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Analysis Of Ketone Formation In The Urine Of Futsal Players Following A Simulated Match Using Ketostix

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Abstract. High-intensity physical activity such as futsal predominantly relies on the anaerobic energy system and has the potential to trigger the formation of ketone bodies as an alternative energy source. While studies on ketones often focus on fasting, ketogenic diets, or metabolic diseases, research on ketone formation due to intense physical activity remains limited. This study aimed to analyze the formation of urinary ketones in futsal players following a simulated match and to assess the distribution of ketone levels produced. This descriptive observational study involved 22 male futsal players aged 18–25 years. Subjects underwent a simulated futsal match (2 × 20 minutes with a 10-minute halftime break). Urine samples were collected approximately 10 minutes post-match and analyzed using Ketostix to detect urinary ketone levels, classified as negative, mild, moderate, or high. All players tested positive for urinary ketones. A total of 90.9% exhibited mild ketonuria (0.5 mmol/L), 9.1% showed moderate ketonuria (1.0 mmol/L), and no cases of severe ketonuria were observed. These findings indicate that high-intensity futsal activity is sufficient to induce metabolic changes leading to ketogenesis. High-intensity futsal activity can stimulate endogenous ketone production as a response to glycogen depletion. Monitoring ketone levels may serve as an important indicator in designing training, recovery, and nutritional strategies in high-intensity sports. Further studies are recommended to explore the relationship between ketone levels, athletic performance, and metabolic risk in athletes.

Keywords: ketone bodies; high-intensity exercise; metabolism; sports nutrition; performance monitoring.

1 Introduction

High-intensity physical activities such as futsal fall into the category of sports with a predominance of anaerobic energy systems, especially in moments of sprinting, rapid maneuvers, and explosive changes of direction. During high-intensity activities, muscle

contractions use glycogen as the main energy source (Vandoorne et al., 2017). Muscle glycogen plays an important role in determining the body's ability to undergo endurance exercise, and a significant decrease in glycogen can hinder the muscle's ability to perform physical work (Holdsworth et al., 2017). Anaerobic systems, which rely on muscle glycogen as the main source of energy, at certain intensities can cause the body to experience an energy deficit which then triggers the utilization of fat as an alternative energy source through the process of lipolysis. In addition to serving as a substitute for glucose to meet the energy needs of the brain, skeletal muscle and heart, ketone bodies also act as an important signal that encourages the body to conserve carbohydrate reserves by utilizing fat as a more available primary energy source (Holdsworth et al., 2017). One of the end results of this process is the formation of ketone bodies. Ketone body metabolism is an adaptive mechanism maintained by higher organisms to support survival when experiencing energy deprivation or metabolic disturbances (Cox et al., 2016) Ketone bodies (KB), such as acetoacetate (AcAc) and β-hydroxybutyrate (βHB), are water-soluble organic compounds derived from fat and mostly produced in the liver. The production of ketone bodies increases significantly under physiological conditions when carbohydrate (CHO) supply is limited. (M. Evans et al., 2022; Holdsworth et al., 2017)

Ketones are compounds resulting from fatty acid metabolism that are formed when glucose as the main energy source is not available in sufficient quantities. Ketone bodies are acetyl-CoA-derived compounds produced by the liver during conditions of low carbohydrate and high fatty acid availability (E. Evans et al., 2023). The presence of ketones can be utilized as an alternative energy source, especially for the brain and muscles. Ketone bodies (KB), which include acetoacetate (AcAc), acetone, and β-hydroxybutyrate (βHB), are alternative energy sources to carbohydrates (CHO) and fat, produced in the liver during certain physiological conditions (M. Evans et al., 2017). Drastic restriction of carbohydrate intake can stimulate increased production of ketone bodies, which act as an additional energy source for the brain and skeletal muscles (Pinckaers et al., 2017). However, the benefits of ketone bodies do not seem to offset the negative impact of low carbohydrate availability on high-intensity exercise performance, which usually accompanies the strategy of naturally (endogenously) increasing ketone production (Robberechts & Poffé, 2024). The accumulation of ketones in the body can be an early indicator of extreme physiological conditions, including the risk of ketoacidosis, a state in which the blood pH becomes too acidic. Evans in (Leckey et al., 2017) says high levels of ketone bodies in the blood can interfere with the utilization of other substrates in muscle, such as decreasing muscle carbohydrate oxidation suppressing fat breakdown in adipose tissue. Adaptation to a state of ketosis can impair the ability of muscle to use glycogen for oxidative processes, thereby inhibiting the use of more economical energy sources when oxygen supply becomes limited and, thus, inhibiting high-intensity exercise performance (Burke, 2021). Although the state of ketosis is able to reduce carbohydrate dependence by relying on fat oxidation as the main source of energy, it becomes a bottleneck in high-intensity exercise. The anaerobic energy system, which is dominant in explosive activities such as sprinting and acceleration, relies heavily on the availability of carbohydrates. As a result, ketosis can limit the body's ability to achieve maximum performance in high-energy, fast-paced situations.

