

**Exercise testing in running: Merging traditional and novel  
concepts to assess physiology and performance**

Masterthesis  
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## Zusammenfassung

**Ziel:** Diese Thesis hatte zum Ziel, den Einfluss von traditionellen und neuen physiologischen sowie leistungsbezogenen Parametern auf 1-, 2-, 3-km Time-Trial Leistung im Laufen zu untersuchen. Auch Zusammenhänge zwischen erhobenen Parametern sowie die Übereinstimmung unterschiedlicher Schwellenkonzepte wurde untersucht.

**Methodik:** Eine Gruppe von Sprintern ( $n = 6$ ), Mittel- und Langstreckenläufern ( $n = 16$ ) und Ultraläufern ( $n = 3$ ) absolvierte mehrere spiroergometrische Tests, einen Sprinttest sowie Time-Trials (TT) über Distanzen von 1, 2 und 3 km. Dabei wurde der Einfluss von physiologischen ( $\dot{V}O_2\text{max}$ , RE,  $\% \dot{V}O_2\text{max}$ , MFO,  $\dot{V}La_{\text{max}}$ ,  $\Delta La_{100}$ ) und leistungsbezogenen Parametern ( $v\dot{V}O_2\text{max}$ , vMLSS, CV,  $Fat_{\text{max}}$ ,  $D'$ , ASR, SRR) auf die jeweiligen Time-Trial ermittelt. Zusätzlich wurden alle Parameter auf mögliche Zusammenhänge untersucht und die Übereinstimmung verschiedener Schwellenkonzepte (vOBLA, vMLSS and CV) überprüft.

**Ergebnisse:** Die größten Zusammenhänge wurden zwischen 2-km ( $r = 0,81$ ;  $r = 0,84$ ) und 3-km ( $r = 0,89$ ;  $r = 0,98$ ) TT-Leistung und  $\dot{V}O_2\text{max}$  sowie CV ermittelt. Für  $\dot{V}La_{\text{max}}$ ,  $\Delta La_{100}$ ,  $D'$ , ASR und SRR ergaben die Untersuchung positive Zusammenhänge mit der Sprintgeschwindigkeit ( $r = 0,73$ ;  $r = 0,54$ ;  $r = 0,69$ ;  $r = 0,56$ ;  $r = 0,43$ ) und negative Zusammenhänge mit 2- ( $r = -0,41$ ;  $r = -0,46$ ;  $r = -0,37$ ;  $r = -0,71$ ;  $r = -0,81$ ) und 3-km ( $r = -0,50$ ;  $r = -0,53$ ;  $r = -0,62$ ;  $r = -0,85$ ;  $r = -0,91$ ) TT-Leistung sowie vMLSS ( $r = -0,48$ ;  $r = -0,51$ ;  $r = -0,62$ ;  $r = -0,79$ ;  $r = -0,86$ ). Korrelationskoeffizienten waren im Allgemeinen niedriger für 1-km TT-Leistung als 2- und 3-km. Sehr gute Übereinstimmung wurde zwischen den Schwellenkonzepten ermittelt (vMLSS – vOBLA:  $R^2 = 0,94$ ; vMLSS – CV:  $R^2 = 0,83$ ) mit mittleren Abweichungen von  $-0,08$  und  $-0,49 \text{ m}\cdot\text{s}^{-1}$ .

**Fazit** Parameter mit Verbindung zum aeroben Stoffwechsel zeigten die größten Zusammenhänge mit TT-Leistung. Bei 1-km Time-Trial-Leistungen oder ca. 3-minütigen maximalen Laufbelastungen wird vermutet, dass negative und positive Einflüsse der anaeroben anaeroben Stoffwechsels, gemessen als  $\dot{V}La_{\text{max}}$ , in Waage liegen. Bei längeren Maximalbelastungen ist zunehmend von Leistungseinbußen durch  $\dot{V}La_{\text{max}}$  auszugehen. Basierend auf diesen Ergebnissen können neue Parameter traditionelle Parameter der Laufleistungsdiagnostik ergänzen. Es wird empfohlen Untersuchungsparameter

gezielt zur Leistungsvorhersage, Trainingssteuerung und in Abhängigkeit von der Wettkampfdistanz auszuwählen.

## Abstract

**Purpose** Aim of this thesis was the identification and critical review of traditional and novel physiological and performance parameters for different threshold concepts and 1-, 2-, 3-km time-trial (TT) running.

**Methods** Physiological tests and TTs were carried out in a group of sprinters ( $n = 6$ ), middle- and long-distance ( $n = 16$ ) and ultra-runners ( $n = 3$ ). Relationship between TT performance and physiological ( $\dot{V}O_{2\max}$ , RE,  $\% \dot{V}O_{2\max}$ , MFO,  $\dot{V}L_{a\max}$ ,  $\Delta La_{100}$ ) as well as performance parameters ( $v\dot{V}O_{2\max}$ ,  $vMLSS$ , CV,  $Fat_{\max}$ ,  $D'$ , ASR, SRR) was assessed. Additionally, correlations between all investigated parameters and agreement between velocity at different threshold concepts ( $vOBLA$ ,  $vMLSS$  and CV) was analyzed.

**Results**  $\dot{V}O_{2\max}$  and CV presented the strongest positive relationship with 2- ( $r = 0.81$ ,  $r = 0.84$ ) and 3-km ( $r = 0.89$ ,  $r = 0.98$ ) TT performance among physiological and performance parameters respectively.  $\dot{V}L_{a\max}$ ,  $\Delta La_{100}$ ,  $D'$ , ASR and SRR were positively correlated with sprint performance ( $r = 0.73$ ,  $r = 0.54$ ,  $r = 0.69$ ,  $r = 0.56$ ,  $r = 0.43$ ) and negatively with 2- ( $r = -0.41$ ,  $r = -0.46$ ,  $r = -0.37$ ,  $r = -0.71$ ,  $r = -0.81$ ) and 3-km ( $r = -0.50$ ,  $r = -0.53$ ,  $r = -0.62$ ,  $r = -0.85$ ,  $r = -0.91$ ) TT performance and  $vMLSS$  ( $r = -0.48$ ,  $r = -0.51$ ,  $r = -0.62$ ,  $r = -0.79$ ,  $r = -0.86$ ). Correlations coefficients for 1-km TT were lower compared to 2- and 3-km. Strong agreement was found between threshold concepts ( $vMLSS - vOBLA$ :  $R^2 = 0.94$ ;  $vMLSS - CV$ :  $R^2 = 0.83$ ) and mean differences amounted to  $-0.08$  and  $-0.49 \text{ m}\cdot\text{s}^{-1}$ .

**Conclusion** Parameters linked to aerobic metabolism displayed the strongest relationship with TTs. While anaerobic variables correlated positively with sprint performance the relationships became increasingly negative with increasing distance of TT. It can be hypothesized that influence of anaerobic metabolism is in balance for maximal running efforts around three minutes. Efforts slower than this balance point might tend to benefit from anaerobic metabolism while longer efforts might be affected in a detrimental way. Prediction of TT and threshold velocity was more accurate through performance than physiological parameters. Based on these findings, novel parameters can complement traditional test variables in running. Deliberate and differential selection of test parameters is

advised for performance prediction or physiological training prescription in running and depending on race distance.

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## List of abbreviations

ASR	anaerobic sprint reserve
$BMI_{LBM}$	body mass index calculated with lean body mass
CV	critical velocity
D'	duration prime
$Fat_{max}$	velocity associated with maximal fat oxidation
LBM	lean body mass
MFO	maximal fat oxidation
MLSS	maximal lactate steady-state
OBLA	onset of four $mmol \cdot L^{-1}$ in blood lactate
RE	running economy
RPE	rate of perceived exertion
SRR	sprint reserve ratio
TT	time trial
$t_{100}$	100 m sprint time
$\dot{V}La_{max}$	maximal lactate accumulation rate
$v_{MLSS}$	velocity at MLSS
$v_{OBLA}$	velocity at OBLA
$\dot{V}O_{2max}$	maximal rate of oxygen consumption
$v\dot{V}O_{2max}$	velocity associated with $\dot{V}O_{2max}$
$v_{100}$	mean velocity of 100-m sprint
$\Delta La_{100}$	difference between resting and maximal lactate concentration caused by 100-m sprint

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# 1 Introduction

Running is the most popular outdoor activity in the USA with an estimated 55.9 million participants each year (The Outdoor Foundation, 2018). Similar popularity can be observed globally, with marathon participation rising by nearly 50 percent between 2008 and 2018 (J. J. Andersen, 2021). Frequently running depicts a leisurely activity but is also pursued competitively with the aim of performance improvement. Hence, gaining comprehensive knowledge about the physiology of running, performance determinants and deliberate training prescription can be of particular interest for the general population, competitive athletes or coaches. Determinants of running performance and training prescription have been subject of extensive research in the past decades (Hale, 2008). In this time span, the importance of aerobic fitness for endurance performance, represented in maximal oxygen consumption ( $\dot{V}O_2\text{max}$ ) and running economy (RE), has been underlined and found mutual agreement (Bassett, 2000; Joyner & Coyle, 2008). Parameters targeting oxygen consumption have a high relevance, due to the increasing contribution of the aerobic metabolism as a function of time (Gastin, 2001).

In addition to the above mentioned parameters of the aerobic metabolism, concepts such as lactate or ventilatory threshold have long tradition in endurance performance testing (Cerezuela-Espejo et al., 2018). Three widely accepted methods of threshold determination are onset of blood lactate (OBLA) (Mader et al., 1976), maximal lactate steady- state (MLSS) (Beneke, 2003a) and critical velocity (CV) (Patoz et al., 2021) , although their legitimacy has been subject to recent debate (Jones et al., 2019; Nixon et al., 2021).

Apart from these traditional measures, novel parameters have been proposed and established in endurance performance testing more recently. Research on fat oxidation has emerged around longer endurance events, limited by glycogen availability (Maunder et al., 2018), while parameters connected to the anaerobic metabolism and maximal running speed have been found to be relevant for middle-distance running (Bellinger et al., 2021a; Quittmann, Appelhans, et al., 2020; Sandford et al., 2021). It is the aim of this thesis to investigate prospects and limitations of both traditional and novel concepts for the assessment of running performance.

Experimental data was collected with the intent to answer the following questions:

1. How well do physiological and performance parameters predict 100-m sprint and 1-, 2-, and 3-km time-trial running performance and how do they differ?
2. How can threshold concepts (MLSS, CV and OBLA) be applied for training prescription in running and how are they related.
3. What are the prospects and limitations of traditional and novel diagnostic parameters for monitoring and enhancing endurance performance.

## 2 Methods

### 2.1 Participants

A total of N = 25 well-trained male athletes (age  $25.5 \pm 4.7$  years, height  $181 \pm 6$  cm, body mass  $69.2 \pm 6.4$  kg,  $\dot{V}O_2\text{max}$ :  $66.0 \pm 5.7$  mL·min<sup>-1</sup>·kg<sup>-1</sup>, RE:  $222.0 \pm 11.1$  mL·kg<sup>-1</sup>·km) volunteered to participate in this study. We recruited participants in local sport clubs and at the university. Requirement for participation was a minimum of three training sessions per week. For investigation of athletes with distinguished metabolic profiles and heterogenic endurance performance level, participants with backgrounds in sprint (SP; 200 to 400 m; n = 6), middle- and long-distance (MD-LD; 800 m to 21.1 km; n = 16), and (ultra-)marathon running (M-UM; 42.2 km and above; n = 3). Participants with protocolled training (n = 22) reported mean weekly training time and running distance in the 12 weeks leading up to the study of  $6.31 \pm 3.47$  h and  $39.55 \pm 20.26$  km, respectively.

**Table 1** Anthropometric data displayed for all participants and as sub-cohorts of sprinters (SP), middle- and long-distance runners (MD-LD) and (ultra-)marathon runners (M-UM). BMI<sub>LBM</sub> represents body mass index (BMI) calculated with lean body mass (LBM). Values are reported as mean and standard deviation (SD).

		body height	body mass	body fat	lean body mass	BMI <sub>LBM</sub>
		[cm]	[kg]	[%]	[kg]	[kg·(m <sup>2</sup> ) <sup>-1</sup> ]
<b>SP</b> (n = 6)	mean	180,62	74,22	11,31%	65,14	19,95
	SD	5,27	7,34	2,19%	5,36	1,07
<b>MD-LD</b> (n = 16)	mean	180,23	66,71	10,94%	59,36	18,29
	SD	6,29	4,76	2,18%	3,74	0,93
<b>M-UM</b> (n = 3)	mean	182,03	72,57	11,73%	64,05	19,30
	SD	2,63	6,63	1,72%	5,89	1,25
<b>Total</b> (n = 25)	mean	180,54	69,21	11,31%	61,31	18,81
	SD	5,61	6,40	2,19%	4,97	1,20

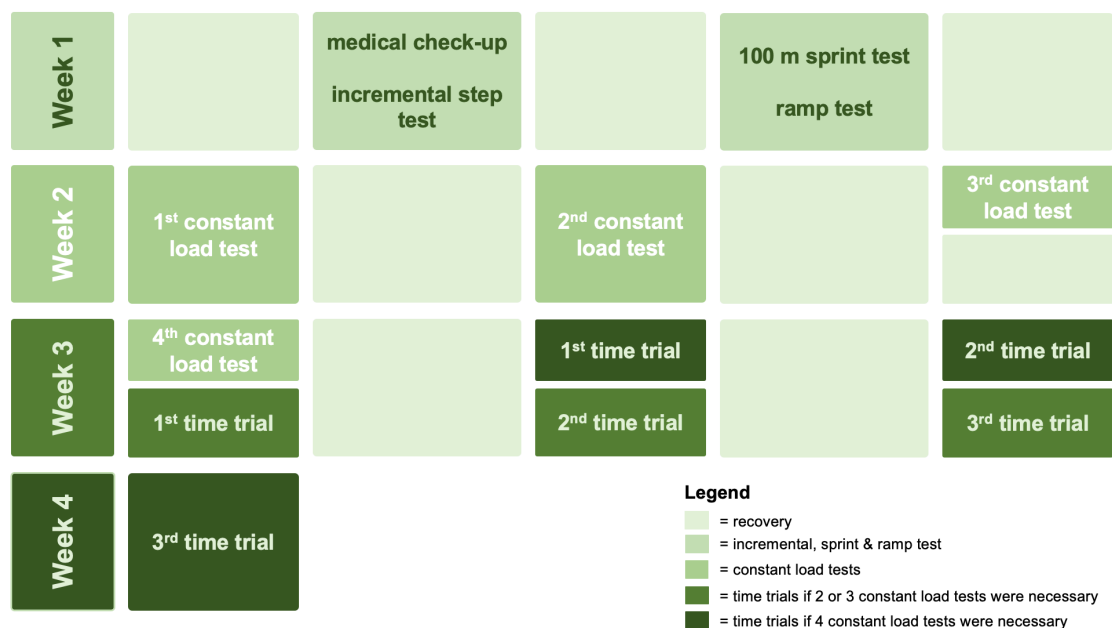
A medical check-up based on the guidelines of the European Society of Cardiology, was carried out for each participating athlete before commencement of any physical activity related to the study. The check-up included notation of medical, family and personal history, a physical examination and a resting electrocardiogram (Corrado et al., 2005). Only participants without positive findings were included. Due to the ongoing COVID-19 pandemic, precautions were made for the safety of participants and staff (Nieß et al., 2020). This included clarification of a given suspected disease, temperature measurements via forehead thermometer and proof of full vaccination or negative COVID test (PCR

or antigen) before every visit to the facilities. Additionally, closed spaces were well ventilated, distance regulations (2 m) kept when possible, surfaces disinfected after each participant and staff was required to wear FFP2-masks at all times. All procedures received institutional ethics approval (No. 092/2021) according to the Declaration of Helsinki. Before the investigation, participants were personally informed about the aims, procedures and potential risks of this study, and gave their written consent.

