

Research article

Effects of sodium phosphate loading on aerobic power and capacity in off road cyclists

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Abstract

The main aim of this paper was to evaluate the effects of short-term (6 days) phosphate loading, as well as prolonged (21 days) intake of sodium phosphate on aerobic capacity in off-road cyclists. Nineteen well-trained cyclists were randomly divided into a supplemental (S) and control group (C). Group S was supplemented for 6 days with tri-sodium phosphate, in a dose of 50 mg·kg⁻¹ of FFM/d, while a placebo was provided for the C group. Additionally, group S was further subjected to a 3-week supplementation of 25 mg·kg⁻¹ FFM/d, while group C received 2g of glucose. The results indicate a significant ($p < 0.05$) increase in $\dot{V}O_{2max}$, $\dot{V}E_{max}$, and O_2/HR , due to sodium phosphate intake over 6 days. Also a significant ($p < 0.05$) decrease in HR_{rest} and HR_{max} occurred. The supplementation procedure caused a significant increase ($p < 0.05$) in P_{max} and a shift of VAT towards higher loads. There were no significant changes in the concentration of 2,3-DPG, acid-base balance and lactate concentration, due to phosphate salt intake.

Key words: Tri-sodium phosphate, 2,3-diphosphoglycerate, oxygen uptake, off road cyclists.

Introduction

The theoretical basis for treating phosphate salts as an ergogenic substance is based upon its metabolic functions. Phosphate salts in both inorganic and organic forms play important roles in human metabolism, particularly as related to sport performance. These phosphate compounds may significantly influence both aerobic and anaerobic energy systems.

Phosphates are also a part of the phosphate buffer, thus their intake may enhance performance at or above the lactate threshold (LT). This concept was confirmed by Cade et al. (1984), who observed a significant decrease in lactate concentration during submaximal exercise after phosphate loading. In several other research projects with sodium phosphate intake, a shift in anaerobic threshold towards higher loads was registered (Kreider et al., 1990; Kreider, 1992; Miller et al., 1991).

The improvement of aerobic metabolism through phosphate loading is most likely caused by the impact of phosphates in the formation of 2,3-diphosphoglycerate (2,3-DPG), a compound in the red blood cells that facilitates the release of oxygen to the tissues. One hypothesis states that the erythrocyte increase in 2,3-DPG, due to phosphate loading, allows for improved oxygen supply to the working muscles involved in prolonged endurance exercise (Cade et al., 1979; 1984; Farber et al., 1984;

1987; Gibby et al., 1978). Unfortunately, research results in this area are ambiguous. Some research support this hypothesis, while other studies do not show significant changes in the level of erythrocyte 2,3-DPG following sodium phosphate supplementation (Bredle et al., 1988; Kreider et al., 1990). The discrepancies in the effectiveness of phosphate intake are most likely caused by training status (novice vs. highly trained subjects) and adaptive possibilities (responders and nonresponders) of subjects submitted to supplementation. Although all humans share similar anatomical and physiological traits, they possess biological individuality due either to basic hereditary differences or environmental modifications (Williams, 1998).

Phosphate deficiencies in the body decrease the contractile properties of the heart muscle, which significantly decreases its stroke volume (Fuller et al., 1978). Once again, theoretically, phosphate salt intake should increase cardiac output at rest and during exercise. This hypothesis has been supported by several research projects, whereby phosphate loading caused a decrease in heart rate during continuous endurance exercise (Farber et al., 1984; Moore and Brewer, 1981; Lunne et al., 1990), and an increased stroke volume in such efforts (Kreider et al., 1992).

Current research results in the area of phosphate loading are, thus, very controversial. There are researchers who support the ergogenic effects of phosphate salts on aerobic endurance and those that deny such ergogenic effects. It should be noted that most research data conform to short-term supplementation (3-6 days), with very little data on prolonged phosphate loading (20 days or more). The research on prolonged intake of sodium phosphate salts seems fully justified since many stage races in cycling last from 2 to 3 weeks (ex. Tour de France). Results of such research could have serious practical applications in competitive sports. There is also very limited research on phosphate intake and phosphate-calcium metabolism. The review of literature showed that in most previous experiments, the research material varied, with subjects participating in the studies differing significantly in power output and $\dot{V}O_{2max}$ (Brennan et al., 2001; Cade et al., 1979; Duffy et al., 1986; Mannix et al., 1990; Kreider et al., 1990; Stewart et al., 1990).

Considering the aforementioned issues, the objective of this work was to evaluate the effects of short-term (6 days) phosphate loading, as well as prolonged (21 days) intake of sodium phosphate on aerobic capacity in elite off-road cyclists.

Methods

Subject characteristics

The participants were 20 elite mountain bike cyclists, with at least 5 years of national and international competition. The research was conducted during the competitive season thus the values of aerobic capacity were at maximum or near maximum levels. One of the subjects resigned during the experiment due to injury. All subjects were randomly divided into a supplemented (S) group ($n = 10$; age, 25 ± 1.29 years; VO_{2max} , 73.53 ± 4.46 ml·kg⁻¹·min⁻¹; body height (BH), 1.89 ± 0.06 m; body mass (BM), 70.33 ± 5.36 kg; fat free mass (FFM), 65.22 ± 5.02 kg; fat content (FAT%), 7.01 ± 1.33 %), which received sodium phosphate salts, and a control (C) group which was given a placebo ($n = 9$; age, 24.5 ± 4.31 years; VO_{2max} , 73.88 ± 4.25 ml·kg⁻¹·min⁻¹; BH, 1.73 ± 0.04 m; BM, 66.43 ± 2.31 kg; FFM, 61.45 ± 2.6 kg; FAT%, 7.28 ± 2.97 %).

