



# Evaluating the impact of coenzyme Q10 and high-intensity interval training on lactate threshold and Plasma blood gases in rats: a randomized controlled trial

Yavuz Yasul<sup>1</sup> · Büşra Yılmaz<sup>2</sup> · Ömer Şenel<sup>2</sup> · Dursun Kurt<sup>1</sup> · Taner Akbulut<sup>3</sup> · Ayşen Çalığıuşu<sup>4</sup> · Elvan Anadol<sup>5</sup> · Canan Yılmaz<sup>6</sup>

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## Abstract

**Purpose** Coenzyme Q<sub>10</sub> (Q<sub>10</sub>) is a mitochondrial coenzyme that facilitates ATP production via oxidative phosphorylation. This study hypothesized that Q<sub>10</sub> enhances mitochondrial efficiency and lactate threshold, while high-intensity interval training (HIIT) promotes metabolic adaptations, improving glucose utilization and buffering capacity for faster recovery after high-intensity exercise.

**Methods** A randomized controlled trial was conducted using 24 male Sprague–Dawley rats (250.4 ± 6.1 g, 8 weeks old). The rats were allocated into four groups: control (C), coenzyme Q<sub>10</sub> (CoQ<sub>10</sub>), HIIT, and HIIT + Q<sub>10</sub>. The Q<sub>10</sub> administration involved a dosage of 10 mg/kg/day, given 30 min prior to the HIIT protocol. Lactate threshold, blood gas parameters, oximetry values, metabolite levels, and electrolyte status were analyzed utilizing the Radiometer 900 device. The blood samples were collected at the fifth and tenth minutes following the HIIT training trials.

**Results** The HIIT + Q<sub>10</sub> group exhibited a significant reduction in lactate threshold ( $p < 0.01$ ), maintaining values below average. Significant improvements in blood gas parameters, including pH, pO<sub>2</sub>, and pCO<sub>2</sub>, were observed in this group. Enhanced oxygen transport capacity was indicated by improved oximetry parameters (Hb, HCT, sO<sub>2</sub>) and reduced COHb levels. Additionally, positive changes in HCO<sub>3</sub><sup>-</sup> and base values indicated reduced metabolic stress. Q<sub>10</sub> supplementation also stabilized electrolytes, particularly K<sup>+</sup> and Na<sup>+</sup>.

**Conclusion** The Q<sub>10</sub> supplementation supported metabolic balance, improved oxygen transport, and stabilized acid–base levels during HIIT. It reduced lactate accumulation, enhanced glucose availability, and alleviated metabolic stress, thereby improving recovery efficiency and physiological adaptation.

**Keywords** Blood gases · Coenzyme Q<sub>10</sub> · HIIT · Lactate threshold · Oxygenation · Recovery

## Abbreviations

|                       |                                        |
|-----------------------|----------------------------------------|
| Q <sub>10</sub>       | Coenzim Q <sub>10</sub>                |
| MICT                  | Moderate-intensity continuous training |
| HICT                  | High-intensity continuous training     |
| HIIT                  | High-intensity interval training       |
| pCO <sub>2</sub>      | Partial pressure of carbon dioxide     |
| pO <sub>2</sub>       | Partial pressure of oxygen             |
| Hb                    | Hemoglobin                             |
| HCT                   | Hematocrit                             |
| sO <sub>2</sub>       | Oxygen saturation                      |
| COHb                  | Carboxyhemoglobin                      |
| HHb                   | Deoxyhemoglobin                        |
| MetHb                 | Methemoglobin                          |
| Lac                   | Lactate threshold                      |
| Glu                   | Glucose                                |
| O <sub>2</sub> (vol%) | Oxygen concentration                   |

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✉ Yavuz Yasul  
yavuz.yasul@omu.edu.tr

<sup>1</sup> Bafra Vocational School, Ondokuz Mayıs University, 55400 Bafra, Samsun, Türkiye

<sup>2</sup> Faculty of Sport Sciences, Gazi University, Ankara, Türkiye

<sup>3</sup> Faculty of Sport Sciences, Fırat University, Elazığ, Türkiye

<sup>4</sup> Department of Anatomy, Ege University, İzmir, Türkiye

<sup>5</sup> Laboratory Animals Breeding and Experimental Researches Center, Gazi University, Ankara, Türkiye

<sup>6</sup> Faculty of Medicine, Department of Medical Biochemistry, Gazi University, Ankara, Türkiye

|                                    |                                    |
|------------------------------------|------------------------------------|
| Base(Ecf)                          | Base status in extracellular fluid |
| Base(B)                            | Base Status in blood               |
| HCO <sub>3</sub> <sup>-</sup> (st) | Standard bicarbonate               |
| HCO <sub>3</sub> <sup>-</sup>      | Bicarbonate                        |
| K <sup>+</sup>                     | Potassium                          |
| Na <sup>+</sup>                    | Sodium                             |
| Ca <sup>+</sup>                    | Calcium                            |

## Introduction

The greatest challenge for coaches and athletes is to identify the optimal combination of exercises during training sessions. Considering its positive effects, high-intensity interval training (HIIT) has recently emerged as one of the popular alternatives. Interval training is defined as the repetition of relatively intense efforts interspersed with periods of lighter activity or rest. This method is generally applied in the context of athletic performance and forms the foundation of training programs for elite athletes (Fox et al. 1973; Altinel et al. 2024).

The fundamental principle of HIIT is to accumulate a greater workload at a higher intensity than what can be achieved through continuous exercise at a steady intensity. The workload capacity developed through HIIT plays a critical role in achieving success in sports and activities such as middle and long-distance running, cycling, swimming, rowing, and cross-country skiing (Hellard et al. 2019; Leo et al. 2020; Torvik et al. 2021; Casado et al. 2022). This perspective is based on the assumption that HIIT strengthens physiological responses, enhances the ability to sustain higher workloads, and increases resistance to fatigue during competition (Laursen and Jenkins 2002). In this context, HIIT serves as a powerful stimulus that triggers physiological remodeling, enhances exercise performance, and improves oxidative capacity (Gibala and Jones 2013). In contrast, HIIT induces both mechanical and metabolic stress during the session (Leite et al. 2023). Unlike aerobic exercise, HIIT places a significantly greater demand on both aerobic and anaerobic energy systems. The repeated high-intensity efforts require rapid ATP resynthesis, leading to increased reliance on anaerobic glycolysis, and heightened metabolic acidosis due to accelerated lactate accumulation. Unlike continuous moderate-intensity exercise, where energy production remains stable, HIIT induces repeated metabolic fluctuations that challenge energy homeostasis. These extreme demands may necessitate additional physiological support to optimize recovery, buffer acidosis, and sustain performance across repeated high-intensity bouts (MacInnis and Gibala 2017). The new metabolic load resulting from HIIT is monitored through indicators such as blood lactate threshold (Lac) levels and blood gas exchange parameters, which help determine the limits of exertion. Furthermore, the

accumulation of Lac above its baseline level and the limitations in oxygen utilization define the boundaries of power and speed performance (Smith and Jones 2001). Therefore, plasma Lac levels and blood gas exchange markers reflect the highest sustainable intensity, where a significant increase in non-oxidative metabolism associated with exercise intolerance is observed (Poole et al. 2016).

