Effects of Phosphatidylserine on Exercise Capacity during Cycling in Active Males

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ABSTRACT

KINGSLEY, M. I., M. MILLER, L. P. KILDUFF, J. MCENENY, and D. BENTON. Effects of Phosphatidylserine on Exercise Capacity during Cycling in Active Males. *Med. Sci. Sports Exerc.*, Vol. 38, No. 1, pp. 64–71, 2006. **Purpose:** The purpose of the study was to investigate the effects of 750 mg of soybean-derived phosphatidylserine, administered daily for 10 d, on exercise capacity, oxygen uptake kinetic response, neuroendocrine function, and feeling states during exhaustive intermittent exercise. **Methods:** Following preliminary testing, fourteen active males completed a staged intermittent exercise protocol on two further occasions (T1 and T2) separated by 16 ± 1 d. The protocol consisted of three 10-min stages of cycling at 45, 55, and 65% $\dot{V}O_{2max}$, followed by a final bout at 85% $\dot{V}O_{2max}$ that was continued until exhaustion. Approximately 5 d after T1 the subjects were assigned, in a double-blind manner, to either phosphatidylserine (PS) or placebo (P). Breath-by-breath respiratory data and heart rate were continually recorded throughout the exercise protocol, and blood samples were obtained at rest, during the rest periods within the protocol (Post-55, Post-65), at the end of exercise (Post-85), 20 min after the completion of exercise (postexercise), and the day following exercise (Post-24 h). **Results:** The main finding of this study was that supplementation had a significant effect on exercise time to exhaustion at 85% $\dot{V}O_{2max}$ ($P = 0.005$). The exercise time to exhaustion in PS increased following supplementation (7:51 ± 1:36 to 9:51 ± 1:42 min:s, $P = 0.001$), whereas P remained unchanged (8:09 ± 0:54 to 8:02 ± 0:54 min:s, $P = 0.670$). Supplementation did not significantly affect oxygen kinetic mean response times (MRT_{on} and MRT_{off}), serum cortisol concentrations, substrate oxidation, and feeling states during the trial. **Conclusion:** This is the first study to report improved exercise capacity following phosphatidylserine supplementation. These findings suggest that phosphatidylserine might possess potential ergogenic properties. **Key Words:** PHOSPHOLIPIDS, SUPPLEMENTATION, ERGOGENIC AID, ENDURANCE, HUMANS

Phosphatidylserine (PtdSer) is a naturally occurring phospholipid nutrient that appears to be essential to the functioning of all mammalian cells (5). In humans, PtdSer is most concentrated in organs with high metabolic activity, such as brain, heart, skeletal muscle, and liver (8). The biochemical actions of PtdSer include: 1) regulation of calcium ion uptake, 2) regulation of substrate binding (e.g., opiate and glutamate), and 3) the stimulation of specific enzymes activities (e.g., ATPase and acetylcholinesterase) (20). The discovery of these actions has led investigators to examine the pharmacological effects of PtdSer supplementation in humans. Traditionally, PtdSer supplements were derived from bovine cortex (BC-PtdSer); however, this source is now considered unsuitable because of the risk of transferring infectious diseases, and soy-derived PtdSer (S-PtdSer) has emerged as a safe alternative. Although Blokland et al. (5) found no differences between the bioactivity of S-PtdSer and BC-PtdSer in rats, it remains unclear whether the source of the supplement is an important factor in determining the pharmacological actions of PtdSer in humans.

The majority of PtdSer supplementation studies have evaluated higher brain functions; indeed, BC-PtdSer has been shown to improve a variety of cognitive functions that include memory and verbal response in aging patients (2,6), and more recently S-PtdSer has been demonstrated to enhance mood in a subsection of young individuals during mental stress (4). Few studies have investigated the effects of PtdSer supplementation before exercise (10,16,17), and the available data suffers from the methodological limitation of using crossover supplementation designs while the washout kinetics of PtdSer remains unclear (20). Orally administered bovine cortex-derived PtdSer (800 $mg \cdot d^{-1}$) has been reported to attenuate the serum cortisol response to acute exercise stress (17). Additionally, Fahey and Pearl (10) reported that oral supplementation with 800 $mg \cdot d^{-1}$ S-PtdSer was effective in reducing serum cortisol concentration following intensive resistance training. These findings suggest that both BC-PtdSer and S-PtdSer act to partly counteract the stress-induced activation of the hypothalamo–pituitary–adrenal (HPA) axis, although the significance of altering this neuroendocrine response on metabolic function (such as substrate utilization) during exercise has not been evaluated.

Assuming that exogenous PtdSer is effectively incorporated into cardiac and skeletal muscles, the aforementioned biochemical actions of PtdSer have the potential to influence mitochondrial enzymatic activity. Therefore, PtdSer

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TABLE 1. Subject characteristics for phosphatidylserine supplementation group (PS) and placebo group (P).

