

Research article

## The Relationships between Simulated Tennis Performance and Biomarkers for Nitric Oxide Synthesis

Tolga Akşit ✉, Faruk Turgay, Emine Kutlay, Mehmet Z. Özkol and Faik Vural

Coaching Education Department, Ege University School of Physical Education and Sports, Bornova, Izmir, Turkey

### Abstract

Tennis performance requires a good aerobic endurance and recovering capacity. Nitric oxide (NO) is a gas which is not only a vasodilator and antioxidant but it also regulates the use of oxygen and glucose. The aim of this study was to examine the relationships between simulated tennis performance test (PT) and NOx (sum of nitrate+nitrite) levels and lactate elimination speed (LES). Twenty well trained male tennis players with game levels of ITN 4 (International Tennis Number) and lower (mean  $\pm$  SD; age  $22.9 \pm 2.6$  yrs; height  $1.82 \pm 0.06$  m and mass  $75.7 \pm 8.0$  kg) participated in the study. Participants performed three 4-min bouts and a 2-min continuous groundstroke against balls projected from a tennis ball machine at speeds of 50, 55, 62 and 70 km·h<sup>-1</sup>. After this exercise, subjects were given a 20 min passive rest. After each period and at during the recovery phase; plasma NOx, glucose (GLU) and lactate (LA) levels were determined. LES was calculated during passive recovery. GLU, LA and heart rate (HR) showed a linear increase in comparison to the values in the previous step while PT decreased significantly. Following each period NOx and glucose levels increased independently, but their decreasing rates in recovery phase were related ( $r = 0.470$ ,  $p < 0.05$ ). The successive increase in NOx and GLU parameters between the third and the fourth periods was significant ( $p < 0.05$ ). Only in the third period was there a significant relation between PT and NOx ( $r = 0.494$ ;  $p < 0.05$ ). In the present study, no significant relationship was found between PT and GLU, LA levels and LES. No significant correlation was found between simulated tennis performance and blood NOx levels. However the addition of loads like those in the third period in tennis trainings can be beneficial for performance in trained tennis players. It is recommended that the relationships between tennis performance with NOx and GLU are studied during a real tennis match.

**Key words:** Blood lactate, heart rate, nitric oxide, stroke performance, tennis.

### Introduction

In a sport like tennis which can last 2-6 hours and has short rest periods, performance depends on the player's capacity to perform the intensive exercise intermittently. Therefore, recovery potential is important in tennis. Moreover, performance in tennis is closely related to groundstroke accuracy, service speeds and the percentage of successful hits (Ferrauti et al., 2001).

Studies show that both aerobic and anaerobic metabolisms play an important role in terms of metabolic and functional responses in tennis (Juan et al., 2007). As for energy system, energy adenosine triphosphate (ATP) during the short term bursting efforts is dominantly supplied by anaerobic sources such as creatine phosphate

(CP) degradation and glycolysis. A small amount is yet produced by aerobic metabolism (Glaister, 2005).

Rally periods are approximately 7-10 seconds in modern professional tennis (Kovacs, 2007). Although average levels of blood LA are approximately 3-4 mM in well-trained tennis players at the end of the match, rallies which are long and close to maximum intensity during the match can increase blood LA levels up to 6 mM and over (Lees, 2003). During rest periods, aerobic energy system helps recovery (Christmass et al., 1998; Glaister, 2005; Konig et al., 2001; Kovacs, 2007; Smekal et al., 2001).

High levels of lactic acid prevent fat acid oxidation and inhibit glycolysis (Ahmaidi et al., 1996; Shephard, 1984). Therefore, moving away of the lactate from the blood following an exercise is important for the upcoming performance especially if it is an intermittent and high intensity one.

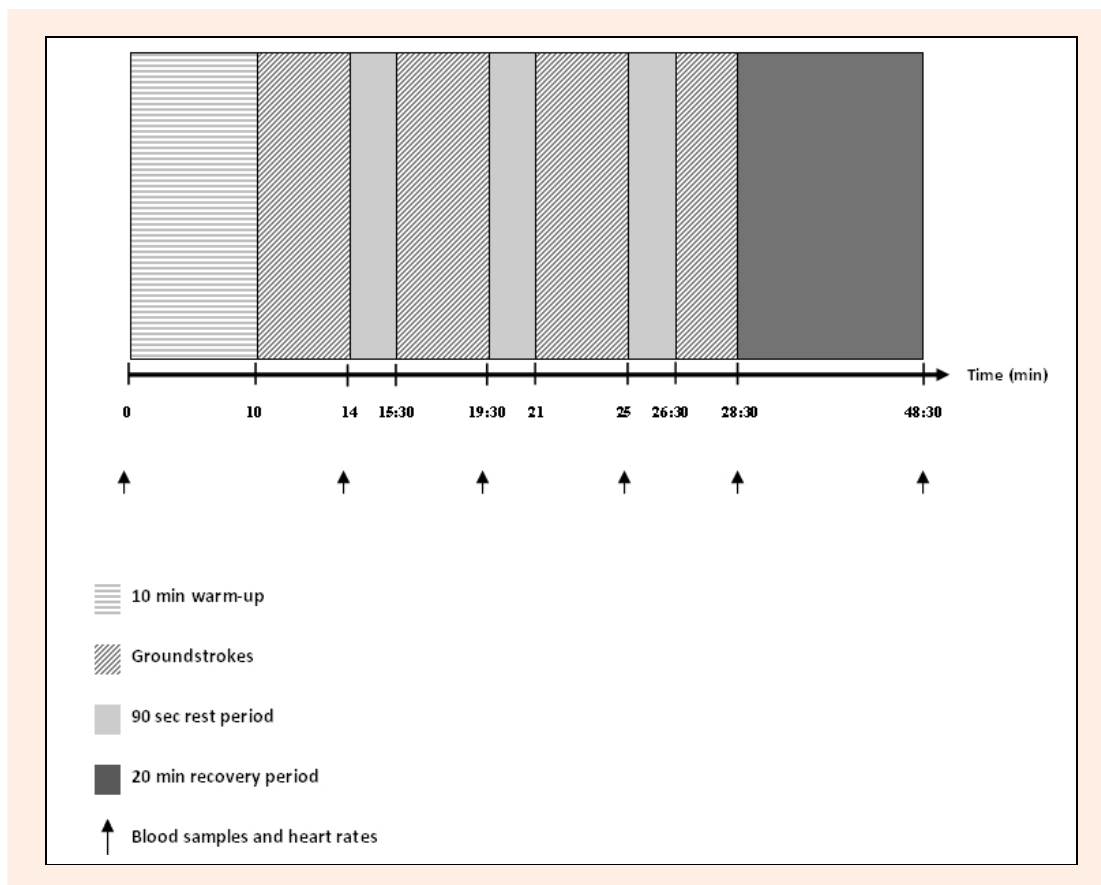
As tennis is a long term activity, glycogen storage may be significantly depleted and lactic acid levels may increase (Lees, 2003). For example, during a game of football which is a sport continuing for a long time like tennis, it is stated that muscle glycogen stores are largely emptied and that this may be associative with the fatigue during football (Fohrenbach et al., 1986). Therefore, decreasing glycogen and increasing lactic acid levels may play a role in fatigue in long-duration sports as tennis activities (Kovacs, 2007).

