#### ORIGINAL RESEARCH

# The Effect of Intensity Soccer Training Sessions on Marked Biochemical Indicators of Blood Acidity of Saudi Young Soccer Players

Ghareeb O Alshuwaier 1, Hadeel Ali Ghazzawi 2, Abdulrahman I Alaqil 3,4, Yahya R Alsharif 1, Mohammed Shaab Alibrahim 3, Khalid S Aljaloud 1

<sup>1</sup>Department of Exercise Physiology, College of Sport Sciences and Physical Activity, King Saud University, Riyadh, ZIP 4545, Saudi Arabia; <sup>2</sup>Department of Nutrition and Food Technology, School of Agriculture, The University of Jordan, Amman, 11942, Jordan; <sup>3</sup>Department of Physical Education, College of Education, King Faisal University, Al-Ahsa, ZIP 400, Saudi Arabia; <sup>4</sup>Department of Sports Science and Clinical Biomechanics, University of Southern Denmark, Odense, 5230, Denmark

Correspondence: Abdulrahman I Alaqil, Department of Physical Education, College of Education, King Faisal University, P.O. BOX 31982, Al-Ahsa, ZIP 400, Saudi Arabia, Tel +966 555239166, Email aialaqil@kfu.edu.sa

**Purpose:** To assess the effect of low-to-moderate-intensity exercise on specific blood biomarkers and blood acidity in young Saudi Arabian professional soccer players.

**Methods:** A total of 43 professional soccer players participated in the current study. A cross-sectional research method was used to compare the changes in the following blood biomarkers: calcium, magnesium, glucose, anion-gap metabolic acidosis, and carbon dioxide, after a range of low-to-moderate intensity training sessions. Measurements were taken in two soccer training sessions (Day 1, and Day 2). The exercise intensity was estimated by heart rate percentage of maximal heart rate. Wilcoxon signed-rank testing was used to analyze the results.

**Results:** Significant differences were found between and pre-and post-training sessions for anion-gap metabolic acidosis: Day 1: 12.55 vs 15.4 mmol/L; Day 2: 14.15 vs 16.35 mmol/L; and magnesium: Day 1: 0.82 vs 0.74 mmol/L; Day 2: 0.85 vs 0.74 mmol/L. Exercise caused anion-gap metabolic acidosis concentrations to rise post-training; magnesium levels decreased after training sessions on Day 1 and Day 2. **Conclusion:** The concentrations of anion-gap metabolic acidosis and magnesium were significantly affected by exercise intensity in the subjects, the former rose post-training while the latter fell. More studies are required to investigate the effects of different exercise intensities on other blood biomarkers in soccer players.

Keywords: blood biomarkers, exercise intensity, acidity, serum

#### Introduction

Football, called soccer in some countries, is the most popular sport in the world.<sup>1</sup> It is a vigorous team sport that requires numerous technical and tactical skills as well as concentration, precision, speed, acceleration, and rapid changes of direction.<sup>2</sup> During an average 90-minute match, soccer players perform between 1300–1400 different motor activities.<sup>3</sup> Major changes in acid-base balance occur during such high-intensity repeated motor activity, causing gradual fatigue. Players' can reach up to 85–90% of their maximal heart rate during elite-level matches, combined with glycogen depletion and increases in blood lactate concentrations.<sup>4</sup>

Monitoring the intensity of soccer training is crucial for increasing its effectiveness and matching players' current levels. An exact assessment of training intensity allows coaches to prevent undertraining or overtraining and guarantee that players are in top condition for competition. Heart rate monitoring is an established technique for checking training intensity load in soccer.<sup>5</sup> Different motor movements involve players covering 10,500–12,000 meters during training sessions varying between walKing, jogging, running backwards, striding, and sprinting. Crucially, the ability to sprint repeatedly in soccer is a key component of success for players.<sup>3</sup>

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Exercise intensity differs between friendly training matches and competition matches. During the former, players perform at a moderate to high exercise intensity, while in the latter they tend to perform at a high exercise intensity for the duration of the match. Technical training sessions involve players exercising at a lower intensity compared to training and competition matches.<sup>6</sup>