The research project was approved by the Ethics Committee for Scientific Research at the Academy of Physical Education in Katowice, Poland.

Experimental design

The experiment had three phases. The supplementation procedure was not double blinded, for safety precautions, since the experiment was carried on after the first phase. Before the start of the experiment, initial values of body mass and body composition (BM, FFM, FAT% and total body water (TBW)) were evaluated with the use of electrical impedance. To increase the reliability and validity of body composition measurements by electrical impedance all tested subjects were evaluated under the same conditions during all 3 phases of the experiment (measurement during the same time of the day 7-8 am, active rest the day before testing, full hydration of the body, last meal at 8pm on the day before evaluation).

Resting blood samples were drawn from the antecubical vein and from the fingertip to determine several biochemical variables. A progressive ergocycle test was administered to determine maximal oxygen uptake. The second phase of the experiment included 6 days of supplementation with sodium phosphate for group S, while a placebo in the same dose was provided to group C. The subjects in group S were given tri-sodium phosphate in a dose of 50mg/kg of fat free mass (FFM) per day, divided into four portions. The control group received a placebo in the form of glucose gelatin caps, also administered 4 times daily. After the supplementation the exercise tests were repeated.

During the third phase of the experiment, group S was further subjected to a 3 week intake of sodium phosphate with a dose of 25mg·kg⁻¹ of FFM per day. Group C received 2g of glucose, separated into portions, 4-times per day. After 3 weeks of supplementation, the exercise procedures were repeated once again. One of the safety issues related to prolonged sodium phosphate intake was related to the disturbance of phosphate-calcium balance, thus blood concentration of calcium and phosphate was monitored throughout the experiment.

Most other research projects used a constant dose of 4g of phosphates per day for up to 6 days. In our re-

search the doses of tri-sodium phosphate in the second and third phases of the experiment were chosen in accordance with Minson's (2000) investigation, considering FFM of the subjects. The atmospheric conditions in regards to air pressure (Ist phase-1037; IInd phase-1051; IIIrd phase-1027 hPa) temperature (Ist phase-18,9; IInd phase-18,5; IIIrd phase-19,2 °C) and humidity (Ist phase-65; IInd phase-61; IIIrd phase 66%) were held constant to increase the reliability of measurements. Because both training intensity and volume were rather high during the competitive period, all tested athletes were on a high carbohydrate diet.

Experimental testing

Body mass and body composition were evaluated with electrical impedance (Inbody 720, Biospace Co., Japan) before each phase of the experiment. Capillary blood samples were drawn at rest, after each load and during the 3rd, 6th, 9th, and 12th min of recovery for the evaluation of lactate concentration. Capillary rest and post exercise blood samples were also used to determine acid-base equilibrium and oxygen saturation of hemoglobin. Venous blood samples were drawn, before and after the exercise protocol to determine hemoglobin concentration (Hb), haematocrit value (Hct), number of erythrocytes (RBC), concentration of non-organic phosphates, serum calcium concentration, and the level of 2,3-DPG.

After blood samples were drawn for biochemical evaluations, a progressive maximal exercise test was applied to determine maximal oxygen uptake and the lactate threshold (LT). The LT was determined by the D-max method (Cheng et al. 1992). The test was performed on an ergocycle (Excalibur Sport, Lode BV, The Netherlands), beginning with a work load of 40W, which was increased by that value every 3 minutes until volitional exhaustion. Some authors suggest that beginning at such loads violates the guidelines for measurement of VO_{2max} , since it prolongs the test up to 35-40min, yet our observations indicate that for elite cyclists the loads up to 200W are treated as a warm-up since exercise heart rate rises only up to 120-130bpm and lactate values remain similar to resting level or even lower. Starting at higher loads and increasing them by greater values (50-60W) would not allow for a precise determination of the anaerobic threshold, which was one of the major objectives of this paper. The criteria of reaching VO_{2max} included a gradual decrease in peak VO_2 during maximal workload.

During the exercise protocol, the following variables were constantly registered: heart rate (HR) (Polar Electro Belt compatible with Oxycon Alpha gas analyzer, Finland), minute ventilation (VE), oxygen uptake (VO_2) and expired carbon dioxide (CO_2) (Oxycon Alpha, Jaeger, Germany). At the end of each load, capillary blood samples from the finger tip were drawn to determine lactate concentration. These values allowed to determine the lactate thresholds by the Dmax method for each athlete. The ventilatory threshold was determined by the V-slope method, which was assessed as the first breaking point from linearity of carbon dioxide output (VCO_2) plotted against oxygen uptake (VO_2). One minute after the test, venous blood samples were drawn to determine changes in hemoglobin concentration (Hb), haematocrit value

Table 1. Mean (\pm SD) values of considered variables in the supplemented (S) and control (C) groups during the 3 phases of the experiments.

