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# Concurrent sprint and aerobic training in swimming: Influence of exercise sequence on physiological responses and perceived exertion

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## ABSTRACT

The study examined the effect of aerobic and sprint sets sequence on physiological responses and perceived exertion during concurrent training sessions. Twelve male highly trained swimmers performed four sessions in randomized order, using combinations of the following training sets: (a) lactate threshold training ( $8 \times 200\text{-m}$  at a speed corresponding to lactate threshold with 30-s recovery; LT), (b) high-intensity aerobic training ( $8 \times 100\text{-m}$  at the maximal aerobic speed with 30-s recovery; MAS) and (c) repeated-sprints training ( $8 \times 25\text{-m}$  repeated sprints with 2-min recovery; SPR). The four combinations used were as follows: LT-SPR, SPR-LT, MAS-SPR, SPR-MAS. Blood lactate (BL), pH, base excess (BE), bicarbonate, heart rate (HR), HR variability, objective [training impulse (iTRIMP)] and subjective training load [session's rating of perceived exertion (sRPE)] were measured. Between session pH and BE were no different, but mean BL was higher in sessions starting with repeated sprints compared with the reverse order (SPR-LT:  $6.3 \pm 3.6$ , LT-SPR:  $5.3 \pm 3.7$   $\text{mmol}\cdot\text{L}^{-1}$ ,  $p = 0.03$ ; SPR-MAS:  $7.2 \pm 3.9$ , MAS-SPR:  $6.0 \pm 3.7$   $\text{mmol}\cdot\text{L}^{-1}$ ,  $p = 0.05$ ). Bicarbonate in SPR-LT was lower compared with LT-SPR ( $p = 0.03$ ). sRPE, but not iTRIMP, was higher in sessions starting with SPR compared with the reverse order ( $p = 0.02$ ). Anaerobic-aerobic set sequence, compared with the reverse order, augments BL response and increases perceived training load but not the training impulse.

## ARTICLE HISTORY

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## KEYWORDS

Repeated sprints; interval training; heart rate variability; internal load; training impulse

## Introduction

In swimming training practice, various sets performed with different intensities are applied in different order in the same training session (Hermosilla et al., 2021). In this context, aerobic dominated sets may precede or follow sets that have been planned to activate the anaerobic metabolism and vice versa. The intensity, volume and duration of the exercise stimulus may influence the physiological responses (Skorski et al., 2012) and the perceived exertion of swimmers (Barroso et al., 2015). By altering the sequence of training sets, the physiological responses and perceived effort may be modified, thereby affecting the total training load (Nikitakis et al., 2023b).

Intense swimming training has a distinct impact on exercise metabolism and physiological responses which, in turn, have an impact on the autonomic nervous system (Buchheit, 2014; Buchheit & Laursen, 2013). For instance, during repeated sprint training, the length of the intervals and the work:rest ratio may induce different autonomic nervous system response to exercise (Lloria-Varela et al., 2023). The degree of physiological and metabolic load of training may alter autonomic function, and monitoring these responses through heart rate variability (HRV) may be used to evaluate the stress-recovery status (Lloria-Varela et al., 2023).

Previous studies have examined the effect of set sequence in training sessions that include an aerobic training set and a repeated sprint training set, usually including four to eight 50-m repetitions at near maximum speed (Nikitakis et al.,

2023a, 2023b). When repeated sprints preceded from a mainly aerobic training set, a higher training load was induced, compared to the reverse sequence (Nikitakis et al., 2023a, 2023b). However, it is unknown whether sprint duration (i.e., 25 m vs. 50 m sprints) modifies the physiological responses and perceived exertion to this type of exercise in a different way. The higher dependence on phosphocreatine (PCr) breakdown and the expected lower glycolytic contribution of repeated 25-m sprints when applied with 2-min intervals compared to longer efforts (e.g., 50-m) may induce a milder metabolic milieu and thus lower fatigue (Bogdanis et al., 1998). The impact of the chosen set sequence on the overall physiological responses and the rating of perceived exertion is of great importance for the practitioners who aim to manipulate training load to optimize adaptations and long-term performance in swimming training (Mujika et al., 2018). Furthermore, there is limited information regarding the different methods of internal load calculation, and their relationship with the metabolic responses during aerobic and repeated sprints training set applied in different order (Nikitakis et al., 2023a, 2023b).

The purpose of this study was to investigate the effect of performing sequences of aerobic and repeated sprints sets in different order on session's physiological responses and perceived exertion. It was hypothesized that a training session including repeated sprints followed by a training set that mainly activates the aerobic metabolism will lead to higher overall metabolic and physiological responses and session's

rating of perceived exertion (sRPE), compared with the reverse order.

## Materials and methods

### Participants

Twelve male swimmers (age:  $19 \pm 3$  years, body mass:  $77.1 \pm 11.0$  kg, height:  $180.0 \pm 5.6$  cm) participated in the study. The swimmers were highly trained/national level (McKay et al., 2022) with best performance in 200 m front crawl at or above the 90th percentile of the national record and 84th percentile of the world record ( $121.5 \pm 6.6$  s,  $615 \pm 88$  World Aquatics points). All tests were carried out during the mesocycle of specific preparation (10–15 weeks before the national championship) and applied the same time of day, at least 48 h apart, in a 25-m indoors swimming pool with constant water and ambient temperature ( $24\text{--}25^\circ\text{C}$ ,  $27\text{--}28^\circ\text{C}$ , respectively). Participants recorded their diet 2 days before the first testing session and were asked to follow the same diet 2 days before each of the following testing sessions. The swimmers agreed to participate by signing written informed consent before the commencement of the study, which had received approval from the institutional ethical committee (1351/3 March 2022).