The concentration of ionized calcium in the blood is affected by blood pH as hydrogen ions and calcium compete for binding sites on albumin as well as other proteins. However, the relationship between pH and the concentration of ionized magnesium is not well understood. The concentrations of both ionized calcium and ionized magnesium decrease as blood pH increases, indicating the stronger bonds between these ions with proteins in a more alkaline environment.<sup>7</sup>

Due to the impact of thermal stress caused by high body and environmental temperatures and humidity, the physical performance of soccer players decreases,<sup>8</sup> causing reductions in the distances covered and sprinting ability.<sup>9</sup>

The present study aimed to investigate the effect of training intensity on blood biomarkers that affect blood acidity levels in young Saudi Arabian soccer players. To the best of the researcher's knowledge, this is the first study to assess the impact of low-to-moderate-intensity exercise on the selected blood biomarkers (ie, calcium (Ca), magnesium (Mg), glucose (GLUC), anion-gap metabolic acidosis (AGAP), and carbon dioxide (ECO2)) in Saudi Arabia. We hypothesize that significant differences will be found between the selected blood biomarkers pre-and post-training on Day 1 and Day 2.

#### **Materials and Methods**

The study used a cross-sectional research design to compare the above-mentioned blood biomarkers in young professional soccer players in Riyadh, Saudi Arabia. Two days of measurements were conducted: Day 1 (1st training soccer session) and Day 2 (2nd training soccer session). The period between the measurements taken on Day 1 and Day 2 was 48 hours. All measurements were taken before and after (at rest) the training soccer session in each day and in-season period. The ethics committee at King Saud University approved the study (4/67/352673) and conducted in the guidelines of the Declaration of Helsinki.<sup>10</sup> The researcher informed all subjects of the purpose and procedures involved and signed consent forms were collected.

#### **Participants**

A total of 43 young Saudi professional soccer players participated (mean  $\pm$ : age 20.2  $\pm$  0.84 y, bodyweight 66.0  $\pm$  5.25 kg, height 1.75  $\pm$  0.04 m, and body fat 6.7  $\pm$  1.66%). These players train daily and compete once a week in the Saudi professional league 2016. All measurements were collected in the month of October 2016.

#### Anthropometry Measurement

Subjects' body weight was measured before beginning the study using a digital scale to the nearest 0.1 kg (SeCa813, Germany); height was measured to the nearest 0.01 cm (Seca, 213 Germany). Body fat percentage was measured using skinfold thickness at four sites: triceps, suprailiac, thigh, and abdominals using a Holtain skinfold caliper (Holtain Ltd., Crymych, UK). The skinfold was taken by a well-trained expert in the technique. The sum of these four skinfold measurements was used to calculate total body fat percentage using the following equation:<sup>11</sup> body fat percentage =  $(0.29288 \text{ x sum of skinfolds}) - (0.0005 \text{ x square of the sum of skinfolds}) + (0.15845 \text{ x age}) - 5.76377.^{11}$ 

## **Blood Samples**

Blood samples were obtained from all players on Day 1 and Day 2. The first blood samples were taken 30 minutes before training; the second blood samples were taken 30 minutes after training. All players were asked to visit the medical room in the club pre-and post-training to collect blood samples. All blood samples were collected in 5 mL tubes containing a clot activator (BD Vacutainer System, Plymouth, UK). The blood tubes were placed in cold storage before being immediately sent to the laboratory for analysis. Siemens QuikLYTE<sup>®</sup> Integrated Multisensor was used to analyze (Ca, Mg, AGAP, GLUC, and ECO2). The laboratory calibrated the machine before running the analysis with control samples from the company.

## **Training Sessions**

On Day 1 and Day 2, the sessions consisted of moderate training activities: a warm-up, small side games (5 vs 5, 7 vs 7), and tactics practice. The duration of all training sessions on both days was 110 min. Training sessions were held between 3 pm–5 pm. Each player was fitted with a Polar Pro Team 2 (Polar, Finland) heart rate sensor for each training session to monitor the intensity of exercise. Training intensity was assessed via heartbeat recordings, which were used to calculate the percentage of heart rate achieved during the sessions based on maximal heart rate.

