The use of near-infrared spectroscopy (NIRS) in the physiological assessment of sprint triathlon.

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Thesis Abstract

Sprint triathlon is a high intensity endurance discipline with ever-growing participation rates, but the sport currently remains under researched. The requirement to complete swim, cycle and run stages consecutively places unique physiological demands upon triathletes that are superior to that of completing identical distances singularly. Despite the knowledge that sprint triathlon is greater than the sum of its parts, the inclusion of all three disciplines in triathlon research is rare. A review of the literature identified a wide variety of measurement tools that have been utilised to observe the global physiological responses (oxygen consumption, heart rate, blood lactate concentration among others) that occur during participation. Despite this wealth of information, it is complex to piece together the findings into a larger picture of performance, with a particular lack of swimming related research. A single study from the review looked to utilise Near-Infrared Spectroscopy (NIRS) within triathletes. NIRS is a light-based technology that reports on the relationship between oxygen delivery and utilisation at the site of gas exchange in the muscle, a key indicator of performance in aerobic events such as sprint triathlon. A laboratory based experimental study explored the utility of multi-site NIRS as a measurement tool within triathlon using recreational male triathletes (n=11). A comprehensive profile of global and peripheral responses across the triathlon simulation was created, identifying the measures associated with performance. NIRS yielded different oxygenation responses between upper and lower limbs throughout and identified a greater peripheral measurement variability between participants compared to global physiological measures. It is suggested this variability highlights differences in efficiency between athletes, although no direct correlations were found between NIRS data during swim, cycle and run and overall performance. As a measurement tool NIRS has the potential to increase the specificity of physiological information available when creating strategies to be applied in sprint triathlon training and competition.

Key words: Sprint triathlon, NIRS, Physiological Profile, Laboratory, Performance

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Abbreviations

ε	Specific Extinction Coefficient
%SMO_2	Skeletal Muscle Oxygen Saturation
ATP	Adenosine Triphosphate
BB	Biceps Brachii
BLC	Blood Lactate Concentration
CW (NIRS)	Continuous Wave NIRS
CYTOX	Cytochrome Oxidase
EMG	Electromyography
FD (NIRS)	Frequency Domain NIRS
Hb	Haemoglobin
HHb	Deoxygenated Haemoglobin
HR	Heart Rate
LD	Latissimus Dorsi
LT	Lactate Threshold
Mb	Myoglobin
MLSS	Maximal Lactate Steady State
MRS	Magnetic Resistance Spectroscopy
NIRS	Near-Infrared Spectroscopy
O_2	Oxygen
O_2Hb	Oxygenated Haemoglobin
PAR-Q	Physical Activity Readiness Questionnaire
PCr	Phosphocreatine
PPO	Peak Power Output
RPE	Rate of Perceived Exertion

SRES	School of Sport, Rehabilitation and Exercise Sciences
SRS	Spatially-Resolved Spectrometers
StO_2	Tissue Oxygen Saturation
T1	Swim-Cycle Transition
T2	Cycle-Run Transition
TD (NIRS)	Time Domain NIRS
tHb	Total Haemoglobin
TOI	Tissue Oxygenation Index
TSI	Tissue Saturation Index
$\dot{V}E$	Minute Ventilation
VL	Vastus Lateralis
$\dot{V}O_2$	Oxygen Consumption
$\dot{V}RMO_2$	Respiratory Muscle Oxygen Consumption
VT	Ventilatory Threshold

1. Thesis Overview

1.1. Thesis Justification

The sequential combination of high intensity swimming, cycling and running performed during a sprint triathlon places unique demands upon the triathlete, different to those experienced in isolated versions of the three disciplines. Despite the knowledge of the residual effects each discipline has on overall performance, much of the laboratory simulated literature has focused on swim-cycle (T1) or cycle-run (T2) transitions (1), with few studies simulating the event in its entirety (2). The necessity to perform well across all three disciplines often leads to high volumes of training as triathletes combine events with discipline-specific skills to replicate the demands of competition (3, 4). However, with a lack of research into the physiological responses of triathletes during a sprint triathlon (5), or recommendations on how to balance the disciplines in training (6), athletes and coaches are forced to translate research from the isolated event literature in which the physiological profile is different (7). With the sprint triathlon discipline being considered an endurance event (8), it is often maximal oxygen consumption, ventilatory and lactate thresholds, and measures of exercise economy that researchers observe (9). However, with new wearable technologies capable of additional bio-feedback it is important that these measures are explored to investigate whether this gives us new insight. Wearable muscle oxygenation sensors that utilise Near-Infrared Spectroscopy (NIRS), a light-based technique that reports on the relationship between oxygen delivery and consumption at the sight of gas exchange, in the muscle, is an example of such technology (10, 11). As sprint distance triathlon is recognized as an aerobic event, oxygen delivery and utilisation should be a key indicator of performance. The portable and non-invasive nature of NIRS devices, paired with its ability to measure in aquatic environments, combats some of the intrinsic limitations with more commonly used physiological measurement tools. In addition, NIRS provides the researcher with a different physiological measurement that may hold value. NIRS is a growing area of research and has previously been applied to swim, cycle and run disciplines individually, and whilst researchers have already shown an interest in using this technology within triathletes, it has yet to be used in triathlon. With the logistical and practical difficulties of carrying out physiological testing in the field, a simulated triathlon is a reliable method in order to investigate the physiological parameters and associated responses during a sprint triathlon via a controlled environment (12). This testing modality allows for the assessment of both global and peripheral measures and gives researchers the ability to collect data specific to the physiological demands placed upon a triathlete. This in turn could be used to inform upon physiological demand, future training and competition strategies.

1.2. Thesis Aims

This thesis aims to understand the applicability and utility of NIRS as a measurement tool within triathlon. In order for this to be achieved, two studies were conducted:

Study 1: Literature review of physiological measurement tools currently used within sprint distance triathlon.

Specific study aims:

1. Identify the physiological measures that have currently been assessed during sprint distance triathlon.
2. Understand the factors that may affect the physiological responses observed in triathletes.

3. Highlight the physiological variables that are considered important for overall performance during sprint distance triathlon.

Study 2: The physiological assessment of simulated sprint distance triathlon, incorporating NIRS to obtain local muscle oxygenation responses.

Specific study aims:

1. Explore the utility of NIRS as a new measurement tool within triathlon.
2. Produce a full oxygenation profile, including swimming response, to triathlon.
3. Create a complete profile of the physiological responses to triathlon.
4. Identify the physiological responses associated with triathlon performance.

1.3. Thesis Outline

Section 1 and the introduction to the literature review provides reasoning for this work, while the main body of the review in Section 2 looks to identify current research understanding of the physiological assessment of sprint distance triathlon. The literature review combined with following NIRS narrative in Section 3 form the rationale for the experimental study. In Section 4, the experimental study is presented and designed to: (a) explore the utility of NIRS as a new measurement tool within triathlon; (b) produce a full oxygenation profile including swimming response; (c) create a profile of the physiological responses to triathlon; and (d) identify the physiological responses associated with triathlon performance. The thesis is concluded in Section 4.7 where the experimental findings are discussed and recommendations for future research are provided.

2. Literature Review

2.1. Abstract

Sprint triathlon has become popular with recreational athletes due to its accessibility, however research into this variation is limited. Each stage of the triathlon challenges the respiratory, cardiovascular and musculoskeletal systems in different ways, with demands different to completing the same distances in an isolated environment. The literature review aimed to identify the physiological measures that have currently been assessed during a sprint triathlon. Studies were selected if they included physiological data regarding relative intensity, influence of prior disciplines, predictors of performance or factors affecting the physiological responses of triathletes. Studies typically observed the global physiological responses (oxygen consumption, heart rate, blood lactate concentration among others) that occur during participation, with frequent calculations of intensity compared to maximal exercise. A large proportion of research explored the effects of the bike to run transition, but the underlying physiological changes are yet to be understood. There is an abundance of literature looking to predict performance through laboratory data but there was a relative lack of research focus on the swim stage. Age, ability and competition strategies were factors considered to alter an athlete's physiological response. Due to inconsistencies of measurement tools and methodologies, it is complex to piece together the findings into a larger picture of performance. A single study from the review looked to utilise Near-Infrared Spectroscopy (NIRS) within triathletes. NIRS is a light-based technology reports on the relationship between oxygen delivery and utilisation in the muscle, a key indicator of performance in aerobic events. NIRS has been used effectively within swim, cycle and run disciplines individually, but has yet to be applied to a full triathlon. The emergence of new technologies suggests that there may be more to uncover within the sport of triathlon, and future work should look to create a physiological profile including both global and peripheral measures.