The most basic way of eliminating lactate is oxidation in working muscles. Skeletal muscle which uptakes a major part of the LA occurring during recovery, the liver, the heart and kidneys play an important role in LA metabolism (Asmussen, 1979; Costill et al., 1997).

Endurance training increases lactate clearance. The reason for this may be the increased lactate transport and oxidative capacity of the muscle or the fall of glycolysis (Stallknecht et al., 1998). A statistically significant difference has been expressed between endurance training level and maximum lactate elimination (clearance). Therefore, maximum lactate level can be used to define the endurance level of an athlete (Messonnier et al., 2001; Francaux et al., 1993). Significant relationships were observed between endurance capacity and LA elimination and NOx levels (Jungersten et al., 1997; Tomlin and Wenger, 2001; Dumlapinar et al., 2006).

In addition, it was shown that intravenous L-arginine significantly reduced exercise-induced increase in plasma lactate and ammonia and enhances the L-arginine NOx pathway during exercise (Schaefer, 2002).

In biological systems NOx is produced via nitric oxide synthase (NOS) enzyme. There are three different



**Figure 1.** Schematic illustration of the experimental protocol.

isoforms of NOS enzyme which cause NO<sub>x</sub> production from L-arginine amino acid. One of these isoforms mentioned is in the vascular endothelium which is called the endothelial NOS (eNOS), it secretes NO<sub>x</sub> which enables blood vessels to relax. Thus, it plays an important role in blood pressure. Neuronal NOS, however, exists in the brain, in the neuronal tissue and in tissues such as skeletal muscles. Inducible NOS (iNOS) is produced in macrophage and other cells against inflammation and infection. eNOS and nNOS are expressed in the body and nNOS release goes up with crush injuries, severe injuries and muscular activity occurring with effort (Kingwell, 2000).

As tennis is a sports activity which is both intermittent (Daniel et al., 2007; Reid et al., 2008) and has short term bursting efforts, NO<sub>x</sub> can be produced in large amounts during a game.

Jungersten et al. (1997), studied healthy groups with different physical fitness levels (exercise capacity) after a jogging exercise of approximately 2 hours and found that plasma nitrate was significantly higher in endurance and sedentary athletes than their resting levels. NO levels were shown to be high after exercise especially in trained athletes (Maeda et al., 2001). These may from changes in oxidative balance and (or) increased endothelial shear stress induced by exercise. Shear stress is the potent activator of NO synthesis activity (Kingwell, 2000).

Aerobic capacity is an important factor in elimination of the lactate in blood both during the exercise and the recovery after the exercise (Astrand and Rodahl,

1986). Therefore, with the increased blood flow due to the high NO<sub>x</sub> to important organs such as the liver, heart and kidneys for lactate metabolism as well as the muscle which is an important lactate (Astrand and Rodahl, 1986, Costill et al., 1997), recovery may become more efficient via the transport of more substrate and hormones. Thus, in athletes with a greater vasodilator reserve dependent on endothelium lactate is expected to be metabolized faster. Moreover, it is stated that NO, which has an antioxidant feature, increases blood glucose uptake independent of blood flow, inhibits glycolysis and CP collapse and therefore contribute to the preservation of intracellular muscle energy stores (Kingwell, 2000).

NO<sub>x</sub> may affect blood GLU and LA levels and during tennis match. Moreover, due its other features mentioned, it may also have a role in tennis performance. However, no study has been found examining the relationship of NO<sub>x</sub> and GLU with LA elimination together with NO<sub>x</sub> and performance relation in tennis. In the present study, it was aimed to analyze the relationship of simulated the tennis performance with NO<sub>x</sub>, LA and GLU parameters.

## Methods

### Participants

Twenty well-trained tennis players participated in the present study. Players whose game levels are ITN 4 and lower, which is the official on court assessment of the International Tennis Federation, participated in the study.