## Statistical Analysis

The data were presented as mean  $\pm$  SD values for the normally distributed variables and as median (range) values for the variables with a skewed distribution. A Wilcoxon signed-rank test was used to compare the differences between the pretraining measurement Day 1 and Day 2, and between the post-training measurements on Day 1 and Day 2. The interquartile range was reported at the 25th and 75th percentiles. All data analysis was carried out using IBM SPSS (version 27, SPSS, Inc. Chicago, Illinois). *P*-values < 0.05 were deemed to indicate statistical significance.

# Results

43 subjects aged between 19–22 years old participated. Table 1 provides a summary of the subjects' characteristics: age, height, weight, body fat percentage, and heart rate percentage for the training sessions on Day 1 and Day 2.

The results of the Wilcoxon signed-rank testing revealed a significant increase in AGAP concentrations between preto post-training on Day 1 (12.55 and 15.4 mmol/L) and Day 2 (14.15 and 16.35 mmol/L), respectively. In addition, the concentration of Mg was lower post-training compared to pre-training on Day 1 and Day 2. However, there were no significant differences in the concentrations of Ca, GLUC, and ECO2 between pre-and post-training (Table 2). In general, intensity training session affected the levels of AGAP and Mg, whereas the levels of Ca, ECO2, and GLUC were not affected by intensity soccer training session.

## Discussion

This study aimed to investigate the impact of the intensity of soccer training exercise on blood biomarkers that may affect blood acidity (eg, the alkaline minerals Ca and Mg) and CO2 and AGAP, as key blood pH indicators. GLUC concentration reflects the fuel used by the body during Physical exercise; its availability prevents the depletion of glycogen stores. The researcher hypothesized that significant differences would be evident among the selected blood biomarkers between pre- and post-training on Day 1 and Day 2.

## Effect on Blood Glucose

Exercise duration, exercise intensity, and the health status of players are the most important determinants of blood GLUC levels.<sup>12</sup> Exercise-induced hypoglycemia caused by overtraining and/or malnutrition poses major risks to health and performance and can be prevented by adequate nutrition and suitable training intensity and rest periods.<sup>13</sup>

Variable	Mean ± SD	Min/Max				
Age (years)	20.2 ± 0.8	(19–22)				
Height (cm)	1.75 ± 0.03	(1.63–1.82)				
Bodyweight (kg)	66 ± 5.3	(55.1–73.7)				
Body Fat (%)	6.67 ± 1.66	(3.90–13.70)				
Heart rate dayI(%)	64.88 ± 7.09	(52–80)				
Heart rate day 2 (%)	71.9 ± 6.6	(57–87)				

Table I	Participant	Variables:	Mean	±	SD,	Minimum	(Min),	and
Maximum	(Max)							

Table 2 Wilcoxon Signed-Rank Testing Analysis of the Subjects' Blood Profiles for Day 1 and Day 2 of Training: Median (25th and 75th
Interquartile Percentile Range)

Variables	Day I		Day 2		
	Pre	Post	Pre	Post	
AGAP (mmol/L)	12.55 (11.65–14.48)*	15.4 (12.83–18.42)	14.15(11.6–15.18)*	16.35(14.63-18)	
Mg (mmol/L)	0.82 (0.73–0.85)*	0.74 (0.69–0.78)	0.85(0.8–0.87)*	0.74(0.69–0.78)	
Ca(mmol/L)	2.35 (2.18–2.42)	2.41(2.36–2.51)	2.37(2.34–2.42)	2.42(2.31–2.49)	
ECO2 (mmol/L)	23.37 (22.4–25)	22.7 (21.4–24.1)	23.6(22.2–24.4)	22.27(20.2–23.5)	
GLUC (mmol/L)	4.92 (4.39–5.24)	5.06 (4.76–5.38)	4.99(4.33–5.63)	4.81 (4.25–5.57)	

Note: \*P<0.05 two-tailed, significant from pre- to post-training.

Abbreviations: AGAP, anion-gap metabolic acidosis; Mg, magnesium; Ca, calcium; ECO2, carbon dioxide; GLUC, glucose.

