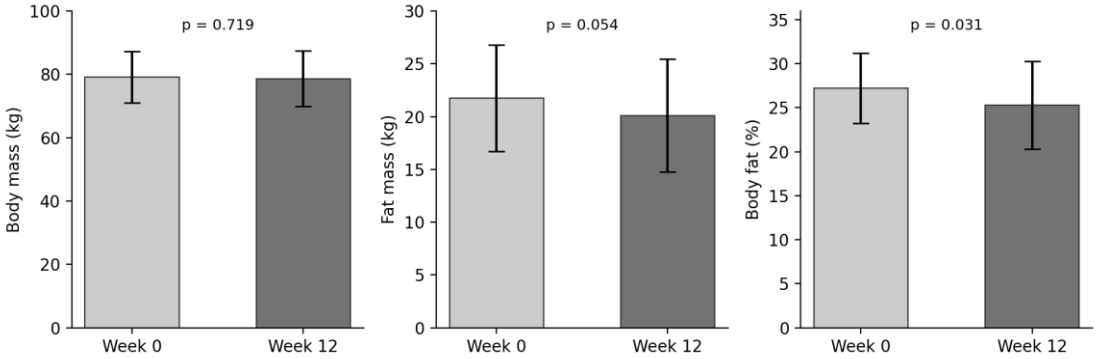


Figure 1. Body mass, fat mass, and body fat percentage before and after the 12-week programme. Bars show mean ± SD (n=6).

Serum FGF21 and irisin

No statistically significant within-phase or between-phase comparison was detected for FGF21 (Table 2). In the acute phase, the 3-hour concentration was 70.7 pg/mL (16.5%) below the pre-exercise value. In the chronic phase, the pre-exercise concentration was lower than its acute-phase counterpart, while the 3-hour value was 149.8 pg/mL (38.5%) above its own pre-

exercise value and was the highest mean concentration recorded. The chronic-phase pre-to-3-hour comparison did not reach statistical significance (p=0.144).

Table 3. Serum FGF21 concentrations (pg/mL; mean ± SD) in the acute phase (week 1) and the chronic phase (week 12).

Time point	Acute phase	p vs pre	Chronic phase	p vs pre	p acute vs chr
Pre-exercise	429.07 ± 284.52	-	388.73 ± 205.21	-	0.681
Immediately post	431.00 ± 163.91	0.981	468.88 ± 300.86	0.161	0.805
1 h post	357.09 ± 182.40	0.399	394.68 ± 249.98	0.917[a]	0.753[a]
3 h post	358.39 ± 128.80	0.465	538.56 ± 328.98	0.144	0.181

[a] Wilcoxon signed-rank test; other comparisons used paired t-tests.

Irisin remained stable in both phases (Table 3). Mean concentrations varied by less than approximately 3% from the pre-exercise value, and no comparison reached statistical significance. Dispersion was greater in the chronic phase than in the acute phase.

Table 4. Serum irisin concentrations (pg/mL; mean ± SD) in the acute phase (week 1) and the chronic phase (week 12).

Time point	Acute phase	p vs pre	Chronic phase	p vs pre	p acute vs chr
Pre-exercise	1361.50 ± 83.42	-	1358.59 ± 229.79	-	0.968
Immediately post	1366.03 ± 158.58	0.911	1404.68 ± 291.94	0.183	0.511
1 h post	1316.49 ± 94.34	0.060	1397.51 ± 325.88	0.410	0.448
3 h post	1339.55 ± 80.32	0.257	1394.29 ± 281.21	0.223	0.610

Serum FGF21 and irisin time courses in both phases are shown in Figure 2.

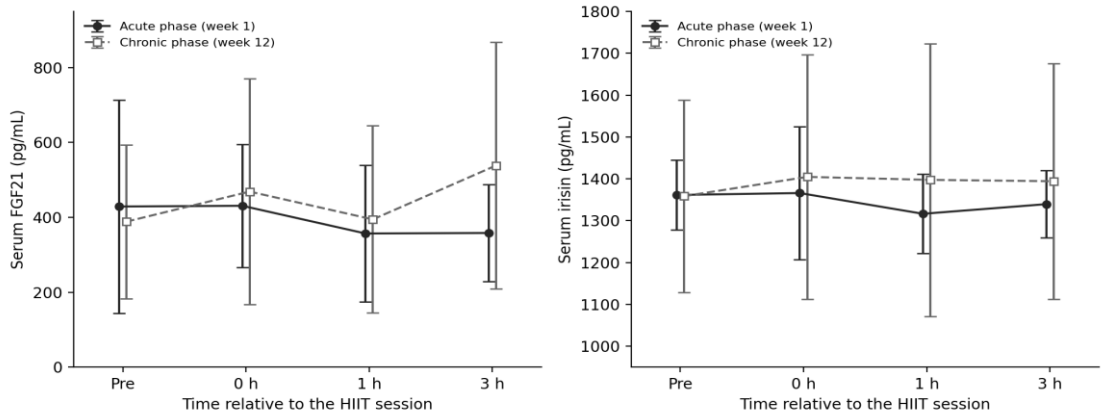


Figure 2. Serum FGF21 (left) and irisin (right) before, immediately after, and 1 and 3 hours after a HIIT session, in the acute phase (week 1) and the chronic phase (week 12). Values are mean ± SD (n=6).