Athletes frequently use dietary supplements to achieve high levels of athletic performance and to enhance sustainability by reducing metabolic intolerance (Mason et al. 2020). Despite potential side effects, sodium bicarbonate, sodium citrate, and beta-alanine-based supplements are commonly preferred by athletes (Gencoglu et al. 2022). Sodium bicarbonate functions as an extracellular buffer, neutralizing hydrogen ions to delay fatigue, while beta-alanine enhances intracellular buffering by increasing muscle carnosine levels. However, both agents have limitations, including gastrointestinal discomfort and dosage sensitivity. Coenzyme Q<sub>10</sub> (Q<sub>10</sub>), along with its various effects, functions as a coenzyme for mitochondrial enzymes in the electron transport system, playing a crucial role in energy production within the mitochondrial respiratory chain and facilitating ATP synthesis via oxidative phosphorylation. Additionally, Q<sub>10</sub> contributes to lactate metabolism by enhancing the efficiency of the pyruvate dehydrogenase complex, promoting aerobic metabolism over anaerobic glycolysis. Furthermore, its antioxidant properties help mitigate oxidative stress, which can otherwise impair mitochondrial function during high-intensity exercise (Goncalves et al. 2015; Mason et al. 2020). Unlike traditional buffering agents, Q<sub>10</sub> not only aids in pH regulation but also supports mitochondrial efficiency and energy availability, making it a multifunctional alternative with added metabolic benefits. This evidence strengthens the notion that Q<sub>10</sub> could serve as a novel buffering alternative due to its bioenergetic role in muscle contraction, muscle-protective effects, and involvement in inflammatory processes (Drobnic et al. 2022). The studies focusing on this topic have reported that Q<sub>10</sub> supplementation improved energy production and athletic performance while reducing lactate accumulation levels in male middle-distance runners who received 5 mg/kg for 14 days prior to competition (Armanfar et al. 2015), cyclists who were supplemented with 20 mg/kg for 12 days (Broome et al. 2021), and runners who consumed 60 mg/kg for 10 days (Ovchinnikov et al. 2022).

Research has generally focused on pre-exercise conditions, moderate-intensity exercise distributions, and short-term persistent responses. However, the effects of Q<sub>10</sub> supplementation on lactate accumulation during high-intensity exercise conditions and HIIT have yet to be fully elucidated (Yasul et al. 2024). Additionally, the effects of Q<sub>10</sub> supplementation on lactic acid accumulation and other plasma blood gases during HIIT have not been sufficiently clarified. Based on these considerations, we hypothesize that CoQ<sub>10</sub>

supplementation will facilitate a faster metabolic recovery post HIIT by lowering lactate levels, optimizing blood gas parameters and enhancing mitochondrial efficiency. This study aims to evaluate these effects by analyzing post-exercise recovery dynamics in rats undergoing HIIT with and without CoQ<sub>10</sub> supplementation.

## Methods

### Experimental design

The study was conducted using a randomized experimental controlled design with 24 male Sprague Dawley rats, aged 8 weeks and weighing  $250.4 \pm 6.1$  gr. The research was carried out at the Gazi University Laboratory Animal Breeding and Experimental Research Center in compliance with the National Institutes of Health (NIH) guidelines for the care and use of laboratory animals. The research protocol was approved by the Gazi University Animal Experiments Ethics Committee on April 5, 2024 (Decision no: E-66332047-604.01-923246) and was conducted in accordance with the principles of the Helsinki Declaration, ensuring animal rights and welfare were upheld. According to G-power analysis, with a Type I error (alpha) of 0.05, a power (1-beta) of 0.8, an effect size of 1.88, and a two-tailed alternative hypothesis (H1), the minimum sample size required to detect a significant difference using this test was determined to be at least six rats per group. Consequently, the rats were randomly divided into four groups: control group (C), coenzyme Q<sub>10</sub> group (CoQ<sub>10</sub>), HIIT group, and HIIT + Q<sub>10</sub> group. The rats were housed in standard cages (two rats per cage) under controlled environmental conditions of 22–25 °C, with a 12-h light/12-h dark cycle, and were fed ad libitum. The C group was fed ad libitum without supplementation or exercise. The exercise groups underwent a four-stage preparatory phase before receiving supplementation and HIIT trials. The rats completed four stages of training: treadmill acclimatization in the first stage (KN-73; Natsume Seisakusho Co., Ltd., Tokyo, Japan), moderate-intensity continuous training (MICT) acclimatization in the second stage, high-intensity continuous training (HICT) acclimatization in the third stage (Soya et al. 2007; Yasul et al. 2025), and HIIT acclimatization in the fourth stage (Okamoto et al. 2021). The structured progression aligns with previous findings that emphasize its role in enhancing physiological resilience and optimizing adaptation to high-intensity exercise. During the I week of HIIT trials, only exercise was performed for the HIIT group and the HIIT + Q<sub>10</sub> group. In the II week, the HIIT group continued with exercise only, while the HIIT + Q<sub>10</sub> and CoQ<sub>10</sub> groups received a 10 mg/kg/day Q<sub>10</sub> supplement 30 min before exercise via gavage, a timing selected based on its pharmacokinetics, ensuring peak

plasma levels coincide with the exercise period to enhance mitochondrial function and metabolic efficiency. This protocol was repeated in the III and IV weeks. The HIIT protocol was designed based on the principle of progressive overload, with the exercise session scheduled at 10:30 AM (Table 1). Additionally, during the HIIT trials weeks, blood samples were collected using capillary tubes at the fifth (first observation time) and tenth minutes (second observation time) after exercise (Fig. 1), as these time points capture early-phase metabolic recovery, reflecting rapid lactate threshold and blood gas stabilization, which are critical for evaluating post-exercise physiological responses. The blood gas values (pH, pCO<sub>2</sub>, pO<sub>2</sub>), oximetry values (Hb, HTC, sO<sub>2</sub>, COHb, HHb, MetHb), metabolite values (Lac, Glu), temperature corrected values (pH (T), pCO<sub>2</sub> (T), pO<sub>2</sub> (T)), oxygen status (p50, O<sub>2</sub>), acid base status (Base (Ecf), Base (B), HCO<sub>2</sub><sup>-</sup> (st), HCO<sub>2</sub><sup>-</sup>), electrolyte values (K<sup>+</sup>, Na<sup>+</sup>, Ca<sup>+</sup>) markers were evaluated and blood gas analyses were performed using the Radiometer 900.

### Collection ve analysis for blood gases

First, the rats were placed in a special movement-restraining device, and their tails were immersed in 40 °C warm water for 5–10 s (Parasuraman et al. 2010) to facilitate vasodilation and minimize potential discomfort during sampling. After vasodilation and the visualization of the veins, approximately 100 µl of blood was collected by inserting a 20–22G needle attached to a syringe into the ventral tail artery at an angle of 20–30 degrees, ensuring minimal distress. The collected blood samples were transferred into capillary tubes (Safeclinitubes; 942-896-D957P-70-100) and analyzed for blood gases using a Radiometer 900 autoanalyzer. To prevent continued bleeding from the tail artery, pressure was applied for a short duration, and the rats were then returned to their cages (Namlı et al. 2023). Blood sampling was performed in a controlled, quiet environment to further limit external stressors and ensure the reliability of physiological measurements.