### Preliminary tests

Before each test, a standardized warm-up was performed (400 m front crawl, 200 m front crawl drills,  $4 \times 50\text{-m}$  front crawl at pace 80% of personal best 400 m, a 12.5 m sprint). On the first visit, swimmers performed two all-out efforts of 25 and 400 m front crawl with push-off from inside the pool, separated by 10 min of active and 20 min of passive recovery. The performance in the 25 and 400 m was used for the assessment of maximal speed and maximal aerobic speed (MAS) (Zacca et al., 2019). On a second visit, five even paced repetitions of 200 m swimming front-crawl ( $5 \times 200\text{-m}$ ) were performed in 5-min cycles with increasing speed in each repetition and starting with push-off from inside the pool (Nikitakis & Toubekis, 2021). The time to complete each 200 m was recorded by two independent researchers, using a digital stopwatch (FINIS 3X300, UK). Fingertip capillary blood was collected in the first 30 s of recovery after each 200-m repetition and analyzed for BL (Lactate Scout<sup>+</sup>, SensLab GmbH, Leipzig, Germany). Individual speed vs. BL curves were drawn and used for the determination of the speed corresponding to LT (Nikitakis & Toubekis, 2021). Heart rate (HR) was continuously recorded with an optical HR sensor (Polar Verity Sense, Polar Electro Oy, Kempele, Finland) placed on the temple under the swimmers' cap.

### Study design

Data collected in preliminary testing were used to plan training sets with the following characteristics. Set LT consisted of eight repetitions of 200 m ( $8 \times 200\text{-m}$ ) with intensity corresponding to the speed at LT separated by 30 s recovery periods (Skorski et al., 2012). Set MAS consisted of eight repetitions of 100 m ( $8 \times 100\text{-m}$ ) with intensity corresponding to MAS separated by 30 s recovery periods (Libicz et al., 2005). Set SPR consisted of

eight repetitions of 25 m ( $8 \times 25\text{-m}$ ) sprint swimming, separated by 2 min recovery periods [i.e., 1:8 swimming:recovery ratio (Toubekis et al., 2005)]. Rest intervals were short (i.e., 30 s) allowing partial recovery but maintaining increased aerobic contribution in LT and MAS sets and long enough (i.e., 2 min) to allow adequate PCr resynthesis in SPR set (Bogdanis et al., 1998; Shimoyama et al., 2003). Four sessions were performed in randomized order including the abovementioned training sets: (i) set LT followed by set SPR (session LT-SPR), (ii) set SPR followed by set LT (session SPR-LT), (iii) set MAS followed by set SPR (session MAS-SPR), (iv) set SPR followed by set MAS (session SPR-MAS). In each session, 10 min of passive recovery was applied between sets allowing adequate but not complete recovery and providing enough time to apply the required measurements after the first set and before the start of the second set.

### Measurements

Before and after the first and the second set, a blood sample (90  $\mu\text{l}$ ) was collected in plastic heparinized tubes (Sarstedt AG & Co., Germany) from a pre-warmed finger and analyzed using an iSTAT biochemical analyser (iSTAT Corporation, Princeton, NJ, USA) for the determination of pH in the blood and the calculation of base excess (BE) and bicarbonate ( $\text{HCO}_3$ ) concentration. BL was assessed before, at the middle and at the end of each set. The area under the BL curve and above the pH curve (because pH decreases after exercise) was calculated based on the trapezoidal rule (Yeh, 2002). Absolute differences ( $\Delta$ ) between time points of measurements (post-set vs. pre-set) were calculated. The time to complete each swimming repetition and the rating of perceived exertion (RPE) were recorded, whereas HR was continuously monitored. The average speed was calculated from the time taken to complete each swimming repetition. Swimming time of each repetition in set SPR was used to calculate the decrement score (DS) (Oliver, 2009).

Session RPE (sRPE) was recorded 30 min after the completion of each session and was used to calculate subjective internal load as the product of sRPE and the duration of the training session in min (Barroso et al., 2015). The individualized training impulse was calculated for each set and for the total session, to estimate objectively the internal load (iTRIMP) (Manzi et al., 2009). The iTRIMP formula combined exercise duration, heart rate reserve ( $\Delta\text{HR}$ ) and a weighting factor  $y$ :  $\text{iTRIMP} = \text{duration} \times \Delta\text{HR} \times y$ , where duration is the time in minutes spent at a specific HR,  $\Delta\text{HR}$  represents the fractions of heart rate reserve during the session calculated as:  $\Delta\text{HR} = \text{HR}_{\text{average}} - \text{HR}_{\text{rest}} / \text{HR}_{\text{maximal}} - \text{HR}_{\text{rest}}$  and  $y$  represents an individual weighting factor calculated when BL plotted against the fractional elevation in HR during the  $5 \times 200\text{-m}$  preliminary test (Manzi et al., 2009). HR<sub>rest</sub> was measured after awakening in supine position as the mean HR during 1 min. For the entire session iTRIMP calculation, the average HR from the start of the first set until 10 min after the second training set was used, including HR in the recovery period between the two sets. In each session, before the first set and after each set, HR variability (HRV) was recorded in a sitting position (Buchheit et al., 2007) for 5 min using an electrocardiographic chest-strap HR monitor (H10 sensor, Polar Electro, Kempele, Finland), paired with a freely available

smartphone application (Elite HRV, Asheville, North Carolina, USA). The logarithm of root mean square successive difference (LnRMSSD) was used to examine the effect of each set on autonomic function (Buchheit, 2014).

Furthermore, R-R intervals were recorded during the night after each session, and HRV was calculated. From this recording and after the removal of the first 30 min, a 4-h recording was chosen for the nocturnal HRV analysis (Hynynen et al., 2010). LnRMSSD was used to examine the effect of the entire session on the autonomic function. All R-R files were exported from the Elite HRV smartphone application and stored onto a separate computer for analysis using Kubios HRV 3.4.1 (Kuopio, Finland). Before analysis, each file was corrected for ectopic beats and artifacts using an artifact correction method provided in Kubios HRV. A very strong level of artifact correction was chosen to help preserve the variability while addressing the presence of any artifacts.

### Statistical analysis

Statistica v.10 software (Stat-Soft Inc, Tulsa, OK, USA) was used for data analysis. Sphericity was verified using Mauchly's test. A two-way analysis of variance for repeated measures was used to examine differences in speed and physiological responses between sessions and sets. When significant main effects were found, a Tukey's honest significant difference post-hoc test was used to identify differences between means. Effect size (ES) was calculated with Cohen's *d* and was categorized as small

(0.20–0.49), medium (0.50–0.80) and large effect [ $>0.80$  (Cohen, 1992)]. Pearson's correlation coefficient was used to examine relationships between parameters. A priori power analysis indicated a required sample size of  $n = 11$ , given error probability (0.05), power (0.80) and a medium effect size (Faul et al., 2007). Significance was set at  $p \leq 0.05$ . Data are presented as mean  $\pm$  SD.