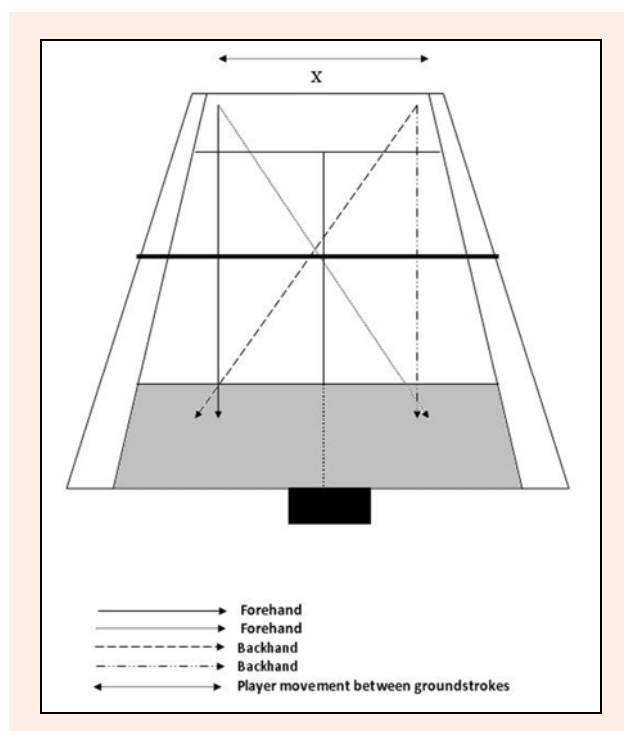
The players trained  $13.7 \pm 3 \text{ h}\cdot\text{wk}^{-1}$  and had a training background of  $12 \pm 2$  years. University School of Medicine's Ethics Committee approved the study and the subjects provided written consent to participate in the study.

### Study design

Anthropometric body measurements of the volunteers were taken (height, body weight, BMI). Anaerobic threshold (AT) test was carried out under laboratory conditions. As of the end of this test, at least a week later, on court groundstroke performance was evaluated; resting HR, basal and exercise NO<sub>x</sub>, GLU and LA values were taken. When the groundstroke performance ended, recovery period started for the volunteers and HR was observed continuously. After 20 minutes, blood was taken from fingertips again and NO<sub>x</sub>, LA and GLU levels were determined. Fluid intake of the athletes was not restricted during test and recovery periods.

### On court groundstroke performance test pattern

NO<sub>x</sub>, LA and GLU levels (in rest period) of the participants were determined in the blood taken from their fingertips. After a 10-minute warm up on the court, the first three periods were held for 4 minutes and the last period for 2 minutes for the groundstroke performance test. 90-second-breaks were taken between each period and meanwhile blood was taken to determine the parameters. For performance assessment, targets were rated as "1" on the court and were designed as in Figure 1. Balls were hit in the periods as forehand parallel, backhand parallel, forehand cross-court and backhand cross-court respectively (Figure 2 and 3). PT is the rate of accuracy of the hits in each period.

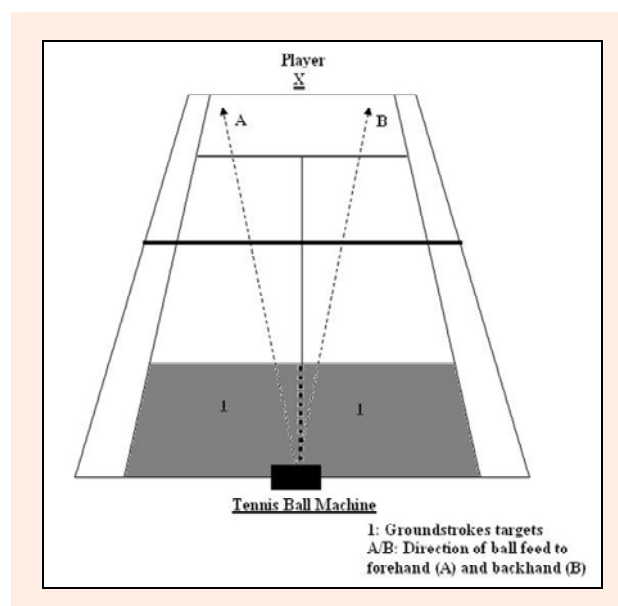


**Figure 2.** Forehand/Backhand performance test pattern schematic representation.

### Measurements

Height and body weight were measured in shorts without shoes by means of height and weight scales.

The measurement of anaerobic threshold speed (ATS): ATS is frequently used both as the criteria for aerobic endurance and the training loads to improve it and to observe the development in endurance performance. The ATS values of the athletes were determined by the each step of which continued until voluntary fatigue with increased loads. Each step of the exercise was designed as 5 minutes and continued by increasing the speed by  $1.2 \text{ km}\cdot\text{h}^{-1}$ . In the one-minute-passive resting periods between each step blood was taken from finger tips for LA analysis. ATS was calculated by (speed corresponding to blood 4 mM LA value) LA-speed graph.



**Figure 3.** Forehand/Backhand performance test pattern schematic representation.

In the recovery period, blood was from each taken tennis player's finger tip twice; at the second and the twentieth minutes after the end of the test. Athletes rested for a total of 20 minutes by sitting. The values obtained by the division of the difference between the values of the second and the twentieth minutes into the time passed (minutes) were called  $\Delta\text{NO}_x$  and  $\Delta\text{GLU}$ .

During the test on the court and 20 minutes after the last step, blood was taken from fingertips into 4 ice-stored heparin containing capillary tubes. Approximately 75 micro liter of blood was taken into each capillary tube. In order to obtain plasma, three of them were emptied into an eppendorf tube without hemolysis. For lactate analysis, the blood in the other capillary tubes was emptied into the special blood lactate preservative tubes of YSI 1500 Sport Lactate analyzer (YSI incorp,USA) and these tubes were stored at  $+4^\circ\text{C}$  during lactate analysis. The eppendorf tubes were centrifuged immediately for 10 minutes at 2500 revs/min. The upper plasma (Approximately 100 microliter) party was transferred into another empty tube with a pipette and stored at  $+4^\circ\text{C}$  for NO<sub>x</sub> and GLU analysis.