### Statistical analysis

The study was conducted using a factorial experimental design in randomized blocks with six replications, allowing for the assessment of both main effects and interaction effects. Factorial ANOVA was performed to analyze the effects of group (C, CoQ<sub>10</sub>, HIIT, HIIT + Q<sub>10</sub>), time (5 min, 10 min), and week (I–IV) at both individual and interaction levels. The data were subjected to variance analysis (ANOVA) using JMP 13.0 software, and significant differences between means were identified using the LSD (Least Significant Difference) multiple comparison test, which was applied only when significant main or interaction effects

**Table 1** presents a structured high-intensity interval training (HIIT) protocol that includes a pre-training phase, progressive training phases, and a comparison of conditions with and without Coenzyme Q10 (Q10) supplementation

High-intensity interval training (HIIT) and HIIT + Q<sub>10</sub> supplementation training protocols

Pre-HIIT phase

| Phases                                                | Training modality | Days  | Reps | Sets | Running speed | Exercise duration | Rest between reps. (sec/min) | Rest between sets (min/hour) |
|-------------------------------------------------------|-------------------|-------|------|------|---------------|-------------------|------------------------------|------------------------------|
| Phase 1                                               | Treadmill Drills  | 0–5   | 1    | –    | 6–10 (m/min)  | 8–12 min/hour     | –                            | –                            |
|                                                       | MICT Drills       | 6–10  | 1    | –    | 11–15 (m/min) | 14–20 min/hour    | –                            | –                            |
| Phase 2                                               | HICT Drills       | 11–15 | 1    | –    | 18–25 (m/min) | 14–20 min/hour    | –                            | –                            |
| Phase 3                                               | HIIT Drills       | 16–20 | 2    | 2    | 25–35 (m/min) | 20 s/min          | 80–100                       | 3–4                          |
| HIIT training without Q <sub>10</sub> supplementation |                   |       |      |      |               |                   |                              |                              |
| Week I                                                | HIIT              | 21–26 | 2    | 3    | 30–40 (m/min) | 30 s/min          | 120–150                      | 3–4                          |
| HIIT Training with Q <sub>10</sub> Supplementation    |                   |       |      |      |               |                   |                              |                              |
| Week II                                               | HIIT              | 27–32 | 3    | 3    | 30–40 (m/min) | 30 s/min          | 120–150                      | 3–4                          |
| Week III                                              | HIIT              | 33–38 | 4    | 4    | 30–40 (m/min) | 30 s/min          | 120–150                      | 3–4                          |
| Week IV                                               | HIIT              | 39–44 | 5    | 4    | 30–40 (m/min) | 30 s/min          | 120–150                      | 3–4                          |

Training intensity progressively increases over four distinct phases before transitioning into the main HIIT intervention. Rest intervals are systematically adjusted to optimize recovery and adaptation. The supplementation phase begins from Week II, allowing direct comparisons between supplemented and non-supplemented conditions

presents the characteristics of exercise intensity for the two groups undergoing high-intensity interval training

*HIIT* high-intensity interval training group, *HIIT* + Q<sub>10</sub> High-intensity interval training + coenzyme Q<sub>10</sub> supplementation group, *MICT* moderate-intensity continuous training, *HICT* High-intensity continuous training

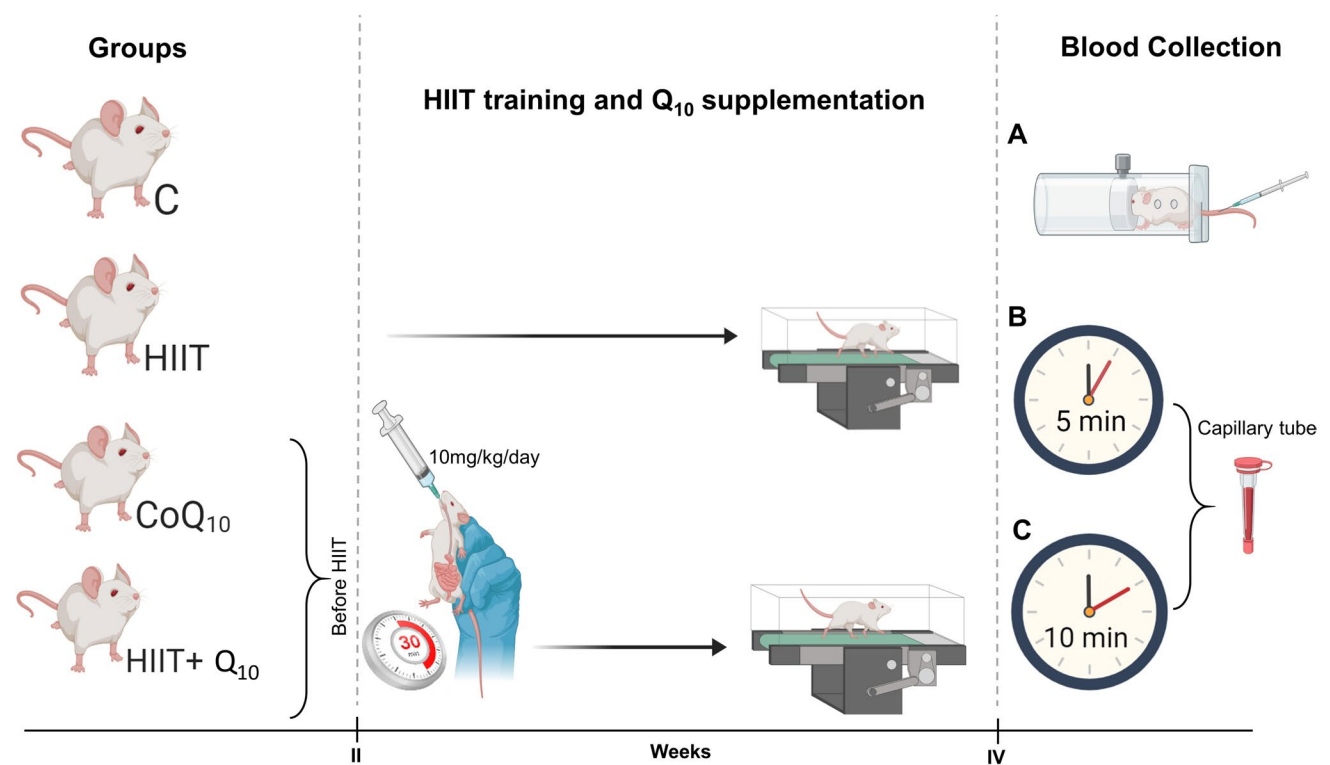


**Fig. 1** Presents a schematic representation of the experimental protocol, illustrating the timing of Q10 supplementation, HIIT interventions, and blood collection at weeks I, II, III, and IV. *C* Control group, *HIIT* High-Intensity Interval Training, *Q<sub>10</sub>* Coenzyme Q<sub>10</sub>

supplement, *A* Blood collection from the tail of rats using a special device, *B* Observation time; 5 min, blood sample collected in a capillary tube, *C* Observation time; 10 min, blood sample collected in a capillary tube

were detected. This approach ensured a comprehensive evaluation of the combined influence of  $Q_{10}$  supplementation and HIIT on physiological parameters. Relationships between parameters were analyzed using Pearson's correlation analysis, while multivariate relationships and scaling between treatments and physiological parameters were explored using PCA-Biplot representation, facilitated by XLSTAT (Kurt 2023). The first two principal components accounted for 67.61% of the total variance, with eigenvalues greater than 1.0, justifying their retention for analysis. This allowed for a clearer interpretation of key physiological parameters that differentiated between experimental groups.