Key words: Sprint triathlon, NIRS, Physiological Profile, Laboratory, Performance

2.2. Introduction

Triathlon consists of three disciplines and two transitions where athletes swim, cycle, and run consecutively over a variety of distances. This varies from short distance (super sprint/sprint, ~1 hour) to long course (Ironman, 14+ hours) and the recent addition of a mixed team sprint relay (4x ~ 20 minute legs) (13). The sprint distance triathlon (0.75 km swim, 20 km bike, 5 km run) is a high intensity discipline which places very different physiological demands on the athlete in comparison to the long steady state efforts of an Ironman. This shorter modality has become popular with recreational athletes due to its accessibility, however research into this variation is limited (5). Between disciplines athletes enter a transition area where they have to efficiently prepare themselves for the upcoming stage of the race, changing clothing and equipment for swim-cycle, T1, and cycle-run, T2. The ability to transition effectively and cope with the change in demands that this brings has been recognized as an important part of success within the sport (3, 14), with importance emphasized in shorter disciplines such as the sprint triathlon. Each section of the triathlon challenges the respiratory, cardiovascular and musculoskeletal systems in different ways, with these demands differing to those observed when completing the same distances in an isolated environment, making the triathlon greater than the sum of its parts. Despite increases in its popularity since its introduction to the Olympics in Sydney 2002, coaches are often having to translate findings from single sport research to improve their athletes due to a lack of triathlon specific research (7). Previous research has examined the physical characteristics of a triathlete (8, 15, 16), their training loads (17-19) and pacing (20-24) and nutritional strategies (19, 25, 26), as well as investigating injury rates (19, 27, 28), sex differences (29-32), and age-related declines in performance (31-34). Improvements in technology have made it possible for researchers to monitor physiological responses to a given exercise, both in the laboratory and in the field, in real time. As well as being able to observe global measures of oxygen consumption ($\dot{V}O_2$), heart rate (HR) and blood lactate concentration (BLC) to assess the physiological changes that occur with participation in the sport, new technologies also allow for the accurate measurement of peripheral measures such as specific muscle oxygenation. Therefore, the aim of this literature review is to identify the physiological measures that have currently been assessed during a triathlon, understand factors that may affect the physiological response, and highlight variables important to overall performance. The purpose is to provide researchers, triathletes and coaches with the current understanding of the demands of the sport which can be used to inform future training and competition strategies.

2.3. Methods

A search of relevant databases was conducted to identify articles suitable for this review: Web of Science, SPORTDiscus and Pubmed. Reference lists of the selected articles were examined. The search terms used were: ‘sprint distance triathlon’ OR ‘Olympic Triathlon’ OR ‘simulated triathlon’ OR ‘triathletes’, AND: ‘profile’ OR ‘physiological’ OR ‘cardiorespiratory’ OR ‘cardiovascular’ OR ‘ventilatory’ OR ‘Internal load’ OR ‘oxygen uptake’ OR ‘energy cost’ OR ‘economy’ OR “muscle oxygenation” OR ‘heart rate’ OR ‘lactate’ OR ‘threshold’ OR ‘exertion’ OR ‘transition’ OR ‘power’ OR ‘characteristics’ OR ‘performance’, NOT: ‘injury’ OR ‘illness’ OR ‘ironman’ OR ‘heat’ OR ‘altitude’. Studies were selected if they included physiological data regarding relative intensity, influence of prior disciplines, predictors of performance or factors affecting the physiological responses of triathletes. Research in peer-reviewed journals concerning swim, cycle and/or run stages, as well as a complete triathlon effort from Sprint and Olympic distances are discussed. An initial search looking at sprint distance triathlon exclusively resulted in 4 eligible articles and was not suitable to review, therefore the search was widened to include Olympic and simulated distances. A total of 75 studies were included in this review and the selection process is illustrated by a flow chart in Figure 1.

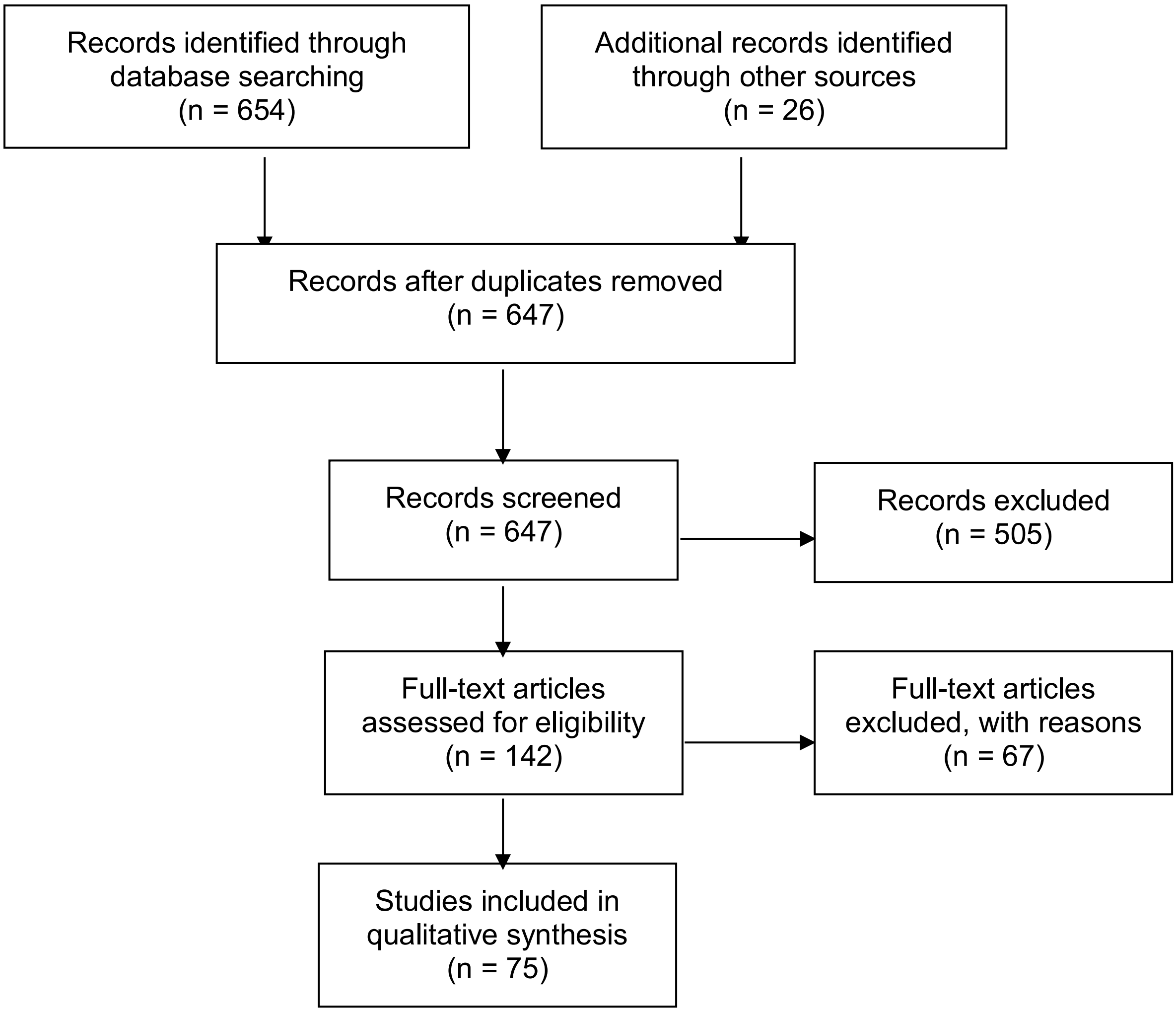


Figure 1. Flow chart summary of the study selection process.