Research on ketone formation is usually focused on fasting conditions, ketogenic diets, or metabolic diseases such as diabetes mellitus. However, studies on ketone formation due to intense and anaerobic physical activities such as futsal are still rare. Therefore, this study aims to determine whether futsal activities can trigger the formation of ketones in the urine of players, as well as to see the variation in ketone levels that appear after match simulation.

2 Method

This study used a descriptive observational approach with the aim of knowing the formation of ketones in the urine of futsal players after performing high-intensity physical activity. The research subjects consisted of 22 male futsal players, with an age range of 18-25 years, who were purposively selected based on active involvement in futsal training and matches.

The research design used was observation after treatment (post-treatment only). Subjects underwent a simulated futsal match according to official rules (2 × 20 minutes with a 10-minute break), as a form of physical activity with the dominance of the anaerobic energy system. After the match simulation was completed, players were asked to provide a urine sample (±10 minutes after the match) using a sterile urine sample container. The urine samples were then analyzed using the Ketostix test kit, which is a special indicator strip to detect the presence of ketone bodies in urine. The color indicator on the strip is matched with a standard scale to determine ketone levels, which are divided into:

- Negative (0 mmol/L)
- Mild (0.5 mmol/L)
- Moderate (1.0-1.5 mmol/L)
- High (>4 mmol/L) (Package Insert, Bayer Ketostix).

In this study, all test results were recorded in tabular form and analyzed descriptively to see the distribution of ketone levels in urine after futsal activity. Data from the measurement of ketone levels were analyzed quantitatively descriptively to determine the number of subjects who experienced ketone formation and its level. The results were then interpreted in the physiological context of ketone formation due to anaerobic physical activity.

3 Result

Based on the observation of ketone levels in the urine of 22 futsal players after simulating a high-intensity match, all subjects showed ketone formation. The distribution of detected ketone levels is as follows:

Tabel 1. Sebaran Keton

Keton Level (mmol/L)	Number of Players	Percentage
0,5 (mild ketonuria)	20	90%
1,0 (moderat ketonuria)	2	9,1%
>1,0 (severe ketonuria)	0	0%
Total	22	100

All players showed positive results for ketones, with the lowest level being 0.5 mmol/L, and two players reaching 1.0 mmol/L. No players showed negative results or high levels (>1.5 mmol/L).

The results of this study indicate that futsal activities, although classified as short-duration sports, have high intensity and dominance of anaerobic energy systems that are able to stimulate the ketogenesis process.

4 Discussion

Ketogenesis is a condition in which the body begins to break down fat into fatty acids and produce ketone bodies as an alternative energy source, especially when muscle glycogen reserves begin to deplete due to repeated explosive activity (Dearlove et al., 2019a; Myette-Côté et al., 2018). In this study, 90.9% of players showed mild ketone levels, indicating an early utilization of fat as an alternative energy substrate. This indicates that, although the duration of futsal matches is not as long as aerobic sports such as marathon running, the continuous high intensity is sufficient to create typical metabolic conditions that promote ketone formation.

High-intensity physical activity causes a rapid increase in energy demand, which is initially met by muscle glycogen reserves through anaerobic pathways. When these reserves begin to decline, the body will activate fat breakdown (lipolysis) to meet the energy demand, ultimately producing ketone bodies (ketogenesis) (Dearlove et al., 2019; Myette-Côté et al., 2019), (Dearlove et al., 2019) also noted that intense exercise can lead to elevated levels of ketones in the blood and urine, and in some cases can even trigger a mild ketoacidosis condition, although these events are rare in healthy individuals.