The LA preservative tubes mentioned preserve the glycolysis and coagulation in blood samples for at least 1-2 days in the refrigerator (without freezing). The reason for taking into preservative tubes and testing later was protection from glycolysis and saving time. This procedure lyses the red blood cells and produces higher blood lactate values. The results obtained are of a comparable character with those in the literature.

A heart rate monitor (RS 400; Polar Electro Oy, Kempele, Finland) was employed to continuously measure heart rate during the tests. Real time heart rate data were sent to the monitor from the chest strap by radio waves.

Plasma glucose levels were determined spectrophotometrically via an enzymatic-colorimetric method. GLU results were given as  $\text{mg}\cdot\text{dl}^{-1}$ .

NO has a half-life of a few seconds and in blood it soon oxidizes into nitrite containing vasoinactive and stable metabolites. Nitrite in whole blood is quickly transformed into nitrate. Therefore, the stable metabolites of NO (nitrite and nitrate) were measured to analyze blood NOx (Kingwell, 2000). Serum Plasma nitric oxide analyses for the study were performed using Oxis kits (Oxis International Inc., Beverly Hills, CA, USA). The method is based on spectrophotometric determination of the absorbance of pink azo dye derived from exposure of nitrite to the Griess reagent. Nitrite ( $\text{NO}_2$ ), on the other hand, is generated by reduction of nitrate ( $-\text{NO}_3$ ) the basic metabolite of nitric oxide using cadmium ( $\text{Cd}^{2+}$ ). Using this method, nitrite inherent in the sample and nitrite reduced from nitrate were measured cumulatively. Thus, total nitrite levels were used in the current studies as representatives of blood NO level ( $\mu\text{M}$ ).

### Statistical analyses

Values are expressed as the mean  $\pm$  SD. The Kolmogorov-Smirnov test was used to determine whether the data were normally distributed and it showed a normal distribution ( $p > 0.05$ ). Accordingly, parametric analysis methods were used. For the differences between the repeated measurements of the variables ( $\text{NO}_{\text{R-1-2-3-4}}$ ,  $\text{GLU}_{\text{R-1-2-3-4}}$ ,  $\text{LA}_{\text{R-1-2-3-4}}$ ,  $\text{PT}_{1-2-3-4}$ ,  $\text{HR}_{1-2-3-4}$ ) "One way repeated measurement with ANOVA" test was used. LSD test was used in order to determine which measurement the difference results from. Relationships between blood NOx levels with other parameters were investigated using correlation analysis (Pearson correlation coefficient). SPSS 11.0 (SPSS Inc., Chicago, IL, USA) was used analyze the data. Statistical significance was defined as  $p < 0.05$  and  $p < 0.01$ .

### Results

Anthropometric measurements of the group are given in Table 1.

The total number of the balls thrown by the ball machine was 48 in the first period, 60 in the second, 80 in the third and 48 in the fourth. The total number of the balls hit into the target by the volunteers and the success percentages were found to be as follows: In the first period  $39.6 \pm 6.6$  and 82% success, in the second  $39.5 \pm 7.6$

and 60% success, in the third period  $44.5 \pm 9.1$  and 55% success, in the fourth  $23.1 \pm 8.5$  and 48% success.

**Table 1.** The characteristics of the participants. Data are means ( $\pm$  SD) ( $n=20$ ).

Variables	
Age (yrs)	22.9 (2.6)
Height (m)	1.82 (0.06)
Body mass (kg)	75.7 (8.0)
BMI ( $\text{kg}\cdot\text{m}^2$ )	22.9 (2.1)
ATS ( $\text{km}\cdot\text{h}^{-1}$ )	11.0 (1.2)
ATHR (bpm)	170.7 (3.7)
LES ( $\text{mM}\cdot\text{min}^{-1}$ )	.21 (.09)
HR <sub>R</sub> (bpm)	71.8 (3.7)
GLUe ( $\text{mg}\cdot\text{dl}^{-1}\cdot\text{min}^{-1}$ )	1.06 (1.02)
$\Delta\text{NOe}$ ( $\mu\text{M}\cdot\text{min}^{-1}$ )	1.77 (2.03)

BMI= body mass index; ATS= anaerobic threshold speed; ATHR= heart rate corresponding to anaerobic threshold speed; LES= lactate elimination speed; HR<sub>R</sub>= resting heart rate; GLUe= glucose change rate per minute;  $\Delta\text{NOe}$ = nitric oxide change rate per minute.

GLU values were observed to increase in the fourth period, while blood NOx levels showed a non-significant decrease (% 12.5) at the end of the second period unlike the others in comparison to those occurring at the end of the first period. In the later periods, an increase was observed in comparison to the resting period.

Resting HR of the group before groundstroke performance was  $73 \pm 5$   $\text{beats}\cdot\text{min}^{-1}$  while it was as  $153 \pm 17$ ,  $173 \pm 11$ ,  $186 \pm 7$  and  $196 \pm 6$  bpm respectively at the end of the periods. After 20 minutes of recovery, HR was found to be  $98 \pm 11$  bpm. While the resting LA values before groundstroke performance was  $1.5 \pm .2$  mM, at the end of the periods it was measured to be  $4.1 \pm 1.3$ ,  $5.4 \pm 2.2$ ,  $7.9 \pm 2.7$  and  $11.2 \pm 2.4$  mM respectively. After 20 minutes of recovery LA was found to be  $7.0 \pm 2.7$  mM.

No significance was found in the correlation analyses of PT, LA, NOx, GLU, HR, AT and ATHR levels of the participants in the first and the fourth periods.