At participant level, change in fat mass did not clearly correspond with change in resting FGF21 (Spearman ρ=+0.54, p=0.27) or with the size of the chronic-phase post-exercise excursion (ρ=-0.03, p=0.96). With n=6, these correlations are descriptive only.

Main findings

The main finding was the absence of a detectable acute increase in serum FGF21 or irisin, in both the acute and the chronic phase. At the same time, body fat percentage decreased in every analysed participant while body mass remained stable. The biomarker and body-composition findings therefore did not move in parallel. These data do not demonstrate

adipose-tissue browning and should not be interpreted as evidence for or against tissue-level browning, because neither UCP1 expression nor brown/beige adipose tissue activity was measured.

Heart rate is an imperfect record of the effort delivered in this protocol. Mean peak heart rate was 83% of age-predicted maximum, just below the 85% threshold, and this probably understates the effort achieved. Heart rate rises with a lag of several tens of seconds after the onset of intense work, so in 8-second maximal bouts it cannot reach its steady-state value before the bout ends, and heart-rate thresholds derived from longer intervals are a poor index of intensity for very short sprints (Buchheit & Laursen, 2013). Age-predicted maximal heart rate also carries an individual error of around ten beats per minute (Tanaka et al., 2001), enough to move a bout across the 85% threshold. Even so, the achieved stimulus cannot be verified, and the negative biomarker findings therefore have two possible explanations that cannot be separated here: the circulating markers may not respond consistently to this type of exercise in humans, or the achieved exercise stimulus may have been insufficient or too variable to produce a measurable response.

FGF21 response

Acute FGF21 responses to exercise vary across studies. Meta-analytic data suggest that FGF21 can increase in the early post-exercise period, but the size and timing of the response depend on the exercise stimulus and participant characteristics (Khalafi et al., 2021). Other human studies have reported increased FGF21 after acute or repeated exercise (Cuevas-Ramos et al., 2012; Kim et al., 2013), and a recent meta-analysis found that longer exercise interventions may reduce resting FGF21 (Liu et al., 2024). These findings show why a single resting sample cannot fully describe training-related changes in FGF21 physiology.

In the present study, the acute-phase and chronic-phase FGF21 profiles differed descriptively. FGF21 was lower at 3 hours than before exercise in the acute phase, whereas it was higher at 3 hours in the chronic phase. Neither comparison was statistically significant. The pattern may reflect differences in the post-exercise response after training, but the present sample is too small to establish this as a training effect. The repeated sampling schedule was useful for showing the time course rather than relying only on a single resting measurement.

One possible explanation is altered FGF21 regulation after training. Obesity has been described as a state of relative FGF21 resistance (Fisher et al., 2010), and exercise may lower resting FGF21 in some populations (Azali Alamdari & Khalafi, 2018; Liu et al., 2024). However, serum concentration alone cannot distinguish altered secretion from altered tissue responsiveness. The liver, skeletal muscle, and adipose tissue can all contribute to circulating FGF21 (Kim et al., 2013), and target-tissue measures such as β-Klotho/FGFR1c signalling were not available. The observed chronic-phase pattern should therefore be treated as a hypothesis for a future controlled study, not as evidence of restored FGF21 sensitivity.

Irisin and measurement limitations

Irisin showed little change across the sampling period. Previous studies have reported transient increases after intense endurance or strength exercise, later increases, or no acute change (He et al., 2018; Huh et al., 2012; Nygaard et al., 2015; Pekkala et al., 2013; Tsuchiya et al., 2014). Long-term exercise studies are similarly heterogeneous (Jandova et al., 2021). This inconsistency may reflect differences in exercise modality and intensity, but analytical

limitations are also important. Commercial irisin antibodies may cross-react with unrelated proteins, and mass spectrometry has produced concentrations that differ substantially from many ELISA-based estimates (Albrecht et al., 2015; Jedrychowski et al., 2015). Consequently, the present irisin results are best interpreted as ELISA-derived measurements rather than definitive quantification of circulating irisin. Indonesian data on this marker remain limited; circulating irisin has been examined in relation to exercise intensity in adults with overweight and obesity (Abdulloh et al., 2021), but we are not aware of a previous report measuring FGF21 and irisin together across a post-exercise time course in this population

Body composition and its relation to the myokine findings

Body mass did not change, but fat mass decreased and fat-free mass increased. This pattern is plausible in previously inactive young men beginning structured exercise, and interval training of similar frequency and duration has previously been reported to leave body mass unchanged in sedentary adults (Astorino et al., 2013). It also illustrates why body mass alone may underestimate training-related changes in body composition (Gaesser & Angadi, 2021). Compensatory changes in appetite or non-exercise activity could also have limited weight loss, but neither diet nor free-living activity was measured (Batitucci et al., 2022; Melanson et al., 2013). One further point of classification is relevant: although a BMI of 25.0-29.9 kg/m² denotes overweight under the standard WHO categories, the Asia-Pacific criteria set the obesity threshold at 25 kg/m² because body fat and cardiometabolic risk are higher at a given BMI in Asian populations (WHO Expert Consultation, 2004), so every participant here would be classified as having obesity class I under the criteria applicable to them.