Characteristic	PS ($N = 7$)	P ($N = 7$)	P value
Age (dec. yr)	23.4 ± 1.9	22.2 ± 1.1	0.638
Mass (kg)	84.9 ± 3.8	87.7 ± 3.2	0.630
Height (m)	1.79 ± 0.01	1.81 ± 0.02	0.528
Maximal oxygen uptake ($mL \cdot kg^{-1} \cdot min^{-1}$)	43.8 ± 2.0	42.4 ± 1.7	0.645

Values are mean ± SEM. P value calculated using independent sample t-test.

supplementation might affect "metabolic inertia," a proposed determinant of the primary oxygen uptake ($\dot{V}O_2$) kinetic response to exercise (12). Consequently, it was hypothesized that PtdSer supplementation could reduce accumulated oxygen deficit and improve exercise tolerance.

Therefore the purpose of the current study was to investigate the efficacy of chronic S-PtdSer supplementation on exercise capacity, neuroendocrine function, oxygen uptake kinetic response, and perceived feeling states during and immediately following intermittent cycling.

METHODS

Subjects. Fourteen healthy male subjects (Table 1) volunteered to participate in this study and completed all the study requirements. All subjects were informed about the potential risks of the study and gave written informed consent for their participation in the study, which was approved by a university ethics committee. In addition, all subjects were informed that they might be asked to consume a supplement rich in S-PtdSer; however, there are currently no reported, or inferred, side effects associated with S-PtdSer supplementation (13,20). The experimental procedures were in accordance with the policy statement of the American College of Sports Medicine. No subject had prior history of cardiovascular or respiratory disease and all subjects were nonsmokers. Potential subjects attended an interview before undertaking the study and were subsequently excluded if they had taken nutritional supplements in the last 8 wk.

Experimental design. Before the main exercise trials, all subjects visited the laboratory on two occasions to complete an incremental exercise test and a familiarization trial. Subjects then performed two main exercise trials, which consisted of staged intermittent cycling, separated by 16.0 ± 1.3 d. Approximately 5 d after completion of the first main exercise trial the subjects were assigned, in a randomized double-blind fashion, to either a PS group or a placebo (P) group, and instructed to take supplements for the 10 d before trial 2. The experimental design is illustrated in Figure 1.

The PS group ingested 750 $mg \cdot d^{-1}$ S-PtdSer, and the P group ingested a weight-matched glucose polymer. The S-PtdSer supplements were manufactured using the method of specific transesterification of soybean lecithin, and then blended with additional soybean lecithin to provide a concentration of 20% PtdSer (Lucas Meyer; Hamburg, Germany). Both supplements were administered in capsules and placed in generic packaging. Subjects were instructed to maintain their normal diet and activity patterns throughout

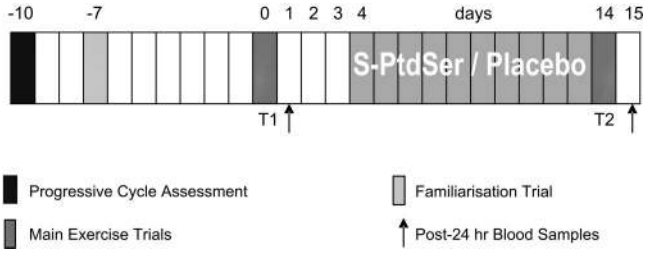

FIGURE 1—Schematic diagram of the experimental design.

the study. Food was weighed and recorded by the subjects for 2 d before each exercise trial and for the day afterwards. Analysis using commercial software (CompEat v5.8.0; Nutrition Systems, UK) revealed that there were no differences in energy intake or dietary composition between supplementation groups or trials. The daily diet comprised of 11.2 ± 0.6 $MJ \cdot d^{-1}$, of which 52 ± 3, 30 ± 1, 16 ± 1, and 2 ± 1% of energy intake was obtained from carbohydrates, fat, protein, and alcohol, respectively. In addition, the subjects were instructed to abstain from strenuous exercise for 3 d before and 2 d following each trial. At the completion of the study all subjects gave their verbal assurance that they had complied with all instructions.

Procedures. During their initial visit to the laboratory the subjects completed an incremental exercise test on an electromagnetically braked cycle ergometer (Lode Excalibur Sport; Lode, Holland). The work rate began at 60 W and thereafter increased in 30-W increments every 3 min until volitional exhaustion. Subjects were instructed to pedal at a constant cadence between 75 and 85 rpm. Heart rate (Polar S810; Polar Electro Oy, Finland), and breath-by-breath respiratory parameters (Oxycon Pro; Jaeger, Germany) were simultaneously recorded. The test was used to identify maximal oxygen uptake ($\dot{V}O_{2max}$) and also to calculate, using linear regression, the work rates required for the staged intermittent exercise protocol.

