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Time Matters: Acute Physiological and Psychological Effects of Different Exercise Durations in Prediabetes

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Abstract

Background: Exercise is a key non-pharmacological strategy for improving metabolic and psychological health in individuals with prediabetes. While the effects of exercise intensity are well documented, the acute impact of *exercise duration* on metabolic biomarkers and perceived stress remains unclear. This study examined the immediate effects of four different exercise durations on serum cholesterol, Irisin levels, and perceived stress in sedentary adults with prediabetes. **Methods:** Twenty-two adults with prediabetes completed a laboratory-based crossover trial consisting of four treadmill exercise sessions of increasing duration: 15 minutes (Visit 1), 30 minutes (Visit 2), 45 minutes (Visit 3), and 60 minutes or maximum tolerable duration (Visit 4). All sessions were performed at moderate intensity following an isocaloric breakfast. Blood samples were collected immediately before and after each session to measure total cholesterol and Irisin concentrations. Perceived Stress Scale (PSS-10) scores were recorded pre- and post-exercise. Paired t-tests assessed within-visit changes, and repeated measures ANOVA compared changes across durations. **Results:** Total cholesterol decreased significantly after each exercise duration

(Visit 1: −23.2 mg/dL, p=0.001; Visit 2: −16.6 mg/dL, p<0.001; Visit 3: −12.4 mg/dL, p=0.001; Visit 4: −21.2 mg/dL, p=0.006). Irisin levels increased after all sessions, with significant rises in Visits 2–4 (p<0.05) and a non-significant increase during the shortest duration (Visit 1: p=0.120). Repeated measures ANOVA showed no significant differences in delta cholesterol or delta Irisin across visits (p>0.05). PSS-10 scores did not change significantly within individual visits but improved significantly from Visit 1 to Visit 4 (p=0.02), indicating cumulative psychological benefit. **Conclusion:** Acute bouts of exercise, even as short as 15 minutes, produce meaningful reductions in cholesterol, while longer durations elicit stronger Irisin responses and gradual improvements in perceived stress. These findings highlight the effectiveness of varying exercise durations in improving metabolic health in prediabetic adults and support flexible, tolerance-based exercise prescriptions.

INTRODUCTION

Prediabetes represents a critical intermediate state between normoglycemia and type 2 diabetes mellitus (T2DM), characterized by impaired fasting glucose, altered insulin sensitivity, and early metabolic dysfunction (1,2). With global prevalence rising sharply, affecting an estimated 374 million adults by 2045 prediabetes is now recognized as one of the most urgent metabolic health challenges worldwide (3). Individuals with prediabetes exhibit a markedly increased risk of progressing to T2DM, cardiovascular disease, dyslipidemia, and chronic inflammation, thereby necessitating early lifestyle-based interventions (4,5). Among these interventions, structured aerobic exercise is considered one of the most effective non-pharmacological strategies for improving metabolic health and preventing progression to diabetes (6).

While the beneficial effects of exercise on insulin sensitivity, glycemic control, lipid metabolism, and inflammatory markers are well established, the optimal exercise dose—particularly the role of exercise duration—remains inadequately understood (7,8). Previous studies have often emphasized exercise intensity or frequency, leaving gaps regarding how variations in session length influence acute hormonal and metabolic responses. Exercise duration is a key determinant of total energy expenditure and muscular workload, both of which influence lipid oxidation, substrate mobilization, and

secretion of myokines such as Irisin, a muscle-derived hormone that promotes browning of white adipose tissue and enhances glucose homeostasis (9–11). Irisin has emerged as an important biomarker linking physical activity with metabolic improvements, yet findings on its responsiveness to different exercise durations remain inconsistent. Some studies suggest that longer exercise bouts elicit greater Irisin release due to increased muscular activation and thermogenic demand (12,13), while others report no clear dose–response relationship (14). Understanding this relationship is especially important for populations with sedentary lifestyles and prediabetes, who may have reduced exercise tolerance and altered myokine profiles.