Variables	Phase 1		Phase 2		Phase 3	
	Group S	Group C	Group S	Group C	Group S	Group C
VO _{2max} (ml·min ⁻¹)	5165 (319)	4907 (328)	5420 (341) *	4911(328)	5457 (376) *	4901 (333)
VO _{2max} (ml·kg ⁻¹ ·min ⁻¹)	73.5 (4.5)	73.9 (4.3)	77.5 (6.0) *	74.0 (4.4)	77.5 (6.0) *	73.3 (4.9)
VE _{max} (l·min ⁻¹)	183 (23)	170 (10)	193 (22) *	171 (9)	196 (18) *	175 (9)
HR _{rest} (bpm)	68 (7)	68 (4)	62 (6) *	69 (3)	62 (8) *	67 (2)
HR _{max} (bpm)	190 (7)	195 (4)	185 (6) *	195 (4)	187 (6) *	194 (3)
RER _{rest}	.83 (.07)	.84 (.11)	.81 (.09)	.85 (.10)	.83 (.08)	.86 (.11)
RER _{max}	1.15 (.05)	1.15 (.04)	1.13 (.03)	1.15 (.04)	1.15 (.04)	1.14 (.03)
O ₂ /HR (ml·bpm ⁻¹)	27.7 (1.8)	25.4 (2.0)	29.3 (1.6) *	25.2 (1.9)	29.2 (2.5) *	25.3 (1.9)
HR _{LT} (bpm)	173 (6)	176 (7)	170 (5) *	175 (6)	169 (1) *	177 (6)
VO _{2LT} (ml·min ⁻¹)	4469 (393)	4354 (491)	4709 (304)	4355 (469)	4624 (326)	4400 (379)
HR _{VAT} (bpm)	163 (6)	171 (7)	160 (8)	172 (7)	161 (8)	170 (7)
VO _{2VAT} (ml·min ⁻¹)	3982 (368)	3894 (291)	4315 (290) *	3908 (263)	4257 (376) *	3929 (246)
P _{max} (W)	404 (28)	390 (19)	415 (267)	390 (19)	425 (32) *#	400 (21)
P _{LT} (W)	320 (28)	291 (19)	329 (24)	290 (16)	328 (22)	194 (19)
P _{VAT} (W)	280 (26)	273 (11)	295 (19) *	274 (11)	295 (22) *	276 (10)

* significantly different from 1st phase (p<0.05); # significantly different from 2nd phase (p<0.05)

(Hct), number of erythrocytes (RBC), the level of 2,3-DPG, acid-base balance variables and oxygen saturation of hemoglobin. These blood samples were necessary for hematological variables which were used to calculate 2,3-DPG concentration per erythrocyte. To determine the level of 2,3-DPG in erythrocytes, the spectrophotometry method was used with Roche Diagnostics kits (Germany).

Statistical analysis

The obtained data were analyzed statistically with the use of Statistica 8.0 (StatSoft). The results were presented as arithmetic means (X) and standard deviations (SD). To determine the influence of tri-sodium phosphate on chosen physiological and biochemical variables, the two way ANOVA (group & treatment) with repeated measures was applied. When significant differences in F ratio were found, the post-hoc Bonferroni test was used. The relationships between particular variables were determined by calculating the Pearson's correlation coefficients. The level of statistical significance was set at p < 0.05.

Results

The average values of chosen physiological variables obtained, as well as the results of ANOVA are presented in Table 1.

The two-way analysis of variance showed a statistically significant effect of the two main factors (group & treatment) on physiological variables such as: VO_{2max}, VE_{max}, resting and exercise HR, O₂/pulse, HR_{LT}, VO_{2VAT}, P_{max}, P_{VAT}. No significant changes were observed in BM, FFM, FAT% and TBW during the experiment in both groups. Additionally a significant interaction of independent factors was observed (Table 1). A statistically significant effect in the main factor „treatment” was registered only in case of HR_{rest}, VO_{2VAT} and P_{VAT}. A statistically significant effect in the other main factor „group” occurred in HR_{VAT} and P_{LT} (Table 1). The supplementation with sodium phosphate did not affect significantly the rest and post exercise values of RER (Table 1). The supplementation procedure applied in this research did not

affect significantly the considered biochemical variables (Table 2).

Table 2. Correlation coefficients between analyzed variables and the concentration of 2,3-DPG at rest and plasma phosphorus (P) concentration during the experiment

Paired Variables	R	p
VO _{2max} & 2,3-DPG _{rest}	.47	.01
VE _{max} & 2,3-DPG _{rest}	.59	.001
O ₂ /HR & 2,3-DPG _{rest}	.41	.05
VO _{2max} & P	.51	.01
VE _{max} & P	.49	.01
HR _{max} & P	-.42	.01
O ₂ /HR & P	.64	.001
pH _{rest} & P	.48	.01
Ca & P	-.43	.01
2,3-DPG _{rest} & P	.48	.01
P _{max} & P	.48	.01
VO _{2VAT} & P	.56	.001
P _{VAT} & P	.58	.001

Post hoc analysis

A significant (p < 0.05) increase in maximal oxygen uptake (VO_{2max}) was observed, as well as oxygen uptake at the ventilation threshold (VO_{2VAT}) in the supplemented group. During the 3rd phase of research these values were significantly smaller in the control group in comparison to the supplemented one (Table 1). Maximal oxygen uptake increased significantly (p < 0.05) by 5.3% after 6 days (2nd phase) of sodium phosphate intake. Continuation of supplementation for 3 weeks (25mg·kgFFM⁻¹·d⁻¹) in group S did not cause further changes in VO_{2max}, but this value was significantly (p < 0.05) higher (5.6%) in comparison to baseline levels.