## Results

### Blood gas values

When examining the effects of  $Q_{10}$  supplementation under HIIT conditions on pH,  $pCO_2$ , and  $pO_2$  levels, varying degrees of significance were identified in terms of interventions and their interactions. All three parameters were significantly influenced by groups (G), weeks (W), and their two-way interaction ( $G \times W$ ), as well as by the  $G \times M$  interaction for  $pO_2$ , with at least a significance level of  $p < 0.05$ . While the time had no significant effect on the changes, the three-way interaction ( $G \times W \times M$ ) significantly affected the pH value at the  $p < 0.05$  level (Table SM-1). The mean pH value, which was 7.36 mmHg, reached its highest level of 7.418 mmHg in the HIIT +  $Q_{10}$  group during the second observation time of the IV week. However, when only groups were considered, the C group and the HIIT +  $Q_{10}$  group were statistically classified in the same group, with the lowest pH value observed exclusively in the HIIT group at 7.34 mmHg. The average  $pCO_2$  level, determined to be 46.05 mmHg, reached its highest value (51.48 mmHg) in the III week of the group Co $Q_{10}$ . Similarly, for  $pO_2$ , the highest value (59.96 mmHg) was observed in the IV week of the group Co $Q_{10}$ . When interactions were disregarded the highest  $pO_2$  levels were predominantly identified in the HIIT +  $Q_{10}$  group (Table SM-6).

### Oximetry values

When examining the effect of  $Q_{10}$  supplementation under HIIT conditions on Hb, HCT,  $sO_2$ , COHb, HHb, and MetHb values, significant differences were identified at various levels regarding the interventions and their interactions. For all parameters, the groups, weeks, and their two-way interaction ( $G \times W$ ) showed at least a significance level of  $p < 0.05$ . However, weeks were non-significant for  $sO_2$ , and groups were non-significant for HHb. Except for MetHb, time (M) had no significant effect on the changes, but the three-way

interaction ( $G \times W \times M$ ) significantly influenced HCT levels at  $p < 0.05$  (Tables SM-1, 2) (Tables SM-1, 2). For Hb, with an overall mean value of 14.68 g/dl, the highest level, 16.36 g/dl, was reached in the III week in the HIIT +  $Q_{10}$  group. When only groups were considered, the HIIT +  $Q_{10}$  group had the highest Hb level at 15.22 g/dl. Similarly, for HCT, which had an overall mean of 44.54, the HIIT +  $Q_{10}$  group recorded the highest value (47.19), while the C group had the lowest value (41.92). In the two-way interaction ( $G \times W$ ), the HIIT +  $Q_{10}$  group exhibited the highest HCT value (50.13), while the C group showed the lowest. In the three-way interaction ( $G \times W \times M$ ), particularly during the second observation time in the II week of supplementation, the HIIT +  $Q_{10}$  group demonstrated significant increases. The highest Hb value, 15.22 g/dl, was identified in the HIIT +  $Q_{10}$  group. For  $sO_2$ , the Co $Q_{10}$  group, HIIT group, and HIIT +  $Q_{10}$  group belonged to the same statistical category, while the lowest value, %50.41, was observed in the C group. Regarding COHb, which had an overall mean of %1.45, the lowest value, %1.07, was statistically distinct and observed in the HIIT +  $Q_{10}$  group. In the  $G \times W$  interaction, COHb reached its highest level during the I week in the HIIT group (3.02) and the II week in the Co $Q_{10}$  group (2.81). The lowest COHb value, %0.53, was recorded in the III week in the HIIT +  $Q_{10}$  group. For HHb, which had an overall mean of %43.65, the highest values were observed in the II (%46.12) and III (%47.42) weeks, while the lowest value occurred in the IV week (%38.68). MetHb, with an overall mean of %0.42, reached its highest value in the IV week in the Co $Q_{10}$  group (%1.17). Among the groups, the HIIT group recorded the lowest MetHb value at %0.16 (Table SM-7).

### Metabolite values

The effect of  $Q_{10}$  supplementation under HIIT conditions on Lac was significant at the  $p < 0.01$  level for all factor and their two-way and three-way interactions. For Glu, significance was observed in G, W,  $G \times W$ , and  $G \times W \times M$  interactions (Tables SM-2, 3). The average Lac value, calculated as 3.72 mmol/L, reached its highest level of 7.76 mmol/L in the HIIT group during the first observation time in the III week. However, the HIIT +  $Q_{10}$  group had the lowest Lac level at the same observation time, with a value of 2.42 mmol/L. At the second observation time in the IV week, the HIIT group exhibited the highest Lac value at 4.68 mmol/L, while the HIIT +  $Q_{10}$  group recorded the lowest Lac value during the second observation time in the II week. Furthermore, when only the groups were compared, the HIIT +  $Q_{10}$  group and the Co $Q_{10}$  group were found to belong to the same statistical category, while the highest Lac value, 5.33 mmol/L, was exclusively observed in the HIIT group. Regarding weeks, the Lac level, which was at its peak during the I week,

declined in subsequent weeks following supplementation intervention (Fig. 2). As for Glu, the overall study average was 186.24 mg/dL, with the CoQ<sub>10</sub> group reaching the highest value (259.68 mg/dL) in the IV week. Among the groups, the highest Glu value (205.71 mg/dL) was identified in the HIIT + Q<sub>10</sub> group (Table SM-8).

### Temperature corrected values

When examining the effect of Q<sub>10</sub> supplementation under HIIT conditions on pH (T), pCO<sub>2</sub> (T), and pO<sub>2</sub> (T) values, varying levels of significance were identified in terms of the interventions and their interactions. Among these, the parameters pH (T), pCO<sub>2</sub> (T), and pO<sub>2</sub> (T) showed significant differences between groups and weeks, as well as in their interaction (G × W) at the  $p < 0.01$  level, while pCO<sub>2</sub> (T) was influenced by weeks at the  $p < 0.05$  level (Table SM-3). The mean pH (T) value, recorded at 7.36, reached its highest level of 7.40 in the IV week in the HIIT + Q<sub>10</sub> group. When only groups were considered, the CoQ<sub>10</sub> group, HIIT group, and HIIT + Q<sub>10</sub> group were found to belong to the same statistical group, with the highest pH (T) value observed exclusively in the C group at 7.38 mmHg. In terms of weeks, the lowest pH (T) value was observed in the I

week, while the highest was recorded in the IV week. The mean pCO<sub>2</sub> (T) value, calculated at 45.95, was found to be highest in the CoQ<sub>10</sub> group (47.64) when only groups were analyzed. In terms of weeks, the highest pCO<sub>2</sub> (T) value was observed in the III week. Regarding the interaction (G × W), the HIIT group (48.82) and the HIIT + Q<sub>10</sub> group (46.67) were above the overall mean in the I week, while the HIIT + Q<sub>10</sub> group was below the mean in the II and III weeks. The mean pO<sub>2</sub> (T) value, recorded at 46.94, showed the highest value in the HIIT group (51.15) when only groups were considered. In terms of weeks, the highest pO<sub>2</sub> (T) value was observed in the IV week (Table SM-9).

### Oxygen status

The effects of Q<sub>10</sub> supplementation under HIIT conditions on p50 and O<sub>2</sub> (vol%) values were analyzed, revealing significant differences based on G, W, M, and their interactions. The p50 value was significantly influenced by G, W, T, and pairwise interactions (G × W and G × M) at  $p < 0.01$ , and by the three-way interaction (G × W × M) at  $p < 0.05$  (Table SM-3,4). The mean p50 value was 37.35 mmHg, with the highest value (41.71 mmHg) recorded in the HIIT + Q<sub>10</sub> group during the first observation time of the I week.

