Sauna effect on blood oxygen transport and prooxidant-antioxidant balance in athletes

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Introduction. Study of sauna effects on the athletes' organism is an important problem, but the data on sauna impact on the blood O_2 transport and lipid peroxidation processes in the athletes' body in the literature is lacking. The purpose of study was to evaluate the state of blood oxygen transport and prooxidant-antioxidant balance in athletes after a single sauna exposure as well as after a sauna course. *Material and Methods*. The effect of sauna (temperature 85-90°C, relative humidity 10-15%) on blood oxygen transport and prooxidant-antioxidant balance in qualified male athletes-wrestlers (age=18-25; n=16) was investigated. *Results*. A single sauna procedure caused respiratory alkalosis (increased blood pH by 0.8% (p<0.001)), elevation of O_2 concentration in venous blood by 53.3% (p<0.001), increase P_{50} (P_{O_2} at which hemoglobin is 50% saturated) by 10.99% (p<0.001), thus reflecting the right shift of oxyhemoglobin-dissociation curve, development of oxidative stress. After a sauna course (1 procedure every week for five months), there was a similar pattern of blood oxygen transport parameters change and normalization of prooxidant-antioxidant balance. The growth of nitric oxide (NO) production, according to increased plasma nitrate/nitrite concentration after the first procedure by 26.2%, after a final sauna by 25.5%, reflected its contribution to the oxygen-dependent processes under thermal exposure. *Conclusion*. The sauna provides adaptations of oxygen-binding blood properties and mechanisms of antioxidant protection via NO-dependent processes, which can increase the athletes body's resistance to various stress-inducing factors.
Key words: *sauna, blood oxygen transport, lipid peroxidation, antioxidants, nitric oxide, athletes*.

Introduction

Physical activity induces significant changes in metabolism, gas exchange and acid-base balance (1,2), determining changes in blood oxygen transport mechanisms functioning. Thus, 3 hours of playing volleyball increase P_{50} (blood P_{O_2} at its 50% O_2 saturation) from 26±0,7 to 29±0,8mmHg in low-qualified athletes and from 28±0,7 to 30±0,8mmHg in highly qualified athletes, indicating the increase of O_2 flow to tissues during physical activity (3).

In addition, high-intensity exercises are accompanied by lipid peroxidation activation and increased free-radical production (4), reflecting oxidative stress development.

Study of sauna effects on the athletes' organism is an important problem (5,6). Sauna causes dehydration, changes in blood volume, increases the cardiovascular and respiratory systems performance, increases anaerobic metabolism threshold and improves neuromuscular and central nervous system activity (7). Sauna significantly changes the organism's oxygen supply. Thus, Nguyen Y. (8) reported what O_2 consumption is increased by 20% during sauna exposure.

However, the data on sauna impact on the blood O_2 transport and lipid peroxidation processes in the athletes' body in the literature is lacking. In this regard the aim of study was to evaluate the state of blood oxygen transport and prooxidant-antioxidant balance in athletes after a single sauna exposure as well as after a sauna course.

Material and Methods

Subjects and Ethical Approval. We examined healthy qualified male athletes-wrestlers (n=16) aged from 19 to 25 years. The Grodno State Medical University Ethics Committee approved all procedures and each subject provided informed consent prior to participating.

Experimental Design. Sauna procedure included two thermal exposures (5 and 10 minutes) at a temperature of 85-90°C, relative humidity 10-15% with a 5-minute interval at a temperature of 20-21°C.

The course of thermal exposure consisted of one procedure provided once a week during 5 months. Blood samples (8 ml) from the cubital vein were obtained before and after the first (n=16) and final procedure (n=12).

Blood Oxygen Transport Analysis

The body temperature was measured with the electronic thermometer MT 1831 «Microlife» in the left axilla. Oxygen tension (P_{O_2}), oxygen content (Cv_{O_2}), oxygen saturation (Sv_{O_2}), hemoglobin and methemoglobin level, blood oxygen capacity (BOC) and acid-base balance (pH), venous carbon dioxide pressure (P_{CO_2}), concentration of bicarbonate (HCO_3^-), the concentration of total carbon dioxide (T_{CO_2}), the actual excess of buffer bases (ABE), the standard excess of buffer bases (SBE) and the standard bicarbonate (SBC) were measured with a micro gas analyser Synthesis-15 (Instrumentation Laboratory) at a temperature of 37 °C.