2.4. Discussion

2.4.1. Swim

Sprint triathlon events are typically initiated by a mass start in the 750m swim, this is the shortest stint of the race and typically lasts around 10 minutes (35). There has been minimal research studies that have investigated the physiological demands of triathlon swimming (2, 12, 35), which is likely to be influenced by early suggestions that swim completion time had little effect on overall race time (36, 37). However, more recently these thoughts have been reversed with several studies highlighting the importance of the swim in Olympic distance triathlon (2, 38), recognizing it as crucial within sprint distance triathlons (24). The swim section of a sprint distance triathlon accounts for the least proportion of variance in overall completion time (9%), but is an important part of the race for tactical reasons (39). These shorter disciplines are typically draft-legal events, allowing athletes to benefit from the effects of drafting in the swim and cycle stage of the race. Therefore, the swim holds great importance as exiting the water within the lead group allows athletes to execute drafting tactics, increase output at a given intensity and provide them a significant advantage on both the bike and subsequent run (1, 38, 40, 41). Those who do not make it out of the water in the lead group are required to put in greater initial cycling effort, a strategy that negatively effects overall performance (38).

The mean intensity observed during an triathlon swim has been profiled at an average intensity of 85.2 ± 1.5% HRmax and 5.7 ± 1.8 - 6.5 ± 1.1 mmol/L BLC upon completion of the swim stage (12, 42, 43). It must be noted that HRmax was calculated from a treadmill test (42), a different exercise modality that will have a modality specific response. The initial accumulation of BLC is carried over into subsequent stages in the race (12, 43), meanwhile a single study measuring ratings of perceived exertion (RPE) rated the effort a 15 ± 1 and 13 ± 2 in non-drafting and drafting swims respectively (43).

As swimming is the first of the three disciplines athletes have to conserve as much energy as possible, whilst swimming at a competitive pace. Research has proposed triathletes complete the swim at a lower intensity, 80% of time trial pace, to improve the physiological condition of the athlete in the first transition. In doing so BLC is significantly reduced upon the completion of the swim (5.2 ± 2.5 versus 9.1 ± 2.7 mmol/L, $p< 0.05$), with carry over effects into the first 5 minutes of the cycle stage (8.1 ± 4.4 versus 11.3 ± 2.5 mmol/L, $p< 0.05$) (2). Selecting this reduced swimming intensity also leads to lower RPE at the end of both swim and cycle stages, $p< 0.01$, but no differences are observed at the completion of the run, $p< 0.05$. Despite research support this strategy does not translate to what is typically seen in competition (23). Further altering the physiological demands of the swim, performance can be influenced by wearing a wetsuit, as well as drafting behind competitors (44). A brief outline of the impacts of drafting on physiological measures is discussed in section 2.5.5. Economical gains can be made through the use of a wetsuit, with 3.2% improvements in 750m swim time being observed in comparison to standard swimming attire; such gains are made with no additional work required from the athlete, indicated by comparable levels of BLC (7.1 ± 0.9 versus 7.8 ± 1.1 mmol/L) (45). As well as BLC, the use of wetsuits have been shown to reduce HR, and RPE to large effect sizes (Cohens *d*: BLC 2.55; HR 11.93; RPE 1.23) (46), with 6% improvement in velocity seen over the first 400m of triathlon swimming (47). The use of a wetsuit, or executing drafting tactics, gives the athlete the ability to reduce work rate for a given speed during the swim, and therefore providing an energy reserve that can be utilised to improve performance in cycle and run stages (2, 41).

Conducting reliable physiological testing of swimming brings logistical and practical difficulties. Some researchers have acquired access to a flume pool in order to complete their studies or used tethered swimming to keep their participants in the water, whilst others have used dry-land swim ergometry techniques. The differences in physiological response to these variations in swimming modalities must be considered when comparing the results of the literature. Although possible, global oxygen data is rarely recorded due to the complexity of measurement.

Table 1. Methodologies used in swimming

Swim method	Purpose of swim	Study
25m Pool	Determining exercise intensities at Maximal Lactate Steady State	Van Schuylenbergh et al. (48)
	Swim-cycle trial – Drafting	Bentley et al. (44)
	Swim-cycle trial – Wetsuit	Peeling & Landers (45)
	Simulated sprint triathlon	Taylor et al. (12)
	Simulated sprint triathlon	Binnie et al. (35)
	Simulated sprint triathlon	Peeling et al. (2)
	Swim time trial – Predicting triathlon performance	Schabort et al. (37)
	Sprint triathlon pacing	Wu et al. (24)
	Time trial to determine pace	Gay et al. (47)
50m Pool	Simulated Olympic triathlon	Lopes et al. (42)
	Swim-cycle trial	Delextrat et al. (49)
	Swim-cycle trial	Delextrat et al. (41)

	Swim-cycle trial – Drafting	Delextrat et al. (43)
Open Water Swim	Observing Olympic triathlon competition	Horne. (39) Landers et al. (16) Vleck et al. (38)
Swim Ergometer (Vasa)	Muscle activity wearing wetsuit	Agnelli & Mercer. (50)
Swimming Flume (Endless Pool Elite Techno Jet Swim 7.5 HP)	Time trial (influence of wetsuit on physiology)	Gay et al. (47)
Tethered Swimming	Understanding physiological responses to maximal exercise	Sleivert & Wenger (51)

Table 2. Measurement tools used in swimming

Measure recorded	Measurement tool	Study
Blood Lactate Concentration	Blood gas analyser – Earlobe (ABL 625)	Binnie et al (35)
		Delextrat et al (41)
		Delextrat et al (43)
		Peeling & Landers (45)
		Hausswirth et al (52)
		Taylor & Smith (53)
	Blood gas analyser – Fingertip (Lactate Pro)	Taylor et al (12)
		Bentley et al (44)
		Gay et al (47)
	Blood gas analyser – Undetermined location (Lactate Pro, YSP 1500 STAT)	Lopes et al (42)
		Taylor & Smith (53)
Heart Rate	Heart rate monitor (CardioSwim, Polar Accurex, Polar Electro, Polar RS200, Polar S610, Polar S810i, Polar Vantage)	Binnie et al (35)
		Delextrat et al (41)

		Delextrat et al (43)
		Gay et al (47)
		Hausswirth et al (52)
		Lopes et al (42)
		Peeling & Landers (45)
		Taylor et al (12)
		Wu et al (24)
		Taylor & Smith (54)
Rating of Perceived Exertion	Borg Scale (6-20)	Bentley et al (44)
		Binnie et al (35)
		Delextrat et al (41)
		Gay et al (47)
		Olcina et al (55)
		Taylor & Smith (53)
		Wu et al (24)

2.4.2. Swim-to-Bike Transition (T1)

Following the swim triathletes enter into the first transition, T1. Here they start to remove their wetsuit whilst running to their personal transition area, before putting on their cycling shoes and helmet and setting off for the cycle stage of the race. There is no uniform distance for a transition area, as these vary from course to course, although typical time spent in each transition is around one minute (56). The physiological cost of this section of the race has been investigated, finding a heightened $\dot{V}O_2$ (~3%) when running with the wetsuit halfway down versus wearing the wetsuit fully up or running whilst carrying the wetsuit ($p < 0.05$) (57). HR was unchanged in each condition in this study. The triathlete must however weigh up the benefits of energetic efficiency versus time efficiency when selecting a transition strategy to be used in competition.

The predominant use of the upper body muscle groups when swimming can result in blood pooling in the arms, causing difficulties when transitioning to lower body focussed cycling, and delaying the redistribution of blood when moving from prone (swimming) to erect (running) and seated (cycling) position (13, 40). Although stated within the research it is important to note no measurement of blood flow was taken, and instead the theory proposed from the knowledge of basic physiology. The effect on swimming on subsequent cycling has been researched, although somewhat lightly in comparison to the cycle to run transition, thought to be a result of a proposed weak correlation between swim duration and overall race time (r= 0.30) (36, 39). It has been shown that short duration high intensity swimming may significantly influence subsequent cycling and overall triathlon time and physiological cost (1, 2, 49). In comparison to an isolated cycle trial, prior swimming has shown reduced efficiency (~13%), increased BLC (~56%), minute ventilation ($\dot{V}E$) (~16%), and elevated $\dot{V}O_2$

(~5%) in the first 5 minutes of cycling (49). In this trial only BLC (~44%) and $\dot{V}E$ (~8%) remained significantly different to isolated cycle exercise after 30 minutes, showing the effects are most pronounced at the onset of cycle exercise but do not last across the whole cycle stage. In a simulated sprint distance triathlon opting for 80% or 90% intensity of a prior swimming time trial effort was shown to improve subsequent cycling time, although only the 80% intensity lead to improvement in overall triathlon time when compared to 100% time trial pace $p < 0.05$ (2). It was inferred that triathletes should aim to increase their swimming ability so that they could reach the speeds required to be in the first pack of swimmers whilst working at 80 - 90% of maximal intensity. However previous knowledge suggests that a constant pacing strategy may not be best suited to the mass start nature of the sprint triathlon (24).