In addition to metabolic outcomes, exercise also influences psychological parameters, including perceived stress, emotional state, and cognitive performance. Although acute bouts of exercise often yield mood improvements, the extent to which these changes relate to exercise duration remains unclear (15,16). The Perceived Stress Scale (PSS-10) is widely used to assess psychological stress but is more sensitive to long-term patterns than immediate changes, making it important to analyze whether session duration can meaningfully impact acute psychological responses.

Given these knowledge gaps, the present study was designed to investigate the acute effects of four different exercise durations (15, 30, 45, and 60 minutes) on (1) total cholesterol concentration, (2) circulating Irisin levels, and (3) perceived stress scores among sedentary adults with prediabetes. Using a controlled, laboratory-based crossover design, this study aimed to determine whether increasing duration enhances the acute metabolic and psychological benefits of exercise. The findings are expected to contribute to the development of personalized exercise prescriptions for prediabetic individuals, particularly in settings where sedentary lifestyles and metabolic disorders are highly prevalent.

METHODS

Study Design

This study was conducted as an experimental, laboratory-based crossover trial aimed specifically at examining the acute effects of different exercise durations on biochemical and psychological outcomes among adults with prediabetes. Each participant completed four exercise sessions, each

separated by a seven-day washout period to eliminate any residual physiological effects from prior sessions. The crossover design ensured that every participant served as their own control, thereby strengthening internal validity and reducing inter-individual variability.

Study Setting and Ethical Considerations

All exercise interventions were conducted in the Physiology Skill Laboratory of the Institute of Basic Medical Sciences (IBMS), Khyber Medical University (KMU), Peshawar. Blood collection and laboratory analyses were carried out in the Physiology Research Laboratory using established aseptic procedures. Ethical approval for the study was obtained as part of an already sanctioned postgraduate research project from the Graduate Study Committee, the Advanced Studies and Research Board, and the Institutional Review Board and Ethics Committee of KMU. Prior to participation, written informed consent was obtained from all volunteers after a detailed explanation of the study objectives, procedures, and risks.

Participants

A purposive sampling technique was employed due to the specific metabolic characteristics required for inclusion. Participants were adults between 25 and 35±5 years of age who met the diagnostic criteria for prediabetes, defined as fasting plasma glucose levels between 100 and 125 mg/dL and HbA1c values between 5.7% and 6.4%. Additional criteria included a sedentary lifestyle, non-smoking status, and absence of cardiovascular, thyroid, or musculoskeletal disorders. Individuals who were pregnant or lactating, those with anemia, and those taking lipid-modifying or metabolic medications were excluded. Although the power analysis indicated a minimum sample of 12 participants, 25 individuals were recruited to improve the robustness of the findings; ultimately, 22 participants completed all four sessions of the duration protocol.

Orientation and Baseline Assessments

Before beginning the exercise sessions, participants attended an orientation visit during which written consent was obtained, demographic and medical history forms were completed, and physical activity levels were assessed using the International Physical Activity Questionnaire (IPAQ). Baseline anthropometric measurements including weight, height, body mass index (BMI), waist circumference, and hip circumference were recorded using

standardized equipment. On this day, participants were familiarized with treadmill walking procedures, safety protocols, and monitoring devices. Baseline laboratory investigations, including HbA1c, complete blood count, renal and liver function tests, and cholesterol levels, were also performed to verify eligibility and ensure participant safety.

Duration-Based Exercise Intervention

The duration protocol was designed to determine the optimal exercise duration capable of eliciting favorable acute changes in Irisin levels, cholesterol concentration, and perceived stress. All participants completed four exercise sessions scheduled seven days apart. Each session was conducted 40 minutes after an isocaloric breakfast of approximately 250 kcal, as this timing had been optimized in earlier phases of the study. Exercise was performed on a treadmill at a moderate intensity, predetermined using each participant's predicted maximum heart rate. The duration of each session differed across visits: participants walked for 15 minutes in the first session, 30 minutes in the second, 45 minutes in the third, and 60 minutes or maximum tolerable duration in the fourth. Each session included a standardized 5-minute warm-up and 5-minute cool-down, which were not included in the calculated exercise duration. Heart rate was continuously monitored via a Garmin heart rate belt linked to an analyzer to ensure consistency in exercise intensity.