## 2.2 Experimental design

### 2.2.1 Testing procedure and equipment

Measurements were performed from March to October 2021. The participants performed various exercise tests to determine a profound metabolic profile and measure middle-distance time trial performances over the course of approximately three weeks (Figure 1).



**Figure 1** Schematic illustration of experimental design and procedures of individual participants

Between each test, a minimum of 48 h was provided to minimize fatigue effects. On their first visit to the laboratory, each athlete was informed about the procedures, received the medical check-up and underwent a ten-site skinfold thickness measurement (Harpenden Skinfold Caliper, Baty Int., West Sussex, United Kingdom) to determine body fat percentage (Parízková & Bůžková, 1971).



Afterwards, the participants performed an incremental step test on a motorized treadmill (saturn 300/100, h/p/cosmos sports & medical GmbH, Nussdorf-Traunstein, Germany) until the participants' lactate concentration exceeded the onset of four  $\text{mmol}\cdot\text{l}^{-1}$  blood lactate (OBLA). Two days later, a 100-m all-out sprint test was performed on a straight 100-m indoor track, followed by a ramp test until subjective exhaustion on a treadmill approximately one hour later.

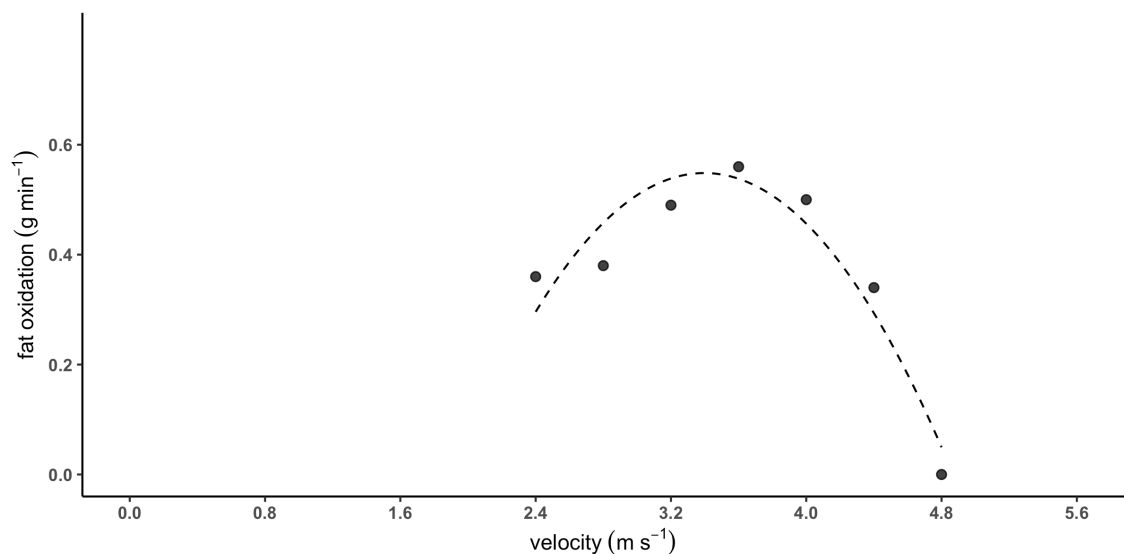
In the second test week, several constant load tests (with a resting period of 48 h between tests) were performed on a treadmill to determine maximal lactate steady-state (MLSS). During the third and final test week, the participants performed time trials (TT) over 1, 2, and 3 km in a randomized order on an outdoor track to determine their middle-distance TT-performance.

All participants were instructed to refrain from caffeinated beverages for at least 8 h before testing and to avoid any vigorous physical activity on the testing day and the day before. For gas exchange analyses in the laboratory, participants had to arrive in a well-fed state, but with a fasting period of at least two hours. To avoid influences based on circadian rhythm, the participants performed the laboratory and field tests at approximately the same time of the day. Testing in the laboratory was performed with a constant treadmill gradient of 1% (Jones & Doust, 1996). The participants wore a safety harness, which was connected to the automatic security brake system of the treadmill. During all trials on the treadmill, participants ran with open windows and a fan for cooling. Heart rate was monitored during all tests by a heart rate monitor (HRM Tri, Garmin International, Inc., Olathe, KS, United States of America).

### *2.2.2 Incremental step test*

The incremental step test started with an initial velocity of  $2.0 \text{ m}\cdot\text{s}^{-1}$  ( $7.2 \text{ km}\cdot\text{h}^{-1}$  or  $8:20 \text{ min}\cdot\text{km}^{-1}$ ), which increased by  $0.4 \text{ m}\cdot\text{s}^{-1}$  ( $1.44 \text{ km}\cdot\text{h}^{-1}$ ) every five minutes. At the end of every step, the treadmill stopped for 30 seconds in which ratings of perceived exertion (Borg, 1982) were noted and a blood sample ( $20 \mu\text{l}$ ) was collected from the earlobe to determine lactate concentration immediately (Biosen C-Line, EKF-diagnostic GmbH, Barleben, Germany). The incremental step test was terminated when OBLA was reached.

Throughout the incremental test, participants wore an airtight silicone oro-nasal mask (7450 Series, V2™, Hans-Rudolph, Inc., Shawnee, KS, United States of America) and breath-by-breath oxygen consumption and expired carbon dioxide was measured by a spirometric device (ZAN 600 USB, nSpire Health, Inc., Longmont, CO, United States of America). Flow sensors were calibrated manually by using a standardized 3000 mL high precision syringe (nSpire Health, Inc., Longmont, CO, United States of America). Gas concentration was calibrated under laboratory conditions as well as a gas mixture of 15% O<sub>2</sub> and 6% CO<sub>2</sub>. Oxygen consumption, expired carbon dioxide, heart rate, carbohydrate consumption and fat oxidation measures were based on the average during the last two min of every five-minute step. Values of substrate utilization were calculated as previously recommended (Jeukendrup & Wallis, 2005) and maximum fat oxidation (MFO) and velocity associated with MFO (Fat<sub>max</sub>) were calculated from polynomial fitted data.



**Figure 2** Example of polynomial fitting of fat oxidation curve from incremental step test. Black dots represent results from each velocity increment and dashed line displays polynomial fitted curve.

### 2.2.3 100-m all-out sprint test

The participants performed the 100-m all-out sprint test and the standardized warm-up of 15 minutes including technical drills and starts as described previously (Quittmann, Appelhans, et al., 2020). Throughout the sprints, participants were verbally encouraged by the examiners. The time to cover the 100 meters ( $t_{100}$ ) was determined using a start pedal and a double infrared photoelectric light barrier (Sportronic Electronic Sports Equipment, Winnenden-

Herthmannsweiler, Germany). Blood samples were collected at the participants' arrival at the test site, at the end of the warm-up, immediately before and after the sprint test, as well as every minute after the sprint for 10 minutes.

Maximal lactate accumulation rate ( $\dot{V}La_{max}$ ) was calculated as the difference between the measured maximal post-exercise lactate concentration and resting lactate concentration ( $\Delta La_{100}$ ), divided by the difference between  $t_{100}$  and the period at the beginning of exercise for which no lactate formation is assumed ( $t_{alac}$ ) (Eq. 1) (Heck et al., 2003; Mader, 1996; Quittmann, Schwarz, et al., 2020).

$$\dot{V}La_{max} = \frac{La_{max} - La_{rest}}{t_{100} - t_{alac}} \quad (1)$$

As a representation of phosphocreatine metabolism,  $t_{alac}$  was interpolated according to previous research (Eq. 2) (Heck et al., 2003; Quittmann, Appelhans, et al., 2020)

$$t_{alac} = t_{100} \cdot 0.0909 + 2.0455 \quad (2)$$

After the last blood sample was collected, participants performed an individual cool-down for 10-min at a self-determined intensity and had to arrive at the laboratory approximately 45 min afterwards.

#### 2.2.4 Ramp test

As a warm-up preceding the ramp test protocol, the participants performed eight minutes at  $2.8 \text{ m}\cdot\text{s}^{-1}$  ( $10.08 \text{ km}\cdot\text{h}^{-1}$  or  $5:57 \text{ min}\cdot\text{km}^{-1}$ ) without spirometric measurement. After a short break for attaching the mask, the ramp test protocol started with an initial velocity of  $2.8 \text{ m}\cdot\text{s}^{-1}$  for another 2 minutes. Afterwards, velocity increased by  $0.15 \text{ m}\cdot\text{s}^{-1}$  ( $0.54 \text{ km}\cdot\text{h}^{-1}$ ) every 30 seconds until volitional exhaustion of the participants.  $\dot{V}O_{2max}$  was determined by the highest 30 s rolling average of breath-by-breath data and corresponding velocity ( $v\dot{V}O_{2max}$ ) as the velocity increment in which the highest average occurred. Anaerobic speed reserve (ASR) and sprint reserve ratio (SRR) were calculated as the difference between the average velocity during the 100-m all-out sprint ( $v_{100}$ ) and  $v\dot{V}O_{2max}$  as well as the ratio of  $v_{100}$  and  $v\dot{V}O_{2max}$  (Sandford, Allen, et al., 2019). Blood lactate concentration was determined before and after performing the warm-up

as well as immediately after the ramp test protocol ( $L_{RT}$ ). For valid detection of  $\dot{V}O_{2max}$  at least one of the criteria described by [Howley et al., 1995](#) needed to present apart from volitional exhaustion. The criteria include the presence of a plateau of  $\leq 150 \text{ mL}\cdot\text{min}^{-1}$ , RPE  $\geq 19$ , respiratory exchange ratio of  $\geq 1.05$  and a post-exercise lactate concentration of  $\geq 8 \text{ mmol}\cdot\text{L}^{-1}$ .

### 2.2.5 Constant load tests

The intensity for the first constant load test was determined as the velocity according to OBLA with an accuracy of  $0.1 \text{ m}\cdot\text{s}^{-1}$ . The protocol started with a five-minute warm-up at 50% of the target velocity. Afterwards, the participants ran at target velocity for 30 min which were interspersed every five minutes by 30 s resting periods for collection of blood samples. Gas exchange analyses were performed throughout the test. The criterion of lactate steady-state was defined as an increase in lactate concentration during the final 20 min of no more than  $1 \text{ mmol}\cdot\text{L}^{-1}$  (Beneke, 2003b). If the lactate concentration stayed within the criterion, target velocity in the following trial increased by  $0.1 \text{ m}\cdot\text{s}^{-1}$ . In the case of exceeding the criterion, target velocity in the following trial decreased by  $0.1 \text{ m}\cdot\text{s}^{-1}$ . If a participant was unable to finish a trial due to premature fatigue, the intensity for the next test decreased by  $0.2 \text{ m}\cdot\text{s}^{-1}$ . vMLSS was defined as the highest velocity at which the steady-state criterion was met. RE and  $\% \dot{V}O_{2max}$  were based on the mean  $\dot{V}O_2$  during the final 20 min of the constant load trials. From here on, the abbreviations RE and  $\% \dot{V}O_{2max}$  describes RE or  $\% \dot{V}O_{2max}$  associated with MLSS or are used to discuss these parameters in general terms.

### 2.2.6 Time trials

Additionally, a subcohort ( $n = 20$ ) completed TT over 1, 2, and 3 km in a randomized order and with about 48 h separating the trials. A standardized warm-up of about 20 min preceded each trial. The participants started with an easy jog for 10 min at a self-determined intensity on the 400-m track. Afterwards, the participants performed various technical drills for approximately 7 to 10 minutes, followed by four controlled accelerations of 50 meters. Before performing the trials, participants had a passive recovery of about 5 minutes. The participants completed TT without other participants on the track and were instructed to achieve the best time possible over the respective distance and received verbal

encouragement as well as feedback regarding the remaining distance and split times. Split times were hand-stopped every 100 m by using standardized optical reference markers (weighted 1.5 m poles) placed close to the respective markings on the track. Lactate concentration was determined before and after the warm-up, immediately before starting and after TT completion, as well as three and six minutes post-performance.

For each athlete critical velocity (CV) and distance prime (D') were calculated from the distance-duration and respective velocity-distance relationship established by the three TT. CV represents the asymptote of velocity-distance relationship, while D' describes the curvature constant. Transferred to running performance, CV represents a maximum sustainable velocity over longer running duration and D' the amount of energy that can be expended above CV (Monod & Scherrer, 1965; Patoz et al., 2021).

### 2.3 Data processing

Raw data was collected in Excel (version 16.59). Further processing and statistical analysis were carried out with RStudio (version 1.4.1717). Complete access to data processing and statistical analysis is available via GitHub (<https://github.com/yannickmaxschwarz/SimProRun>)

### 2.4 Statistical analysis

Descriptive data is presented as mean  $\pm$  SD. Normality of data was assessed visually by independent and blinded reviewers rating Q-Q plots of residuals prior to bivariate correlation calculation. In the case of normality Pearson's correlations coefficients and Fisher-z confidence intervals were reported. If normality was denied Spearman's coefficients and confidence intervals with corrected standard error according to Fieller were calculated. Results were considered significant if calculated alpha error was below five percent ( $p \leq 0.05$ ). Subsequently significance is also reached if zero is not included within confidence intervals (95%). Correlations are interpreted as "weak" ( $r = 0.1 - 0.3$ ), "moderate" ( $r = 0.3 - 0.5$ ), "strong" ( $r = 0.5 - 0.7$ ), "very high" ( $r = 0.7 - 1.0$ ). No correlation was assumed below coefficients of 0.1 (Hopkins et al., 2009).

For forward stepwise multiple regression analysis, variables were entered into the model by order of their change in  $R^2$  and if there was a significant ( $p \leq 0.05$ ) change in Akaike information criterion (AIC).