### Results

Mean swimming speed in each set LT, MAS or SPR was unaffected by set sequence (Table 1). Swimmers in the SPR set maintained speed corresponding to 97–99% of the maximal speed attained in the preliminary test with no difference between sessions (LT-SPR:  $97.8 \pm 2.8$ , SPR-LT:  $98.4 \pm 2.2$ , MAS-SPR:  $98.9 \pm 2.6$ , SPR-MAS:  $98.4 \pm 2.7\%$ ,  $p = 0.40$ ). Furthermore, DS in SPR set did not differ, irrespective of the applied sequence (LT-SPR:  $2.18 \pm 0.66$ , SPR-LT:  $2.05 \pm 0.95$ , MAS-SPR:  $1.84 \pm 0.81$ , SPR-MAS:  $1.66 \pm 0.46\%$ ,  $F_{1,33} = 1.2$ ,  $p = 0.33$ ). There was no HR difference between sessions (LT-SPR vs. SPR-LT:  $p = 0.92$ , MAS-SPR vs. SPR-MAS:  $p = 0.80$ ) and between sets (LT-SPR and SPR-LT sessions, set LT:  $p = 0.90$ , set SPR:  $p = 0.94$ , MAS-SPR and SPR-MAS sessions, set MAS:  $p = 0.63$ , set SPR:  $p = 0.90$ , Table 1). The reported RPE was higher at the start of sets LT and MAS in sessions SPR-LT and SPR-MAS compared with the reverse sequence (Table 2). Furthermore, at the start of set SPR, RPE was higher in LT-SPR compared with SPR-LT session (Table 2). During sets LT and MAS, RPE was higher in SPR-LT and SPR-MAS

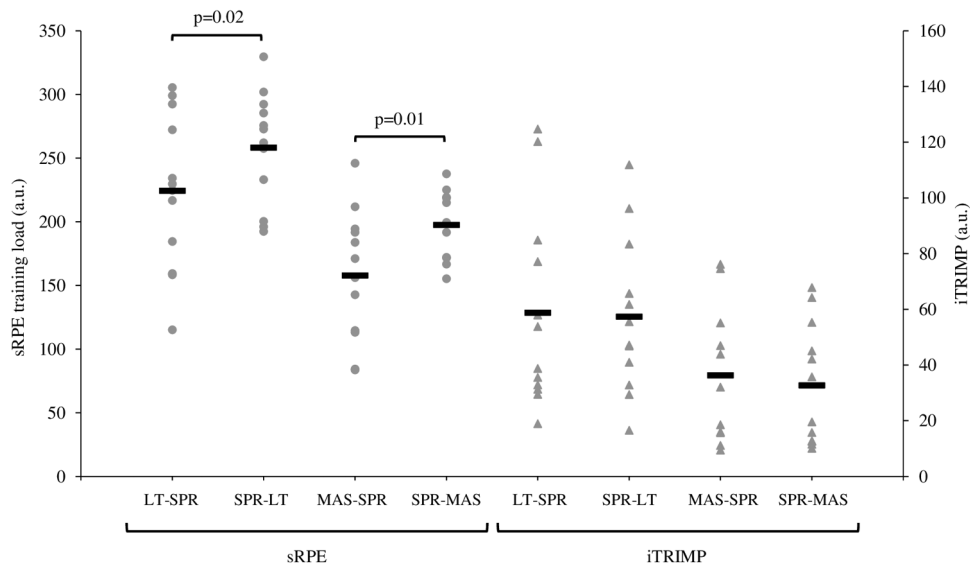
**Table 1.** Swimming speed, heart rate (HR) and individualized training impulse (iTRIMP) in each set LT, MAS, SPR, applied in four sessions of the present study (sessions LT-SPR, SPR-LT, MAS-SPR, SPR-MAS). Effect size (ES) refers to the comparison between the same sets.

	Session LT-SPR		Session SPR-LT		Between sessions ES	
	set LT	set SPR	set SPR	set LT	ES for set LT	ES for set SPR
Speed ( $m \cdot s^{-1}$ )	$1.40 \pm 0.08$	$1.98 \pm 0.08$	$1.99 \pm 0.08$	$1.39 \pm 0.08$	0.04	0.17
HR ( $beats \cdot min^{-1}$ )	$170 \pm 11$	$150 \pm 12$	$149 \pm 9$	$172 \pm 9$	0.17	0.13
iTRIMP (a.u.)	$79.9 \pm 32.0$	$11.6 \pm 8.8$	$10.8 \pm 7.4$	$84.3 \pm 19.2$	0.17	0.09
	Session MAS-SPR		Session SPR-MAS		Between sessions ES	
	set MAS	set SPR	set SPR	set MAS	ES set MAS	ES set SPR
Speed ( $m \cdot s^{-1}$ )	$1.49 \pm 0.07$	$2.00 \pm 0.08$	$1.99 \pm 0.08$	$1.49 \pm 0.07$	0.01	0.12
HR ( $beats \cdot min^{-1}$ )	$170 \pm 11$	$151 \pm 11$	$150 \pm 9$	$172 \pm 9$	0.18	0.04
iTRIMP (a.u.)	$38.7 \pm 19.9$	$11.8 \pm 8.5$	$8.7 \pm 6.7$	$45.0 \pm 12.3$	0.39	0.42

**Table 2.** Logarithm of root mean square successive difference (LnRMSSD) and rating of perceived exertion (RPE) before and after each training set LT, MAS, SPR and nocturnal LnRMSSD after each training session (sessions LT-SPR, SPR-LT, MAS-SPR, SPR-MAS). Effect size (ES) between sessions.