Blood Sampling and Handling

Venous blood samples were collected immediately before and immediately after each exercise session. A 20G cannula was inserted into the antecubital vein and secured with a three-way connector to facilitate repeated sampling. Blood was collected into gel tubes and allowed to clot for 15–20 minutes at room temperature. Samples were then centrifuged at 1000 rpm for 15 minutes, and the serum obtained was aliquoted into labeled Eppendorf tubes. All serum samples were stored at –80°C in cryoboxes until further biochemical analyses were performed. This standardized collection process was consistent across all four visits to ensure reliability of pre- and post-exercise measurements.

Biochemical Analyses

Serum Irisin concentrations were quantified using a commercially available human Irisin ELISA kit based on the sandwich principle. Samples and standards were added to microplate wells pre-coated with anti-Irisin antibodies, followed by incubation with biotinylated detection antibodies and horseradish peroxidase conjugate. After substrate addition, colorimetric changes were measured at 450 nm using a microplate reader, and concentrations were calculated through standard curves generated in GraphPad Prism. Total serum cholesterol levels were measured using an enzymatic-colorimetric method in which the production of a red dye, following enzymatic oxidation of cholesterol, was quantified spectrophotometrically. Both analyses were conducted according to the manufacturers' protocols.

Assessment of Perceived Stress

Psychological stress was assessed using the Perceived Stress Scale-10 (PSS-10), a validated instrument widely used to measure perceived stress levels over the preceding month. The questionnaire was administered immediately before and after each exercise session to evaluate acute changes in stress response. Four positively stated items were reverse-scored before summing the total score. Although the scale captures long-term perceived stress, repeated administration allowed detection of short-term changes associated with exercise.

Statistical Analysis

Data were analyzed using SPSS version 24. Descriptive statistics were expressed as mean ± standard deviation. Normality of all continuous variables was verified using the Shapiro–Wilk test. Changes in Irisin levels, cholesterol concentration, and PSS scores before and after each exercise duration were examined using paired t-tests. To compare the magnitude of change across the four exercise durations, repeated measures ANOVA was applied. A p-value of less than 0.05 was considered statistically significant for all analyses.

RESULTS

Participant Characteristics

A total of 22 participants completed all four visits of the duration protocol. Baseline demographic and metabolic characteristics are presented in Table 1.

Table 1: *Baseline Demographic and Anthropometric Characteristics*

Variable	Mean ± SD / n (%)
Age (years)	34.5 ± 4.14
• Male	10 (45.5%)
• Female	12 (54.5%)
Height (cm)	170.3 ± 7.13
Weight (kg)	88.76 ± 15.21
Body Mass Index (kg/m^2)	30.66 ± 4.41
Waist Circumference (cm)	102.50 ± 12.94
Hip Circumference (cm)	105.97 ± 13.14
Fasting Plasma Glucose (mg/dL)	112.4 ± 6.8
HbA1c (%)	5.84 ± 0.14
Physical Activity Level	Sedentary (100%)
Smoking Status	0 (0%)
Family History of Diabetes	13 (59.1%)

Effects of Exercise Duration on Cholesterol Levels

Exercise duration was progressively increased across four visits. Cholesterol levels decreased significantly after each exercise session (Table 2). Although the magnitude of reduction varied, all durations of exercise produced significant acute lipid improvements.

Table 2: *Cholesterol and Irisin Levels Before and After Exercise (Duration Protocol: Visits 1–4)*

Visit	Variable	Before Exercise	After Exercise	Mean Difference	p-value
Visit 1 (15 min)	Cholesterol	194.5 ± 30	170 ± 15	−23.2 ± 26.6	0.001
	Irisin	320 ± 192	364 ± 192	+44.5 ± 129	0.120
Visit 2 (30 min)	Cholesterol	186 ± 24	169 ± 15.7	−16.6 ± 17.7	<0.001
	Irisin	287 ± 158	358 ± 195	+71.7 ± 128	0.010
Visit 3 (45 min)	Cholesterol	185 ± 22	173 ± 12.4	−12.4 ± 14.9	0.001