## 3 Results

### 3.1 Descriptive results

#### 3.1.1 Physiological parameters

Descriptive results of physiological parameters measured during, incremental step test, sprint test, ramp test and constant load tests are displayed for all athlete groups and the entire study cohort in Table 2. Group differences were not tested for significance due to insufficient sample size of groups. Mean  $\dot{V}O_2\text{max}$  from ramp test amounted to  $66.00 \pm 4.21 \text{ mL}\cdot\text{kg}^{-1}\cdot\text{km}^{-1}$ . Incremental step test results for RE and MFO averaged to  $222.99 \pm 11.08 \text{ mL}\cdot\text{kg}^{-1}\cdot\text{km}^{-1}$  and  $0.45 \pm 0.11 \text{ g}\cdot\text{s}^{-1}$ . Average  $\dot{V}La_{\text{max}}$  and  $\Delta La_{100}$  of  $0.73 \pm 0.18 \text{ mmol}\cdot\text{L}^{-1}\cdot\text{s}^{-1}$  and  $7.01 \pm 1.26 \text{ mmol}\cdot\text{L}^{-1}$  were reached during the sprint test and mean result of  $\% \dot{V}O_2\text{max}$  was  $83.35 \pm 4.46$  percent.

**Table 2** Physiological parameters including  $\dot{V}O_2\text{max}$ , RE,  $\% \dot{V}O_2\text{max}$ , MFO,  $\dot{V}La_{\text{max}}$ , and  $\Delta La_{100}$ , displayed for all investigated participants and respective of athlete groups.

		$\dot{V}O_2\text{max}$	RE	$\% \dot{V}O_2\text{max}$	MFO	$\dot{V}La_{\text{max}}$	$\Delta La_{100}$
		[ $\text{mL}\cdot\text{min}^{-1}\cdot\text{kg}^{-1}$ ]	[ $\text{mL}\cdot\text{kg}^{-1}\cdot\text{km}^{-1}$ ]	[%]	[ $\text{g}\cdot\text{min}^{-1}$ ]	[ $\text{mmol}\cdot\text{L}^{-1}\cdot\text{s}^{-1}$ ]	[ $\text{mmol}\cdot\text{L}^{-1}$ ]
<b>SP</b> (n = 6)	mean	61.16	233.91	81.12	0.41	0.98	8.45
	SD	3.68	7.85	5.44	0.07	0.12	0.86
<b>MD-LD</b> (n = 16)	mean	67.51	217.34	83.60	0.45	0.66	6.49
	SD	5.67	8.59	3.40	0.12	0.11	0.95
<b>M-UM</b> (n = 3)	mean	67.65	222.90	86.49	0.51	0.65	6.93
	SD	4.83	12.84	6.87	0.12	0.12	1.44
<b>Total</b> (n = 25)	mean	66.00	221.99	83.35	0.45	0.73	7.01
	SD	5.71	11.08	4.46	0.11	0.18	1.26

#### 3.1.2 Performance parameters

Results of performance parameters are listed in Table 3 and 4. Performance parameters we defined as variables representing actual performance ( $v_{100}$ ), velocities associated with a physiological phenomenon ( $v\dot{V}O_2\text{max}$ ,  $Fat_{\text{max}}$ ,  $MLSS$ ) or measures calculated from other performance variables ( $ASR$ ,  $SRR$ ,  $CV$ ,  $D'$ ). Furthermore, the term “performance” will be used synonymously for mean velocity hereafter. Due to calculation from TT, results for  $CV$  and  $D'$  have a lower sample size (indicated in Table 3) than parameters not derived from TT.

Mean  $v_{100}$ ,  $v\dot{V}O_2\text{max}$ ,  $vMLSS$  and  $Fat_{\text{max}}$  were measured at  $7.78 \pm 0.56 \text{ m}\cdot\text{s}^{-1}$ ,  $5.49 \pm 0.42 \text{ m}\cdot\text{s}^{-1}$ ,  $4.14 \pm 0.50 \text{ m}\cdot\text{s}^{-1}$  and  $3.04 \pm 0.45 \text{ m}\cdot\text{s}^{-1}$ .  $CV$  and  $D'$  derived

from time trial distance-duration relationship averaged at  $4.67 \pm 0.41 \text{ m}\cdot\text{s}^{-1}$  and  $203.7 \pm 62.98$  meters.

**Table 3** Performance parameters including vMLSS, CV, Fatmax, D', for all investigated participants and respective of athlete groups. \*CV and D' are only reported for athletes who participated in TT. Respective reduction in sample size is displayed for each group (n = all athletes/ TT athletes)

		<b>vMLSS</b>	<b>CV *</b>	<b>Fat<sub>max</sub></b>	<b>D' *</b>
		[m·s <sup>-1</sup> ]	[m·s <sup>-1</sup> ]	[m·s <sup>-1</sup> ]	[m]
<b>SP</b> (n = 6 / 4*)	mean	3.53	4.15	2.43	285.84
	SD	0.27	0.15	0.34	46.14
<b>MD-LD</b> (n = 16 / 15*)	mean	4.33	4.78	3.23	186.68
	SD	0.41	0.37	0.28	52.47
<b>M-UM</b> (n = 3 / 1*)	mean	4.37	4.95	3.24	167.08
	SD	0.21	0.21	0.23	19.52
<b>Total</b> (n = 25 / 20)	mean	4.14	4.67	3.04	203.70
	SD	0.50	0.41	0.45	62.98

Average ASR and SRR were calculated to be  $2.29 \pm 0.77 \text{ m}\cdot\text{s}^{-1}$  and  $1.43 \pm 0.17$ .

**Table 4** Performance parameters including velocities associated with  $\dot{V}O_{2\text{max}}$ , 100 m sprint, anaerobic speed reserve, as well as sprint reserve ratio displayed for all investigated participants and respective of athlete groups.

		<b>v<math>\dot{V}O_{2\text{max}}</math></b>	<b>v100</b>	<b>ASR</b>	<b>SRR</b>
		[m·s <sup>-1</sup> ]	[m·s <sup>-1</sup> ]	[m·s <sup>-1</sup> ]	
<b>SP</b> (n = 6)	mean	5.10	8.54	3.44	1.68
	SD	0.29	0.33	0.45	0.12
<b>MD-LD</b> (n = 16)	mean	5.65	7.62	1.97	1.35
	SD	0.36	0.36	0.37	0.08
<b>M-UM</b> (n = 3)	mean	5.45	7.17	1.72	1.32
	SD	0.53	0.20	0.55	0.13
<b>Total</b> (n = 25)	mean	5.49	7.78	2.29	1.43
	SD	0.42	0.56	0.77	0.17

Mean finishing times for 100 m sprint and 1000, 2000 and 3000 m TT were  $12.91 \pm 0.91$ ,  $173.0 \pm 10.6$ ,  $383.9 \pm 24.7$  and  $606.0 \pm 45.5$  s respectively for athletes who participated in TT.

## 3.2 Correlation analysis

### 3.2.1 Relationships between anthropometric and physiological parameters

Influence of anthropometric parameters on key physiological parameters were examined via correlation analysis (Table 5). Absolute  $\dot{V}O_{2\text{max}}$  correlated



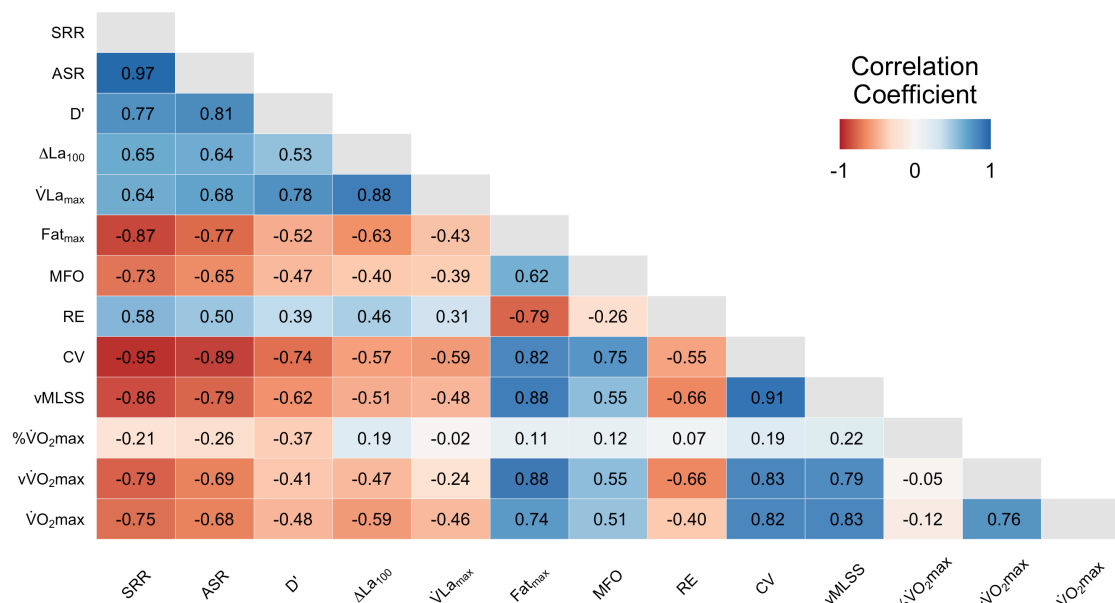
significantly with weight ( $R^2 = 0.57$ ;  $p < 0.01$ ) and height ( $R^2 = 0.50$ ;  $p = 0.03$ ). Furthermore, a significant correlation was found between BMI calculated with lean body mass ( $BMI_{LBM}$ ) and RE ( $R^2 = 0.73$ ;  $p < 0.001$ ), as well as  $\% \dot{V}O_{2max}$  ( $R^2 = 0.41$ ;  $p = 0.04$ ). No relationship was found between anthropometric variables and MFO or  $\dot{V}La_{max}$ .

**Table 5** Relationship between anthropometric and physiological parameters listed as correlation coefficients and respective p-value

anthropometric parameters		$\dot{V}O_{2max}$	RE	$\% \dot{V}O_{2max}$	MFO	$\dot{V}La_{max}$
		[mL·min <sup>-1</sup> ]	[mL·kg <sup>-1</sup> ·km <sup>-1</sup> ]	[%]	[g·min <sup>-1</sup> ]	[mmol·L <sup>-1</sup> ·s <sup>-1</sup> ]
weight [kg]	r	0.57	0.20	0.41	-0.04	0.36
	p	< 0.01	0.40	0.07	0.88	0.12
height [cm]	r	0.50	-0.38	-0.15	-0.07	0.28
	p	0.03	0.10	0.53	0.77	0.23
body fat [%]	r	0.07	0.20	0.12	-0.15	0.06
	p	0.77	0.41	0.62	0.52	0.81
$BMI_{LBM}$	r	0.18	0.73	0.48	-0.06	0.17
	p	0.44	< 0.001	0.03	0.81	0.46

### 3.2.2 Relationships between physiological and performance parameters

Relationship between all individual physiological and performance parameters were investigated by means of correlation analysis and are displayed as a matrix (Figure 3). Positive correlations were found between various parameters influenced by the anaerobic metabolism, such as ASR, SRR, D',  $\dot{V}La_{max}$ ,  $\Delta La_{100}$ . These parameters correlated negatively with aerobically determined performance parameters such as  $v\dot{V}O_{2max}$ ,  $vMLSS$ , CV and  $Fat_{max}$ , as well as aerobically determined physiological parameters including  $\dot{V}O_{2max}$  and MFO. Threshold concepts such as  $vMLSS$  and CV correlated positively between each other (Figure 7) and with other performance and physiological variables such as  $v\dot{V}O_{2max}$ ,  $Fat_{max}$ ,  $\dot{V}O_{2max}$  and MFO. No statistically significant relationships were observed between  $\% \dot{V}O_{2max}$  and any variables.



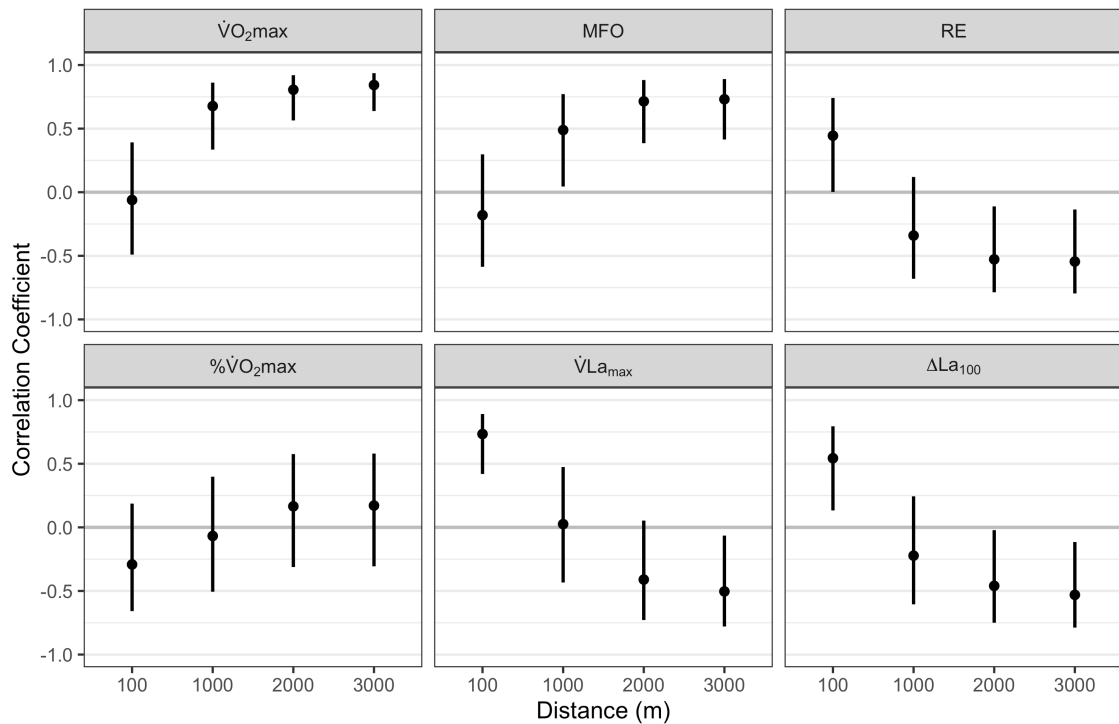
**Figure 3** Correlation matrix displaying correlation coefficients for all investigated parameters. \*Participants without TT completion were excluded in this analysis resulting in a reduced sample size ( $n = 20$ ).

$\dot{V}La_{max}$  only showed significant correlations with  $\dot{V}O_{2max}$  ( $p = 0.04$ ), vMLSS ( $p = 0.03$ ), CV ( $p < 0.01$ ), ASR ( $p < 0.01$ ), SRR ( $p < 0.01$ ) and D' ( $p < 0.001$ ). All other correlations were statistically significant except relationships between MFO and RE ( $p = 0.27$ ), MFO and  $\Delta La_{100}$  ( $p = 0.08$ ), as well as D' with  $v\dot{V}O_{2max}$  ( $p = 0.07$ ) and RE ( $p = 0.29$ ).