Similar changes were observed in values of oxygen uptake, measured at the ventilation threshold (VO_{2VAT}) in the supplemented group. During the second phase of the experiment, VO_{2VAT} significantly (p < 0.05) increased by 8.3% in group S. However, in the third phase of the experiment there was a non-significant decrease in this variable. Additionally, a significant (p < 0.05) increase, by 5.1% in maximal minute ventilation (VE_{max}) was observed after 6 days of phosphate loading. The changes in

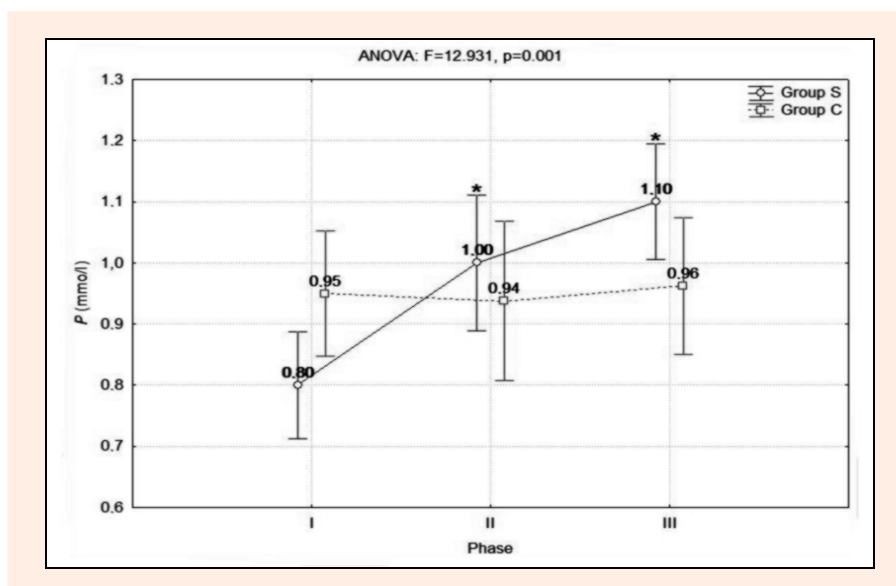


Figure 1. Changes in the serum concentration of non-organic phosphates (P) in the supplemented (S) and control (C) group; * significantly different from 1st phase ($p < 0.05$).

VE_{max} , after the next 3 weeks of supplementation (25mg/kgFFM/d), were significantly ($p < 0.05$) higher (7%) compared to baseline values.

Also a significant ($p < 0.05$) decrease in resting (9.6%) and maximal exercise heart rates (2.7%) occurred after 6 days of supplementation. This was also true for heart rates measured at the lactate threshold (HR_{LT}), which decreased by 1.7%. The changes in HR_{rest} , HR_{LT} and HR_{max} in the third phase of the experiment were non-significant, in comparison to the previous phase of the experiment (Table 1).

A significant ($p < 0.05$) increase in VO_{2max} and a ($p < 0.05$) decrease in HR_{max} caused a significant ($p < 0.05$) increase in oxygen pulse (O_2/HR) after 6 days of trisodium phosphate intake.

The general intake of sodium phosphate in the experiment (6 days in a dose of $50mg \cdot kgFFM^{-1} \cdot d^{-1}$ and 3 weeks in a dose of $25mg \cdot kgFFM^{-1} \cdot d^{-1}$) allowed for a sig-

nificant ($p < 0.05$) increase in maximal power (P_{max}) and an increase in the ventilation threshold (VAT) towards a higher load in the third phase of the experiment.

Also, a significant ($p < 0.05$) increase in the serum concentration of non-organic phosphates (P) was observed (Figure 1), which was accompanied by a decrease in serum calcium (Ca) concentration in the third phase of research in group S (Figure 2). Additionally, the changes in the resting and post-exercise concentration of 2,3-DPG were insignificant in both the S and C group, yet the supplementation procedure showed a tendency for increased values of this variable (Figure 3), while the delta (Δ) values of 2,3-DPG in the S group decreased (Figure 4). The acid-base balance, as well as post-exercise lactate values, were not affected by sodium phosphate supplementation (Table 1). The values of chosen physiological and biochemical variables obtained, as well as the significance of differences between particular phases of the

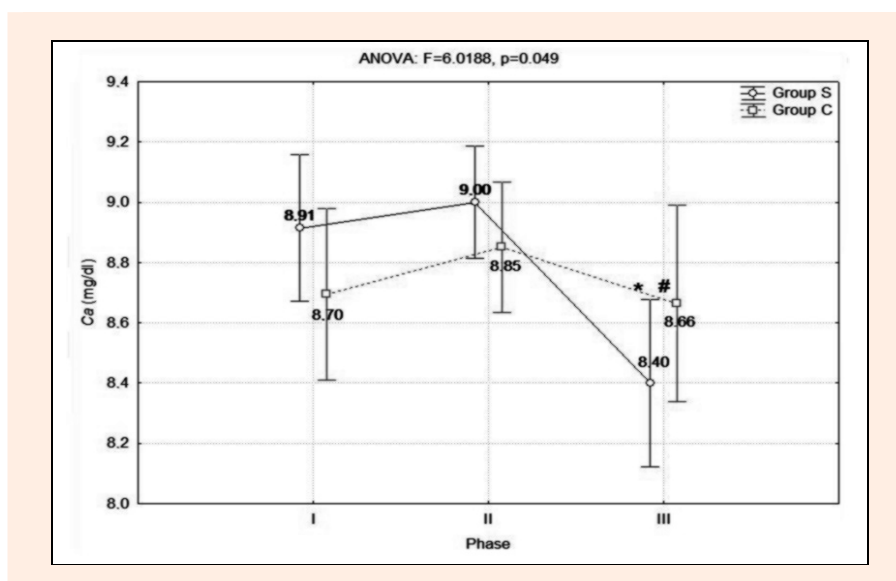


Figure 2. Changes in the serum concentration of calcium (Ca) in the supplemented (S) and control (C) group. * significantly different from 1st phase ($p < 0.05$); # significantly different from 2nd phase ($p < 0.05$).