Bicarbonate, standard bicarbonate and total carbonic acid concentration, buffer basis deficiency/redundancy actual and standard level where measured with the Siggaard-Andersen nomogram.

Hemoglobin-oxygen affinity was evaluated by P_{50} (blood P_{O_2} at its 50% O_2 saturation) determined spectrophotometrically at 37 °C, pH=7.4 and P_{CO_2}=40 mmHg ($P_{50stand}$). The P_{50} at actual pH, P_{CO_2} and temperature (P_{50act}) was calculated from $P_{50stand}$ from the Severinghaus' formula (9) using the temperature coefficient of 0.024. Oxygen dissociation curves were calculated with Hill's equation with n=2.8.

Lipid Peroxidation Measurement

Conjugated diene (CD) concentraion in plasma and erythrocytes was estimated by UV absorption at 232-234 nm (10). The Schiff bases (SB) level was determined by fluorescence intensity of chloroform extracts at excitation and emission wavelengths of 344 and 440 nm, respectively (11).

Antioxidants Activity Estimation

Antioxidant system: α-tocopherol (α-T) concentration was evaluated by fluorescence intensity of hexane extracts at excitation and emission wavelengths of 295 and 326 nm respectively (12); catalase activity (CA) was determined by measurement of the rate of decomposition of hydrogen peroxide capable of generating a stable colored complex at wavelengths of 410 nm (13).

Definition of Nitric Oxide Production

Nitric oxide level was determined by the concentration of its metabolites-nitrates and nitrites (NO_3^-/NO_2^-) in blood plasma. Nitrates/nitrites level was measured by spectrophotometric method at a wavelength of 540 nm using Griess reagent (14).

Data Analysis.

The results obtained were analysed statistically using «Statistica» software. The data distribution was assessed using Shapiro-Wilk's test. All values are reported as median (25 percentile-75 percentile). Statistical significance differences of the data were determined using Wilcoxon test for dependent groups. For all analyses p<0.05 was considered significant.

Results

Sauna increases body temperature, which is less evident after the course of thermal exposure: the increase of athletes' body temperature by 2.6 °C (p<0.001) after a first sauna procedure and by 1.9 °C (p<0.002) after a finale procedure was observed (Figure 1).

Blood oxygen transport characteristics are listed in Table I.

After a first procedure acid-base balance was characterized by increased blood pH by 0.8% (p<0.001). Carbon dioxide pressure was reduced by 22.2% (p<0.001), concentration of total carbon dioxide was reduced by 6.5% (p<0.001), concentration of bicarbonate by 6.01% (p<0.001), and standard excess of buffer bases was reduced by 20.3% (p<0.001).

There was detected the increase in venous oxygen content by 53.3% (p<0.001), oxygen tension by 42.6% (p<0.001), and blood oxygen saturation by 49.4% (p<0.001). There was an increase in hemoglobin concentration by 5.2% (p<0.001), in methemoglobin level by 18.8% (p<0.001), in oxygen capacity of blood by 5.2% (p<0.001). P_{50} under standard value of pH, P_{CO_2} and temperature increased by 3.3% (p<0.020). P_{50} under actual pH, P_{CO_2} and temperature increased by 10.99% (p<0.001) as compared with initial values, thus reflecting the right shift of oxyhemoglobin-dissociation curve (Figure 2).

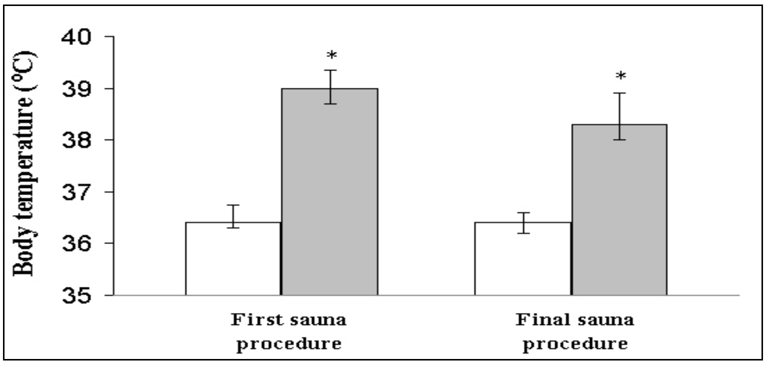

Figure 1. Body temperature before (white bars) and after (gray bars) the first and the final sauna procedures (*: p<0.01).