### 3.2.3 Relationships between time-trial performance and physiological parameters

Correlations coefficients and confidence intervals of physiological variables with TT and sprint performance, are presented in Figure 4.  $\dot{V}O_{2max}$  demonstrated the strongest relationship with performance for all TT distances from 1000 to 3000 m ( $r = 0.68$ ,  $p = 0.001$ ;  $r = 0.81$ ,  $p < 0.001$ ;  $r = 0.84$ ,  $p < 0.001$ ), while no significant correlation was found with sprint performance ( $r = -0.06$ ,  $p = 0.80$ ). Congruent with  $\dot{V}O_{2max}$ , correlation coefficients of MFO increased with rising TT distance ( $r = 0.49$ ,  $p = 0.03$ ;  $r = 0.71$ ,  $p < 0.001$ ;  $r = 0.73$ ,  $p < 0.001$ ). Lower RE, expressed in oxygen consumption, resulted in significantly faster mean velocity for 2000 and 3000-m ( $r = -0.53$ ,  $p = 0.017$ ;  $r = -0.55$ ,  $p = 0.013$ ) but only trended in this direction for 1000-m ( $r = -0.34$ ,  $p = 0.14$ ). Additionally, a trend of higher RE in participants with faster sprint times could be observed ( $r = 0.44$ ,  $p = 0.05$ ).  $\dot{V}La_{max}$  was the only physiological variable significantly correlated with 100-m performance ( $r = 0.73$ ,  $p < 0.001$ ). In terms of TT performance an increasingly negative influence of  $\dot{V}La_{max}$  can be observed, but level of significance was only reached for 3000-

m ( $r = 0.03$ ,  $p = 0.92$ ;  $r = -0.41$ ,  $p = 0.07$ ;  $r = -0.50$ ,  $p = 0.03$ ).  $\Delta La_{100}$  produced similar results  $\dot{V}La_{max}$  in terms of sprint ( $r = 0.54$ ,  $p = 0.01$ ) and TT performance ( $r = -0.22$ ,  $p = 0.35$ ;  $r = -0.46$ ,  $p = 0.04$ ;  $r = -0.53$ ,  $p = 0.02$ ). No significant relationship can be reported for  $\% \dot{V}O_{2max}$  and performance over any investigated distances.

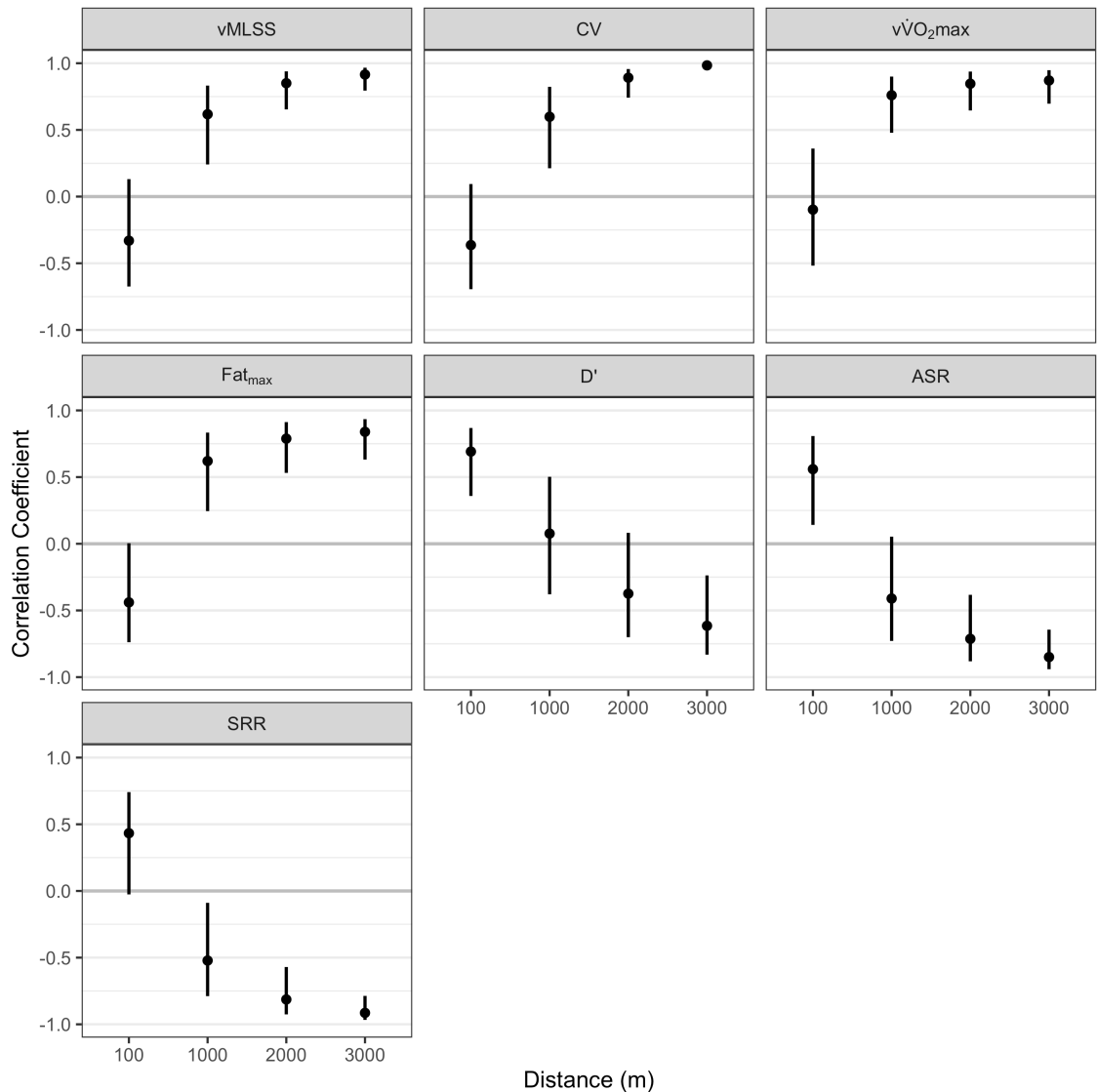


**Figure 4** Correlation coefficients of physiological parameters and 100 m sprint and 1, 2, and 3 km TTs are displayed as solid dots and bars indicating respective confidence intervals. Intersection of confidence intervals with zero corresponds to p-values exceeding 0.05.

### 3.2.4 Relationships between time-trial performance and performance parameters

Correlations coefficients and confidence intervals of performance variables with TT and sprint performance, are presented in Figure 5. Strong to very strong relationships of performance from 1 to 3 km were observed with  $vMLSS$  ( $r = 0.62$ ,  $p < 0.01$ ;  $r = 0.85$ ,  $p < 0.0001$ ;  $r = 0.92$ ,  $p < 0.0001$ ),  $CV$  ( $r = 0.60$ ,  $p < 0.01$ ;  $r = 0.89$ ,  $p < 0.0001$ ;  $r = 0.98$ ,  $p < 0.0001$ ),  $v\dot{V}O_{2max}$  ( $r = 0.76$ ,  $p < 0.01$ ;  $r = 0.85$ ,  $p < 0.0001$ ;  $r = 0.87$ ,  $p < 0.0001$ ) and  $Fat_{max}$  ( $r = 0.62$ ,  $p < 0.01$ ;  $r = 0.79$ ,  $p < 0.0001$ ;  $r = 0.84$ ,  $p < 0.0001$ ). No significant correlations were found between sprint performance and  $vMLSS$ ,  $CV$ ,  $v\dot{V}O_{2max}$  or  $Fat_{max}$ . In contrast, significant positive relationships were found between  $v_{100}$  and  $D'$  ( $r = 0.69$ ,  $p < 0.001$ ),  $ASR$  ( $r = 0.56$ ,  $p = 0.01$ ). A continuously negative trend was observed for correlations coefficients from 1- to 3-km TT performance and  $D'$  ( $r = 0.08$ ,  $p = 0.75$ ;  $r = -0.37$ ,

$p = 0,10$ ;  $r = -0.62$ ,  $p < 0.01$ ), ASR ( $r = -0.41$ ,  $p = 0.07$ ;  $r = -0.71$ ,  $p < 0,001$ ;  $r = -0.85$ ,  $p < 0.0001$ ) and SRR ( $r = -0.52$ ,  $p = 0.02$ ;  $r = -0.81$ ,  $p < 0.0001$ ;  $r = -0.91$ ,  $p < 0.0001$ ). Mean velocity in 1-km TT correlated positively with 2- and 3-km TT velocity ( $r = 0.85$ ,  $p < 0.0001$ ;  $r = -0.73$ ,  $p < 0.001$ ) and 2-km with mean 3-km velocity ( $r = 0.95$ ,  $p < 0.0001$ ).



**Figure 5** Correlation coefficients of performance parameters and 100 m sprint and 1, 2, and 3 km TTs are displayed as solid dots and bars indicating respective confidence intervals. Intersection of confidence intervals with zero corresponds to p-values exceeding 0.05.

### 3.3 Stepwise regression analysis

#### 3.3.1 Stepwise regression analysis of time-trial performance and physiological parameters

Stepwise regression was conducted to find the best prediction for sprint and TT performances with multiple physiological parameters including  $\dot{V}O_{2\max}$ , RE, %  $\dot{V}O_{2\max}$ , MFO and  $\dot{V}La_{\max}$  (Table 6). Calculations revealed, performance predictions with least amount of remaining variance ( $R^2 = 0.93$ ) and lowest residual standard error ( $0.11 \text{ m}\cdot\text{s}^{-1}$ ) can be made for the 3000-m TT. Coefficient of determination after stepwise regression was successively lower for 2000 m ( $R^2 = 0.85$ ) and 1000 m TT ( $R^2 = 0.62$ ).  $\dot{V}O_{2\max}$  represented the best single linear model over all TT distances ( $R^2 = 0.46$ ;  $0.65$ ;  $0.71$ ) and was complemented by MFO and RE for further explanation of variance in performance over 2000 and 3000 m.  $\dot{V}La_{\max}$  substantially improved predictions of 1000 m velocity ( $\Delta R^2 = 0.07$ ), when it was added to the single variable model. It also accounts for the greatest variance in 100-m performance as a single predictor ( $R^2 = 0.60$ ). All results were highly significant with alpha error results of 0.004 or lower.

**Table 6** Forward stepwise regression models of physiological parameters for sprint and TT velocity are displayed including coefficient of determination ( $R^2$ ), change of  $R^2$  and relation to inferior model ( $\Delta R^2$ ), residual standard error ( $\text{m}\cdot\text{s}^{-1}$ ), probability of alpha error ( $p$ ). Akaike's Information Criterion (AIC) was used for successive selection of added variables.

Time-trial	Model	$R^2$	$\Delta R^2$	Resid. Std. Error	$p$	AIC
<b>100 m</b>	$\dot{V}La_{\max}$	0.60		0.31	< 0.0001	-45.22
	$\dot{V}La_{\max} + \Delta La_{100}$	0.97	0.12	0.08	< 0.0001	-96.96
<b>1000 m</b>	$\dot{V}O_{2\max}$	0.46		0.26	0.001	-52.56v
	$\dot{V}O_{2\max} + \dot{V}La_{\max}$	0.53	0.07	0.25	0.003	-53.39
	$\dot{V}O_{2\max} + \dot{V}La_{\max} + \text{MFO}$	0.58	0.05	0.24	0.004	-53.52
	$\dot{V}O_{2\max} + \dot{V}La_{\max} + \text{MFO} + \text{RE}_{\text{MLSS}}$	0.62	0.05	0.23	0.004	-53.88
<b>2000 m</b>	$\dot{V}O_{2\max}$	0.65		0.20	< 0.0001	-61.86
	$\dot{V}O_{2\max} + \text{MFO}$	0.78	0.13	0.16	< 0.0001	-69.55
	$\dot{V}O_{2\max} + \text{MFO} + \text{RE}_{\text{MLSS}}$	0.83	0.05	0.15	< 0.0001	-72.36
	$\dot{V}O_{2\max} + \text{MFO} + \text{RE}_{\text{MLSS}} + \dot{V}La_{\max}$	0.85	0.02	0.14	< 0.0001	-73.28
<b>3000 m</b>	$\dot{V}O_{2\max}$	0.71		0.21	< 0.0001	-61.32
	$\dot{V}O_{2\max} + \text{MFO}$	0.83	0.12	0.16	< 0.0001	-70.53
	$\dot{V}O_{2\max} + \text{MFO} + \text{RE}_{\text{MLSS}}$	0.88	0.05	0.14	< 0.0001	-75.33
	$\dot{V}O_{2\max} + \text{MFO} + \text{RE}_{\text{MLSS}} + \% \dot{V}O_{2\max}$	0.93	0.05	0.11	< 0.0001	-83.00

### 3.4.2 Stepwise regression analysis of time-trial performance and performance parameters

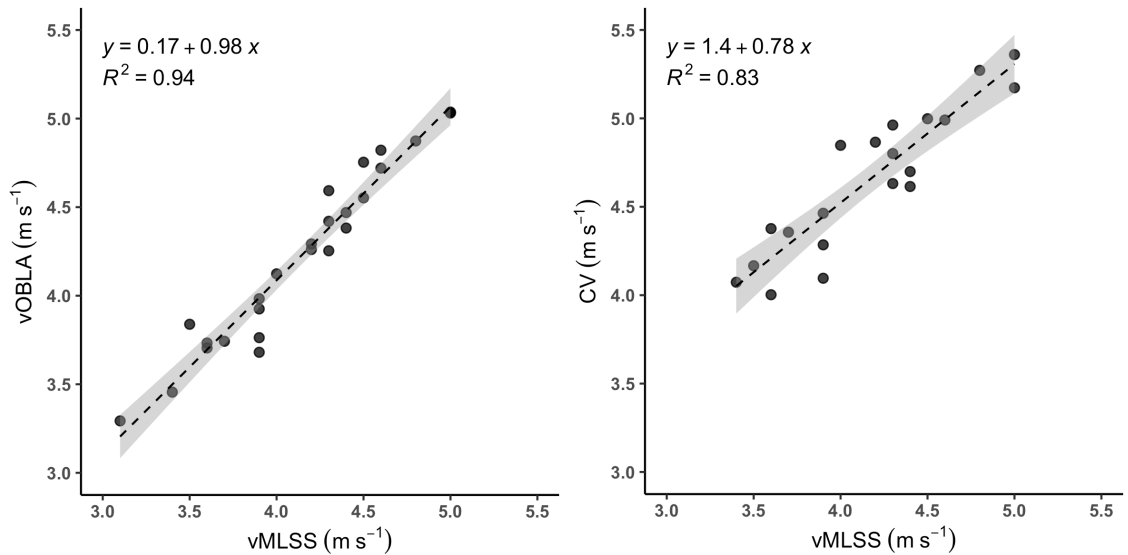
CV, vMLSS,  $\dot{V}O_{2\max}$ ,  $Fat_{\max}$ ,  $D'$ , ASR, SRR, v100 were included in stepwise regression of TT performance and performance parameters (Table 7). In the case of 100 m performance, ASR, SRR, and v100 were excluded from the model due their calculation on the basis of v100. Near perfect predictions of mean velocity for 2000 and 3000 m by means of stepwise regression were reached with CV and  $D'$  ( $R^2 = 0.98$ ; 1.00).  $\dot{V}O_{2\max}$  represented the single best predictor of 1000 m performance ( $R^2 = 0.58$ ) out of all performance parameters. While TT reached near perfect coefficients of determination, 100 m performance did not reach comparable accuracy of prediction. Sprint performance was best predicted by  $D'$  ( $R^2 = 0.48$ ) with single and by  $D'$  and CV ( $R^2 = 0.53$ ) with stepwise regression analysis. All results were highly significant with alpha error results of 0.002 or lower.