2.4.3. Bike

The 20 km cycle is the stage of the longest duration of the sprint triathlon and lasts around 40 minutes (35), although this will vary from course to course, and accounts for the greatest proportion of variance in overall completion time (59%) (39). It has been proposed that this stage of the race should be performed at maximal intensity in order to optimize overall triathlon time (8). The mean intensity of the cycle stage within a simulated sprint triathlon equates to 86 - 90% HRpeak, 74 - 82% $\dot{V}O_2$peak and 6.98 ± 0.74mmol/L BLC. Further to this triathletes operate at 68.2 ± 7.2% peak power output (PPO (Watts)), where a 10% decrease in power output from the first third to final third of cycle stage is reported (12, 14, 42, 58). Due to the requirement for short high intensity bursts, and variable race dynamics during the cycle stage, the true workload for this stage is greater than that of the mean power output (58). It has been shown that after 5 minutes triathletes' HR and $\dot{V}E$ increase progressively throughout ($p<$

0.05), despite power output and $\dot{V}O_2$ remaining constant. It has also been outlined that triathletes opt for a cycling cadence of ~90 rpm (59, 60). This cadence has been observed during simulated sprint triathlons despite it being greater than the 60-80rpm cadence considered metabolically optimal (52). When comparing 60, 80 and 100rpm cadences, the physiological response of the latter was categorized by higher HR (~5%) and $\dot{V}E$ (~12%) ($p < 0.05$), with BLC values of 7.0 ± 2.0 mmol/L (100rpm) versus 4.6 ± 2.1 mmol/L (60rpm) suggesting varying contributions of anaerobic metabolism and overall oxygen cost is dependent on cadence selection (61). It is recommended that triathletes cycle at a lower intensity in the final section of the cycle stage, in order to optimise running performance (25), although this is an unlikely strategy in competition. A substantial increase in pace is typically seen in the closing stages of the cycle (~1 km) in attempt to attain a useful position before entering the second transition area, T2, where being positioned near the front of a group would decrease time spent in transition (56). The ability to efficiently dismount and rack the bicycle, take off the helmet and put on running shoes can be crucial to overall finishing position (56).

Table 3. Measurement tools used in cycling

Measure recorded	Measurement tool	Study
Blood Lactate Concentration	Blood gas analyser – Earlobe (ABL 625)	Bernard et al (61)
		Binnie et al (35)
		Delextrat et al (41)
		Delextrat et al (43)
		Delextrat et al (49)
		Hausswirth et al (20)
		Hue et al (62)
		Peeling & Landers (45)
		Peeling et al (2)
	Blood gas analyser – Fingertip (Lactate Pro)	Bentley et al (44)
		Hausswirth et al (52)
		Taylor et al (12)

	Blood gas analyser – Undetermined location (Lactate Pro, YSP 1500 STAT)	Bonacci et al (63)
		Lopes et al (42)
		Taylor & Smith (53)
Core Temperature	Ingestible temperature measurement pill (CorTemp)	Binnie et al (35)
		Taylor & Smith (53)
Heart Rate	Heart rate monitor (BHL-6000, Garmin, Polar Accurex, Polar Accurex plus, Polar Electro, Polar Racer, Polar RS200, Polar S810i, Polar Vantage, Polar Vantage XL)	Bentley et al (44)
		Bernard et al (61)
		Binnie et al (35)
		Delextrat et al (41)
		Delextrat et al (43)
		Delextrat et al (49)
		Hausswirth et al (52)
		Hausswirth et al (64)
		Hue et al (14)
		Hue et al (62)
		Lepers et al (60)

		Lopes et al (42)
		Millet et al (65)
		Olcina et al (55)
		Peeling & Landers (45)
		Taylor & Smith (54)
		Taylor et al (12)
		Walsh et al (66)
		Wu et al (24)
Muscle Activation	Electromyography (EMG) (Delsys, Mega electronics, Noraxon)	Bonacci et al (63)
		Heiden & Burnett (67)
		Walsh et al (66)
Muscle Oxygenation	Near-infrared Spectroscopy (NIRS) (Moxy)	Olcina et al (55)
Power Output	Cycle ergometer (Cyclemax, Lode Excalibur, TacX Fortius)	Bernard et al (61)
		Binnie et al (35)
		Peeling & Landers (45)
		Peeling et al (2)

		Schabort et al (37)
		Wu et al (24)
	Power meter (SRM crankset)	Bentley et al (44)
		Etxebarria et al (58)
Rating of Perceived Exertion	Borg Scale (6-20)	Bentley et al (44)
		Binnie et al (35)
		Bonacci et al (63)
		Delextrat et al (43)
		Olcina et al (55)
		Peeling et al (2)
		Taylor & Smith (53)
		Wu et al (24)
Minute Ventilation	Respiratory gas analyser (Cosmed K4, CPX, MGA-1100)	Bernard et al (61)
		Delextrat et al (41)
		Delextrat et al (49)
		Hausswirth et al (20)

		Hausswirth et al (52)
		Hue et al (14)
		Hue et al (62)
		Lepers et al (60)
		Millet et al (65)
		Taylor & Smith (54)
		Taylor et al (12)
		Vercruyssen et al (68)
Oxygen Consumption	Respiratory gas analyser (Ametek Gas analyser, Cortex Metalyser, Cosmed K4, CPX, MGA-1100, Parvo TrueMax 2400)	Bentley et al (44)
		Bernard et al (61)
		Delextrat et al (41)
		Delextrat et al (43)
		Delextrat et al (49)
		Hausswirth et al (20)
		Hausswirth et al (52)
		Hausswirth et al (64)

Hue et al (14)

Hue et al (62)

Lepers et al (60)

Millet et al (65)

Peeling et al (2)

Schabort et al (37)

Taylor & Smith (54)

Taylor et al (12)

Vercruyssen et al (68)

Walsh et al (66)

2.4.4. Bike-to-Run Transition (T2)

A large area of triathlon cycling research is the cycle to run transition where effective transitioning is complex and is yet to be fully understood (69). Following the cycle stage triathletes have to negotiate T2, dismounting their bike and placing it on their allocated rack, removing their helmet and swapping footwear, before setting off for the final stage of the race. The transition from non-weight bearing cycling to the impact forces created in running, and the associated change in movement frequency, often leads to recreational athletes experiencing a loss of coordination in this transition (56, 64, 67). It appears this phenomenon may be trainable, where coordination loss has not been reported in elite athletes (65), and multiple high intensity cycle-run repetitions has been reported to improve the transition between disciplines (70). The stacking of multiple disciplines within an exercise routine, known as ‘brick training’, is often used in triathletes training programs to aid transition performance and combat the potential negative effects it can cause (55).