The body-composition findings did not provide evidence that FGF21 or irisin mediated the reduction in fat mass. Fat loss was not clearly related to resting FGF21 or to the post-exercise FGF21 excursion, and irisin remained nearly unchanged. More importantly, circulating myokines are indirect markers of adipose-tissue phenotype. Human studies using adipose-tissue sampling or imaging have not consistently shown exercise-induced browning (Martinez-Tellez et al., 2022; Tsiloulis et al., 2018). Thus, the safer interpretation is that body composition changed during the programme without a parallel detectable change in these circulating biomarkers.

Strengths, limitations, and implications for future studies

A strength of the study was the use of the same four-point blood-sampling schedule at the beginning and end of the 12-week programme, a design rarely used in this literature, where training effects on these myokines have usually been assessed from a single resting sample before and after a programme (Azali Alamdari & Khalafi, 2018; Tofighi et al., 2017). This allowed both the acute post-exercise profile and the change after training to be described in the same participants. The study also evaluated body composition together with two circulating factors linked to the proposed browning pathway.

The limitations are substantial. Only six participants were analysed, and there was no non-exercising control group. One of seven completers was excluded post hoc based on the primary biomarker values, which can bias estimates in a sample this small. Training adherence could not be established because most prescribed sessions were not logged, and heart rate could verify the prescribed intensity only partially in 8-second bouts. Maximal heart rate was estimated rather than measured directly. Diet and non-exercise activity were not recorded.

Body composition was assessed by bioelectrical impedance rather than DXA (Achamrah et al., 2018). Irisin was measured by ELISA without mass-spectrometric confirmation (Albrecht et al., 2015; Jedrychowski et al., 2015). Sampling ended at 3 hours, so later responses may have been missed, and no time-matched non-exercise day was included to separate exercise-related FGF21 change from its diurnal rhythm (Yu et al., 2011). Pandemic restrictions also affected supervision and retention.

Future studies should include a larger sample, a control group, and a time-matched non-exercise sampling day. Exercise intensity and adherence should be verified prospectively. Later sampling points may be useful for irisin, while DXA would strengthen body-composition assessment. If adipose-tissue browning is the primary biological question, direct tissue or imaging endpoints should be included rather than inferred from circulating FGF21 and irisin alone (Martinez-Tellez et al., 2022; Tsiloulis et al., 2018).

CONCLUSION

In this exploratory study, 12 weeks of progressive high-intensity interval training (HIIT) was associated with a reduction in body fat percentage without a statistically significant change in body mass among young men with overweight. Serum fibroblast growth factor 21 (FGF21) and irisin concentrations did not exhibit statistically significant acute increases following exercise during either the initial (week 1) or final (week 12) assessment. Although the temporal profile of serum FGF21 concentrations showed descriptive differences following the training intervention, these changes were not statistically significant. Therefore, the observed patterns should be interpreted cautiously and require confirmation in larger, adequately powered studies.

Overall, the findings did not establish whether the observed reduction in body fat percentage was mediated by FGF21- or irisin-associated adipose tissue browning. The small sample size, absence of a control group, incomplete training records, and variability in the exercise intensity achieved by participants limited the interpretation and generalizability of the results. Future studies should incorporate larger samples, appropriate control groups, standardized monitoring of exercise intensity, and direct assessments of adipose tissue browning, including uncoupling protein 1 (UCP1) expression and thermogenic activity. Such investigations are necessary to clarify the potential roles of FGF21 and irisin in exercise-induced metabolic adaptations and to determine whether HIIT promotes adipose tissue browning in humans.

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CONFLICT OF INTEREST

The author states that there are no conflicts of interest, whether personal, commercial, or financial, that can affect the implementation and results of this study.

AUTHOR'S CONTRIBUTION

Author contributes to research design, data collection and processing, system development, result analysis, and article manuscript preparation.

GENERATIVE ARTIFICIAL INTELLIGENCE USAGE STATEMENT

In the process of compiling this article, the author uses generative artificial intelligence (AI) as a tool in language refinement, writing structure, and grammar improvement. The use of AI is carried out on a limited basis and remains under the supervision of the author. All data, analysis, interpretation, and scientific substance in this article have been checked and verified by the author. The author is solely responsible for the accuracy, authenticity, and final content of the manuscript.

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