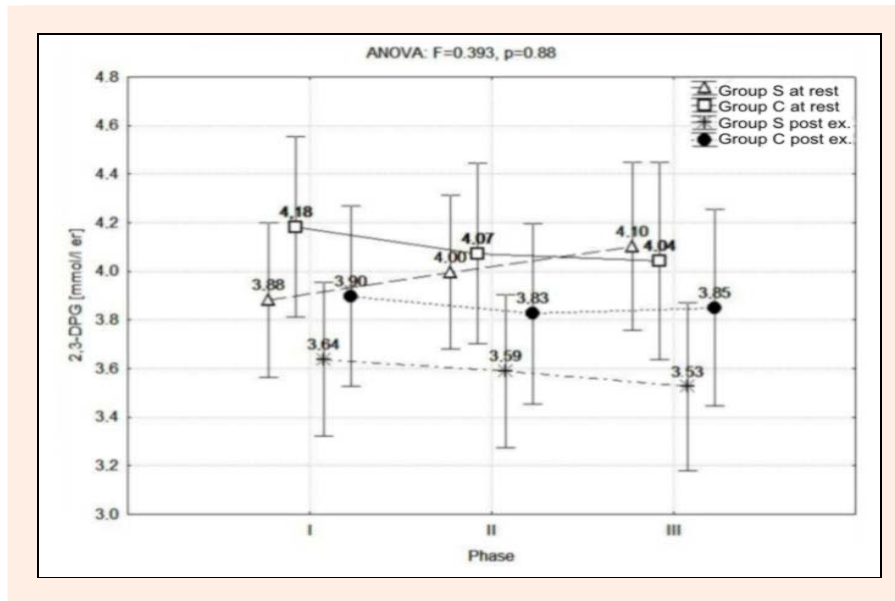


Figure 3. Changes in the resting and post-exercise level of 2,3-DPG in the supplemented (S) and control (C) groups.

experiment in the supplemented and control groups are presented in Table 1.

Correlation coefficients between analyzed variables and the concentration of 2,3-DPG at rest and plasma P concentration in the tested athletes are presented in Table 2.

The results indicate a significant correlation ($r = 0.47$; $p < 0.01$) between the level of 2,3-DPG at rest (2,3-DPG_{rest}) and VO_{2max} , and also between 2,3-DPG_{rest} and VE_{max} ($r = 0.59$; $p < 0.001$). Positively significant correlations between plasma P concentration and VO_{2max} ($r = 0.51$; $p < 0.01$), and between plasma P concentration and O_2/HR ($r = 0.64$; $p < 0.001$) were also observed. There was also a negative significant correlation ($r = -0.42$; $p < 0.01$) between plasma P concentration and HR_{max} , as well as between plasma P concentration and plasma Ca concentration ($r = -0.43$; $p < 0.01$).

Discussion

Although research findings are inconclusive, several well-

controlled studies support the theory that phosphate salt supplementation may enhance functional capacity of the aerobic energy system. The results of these studies indicate that the improvement in aerobic metabolism is caused by an increase in erythrocyte 2,3-DPG, which decreases the affinity of hemoglobin for oxygen, what facilitates the release of oxygen to muscle tissue during exercise (Cade et al., 1979; 1984; Farber et al., 1984;1987; Gibby et al., 1978). Other authors that conducted similar research did not show any changes in this metabolite after phosphate salt intake (Bredle et al., 1988; Kreider et al., 1990).

Most of the current research evaluating the ergogenic effects of phosphate salts refers to the early experiments of Cade et al. (1984). They reported a significant ($p < 0.05$) increase in the concentration of erythrocyte 2,3-DPG (13.00 vs. 13.92 $mg \cdot g^{-1} Hb^{-1}$) in a group supplemented with phosphate salts. Additionally, a 6 to 12% increase in VO_{2max} was observed for subjects given phosphate salts. Similar results were reported by Stewart et al. (1990), who evaluated the effects of sodium

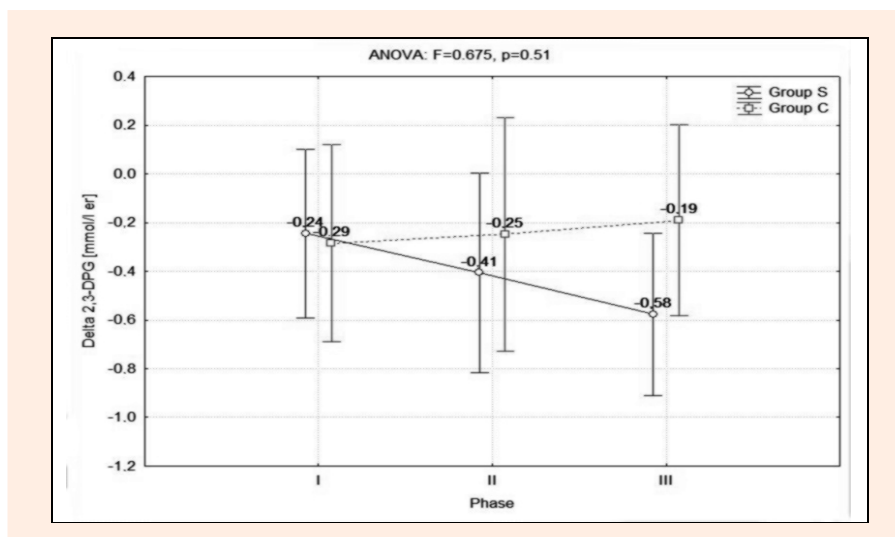


Figure 4. Changes in values of delta (Δ) 2,3-DPG in the supplemented (S) and control (C) groups.

phosphate intake on $\text{VO}_{2\text{max}}$, time to volitional exhaustion, the concentration of 2,3-DPG, and serum inorganic phosphate concentration in 8 well-trained cyclists. The experimental procedure in this study included sodium phosphate intake of $3.6 \text{ g}\cdot\text{day}^{-1}$ or a placebo over a 3-day period. After the supplementation protocol, the exercise tests were repeated, and a 7-day rest period was incorporated. Following the 7-day rest period, the entire procedure was performed once again.