The accumulated fatigue of the prior disciplines, coupled with the stresses involved with T2, results in triathlon running having different physiological demands and responses to that of isolated running (40, 66). Early research showed how $\dot{V}O_2$ (~7%) (14, 56, 64, 71), $\dot{V}E$ (~16%) (14, 56, 62, 65) and HR (~4%) (14, 40, 56) are all elevated in comparison to an isolated running control ($p < 0.05$). Some authors have used video analysis to suggest biomechanical measures remain unchanged (14), proposing metabolic factors such as fatigue constitute to the greater energy cost of running during a triathlon. However others using video replays have observed decreased stride length and step cadence following prior cycling, which altered muscular recruitment, reduced efficiency of the stretch-shortening cycle, and therefore impacted the overall metabolic cost of running by up to 12% (4, 56, 69). It is speculated that these changes are evoked partly due to reduction of glycogen stores during the earlier part of the race that affect running performance of the triathletes (14, 64, 72). Researchers have used mathematical formulas using $\dot{V}E$ to calculate the work of breathing and subsequently used this to establish the oxygen uptake in respiratory muscles ($\dot{V}RMO_2$). A 38% increase of $\dot{V}RMO_2$, influenced by fatigue of the inspiratory muscles, has been observed to account for 8.5% variance in the energy cost of triathlon running in middle-level athletes (65), and is another possible factor in the cycle-run transition. Here energy cost was calculated as $\dot{V}O_2$ above basal metabolic rate, as a function of running velocity. Combined EMG and video analysis has shown changes in kinematic variables and associated ground reaction forces, such as the ankle angle at foot contact, to explain 67.1% of the variance in $\dot{V}O_2$ when running after cycling (63), and hence provides a greater understanding of the complexities experienced during this stage of the race.

It must be noted that the negative effects of cycling on subsequent running performance are more pronounced in recreational triathletes compared to those who are highly trained (25, 56, 63, 65). Prior moderate intensity cycling does not cause significant changes to EMG activity, $\dot{V}O_2$ or HR values during subsequent running in highly competitive triathletes (66). Despite differences in physiological demands, perceptual responses of: initial RPE, rate of RPE increase and final RPE values remain comparable between isolated and triathlon running (53). Researchers have suggested that RPE values may be influenced by local muscle discomfort at high work intensities (73), and therefore it could be that a change in active musculature between disciplines resets RPE and results in this similarity.

The greatest elevation in the physiological cost of $\dot{V}E$, HR and BLC are seen in the initial portion of the run (74), in the first 500m following transition, where initial running speed is related to prior cycling cadence. This increase in speed has been observed when comparing 60, 80 and 100rpm cycling cadences, but appears to be transitory, with no significant differences in overall running performance (61). Athletes who cycle at their freely chosen cadence (81.2 ± 7.2 rpm) during the cycle experience increased $\dot{V}O_2$ uptakes (10.4%) when compared to an isolated run (68). However, with energetically optimal cadence (72.5 ± 4.6 rpm) during the cycle showing no adverse effects on the subsequent run, it is evident that cadence and intensity alters the physiological response to triathlon. The fraction of $\dot{V}O_2max$ at which triathletes can sustain during the run is also influenced by cycling cadence, 92% (60rpm) versus 84% (100rpm), suggesting triathletes may start to feel the effects of fatigue earlier if they select a high cadence during the cycle due to metabolic acidosis (61). These results add to the proposals that the sum of the optimal strategies for each individual discipline may not be equal to the optimal strategy for the whole triathlon race. It has also been observed that skeletal muscle oxygen saturation (%SmO_2) is higher in the active muscles during running when proceeded by cycling to a large effect (Cohens *d*: 1.63, $p < 0.001$), with the previous exercise causing an inability to peripherally utilise oxygen in the muscles and therefore hindering the capacity to increase exercise intensity (55). %SmO_2 is the volume of oxyhemoglobin (O_2Hb) relative to O_2Hb + deoxyhemoglobin (HHb) and therefore provides insight on the oxygen status within the muscle. These changes have seen during brick training whilst no other effects on kinematics, physiological or perceptual parameters were evident, although this may be explained by the exclusion of swimming prior to the cycling effort. Despite the understanding of the effects of prior cycling on running performance, the underlying physiological, biochemical and neuromuscular responses are not fully understood (40), but are likely to be influenced by the complex build-up of central and peripheral fatigue mechanisms (13).

2.4.4. Run

The 5 km run is the final stage of the sprint triathlon and is typically completed in around 20 minutes (35), accounting for the second greatest proportion of variance in overall completion time (31%) (39). With a greater variability in athletes' performance compared to the prior disciplines and a strong correlation between running speed and finishing position (38), it holds a critical importance to the overall race achievement (16, 38). This final section of the race is completed at an intensity corresponding to 91.9 ± 1.9% HRpeak, 87.5 ± 3.0% $\dot{V}$Epeak, 83-90% $\dot{V}O_2$peak, and 7.6 ± 0.9 mmol/L BLC (12, 14). There is research support for running at 5% slower than 10 km pace during the first kilometre of the run, in order to reduce fatigue development and improve overall running performance (20). It would be optimal to slowly increase running speed until ventilatory, cardiovascular, and neuromuscular steady state is achieved, and triathletes should avoid an aggressive strategy despite a motivation to follow direct opponents (21). However, such tactics are not realistic in competition and it appears that the fast start strategy is a more effective strategy during sprint distance racing (23, 71). Even during maximal effort running it has been observed that triathletes possess a reserve capacity which has been obtained by deception of running speed over the first 1.66 km. This was seen to improve overall running performance by 1% (54), a worthwhile enhancement (75), and provides support for the selection of an aggressive fast start strategy previously opposed in the literature (20, 21)

Table 4. Measurement tools used in running

Measure recorded	Measurement tool	Study
Blood Lactate Concentration	Blood gas analyser – Earlobe (ABL 625)	Binnie et al (35) Hue et al (62) Hausswirth et al (20)
	Blood gas analyser – Fingertip (Lactate Pro)	Taylor et al (12) Schabort et al (37) Taylor & Smith (54)
	Blood gas analyser – Undetermined location (Lactate Pro, YSP 1500 STAT)	Lopes et al (42) Taylor & Smith (53)
Core Temperature	Ingestible temperature measurement pill (CorTemp)	Binnie et al (35) Taylor & Smith (53)
Heart Rate	Heart rate monitor (BHL-6000, Garmin, Polar Electro, Polar RS200, Polar S810i, Polar Vantage XL)	Binnie et al (35) Hausswirth et al (52) Hausswirth et al (64)

		Hue et al (62)
		Lopes et al (42)
		Millet et al (65)
		Olcina et al (55)
		Taylor & Smith (54)
		Taylor et al (12)
		Wu et al (24)
Muscle Activation	Electromyography (EMG) (Delsys)	Walsh et al (66)
Rating of Perceived Exertion	Borg Scale (6-20)	Binnie et al (35)
		Millet et al (65)
		Taylor & Smith (53)
		Wu et al (24)
Minute Ventilation	Respiratory gas analyser (Cosmed K4, CPX, MGA-1100)	Hausswirth et al (20)
		Hausswirth et al (52)
		Hue et al (14)
		Hue et al (62)

		Millet et al (65) Taylor & Smith (54) Taylor et al (12) Vercruyssen et al (68)
Oxygen Consumption	Respiratory gas analyser (Cosmed K2, Cosmed K4, CPX, MGA-1100, Parvo TrueMax 2400)	Guezennec et al (72) Hausswirth et al (20) Hausswirth et al (52) Hausswirth et al (64) Hue et al (14) Hue et al (62) Millet et al (65) Schabort et al (37) Taylor & Smith (54) Taylor et al (12) Vercruyssen et al (68) Walsh et al (66)

2.5. Predicting triathlon time through physiological variables

The sprint triathlon event requires high intensity effort over a long period of time (~1 hour), and as such is classified as an aerobic endurance event (8). The gold standard measure of aerobic endurance is a laboratory $\dot{V}O_2max$ test, therefore there is an abundance of research of this kind. Typically athletes obtain higher $\dot{V}O_2peak$ values when running on a treadmill, compared to cycle ergometry (7-20% lower), with tethered swimming observed at ~70% of cycle ergometer values (73, 76). Previously recorded values of $\dot{V}O_2max$ in highly trained triathletes have ranged from 39-49 $ml \cdot kg^{-1} \cdot min^{-1}$ in swimming, 57-61 $ml \cdot kg^{-1} \cdot min^{-1}$ in cycle ergometry, and 61-85 $ml \cdot kg^{-1} \cdot min^{-1}$ in treadmill running (76). There is a discussion within the literature concerning whether absolute or relative $\dot{V}O_2max$ is a more appropriate way to normalize the data in triathletes. With body weight being supported during both swimming and cycling, these are generally reported in absolute terms as it deemed to be advantageous (8), however $\dot{V}O_2max$ in runners are typically reported in relative terms with extra body mass being considered as a hindrance to performance (77). In comparison to single-discipline athletes, triathletes have shown similar values of $\dot{V}O_2max$ (approximate differences ~5 $ml \cdot kg^{-1} \cdot min^{-1}$) in running and cycling across a range of competitive levels (78, 79), attributed to the multi-discipline nature of their training routines. As previously highlighted, maximal oxygen uptake is modality specific and therefore triathletes should complete tests across all three disciplines to best understand their abilities and to accurately inform training. $\dot{V}O_2max$ has correlated significantly with overall triathlon completion time ($r = -0.80$, $p < 0.001$) (14) and swim performance ($r = -0.48$) (51), however it is known that individuals who obtain similar $\dot{V}O_2peak$ values in physiological tests can vary greatly in performance. Early research highlighted that in male triathletes $\dot{V}O_2max$, HR, BL, PPO and ventilatory thresholds (VT) during a triathlon could not be correlated to overall performance (51, 80). Of all the global physiological measures recorded across both studies, notably no measures of global physiology were significantly related to cycling performance. However, it must be noted that the 'short course' used (1 km swim, 30 km cycle and 9 km run) is not currently a recognised distance and sits between sprint distance and Olympic distance triathlon formats.