Ketone bodies themselves have a number of physiological benefits. (Stubbs et al., 2019) explained that ketone bodies can be an efficient alternative energy source for muscles and the brain, while (Myette-Côté et al., 2019) showed that the use of ketones can increase the efficiency of fat metabolism. Thus, the athlete's body can optimize the use of fat as energy without having to fully rely on carbohydrates, which in high-intensity exercise are quickly depleted. However, it is important to note that the accumulation of excessive amounts of ketones may carry health risks. Ketoacidosis, a condition in which ketone levels are too high and cause a drop in blood pH, is a serious complication that can occur especially if fluid and electrolyte management is not optimal (Dearlove et al., 2019a). In this study, although no cases of severe ketonuria were found, the presence of mild ketonuria suggests that intense sports activities such as futsal are enough to significantly alter the body's metabolic profile.

Interestingly, recent research findings related to the use of exogenous ketones as a sports supplement provide an additional perspective to these results. Studies (Myette-Côté et al., 2018; Myette-Côté et al., 2019) showed that consumption of ketone monoester beverages can decrease the glycemic response to a glucose challenge, suggesting a shift in energy substrates towards ketone utilization. However, in terms of exercise performance, results are not always consistent. (M. Evans et al., 2019) reported that consumption of ketone monoester supplements did not significantly benefit 10 km running performance, while (Waldman et al., 2018, 2020) found that exogenous ketones did not improve cognitive function after high-intensity exercise. This suggests that while ketones have potential as an energy source, their benefits on improving exercise performance have yet to be confirmed.

In addition, (Stubbs et al., 2018) reported that consumption of ketone esters may decrease levels of ghrelin, a hormone that regulates appetite, thus potentially aiding in the regulation of post-exercise energy intake. However, (Stubbs et al., 2019) also noted that consumption of exogenous ketones may cause gastrointestinal side effects, although they are generally mild and rare. In the context of sports performance, (Scott et al., 2019) and (Shaw et al., 2019) showed that neither 1,3-butanediol supplementation nor the combination of ketones and carbohydrates always provided significant performance improvements in sports such as running or cycling. (Faull et al., 2019) even noted that the state of ketosis may influence athletes' subjective perception of effort levels, which may have implications for pacing strategies or training intensity settings.

These findings suggest that the presence of ketones in the body, whether endogenously produced through physical activity or obtained from exogenous supplementation, has complex metabolic implications. While ketones may serve as an alternative energy source, their presence does not necessarily translate to improved exercise performance. Therefore, monitoring ketone levels in athletes, both through urine and blood, can be one of the important metabolic indicators that help coaches, sports physicians, and nutritionists in designing more targeted training programs, recovery, and nutritional strategies, especially in high-intensity sports such as futsal.

Furthermore, the results of this study also open up opportunities for further research into the influence of pre-exercise duration, intensity and nutritional status on endogenous ketone formation. Future research could also explore whether real-time ketone monitoring can help prevent metabolic risks such as ketoacidosis or even be a tool to optimize fat burning during exercise. Thus, this study provides an important initial contribution to understanding the energy metabolism aspect of futsal, which has been mostly focused on the technical and tactical aspects of the game.

5 Conclusion

This study shows that high-intensity and predominantly anaerobic physical activities such as futsal games can trigger the formation of ketones in the urine of players. Of the 22 subjects observed, all showed positive results for the presence of ketone bodies, with 90.9% being at a mild level of ketonuria (0.5 mmol/L) and 9.1% at a moderate level (1.0 mmol/L). This finding proves that despite its short duration, futsal activity is sufficient to promote fat metabolism through the process of ketogenesis.

Ketone formation as a result of alternative energy metabolism has potential benefits in supporting physical performance, but also carries risks if ketone levels increase without control, such as the occurrence of disturbances in the body's acid-base balance (ketoacidosis). Therefore, monitoring ketone levels can be an important indicator in developing training and recovery strategies, especially in high-intensity sports such as futsal.

There are several suggestions from the author that can be considered such as regular ketone monitoring, athletes or futsal players who undergo intensive training should check urine ketone levels regularly, especially when the intensity of training increases or when undergoing a low-carbohydrate diet. Coaches and sports nutritionists are advised to pay attention to the balance between carbohydrate, fat and fluid intake to prevent excessive ketone elevation and the risk of metabolic acidosis. There is a need to educate players about the body's metabolic

response to strenuous exercise, including the risks and benefits of ketone formation, so that they can recognize signs of metabolic fatigue or physiological disorders early.

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