The obtained results showed insignificant changes in resting 2,3-DPG concentration, yet the post-exercise 2,3-DPG values were significantly ($p < 0.05$) higher in the group supplemented with sodium phosphate. Additionally, a significant ($p < 0.01$) increase in $\text{VO}_{2\text{max}}$ was registered in the subjects that were given phosphate salts.

A similar experiment was conducted by Kreider et al. (1990), where the effects of phosphate salt intake on $\text{VO}_{2\text{max}}$, VO_2 at the ventilation threshold, and the 5-mile run time were evaluated. The results of this experiment showed a 9% increase in $\text{VO}_{2\text{max}}$ (73.9 ± 5.0 vs. $80.3 \pm 4.0 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$) and a 12% improvement in $\text{VO}_{2\text{VAT}}$ (58.0 ± 4.0 vs. $64.8 \pm 2.0 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$) in subjects supplemented with sodium phosphate. The concentration of 2,3-DPG was not considered in this research.

On the contrary, research conducted by Bredle et al. (1988) showed no changes in 2, 3-DPG and $\text{VO}_{2\text{max}}$ in a group of athletes supplemented with phosphate salts for 4 days, with a dose of $5.7 \text{ g}\cdot\text{day}^{-1}$. Brennan et al. (2001) documented similar findings to the Bredle study in a group of well-trained cyclists ($\text{VO}_{2\text{max}} = 60.6 \pm 4.4 \text{ ml}\cdot\text{kg}^{-1}\cdot\text{min}^{-1}$), who were supplemented with sodium diphosphate ($4 \text{ g}\cdot\text{day}^{-1}$).

The results of our study are in accordance to those obtained by Cade et al. (1984), Stewart et al. (1990) and Kreider et al. (1990). The experiment showed a significant ($p < 0.05$) increase in $\text{VO}_{2\text{max}}$ following sodium triphosphate intake for 6 days, with a dose of $50\text{mg}\cdot\text{kgFFM}^{-1}\cdot\text{d}^{-1}$. Significant ($p < 0.05$) changes in $\text{VO}_{2\text{max}}$ were registered for both absolute and relative values. Further supplementation with phosphate salts, with a dose of $25\text{mg}\cdot\text{kgFFM}^{-1}\cdot\text{d}^{-1}$, over a period of 21 days, did not increase the level of aerobic power, yet in comparison to baseline level, the changes in absolute and relative values of $\text{VO}_{2\text{max}}$ were significant, respectively.

Our research confirms that changes in $\text{VO}_{2\text{max}}$ obtained in a short-term supplementation procedure can be maintained for a longer period of time by continued intake of phosphate salts in smaller doses. This protocol also increased the ventilation threshold. $\text{VO}_{2\text{max}}$ decreased in the 3rd phase of the research by 1.4%, in comparison to the second phase of the experiment, yet these values were significantly higher in relation to baseline values.

A significant improvement in $\text{VO}_{2\text{VAT}}$ in the group supplemented with phosphate salts caused a shift in VAT towards much higher loads. In the 2nd and 3rd phases of the experiment, a 5.4% increase in P_{VAT} in comparison to baseline values was registered (280.4 vs. 295W). The intake of sodium triphosphate caused a delay in the drastic increase of carbon dioxide concentration in the blood ($p\text{CO}_2$), stimulating respiration. The delay in hyperventilation, aimed at the removal of ex-

cess CO_2 from the body, indicates a better supply of oxygen to muscle tissues in the supplemented group.

One of the indexes of tissue oxygen saturation includes oxygen pressure ($p\text{O}_2$) in capillarized blood. According to Dempsey et al. (1971), an increase in erythrocyte 2,3-DPG is accompanied by a simultaneous rise in capillary $p\text{O}_2$. The changes in rest and post-exercise values of capillary $p\text{O}_2$ were insignificant in the group that was given sodium phosphate, yet a tendency for an increase in this variable, due to supplementation, was observed. A similar tendency was registered in the resting concentration of 2,3-DPG in group S.

The statistical analysis showed a significant relationship between the resting concentration of 2,3-DPG ($2,3\text{-DPG}_{\text{rest}}$) and $\text{VO}_{2\text{max}}$. There were no significant changes in post-exercise concentration of 2,3-DPG ($2,3\text{-DPG}_{\text{max}}$) and values of delta (Δ) 2,3-DPG in group S, yet a slight decrease in these variables occurred in the second and third phases of the experiment. The decrease in these variables could have been caused by a significant ($p < 0.05$) increase in peak power output (P_{max}) in group S. After 6 days of supplementation ($50\text{mg}\cdot\text{kgFFM}^{-1}\cdot\text{d}^{-1}$), a insignificant rise in P_{max} occurred, yet the continued intake of sodium phosphate for 3 weeks ($25\text{mg}\cdot\text{kgFFM}^{-1}\cdot\text{d}^{-1}$) caused a significant ($p < 0.05$) increase in this variable. An increase in P_{max} caused a drop in post-exercise pH, which may have influenced the concentration of 2,3-DPG ($2,3\text{-DPG}_{\text{post}}$). According to Bard and Teasdale (1979), a decrease in blood pH by 0.010 units causes a simultaneous (4%) drop in 2,3-DPG concentration.