**Table 7** Forward stepwise regression models of performance parameters for sprint and TT velocity are displayed including coefficient of determination ( $R^2$ ), change of  $R^2$  and relation to inferior model ( $\Delta R^2$ ), residual standard error ( $m \cdot s^{-1}$ ), probability of alpha error ( $p$ ). Akaike's Information Criterion (AIC) was used for successive selection of added variables.

Time-trial	Model	$R^2$	$\Delta R^2_{adj}$	Resid. Std. Error	$p$	AIC
100 m	$D'$	0.48		0.35	< 0.001	-39.85
	$D' + CV$	0.53	0.05	0.34	0.002	-39.88
1000 m	$\dot{V}O_{2\max}$	0.58		0.23	0.0001	-57.53
	$\dot{V}O_{2\max} + D'$	0.76	0.18	0.18	< 0.0001	-66.86
	$\dot{V}O_{2\max} + D' + CV$	0.97	0.21	0.06	< 0.0001	-106.84
2000 m	CV	0.80		0.16	< 0.0001	-72.60
	CV + $D'$	0.98	0.18	0.05	< 0.0001	-118.70
3000 m	CV	0.97		0.06	< 0.0001	-105.55
	CV + $D'$	1.00	0.03	0.02	< 0.0001	-165.15

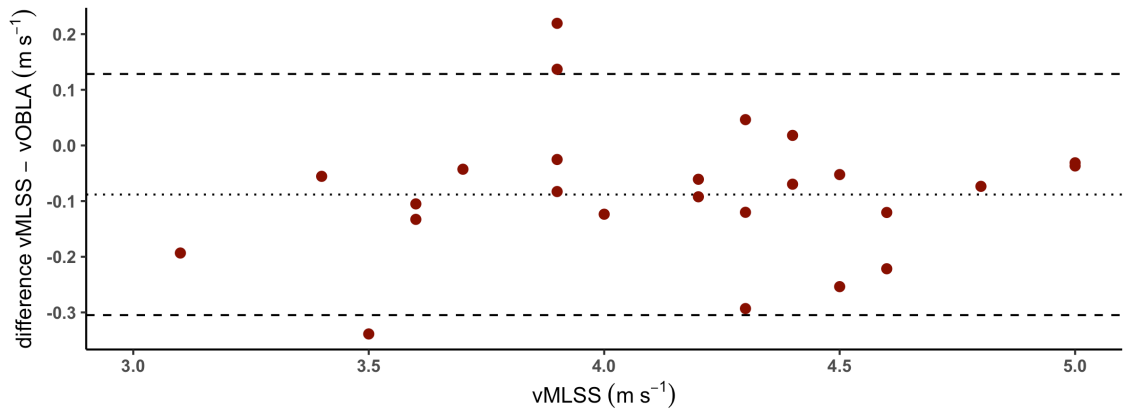
### 3.4 Analysis of test parameters with similar theoretical foundations

Threshold velocity calculated based on different concepts including MLSS, CV and OBLA were analyzed in greater detail for evaluation of agreement (Figure 6). A high amount of variance in vMLSS could be explained by vOBLA ( $R^2 = 0.94$ ,  $p < 0.0001$ ) and CV ( $R^2 = 0.83$ ,  $p < 0.0001$ ).

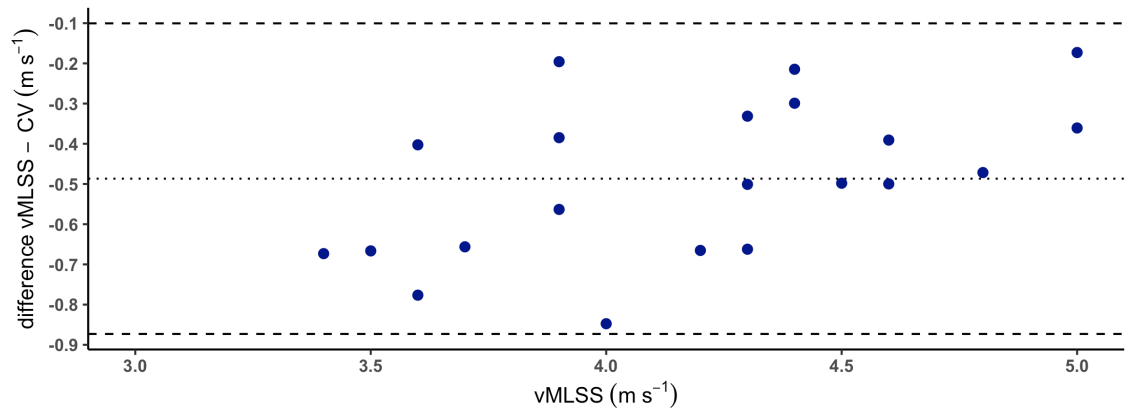


**Figure 6** Relationship of vMLSS with vOBLA and CV. Dashed line indicates linear regression line and grey area confidence interval.

Mean difference between vMLSS and vOBLA amounted to  $-0.08 \text{ m}\cdot\text{s}^{-1}$ , with limits of agreement from  $0.15$  to  $-0.32 \text{ m}\cdot\text{s}^{-1}$ (Figure 7). A larger mean difference of  $-0.49 \text{ m}\cdot\text{s}^{-1}$  and broader limits of agreement ranging from  $-0.10$  to  $-0.87 \text{ m}\cdot\text{s}^{-1}$  were present between vMLSS and CV (Figure 8).

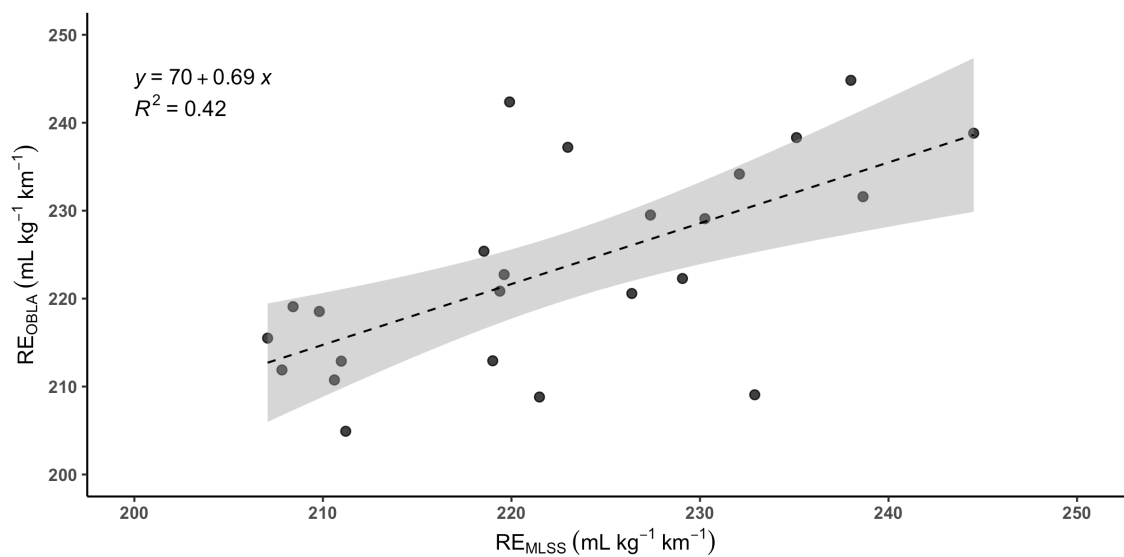


**Figure 7** Bland-Altman plot for analysis of agreement between vMLSS and vOBLA. Dotted line indicates mean difference and dashed lines represent upper and lower limits of agreement



**Figure 8** Bland-Altman plot for analysis of agreement between vMLSS and CV. Dotted line indicates mean difference and dashed lines represent upper and lower limits of agreement

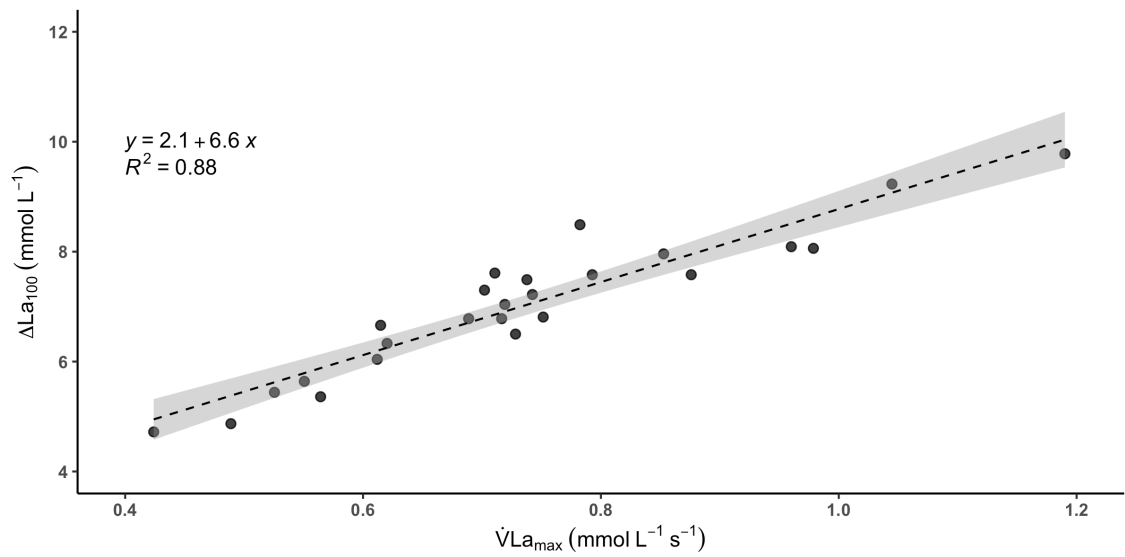
Despite a mean difference of  $-0.49 \text{ mL} \cdot \text{kg}^{-1} \cdot \text{km}^{-1}$ , comparison of RE at MLSS and OBLA revealed a weaker relationship ( $R^2 = 0.42$ ,  $p < 0.001$ ) than the comparison of velocities (Figure 9).



**Figure 9** Relationship of  $RE_{MLSS}$  with  $RE_{OBLA}$ . Dashed line indicates linear regression line and grey area confidence interval.

Correlation analysis revealed a very strong relationship between  $\dot{V}La_{max}$  and  $\Delta La_{100}$  (Figure 10).





**Figure 10** Relationship of  $\dot{V}La_{max}$  and  $\Delta La_{100}$ . Dashed line indicates linear regression line and grey area confidence interval.

## 4 Discussion

### 4.1 Predictors of time-trial performance

#### 4.1.1 *Physiological predictors of time trial performance*

In line with current literature on determinants of endurance performance (Bassett, 2000; Joyner & Coyle, 2008),  $\dot{V}O_2\text{max}$  represented the strongest physiological predictor for all TT distances. High relevance of maximal aerobic power for running performance has been well-established previously (Bacon et al., 2013; Boileau et al., 1982; Brandon, 1995; Levine, 2008; Lundby et al., 2017; Lundby & Robach, 2015). Furthermore, increasing correlation coefficients with rising time trial distance confirm present evidence of increased aerobic energy contribution and parallel importance of aerobic capacity as function of time in endurance running (Busso & Chatagnon, 2006; Gastin, 2001; Heck et al., 2003; Peronnet & Thibault, 1989; Spencer & Gastin, 2001). High predictive value has previously been found for  $\dot{V}O_2\text{max}$  ranging from middle-distance (Brandon, 1995) to ultra-running running (Pastor et al., 2022). Mathematical simulations estimate anaerobic energy system contribution of about 30% and 10% in correspondence to the mean 1- and 3-km TT finishing times in our study (Gastin, 2001; Peronnet & Thibault, 1989). Along these lines, previous comparative studies have reported higher  $\dot{V}O_2\text{max}$  in long-distance compared to middle-distance runners (Boileau et al., 1982; Brandon, 1995; Crielaard & Pirnay, 1981; Daniels & Daniels, 1992; Schnabel & Kindermann, 1983; Svedenhag & Sjödin, 1984). Nevertheless, it is comprehensible that even shorter running durations such as a 1-km TT are strongly dependent on  $\dot{V}O_2\text{max}$  since higher levels of  $\dot{V}O_2\text{max}$  enable higher total energy release and higher sustainable velocity. Minimal aerobic energy contribution in sprint compared to endurance running (Peronnet & Thibault, 1989; Spencer & Gastin, 2001), coincides with the absence of a statistical relationship between  $\dot{V}O_2\text{max}$  and 100-m performance in our investigation.

In contrast to  $\dot{V}O_2\text{max}$ , research concerning its fractional utilization at MLSS has been scarce and rather equivocal. The lack of correlation for  $\% \dot{V}O_2\text{max}$  with TT or sprint performance in our study is supported by previous investigations, which concluded  $\% \dot{V}O_2\text{max}$  not to be a valid predictor of performances below 20 min (Støa et al., 2010; Tanji, Shirai, et al., 2017), ~ 60 min (McLaughlin et al., 2010)

and for marathon running (Gordon et al., 2017). However other studies have observed higher  $\% \dot{V}O_{2\max}$  in athletes exhibiting superior endurance performance (Sjödín & Svedenhag, 1985), leading to the assumption that  $\% \dot{V}O_{2\max}$  is a performance relevant measure (Coyle, 1995). Yet, Støa et al. argue, that this could possibly be a misconception due to decreasing  $\% \dot{V}O_{2\max}$  as a function of time (Støa et al., 2010). Consequently, higher  $\% \dot{V}O_{2\max}$  in highly trained runners might simply be the result of a shorter performance duration, and not a determinant of running performance. Thus, it can be hypothesized that enabling higher rates of aerobic energy release by increasing  $\% \dot{V}O_{2\max}$  in competition could potentially be beneficial for performance. To date insufficient evidence exists to confirm such a hypothesis (Brandon, 1995). Further research on  $\% \dot{V}O_{2\max}$ , its underlying mechanisms and role for running performance should be conducted.

Robust evidence exists on the importance of RE for running performance (Barnes & Kilding, 2015b). Athletes with better RE can outperform competitors with similar  $\dot{V}O_{2\max}$  and athletes of the same performance level might compensate lower  $\dot{V}O_{2\max}$  with better RE (Daniels & Daniels, 1992; Jones, 2006; Lucia et al., 2008; Weston et al., 2000). Even though significant correlations between RE, 2- and 3-km TT performance were found in our study, the respective correlation coefficients were only moderate and did not reach the level of significance for 1 km. Hence it can be hypothesized, that on the lower end of the middle-distance spectrum, the ability to generate higher rates of energy release through  $\dot{V}O_{2\max}$  is more relevant than the efficient usage of oxygen. However a study by Tanji, Shirai, et al., 2017 reported considerably higher correlation coefficients between RE and middle-distance performance in a homogenous group of well-trained middle-distance runners. Yet, substantial differences in methodology compared to our study might explain this disparity. Tanji and colleagues determined RE from energy equivalents of both oxygen consumption and lactate, to account for aerobic and anaerobic energy expenditure and assessed RE at velocities below (65%  $\dot{V}O_{2\max}$ ) and above (90%  $\dot{V}O_{2\max}$ ) intensities usually corresponding with lactate threshold (Jones et al., 2019).