	Session LT-SPR					Session SPR-LT				ES	
	pre-set LT	post-set LT	pre-set SPR	post-set SPR	LnRMSSD (nocturnal)	pre-set SPR	post-set SPR	pre-set LT	post-set LT	LnRMSSD (nocturnal)	LnRMSSD nocturnal
Ln RMSSD (ms)	2.7 $\pm 0.2$	2.1 $\pm 0.4$	–	2.6 $\pm 0.3$	$3.3 \pm 0.2$	2.9 $\pm 0.2$	2.4 $\pm 0.4$	–	2.1 $\pm 0.5$	$3.3 \pm 0.1$	0.08
RPE (a.u.)	1.6 $\pm 1.4$	4.5 $\pm 1.6$	3.2 $\pm 1.7$	5.1 $\pm 1.5$		1.7 $\pm 1.4^*$	5.0 $\pm 1.4$	3.7 $\pm 2.0^*$	6.0 $\pm 1.6^*$		
	Session MAS-SPR					Session SPR-MAS				ES	
	pre-set MAS	post-set MAS	pre-set SPR	post-set SPR	LnRMSSD (nocturnal)	pre-set SPR	post-set SPR	pre-set MAS	post-set MAS	LnRMSSD (nocturnal)	LnRMSSD nocturnal
Ln RMSSD (ms)	2.7 $\pm 0.4$	2.3 $\pm 0.5$	–	2.2 $\pm 0.5$	$3.3 \pm 0.2$	2.8 $\pm 0.2$	2.5 $\pm 0.4$	–	2.3 $\pm 0.6$	$3.3 \pm 0.2$	0.14
RPE (a.u.)	1.7 $\pm 1.5$	4.4 $\pm 1.8$	1.7 $\pm 1.6$	4.9 $\pm 1.6$		2.1 $\pm 1.3$	4.7 $\pm 1.6$	3.8 $\pm 1.5^*$	6.3 $\pm 1.3^*$		

\*:  $p < 0.05$  between the same training set in each session.



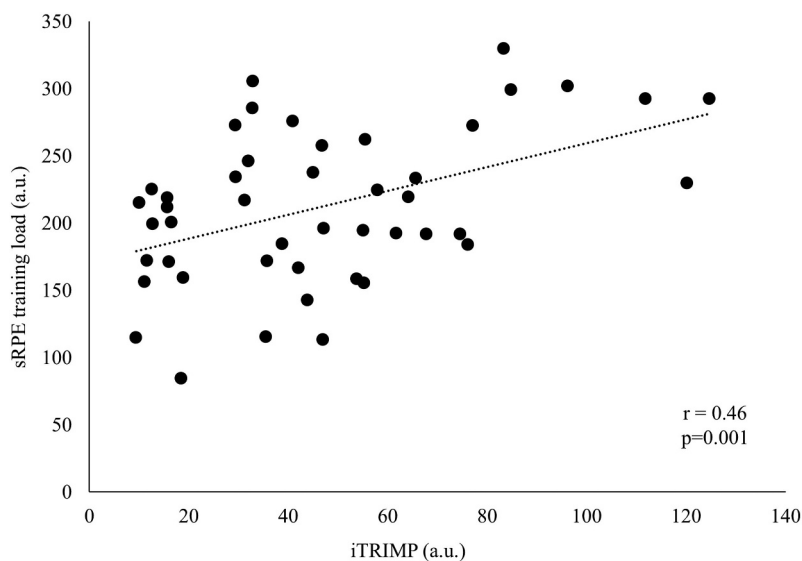
**Figure 1.** Session RPE (sRPE) training load in circles and individualized training impulse (iTRIMP) in triangles after each training session (LT-SPR, SPR-LT, MAS-SPR, SPR-MAS).  $p$  value corresponds to the comparison between sessions in sRPE training load.

sessions compared with reverse sequence ( $p = 0.01$ ) while during set SPR, RPE was not affected by the applied sequence ( $p = 0.99$ , Table 2).

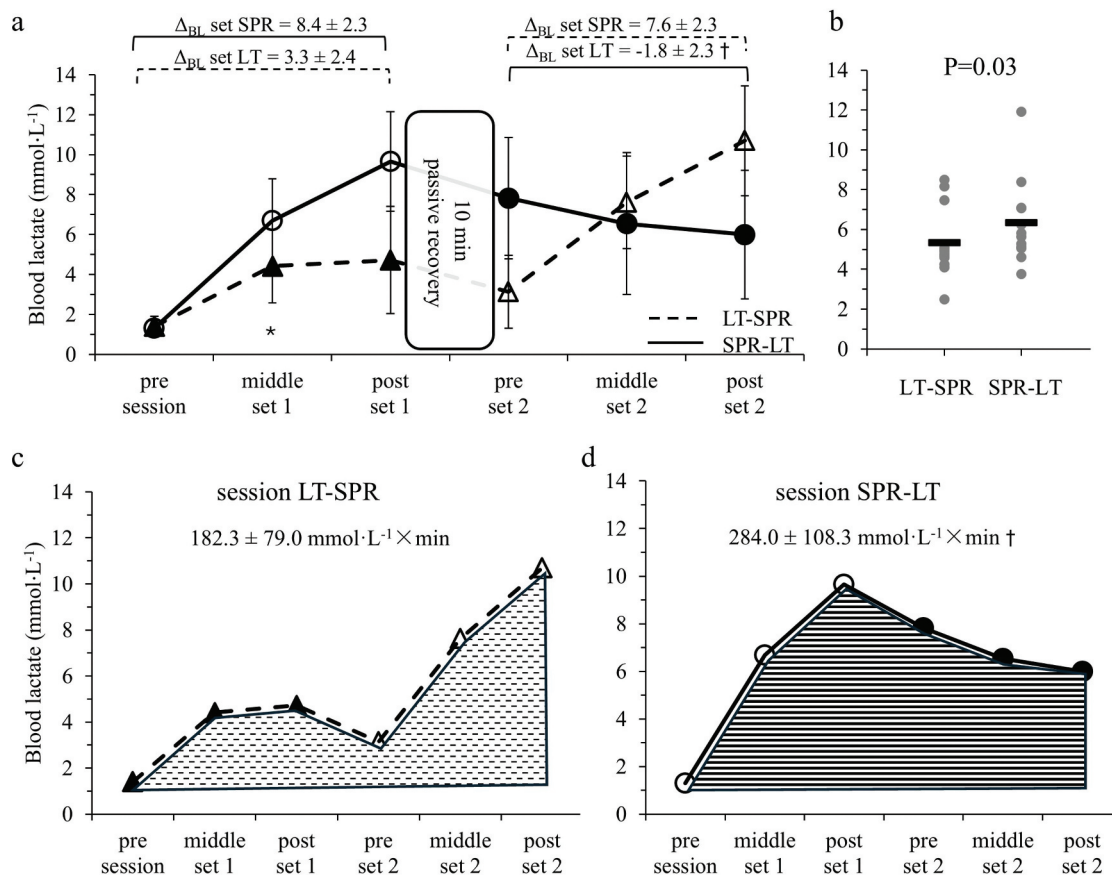
Subjective (sRPE) training load was higher in SPR-LT and SPR-MAS sessions compared with the reverse sequence (LT-SPR vs SPR-LT,  $ES = 0.64, t = -2.8, p = 0.02$ , MAS-SPR vs SPR-MAS,  $ES = 1.00, t = -3.8, p = 0.01$ , Figure 1). iTRIMP in each set (LT, MAS, or SPR) did not differ as a result of the applied sequence (Table 1). Entire session iTRIMP did not differ depending on the applied set sequence (LT-SPR vs SPR-LT,  $ES = 0.05, t = 0.3, p = 0.75$ , MAS-SPR vs SPR-MAS,  $ES = 0.16, t = 2.06, p = 0.07$ , Figure 1). sRPE training load and iTRIMP were positively correlated ( $r = 0.46, p = 0.01$ , Figure 2). The effect of each set separately on autonomic function and the nocturnal