Table I. Sauna effect on blood oxygen transport in athletes

	First sauna procedure		**Final sauna procedure**	
	Before	After	Before	After
P_{O_2} (mmHg)	27,00 (23,00-31,50)	38,50* (31,00-46,00)	26,00 (23,00-30,50)	35,00$^{\#}$ (28,50-41,50)
Cv_{O_2} (Vol % O_2)	9,85 (7,15-11,10)	15,10* (11,80-1815)	10,50 (8,30-13,00)	16,80$^{\#}$ (12,10-19,45)
Hemoglobin (g/l)	145,50 (140,00-154,00)	153,00* (146,00-159,00)	156,50* (152,50-166,50)	164,50$^{\#\dagger}$ (158,00-175,50)
Sv_{O_2} (%)	47,45 (36,40-52,55)	70,90* (60,50-85,05)	45,35 (39,15-53,00)	64,25$^{\#}$ (47,35-75,90)
BOC (Vol % O_2)	19,79 (19,04-20,94)	20,81* (19,86-21,62)	21,28* (20,74-22,64)	22,37$^{\#\dagger}$ (21,49-23,87)
Methemoglobin (%)	0,80 (0,70-0,90)	0,95* (0,90-1,20)	0,75 (0,65-1,10)	1,10$^{\#}$ (0,95-1,15)
$P_{50_{act}}$ (mmHg)	27,29 (26,16-28,42)	30,29* (28,59-30,83)	26,67 (26,17-26,92)	29,49$^{\#}$ (28,85-30,21)
$P_{50_{stand}}$ (mmHg)	26,20 (25,17-26,90)	27,09* (26,58-27,55)	26,30 (26,02-27,05)	27,56$^{\#}$ (26,40-28,86)
P_{CO_2} (mmHg)	58,35 (52,35-61,75)	45,40* (41,75-49,65)	58,45 (51,45-61,50)	48,30$^{\#}$ (38,60-58,00)
pH (units)	7,338 (7,286-7,364)	7,396* (7,378-7,416)	7,349 (7,328-7,367)	7,397$^{\#}$ (7,348-7,445)
ABE (mM/l)	3,50 (1,65-4,95)	3,05 (2,70-4,10)	4,45* (3,55-6,45)	4,30 (3,75-4,70)
HCO_3^- (mM/l)	29,95 (28,70-32,50)	28,15* (27,15-29,10)	32,25 (30,45-33,85)	29,90$^{\#}$ (26,95-32,20)
T_{CO_2} (mM/l)	31,60 (30,30-34,40)	29,55* (28,45-30,55)	34,15* (32,15-35,75)	31,35$^{\#}$ (28,15-34,05)
SBE (mM/l)	3,70 (2,10-6,25)	2,95* (2,40-3,70)	5,50* (4,85-8,10)	4,85$^{\#\dagger}$ (2,95-6,10)
SBC (mM/l)	26,45 (24,75-27,05)	26,60 (26,00-27,55)	26,75 (26,15-27,60)	27,45 (26,95-27,80)

Data are Me (25‰-75‰). *Significantly different to before first sauna procedure (*P*<0.05). $^{\#}$Significantly different to before final sauna procedure (*P*<0.05). †Significantly different to after first sauna procedure (*P*<0.05).

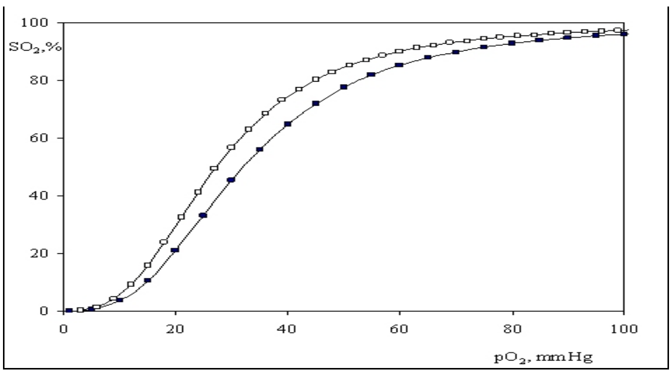

Figure 2. Oxyhemoglobin dissociation curves at actual pH, P_{CO_2} and temperature before (□) and after (♦) the first sauna procedure