This study found no significant differences in blood glucose concentrations between pre- and post- training. Blood glucose availability is linked to cognitive function and may influence players' soccer skills and tactical proficiency. Recommendations are made to optimize match-day performance by either maintaining blood glucose levels and/or taking ergogenic carbohydrates to enhance performance.<sup>14</sup> The findings also showed that the training strategy followed by the coaches did not lead to excessive fatigue and was effective at maintaining the players' performance.

Circulating glucose levels during exercise rely on energy status, food consumption, exercise intensity, and glycogen loading levels. Decreased glycogen accessibility is ordinarily related to tiredness. When faced with glucose-depleting, high-intensity activity, carbohydrate intake prior to or during prolonged exercise has been shown to restock glycogen reserves, maintain blood glucose levels, and improve performance. Monitoring and checking fasting and longer-term blood glucose levels via measuring the levels of blood biomarkers might help individual competitors improve the suitability of their dietary regimens. Even though fasting blood glucose levels are not directly related to physical performance, competitors will, in general, have lower fasting blood glucose levels, which are related to the intensity of their training routine.<sup>13</sup>

During short-duration, low-to-moderate-intensity exercise (60% VO2 max), blood glucose levels remain unaffected by increasing the production of hepatic glucose to supply the muscles with glucose. Muscle fuel is exclusively glucose during high-intensity exercise. During high-intensity exercise (>80% VO2max), a small rise in blood glucose is seen immediately on exhaustion; it continues for up to one hour and is corrected by an increase in plasma insulin and restoration of muscle glycogen;<sup>15</sup> this accounts for the unchanged concentrations of blood glucose in the current study pre- and post-training.

Pietras' 2013 results concur with those of the present study; it demonstrated that in healthy individuals, blood glucose responses pre-, during, and post-training were similar despite changing from moderate to high-intensity exercise.<sup>16</sup> This consistency in blood glucose concentrations demonstrates that the subjects were well-nourished, trained at a suitable intensity, and had well-suited exercise economy and efficiency.

#### Effect on Blood Anion-Gap Metabolic Acidosis

Ions are the charged molecules (electrolytes) within the cells, intracellular fluid, and blood plasma. Cations are positively charged electrolytes while anions are negatively charged electrolytes. Plasma electrolytes are the most affected electrolytes by exercise. Bone mass gain is positively affected by physical exercise, especially at load-bearing bone sites. Physical exercise continuously interrupts the plasma calcium ion concentration, consequently changing the plasma electrolyte balance.<sup>17</sup>

Emenike's 2014 study investigated the effect of short durations of exercise (45 minutes) on blood electrolytes among random samples of athletes including volleyball players, football players, and handball players pre-and post-exercise.

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Emenike's results concur with the findings of the present study: short-duration exercise that involves sweating causes the levels of serum electrolytes to differ.<sup>18</sup>

The anion gap refers to the difference between the levels of cations (positively charged ions such as K+ and Na+) and anion levels (negatively charged ions such as HCO3- and Cl-).<sup>19</sup> A greater serum anion gap was associated with an odds ratio for low fitness levels.<sup>20</sup> The present study was conducted during warm weather, which lead to sweating; the results indicated that AGAP levels increased from pre- to post-training on Day 1 and Day 2.

#### Effect on Blood Calcium

Blood calcium plays an important function in muscle contraction.<sup>21</sup> Temur, in 2018, found that during exercise, calcium may become infiltrated into the tissue to produce muscle contractions, and consequently blood levels of calcium may decrease.<sup>22</sup>

Exercise can cause a decrease in blood calcium (Ca) and an increase in parathyroid hormone (PTH) and bone resorption. Bone resorption is stimulated early on during exercise to defend against decreases in blood Ca. Vascular Ca content decreases early on during exercise.<sup>23</sup> In contrast, during moderate-intensity exercise, increased levels of serum PTH have been reported, suggesting that such exercise may stimulate bone resorption.<sup>24</sup> The present study found no significant changes in blood calcium due to low-to-moderate-intensity exercise in combination with average training times (110 minutes).