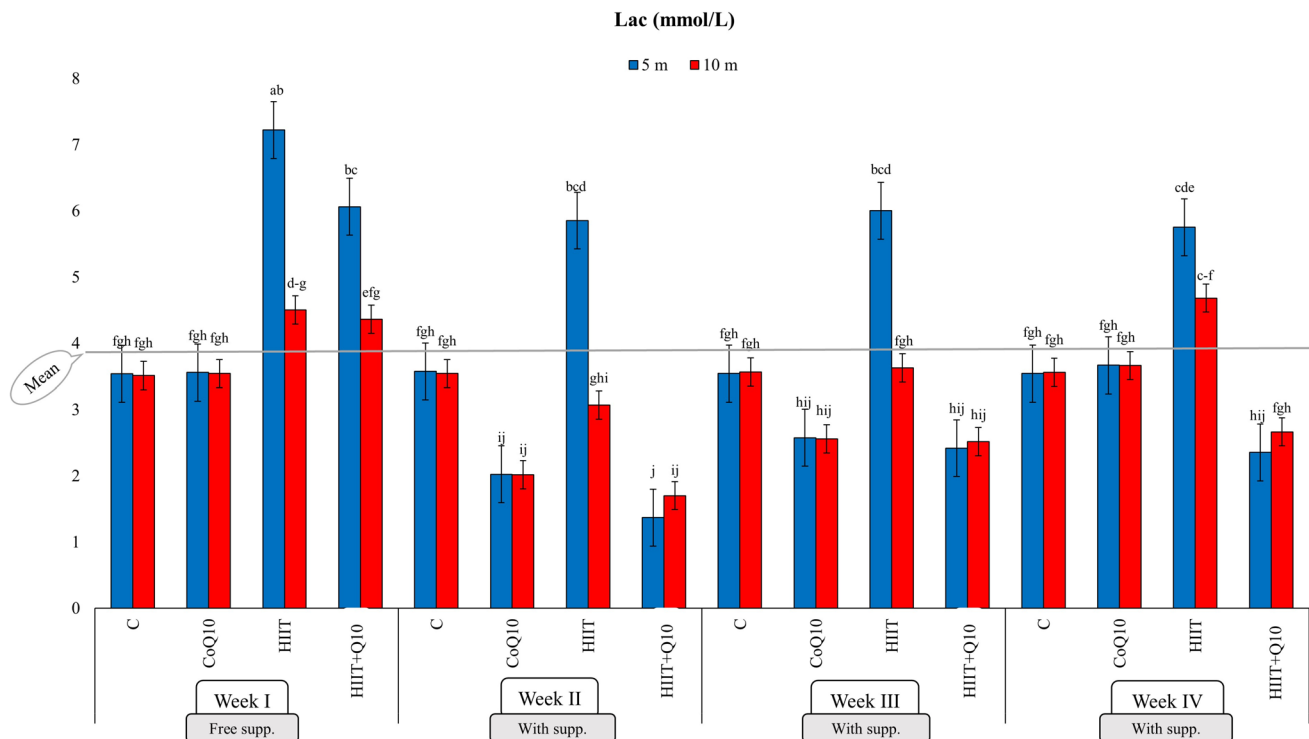


**Fig. 2** Four-week follow-up of the lactate (Lac) mmol/L value at two different observation times. Lac Lactate concentration in blood, C Control group, CoQ<sub>10</sub> Coenzyme Q<sub>10</sub> supplement group, HIIT High-Intensity Interval Training group, HIIT + Q<sub>10</sub> High-Intensity Interval Training + Coenzyme Q<sub>10</sub> supplement group, 5 m: 5th minute (first

observation time), 10 m: 10th minute (second observation time), the letters above the columns (a, b, c, d, e, f, g, h, j) indicate statistical differences between groups analyzed by the One-Way ANOVA test followed by the LSD multiple comparison test

Among the groups, the lowest p50 value (35.27 mmHg) was observed in the C group. The  $O_2$  (vol%) value was significantly affected by G and W at  $p < 0.05$  and by pairwise interactions ( $G \times W$ ) at  $p < 0.01$ . The mean  $O_2$  (vol%) value was 12.40, with the lowest value (11.11) observed in the C group and the highest value (13.65) recorded in the IV week. The CoQ<sub>10</sub> group and HIIT + Q<sub>10</sub> group reached the highest  $O_2$  (vol%) values during the IV week, placing them in the same statistical group (Table SM-10).

### Acid–base status values

The effects of Q<sub>10</sub> supplementation on metabolic parameters under HIIT conditions were evaluated. When examining Base (Ecf), Base (B),  $HCO_3^-$  (st), and  $HCO_3^-$  values, statistically significant differences were identified in terms of interventions and interactions at least at the  $p < 0.05$  and  $p < 0.01$  levels (Table SM-4). The overall mean of  $HCO_3^-$  (st) was 23.38 mmol/L, showing an increase from the I week and reaching its peak level. In the HIIT + Q<sub>10</sub> group, the lowest value was recorded in the I week, followed by an increase in subsequent weeks. The mean Base (Ecf) value was 0.67 mmol/L, with the negative value in the I week transitioning to a positive increase. The lowest positive value was observed in the HIIT + Q<sub>10</sub> group, while the highest was recorded in the CoQ<sub>10</sub> group. The mean Base (B) value was 0.13 mmol/L, where the initially negative value in the I week turned positive in subsequent weeks. The mean  $HCO_3^-$  value was calculated as 25.51 mmol/L, with the maximum level reached in the IV week in the HIIT + Q<sub>10</sub> group (Table SM-11).

### Electrolyte values

When examining the effects of Q<sub>10</sub> supplementation under HIIT conditions on  $K^+$ ,  $Na^+$ , and  $Ca^{+}$  levels, significant differences were identified at various levels in terms of treatments and their interactions. The  $K^+$  and  $Ca^{+}$  levels were influenced by G, W, and their two-way interaction ( $G \times W$ ), with  $Ca^{+}$  also being affected by the interaction ( $G \times M$ ), and  $K^+$  by the interaction ( $W \times M$ ), all at a significance level of at least  $p < 0.05$ . The  $Na^+$  level was significantly influenced by G and W ( $p < 0.05$ ) and by two-way interactions ( $G \times W$  and  $W \times M$ ) at a significance level of at least  $p < 0.01$  (Table SM-5). The mean  $K^+$  level, recorded as 3.96 mmol/L, showed that the HIIT group and the HIIT + Q<sub>10</sub> group were within the same statistical category, with the highest  $K^+$  level observed in the C group at 4.64 mmol/L. The  $Na^+$  level, with an overall mean of 142.46 mmol/L, was lowest in the HIIT group at 141.47 mmol/L. For  $Ca^{+}$ , the highest level (1.42 mmol/L) was observed in the C group, while the lowest level (1.32 mmol/L) was found in the HIIT + Q<sub>10</sub> group. At the second observation time, the HIIT group and

HIIT + Q<sub>10</sub> group were within the same statistical category, although the  $Ca^{+}$  level in the HIIT + Q<sub>10</sub> group was lowest (1.33 mmol/L) at the first observation time (Table SM-12).

### Correlation

The results of the Pearson correlation analysis demonstrated statistically significant but low-level positive and negative correlations among biochemical parameters. Specifically, weak positive or negative correlations were identified among pH,  $Na^+$ ,  $HCO_3^-$ , Lac, COHb, Hb, and pO<sub>2</sub>, while pCO<sub>2</sub> was found to be associated with Glu and HTC. Hb exhibited correlations with  $HCO_3^-$  and HHb, whereas HTC showed low-level positive relationships with parameters such as  $Na^+$ ,  $HCO_3^-$ , and MetHb. Positive correlations were detected between Lac and  $Ca^{+}$ , while negative correlations were observed between Lac and  $HCO_3^-$  or pH. Additionally, Glu exhibited a negative correlation with  $Ca^{+}$  and a positive correlation with p50. pO<sub>3</sub> was positively associated with  $Na^+$ , Base(B), and Base(Ecf), while significant relationships were identified between pCO<sub>2</sub>(T) and  $O_2$ (Vol%). These findings are illustrated in Fig. 3.