In the second period no significant relationship was found between the performance and LA, NOx, GLU and HR parameters. A positive ( $r = 0.501$ ,  $p < 0.05$ ) significant relationship was found between HR and the average LA value in the second period.

Looking at the relationship between performance in the third period and LA, NOx, GLU and HR parameters, it was found out that there was a statistically significant ( $r = 0.494$ ,  $p < 0.05$ ) relation between PT and NOx variables.

The findings obtained showed that  $\text{LA}_1$ - $\text{LA}_2$ - $\text{LA}_3$  and  $\text{LA}_4$  values were significantly different from each other in all periods ( $p < 0.01$ ) (Table 2).

The findings obtained showed that there were significant difference between  $\text{NO}_2 - \text{NO}_4$  and  $\text{NO}_3 - \text{NO}_4$  values.  $\text{NO}_4$  value was observed to be higher than  $\text{NO}_2$  and  $\text{NO}_3$  conversion values in a statistically significant way ( $p < 0.01$ ). No significant difference was found in the NOx values obtained in other periods ( $p < 0.05$ ) (Table 2).

There was a significant difference between the GLU values obtained in the first and the third periods ( $p < 0.05$ ). Also, the differences between GLU values

**Table 2. Comparison results of the variables. Data are means ( $\pm$  standard error) (n=20).**

Variables	Resting (R)	Period 1 (1)	Period 2 (2)	Period 3 (3)	Period 4 (4)
NOx ( $\mu$ M)	43.87 (3.85) <sup>1,†,2#,3#,4,†</sup>	67.3 (21.5) <sup>R,4#</sup>	58.9 (21.6) <sup>R</sup>	73.7 (39.6) <sup>R,4*</sup>	83.3 (39.8) <sup>R,2,3</sup>
LA (mM)	1.55 (.05) <sup>1,2,3,4,†</sup>	4.11 (1.39) <sup>R,2#,3,†,4,†</sup>	5.47 (2.20) <sup>R,1,3,†,4,†</sup>	7.97 (2.76) <sup>R,1,2,4,†</sup>	11.27 (2.40) <sup>R,1,2,3</sup>
GLU (mg-dl <sup>-1</sup> )	91.7 (19.4) <sup>4*</sup>	89.4 (15.5) <sup>4#</sup>	91.3 (15.8) <sup>4#</sup>	95.1 (13.9) <sup>4*</sup>	107.8 (24.9) <sup>R,1,2,3</sup>
Accuracy (%)	-	82.1 (10.8) <sup>2,†,3,†,4,†</sup>	66.0 (12.7) <sup>1,3,†,4,†</sup>	55.6 (11.4) <sup>1,2,4*</sup>	45.8 (15.3) <sup>1,2,3</sup>
HR (bpm)	-	153 (17) <sup>2,†,3,†,4,†</sup>	173 (11) <sup>1,3,†,4,†</sup>	186 (7) <sup>1,2,4,†</sup>	196 (6) <sup>1,2,3</sup>

Superscripts denote significant differences among the periods. \*  $p < 0.05$ , #  $p < 0.01$ , †  $p < 0.001$ .

NOx = nitric oxide; GLU = glucose; HR = heart rate; GLU<sub>R</sub> = resting glucose level; NO<sub>R</sub> = resting nitric oxide level; LA = lactate; LA<sub>R</sub> = resting lactate level; PT = simulate the tennis performance test; Accuracy = percentage of the balls hitting the target.

between the first and the fourth, the second and the fourth and the third and fourth periods were significantly greater ( $p < 0.01$ ) (Table 2).

Percentage values in terms of performance showed a significant negative difference ( $p < 0.01$ ). There was a negative difference in the performance values between the third and the fourth periods ( $p < 0.05$ ) (Table 2).

The findings obtained from the group showed that HR<sub>1</sub>-HR<sub>2</sub>-HR<sub>3</sub> and HR<sub>4</sub> values were significantly different from each other ( $p < 0.01$ ) in all periods (Table 2).

In the group, no significant difference was found between GLU<sub>R</sub>-GLU<sub>2</sub> and GLU<sub>R</sub>-GLU<sub>3</sub> values. Significant differences were found only between GLU<sub>R</sub>-GLU<sub>4</sub> and GLU<sub>3</sub>-GLU<sub>4</sub> values ( $p < 0.05$ ). Findings obtained from the group showed that in all periods, NOx and LA values; and NO<sub>R</sub> and LA<sub>R</sub> values were significantly different from each other ( $p < 0.01$ ) (Table 2).

Positive correlations were found between the decrease rates of  $\Delta$ NO and  $\Delta$ GLU concentrations ( $r = 0.470$ ,  $p < 0.05$ ); and between the decrease rates  $\Delta$ NO<sub>x</sub> and the maximum NOx level at the end of the exercise ( $r = 0.876$ ,  $p = 0.000$ ).

No significant relation was found ( $r = 0.107$ ) in the correlation analyses of the differences between NO<sub>3</sub>-NO<sub>4</sub> and GLU<sub>3</sub>-GLU<sub>4</sub>.

No significant relation was found ( $r = 0.239$ ) in the correlation analyses of the differences between NO<sub>2</sub>-NO<sub>4</sub> and GLU<sub>2</sub>-GLU<sub>4</sub>.

## Discussion

In a previous study, average HR levels of trained athletes between the ages of 20 and 30 were found to be 140-160 bpm during a tennis match (Bergeron et al., 1991). In well-trained tennis players the average LA level was measured as 3-4 mM during a singles match. During long and fast rallies; however, LA level was recorded as 6 mM (Konig et al., 2001). When blood lactate levels reach 7-8 mM technical skills and performance levels were shown to be negatively affected (Davey et al., 2002). These examples show that glycolytic system is included in tennis to varying extents and might have an influence on tennis performance.