The subjects then completed a staged intermittent exercise protocol on three separate occasions. The intermittent protocol required subjects to complete three 10-min stages of cycling at work rates calculated to elicit 45, 55, and 65% $\dot{V}O_{2max}$, followed by a final exercise bout at 85% $\dot{V}O_{2max}$ that was continued until exhaustion (an inability to maintain a cadence of 60 rpm despite verbal encouragement). All exercise stages were interspaced with 5-min passive rest periods. The first exercise trial served to familiarize the subjects with the intermittent exercise protocol, and the subjects remained blind to their exercise times to exhaustion throughout the study to prevent an anticipated trial–order effect. The subsequent main exercise trials (T1 and T2) were completed 14–21 d apart. Breath-by-breath respiratory data (Oxycon Pro; Jaeger, Germany) and heart rate (Polar S810; Polar Electro Oy, Finland) were monitored throughout T1 and T2. Exercise-induced feeling inventory (EFI (11)) was completed at Preexercise and following each exercise stage. Blood samples were taken by venepuncture (Vacutainer system; Becton-Dickinson Ltd, UK) from an antecubital vein at Preexercise, on completion of the 55% $\dot{V}O_{2max}$

(Post-55%) and 65% $\dot{V}O_{2max}$ (Post-65%) exercise stages, 20 min after the completion of the trial (postexercise), and 24 h after the trial (Post-24 h). An additional capillary blood sample was taken immediately after exhaustion (Post-85%) for the analysis of blood lactate and glucose concentrations.

Analysis of oxygen uptake and heart rate. Breath-by-breath oxygen uptake, carbon dioxide production, and 5-s heart rate data were averaged for the last minute of exercise for all exercise intensities. Subsequently nonprotein respiratory exchange ratios were used to calculate the rates of carbohydrate and fat oxidation for Preexercise and during each exercise stage up to and including 65% $\dot{V}O_{2max}$ as previously described (29), using equations derived by Peronnet and Massicotte (21). Linear regression was applied to the final 3 min of oxygen uptake data for each of these stages, and the 95% confidence interval for the slope was inspected; steady state was assumed if the 95% confidence interval included zero.

In addition, breath-by-breath oxygen uptake data were initially edited to remove occasional errant breaths (from coughs, sighs, or swallows) when values were greater than four standard deviations from the local mean (22). The data for all exercise stages and rest periods were interpolated, using a cubic spline, in 1-s intervals. The data for moderate-intensity exercise (45–65% $\dot{V}O_{2max}$) and subsequent rest periods during each trial were normalized (normalized to the respective last-minute $\dot{V}O_2$ values), time-aligned, and averaged. The data for the final bout (85% $\dot{V}O_{2max}$) were modeled separately, as the oxygen uptake response to different intensity domains (below and above lactate threshold) have been suggested to differ (19). The traditionally defined phase II oxygen uptake on-kinetic response (30) was determined for 45–65% $\dot{V}O_{2max}$ and 85% $\dot{V}O_{2max}$. The end of phase I was set to 20 s after the onset of constant-load exercise, and a monoexponential function (Equation 1) was fitted between the end of phase I and 3 min of exercise (23) using iterative nonlinear regression techniques (SPSS version 12.0; SPSS Inc., IL). Similarly, the off-kinetic response to each exercise stage was determined using a monoexponential function (Equation 2) after omission of the first 20 s of postexercise data

$$\dot{V}O_2(t) = \dot{V}O_2(B) + G \cdot (1 - e^{-(t-TD)/\tau}) \quad [1]$$

$$\dot{V}O_2(t) = \dot{V}O_2(EE) + G \cdot (e^{-(t-TD')/\tau'}) \quad [2]$$

where $\dot{V}O_2(t)$ is oxygen uptake at time t, $\dot{V}O_2(B)$ is baseline oxygen uptake (average $\dot{V}O_2$ during the minute before the onset of exercise), $\dot{V}O_2$ (EE) is end of exercise oxygen uptake (average $\dot{V}O_2$ during the last minute of exercise), G is the primary gain (i.e., the calculated change in oxygen uptake), TD is the on-kinetic time delay, and τ is the time constant (prime mark designates off-kinetics parameters). Mean response times for the on-kinetic responses (MRT_{on}) and off-kinetic responses (MRT_{off}) were calculated as TD + τ and TD′ + τ′, respectively.

Blood sampling and analysis. Venous blood was collected in a 5-mL container (Becton-Dickinson Ltd, UK) containing the anticoagulant ethylenediaminetetra-acid (EDTA). Several small aliquots were removed for the triplicate determination of blood lactate and glucose concentration (YSI 2300 Stat, Yellow Springs Instruments, U.S.), blood hemoglobin (Hb) concentration (Hemocue Ltd, UK), hematocrit (Hct) (Micro Hct MK IV, Hawksley, England) and changes in plasma volume as previously described (9). The remaining blood was centrifuged at 3000 × *g* for 15 min to obtain plasma, which was subsequently dispensed and frozen at −70°C. Two additional 7-mL blood samples were collected in serum separation tubes (Becton-Dickinson Ltd, UK), left to stand for 15 min, then centrifuged at 3000 × *g* for 15 min to obtain serum. The serum was transferred to appropriate containers and subsequently frozen at −70°C. Serum cortisol concentrations were determined using an automated time-resolved fluoroimmunoassay (AutoDELFIA™ Cortisol kit, Perkin Elmer, Life Sciences, UK).