Others have proposed running and swimming speed at lactate threshold (LT) combined to be the strongest predictors in performance, explaining ~98% of variance in overall triathlon time (16, 48). With LT occurring at a lower exercise intensity during cycling than that obtained during treadmill running, it suggests that LT is specific to the exercise and the active musculature involved (81). However, more studies have investigated the VT, rather than LT, in triathletes (8, 56, 78, 79). It has been observed that the VT occurs at a similar intensity of $\dot{V}O_2max$ between running and cycling 65 - 85% versus 61 - 84%, with vast differences caused by the various methods used to determine VT. This likeness is carried over to competition performance, with triathletes working at comparable levels of intensity in running and cycling during a triathlon (81). Research into the VT during triathlon swimming is limited, reporting VT occurs at ~70% of $\dot{V}O_2max$ (8), whilst others could not correlate any physiological variable during swimming to discipline or overall triathlon performance (37). It is important to note that differences in methods used to determine thresholds complicates the ability to make direct comparisons between studies.

BLC at the conclusion of the cycle stage ($r = 0.83$, $p < 0.05$), during steady state cycling at 4 $W \cdot kg^{-1}$ ($r = 0.92$), and when running at 15 $km \cdot h^{-1}$ ($r = 0.89$) have also been correlated to overall completion time (37, 80). It was calculated that BLC during submaximal cycling and peak treadmill running velocity could account for 81% of the variance in overall completion time. Running velocity has been subject to further research, with peak treadmill running velocity (r= 0.85) (37), running velocity during competition (r= -0.92, $p< 0.001$) (14), running velocity at VT (r= -0.78) (51), and the running velocity at the power output of optimal lactate removal (r= 0.87, explaining 76% variance in overall completion time) being other physiological predictors of overall performance (82). Other studies have also correlated PPO during cycling to overall completion time (r= 0.86) (37), (r= -0.71, $p< 0.05$) (14). There has been no success in research looking to associate specific anthropometric characteristics as predictive factors of triathlon performance (15).

With a wealth of literature on the topic of predicting performance from laboratory tests, it is therefore accepted that $\dot{V}O_2$max, LT, VT, BLC, running velocity and PPO are important variables within triathlon. Whilst the evidence here appears strong, a select group of studies were unable to determine links between laboratory variables and performance, although these were generally earlier studies with unrecognized distances and using technology that was less developed. Despite methodological limitations these results cannot be merely ignored. With suggestions that triathlon performance cannot be entirely explained by $\dot{V}O_2$max, HR, BLC or VT alone this highlights the importance in further research investigating factors outside of these commonly used global physiological variables. The vast nature of the findings in this section come as a result of the variation in testing protocols, variables measured, and statistical methods used to analyse the results. The above predictions are generally based on data gathered in the laboratory or from retrospective analysis from triathlon competition. New technology now allows for the analysis of physiological responses during both training and competition and would be an interesting avenue of future research (7).

2.6. Factors affecting physiological response

2.6.1. Sex difference

It should be noted that there is a sex difference in triathlon performance with males outperforming females by 10-20%, proposed to be influenced by physiological and societal factors such as $\dot{V}O_2$max and lower participation rates, although the gap has narrowed in recent years (30). The greatest differences in performance exist within overall power output during the swim (41%) and completion for the run (15%), with the gap widest in younger (18 - 24 years old) and older (55 - 59 years old & 60 - 64 years old) age groups (30).

2.6.2. Age difference

There are also age-related declines in sprint distance triathlon performance which have been linked to a reduction in $\dot{V}O_2$max and ability to sustain a high fraction of $\dot{V}O_2$max, as well as a general decrease in training volume (23). Age-related cellular alterations are proposed to influence the age related declines in cycling efficiency (11.2%) and subsequent increase in energy cost of running (10.8%) between well trained young versus masters triathletes (33). Further to this, a reduced muscle mass and associated strength increases the energy cost of running in aging triathletes, adding to the age-related decline (34). However, a single study comparing triathletes of 27.75 ± 3.37 years old versus 39.7 ± 4.81 years old failed to observe age-related changes in performance (5). With no significant differences in training habits between groups, and endurance performance thought to be maintained until approximately 40 years of age (29), it may be a failure in recruiting appropriately aged participants that caused this finding.

2.6.3. Ability

Training status and performance levels are also seen to alter the physiological response to triathlon, particularly when concerning the energy cost of exercise. The effects of cycling on the energy cost of running is greater in middle-level triathletes, comparing to their elite counterparts ($p < 0.01$) (65), with reasoning attributed to better leg stiffness regulation reducing the vertical displacement of running increasing exercise economy. This lack of experience and ability to effectively switch between movement patterns from cycling to running leads to more adverse mechanical alterations, and therefore increases the cost of running to a greater extent in these athletes (63). The cost of running in middle-level athletes has also been proposed to be exaggerated due to greater fatigue of the inspiratory muscles. However, it may be that elite athletes typically spend more time training and it is the mere accumulation of training hours that gives them a better adapted cardiorespiratory response to this transition, evident when comparing different age young professional triathletes of the same ability (15.2 ± 0.7 years old versus 23.8 ± 5.6 years old) (83). It must be recognised that when comparing between junior and senior elite triathletes (19.1 ± 1.5 years old versus 24.8 ± 2.6 years old), this better adapted cardiorespiratory response was only seen in elite female triathletes, and instead it was a higher ventilatory threshold that distinguished the senior males triathletes from their junior counterparts (84).

2.6.4. Competition strategies

When analysing an individual's physiological response to triathlon the researcher must account for competition strategies. Explained simply, despite being matched in completion time, a positive swim pacing elicits a greater RPE than a negative pacing strategy (9 ± 2 versus 11 ± 2), and this has carry over effects in the cycling discipline (24, 44). Further explanation can be seen where swimming at 90% of time trial pace causes significantly lower BL than at 100% (3.8 ± 0.9 mmol/L versus 7.3 ± 2.4 mmol/L, $p < 0.01$) (44). Cycling at a higher intensity (96 - 100% versus 81 - 85% of time trial effort) decreases running performance, although overall performance is maximised when the cycle is completed at the highest sustainable intensity (85). It is observed that $\dot{V}O_2$, HR and BL increase at higher cycling intensities, however despite this no differences are seen in the subsequent run. Further complications in interpreting data arise when comparing variable versus constant power cycling efforts (86, 87). Variable power efforts, which are further influenced by the geographical characteristics of the course, lead to a substantially heightened physiological response of HR, $\dot{V}E$, BL and RPE. These increases are moderately-highly correlated to impaired running performance when compared to constant cycling matched for time and mean power output ($r = 0.51 - 0.96$). Research has suggested that triathlon coaches encourage their riders to 'spin' their legs at the end of the cycle stage, performed by decreasing resistance and increasing cadence, despite there being no evidence to support this proposal as a performance aiding strategy (69). With previous findings showing cadence selection alters physiological response whilst cycling (68), it would be inferred that any conscious change in approach would alter the physiological response, and researchers must be aware of this when trying to understand the demands of the cycle stage. Similar research exists within the running stage of the triathlon, with significant effects of pacing on $\dot{V}O_2$, $\dot{V}E$ and HR during the initial kilometre when speed is manipulated by -10%, -5% and +5% compared to an isolated control run ($p < 0.05$) (20). The authors propose a reduction in running pace at the onset of the run is most effective in terms of overall performance. This study also furthered support for relationship between speed and $\dot{V}O_2$, and therefore authors must consider pacing strategies when deciding how to analyse their data.