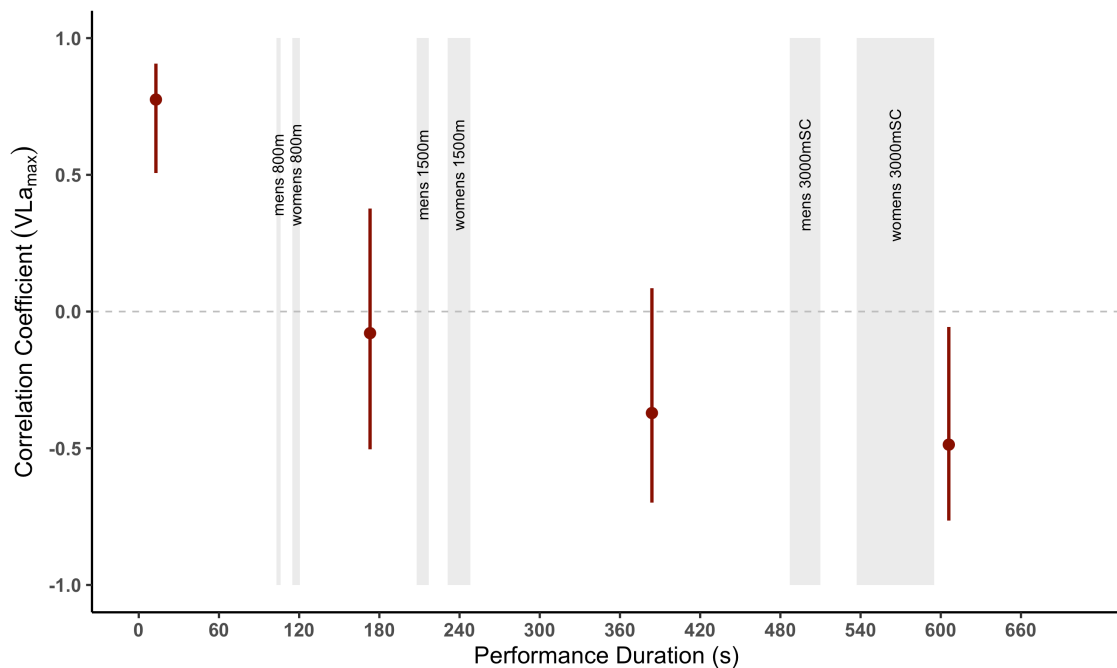
Positive correlations between  $v_{100}$  and RE could allow the assumption that higher oxygen consumption at submaximal running intensity enhances sprint

performance. Accordingly, [Vučetić et al., 2007](#) found significantly higher metabolic cost in sprinters than middle- and long-distance runners. Nonetheless, the causality of this relationship should be questioned due to the minimal aerobic energy contribution in sprinting (Gastin, 2001; Peronnet & Thibault, 1989). It seems more plausible that higher proportion of type-II muscle fibers and greater muscle mass typically found in sprinters causes higher metabolic cost of running, rather than RE taking direct influence on sprint performance. While contractile properties of type-II muscle fibers and increased muscle mass have positive effects on forward propulsion and peak velocity in sprint running (Miller et al., 2020; Nuell et al., 2021), they are detrimental for RE due to increased oxygen and energy consumption (Bellinger et al., 2021a; Black et al., 2020; Pringle et al., 2003). In our study, influence of greater muscle mass on RE might be reflected by the very strong relationship found between RE and lean body mass in proportion to body height squared ( $BMI_{LBM}$ ).

It is anaerobic energy release that permits high muscular power output relevant to sprint running (Arsac & Locatelli, 2002). Consequentially, a strong correlation between 100 m sprint performance and  $\dot{V}La_{max}$  as well as  $\Delta La_{100}$  were found in this investigation. Similar correlation coefficients have been reported in preceding studies (Quittmann, Appelhans, et al., 2020; Quittmann, Schwarz, et al., 2020). Withal, the bias through incorporation of 100 sprint time in the calculation of  $\dot{V}La_{max}$  is a limitation that should be noted. In spite of this limitation, similar results were found for  $\Delta La_{100}$ , having no mathematical link to  $v_{100}$ .

In contrast, correlations of  $\dot{V}La_{max}$  and  $\Delta La_{100}$  with TT trended negatively with increasing distance. While the negative correlation of  $\dot{V}La_{max}$  and  $\Delta La_{100}$  with 3-km TT performance was statistically significant, correlations with 1- and 2-km TT were weak to moderate and not significant. At first sight this might imply irrelevance of  $\dot{V}La_{max}$  and  $\Delta La_{100}$  for 1- and 2-km running. Contemplating the whole continuum from 100 m sprint to 3 km however, there seems to be a crossing point at which positive and negative influence of  $\dot{V}La_{max}$  and  $\Delta La_{100}$  are in balance. This crossing point seems to occur around the velocity associated with 1-km TT or maximal running of approximately three minutes. Running performances shorter than this crossing point seem to benefit from higher  $\dot{V}La_{max}$

and  $\Delta La_{100}$ , while longer performance experience an increasingly detrimental effect (Figure 11)



**Figure 11** Conceptual data displaying assumed relationship of  $\dot{V}La_{max}$  and world-class male and female 800 m, 1500 m, 3000 m steeplechase running. Correlation coefficients are placed at mean velocities for 100 m and 1-, 2- and 3-km TTs examined in this study. Shaded areas represent the range of respective TOP50 performances for 2021. Red bars represent confidence intervals.

The beneficial and detrimental effects of  $\dot{V}La_{max}$  and  $\Delta La_{100}$  are likely caused by their link to the anaerobic metabolism. Anaerobic breakdown of ATP, high-energy phosphates, and glucose (Heck et al., 2003) can be beneficial, when high rates of energy are needed for example in high velocity running. However, the accumulation of hydrogen protons as a result of these metabolic pathways lead to muscular acidosis and subsequent impairment of muscular function (Robergs et al., 2004). Despite common acceptance of these metabolic processes, in the past scientist have not clearly investigated and identified the running duration or distance at which beneficial and detrimental effects are leveled. Distinctions in anaerobic power and capacity have been reported for athletes competing in different running event though (Crielaard & Pirnay, 1981; Schnabel & Kindermann, 1983; Svedenhag & Sjödín, 1984). For example Schnabel & Kindermann, 1983 found lactate concentrations were highest in elite sprinters and lowest in elite marathon runners after a supramaximal run to exhaustion. Based on these results it can either be assumed that athletes are physiologically

adapted to the requirements of their running event or success in respective running events requires a distinct metabolic profile.

While the anaerobic metabolism is driven by breakdown of high energy phosphates and glucose, energy can also be supplied by breakdown of lipids up until approximately vMLSS or CV (Maunder et al., 2018). From resting levels, absolute fat oxidation increases during lower intensities until MFO, from where it decreases as lipid metabolism cannot meet high rates of energy release required during higher intensities (Jeukendrup & Wallis, 2005). MFO represented the second-best single physiological predictor of all TT performances, which were run well above MLSS or critical velocity. Additionally, MFO added substantial explanation of variance to stepwise regression models of all TT, despite lipid oxidation being likely neglectable during these intensities. Multiple studies however, have shown MFO is an expression of endurance training status (Maunder et al., 2018) and especially relevant during ultra-endurance running (Frandsen et al., 2017; Pastor et al., 2022). Athletes with higher endurance performance capacity display higher MFO, presumably due to higher absolute training volume in the low-intensity domain. Due to strong correlations between MFO and  $\dot{V}O_2\text{max}$ , this could also hold true for our athlete cohort, rather than MFO playing a decisive role within the range of 3 to 10 min maximal running.

#### *4.1.2 Performance predictors of time trial performance*

Investigating the influence of performance predictors on 1-, 2-, 3-km TT and 100-m sprint performance and comparing the results with physiological predictors is a further objective of this thesis. Due to the complex interaction of physiological mechanisms in human exercise (Coffey & Hawley, 2007; Flück, 2006; Joyner & Coyle, 2008; Lavin et al., 2022), single physiological parameters often leave some amount of variation in performance unexplained (Grant et al., 1997). An advantage of this approach is the finding concerning the function and influence of single physiological mechanisms for the selected performance. The disadvantage on the other hand is the lack of predictive accuracy, when examining a specific part of a complex system. Higher accuracy for predicting performance can often be achieved through observations of other similar performances or performance-related parameters (Lerebourg et al., 2022; Röcker et al., 1998). In support of this, high accuracy of prediction was achieved

for single TT performance through the other TT performances in this study. Furthermore, performance-related parameters such as velocities associated with  $\dot{V}O_2\text{max}$  and MFO displayed higher correlation coefficients than the respective physiological variables alone.

In addition, mathematical models such as CV model can be applied for predictions of running performance (Fukuba & Whipp, 1999; Kramer et al., 2020; Patoz et al., 2021). The linear velocity-distance relationship has previously been used for accurate predictions of endurance running performance (Bundle et al., 2003; Nimmerichter et al., 2017) and even pacing behavior (Kirby et al., 2021). These previous findings can be confirmed by our results, where CV and D' were calculated from 1-, 2-, and 3-km TTs. CV represented the single best predictor of 2- and 3-km TTs and when complemented by D' in multiple regression analysis near perfect predictions with low residual standard errors were reached. This was not the case for the 1-km TT, which was best predicted by  $v\dot{V}O_2\text{max}$  alone and by  $v\dot{V}O_2\text{max}$ , D' and CV in multiple regression. High predictions of variance were reached in multiple regression, nonetheless.

Congruent with the results of this study, research has shown that  $v\dot{V}O_2\text{max}$  is decisive for middle-distance (Boileau et al., 1982; Brandon, 1995; Sandford, Rogers, et al., 2019a), long-distance (Jones & Doust, 1998; Morgan et al., 1989) and even ultra-running (Coates et al., 2021). In agreement to our findings Ingham et al., 2008 showed significant correlations between  $v\dot{V}O_2\text{max}$  and 800- and 1500-m performance, yet a stronger relationship in the case of 1500-m. This confirms lower correlation coefficients found in this study for 1-km TT compared to longer TTs and reveals a potentially decreasing influence of aerobically determined performance at this distance.

Like CV and  $v\dot{V}O_2\text{max}$ ,  $v\text{MLSS}$  also showed a strong relationship with TT performance. This could be due to their similarity of representing velocities that can only be sustained for a finite amount of time (Jones et al., 2019) and hence being very similar to an endurance performance observed in the field. Strong correlations between  $v\text{MLSS}$  or similar threshold concepts and middle- and long-distance running have been continuously observed in the past (Alvero-Cruz et al., 2020; Jones & Doust, 1998; Sjödín & Jacobs, 1981).

Even  $Fat_{max}$ , a submaximal velocity, has produced correlations with TT performance similar to  $v\dot{V}O_{2max}$ , CV and  $vMLSS$ . Unfortunately, scarce literature exists on the influence of  $Fat_{max}$  on running performance (Maunder et al., 2018). Based on the strong correlations with determinants of endurance performance such as  $\dot{V}O_{2max}$ , CV  $vMLSS$  and TT performance, it can be assumed that runners with higher overall endurance fitness have a higher  $Fat_{max}$ . Without any biological or biomechanical proximity to high intensity running it seems unlikely though that  $Fat_{max}$  is a key determinant of running performance up to ~ 10 min.

Apart from estimation of threshold velocity, the CV model can also give indirect insight on anaerobic capacity through the curvature constant of the velocity-distance relationship or  $D'$ . It describes the observed increase in velocity with decreasing TT running distance or the ability to run above CV. Since additional energy release through anaerobic pathways permits a velocity increase for a given distance above CV,  $D'$  is often used as a synonym for anaerobic capacity. In line with these assumptions,  $D'$  correlation coefficients with sprint and TT performance were coincided with the anaerobic parameters of  $\dot{V}La_{max}$  and  $\Delta La_{100}$ . Continuous decline from significant positive correlations with sprint performance, over non-significant weak-positive to moderate negative correlations with 1- and 2-km TTs to a significant negative influence on 3-km performance was observed. Like for  $\dot{V}La_{max}$  and  $\Delta La_{100}$  it can be hypothesized, that  $v_{100}$  is positively influenced by  $D'$ , while this influence degrades continuously with increasing performance duration and eventually becomes a limitation for endurance performance. Unfortunately, little research attention on  $D'$  makes comparison with other studies difficult. [Blondel et al., 2001](#) found very strong positive correlations ( $r = 0.91 - 0.94$ ) between  $D'$  and supramaximal time to exhaustion tests (120% and 140% of  $v\dot{V}O_{2max}$  with mean times of 122 and 67 s in a study by [Kramer et al., 2020](#) observed a trend for higher  $D'$  in trained compared to untrained individuals but reported only small effect sizes for group comparison. The results of both studies stand in clear contrast to the results of this study. More recently, [Kirby et al., 2021](#) attempted to model bioenergetics of the 2017 men's 10,000 and 5,000 world championship finals with the  $D'$ -balance model. This model assumes that  $D'$  is depleted when running above CV and reconstituted when running below and that once depleted the highest sustainable pace is CV.



Based on these presumptions [Kirby et al., 2021](#) simulated the balance of D' depletion and reconstituted depending on the variation in race velocity and individual CV. They concluded that in “slow” races athletes with the highest remaining D' going into the final were able to run a faster final lap. Based on this observations D' could be interpreted as an indirect measure of the maximal anaerobic capacity. However, this has been partly refuted due to the lack of a correlation of D' cycling-equivalent W' with muscle fiber type distribution [Vanhatalo et al., 2016](#) and reduction of W' under hyperoxic conditions (Vanhatalo et al., 2010). More research is needed to gain better understanding of the underlying physiological mechanisms of D'.

ASR and SRR are two further performance parameters theoretically linked to anaerobic energy release. High ASR or SRR can be achieved through fast sprint and relatively slow maximal aerobic speed or  $\dot{V}O_{2\max}$ . Since sprint performance is predominately fueled by the anaerobic metabolism a connection with ASR and SRR seems logical. A tendency for higher proportion of type-II muscle fibers connected to ASR has been documented recently (Bellinger et al., 2021a) and a strong relationship between ASR and v100 in this study speaks in favor of the assumed connection to the anaerobic metabolism. Although, correlations followed a similar continuously negative pattern as  $\dot{V}La_{\max}$ ,  $\Delta La_{100}$  and D', the relationship was already significant for 2-km TT, confidence intervals were narrower and correlation coefficients higher. A potential explanation for the more pronounced negative relationship in comparison to  $\dot{V}La_{\max}$ ,  $\Delta La_{100}$  and D' could be the calculation from two distinct performance observations, as opposed to variables from physiological tests or a mathematical model. However, a positive correlation was found between ASR and world-class 800-m running performance, (Sandford, Allen, et al., 2019) but a moderate negative correlation was present in elite 1500-m runners (Sandford, Rogers, et al., 2019b). This suggests that the time frame of 90 s to 3 min maximal running represents a dynamic metabolic situation associated with highly variable physiological responses and therefore doesn't allow generalization.