	Irisin	296 ± 148	391 ± 206	+95.2 ± 188	0.020
Visit 4 (60 min)	Cholesterol	185 ± 26	164 ± 37	−21.2 ± 32.3	0.006
	Irisin	294 ± 147	355 ± 175	+61.7 ± 72.6	0.001

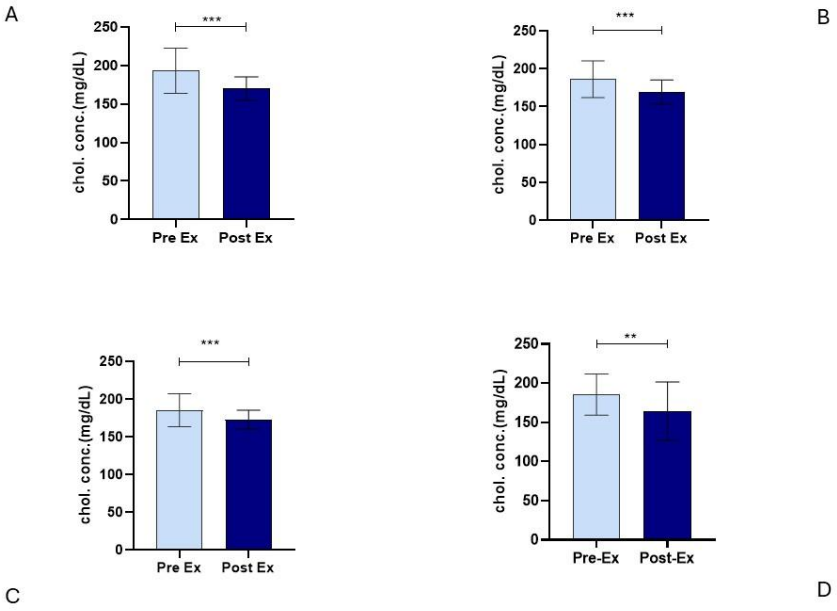


Figure 1A–D illustrates the pre- and post-exercise cholesterol concentrations across Visits 1–4

Panels A–D represent cholesterol levels measured immediately before (light bars) and after (dark bars) exercise of progressively increasing duration: Visit 1 = 15 minutes, Visit 2 = 30 minutes, Visit 3 = 45 minutes, and Visit 4 = 60 minutes or maximum tolerable duration.

A significant reduction in serum cholesterol was observed after each exercise session (Visit 1: p = 0.001; Visit 2: p < 0.001; Visit 3: p = 0.001; Visit 4: p = 0.006). Data are presented as mean ± SD (n = 22).

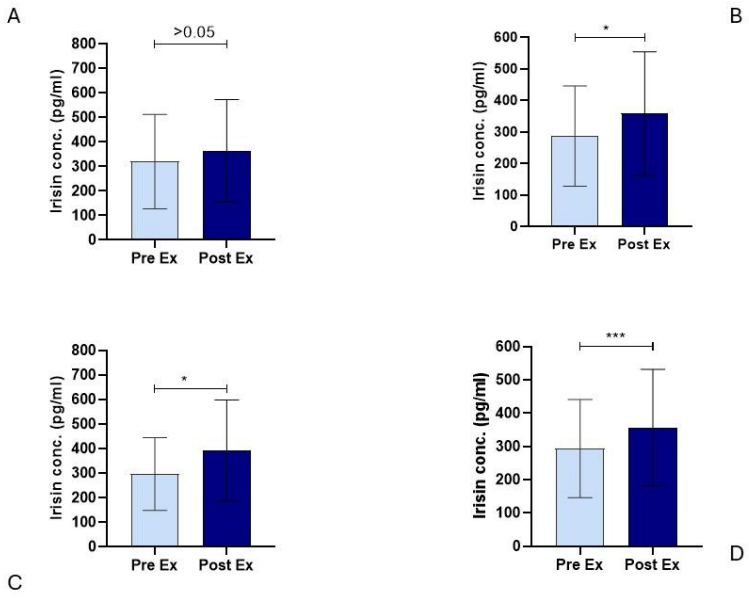
**p < 0.05, **p < 0.005, ***p < 0.001.*

However, when the *change in cholesterol* (delta values) was compared across all four durations using repeated measures ANOVA, the differences were not statistically significant, indicating that while each session reduced cholesterol,

the *incremental increase in duration* did not further enhance the acute cholesterol-lowering response.