LnRMSSD after each training session was independent of the applied set sequence (Table 2).

BL in session SPR-LT was higher compared with LT-SPR ( $F_{1,11} = 6.8, p = 0.03$ ; Figure 3). Mean BL of set LT was higher when applied after set SPR ( $F_{1,11} = 33.6, p = 0.01$ ; Figure 3) and BL at middle set LT was higher in SPR-LT session compared with LT-SPR ( $F_{2,22} = 18.5, p = 0.01$ ; Figure 3). In addition, BL in session SPR-MAS was higher compared with MAS-SPR ( $F_{1,11} = 4.8, p = 0.05$ ) and swimmers performed set MAS in SPR-MAS session with higher BL compared with MAS-SPR ( $F_{2,22} = 25.8, p = 0.01$ ; Figure 4). BL at set SPR did not differ between LT-SPR and SPR-LT sessions ( $p = 0.16$ , Figure 4). However, mean BL during set SPR in MAS-SPR session was higher compared with SPR-MAS ( $F_{1,11} = 48.1, p = 0.01$ ; Figure 4) and at middle set SPR was



**Figure 2.** Correlation between the individualized training impulse (iTRIMP) and session RPE (sRPE) training load, including all the four training sessions (LT-SPR, SPR-LT, MAS-SPR, SPR-MAS) in the study.



**Figure 3.** Blood lactate (BL) in each set of LT-SPR and SPR-LT sessions (a), individually (grey circles) and mean BL (black line) of entire sessions (b) and the area under the BL curve in LT-SPR (c) and SPR-LT (d) session. Continuous line and circles indicate SPR-LT session and dashed line and triangles indicate LT-SPR session. Open circles and triangles indicate set SPR and filled circles and triangles indicate set LT.  $\Delta_{BL}$  set LT: change from pre- to post-set LT.  $\Delta_{BL}$  set SPR: change from pre- to post-set SPR. \*:  $p = 0.02$  in set LT compared with the middle set 2 in SPR-LT session. †:  $p = 0.01$  between sessions.

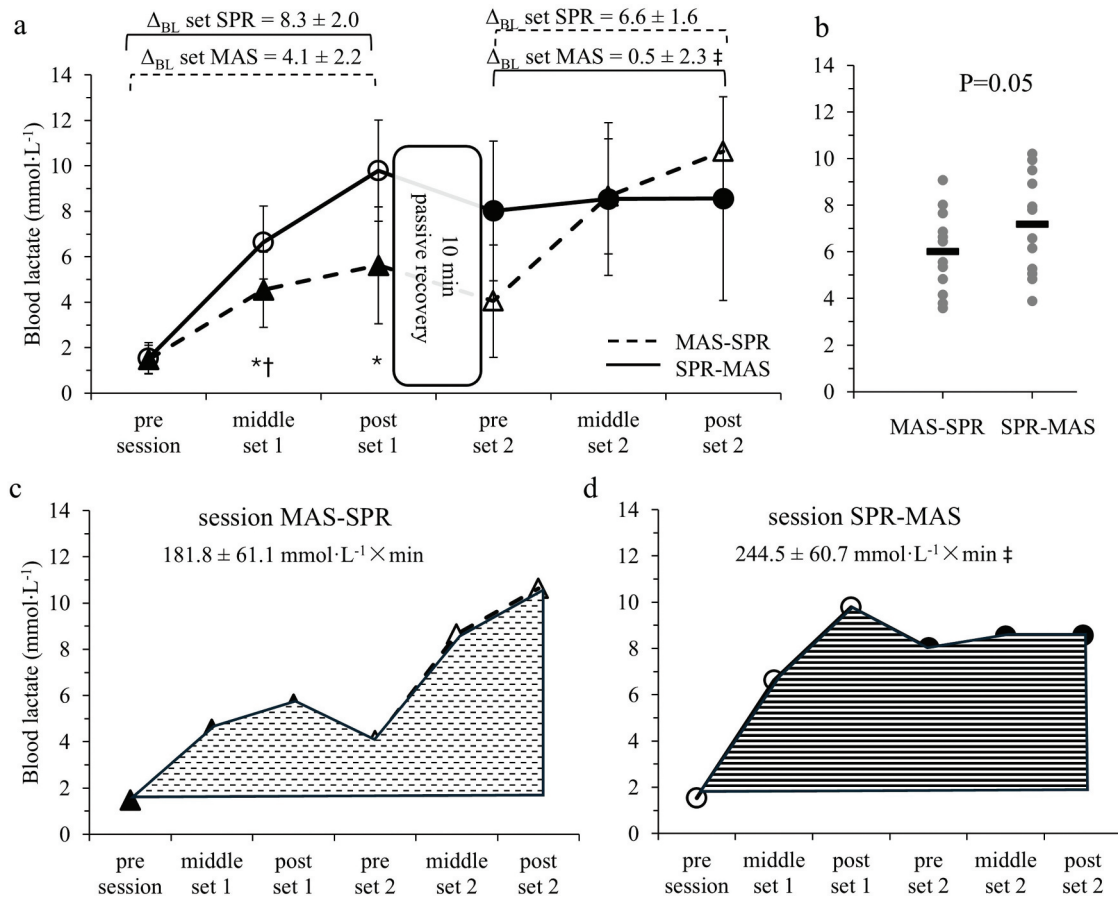
higher in MAS-SPR session compared with SPR-MAS ( $p = 0.01$ , Figure 4). BL difference between before and after sets LT and MAS ( $\Delta_{BL}$ ) was positive in LT-SPR and MAS-SPR while negative in SPR-LT and zero in SPR-MAS sessions (Figures 3, 4).  $\Delta_{BL}$  during set SPR did not differ between sessions (Figures 3, 4). The area under the BL curve in SPR-LT and SPR-MAS was higher compared with the reverse sequence (LT-SPR vs SPR-LT: 36% difference,  $ES = 1.09$ ,  $t = -4.1$ ,  $p = 0.01$ , Figure 3; MAS-SPR vs SPR-MAS: 26% difference,  $ES = 1.03$ ,  $t = -3.2$ ,  $p = 0.01$ , Figure 4). The area under the BL curve and the training load derived from sRPE in the four training sessions were correlated ( $r = 0.63$ ,  $p = 0.001$ , Figure 5).