Perceived feeling states. Subjects were instructed to respond to the EFI as described in detail by Gauvin and Rejeski (11). Briefly, the subjects rated their feelings using the 12-item adjective scale on an analog scale from 0 (do not feel) to 5 (feel very strongly). The appropriate adjectives were averaged to obtain four perceived feeling states (positive engagement, revitalisation, tranquillity, and physical exhaustion) at each time point as previously described (11). The EFI was specifically developed to assess distinct feeling states that occur during stages of exercise and psychometric studies have indicated concurrent and discriminant validity (11). Moreover, the EFI has been demonstrated to be sensitive to interventions involving recreational active individuals (15).

Statistical analysis. Statistical analysis was carried out using SPSS software (version 12.0; SPSS Inc., IL). Group data were expressed as mean ± SEM and statistical significance was set at the $P < 0.05$ level. Subject characteristics were compared under supplementation groups using independent samples *t*-tests (Table 1). The exercise times to exhaustion during familiarisation, T1 and T2, were assessed using mixed-model repeated measures ANOVA (within-subject factors: trials; between-subject factor: supplementation groups) followed by simple main effect analysis. The remaining data, which contained multiple time points during each trial, were analyzed using mixed-model repeated measures ANOVA (within-subject factors: trial × time of sample; between-subject factor: supplementation groups). Mauchly's test was consulted and Greenhouse–Geisser correction was applied if the assumption of sphericity was violated. If a significant *P* value was identified for the three-way interaction effect (supplementation group × trial × time of sample), supplementation was deemed to have a significant effect. If a significant *P* value was identified for the main effect of time (time of sample), multiple pairwise comparisons were made using Bonferonni confidence interval adjustment, with statistical significance set at $P < 0.01$.

RESULTS

Exercise times to exhaustion at 85% $\dot{V}O_{2max}$ were similar between groups during familiarization and T1 (PS, P: 8:02

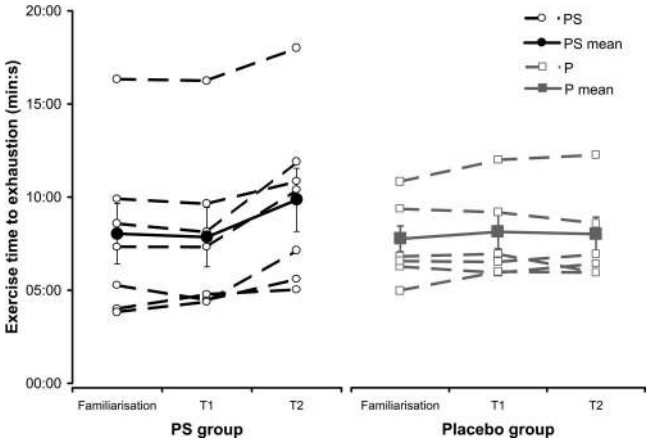

FIGURE 2—Exercise times to exhaustion at 85% $\dot{V}O_{2max}$ completed at the end of each main exercise trial. Individual data are presented as open shapes and mean data are presented as filled shapes. PS, phosphatidylserine group; P, placebo group. (supplementation group × trial interaction, $P = 0.007$).

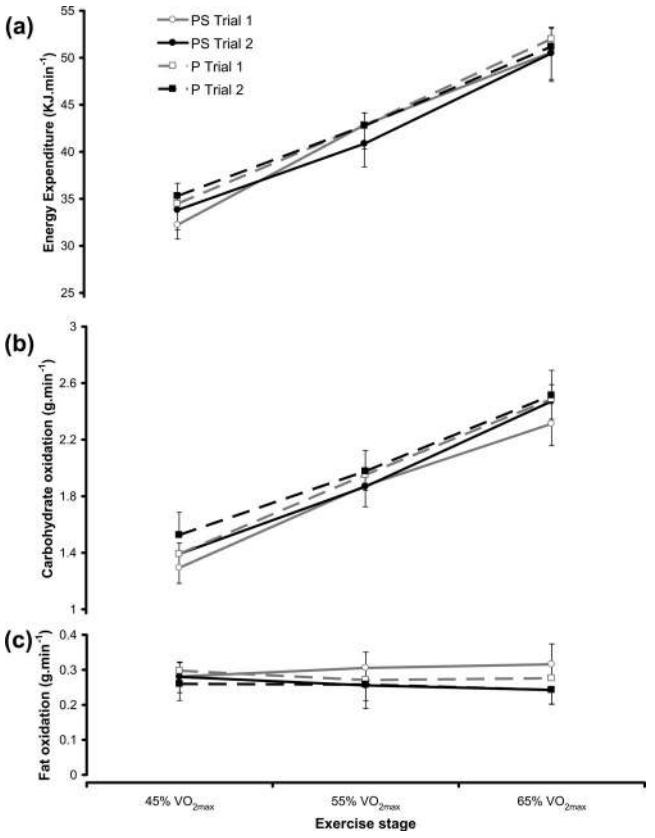

FIGURE 3—(a) Energy expenditure, (b) carbohydrate oxidation rates, and (c) fat oxidation rates during each main exercise trial. Values represent mean ± SEM ($N = 7$). PS, phosphatidylserine group; P, placebo group. Trial 1: presupplementation; Trial 2: postsupplementation.