Effects of Exercise Duration on Irisin Concentration

Irisin concentrations increased after all exercise sessions, with longer durations generally producing greater changes (Table 2). The non-significant change in Visit 1 supports the observation that exercise duration influences Irisin release, as shorter bouts of activity may not sufficiently stimulate myokine production.



Figures 2A–D depict the rise in Irisin concentrations across all four visits.

Panels A–D show pre-exercise (light bars) and post-exercise (dark bars) Irisin levels following 15-, 30-, 45-, and 60-minute exercise sessions, respectively. Irisin increased after all sessions; however, the rise was not significant after the shortest duration (Visit 1; p = 0.120). Significant increases were seen in Visit 2 (p = 0.010), Visit 3 (p = 0.020), and Visit 4 (p = 0.001). Data are presented as mean ± SD (n = 22).

**p < 0.05, **p < 0.01, ***p < 0.001.*

Despite significant pre–post increases in Visits 2–4, repeated measures ANOVA demonstrated no significant difference in delta Irisin across the four durations, indicating individual variability in Irisin responsiveness.

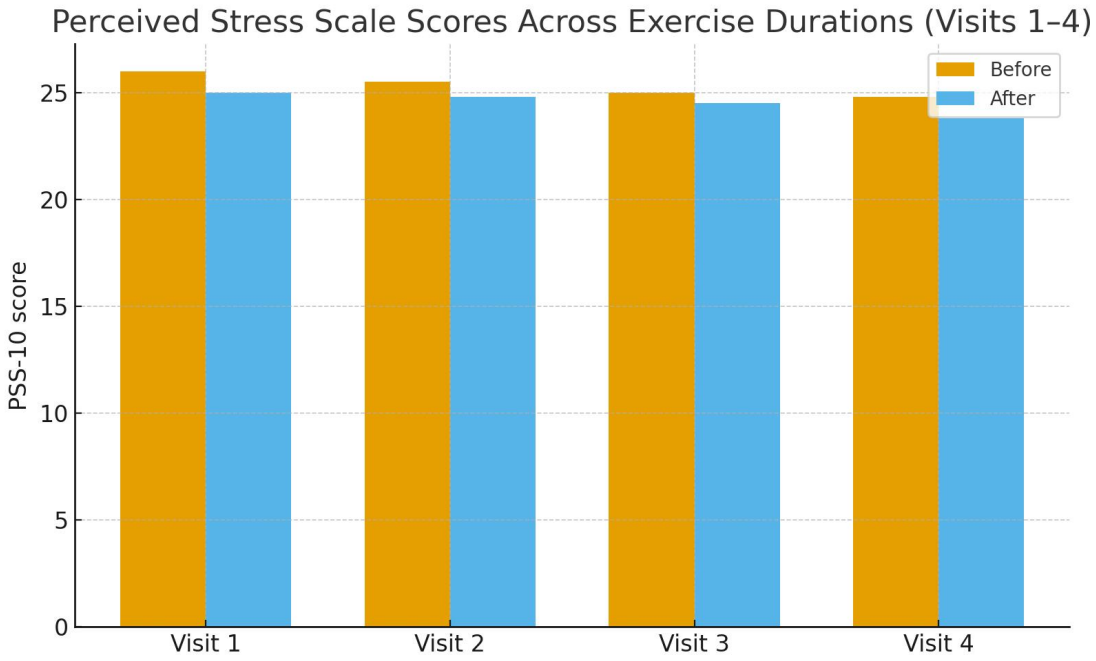


Figure 3. Perceived Stress Scale scores across duration protocol (Visits 1–4).
Mean PSS-10 scores recorded immediately before (light bars) and after (dark bars) each exercise session of increasing duration (15, 30, 45, and 60 minutes). No statistically significant pre–post change was observed at the individual visit level ($p > 0.05$).