Mean pH and BE (pH,  $F_{1,11} = 2.0$ ,  $p = 0.19$ ; BE,  $F_{1,11} = 4.5$ ,  $p = 0.06$ ) were similar in SPR-LT and LT-SPR session, while  $\text{HCO}_3^-$  in SPR-LT was lower compared with LT-SPR ( $F_{1,11} = 6.3$ ,  $p = 0.03$ , Table 3). Nevertheless, set LT or set SPR separately showed similar response independent of the applied sequence (Table 3). Mean pH, BE and  $\text{HCO}_3^-$  (pH,  $F_{1,10} = 1.9$ ,  $p = 0.19$ ; BE,  $F_{1,10} = 3.6$ ,  $p = 0.09$ ;  $\text{HCO}_3^-$ ,  $F_{1,10} = 3.5$ ,  $p = 0.09$ , Table 3) were similar in SPR-MAS and MAS-SPR sessions. Measured pH, BE and  $\text{HCO}_3^-$  after set MAS were lower in SPR-MAS session compared with the reverse order (Table 3). Delta values in pH, BE and  $\text{HCO}_3^-$  (Table 3) at sets LT and MAS were negative in LT-SPR and MAS-SPR while positive (or approximately zero) in SPR-LT and SPR-MAS sessions. The area above the pH curve in SPR-LT and SPR-MAS was higher compared with the reverse sequence (LT-

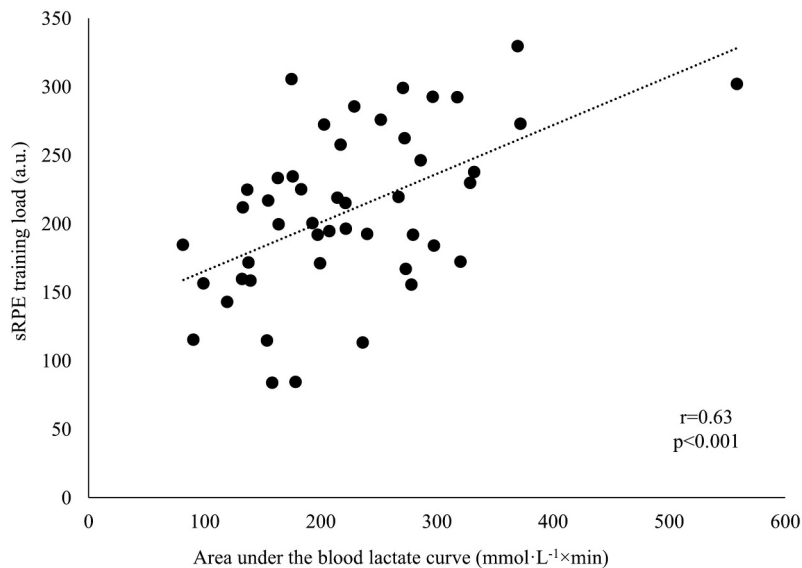
SPR:  $3.0 \pm 2.1$  vs SPR-LT:  $4.7 \pm 2.1$ , 36% difference,  $ES = 0.81$ ,  $t = 3.9$ ,  $p = 0.01$ ; MAS-SPR:  $3.0 \pm 1.7$  vs SPR-MAS:  $3.9 \pm 1.9$ , 24% difference,  $ES = 0.53$ ,  $t = 2.2$ ,  $p = 0.05$ ).

## Discussion

The purpose of the study was to investigate the effect of different sequences of training sets performed at intensities corresponding to LT or MAS and repeated sprints applied concurrently within a swimming training session on performance, physiological responses and internal load. The main findings of the study were that (i) performance in each set was maintained irrespective of the applied sets sequence, (ii) the overall session metabolic responses were higher when repeated sprints preceded the LT and MAS sets, (iii) the acid-base balance during the entire session was not influenced by sets sequence. However, the area under the BL and above pH curve was higher in sessions starting with repeated sprints, indicating that a longer time was spent with high BL and low pH in these sessions compared with the reverse sequence, (iv) subjective internal load (sRPE) and objective iTRIMP were correlated but only sRPE was higher in the sessions that repeated sprints preceded the LT and MAS sets, thus the two methods presented different estimated internal load, (v) sRPE-based internal load was correlated with the area under the BL curve.



**Figure 4.** Blood lactate (BL) in each set of MAS-SPR and SPR-MAS sessions (a), individually (grey circles) and mean BL (black line) of entire sessions (b) and the area under the BL curve in MAS-SPR (c) and SPR-MAS (d) session. Continuous line and circles indicate SPR-MAS session and dashed line and triangles indicate MAS-SPR session. Open circles and triangles indicate set SPR and filled circles and triangles indicate set MAS.  $\Delta_{BL} \text{ set MAS}$ : change from pre- to post-set MAS.  $\Delta_{BL} \text{ set SPR}$ : change from pre- to post-set SPR. \*:  $p = 0.01$  in set MAS compared with the middle and post set 2 in SPR-MAS session. †:  $p = 0.01$  in set SPR compared with the middle set 1 in SPR-MAS session. ‡:  $p = 0.01$  between sessions.



**Figure 5.** Correlation between the area under the blood lactate curve and session RPE (sRPE) training load, including all the four training sessions (LT-SPR, SPR-LT, MAS-SPR, SPR-MAS) in the study.