There were observed similar changes in the acid-base balance parameters and blood oxygen transport after the final procedure in our study: pH shifted to the alkaline side by 0.7% (p<0.005), carbon dioxide pressure was reduced by 17.4% (p<0.010), concentration of total carbon dioxide by 8.2% (p<0.011), and hydrocarbonate level by 7.3% (p<0.013). Venous oxygen content increased by 60.0% (p<0.003), oxygen tension by 34.6% (p<0.004), hemoglobin level by 5.4% (p<0.026), methemoglobin level by 46.7% (p<0.043). There was an increase of blood oxygen saturation by 41.7% (p<0.004) and oxygen capacity of blood by 5.1% (p<0.026). We observed an increase of the $P_{50stand}$ by 4.8% (p<0.016) and P_{50act} increase by 10.6% (p<0.004), indicating decreasing hemoglobin-oxygen affinity.

Lipid peroxidation processes (Table II) in blood after a first sauna procedure indicated the increase of conjugated dienes level by 37.5% (p<0.016) and Schiff bases content by 7.1% (p<0.039). Increase of free radical processes in erythrocytes was observed, which manifested in the increase of conjugated dienes concentration by 13.8%, (p<0.002) and Schiff bases content by 2.6% (p<0.001).

Antioxidant system was characterized by α-tocopherol level decrease in plasma by 10.6% (p<0.004) and catalase activity decrease in erythrocytes by 7.4% (p<0.002). There were no significant changes of lipid peroxidation processes and antioxidant defense factors after a thermal exposure course.

Plasma nitrate/nitrite concentration data are shown on Figure 3.

Plasma NO_3/NO_2 level after the first procedure increased by 26.2%, after a final sauna by 25.5%, indicating the increase of nitric oxide production in an organism in sauna.

Table II. *Sauna effect on lipid peroxidation and antioxidant system in athletes*

		First sauna procedure		Final sauna procedure	
		Before	After	Before	After
CD, units/ml	Plasma	1.60 (1.28-1.92)	2.24* (2.07-2.56)	1.65 (1.61-1.99)	1.63† (1.55-1.92)
	RBC	10.43 (9.68-11.08)	11.89* (11.64-13.86)	11.01 (10.49-11.98)	13.08 (10.37-15.30)
SB, units/ml	Plasma	142.55 (134.68-145.77)	152.70* (147.85-157.25)	130.71 (94.89-152.57)	90.33† (80.26-173.99)
	RBC	222.64 (218.32-223.78)	228.33* (224.10-231.62)	198.72* (188.03-213.41)	187.16† (175.80-220.00)
α-T, mkM/l	Plasma	24.13 (22.09-26.14)	21.63* (19.90-22.23)	19.90 (17.40-23.41)	21.03 (16.14-24.03)
CA, MH_2O_2/ min/g Hb	RBC	5.27 (4.95-5.61)	4,88* (4.20-4.99)	5.64 (5.39-6.08)	5.39† (5.02-5.74)

Data are Me (25‰-75‰). *Significantly different to before first sauna procedure (*P*<0.05). †Significantly different to after first sauna procedure (*P*<0.05).

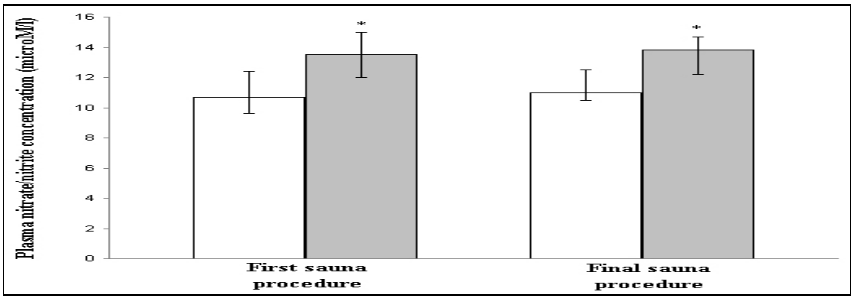

Figure 3. Plasma nitrate/nitrite concentration before (white bars) and after (gray bars) the first and the final sauna procedures (*: p<0.01).