± 1:37, 7:46 ± 0:41; 7:51 ± 1:36, 8:09 ± 0:54 min:s). A significant interaction effect (supplementation group × trial, $P = 0.007$) indicated that supplementation had a significant effect on time to exhaustion; *post hoc* analysis revealed no differences between trials for P ($P = 0.670$) and significant differences between trials for PS ($P = 0.001$). The magnitude of change in exercise times to exhaustion (individual T2 values minus T1 values) in PS were 2:00 ± 0:28 min:s, whereas P remained similar (0:07 ± 0:13 min:s) (Fig. 2).

Supplementation did not significant effect last minute heart rates during the protocol (supplementation group × trial × time of sample, $P = 0.058$). Mean last-minute heart rates increased throughout the stages of the protocol (Table 2), being 119 ± 2, 137 ± 3, 156 ± 3, and 183 ± 2 beats·min^{-1}, respectively at 45, 55, 65, and 85% $\dot{V}O_{2max}$.

Last-minute oxygen uptake data are presented in Table 2. These data increased progressively with exercise intensity (47 ± 1, 58 ± 1, 70 ± 1, and 97 ± 2% $\dot{V}O_{2max}$; time of sample effect, $P < 0.001$) and the supplement × trial × time of sample interaction was not significant ($P = 0.160$). Steady state was confirmed in all stages at exercise intensities up to and including 65% $\dot{V}O_{2max}$. Mean carbohydrate oxidation, fat oxidation, and total energy expenditure were 1.40 ± 0.06, 1.95 ± 0.11, and 2.49 ± 0.14 g·min^{-1}; 0.28 ± 0.02, 0.27 ± 0.02, and 0.27 ± 0.02 g·min^{-1}; and 34.0 ± 0.8, 42.3 ± 1.0, and 51.1 ± 1.1 kJ·min^{-1}, respectively at each exercise stage (Fig. 3). Supplementation had no effect on carbohydrate oxidation (supplementation group × trial × time of sample, $P = 0.596$), fat oxidation (supplementation group × trial × time of sample, $P = 0.187$), or calculated energy expenditure (supplementation group × trial × time of sample, $P = 0.595$).

The mean response times for the on-kinetic response (MRT_{on}) were significantly higher at 85% $\dot{V}O_{2max}$ than at 45–65% $\dot{V}O_{2max}$ (time of sample effect, $P = 0.019$); however, the three-way interaction was not significant ($P = 0.069$) (Fig. 4). Supplementation had no effect on MRT_{off} (supplementation group × trial × time of sample, $P = 0.449$), and exercise intensity had no significant effect (time of sample effect, $P = 0.055$) (Fig. 4).

Estimated plasma volume fell by 7–11% during the first two exercise stages and remained below preexercise values throughout exercise (Table 3); supplementation had no effect (supplementation group × trial × time of sample, $P =$

TABLE 2. Last minute heart rate and oxygen uptake during both exercise trials for phosphatidylserine supplementation group and placebo group.

		Trial 1 45% $\dot{V}O_{2max}$	Trial 1 55% $\dot{V}O_{2max}$	Trial 1 65% $\dot{V}O_{2max}$	Trial 1 85% $\dot{V}O_{2max}$	Trial 2 45% $\dot{V}O_{2max}$	Trial 2 55% $\dot{V}O_{2max}$	Trial 2 65% $\dot{V}O_{2max}$	Trial 2 85% $\dot{V}O_{2max}$	Effect: *P* interaction, *timing*
Heart rate	PS	120 ± 4	136 ± 4	153 ± 3	178 ± 2	116 ± 5	132 ± 5	152 ± 6	183 ± 4	0.081
(beats·min^{-1})	P	120 ± 6	140 ± 6	158 ± 6	185 ± 4	121 ± 4	139 ± 6	160 ± 5	185 ± 3	*<0.001*
Oxygen uptake	PS	1.73 ± 0.10	2.26 ± 0.16	2.59 ± 0.15	3.54 ± 0.15	1.68 ± 0.09	2.07 ± 0.13	2.51 ± 0.13	3.62 ± 0.22	0.160
(L·min^{-1})	P	1.76 ± 0.05	2.16 ± 0.07	2.65 ± 0.07	3.59 ± 0.07	1.76 ± 0.06	2.15 ± 0.07	2.60 ± 0.08	3.45 ± 0.12	*<0.001*

Values are last minute values; presented as mean ± SEM ($N = 7$). PS, phosphatidylserine group; P, placebo group.
Effects present *P* values for supplementation group × trial × timing of sample interaction and timing.