Again, similar negative but even clearer relationships than observed in ASR, were found for SRR and all TTs. Derived from the same variables as ASR, SRR

represents a ratio instead of a velocity. Subsequently, higher sprint speed or lower  $\dot{V}O_2\text{max}$  seem to increasingly inhibit endurance running performance from 1 to 3 km. This could lead to the hypothesis, that the ratio between maximal anaerobic and aerobic velocity might serve as gauge for determining whether athletes are more endurance or sprint trained. Accordingly, (Sandford, Allen, et al., 2019) were able to clearly differentiate between subgroups of sprint or endurance type 800-m runners on a world-class level. It needs to be critically mentioned that despite advantages of profiling athletes using SRR as a performance indicator it poses challenges. The same SRR could be achieved with  $v_{100}$  and  $\dot{V}O_2\text{max}$  of 9.0 and 6.0  $\text{m}\cdot\text{s}^{-1}$  for one athlete and 7.5 and 5.0  $\text{m}\cdot\text{s}^{-1}$  for another, although their performance is clearly lower in the latter.

Both results of ASR and SRR show, that endurance running performance ranging from about three to ten minutes in duration is not exclusively determined by either sprint or endurance capabilities. Multiple investigations have found both sprint (Bachero-Mena et al., 2017; Houmard et al., 1990; Sandford, Kilding, et al., 2019) and endurance (Brandon, 1995; Svedenhag & Sjödín, 1984; Tanji, Tsuji, et al., 2017) capabilities to be relevant for middle-distance running. Low ASR and SRR, associated with superior endurance performance, can theoretically be achieved despite high  $v_{100}$  if  $\dot{V}O_2\text{max}$  is also high. Thus, potential negative influence of  $v_{100}$  on endurance performance seems to be mitigated by high  $\dot{V}O_2\text{max}$  or underlying aerobic mechanisms. Accumulation of lactate and hydrogen protons caused by sprinting or running at velocities above  $v_{MLSS}$  could be delayed in athletes capable of clearing these metabolites through aerobic mitochondrial respiration (Robergs et al., 2004).

Based on these results it can be concluded that the investigated selection of performance or performance-related parameters serve as good predictors for running performance, especially exceeding distances of two km or duration of about six minutes. The accuracy of prediction exceeded predictions made by physiological parameters. In congruence with existing evidence, parameters connected to aerobic metabolism tend to positively influence endurance performance, while the opposite is the case for parameters linked to the anaerobic metabolism. Greatest amount of unexplained variance was found for

1-km TT performance. It seems that physiological response to this exercise intensity can vary substantial between athletes.

## 4.2 Predictors of maximal lactate steady-state and critical velocity

### 4.2.1 Comparison of MLSS and CV

MLSS and CV are two concepts to date which have been used to define and assess maximal metabolic steady-state. The physiological phenomenon of maximal metabolic steady-state describes the highest velocity at which physiological homeostasis can be sustained (Jones et al., 2019). While  $v_{MLSS}$  is defined as the velocity associated steady-state of lactate concentration, CV represents a velocity that allows a steady-state in oxygen consumption but not in lactate (Nixon et al., 2021). Above these defined intensities, a rapid and continuous rise of either lactate concentration or oxygen consumption is observed until exhaustion. As a result of these different definitions of maximal metabolic steady-state, CV is usually found at higher velocities than  $v_{MLSS}$ . Mean difference of  $0.48 \text{ m}\cdot\text{s}^{-1}$  found in this investigation was very similar to previously reported  $0.6 \text{ m}\cdot\text{s}^{-1}$  (Nixon et al., 2021). On an individual level, a broad spectrum of differences was observed in Bland-Altman plots though with limits of agreement from approximately  $-0.1$  to  $-0.9 \text{ m}\cdot\text{s}^{-1}$ . Some subjects reported RPE of 15 corresponding to “somewhat hard”, without staying within criteria for MLSS, whilst other subjects had to end constant load tests prematurely without a delta lactate concentration of exceeding  $1 \text{ mmol}\cdot\text{L}^{-1}$  from 10 min until exhaustion.

It could be speculated that the difference  $v_{MLSS}$  and CV ( $v_{MLSS-CV}$ ) is greater in athletes that have higher anaerobic capacity, due to  $v_{MLSS}$  being limited by lactate accumulation over longer duration of intense running. Moderate correlation between  $\text{Diff}_{v_{MLSS-CV}}$  and  $\text{La}_{RT}$  ( $r = -0.43$ ,  $p = 0.06$ ) as well as high correlations between three and six minutes post 3-km TT and  $\text{Diff}_{v_{MLSS-CV}}$  ( $r = -0.67$ ,  $p = 0.001$ ;  $r = -0.72$ ,  $p < 0.001$ ;) support this speculation. In contrast to these values describing post-exercise lactate accumulation,  $\dot{V}\text{La}_{max}$ , describing the rate of lactate production, did not influence  $\text{Diff}_{v_{MLSS-CV}}$  ( $r = -0.08$ ,  $p = 0.72$ ).

As a consequence of lower velocity at MLSS compared to CV, maximal sustained time at  $v_{MLSS}$  has previously been observed longer ( $\sim 30 - 60 \text{ min}$ ) than CV ( $\sim 20 - 30 \text{ min}$ ) (Jones et al., 2019). From these and other reports it can be assumed

that world class level running events such as 5,000 and 10,000 m are closely related and well-predicted by CV (Kirby et al., 2021), and therefore limited by  $\dot{V}O_2\text{max}$  and steady-state in oxygen consumption. Whereas world-class 15 km, ten mile and half marathon running might be limited by extended accumulation and exposure to muscular acidosis over the course of the race and better predicted by vMLSS (Röcker et al., 1998).

Despite observed differences in velocity and maximal sustained time CV and vMLSS share a very strong relationship (Figure 6). Very similar values were also reported in previous investigations (Galán-Rioja et al., 2020; Iannetta et al., 2022; Nixon et al., 2021). A very strong relationship and low mean difference was also found between vMLSS and vOBLA (Figure 6 & 7), reflecting previous investigations (Heck et al., 1985; Jones & Doust, 1998). Due to this close link and substantially lower assessment time, vOBLA has frequently been used as a surrogate for vMLSS in the past (Faude et al., 2009). Nonetheless, it needs to be considered that individual differences can vary (Figure 7) (Galán-Rioja et al., 2020). In addition, judging from our results RE cannot be assumed to be identical for vMLSS and vOBLA. Respective correlations coefficient was significant but only moderate (Figure 8).

#### *4.2.2 Predictors of MLSS and CV*

Despite recommendation not to use CV and MLSS synonymously, like other threshold concepts, they are both valid indicators of general endurance performance (Alvero-Cruz et al., 2020; Faude et al., 2009; Heuberger et al., 2018; Jones et al., 2019). Accordingly, relationships with aerobic parameters ( $\dot{V}O_2\text{max}$ ,  $v\dot{V}O_2\text{max}$ ,  $Fat_{\text{max}}$ , MFO) were significant and ranged from strong to very strong for both CV and vMLSS. RE was negatively correlated because higher values represent lower efficiency. In opposition, anaerobic parameters ( $\dot{V}La_{\text{max}}$ ,  $\Delta La_{100}$ ,  $D'$ , SRR, ASR) displayed moderate to very strong negative correlations with CV and vMLSS, underlining potential detrimental effects of anaerobic metabolism on endurance performance already observed in TTs. Whether detrimental effects can be isolated to anaerobic energy release and onset of muscular acidosis is unclear. Perhaps these parameters do not reflect anaerobic metabolism alone but muscle characteristics such as fiber type distribution (Costill et al., 1976; Vanhatalo et al., 2016).

### 4.3 Limitations

Unfortunately, small sample sizes of the sprint and (ultra-)marathon cohorts did not allow a meaningful group comparison. Additionally, a lower participation in TTs further reduced examined sample size for investigation of TT performance. From a methodical standpoint it also needs to be taken into account, that mean velocity during the 100-m sprint does not resemble maximal velocity. Maximal velocity is usually assessed during a shorter segment after the acceleration phase (Sandford et al., 2021). Comparison of ASR and SRR with other studies is not advised since maximal sprint speed in these variables was derived from v100. Furthermore, substantially lower sprint performance in endurance athletes might not be a sole result of lower maximal velocity but inferior ability to accelerate. This can cause further bias and is a limitation of assessing sprint performance in endurance athletes. To alleviate this problem, familiarization was a key component of the warm-up program before the 100-m sprint.

The decision to recruit athletes from different competition backgrounds was made deliberately to make more general assumptions on determinants of endurance performance opposed to the majority of evidence focusing on endurance trained or even specialized world-class athletes (Borgen, 2018). In return, this entails the limitation of examining participants with more heterogenous endurance performance, especially due to the inclusion of sprinters. Focusing on developing sprint rather than endurance performance in training (J. L. Andersen et al., 1994; Haugen et al., 2019, 2022; Majumdar & Robergs, 2011), a different metabolic profile and lower endurance performance in sprinters was expected before commencement of the study. Results need to be interpreted accordingly and caution needs to be taken when drawing conclusions for homogenous cohorts of endurance runners. Negative influence of parameters linked to sprint performance and anaerobic metabolism might not be as pronounced when examining endurance athletes only or middle-distance runners more specifically. Other investigation showed contrary results in homogenous cohorts (Tanji, Shirai, et al., 2017; Tanji, Tsuji, et al., 2017).

## 5 Practical applications

From a practical perspective the presented results can be applied in various scenarios and settings of endurance running. Firstly, traditional diagnostic parameters such as  $\dot{V}O_2\text{max}$ , RE and  $v\text{MLSS}$  can still be coined as highly relevant variables of endurance performance. The ability to generate high ATP turnover through aerobic pathways enables higher performance especially for running events which reach  $\dot{V}O_2\text{max}$  or utilize a high fraction of it. A large body of research has been compiled explaining underlining mechanisms and guiding training interventions to enhance  $\dot{V}O_2\text{max}$  (Bacon et al., 2013; Boushel et al., 2011; Esfarjani & Laursen, 2007; Levine, 2008; Lundby et al., 2017; Midgley et al., 2006).  $\dot{V}O_2\text{max}$  is predominantly determined by cardiac output and oxygen transport capacity and to a lesser extent by oxygen extraction linked to mitochondrial function (Daussin et al., 2007). Evidence suggests, high-intensity training seems to cause the greatest adaptation in  $\dot{V}O_2\text{max}$  (Helgerud et al., 2007) and dose-response effect can be observed if intensity and recovery are optimally programmed (Langan & Grosicki, 2021; Rosenblat et al., 2022). While large improvements observed in less trained individuals, enhancements of  $\dot{V}O_2\text{max}$  are of small magnitude or not present at all in elite athletes (Lucia et al., 2008; Zoppirolli et al., 2020). At this point, further improvements of endurance performance has been shown to be driven by submaximal parameters such as exercise economy (Zoppirolli et al., 2020).

In line with those observations, relevance of RE for endurance performance can be confirmed through this investigation. Despite lower correlation coefficients than  $\dot{V}O_2\text{max}$ , RE gained influence with growing duration of maximal running and explained a large amount of variance in endurance performance in a multiple regression model with  $\dot{V}O_2\text{max}$ . Not only does this confirm previous assumed relevance for endurance performance (Borgen, 2018; Weston et al., 2000) but shows the interaction of maximal and submaximal oxygen consumption (Joyner, 1991). Truly excellent RE ( $< 170 \text{ mL} \cdot \text{kg}^{-1} \cdot \text{km}^{-1}$ ) is rarely combined with the highest reported values of  $\dot{V}O_2\text{max}$  ( $\sim 90 \text{ mL} \cdot \text{min}^{-1} \cdot \text{kg}^{-1}$ ) even in world-class athletes (Jones, 2006; Joyner, 1991; Lucia et al., 2006, 2008) and vice versa. This is most certainly due to the fact that, extreme values of  $\dot{V}O_2\text{max}$  with low submaximal

oxygen consumption would only be reached under extreme velocities which are likely not sustainable for longer periods of time due to other limiting factors such as muscular acidosis (Lucia et al., 2002). This needs to be considered when prescribing training or setting physiological benchmarks for athletes. Previously identified factors influencing RE have been anthropometric variables such as body size, fiber type distribution, life time running distance (Barnes & Kilding, 2015b), high-volume low intensity training, uphill running or strength and plyometric training (Barnes & Kilding, 2015a).

Adverse to the common assumptions (Bassett, 2000; Joyner & Coyle, 2008)  $\% \dot{V}O_2\text{max}$  was not identified as a relevant parameter for TT performance, CV or vMLSS. In addition, no clear evidence exists on how to deliberately improve  $\% \dot{V}O_2\text{max}$ . Caution is recommended for practitioners when interpreting  $\% \dot{V}O_2\text{max}$  until further evidence arises. Scientists are called-upon to identify mechanisms, determinants and training interventions concerning  $\% \dot{V}O_2\text{max}$ .

Amongst the traditional diagnostic parameters in running threshold concepts have gained considerable attention for decades (Faude et al., 2009; Heuberger et al., 2018). From the collected data it can be confirmed that CV, vMLSS and vOBLA share strong relationships with endurance performance in the field and physiological parameters connected to the aerobic energy supply. Longitudinally monitoring general performance can be recommended for CV, vMLSS and vOBLA, if tested in a repeated and standardized manner. It is not recommended however, to use different threshold determination concepts interchangeably (Galán-Rioja et al., 2020; Jones et al., 2019). Analyzing and prescribing training based on threshold-derived training zones is common practice in science and coaching, yet also faces its challenges (Burnley et al., 2022a, 2022b; Foster et al., 2022a, 2022b)

Despite close connection to various endurance performance, threshold determination as the only testing objective has the disadvantage of leaving physiological mechanisms underlying endurance performance unknown. Assessing the mentioned traditional as well as novel diagnostic parameters can help practitioners to identify performance determinants and potential training

interventions on an individual basis. Marathon running for example is limited by glycogen availability (Rapoport, 2010). Besides nutritional strategies to delay glycogen depletion, lowering cost of running or RE can reduce glycogen consumption and benefit marathon running. A reduction in glycogen consumption can also be achieved by increasing fat oxidation to meet total energy demand (Rapoport, 2010). Hence, knowledge about substrate utilization of carbohydrates and lipids can be advantageous for running events limited by glycogen availability. Selection of a sustainable race velocity over these longer race durations could be based on  $Fat_{max}$  and MFO.