Discussion

The increase of body temperature during the sauna leads to pulmonary hyperventilation (7), causing respiratory determinated pH shift to the alkaline side due to increased CO_2 excretion and development of respiratory alkalosis thus changing, alongside with other factors, oxygen supply.
Tissue oxygenation depends on several factors: pulmonary ventilation, hemoglobin-oxygen affinity, local microvascular tissue perfusion (15).
Hemoglobin oxygen binding properties are responsible for blood oxygenation in the lungs and blood deoxygination in peripheral tissues capillaries (16). Oxyhemoglobin dissociation curve shift direction is of a compensatory-adaptive character. The reduction of hemoglobin's affinity for oxygen promotes more effective tissue oxygenation under hypoxia (17). However, Kwasiborski P.J. (18) using mathematical models showed that under extreme hypoxemic conditions increased hemoglobin-oxygen affinity improved tissues oxygenation in some cases. According to our data, the increase of O_2 concentration and O_2 tension in the venous blood under thermal exposure is caused by increased oxygen consumption during sauna. The observed elevation of p50 and other oxygen transport parameters after a sauna procedure improved O_2 flow in tissues after the thermal exposure.
Free radical oxidation is a physiological process constantly occurring in the organism, with antioxidants providing certain restriction of reactive oxygen species induced oxidation limiting, in turn, its excessive enhancement (19). Blood O_2 concentration increase leads to high reactive oxygen species production (20). Body temperature also rising promotes free radicals production (21). Prooxidant-antioxidant imbalance caused by increased free radicals generation in antioxidant system mechanisms deterioration leads to oxidative stress (22). We observed the lipid peroxidation intensification and antioxidants activity reduction after the first sauna procedure in our study which reflect the oxidative stress development. After a sauna course normalisation of prooxidant-antioxidant balance is detected indicating the increase of body's adaptation abilities to that kind of exposure.
Regular thermal therapy improves hemodynamic and endothelial function. Our experiment showed that sauna enhances mRNA activity and increases endothelial NO synthase proteins expression (23). A course of sauna reduces concentration of 8-epi-prostaglandin $F_{2\alpha}$ indicating thus its protective effect against oxidative stress (24).
Nitric oxide is involved in blood oxygen-binding properties development (25) and due to its free radical and antioxidant properties in prooxidant-antioxidant balances as well (26). NO synthase inhibiting under hyperthermic conditions leads to more significant oxyhemoglobin dissociation curve shift to the right, activation of free radical oxidation and antioxidant mechanisms deterioration (27). Plasma, erythrocytes and tissues (liver, kidneys, and heart) conjugated dienes and Schiff bases concentration increases with rising body temperature caused by lipopolysaccharide injection after preliminary treatment with NO-synthase inhibitor N^{w}-nitro-L-arginine (28).
L-arginine–NO system determines functional hemoglobin properties by modification of its affinity to oxygen via intraerythrocytic regulation mechanisms, oxygen-dependent processes of NO production, and vascular tone regulation etc. (29). Different hemoglobin/NO compounds could alter hemoglobin affinity to oxygen by changing hemoglobin R-conformation to T-conformation, increasing methemoglobin level, nitrosothiol production and others hemoglobin oxidation products (25). There is a constant interaction between NO and other free radicals, particularly with superoxide anion, interaction under physiological conditions in the body; a shift of O_2^-/NO balance from some cases to the superoxide leading to production of highly toxic peroxynitrite ($ONOO^-$) (30), which can cause prooxidant-antioxidant imbalance. Increased nitric oxide level after sauna seems to reflect its contribution to the development of oxygen-dependent processes under thermal exposure.

Conclusion

Therefore sauna procedure in athletes changes acid-base balance and venous blood oxygen transport, exhibiting itself by respiratory alkalosis, P_{O_2} increase, and diminishing hemoglobin-oxygen affinity, leading to increasing tissue oxygenation. A single sauna procedure causes oxidative stress (free radical processes enhancement and antioxidant defense mechanisms deterioration), with its manifestations diminishing after a course of thermal exposure. The observed increase in NO production during a sauna procedure might influence human body's oxygen-dependent processes (blood oxygen transport and prooxidant-antioxidant balance).

Systematic sauna procedures in athletes provide adaptations of oxygen-binding blood properties and mechanisms of antioxidant protection via NO-dependent processes, which can increase the athletes body's resistance to various stress-inducing factors.

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