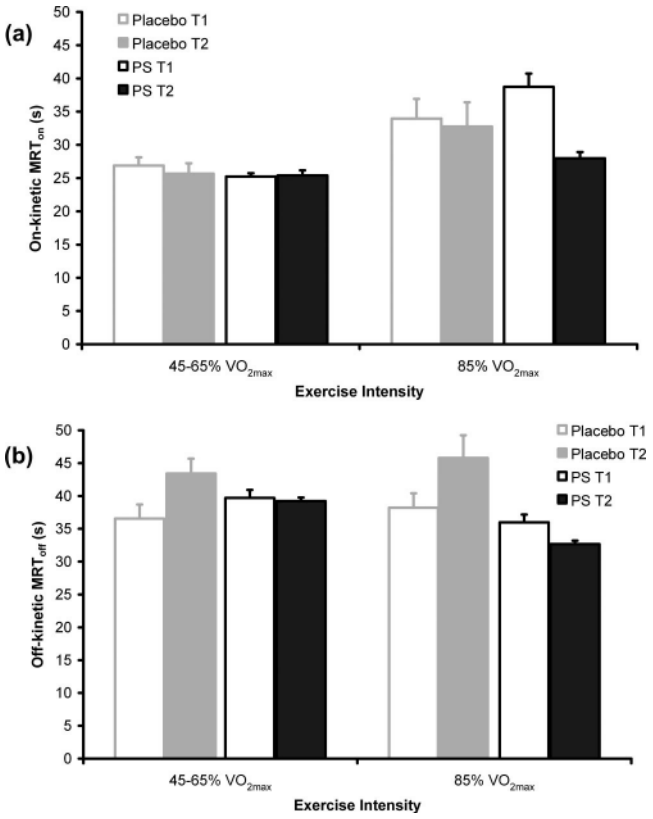

FIGURE 4—(a) On-kinetic mean response times, and (b) off-kinetic mean response times during moderate and very heavy exercise stages. Values represent mean ± SEM ($N = 7$). PS, phosphatidylserine group; P, placebo group. Trial 1: presupplementation; Trial 2: postsupplementation.

0.392), and no statistical differences were identified between T1 and T2 (trial effect: $P = 0.491$).

Blood lactate concentrations were significantly elevated from preexercise values from Post-65% (Table 4) and peaked at Post-85% (mean value for all trials was 7.70 ± 0.33 mmol·L^{-1}). Blood glucose concentrations did not differ significantly from preexercise values at any time point (Table 4), and no differences were identified between groups or as a result of supplementation.

Serum cortisol concentrations were significantly elevated by exercise (time of sample effect, $P < 0.001$) from preexercise values of 378 ± 21 nmol·L^{-1} to postexercise values of 554 ± 32 nmol·L^{-1} (Fig. 5). Serum cortisol concentrations were not significantly affected by supplementation (supplementation group × trial × time of sample, $P = 0.118$).

Significant temporal changes were observed in all of the EFI subscales; subjects reported decreases in revitalisation, positive engagement and tranquility, and increases in physical exhaustion through exercise (Table 5). The three-way interactions were not significant in any of the subscales (Table 5).

DISCUSSION

The primary finding of this investigation was that oral supplementation with 750 mg·d^{-1} S-PtdSer for 10 d significantly affected exercise capacity in a group of recreationally active subjects during a staged intermittent cycling protocol. Furthermore, the enhancements in exercise capacity in PS ranged from 0:15 to 3:47 min:s, whereas the exercise times to exhaustion remained unchanged in P.

It was originally hypothesized that PtdSer would influence the primary oxygen uptake kinetic response and thereby increase exercise capacity. However, supplementation did not significantly affect MRT_{on}. Furthermore, no differences were observed between trial or supplementation group in MRT_{off}; therefore, the current data presents insufficient evidence to support any change in the primary oxygen uptake kinetic response following supplementation.

The causes of fatigue during cycling at 85% $\dot{V}O_{2max}$ (within an intensity domain that has been previously classified as very heavy exercise (19)) have not been fully elucidated and may include central and peripheral components (14). Exercise during the final bout was associated with near maximal oxygen uptakes and heart rates in addition to relatively high blood lactate concentrations; therefore, it can be assumed that heavy demands were placed on both oxidative and nonoxidative phosphorylation. Metabolic acidosis has been implicated as a mechanism of peripheral fatigue either through direct effects on the contractile proteins or through inhibition of key regulatory enzymes such as phosphofructokinase (7); however, it may be more likely that other ionic imbalances contribute to fatigue in this exercise model (1). *In vitro* studies have demonstrated that low concentrations of PtdSer are effective in activating (Na^+-K^+)–dependent ATPase in mammalian kidney (25) and brain (28) preparations. Similarly, Ca^{2+}-ATPase, an enzyme primarily responsible for Ca^{2+} re-uptake from the muscle cytosol into the sarcoplasmic reticulum, is known to require PtdSer (18,24). Therefore, it is plausible that exogenous S-PtdSer delayed the onset of fatigue by maintaining ionic homeostasis for longer during exercise.

In addition, Tibbits et al. (27) reported that extended exercise training increased the levels of phospholipids, especially PtdSer content, in rat cardiac sarcolemma. This adaptation to training might suggest that additional PtdSer within the heart muscle has functional benefits during ex-

TABLE 3. Estimated percentage changes in plasma volume throughout both trials for phosphatidylserine supplementation group and placebo group.