Principal component analysis (PCA) was conducted to determine the relative importance of the parameters and their contributions to total variance. The first two components accounted for 67.61% of the total variance, while the first five components, with eigenvalues  $\geq 1$ , explained 99.39% of the variance. The largest contributions to the F1 component were made by  $K^+$  (9.1%) and  $Ca^{+}$  (8.3%), followed by Hb, sO<sub>2</sub>, pO<sub>2</sub>(T),  $HCO_3^-$ (st), and cBase(B). For the F2 component, the highest contributions were from HTC (16.9%), pH (15.2%), and HHb (13.1%), followed by CoHb,  $O_2$ , pCO<sub>2</sub>, and MetHb. The PCA biplot graph revealed the adaptive capacity of parameters to variable conditions. The parameters located near the origin represented higher adaptability, while  $Na^+$ , Lac, p50, and Glu emerged as significant parameters under HIIT and Q<sub>10</sub> interventions. Notably,  $Na^+$  levels were more stable in the CoQ<sub>10</sub> groups, suggesting a potential role in maintaining electrolyte balance during exercise stress. Meanwhile, Lac levels were significantly lower in the HIIT + Q<sub>10</sub> group compared to the HIIT group, reinforcing the hypothesis that Q<sub>10</sub> enhances lactate clearance and aerobic energy production. Similarly, Glu levels were highest in the HIIT + Q<sub>10</sub> group, indicating enhanced glucose utilization and energy availability, possibly due to Q<sub>10</sub>'s role in mitochondrial efficiency. A strong relationship between the HIIT + Q<sub>10</sub> group and HTC was evident, while pCO<sub>2</sub> demonstrated a notable association with the CoQ<sub>10</sub> group. In the HIIT-only group,  $O_2$  and Lac were prominent in the early phase, whereas Glu dominated in the later phase. This suggests that  $O_2$  emerged as a key parameter in the HIIT + Q<sub>10</sub> group. The vector lengths and

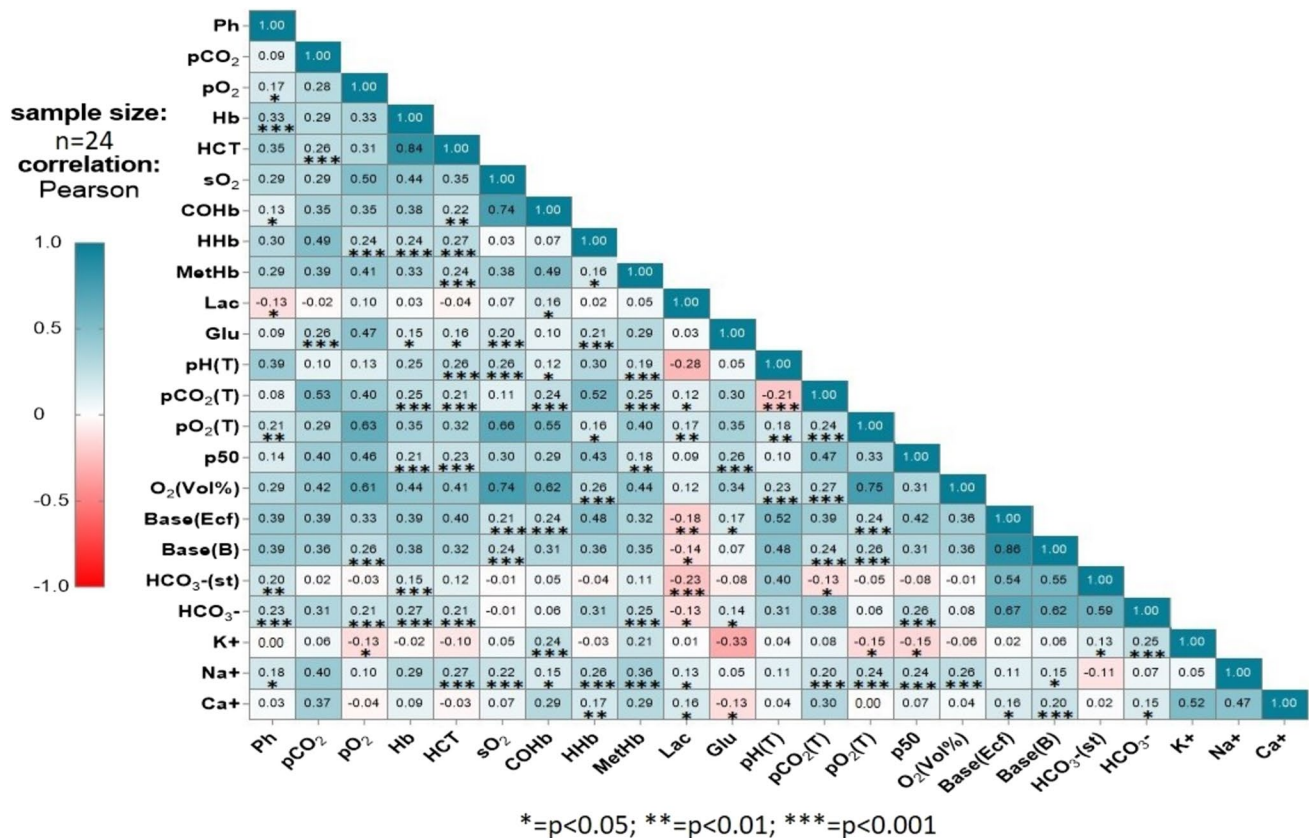


**Fig. 3** Correlation between Lac and other blood gas measurements using Pearson correlation analysis. *pCO<sub>2</sub>* Partial pressure of carbon dioxide, *pO<sub>2</sub>* partial pressure of oxygen, *Hb* hemoglobin, *HCT* hematocrit, *sO<sub>2</sub>* Oxygen saturation, *COHb* Carboxyhemoglobin, *HHb* Deoxyhemoglobin, *MetHb* Methemoglobin, *Lac* Lactate, *Glu*

Glucose, *O<sub>2</sub>(vol%)* Oxygen concentration, *Base(Ecf)* Base excess in extracellular fluid, *Base(B)* Base excess in blood, *HCO<sub>3</sub>-(st)* Standard bicarbonate, *HCO<sub>3</sub><sup>-</sup>* Bicarbonate, *K<sup>+</sup>* Potassium, *Na<sup>+</sup>* Sodium, *Ca<sup>+</sup>* Calcium

angles in the PCA biplot indicated relationships between parameters: angles smaller than 90° indicated positive correlations, while those greater than 90° signified negative correlations. For instance, a strong negative correlation was found between *pO<sub>2</sub>* and *pH*, whereas no significant association was observed with *pCO<sub>2</sub>*. Variations in the responses of parameters within the same group were inferred from the tendency of vectors to diverge from or converge toward the center. These findings suggest that CoQ<sub>10</sub> supplementation may contribute to metabolic optimization during high-intensity exercise, a trend that was clearly visualized through PCA group separation. Notably, MetHb and K<sup>+</sup> were positioned near the C group, whereas pH(T) and HHb distanced themselves from the HIIT and Q<sub>10</sub> factors, occupying distinct regions. The correlation findings indicate that biochemical balance is shaped by multiple factors and that Q<sub>10</sub> supplementation, in combination with HIIT exercise, significantly contributes to physiological adaptation processes (Fig. 4).