In the present study, blood LA values were measured as 4.1 mM in the first period and 11.2 mM in the last period, and HR values were found as 153 and 196 bpm respectively in the same periods. At the end of each period of the exercise model in this study, blood LA, GLU and HR parameters showed a linear increase at more

significant levels in comparison to the previous step, negative decreases of significant levels were observed in groundstroke performances.

The findings of the present study show that the exercise model used is an intermittent maximal exercise the speed of which gradually increases and the periods designed reveal, at least partially, the important physiological stress conditions of a tennis match.

It is stated that there are significant relationships between aerobic capacity levels and LA elimination and CP renewal in the recovery phase following a highly intensive gradually increasing exercise (Tomlin and Wenger, 2001).

In none of the periods of this study were the expected (significant) relations observed between the PT and aerobic endurance (ATS) levels, LA levels and LES parameters of the athletes. Therefore, it can be considered that LA level increase, LA elimination and aerobic endurance do not play a significant role in this simulated tennis performance in this exercise model. But this may not always be true as correlation does not necessarily express causes and effects.

In the studies carried out it was found that L-arginine support, which is the main precursor of nitric oxide, decreases LA levels during a sub maximal exercise in human beings (Schaefer, 2002) and increases the working capacity with the rise in NO production (Maxwell et al., 1998).

In a study which examined the acute effects on exercise (HR-LA levels and oxygen consumption) of L-arginine support, no significant difference was observed in the oxygen consumption and HR increases of this group supported by L-arginine, but LA levels significantly decreased (Schaefer, 2002). These findings support the opinion that L-arginine application decreases plasma LA level and NOx production increases with this support (Schaefer, 2002). However, in the present study, no significant relation was found between blood NOx levels and anaerobic threshold levels and LES. Differences in study design, the number of the subjects participated and the methods of analysis employed could have affected the results of the study.

Blood NOx levels increased significantly in the first period compared with the resting period, whereas at the end of the second period there was an obvious yet non-significant drop in comparison with the value at the end of the first period.

At the end of the continuing third and the fourth periods, both plasma NOx and GLU levels increased

significantly. Plasma blood NO<sub>x</sub> levels at the end of all periods were significantly greater than those in the resting period.

Blood flow during exercise is regulated in order for the oxygen to reach necessary points to meet the needs of the metabolism. It is stated that along with the hyperpolarizing factors released from endothelium during exercise, NO released from skeleton and endothelium. NO<sub>x</sub> plays an important role in many physiological processes such as the regulation of the cellular respiration and vascular tone in resting state. "Shear stress" during the exercise also plays an important role in the production of NO<sub>x</sub> (Green et al., 2004).

These vasodilatation effects in acute period help endothelium function improve in the long term just like in the experimental animals that are given regular exercise. NO<sub>x</sub>, relaxing vessel tonus in resting state, determines the vasodilator capacity during exercise (Bitigen et al., 2006).

Although NO<sub>x</sub> and other blood parameters increased after each step of the exercise, a significant relation was found between blood NO<sub>x</sub> and performance only in the third step. The reason for this may be due to the fact that NO<sub>x</sub> value reached a level at which it can show a significant effect under the physiological conditions of this step only. In this period, LA value is approximately 8 mM, NO<sub>x</sub> 73.7 μM, glucose is 95 mg·dL<sup>-1</sup> which is its normal range and HR in 186 bpm. Considering that the average age of the athletes is 23, maximal heart rate ratio was determined (220-age). As a result, it was observed that the exercise intensity in the 3<sup>rd</sup> period corresponded approximately to the 94% of maximal heart rate ratio. This exercise intensity is similar to the intensity occurring towards the end of a real tennis match.

A study was carried out with healthy men and women on in acute bicycle exercise at several percentages of maximal oxygen use. As a result, it was found out that an exercise of 60% intensity increased NO<sub>x</sub> and vasodilatation dependent on endothelium while a sub maximal exercise (at a 75% of max VO<sub>2</sub>) increased oxidative stress (Goto et al., 2003).

It is difficult to explain exactly the reasons for the relationship between PT and NO<sub>x</sub> in the third period. However, as the conditions of the third period were more suitable for NO increase and function than other conditions, a significant relation might have been observed between NO and PT.

The fact that there was no significant relationship between NO<sub>x</sub> and the performance in the last period might be due to the role of the maximal exercise conditions accompanied by higher NO<sub>x</sub> and LA levels. In the last exercise step, inflammation may occur. In case of inflammation, iNOS, which increases with the rise in macrophages and other immune cells, is stimulated by cytokines and it is not dependent on calcium and calmoduline. High amounts of iNOS have toxic effects on the cell (Kingwell, 2000). It is difficult to know the exact reason for this, but these factors stated in the maximal step may have played a role in finding no significant relation between NO<sub>x</sub> and PT.

Although no significant relation was observed between NO<sub>x</sub> and GLU in any of the steps, after the recov-

ery step, there was a significant relation between the drop rates of NO<sub>x</sub> and GLU per minute.

In a study carried out on trained endurance athletes, during a two-hour cycling exercise and as a result of L-arginin load, which is the precursor of NO<sub>x</sub>; GLU degradation increased while plasma insulin concentration remained unchanged, the increase in fat acid levels dropped and LA levels went up (Carter et al., 1999).

Linearly increasing blood glucose and lactate levels during our exercise periods give the impression that glucose is considerably needed, but the increase in glucose occurs independently of NO<sub>x</sub>. NO<sub>x</sub> has a role in blood flow and glucose uptake so NO appears to be important for glucose transport and uptake.