Additionally, the level of erythrocyte 2,3-DPG can be modified by the serum concentration of inorganic phosphates (P). This is confirmed in our research by the significant relationship between the concentration of inorganic phosphates (P) in blood serum and the level of $2,3\text{-DPG}_{\text{rest}}$ ($r = 0.49$; $p = 0.01$). In case of hypophosphatemia, a drop in the concentration of 2,3-DPG occurs, while under conditions of hyperphosphatemia, the opposite takes place (Lichtman et al. 1971; Card and Brain, 1973). Not all research conducted with phosphate loading confirm this relationship. Cade et al. (1984), after 3 days of supplementation with phosphate salts, observed a significant ($p < 0.05$) increase in the resting level of serum phosphates, as well as a rise in the concentration of 2,3-DPG. In a similar experiment, Kreider et al. (1990) also registered a significant increase (17%) in resting concentration of blood serum inorganic phosphates after supplementation, yet changes in 2,3-DPG were not analyzed. Bredle et al. (1988) also showed a significant increase (35%) in the concentration of blood serum phosphates after 4 days of supplementation with calcium phosphate, however, they did not show significant changes in 2,3-DPG, P_{50} , pH and $\text{VO}_{2\text{max}}$. In a research project conducted by Mannix et al. (1990), with a single intake of calcium phosphate, a significant increase in the concentration of serum phosphates (13%) and 2,3-DPG (11%) occurred, yet no changes in $\text{VO}_{2\text{max}}$ or heart muscle work capacity were registered. In a more recent experiment, Bremner et al. (2002) showed significant relationships between the concentration of inorganic phosphates in the blood and erythrocyte phosphate level, as well as the erythrocyte

concentration of phosphates and the level of 2,3-DPG. No relationship was observed between the concentration of blood serum phosphates and erythrocyte 2,3-DPG level.

The applied 6-day supplementation protocol in our research caused a significant (30%) increase in the concentration of phosphates in blood serum, as well as a 25% rise in erythrocyte 2,3-DPG. The authors suggest that the increase in 2,3-DPG is most likely the effect of increased concentration of erythrocyte phosphates.

In our research project a continuous rise in serum inorganic phosphate (P) concentration was observed in the group supplemented with sodium phosphate. During the second phase of research, a significant blood serum ($p < 0.05$) increase in inorganic phosphates (P) occurred (0.8 ± 0.16 vs. 1.0 ± 0.22 $\text{mmol}\cdot\text{l}^{-1}$). Continued intake of sodium phosphate in the third phase of the experiment caused a further increase in this variable, yet it was insignificant in comparison to the previous phase, however it was significant ($p < 0.05$) in relation to initial values. It must be indicated that the initial concentration of serum inorganic phosphates (P) in group S equaled 0.8 ± 0.16 $\text{mmol}\cdot\text{l}^{-1}$, which indicates a state of hypophosphatemia, which occurs when serum P concentration falls below 0.9 $\text{mmol}\cdot\text{l}^{-1}$. In athletes, such a state is most often caused by incomplete recovery from training and competition, or dietary phosphate deficiency. In the control group, the concentration of P was in the lower range of daily allowance and equaled (0.95 ± 0.09 $\text{mmol}\cdot\text{l}^{-1}$). On the other hand, a significant relationship detected between the serum concentration of inorganic phosphates and $\text{VO}_{2\text{max}}$, as well as P and $\text{VO}_{2\text{VAT}}$, indicates that the effectiveness of phosphate loading depends on the initial concentration of P in the blood.

The available data regarding ergogenic benefits of phosphate salts are predominantly related to short-term supplementation, lasting from 3 to 6 days. The majority of these projects were based on the assumption presented by Cade et al. (1984), who suggested that longer supplementation protocols are not justified, since continued intake of phosphate salts does not further increase the level of 2,3-DPG, nor does it change $\text{VO}_{2\text{max}}$. This phenomenon could be explained by the hormonal regulation of blood serum concentration of inorganic phosphates. A key role is played here by the parathyroid hormone (PTH), which increases the elimination of phosphates through the kidneys. Long-term intake of phosphate salts causes an increased secretion of parathyroid hormone, which increases the elimination of phosphates through urine (Chase and Aurbach, 1968). Our research suggests that the time of phosphate supplementation should consider the initial concentration of blood serum inorganic phosphates and changes in this variable throughout the supplementation protocol.

Calvo (1988) conducted an experiment in which he analyzed the influence of a 1g dose of phosphate salts on the concentration of blood inorganic phosphate and calcium, as well as PTH concentration. The results indicated a significant ($p < 0.05$) increase in the concentration of phosphates, and no changes in the level of calcium and PTH.

Silverberg et al. (1986) also showed a significant increase in the concentration of blood inorganic phos-

phates, and a lack of change in the level of serum calcium and PTH, 1 hour after a single intake of 1g of sodium phosphate. Continued intake of phosphate salts for 5 days, in a dose of 2g/d, caused a significant increase in the concentration of PTH.

Most research (Silverberg et al., 1986; Calvo, 1988), thus, confirm that a transition state of increased blood concentration of P does not cause hypocalcaemia, and does not increase the concentration of PTH. On the other hand, prolonged hyperphosphatemia significantly affects the blood concentration of these variables. In our research, group S showed significant changes ($p < 0.05$) in serum concentration of Ca following long-term sodium phosphate intake (Table 1). A lack of significant changes in the concentration of Ca following the first 6 days of supplementation was most likely the effect of a low blood serum P concentration.