Table 3. pH, base excess (BE) and bicarbonate (HCO<sub>3</sub>) concentration in sets LT, MAS, SPR within the four experimental sessions of the study (sessions LT-SPR, SPR-LT, MAS-SPR, SPR-MAS). Effect size (ES) between sessions.

	Session SPR-LT										
	Session LT-SPR					Session SPR-LT					
	pre-set LT	post-set LT	$\Delta_{set}$ LT	pre-set SPR	post-set SPR	$\Delta_{set}$ SPR	overall session	pre-set LT	post-set LT	$\Delta_{set}$ LT	overall session
pH	7.41 ± 0.02	7.35 ± 0.06	-0.06 ± 0.06	7.37 ± 0.05	7.25 ± 0.07	-0.13 ± 0.06	7.34 ± 0.08	7.31 ± 0.06*	7.34 ± 0.06	0.03 ± 0.03	7.33 ± 0.08
BE (mmol·L <sup>-1</sup> )	0.6 ± 1.5	-5.0 ± 4.7	-5.6 ± 4.1	-3.4 ± 4.2	-13.6 ± 4.4	-10.2 ± 3.8	-5.4 ± 6.4	-9.9 ± 3.9*	-5.6 ± 4.7	4.3 ± 2.6*	-6.9 ± 6.1
HCO <sub>3</sub> (mmol·L <sup>-1</sup> )	25.3 ± 1.3	20.6 ± 3.7	-4.7 ± 3.3	22.1 ± 3.6	13.7 ± 3.2	-8.5 ± 3.0	20.4 ± 5.2	16.3 ± 3.1*	19.8 ± 3.6	3.5 ± 2.2*	19.0 ± 4.9†

	Session SPR-MAS										
	Session MAS-SPR					Session SPR-MAS					
	pre-set MAS	post-set MAS	$\Delta_{set}$ MAS	pre-set SPR	post-set SPR	$\Delta_{set}$ SPR	overall session	pre-set MAS	post-set MAS	$\Delta_{set}$ MAS	overall session
pH	7.40 ± 0.02	7.33 ± 0.07	-0.07 ± 0.06	7.35 ± 0.05	7.22 ± 0.07	-0.13 ± 0.05	7.32 ± 0.08	7.31 ± 0.06*	7.29 ± 0.10	-0.02 ± 0.05	7.31 ± 0.09
BE (mmol·L <sup>-1</sup> )	-1.1 ± 2.1	-7.1 ± 4.7	-6.0 ± 4.1	-5.7 ± 4.1	-15.4 ± 3.3	-9.8 ± 2.8	-7.3 ± 6.3	-10.1 ± 3.6*	-10.4 ± 5.7*	-0.3 ± 3.0*	-8.4 ± 6.0
HCO <sub>3</sub> (mmol·L <sup>-1</sup> )	24.0 ± 2.0	18.9 ± 3.6	-5.0 ± 3.1	20.0 ± 3.4	12.2 ± 2.3	-7.7 ± 2.2	18.8 ± 5.1	16.2 ± 2.7*	16.3 ± 4.2*	0.1 ± 2.3*	17.9 ± 4.7

\*:  $p < 0.05$  between the same training set in each session.  
 †:  $p < 0.05$  between sessions.  
 $\Delta_{set}$  LT: change from pre- to post-set LT.  
 $\Delta_{set}$  MAS: change from pre- to post-set MAS.  
 $\Delta_{set}$  SPR: change from pre- to post-set SPR.

The increased BL when repeated sprints were performed as the first set should not be considered as fatigue agent leading to decreased performance in a subsequent set (Toubekis et al., 2005, 2006), but rather as an energy source (Brooks, 2018). In fact, lactate may be used as a substrate and it represents the main gluconeogenic precursor (Brooks, 2020). The above-mentioned response of blood lactate concentration may induce long-term effects including faster resynthesis of glycogen after exercise (Nalbandian & Takeda, 2016) and this is important especially in periods of high-volume high-intensity training characterized by extensive glycogen depletion. The elevation of BL during training may result in adaptations such as mitochondrial biogenesis and improved metabolic flexibility (Brooks, 2020).

In the present study, the higher area under the BL curve observed in sessions starting with repeated sprints reflects longer duration of elevated lactate levels and is considered important, since lactate is a potential signaling molecule and its presence in blood may promote metabolic and molecular adaptations (Brooks, 2018). Such an increment in SPR-LT and SPR-MAS sessions is attributed to increased BL concentration following SPR set and a likely reduced removal during the subsequent LT and MAS sets. We expected that aerobic exercise at least in the LT set will increase lactate removal acting as active recovery (Greenwood et al., 2008). However, it seems that a longer set duration or a lower swimming intensity may be needed to remove the previously increased lactate concentration. Indeed, previous studies examined BL removal incorporating active recovery of lower intensity (54% of the maximal 25-m swimming speed, Toubekis et al., 2011) compared to 70% of the 25-m speed in the LT set in our study. Furthermore, LT corresponds to an intensity that lactate production and clearance are in equilibrium (Faude et al., 2009). Thus, a lower intensity may be required for a faster lactate removal. Moreover, extending sampling for BL measurement for a longer period after the end of each session would reveal a more complete view of the areas under the curve.

Overall session acid-base balance did not differ depending on the applied set sequence. However, acid-base regulation during exercise includes amino acids, proteins and HCO<sub>3</sub> that consume hydrogen ions to protect the cell against intracellular proton accumulation (Robergs et al., 2004). The lower HCO<sub>3</sub> in SPR-LT session may indicate an increased acid-base regulation protecting the cell from severe acidity (Juel, 2008). Furthermore, lactate itself can also act as an acid-base regulator providing proton efflux from the cell (Robergs et al., 2004); thus, it seems possible that the longer time spent with high BL values may induce long-term adaptations on pH regulation (Juel, 2008). Indeed, previous reports highlight that an athlete's ability to maintain glycolysis activation and simultaneously remove (or use) lactate is essential to improve performance in endurance activities (Brooks, 2018).

The higher RPE during sets LT (8×200-m) and MAS (8×100-m) in SPR-LT and SPR-MAS sessions compared with the reverse sequence indicates a higher perceived effort in order to maintain the required speed. The higher perceived exertion in these training sets may be due to the high BL and the related low pH caused by the preceding repeated sprints (Toubekis et al., 2008). Indeed, swimmers started sets LT and MAS with lower pH when applied after repeated sprints in the training sessions.