The reason for the relationship between the decreasing rates of NO<sub>x</sub> and GLU during recovery is not known. However, considering the fact that glucose is stored in the liver as glycogen after exercise, it seems possible that as well as insulin, NO<sub>x</sub> also plays a role in glycogen production from glucose during recovery stage. For this reason, it is recommended that this relationship should be further studied. As the conditions of the present study and a real tennis match are not the same it is not possible to associate the results observed in the study with performance in tennis.

## Conclusion

In conclusion, no significant correlation was found between simulated tennis performance and blood NO<sub>x</sub> levels. However the addition of loads like those in the third period in tennis trainings can be beneficial for performance in trained tennis players. It is recommended that the relationships between tennis performance with NO<sub>x</sub> and GLU are studied during a real tennis match.

## Acknowledgements

The authors would like to thank the coaches and players who participated in this study. This study was supported by Ege University Research Foundation (Project Number: 2007/BIL/033).

## References

- Ahmaidi, S., Granier, P., Taoutaou, Z., Mercier, J., Dubouchaud, H. and Prefaut, C. (1996) Effects of active recovery on plasma lactate and anaerobic power following repeated intensive exercise. *Medicine Science in Sports & Exercise* **28**(4), 450-456.
- Asmussen, E. (1979) Muscle fatigue. *Medicine and Science in Sports* **11**(4), 313-321.
- Astrand, P.O. and Rodahl, K. (1986) *Textbook of work physiology*. McGraw Hill Co, New York.
- Bergeron, M.F., Maresh, C.M., Kraemer, W.J., Abraham, A., Conroy, B. and Gabaree, C. (1991) Tennis: a physiological profile during match play. *International Journal of Sports Medicine* **12**(5), 474-479.
- Bitigen, A., Türkyılmaz, E. and Özdemir, N. (2006) Blood pressure response to treadmill exercise testing. *Journal of The Turkish Society of Cardiology* **34**(6), 376-381. (In Turkish: English abstract).
- Carter, H., Jones, M. and Doust, J. (1999) Effects of incremental test protocol on the lactate minimum speed. *Medicine Science in Sports & Exercise* **31**(6), 837-845.
- Christmass, M.A., Richmond, S.E., Cable, N.T., Arthur, P.G. and Hartmann, P.E. (1998) Exercise intensity and metabolic response in singles tennis. *Journal of Sports Sciences* **16**, 739-841.

- Costill, D.L., Coyle, E.F., Dalsky, G., Evans, W., Fink, W. and Hoopes, D. (1997) Effects of elevated plasma FFA and insulin on muscle glycogen usage during exercise. *The Journal of Applied Physiology* **43**, 695-699.
- Daniel, J.H., Damian, F., Inigo M. and Warren, Y. (2007) Integrated physiological and performance profile of professional tennis. *British Journal of Sports Medicine* **41**, 531-536.
- Davey, P.R., Thorpe, R.D. and Williams, C. (2002) Fatigue decreases skilled tennis performance. *Journal of Sports Sciences* **20**, 311-318.
- Dumlupınar, C., Turgay, F., Bereket S., Yücel, S. and Çeçen, A. (2006) The relation between serum nitric oxide levels and maximal effort, and recovery blood lactate in professional football players. *Turkish Journal of Sports Medicine* **41**, 37-44. (In Turkish: English abstract).
- Endo, T., Imaizumi, T., Tawaga T., Shiramoto, M., Ando, S. and Takeshita, A. (1994) A role of nitric oxide in exercise-induced vasodilation of the forearm. *Circulation* **90**, 2886-2890.
- Ferrauti, A., Bergeron M., Pluim B. and Weber, K. (2001) Physiological responses in tennis and running with similar oxygen uptake. *European Journal of Applied Physiology* **85**, 27-33.
- Fohrenbach, R., Buschmann, J., Liesen, H., Hollmann, W. and Mader, A. (1986) Speed and endurance in soccer players of various sport classes. *Schweiz Z Sportmed* **34(3)**, 113-119.
- Francaux, M.A., Jacqmin, P.A., Sturbois, X.G. (1993) The maximum lactate clearance: a new concept to approach the endurance level of an athlete. *Arch Int Physiol Biochim Biophys* **101(1)**, 57-61.
- Glaister, M. (2005) Multiple sprint work: physiological responses mechanisms of fatigue and the influence of aerobic fitness. *Sports Medicine* **35(9)**, 757-777.
- Goto, C., Higashi, Y., Kimura, M., Noma, K., Hara, K., Nakagawa, K., Kawamura, M., Chayama, K., Yoshizumi, M. and Nara, I. (2003) Effect of different intensities of exercise on endothelium dependent vasodilation in humans role of endothelium dependent nitric oxide and oxidative stress. *Circulation* **108**, 530-535.
- Green, D., Maiorana, A., O'Driscoll, G. and Taylor, R. (2004) Effect of exercise training on endothelium derived nitric oxide function in humans. *Journal of Physiology* **561(1)**, 1-25.
- Murias, J.M., Lanatta, D., Arcuri, C.R. and Laiño, F.A. (2007) Metabolic and functional responses playing tennis on different Surfaces. *Journal of Strength and Conditioning Research* **21(1)**, 112-117.
- Juan, M., Muriias, S., Damian, L., Carlos, R.A. and Fernando A.L. (2007) Metabolic and functional responses playing tennis on different surfaces. *Journal of Strength and Conditioning Research* **21(1)**, 112-117.
- Jungersten L., Ambring A., Wall B. and Wennmalm, A. (1997) Both physical fitness and acute exercise regulate nitric oxide formation in healthy humans. *The Journal of Applied Physiology* **82**, 760-764.
- Kingwell, B.A. (2000) Nitric oxide-mediated metabolic regulation during exercise effects of training in health and cardiovascular disease. *The Journal of the Federation of American Societies for Experimental Biology* **14**, 1685-1696.
- Konig, D., Huonker, M., Schmid, A., Halle, M., Berg, A. and Keul, J. (2001) Cardiovascular, metabolic, and hormonal parameters in Professional tennis players. *Medicine and Science in Sports and Exercise* **33**, 654-658.
- Kovacs, M.S. (2007) Tennis physiology: training the competitive athlete. *Sports Medicine* **37(3)**, 189-198.
- Laplaud, D., Guinot M., Juvin F.A. and Flore, P. (2006) Maximal lactate steady state determination with a single incremental test exercise. *European Journal of Applied Physiology* **96**, 446-452.
- Lees, A. (2003) Science and the major racket sports. *Journal of Sports Sciences* **21**, 707-732.
- Maeda, S., Miyauchi, T., Kakiyama, T., Sugawara, J., Iemitsu, M., Irukayama-Tomobe, Y., Murakami, H., Kumagai, Y., Kuno, S. and Matsuda, M. (2001) Effects of exercise training of 8 weeks and detraining on plasma levels of endothelium-derived factors, endothelin-1 and nitric oxide, in healthy young humans. *Life Sciences* **69**, 1005-1016.
- Maxwell, A., Schauble, E., Bernstein, D. and Cooke J.P. (1998). Limb blood flow during exercise is dependent on nitric oxide. *Circulation* **98**, 369-374.
- Messonnier, L., Freund, H., Féasson, L., Prieur, F., Castells, J., Denis, C., Linossier, M.T., Geysant, A. and Lacour, J.R. (2001) Blood lactate exchange and removal abilities after relative high-intensity exercise: effects of training in normoxia and hypoxia. *European Journal of Applied Physiology* **84**, 403-412.
- Reid, M., Duffield, R., Dawson, B., Baker, J. and Crespo, M. (2008) Quantification of the physiological and performance characteristics of on-court tennis drills. *British Journal of Sports Medicine* **42(2)**, 146-151.
- Reid, M.B. (2001) Nitric oxide, reactive oxygen species and skeletal muscle contraction. *Medicine and Science in Sports and Exercise* **33(3)**, 371-376.
- Schaefer, A., Piquard, F., Geny, B., Doutreleau, S., Lampert, E., Mettauer, B. and Lonsdorfer, J. (2002) L-Arginine reduce exercise induced increase in plasma lactate and ammonia. *International Journal of Sports Medicine* **23(6)**, 403-407.
- Shephard, R.J. (1984) *Biochemistry of physical activity*. Charles C. Thomas Publ., Springfield, Illinois, USA.
- Smekal, G., Von, Duvillard, S.P., Rihacek, C., Pokan, R., Hofmann, P., Baron, R., Tschann, H. and Bachl, N. (2001) A physiological profile of tennis matchplay. *Medicine and Science in Sports and Exercise* **33**, 999-1005.
- Stallknecht, B., Vissing, J. and Galbo, H. (1998) Lactate production and clearance in exercise. Effects of training. A mini-review. *Scandinavian Journal of Medicine and Science in Sports* **8(3)**, 127-31.
- Tomlin, D. and Wenger, H.A. (2001) The relationship between aerobic fitness and recovery from high intensity intermittent exercise. *Sports Medicine* **31(1)**, 1-11.
- Woodman, C.R., Muller, J.M., Laughlin, M.H. and Price, E.M. (1997) Induction of nitric oxide Synthase mRNA in coronary resistance arteries isolated from exercise-training pigs. *American Journal of Physiology* **273**, 2575-2579.