After the 6-day supplementation protocol with trisodium phosphate, a significant increase in VE_{max} was registered ($p < 0.05$). This variable continued to increase during the next 3 weeks of supplementation, yet the changes were statistically insignificant. When compared to baseline values, the changes in VE_{max} , after the long-term phosphate salt intake were statistically significant ($p < 0.05$). The increase in VE_{max} in group S may be explained by improved function of the diaphragm. This assumption can be partially confirmed by the research of Aubier et al. (1985), where the effects of hypophosphatemia on the function of the diaphragm in patients ($n = 8$) with severe respiratory inefficiency were analyzed. A high relationship ($r = 0.73$) between blood concentration of phosphates and transdiaphragmatic pressure was observed. These results indicate that hypophosphatemia impairs the function of the diaphragm.

Several authors indicate an ergogenic effect of phosphate salt intake on heart efficiency at rest, as well as during exercise. This hypothesis is based on the fact that hypophosphatemia decreases stroke volume (Fuller et al., 1978; O'Connor et al., 1977; Rubin and Naris, 1990). O'Connor et al. (1977) suggest that the increased contractibility of the heart muscle is caused by increased concentration of cell ATP, which is low during hypophosphatemia. Animal research confirmed the data on improved heart work capacity following phosphate salt intake (Darsee et al., 1978; Stoff, 1982). Several other research projects, which used sodium or calcium phosphate intake, showed a significant decrease of cardiac output and stroke volume during exercise of moderate intensity (Farber et al. 1984; Lunne et al. 1990; Moore et al. 1981), and significant improvements in these variables during endurance exercise with maximal intensity (Kreider, 1992). Bredle et al. (1988) indicated a significant ($p < 0.05$) increase in serum inorganic phosphate concentration and heart function, following 4 days of supplementation with 176 $\text{mmol}\cdot\text{day}^{-1}$ of calcium phosphate. A significant ($p < 0.05$) decrease in cardiac output was registered during an endurance exercise protocol, conducted at 70% of $\text{VO}_{2\text{max}}$. There were no changes in 2,3-DPG and $\text{VO}_{2\text{max}}$ values, yet a significant ($p < 0.05$) increase was observed in arteriovenous oxygen difference, which suggests a better supply of oxygen to the tissues. This data indicates that phosphate salt intake

may improve the function of the cardio-respiratory system.

The analysis of heart rate (HR) in our research indicated significant changes in resting and exercising heart rates in the group of cyclists supplemented with sodium phosphate. The changes in exercise heart rate may be explained by increased stroke volume and improved contractibility of the heart muscle. These assumptions can be confirmed by Kreider et al. (1992), who showed that sodium phosphate intake significantly improves the functioning of the heart muscle. Echocardiographic evaluations in a group of cyclists supplemented with phosphate salts, indicated a significant (4%) increase in stroke volume during this period of time.

The analysis of results in group S also showed an improvement in oxygen pulse (O_2/HR), which is a non-invasive index of evaluating work capacity of the cardio-respiratory system; and simultaneously, a good indicator of physical fitness in endurance sport disciplines.

Other than improving the supply of oxygen to the tissues, phosphate salt intake may improve the acid-base balance during intensive exercise. Phosphates are very active in buffering processes and participate in the acid-base balance of blood plasma, as well as inside the muscle cells. The buffering capacity of phosphates is rather low in the extracellular fluids, yet they play a significant role in the intracellular fluids, where the concentration of phosphates is much higher (Avioli, 1988). Some authors suggest that the intake of sodium phosphate may increase the buffering capacity of muscle cells, and may increase work capacity during exercise of high intensity (Cade et al., 1984; Kreider, 1992; Miller et al., 1991). For example, Cade et al. (1984) showed that phosphate salt intake lowered lactate concentration during exercise of submaximal intensity.

Other research projects indicated a shift of lactate threshold towards higher loads (Kreider et al. 1990; 1992; Miller et al. 1991). Similar results were presented by Stewart et al. (1990), who showed a minor but significant decrease in ($p < 0.05$) post-exercise lactate concentration after an endurance exercise protocol, following 3 days of sodium phosphate intake.

The supplementation protocol applied in our research did not confirm the buffering properties of phosphate salts. The analysis of resting and post-exercise lactate concentrations, and the level of LT, showed no significant changes in these variables due to supplementation. There were also no significant changes in the acid-base variables in the S group. The only significant ($p < 0.05$) changes occurred in the resting values of base excess (BE_{rest}) and in the extracellular fluids ($BE_{ecf,rest}$) during the third phase of the experiment.

Conclusion

The most important findings of this work include a significant increase in VO_{2max} , VE_{max} and O_2/HR , due to sodium phosphate intake over 6 days. Additionally, a significant decrease in resting and maximal heart rate (HR) occurred, as well as during each successive incre-

mental load during the progressive exercise protocol. The supplementation procedure caused a significant increase in maximal aerobic power (P_{max}) and a shift of VAT towards higher loads. An increase in the serum concentration of inorganic P was registered, which was accompanied by a decrease in the concentration of blood Ca. During the experiment, no significant changes were noticed in the resting and post-exercise concentration of erythrocyte 2,3-DPG, yet there was a tendency for a slight increase in the resting value of this variable. No changes were registered in the acid-base balance due to phosphate salt intake, as well as in the post-exercise lactate concentration. The results of this research and literature review, allows to conclude that phosphate salt supplementation can be considered a valuable ergogenic aid in endurance sport disciplines.

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Key points

- Studies on bone acute biochemical response to loading have yielded unequivocal results.
- There is a paucity of research on the biochemical bone response to high impact exercise.
- An increase in bone turnover was observed one to two days post exercise.

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