DISCUSSION

This study evaluated the acute effects of progressively increasing exercise duration on cholesterol levels, circulating Irisin concentrations, and perceived stress among sedentary adults with prediabetes. The findings demonstrate three key outcomes: (1) cholesterol levels significantly decreased after all exercise durations; (2) Irisin concentrations rose following each exercise session, with greater increases observed during longer durations; and (3) perceived stress improved subjectively and showed a statistically significant reduction from Visit 1 to Visit 4, despite no per-visit significance. These results highlight the role of exercise duration as an important determinant of acute physiological adaptations in individuals at metabolic risk.

The consistent reduction in cholesterol levels across all exercise durations aligns with established evidence that even a single session of aerobic exercise can significantly enhance lipid metabolism by increasing enzymatic activity and promoting greater lipoprotein lipase function (17,18).

Interestingly, although each duration produced significant declines in cholesterol, the magnitude of reduction did not differ significantly across visits based on repeated measures ANOVA. This suggests that even shorter bouts of exercise (15–30 minutes) may be sufficient to initiate favourable cholesterol-lowering effects, supporting previous reports that moderate-duration exercise sessions can meaningfully improve lipid profiles in the short term (19).

However, larger or accumulated reductions over time may require longer or repeated sessions, as shown in chronic training studies (20). The present findings emphasize that for sedentary prediabetic individuals, acute metabolic benefits begin early, even before sustained endurance capacity is developed. Irisin exhibited a duration-dependent pattern, increasing progressively across 30-, 45-, and 60-minute sessions, while the 15-minute bout showed a non-significant rise. This supports the hypothesis that Irisin secretion is influenced by total exercise duration and muscular workload, consistent with prior studies reporting that longer sessions elicit greater myokine release due to increased thermogenic and metabolic demand (21–23).

Moreover, Irisin has been implicated in browning of white adipose tissue, enhanced mitochondrial function, and improved glucose utilization processes highly relevant for individuals with prediabetes (24,25). The significant increases observed in the present study reinforce the potential role of Irisin as a biomarker of beneficial physiological adaptation to exercise. Nonetheless, despite significant pre–post increases in Visits 2–4, the lack of statistically significant differences in delta Irisin across durations suggests variability in individual hormonal responsiveness, consistent with findings from other human exercise studies (26,27). Factors such as baseline fitness, adiposity, sex, and genetic variability may influence myokine release and warrant further investigation.

Although PSS scores did not change significantly within individual visits, the significant improvement between Visit 1 and Visit 4 reflects a cumulative psychological benefit of repeated exercise exposure. This aligns with literature showing that while acute mood improvements are common, measurable reductions in perceived stress often require repeated sessions (28,29). The PSS-10 scale captures stress appraisal over a one-month period; therefore,

lack of per-visit significance is expected. However, subjective reports of reduced tension and improved mood after each session are consistent with known immediate psychological effects of physical activity, mediated through endorphin release, increased cerebral blood flow, and reduced sympathetic activity (30,31).

A major strength of this study is its crossover design, which minimized inter-individual variability and allowed each participant to serve as their own control. The use of controlled laboratory conditions, uniform timing of exercise after an isocaloric meal, and standardized biomarker processing enhanced internal validity.

Limitations include the modest sample size, which may reduce statistical power to detect subtle duration-dependent differences, and the reliance on self-reported measures for psychological outcomes. Further, as this study focused on acute changes, long-term training effects remain unexplored.

CONCLUSION

The findings demonstrate that even short-duration exercise induces significant metabolic improvements in adults with prediabetes, while longer durations enhance Irisin release and contribute to cumulative psychological benefits. These results underscore the importance of tailoring exercise prescriptions to individual tolerance while recognizing that meaningful physiological benefits can occur across a range of durations. Future studies should investigate long-term adaptations and explore how duration interacts with intensity to optimize metabolic outcomes.

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