### Key points

- In a sport like tennis which lasts 2-6 hours and has short rests, performance depends on the player's capacity to perform the intensive exercise intermittently. Therefore, recovering potential is important in tennis.
- In none of the periods of this study were the expected (significant) relations observed of the athletes. Therefore, it can be claimed that LA increase, LA elimination and aerobic endurance do not play a significant role in the performance and the decrease in performance in this exercise model.

### AUTHORS BIOGRAPHY



#### Tolga AKSIT

#### Employment

Assistant Professor at the Coaching Education Department, School of Physical Education and Sports, Ege University, Bornova, Izmir, Turkey.

#### Degree

PhD

#### Research interests

Strength training and conditioning, tennis physiology, performance analysis in tennis.

**E-mail:** [tolga.aksit@ege.edu.tr](mailto:tolga.aksit@ege.edu.tr)


**Faruk TURGAY**
**Employment**

Assistant Professor at the Coaching Education Department, School of Physical Education and Sports, Ege University, Bornova, Izmir, Turkey.

**Degree**

PhD

**Research interests**

Exercise biochemistry, exercise testing.

**E-mail:** fturgay@yahoo.com


**Emine KUTLAY**
**Employment**

Assistant Professor at the Coaching Education Department, School of Physical Education and Sports, Ege University, Bornova, Izmir, Turkey.

**Degree**

PhD

**Research interests**

Sports nutrition, gymnastics, anthropometric measures.


**Mehmet Zeki OZKOL**
**Employment**

Assistant Professor at the Coaching Education Department, School of Physical Education and Sports, Ege University, Bornova, Izmir, Turkey.

**Degree**

PhD

**Research interests**

Exercise physiology, strength training and conditioning in water polo, game analysis in water polo, statistic methods in sports.

**E-mail:** zeki.ozkol@ege.edu.tr


**Faik VURAL**
**Employment**

Assistant at the Coaching Education Department, School of Physical Education and Sports, Ege University, Bornova, Izmir, Turkey.

**Degree**

MSc

**Research interests**

Strength training and conditioning, performance analysis in soccer, game analysis in soccer.

**E-mail:** faik.vural@ege.edu.tr

**✉ Dr. Tolga Akşit**

Coaching Education Department, Ege University School of Physical Education and Sports, 35100 Bornova, Izmir, Turkey