Potential for individualization of training can also be found in ASR and SRR, especially for middle-distance runners. Like  $\dot{V}La_{max}$  and  $\Delta La_{100}$ , both ASR and SRR have been negatively associated with TT running performance. Yet at the same time acknowledge that negative influence of high maximal anaerobic speed can be mitigated by high aerobic speed. By continuously monitoring ASR and SRR middle-distance coaches can make decisions on whether to focus more on developing anaerobic sprint capabilities or aerobic endurance capabilities. In retrospective it could then be analyzed whether chosen training interventions produced adaptations more in favor of maximal anaerobic or aerobic speed. Pushing sprint capabilities to far without development of aerobic speed could easily be detected and response in training prescription could be made accordingly. One study even investigated the possibility to determine ASR without laboratory from 1500-m race results and came to the conclusion that it can be considered by coaches without access to metabolic testing (Sandford, Rogers, et al., 2019b).

Furthermore, SRR has previously been used to differentiate in speed, hybrid or endurance type 800-m runners (Sandford et al., 2021). A similar categorization could be tested for athletes in longer running competitions or could be used for talent selection. [Sandford et al., 2021](#) even take a step further and suggest individualizing overall training load, training mode (continuous vs. interval) and interval length based on SRR. Their suggestion relies on findings that suggest speed-dominant athletes with fast-twitch muscle typology are at greater risk for overtraining (Bellinger et al., 2020). Unfortunately, to date there are no specific



recommendations concerning the modulation of ASR and SRR based on training intervention studies. However, it does seem plausible that common methods for enhancement of maximal sprint speed (Haugen et al., 2019) or  $\dot{V}O_{2\max}$  (Rosenblat et al., 2018, 2022) can be used accordingly to either increase or decrease ASR and SRR.

The potential of monitoring training-induced adaptations of anaerobic metabolism through  $\dot{V}La_{\max}$  can be confirmed through this and previous investigations (Hommel et al., 2019; Manunzio et al., 2016; Quittmann et al., 2022). In line with the results of this study, all mentioned studies showed an increasing in endurance performance with a parallel decrease in  $\dot{V}La_{\max}$ . The decrease of  $\dot{V}La_{\max}$  in a triathlete with spinal cord injury (Quittmann et al., 2022) and in a ultra-distance cyclists (Manunzio et al., 2016) were achieved by a pyramidal training intensity distribution, with large proportions of training spent within the low-intensity domain. In contrast, Hommel et al., 2019 reported a decrease in  $\dot{V}La_{\max}$  after six weeks of sprint-interval training in cyclists and no change through continuous endurance training. Nitzsche et al., 2020 reported increased  $\dot{V}La_{\max}$  as a result of strength training. In running no training interventions have monitored  $\dot{V}La_{\max}$  to date, despite potential help insights for coaches. For example, maintaining sprint speed but reducing detrimental lactate production or metabolic acidosis, would seem beneficial especially for middle-distance runners and could be monitored regularly by assessing  $\dot{V}La_{\max}$ .

The general observation can be made, that traditional parameters have been concentrated around aerobic metabolism and have neglecting the influence of anaerobic performance on endurance performance (Wackerhage et al., 2022). One potential explanation for the common neglect of anaerobic parameters in endurance exercise could be the fact, that no method for determination of anaerobic capacity or power has found common acceptance. Both maximal accumulated oxygen deficit (Noordhof et al., 2010) and post-exhaustive exercise lactate concentrations (Buchheit & Laursen, 2013) and other proposed tests (Nummela et al., 1996) to determine maximal anaerobic capacity have been heavily criticized (Green & Dawson, 1993). Additionally, anaerobic energy contribution has been simulated to account for as little as ~2% for durations of

maximal running exceeding 13 min (Peronnet & Thibault, 1989) implying little relevance for endurance performance.

Nevertheless, results of this and other studies show that monitoring aerobic and anaerobic parameters side by side holds potential for coaches, athletes and sport scientists. Anaerobic energy contribution increases substantially and gains relevance for middle-distance running events (Peronnet & Thibault, 1989). All investigated parameters with potential link to the anaerobic metabolism revealed a similar negative influence on endurance performance exceeding approximately three minutes and a trend for positive influence below this duration. It is noticeable that the pattern observed in these parameters is very similar to the characteristic curve of the hyperbolic velocity-distance relationship. Hence, it can be speculated that the presumably anaerobic parameters investigated in this study and their underlying mechanisms are at least partly responsible for inevitable decrease in running velocity with increasing race distance or duration. This could be due to the positive relationship found between anaerobic capabilities in runners and abundance of type-II muscle fibers (Bellinger et al., 2021a; Costill et al., 1976). Based on these findings assessment anaerobic parameters could be used to make careful assumptions on muscle typology.

## 6 Future directions

Continuous research in the field of endurance exercise and the development of new testing methods have led to advances in understanding biological mechanisms of endurance exercise and individualized exercise prescription over the last centuries. While examples of more recent developments and test parameters have been discussed in this thesis, the field of exercise science and physiology is moving forward rapidly.

Recently, exciting contributions have been made to the field of exercise science through interaction with molecular biology, genomics, muscle physiology, research on wearable technology and through public availability of large amounts of data. Scientists have identified a plethora of genes associated with superior endurance performance or specific physiological traits that could benefit talent identification or help to better understand dose-response relationships (Eynon et al., 2011; Harvey et al., 2020; Jacques et al., 2020; Pickering et al., 2019; Pickering & Kiely, 2019a, 2019b; Seaborne et al., 2018; Voisin et al., 2020). Furthermore, new non-invasive methods for the assessment of muscle physiology such as near-infrared spectroscopy (Azevedo et al., 2022; Batterson et al., 2020; Ferrari et al., 2011; Hovorka et al., 2021; Neary et al., 1992; Sako et al., 2001; van der Zwaard et al., 2016) or proton magnetic resonance spectroscopy (Baguet et al., 2011; Bellinger et al., 2021a, 2021b; Lievens, 2021; Lievens et al., 2020) have been established and produced promising results in running and endurance sport. Wearable technology has made a giant leap in the past century, enabling more precise exercise prescription as well as automated data collection (García-Pinillos et al., 2019; Yang & Gao, 2019). Advances in diabetic research have made continuous glucose measurement applicable in endurance sport (Gao et al., 2018; Holzer et al., 2022). Continuous measurements of other metabolites such as lactate, which promise big benefits compared to conventional discontinuous measurement, have been tested very recently (Chien et al., 2022; Ming et al., 2021). Finally, public availability of large datasets can address the issue of small sample sizes, common in exercise science (Huber, 2016; Smyth et al., 2022; Smyth & Muniz-Pumares, 2020).

## 7 Conclusions

The findings of this thesis aim to contribute to the existing knowledge on endurance running performance and its underlying physiological mechanisms. Through continuous investigations athletes, coaches and scientists can develop more successful and specific training interventions to improve running performance.

The relationships of 1-, 2- and 3-km TT running performance and physiological parameters were investigated and  $\dot{V}O_{2\max}$  revealed the strongest positive correlation with all TTs. This confirms previously assumed relevance of high maximal aerobic power for endurance running performance. MFO also showcased strong correlations with TT performance but is assumed to represent general status of endurance performance rather than having direct influence on maximal running performance of short durations below 10 minutes. Increasingly negative relationships were found between RE and TT performance, implying negative influence of higher oxygen cost with increasing distance.  $\dot{V}L_{\max}$  and  $\Delta La_{100}$  displayed increasingly detrimental effects on TT performance and positive correlations with sprint performance. Based on these findings, balance of detrimental and beneficial effects of anaerobic energy release is assumed around 1-km TT performance or approximately two to three minutes of maximal running. Athletes might experience more beneficial effects of anaerobic metabolism such as higher total energy release in running events shorter than this crossing point. Detrimental effects such as muscular acidosis seem to prevail during longer durations. In contrast to previous assumptions, no significant relationships was found between  $\% \dot{V}O_{2\max}$  and sprint or TT performance or any other investigated variable.

More accurate prediction of TT performance was achieved through performance parameters such as CV,  $vMLSS$  or  $v\dot{V}O_{2\max}$  in comparison to physiological parameters such as  $\dot{V}O_{2\max}$ , MFO or RE. Positive relationships were found between  $v100$  and  $D'$ , ASR and SRR, but correlation coefficients followed an increasingly negative trend from 1- to 3-km TT performance. Results for ASR and SRR lead to the hypothesize, that detrimental effects of high sprint speed and

connected anaerobic energy release can be mitigated by equally high aerobic capabilities.

Results for prediction of threshold velocity were very similar to the results of 3-km TT performance, independent of detection method (CV, MLSS, OBLA). Agreement between the different threshold concepts was highest between vOBLA and vMLSS, while a higher mean velocity was determined for CV compared to vMLSS. Thus, all concepts can be used singularly but not interchangeably for training prescription and as a measure of general endurance performance.

In conclusion, novel diagnostic parameters such as  $\dot{V}La_{max}$ , CV or ASR have presented new and valuable information to complement traditional parameters. Selection of diagnostic parameters in running are suggested to be based on the aim of the investigation. If practitioners aim to predict competition velocity with high precision, performance parameters are the recommended over single physiological parameters. Especially the duration-distance or velocity-distance relationships represented by CV and D' allow very accurate predictions of running performance. Nonetheless, physiological parameters remain relevant when studying and enhancing endurance running performance. They can help coaches and scientists understand individual physiological differences in athletes and present a potential approach for individualized training prescription.

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# Supplementary data

**Supplementary data 1** Stepwise regression analysis of time trial performance and physiological parameters including model equations

Time-trial	Model	R <sup>2</sup>	Δ R <sup>2</sup>	Resid. Std. Error (m·s <sup>-1</sup> )	p-value	AIC	Equation
velocity 100 m	VL <sub>Bmax</sub>	0.60		0.308	< 0.0001	-45,225	$y = 2.28 (VL_{Bmax}) + 6.16$
	VL <sub>Bmax</sub> + %VO <sub>2max</sub>	0.72	0.12	0.265	< 0.0001	-50,363	$y = 2.36 (VL_{Bmax}) - 3.91 (VO_{2max}) + 9.38$
	VL <sub>Bmax</sub> + %VO <sub>2max</sub> + VO <sub>2max</sub>	0.80	0.08	0.230	< 0.0001	-55,383	$y = 2.79 (VL_{Bmax}) - 3.37 (\%VO_{2max}) + 0.03(VO_{2max}) + 6.77$
velocity 1000 m	VO <sub>2max</sub>	0.46		0.256	0.001	-52,563	$y = 0.04 (VO_{2max}) + 3.08$
	VO <sub>2max</sub> + VL <sub>Amax</sub>	0.53	0.07	0.246	0.003	-53,383	$y = 0.05 (VO_{2max}) + 0.64 (VL_{Amax}) + 2.05$
	VO <sub>2max</sub> + VL <sub>Amax</sub> + MFO	0.58	0.05	0.240	0.004	-53,523	$y = 0.04 (VO_{2max}) + 0.69 (VL_{Amax}) + 0.76(MFO) + 2.14$
	VO <sub>2max</sub> + VL <sub>Amax</sub> + MFO + RE <sub>M,SS</sub>	0.62	0.05	0.234	0.004	-53,884	$y = 0.04 (VO_{2max}) + 0.94 (VL_{Amax}) + 0.78(MFO) - 0.01 (RE) + 4.00$
	VO <sub>2max</sub>	0.65		0.203	< 0.0001	-61,858	$y = 0.05 (VO_{2max}) + 2.04$
velocity 2000 m	VO <sub>2max</sub> + MFO	0.78	0.13	0.164	< 0.0001	-69,552	$y = 0.04 (VO_{2max}) + 1.26 (MFO) + 2.34$
	VO <sub>2max</sub> + MFO + RE <sub>M,SS</sub>	0.83	0.05	0.150	< 0.0001	-72,355	$y = 0.03 (VO_{2max}) + 1.23 (MFO) - 0.01 (RE) + 4.32$
	VO <sub>2max</sub> + MFO + RE <sub>M,SS</sub> + VL <sub>Bmax</sub>	0.85	0.02	0.144	< 0.0001	-73,277	$y = 0.03 (VO_{2max}) + 1.29 (MFO) - 0.01 (RE) + 0.40 (VL_{Bmax}) + 4.36$
velocity 3000 m	VO <sub>2max</sub>	0.71		0.206	< 0.0001	-61,317	$y = 0.06 (VO_{2max}) + 1.26$
	VO <sub>2max</sub> + MFO	0.83	0.12	0.160	< 0.0001	-70,531	$y = 0.04 (VO_{2max}) + 1.35 (MFO) + 1.57$
	VO <sub>2max</sub> + MFO + RE <sub>M,SS</sub>	0.88	0.05	0.139	< 0.0001	-75,327	$y = 0.04 (VO_{2max}) + 1.32 (MFO) - 0.01 (RE) + 3.82$
	VO <sub>2max</sub> + MFO + RE <sub>M,SS</sub> + %VO <sub>2max</sub>	0.93	0.05	0.113	< 0.0001	-83,004	$y = 0.04 (VO_{2max}) + 1.05 (MFO) - 0.01 (RE) + 2.02 (\%VO_{2max}) + 1.98$

**Supplementary data 2** Stepwise regression analysis of time trial performance and performance parameters including model equations

Time-trial	Model	R <sub>2</sub>	R <sub>2adj</sub>	Δ R <sub>2adj</sub>	Resid. Std. Error [m·s <sup>-1</sup> ]	F	p	AIC	Equation
velocity 100 m	D'	0.478	0.449		0.352	16,490	< 0.001	-39,845	$y = 0.005 (D') + 6.753$
	D' + CV	1.000	0.529	0.080	0.340	9,533	0.002	-39,884	$y = 0.007 (D') + 0.387 (CV) + 4.569$
velocity 1000 m	VVO2max	0.578	0.554		0.226	24,620	0.0001	-57,532	$y = 0.642 (VVO2max) + 2.241$
	VVO2max + D'	0.760	0.732	0.178	0.176	26,960	< 0.0001	-66,858	$y = 0.805 (VVO2max) + 0.003 (D') + 0.821$
	VVO2max + D' + CV	0.971	0.965	0.411	0.063	176,200	< 0.0001	-106,840	$y = 0.113 (VVO2max) + 0.006 (D') + 1.077 (CV) - 1.053$
velocity 2000 m	CV	0.795	0.784		0.155	69,750	< 0.0001	-72,596	$y = 0.721 (CV) + 1.875$
	CV + D'	0.982	0.979	0.196	0.048	450,700	< 0.0001	-118,702	$y = 1.110 (CV) + 0.003 (D') - 0.633$
velocity 3000 m	CV	0.968	0.967		0.061	549,100	< 0.0001	-105,554	$y = 0.888 (CV) + 0.847$
	CV + D'	0.999	0.998	0.032	0.015	5816,000	< 0.0001	-165,145	$y = 1.062 (CV) + 0.002 (D') - 0.280$

## Affirmation in lieu of an oath

Herewith I affirm in lieu of an oath that I have authored this thesis independently and did not use any other sources and tools than indicated. All citations, either direct quotations or passages which were reproduced verbatim or nearby-verbatim from publications, are indicated and the respective references are named. The same is true for tables and figures. I did not submit this piece of work in the same or similar way or in extracts in another assignment.

A handwritten signature in black ink, appearing to read 'J. Schz', written above a horizontal line.

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Cologne, 22.08.2022

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Signature