## Discussion

Under HIIT conditions, it was observed that lactate threshold levels showed a significant reduction in the HIIT + Q<sub>10</sub> group receiving Q<sub>10</sub> supplementation, with these levels remaining below average. Additionally, the combination of Q<sub>10</sub> supplementation with HIIT resulted in significant improvements in blood gas parameters (*pH*, *pO<sub>2</sub>*, and *pCO<sub>2</sub>*) and enhanced oxygen transport capacity, as indicated by oximetry parameters (*Hb*, *HCT*, and *sO<sub>2</sub>*). Moreover, the reduction in COHb levels highlighted Q<sub>10</sub> supplementation as an effective strategy for alleviating exercise load. From an acid–base balance perspective, positive changes in HCO<sub>3</sub><sup>-</sup> and base values contributed to a reduction in metabolic stress, with this effect becoming more pronounced over time. Regarding electrolyte balance, regulatory effects were particularly noted in K<sup>+</sup> and Na<sup>+</sup> levels. These findings demonstrate that the integration

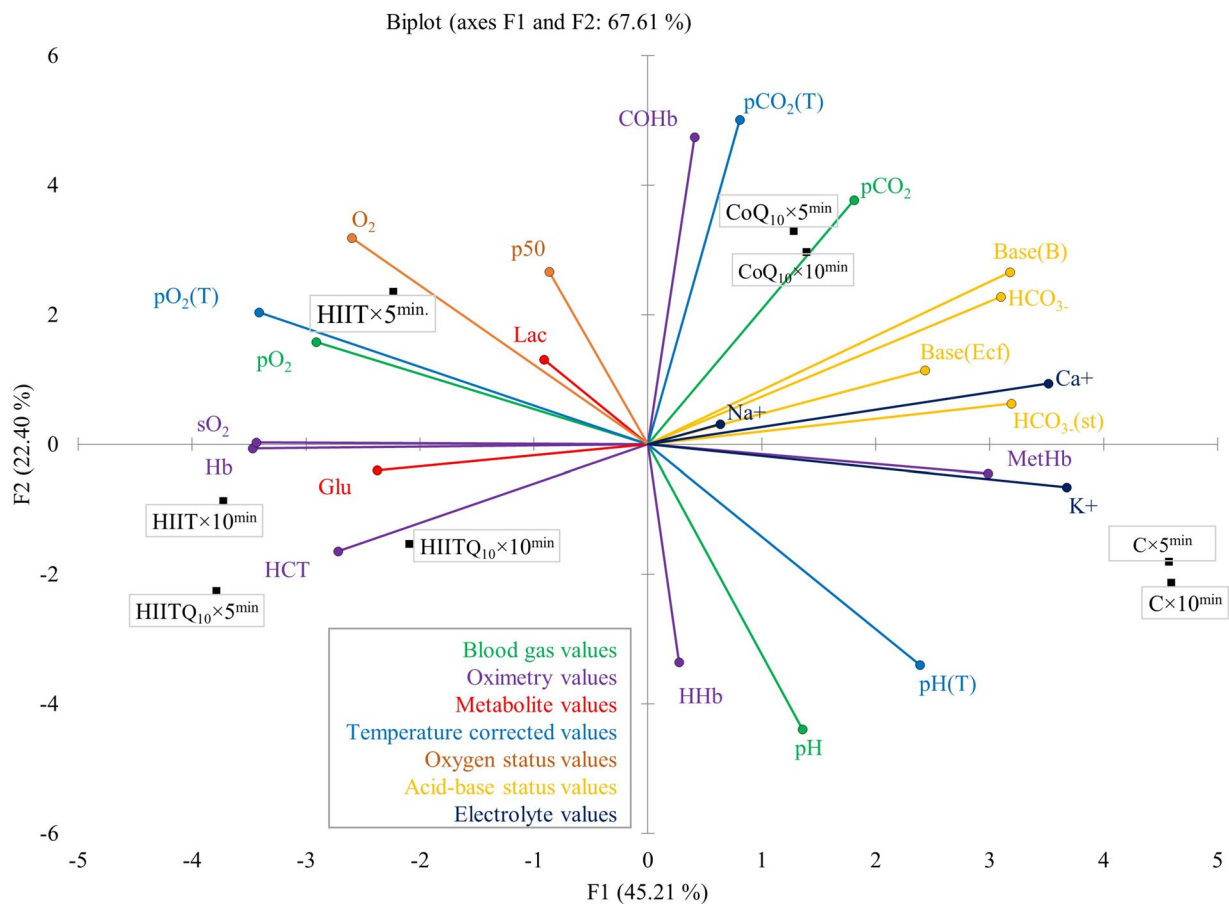


**Fig. 4** Responses of different parameters to changing conditions, differences between groups, and relationships between parameters using principal component analysis (PCA).  $pCO_2$  partial pressure of carbon dioxide,  $pO_2$  partial pressure of oxygen,  $Hb$  hemoglobin,  $HCT$  hematocrit,  $sO_2$  oxygen saturation,  $COHb$  carboxyhemoglobin,  $HHb$

deoxyhemoglobin,  $MetHb$  methemoglobin,  $Lac$  lactate,  $Glu$  glucose,  $O_2(vol\%)$  oxygen concentration,  $Base(Ecf)$  base excess in extracellular fluid,  $Base(B)$  base excess in blood,  $HCO_3-(st)$  Standard bicarbonate,  $HCO_3^-$  bicarbonate,  $K^+$  potassium,  $Na^+$  sodium,  $Ca^{+}$  calcium

of  $Q_{10}$  supplementation with HIIT provides significant benefits on both physiological and metabolic parameters.

The homeostasis of  $H^+$  ions in body fluids and cells is crucial for maintaining healthy cellular biological processes and keeping the pH levels of intracellular and extracellular fluids within narrow limits (Gençoğlu et al. 2022). High-intensity exercises promote  $Lac$  accumulation, triggering  $H^+$  ion release in muscles, which disrupts the body's acid–base homeostasis. This condition shifts pH levels toward acidosis (Fosstveit et al. 2024). In current study, the HIIT group exhibited significant  $Lac$  accumulation and lower pH levels at the first observation time during the I week compared to the C. However, by the second observation time, partial improvement in buffering capacity had brought pH levels to an acceptable range. Cao et al. (2021) reported that prolonged HIIT in rats stimulated lactate production in muscles, increasing blood lactic acid levels. This created an acidic microenvironment, rapidly lowering pH levels, and, when buffering capacity was insufficient, negatively impacted

performance. Thus, when HIIT is performed alone, it disrupts acid–base balance and increases metabolic stress. This situation indicates increased metabolic acidosis associated with impaired acid–base balance during HIIT (Lühker et al. 2018). High-intensity exercises involving the activation of large muscle groups, such as running, negatively affect acid–base parameters like arterial pH,  $Base(Ecf)$ ,  $Base(B)$ ,  $HCO_3-(st)$ , and  $HCO_3^-$  in a short period (Lindinger and Heigenhauser 2012). However, from the second week onward,  $Q_{10}$  supplementation mitigated these effects by significantly limiting  $Lac$  accumulation and enhancing buffering capacity in the HIIT +  $Q_{10}$  group. Specifically,  $Base(Ecf)$ ,  $Base(B)$ ,  $HCO_3-(st)$ , and  $HCO_3^-$  values indicated better maintenance of acid–base balance, with notable improvements in recovery during the HIIT +  $Q_{10}$  group. By the fourth week, the HIIT +  $Q_{10}$  group achieved the highest positive values in metabolic parameters, displaying a profile of effectively managed oxidative stress. These results highlight the beneficial contributions of coenzyme  $Q_{10}$  supplementation in

balancing oxidative stress, improving metabolic parameters, and supporting post-exercise performance. This relationship was also reflected in the correlation analysis results.