	Trial 1 Pre-ex	Trial 1 Post-55%	Trial 1 Post-65%	Trial 1 Post-ex	Trial 1 Post-24 hr	Trial 2 Pre-ex	Trial 2 Post-55%	Trial 2 Post-65%	Trial 2 Post-ex	Trial 2 Post-24 hr	Effect: *P* interaction, *timing*
PS	—	−9.3 ± 1.3	−11.0 ± 1.3	−4.5 ± 0.7	4.9 ± 2.5	3.5 ± 2.7	−8.6 ± 1.4	−8.2 ± 1.2	−6.1 ± 1.8	3.8 ± 2.3	0.392
P	—	−7.3 ± 1.4	−8.9 ± 1.8	−3.1 ± 1.7	4.5 ± 1.4	0.5 ± 2.1	−6.9 ± 2.0	−9.8 ± 1.9	−4.9 ± 2.5	3.3 ± 2.3	*<0.001*

Values are presented as mean ± SEM ($N = 7$). PS, phosphatidylserine group; P, placebo group.
Effects present *P* values for supplementation group × trial × timing of sample interaction and timing.

ercise. An increase in membrane bound PtdSer may have the potential to enhance myocardial excitation–contraction coupling, potentially through the activation of different protein kinase C isoforms (26) and/or enhanced calcium uptake (20). Thus, it is plausible that these mechanisms also may have contributed to delaying fatigue in the present study. However, without corroborating data from further studies that investigate the *in vivo* pharmacological actions of S-PtdSer, the proposed mechanisms remain speculative.

The significant rise in serum cortisol concentration that followed the final bout of exercise suggested that the protocol activated the HPA axis (16). However, supplementation with PtdSer did not significantly influence serum cortisol concentrations (Fig. 5). This finding does not concur with the results of Fahey and Pearl (10), who found that S-PtdSer, using a similar supplementation regime, attenuated serum cortisol concentrations following resistance training. Furthermore, Monteleone et al. (17) reported that 800 $mg \cdot d^{-1}$ BC-PtdSer resulted in significant reductions in plasma cortisol and adrenocorticotrophic hormone (ACTH) concentrations during submaximal cycle exercise in untrained subjects.

The elevation in blood cortisol is a generic response to stress from both psychological and physical origin; consequently, there is considerable interindividual variability in response to exercise. Although the choice of experimental design in the current study investigated individual changes in response (pre- to postsupplementation) and, therefore, reduced the possible effect of subject selection–related bias, the possibility exists that the current dose may have been insufficient to attenuate the cortisol response in these active individuals. Alternatively, the current exercise protocol required that all participants continued the final exercise bout until exhaustion in both trials; therefore, it remains plausible that any effects of PtdSer supplementation on cortisol concentrations were masked as the PS group completed significantly more work in T2 when compared with T1.

Blood glucose concentrations remained unchanged throughout all trials. This finding was in agreement with previous studies using similar exercise protocols (16,17).

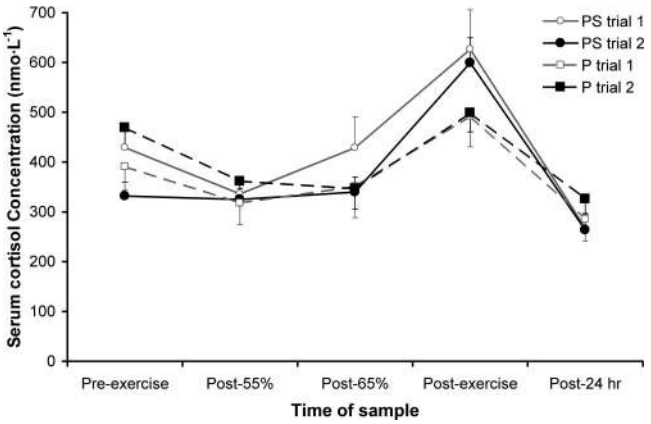

FIGURE 5—Serum cortisol concentrations throughout each exercise trial. Values represent mean ± SEM ($N = 7$). PS, phosphatidylserine group; P, placebo group. Trial 1: presupplementation; Trial 2: postsupplementation.

TABLE 4. Blood lactate and glucose concentrations during both exercise trials for phosphatidylserine supplementation group and placebo group.

		Trial 1 Pre-ex	Trial 1 Post-55%	Trial 1 Post-65%	Trial 1 Post-85%	Trial 1 Post-ex	Trial 2 Pre-ex	Trial 2 Post-55%	Trial 2 Post-65%	Trial 2 Post-85%	Trial 2 Post-ex	Effect: *P* interaction, *timing*
Lactate ($mmol \cdot L^{-1}$)	PS	1.50 ± 0.20	1.57 ± 0.13	2.71 ± 0.32	7.22 ± 0.80	2.49 ± 0.29	1.25 ± 0.17	1.69 ± 0.12	2.47 ± 0.23	8.47 ± 0.86	3.05 ± 0.35	0.155
	P	1.49 ± 0.32	1.95 ± 0.21	3.24 ± 0.14	8.01 ± 0.45	3.13 ± 0.22	1.38 ± 0.19	1.79 ± 0.20	2.69 ± 0.19	7.02 ± 0.50	3.06 ± 0.24	*< 0.001*
Glucose ($mmol \cdot L^{-1}$)	PS	4.22 ± 0.21	4.04 ± 0.09	3.95 ± 0.16	4.36 ± 0.25	4.13 ± 0.11	4.19 ± 0.10	4.06 ± 0.13	3.88 ± 0.06	4.40 ± 0.19	4.12 ± 0.14	0.355
	P	4.24 ± 0.10	4.24 ± 0.07	4.18 ± 0.11	4.49 ± 0.37	4.34 ± 0.20	4.42 ± 0.27	4.18 ± 0.11	4.08 ± 0.15	4.41 ± 0.29	4.41 ± 0.30	*0.281*