2.6.5. Drafting

During swimming the metabolic responses are affected by drafting, with $\dot{V}O_2$ (11%), HR (6%), BLC (38%) and RPE (20%) all significantly reduced compared to a non-drafting position (88). In this study the benefits of drafting were complicated by drafting distance and position, with 0 - 50cm and drafting behind a lead swimmer being more beneficial than drafting 100 - 150cm behind a swimmer and in a lateral drafting formation. Contrary research suggests that drafting does not elicit significant differences in BLC (7.3 ± 2.4 mmol/L versus 7.9 ± 2.4 mmol/L) or $\dot{V}O_2$ (57 ± 5 versus 54 ± 6 $ml \cdot kg^{-1} \cdot min^{-1}$) when compared to a time trial effort, although the use of the a 25m pool to carry out the swim and the number of turns required may have skewed the data (44). This drafting opportunity separates triathlon swimming from traditional pool swimming, where passive drag can be reduced by up to 26% (89), subsequently lowering oxygen uptake (5 - 10%) whilst remaining at the same swimming speed, indicating an increase in exercise economy whilst drafting (41). The sum of these benefits has shown performance improvements of up to 6% in drafting triathlete swimmers (40). Carry over effects have also been researched showing that drafting during swimming results in lower BLC and RPE the subsequent cycle ($p < 0.05$) and therefore has physiological and psychological benefits to performance (90). Much of the cycling literature has focused on drafting, where if utilised correctly triathletes can cycle at a reduced $\dot{V}O_2$ (-14%), $\dot{V}E$ (-31%), HR (-8%) and BLC (4.0 versus 8.4 mmol/L) whilst maintaining the same velocity (52, 56), therefore inducing a metabolic reserve. This energy saving minimizes the negative impact of the cycle-run transition (23, 25, 38, 40, 56) and therefore allows triathletes to leave T2 at a decreased physiological state. This has been seen to enable the maintenance of higher $\dot{V}O_2$, $\dot{V}E$, HR and BLC during the run, improving running performance by up to 4% (4, 52) with carry over effects to total completion time (40, 52).

2.7. Conclusions

Despite the increasing popularity of the sprint distance triathlon, research has yet to react. By widening the search terms to include Olympic and simulated distances the literature is able to inform on the physiological response seen during competition, with relative intensity rising sequentially across the disciplines. It is evident that previous work has utilised a wide variety of measurement tools to provide this physiological data, providing insight into a proportionally large set of performance measures. With each individual study comprising of a unique combination of measurement tools and outcome variables, it becomes problematic when piecing together the findings into a larger picture of overall performance.

With the sprint distance triathlon being classified as an endurance event, there is a large area of research focused on maximal exercise testing and the resultant data such as $\dot{V}O_2$, PPO and ventilatory thresholds. Various metrics have been correlated to both discipline-specific and overall triathlon performance. Combined running and swimming speed at LT appear to be the strongest predictor of completion time. It must be noted that not all studies were able to correlate global physiological measures to performance. Although there is an abundance of research in this area, it is again the wide variations in testing protocols, equipment, and variables measured that renders it complex to gather strong patterns from the data. Researchers must also consider the sex, age and ability of their participants, whilst closely observing drafting and competition strategies in order to accurately draw conclusions from the data.

Due to practical difficulties of testing in water, there is a gap in the literature when it comes to swimming research for both field and laboratory studies. No research was found to report $\dot{V}O_2$ data in this discipline, whilst studies that did report intensities generally calculated these from maximal exercise tests of a different modality. The use of: open water, traditional pool swimming, flume pool swimming and swim ergometry techniques further complicates this area of research.

The sequential combination of high intensity swimming, cycling and running performed during a sprint triathlon places unique demands upon the triathlete, different to those experienced in isolated versions of the three disciplines. The most emergent topic of the literature is the influence of prior cycling on triathlon running, where although the impact on the transition on $\dot{V}O_2$, $\dot{V}E$ and HR are well documented, the physiological changes that occur during the T1 transition (swim to bike) are not fully understood. Despite these studies highlighting the importance of considering prior disciplines when analyzing the physiological responses, many of these studies did not include swimming prior to the cycling efforts. It is within this area a lone study made use of wearable NIRS technology, highlighting the inability of the *Vastus Lateralis (VL)* muscle to utilise oxygen following the bike to run transition.

Although there is a wealth of information on the physiological responses to triathlon, the emergence of new technologies such as wearable NIRS devices suggest that there is more to uncover within the sport of triathlon. Future research should aim to create a complete physiological profile of sprint distance triathlon, looking to include respiratory swim data, utilising new testing methods and accounting for, where possible, the many factors that affect the physiological responses displayed by triathletes.

3. Near-Infrared Spectroscopy (NIRS)

3.1. Justification for exploring the use NIRS in triathlon.

Although the current literature has exhausted a wide variety of measurement tools when observing the physiological response to triathlon, much of the equipment used has limited practical application in the real world. Some of the equipment used has evolved to become portable such as gas analysers and video analysis applications, in the pursuit of ecological measurement and providing real time feedback during training and competition. Whilst there is a wealth of knowledge in certain areas of triathlon, particularly concerning areas applying commonly used measurement tools, because of the developments of new technology it is important to investigate whether this gives us new and helpful insight. The NIRS devices identified in a single study (55) in the above review gives us a new measure that has already been used in multiple sports, including swimming. It is a tool that could feasibly be used within triathlon and therefore its use must be explored further.

The above review outlines the current research that has aimed to characterize the specific physiological demands placed upon an athlete when completing a triathlon.

The monitoring of load has recently gained research interest, possibly as a result of technological advancements allowing for better quantification of activities (91), with this understanding necessary to training and competition strategies to maximize improvements in performance (92). Although, the majority of reviewed studies aimed to take a holistic approach, assessing $\dot{V}O_2$, HR, BL and RPE, each of these variables measures a systemic response to exercise (93). Although widely utilised, useful and informative, these methods have intrinsic limitations where such global measures are unable to separate the responses of the exercising muscles to the rest of body, or differentiate the work carried out by each of the active muscle groups (94). The above literature has identified that each discipline requires different active musculature in order to complete the given task, being able to understand the relative workloads undertaken by these specific muscle groups during exercise could better inform training and competition strategies. Furthermore, issues arise with the process of data collection when using the global measures, where despite being a popular and reliable metabolite tool to indicate exercise intensity (95), BLC collection has to be obtained invasively. As a result, there is an inability to achieve complete monitoring of this measure and it is often recorded sporadically during exercise, frequently requiring enforced rest intervals, and may not accurately portray the full response. Likewise, measures of $\dot{V}O_2$ are also invasive due to the need to wear a fully secured mask, with some athletes potentially developing discomfort, anxiety or claustrophobia when wearing the mask for long periods of time (96). Further difficulties arise when attempting to measure $\dot{V}O_2$ in the field, and despite the creation of portable devices (Cosmed, Cosmed K5, Italy), no such device is available for testing open water swimming and other underwater activities.

Advancements in technology has led to the embedment of load monitoring technology in portable everyday devices, acquiring movement data non-invasively in real time (92). Wearable health technology is gaining ever increasing interest in society with wearable sensors helping individuals track activity in pursuit of a healthy active lifestyle (94). Physiology has gained from such advancements, for example, NIRS devices that combat the restrictions of more widely utilised measures, recording peripheral responses noninvasively and continuously in all environments (94, 97, 98). NIRS is a light-based technology that reports on the relationship between oxygen delivery and consumption or changes in tissue blood volume at the sight of gas exchange in the muscle (10, 11). As sprint distance triathlon is recognized as an aerobic event, oxygen delivery and utilisation should be a key indicator of performance. $\dot{V}O_2$max is generally accepted as the gold standard measure of aerobic endurance, although within triathlon there were inconclusive correlations between this measure and overall triathlon performance. It is also known that individuals who obtain similar $\dot{V}O_2$peak values in physiological tests can vary greatly in performance, due to ventilatory and lactate thresholds, as well as discipline-specific technique and subsequent movement economy. The above review indicated that combined running and swimming speed at LT combined appears to be the strongest predictor of overall completion time (accounting for 98% of the variance), although with the methodological limitations to recording BLC highlighted above, this is not a measure that can be continuously taken during competition. Therefore, use of a non-invasive, continuous, real time measurement of peripheral oxygenation is warranted. There is a need to explore device utility and measurement information within this exercise environment.