Broome et al. (2021) provided evidence showing that well-trained middle-aged male cyclists exhibited better time performance and higher power output in an 8-km time trial after 28 days of coenzyme Q<sub>10</sub> supplementation compared to a placebo group. Similarly, Hostrup et al. (2023) reported that 6 weeks of HIIT, performed three times per week by nine young men, reduced K<sup>+</sup> levels in skeletal muscle. In current study, the parameters such as K<sup>+</sup>, Ca<sup>+</sup>, HCT, pH, and HHb played a negative role in performance in the HIIT group. However, the differentiation of the HIIT + Q<sub>10</sub> group from other groups suggested that Q<sub>10</sub> supplementation provided a biological advantage and improved performance within this group. Notably, the significant reduction in Ca<sup>+</sup> levels observed in the HIIT + Q<sub>10</sub> group was statistically significant, suggesting a potential regulatory role of Q<sub>10</sub> in calcium homeostasis. This aligns with the findings of Petrofsky et al. (2012), who suggested that Q<sub>10</sub> stabilizes calcium channels indirectly, reducing intracellular calcium imbalance. Additionally, Dos Santos et al. (2024) linked decreased serum Ca<sup>+</sup> levels after eight weeks of HIIT to an increase in myofilament sensitivity to Ca<sup>+</sup>, which may enhance cardiomyocyte contractile function. These findings indicate that the observed Ca<sup>+</sup> reduction in the HIIT + Q<sub>10</sub> group may contribute to improved muscle contractility and fatigue resistance, further supporting the role of Q<sub>10</sub> in performance enhancement.

The balanced pCO<sub>2</sub> values observed in the HIIT + Q<sub>10</sub> group compared to other groups indicated that Q<sub>10</sub> supplementation plays a stabilizing role under the strenuous conditions of HIIT, enhancing both performance and metabolic balance. This role was further supported by the positive relationship between the HIIT + Q<sub>10</sub> group and HCT. At the first observation time point, high oxygen demand and elevated lactate levels negatively affected performance, while high glucose 2 demand was a significant limiting factor at the second observation point. Emami et al. (2018), investigating the effects of Q<sub>10</sub> concentration during intense physical exercise in elite swimmers, emphasized that a 14-day Q<sub>10</sub> supplementation acted as a cellular stabilizer, regulated core body temperature, and served as a crucial cofactor in oxidative phosphorylation, thereby increasing the time to exhaustion associated with fatigue. In current study, Q<sub>10</sub> supplementation and HIIT interventions influenced pH(T), pCO<sub>2</sub>(T), and pO<sub>2</sub>(T) values differently across groups and weeks. While Q<sub>10</sub> supplementation alone in the CoQ<sub>10</sub> group showed limited effects, the HIIT + Q<sub>10</sub> group positively impacted both performance and metabolic balance.

One of the key processes for performance enhancement is glucose-driven ATP production. Research exploring the effects of HIIT on energy metabolism consistently reported

significant reductions in glucose levels (Flockhart et al. 2023; Chen et al. 2024). Additionally, Q<sub>10</sub> supplementation has been shown to increase 2,3-diphosphoglycerate levels in erythrocytes, facilitating pO<sub>2</sub> transfer at specific pressures. This mechanism not only enhances oxygen delivery to skeletal muscles but also improves oxygen availability in cardiac and respiratory muscles, potentially boosting ATP synthesis, optimizing glucose utilization, reducing lactate production, and increasing VO<sub>2</sub>max (Zheng and Moritani 2008). In current study, Q<sub>10</sub> supplementation was found to significantly influence Glu levels, with the highest Glu average observed in the HIIT + Q<sub>10</sub> group. This suggests a synergistic effect of Q<sub>10</sub> supplementation and HIIT in regulating energy metabolism and glucose levels, thereby enhancing time to exhaustion and performance output.

## Conclusion

The Q<sub>10</sub> supplementation has been recognized for its effects on metabolic balance and performance during HIIT training. The supplementation provided significant improvements, particularly in pH and pO<sub>2</sub> values, while contributing to the balanced maintenance of pCO<sub>2</sub> levels in the HIIT + Q<sub>10</sub> group. Lactate (Lac) levels remained high in the HIIT group but showed a marked reduction in the HIIT + Q<sub>10</sub> group. Glucose (Glu) levels, on the other hand, reached their highest values (205.71 mg/dL) in the HIIT + Q<sub>10</sub> group. These findings reveal the potential of Q<sub>10</sub> supplementation to reduce metabolic stress and support energy metabolism. In terms of oximetry parameters, Q<sub>10</sub> supplementation enhanced oxygen transport capacity (Hb, HCT, sO<sub>2</sub>) and reduced COHb levels, thereby alleviating exercise load. Additionally, it demonstrated positive effects on acid–base balance by reducing metabolic load, as indicated by increases in HCO<sub>3</sub><sup>-</sup> and base levels. Regarding electrolyte levels, the stabilizing effects on K<sup>+</sup> and Na<sup>+</sup> were notable, while changes in Ca<sup>+</sup> levels were identified as an important parameter for further investigation. PCA analyses revealed that Q<sub>10</sub> supplementation distinctly separated the HIIT + Q<sub>10</sub> group from the others, providing a performance advantage. Pearson correlation analysis identified low but significant relationships among biochemical parameters, further supporting the contribution of Q<sub>10</sub> supplementation to biochemical adaptation processes. In conclusion, the Q<sub>10</sub> supplementation supported metabolic balance, improved oxygen transport, and stabilized acid–base levels during HIIT. It reduced lactate accumulation, enhanced glucose availability, and alleviated metabolic stress, thereby improving recovery efficiency and physiological adaptation.

## Strengths and limitations

This study's strengths include its use of a randomized controlled design, a statistically powered sample size, and advanced statistical methods (PCA and Pearson's correlation analysis), which enhance the reliability of the findings. The well-structured exercise protocol, incorporating progressive overload and acclimatization, ensures consistency, while the use of advanced equipment, such as the Radiometer 900, strengthens the precision of the results. However, the study has several limitations that must be acknowledged. First, although the sample size was statistically sufficient, it remains relatively small compared to similar preclinical studies, potentially affecting the generalizability of the findings. Second, the controlled laboratory setting provided a standardized environment; however, it does not fully replicate the complex physiological and psychological stressors encountered in real-world athletic or clinical settings. Future studies should consider incorporating additional environmental factors to improve external validity. Third, the exclusive use of male Sprague–Dawley rats limits the applicability of these results to female subjects, who may exhibit different physiological responses to HIIT and CoQ<sub>10</sub> supplementation due to hormonal and metabolic variations. To address this, further research is needed to investigate potential sex-based differences and their impact on exercise-induced metabolic adaptations. Additionally, while the short study duration allowed for focused analysis, longitudinal studies are needed to examine long-term physiological adaptations to HIIT and CoQ<sub>10</sub> supplementation. Finally, the reliance on an animal model, while beneficial for mechanistic insights, necessitates caution when extrapolating findings to human populations. These limitations should be addressed in future research to enhance the applicability and external validity of the results.

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## Declarations

**Conflict of interest** The authors declare that there are no conflicts of interest.

**Data availability** The corresponding author is available to provide the primary data upon request.

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