#### Effect on Blood Magnesium

For monitoring recovery efficiency in soccer players, plasma magnesium and iron level measurements are used as an additional tool. Magnesium, which is considered one of the most crucial cofactor minerals, affects muscle contraction and relaxation. Magnesium enhances muscle strength by participating in regulating troponin expression via calcium ion concentration gradients.<sup>25</sup>

On one hand, the present study's results indicated that the serum magnesium concentrations caused by physical activity did not adversely affect magnesium status. On the other hand, serum magnesium tends to remain in the normal range when magnesium intake is adequate,<sup>26</sup> regardless of physical activity levels,<sup>27</sup> which also concurs with the findings of the present study.

The results indicated a negative correlation between magnesium concentration and cumulative match time for players in midfielder/defender positions. Significant variations in magnesium levels in players in other positions were also evident.<sup>28</sup> As magnesium plays an important role in glucose accessibility in the central and peripheral nervous systems, and during exercise, magnesium stimulates lactate release in the muscles, providing magnesium supplements for professional players in the most dynamic positions should be considered.<sup>25</sup>

In response to intense exercise, magnesium moves from the plasma into the red blood cells and is redistributed throughout the body, which increases the loss of magnesium pre-exercise when serum magnesium concentrations remain constant. In addition to previous exercise intensity, the current soccer training session positively affects the magnitude and direction of magnesium redistribution in the body. More intense exercise causes greater requirements for anaerobically and/or glycolytically metabolized energy, causing a greater movement of magnesium into the red blood cells from the plasma.<sup>27</sup> As the present study, magnesium levels concentrations during low-to-moderate-intensity soccer sessions did not significantly differ. These alterations in magnesium were compensated on the day following the training session; specifically, the concentrations of red blood cells, plasma, and urinary magnesium returned to their pre-exercise levels. Therefore, the body's homeostatic mechanisms compensate for the temporary loss of magnesium by equalizing during recovery from a previous exercise session.<sup>27</sup>

#### Effect on Blood Carbon Dioxide

The heart and lungs are the most two important organs to provide the energy required to move the body during exercise. The lungs carry oxygen into the body and remove carbon dioxide, which is the waste product caused by the energy production process. Via the blood, the heart drives oxygen into the required muscles during physical activity. When the muscles are working hard during exercise, the body utilizes more oxygen and produces more carbon dioxide. To adapt to

this additional demand, the breathing rate rises from around 15 times per minute (12 liters of air per minute) at rest, up to around 40–60 times per minute (100 liters of air) during exercise.<sup>29</sup>

The present study showed no significant differences in carbon dioxide (ECO2) during low-to- moderate exercise among the soccer players between pre- and post-training. It is noteworthy that high-intensity exercise leads to a reduction in the body's ECO2 levels (hypocapnia). Respiratory alkalosis disturbance becomes more obvious during recovery or between seasons of intermittent exercise, which leads to neural fatigue state among players.<sup>30</sup>

The body's breathing rate increases in response to exercise, which helps to prevent blood acidity and reduce the pHlowering effects by removing ECO2 from the blood. ECO2 and H+ are released from the muscles into the blood due to the increase in the metabolic rate, setting up concentration gradients in the opposite direction from the O2 gradient. The buffering action of hemoglobin absorbs the extra H+ and ECO2. The carbonic acid equilibrium is affected if the amounts of H+ and CO2 exceed the capacity of hemoglobin. This lowers blood pH, causing acidosis; in response, the lungs and kidneys respond to pH changes by removing ECO2, HCO3-, and H+ from the blood.<sup>31</sup>

A 2001 study indicated that cardiac output can be calculated using ECO2 concentrations (CO2) derived from a combination of venous and arterial blood. This assumed that the ECO2 pressure-concentration relationship is primarily influenced by changes in blood oxyhemoglobin saturation and hemoglobin concentration rather than by changes in pH. The researchers concluded that changes in pH and buffer base affect blood acidity and regulate the CO2 relationship during exercise.<sup>32</sup>

#### Conclusion

This study is the first to assess the effects of low-to-moderate-intensity soccer training session on the above-mentioned blood biomarkers in young Saudi soccer players. AGAP and magnesium were highly affected by low-to-moderate intensity exercise. Moreover, other blood biomarker concentrations were not affected by either low or moderate intensity exercise. Future studies should examine whether high-intensity training could affect other crucial blood biomarkers.

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## Disclosure

The authors report no conflicts of interest in this work.

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