A remarkable exercise-induced metabolic acidosis provides neural feedback mediated by thin-fiber muscle afferents leading to inhibitory feedback of the magnitude of central motor drive (Amann, 2011). This inhibitory feedback is developed to avoid a potential overstraining and a possible harmful consequence for the muscle (Amann, 2011); the former is likely to be expressed by the higher RPE. Furthermore, an impaired shoulder muscle function that has been observed following high-intensity (Dekerle & Paterson, 2016) or repeated sprints swimming (Aujouannet et al., 2006) may confirm the increased perceived difficulty in sets LT and MAS. In any case, swimmers did not pass their individual critical threshold (Amann, 2011) or a sensory tolerance limit (Hureau et al., 2018) which may lead to fatigue development and termination of the task, since they managed to maintain the pre-defined speed during sets LT (8×200-m) and MAS (8×100-m) when applied after repeated-sprints.

The higher sRPE training load observed in sessions started with repeated sprints (SPR-LT and SPR-MAS) may be attributed to the higher physiological impact of these sessions compared with the reverse order. This is confirmed by the significant correlation noticed between the area under the BL curve and the training load. In fact, the longer duration that swimmers are forced to swim with high BL represents a chemical stimulus that activates metabosensitive receptors and this neural feedback is transferred to the central neural system, providing a perception of higher central fatigue (Amann, 2011). The observed positive relationship between training load and area under the BL curve indicates that by changing the training sets' sequence, a lower (or higher) training load may be caused and this can be applied depending on the training period. For instance, repeated sprints could be applied before an aerobic training set in training periods characterized by high training load and vice versa. Whatever the case, the effect that sets' sequence selection can have on internal load should be taken into account because a systematic implementation of training sessions with high load may affect long-term performance (Mujika et al., 2018).

The individually calculated iTRIMP in each training set obtained separately as well as in each training session did not differ depending on the applied set sequence. The abovementioned observation seems reasonable since HR in each training set separately, which was used for iTRIMP calculation, did not differ between sessions. It seems that a long duration of LT and MAS sets masked any difference in iTRIMP caused by the shorter duration SPR set when HR was maintained at lower level especially during the long 2 min recovery period without adding to the iTRIMP as it is confirmed with previous findings (García-Ramos et al., 2015). In contrast, any perception of fatigue caused by both SPR-LT or SPR-MAS sessions was indicated by the swimmers when using the sRPE method. The observed difference in results concerning the two methods confirms that coaches should be aware of a possible underestimation of the induced training load when iTRIMP method is used in training sessions consisting of repeated sprints and aerobic-dominated training sets. The abovementioned rationale arises from the fact that HR may be a less valid indicator of internal load in repeated sprints because of HR lag at the onset of exercise

accompanied with the short-duration efforts (Buchheit & Laursen, 2013). In short-duration maximal-intensity efforts neuromuscular factors are strongly associated with perceived exertion, and RPE may be the best index to reflect such changes in fatigue (Foster et al., 2021). In that case, a subjective tool for internal load monitoring (e.g., sRPE) may be more appropriate.

The relevant literature provides evidence that the higher the exercise intensity, the greater the metabolic acidosis and the lower the HRV (high sympathetic predominance) during recovery (Buchheit, 2014). However, recent study supports that changes in HRV response are intensity-dependent under specific circumstances (Lloria-Varela et al., 2023). In particular, the length of the intervals as well as the work:rest ratio may also affect the autonomic nervous system response to exercise (Lloria-Varela et al., 2023). Indeed, 8 × 50 m [30 s duration, 1:1 work:rest ratio (Nikitakis et al., 2023a)] and 4 × 50 m repeated sprints [30 s duration, 1:4 work:rest ratio (Nikitakis et al., 2023b)] induced higher sympathetic predominance compared with 8 × 25 m in the present study (13 s duration, 1:8 work:rest ratio). When a speed endurance training set consisted of 8 × 50 m was applied as a first set, high sympathetic predominance occurred at a subsequent training set applied at maximal aerobic speed (Nikitakis et al., 2023a). However, in the present study in which repeated sprints of shorter duration were applied (8 × 25 m, 2 min of recovery), the lower autonomic disturbance induced after repeated sprints did not affect the autonomic response observed after the following aerobic-dominated training set. Furthermore, autonomic function that reflects wellness and readiness to perform (Buchheit, 2014) seems to be fully recovered within 24 h of sprint interval training (Lloria-Varela et al., 2023). However, the novel finding of our study is that autonomic function 24 h later was comparable between sessions irrespective of the applied set sequence and this does not depend on the characteristics of training sets, as confirmed by previous studies (Nikitakis et al., 2023a, 2023b).

Nevertheless, a limitation of the study was that the overall duration of each training session was about half of a usual training session performed by highly trained swimmers (90–120 min) and the magnitude of autonomic response may differ depending on the overall volume and accumulated training load. We acknowledge that the effect sizes reported in the results section are rather small. Although such a small effect may be meaningful in repeated training practice, a larger sample size could strengthen our findings.

## Conclusions

Performing repeated sprints before an aerobic swimming training set (LT or MAS) augments BL response and increases perceived exertion, compared with the reverse order. However, heart rate responses and overall acid-base balance are not affected by set order. The higher sRPE load caused by training sessions consisting of repeated sprints followed by an aerobic training set is related to the longer time spent while exercising with high BL. In training periods that require a higher training load, coaches could choose to apply repeated sprints at the very start of the training session, while an inverse approach may reduce training load. The magnitude of the internal load induced when aerobic sets

and repeated sprints are combined may be masked when iTRIMP compared with sRPE method is selected, despite the positive relationship observed between the two methods. When combining repeated sprints and aerobic-dominated training sets systematically, the same training load evaluation method should be chosen. Autonomic function the night after each training session does not differ depending on the applied set sequence. Highly trained swimmers demonstrate a similar level of recovery the night after training sessions consisting of repeated sprints and an aerobic-dominated training set irrespective of the applied set sequence.

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## Author contributions

All authors contributed to the study conception and design. Material preparation, data collection and analysis were performed by ISN. The first draft of the manuscript was written by ISN and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

## Data availability statement

The data that support the findings of this study are available from the corresponding author AGT upon reasonable request.

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