Values are presented as mean ± SEM ($N = 7$). PS, phosphatidylserine group; P, placebo group.
Effects present *P* values for supplementation group × trial × timing of sample interaction and timing.

TABLE 5. Exercise-induced feeling inventory (EFI) subscale scores throughout both trials for phosphatidylserine supplementation group and placebo group.

		Trial 1 Pre-ex	Trial 1 Post-55%	Trial 1 Post-65%	Trial 1 Post-85%	Trial 2 Pre-ex	Trial 2 Post-55%	Trial 2 Post-65%	Trial 2 Post-85%	Effect: *P* interaction, *timing*
Revitalisation	PS	2.2 ± 0.2	1.9 ± 0.1	1.6 ± 0.1	1.7 ± 0.4	2.7 ± 0.1	2.1 ± 0.3	1.8 ± 0.3	2.3 ± 0.2	0.377
(0–5)	P	2.4 ± 0.3	2.2 ± 0.2	1.8 ± 0.3	2.3 ± 0.3	2.4 ± 0.2	2.1 ± 0.3	1.8 ± 0.4	2.0 ± 0.4	*0.006*
Positive Engagement	PS	3.1 ± 0.2	2.5 ± 0.2	2.4 ± 0.2	2.6 ± 0.4	3.1 ± 0.2	2.7 ± 0.2	2.7 ± 0.2	2.8 ± 0.2	0.827
(0–5)	P	2.8 ± 0.3	2.7 ± 0.2	2.5 ± 0.2	2.6 ± 0.3	2.4 ± 0.3	2.3 ± 0.4	2.4 ± 0.5	2.6 ± 0.4	*0.044*
Tranquillity	PS	2.7 ± 0.2	2.3 ± 0.2	2.1 ± 0.3	1.9 ± 0.5	2.7 ± 0.3	2.3 ± 0.3	2.0 ± 0.4	2.6 ± 0.4	0.235
(0–5)	P	2.8 ± 0.3	2.4 ± 0.1	2.1 ± 0.2	2.6 ± 0.3	2.2 ± 0.3	2.3 ± 0.3	2.2 ± 0.4	2.6 ± 0.4	*<0.001*
Physical Exhaustion	PS	1.3 ± 0.3	1.8 ± 0.1	2.3 ± 0.2	1.7 ± 0.4	0.9 ± 0.2	1.6 ± 0.2	2.1 ± 0.2	1.8 ± 0.3	0.496
(0–5)	P	1.4 ± 0.3	1.8 ± 0.2	1.9 ± 0.2	2.2 ± 0.3	1.4 ± 0.3	1.9 ± 0.3	1.6 ± 0.3	1.9 ± 0.5	*<0.001*

Values are mean subscale scores; presented as mean ± SEM ($N = 7$). PS, phosphatidylserine group; P, placebo group.
Effects present *P* values for supplementation group × trial × timing of sample interaction and timing.

The concomitant effects of reduced insulin and elevations in ACTH, cortisol, and epinephrine are responsible for controlling blood glucose during exercise (3). Therefore, any effects that S-PtdSer supplementation may have had on blood ACTH and cortisol did not appear to have overchallenged blood glucose homeostasis during exercise. Furthermore, the calculated rates of carbohydrate, fat, and combined fuel oxidation during the steady-state stages of exercise were similar in all trials, suggesting that any change in the HPA axis induced by S-PtdSer supplementation did not affect substrate oxidation during moderate exercise stages.

All subscales of the EFI were sensitive to change during the exercise. However, the three-way interaction did not reach significance in any subscale; therefore, there was no evidence to suggest that feeling states differed following supplementation in either supplementation group. The participants in the current study provided baseline responses that were similar to those of other recreationally active populations before exercise training (15), indicating that the testing procedures did not induce large changes in feeling states before exercise in these subjects. Benton et al. (4) reported improvements in mood after mental stress within a subgroup of young healthy adults following chronic S-PtdSer supplementation. Nevertheless, these improvements were only identifiable in a subgroup of subjects who scored higher than the median for neuroticism; people who score highly on this dimension are known to display strong emotional reactions to stress (4). Consequently, the baseline emotional state of an individual might influence the efficacy of S-PtdSer in altering feeling states during exercise.

To our knowledge, this is the first study to identify the ergogenic properties of phosphatidylserine; therefore, we suggest that further studies are required to substantiate these findings and to investigate the potential uses of S-PtdSer in exercise and physical activity. In addition, future studies are warranted to investigate the mechanism by which S-PtdSer may act physiologically.

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