3.2. NIRS Technique and Measurement

The NIRS technique utlises the fact that, unlike visible light (~400 – 650 nm), the near infrared light range (~700 – 900 nm) is able to penetrate human tissue (skin, adipose tissue and muscle tissue) (94, 99). This provides access to the light absorbing chromophores: hemoglobin (Hb), myoglobin (Mb) and cytochrome oxidase (cytox) (97), where the absorption of light is oxygen status dependent.

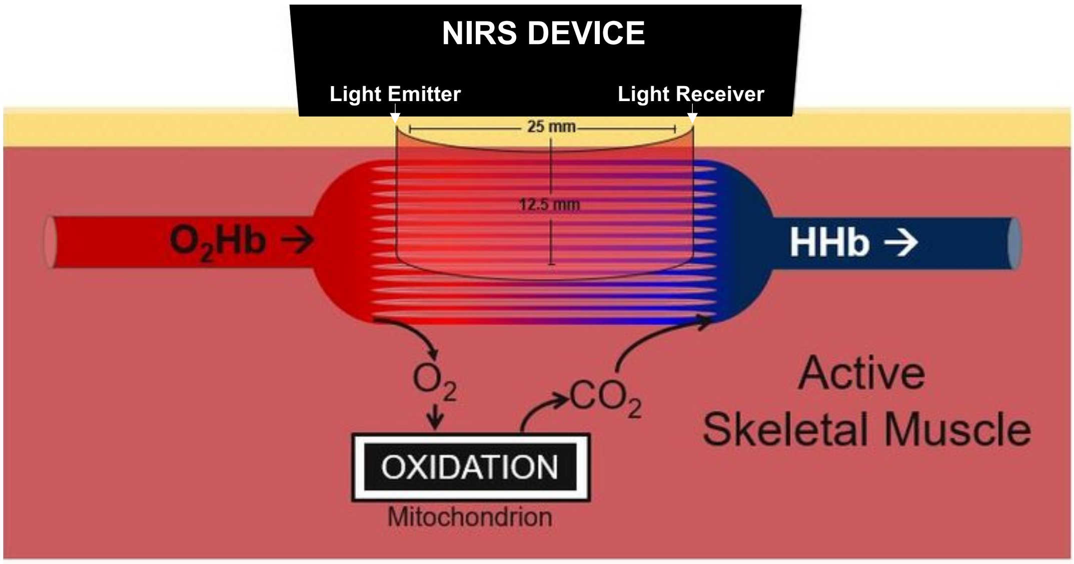


Figure 2. Diagram to illustrate the process of near-infrared spectroscopy (NIRS) (adapted from Luck 2019 (100)).

Currently three types of oximeters have been used in NIRS research, continuous wave (CW) NIRS, time domain (TD) NIRS and frequency domain (FD) NIRS, differing in the intensity and pattern of light emitted (101) (Figure 3). The most commonly used is CW NIRS, where a single, consistent beam of light is emitted, with changes in light intensity recorded by the receiver.

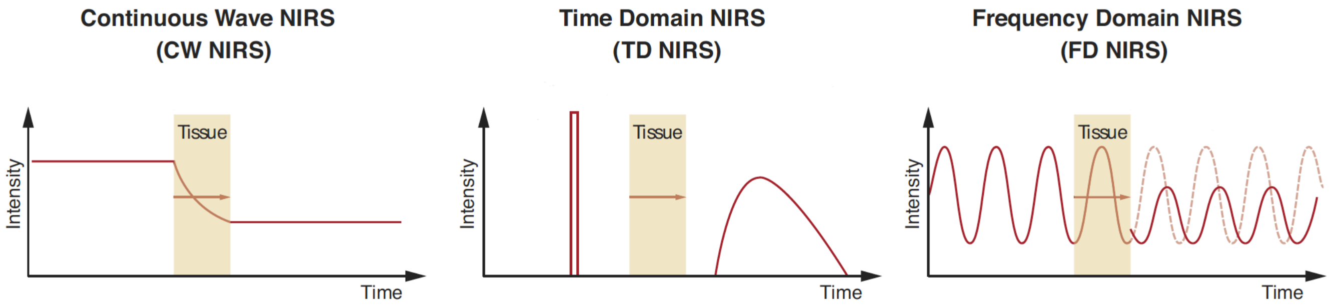


Figure 3. Diagram to illustrate light flow across the three types of muscle oximeter used in near-infrared spectroscopy (NIRS) research (adapted from Barstow 2019 (97)).

Advancements to such technology has included spatially-resolved spectrometers (SRS), where multiple source detectors are used, utilising different wavelengths to differentiate between chromophores. This is possible due to the specific extinction coefficient (ε), and how strongly a substance absorbs light at a given wavelength. Peak absorbency for HHb occurs at shorter wavelengths (~750 nm), whereas this occurs at longer wavelengths (~850 nm) in O_2Hb. The sum of HHb + O_2Hb provides total hemoglobin (tHb) and provides a measure of blood volume, with changes in this measure being attributed to blood flow (102). The O_2Hb measure is also thought to be primarily influenced by blood flow/volume, with cutaneous tissue extracting little oxygen (103). The ratio between O_2Hb and tHb (O_2Hb + HHb) provides the balance between delivery and removal of oxygen and hence estimates skeletal muscle oxygenation as a percentage (94, 104). This is the primary variable recorded using NIRS, this is calculated by:

$$\text{Tissue Saturation Index} = \frac{\text{Oxyhemoglobin}}{\text{Deoxyhemoglobin} + \text{Oxyhemoglobin}} \times 100\ (\%)$$

Equation 1. Tissue Saturation Index (TSI (%)) calculation.

With different NIRS manufacturers deriving their own set of calculations and formulas to approximate similar readings, the basis of such calculations must be understood. As a result, it is important to understand that the literature has referred to tissue oxygenation as: tissue saturation index (TSI), %SmO_2, tissue oxygen saturation (StO_2) and tissue oxygenation index (TOI) which are synonymous terms that inform upon the same measurement. Therefore, as a whole, NIRS is capable of estimating changes in oxyhemoglobin (O_2Hb), deoxyhemoglobin (HHb), total hemoglobin (tHb) and tissue saturation index (TSI) (105, 106) at the superficial level, informing upon the muscle the NIRS device is placed upon. The ability to observe the oxygenation (TSI%) of a muscle is of particular use in sports performance research, specifically in aerobic disciplines that rely on effective oxygen delivery and utilisation.

It is important to note that the NIRS data is not based solely from arterial blood (pulse oximetry), as larger quantities of blood in these vessels lead to near complete absorption of NIRS light. It is instead derived from a mixed venous sample resulted from weighted averages of O_2 saturations of arterioles, capillaries and venules and the Mb heme group of muscle fibres (94, 107). Whilst this may be perceived as a limitation, it has been identified from the literature that change in this arterial oxygen saturation value is largely only seen in hypoxia or disease (108, 109). The combination of sites used by NIRS allows the technology to be able to detect small changes <1% in muscle oxygenation during different exercise conditions (55).

Biological tissue (muscle) is highly scattering and therefore the NIRS light that enters the tissue does not follow a uniform path. The scattering process is complex and subsequently NIRS devices make use of the modified Beer-Lambert Law to account for light that: is lost due to absorption, takes a non-scattering path, scatters between emitter and receiver, and lost due to scattering (101, 110) (Figure 4). As the specific path length of light is unknown, CW devices provide relative values instead of absolute concentrations of chromophores, where changes are tracked from baseline (97). It is possible for researchers to directly compare TSI (%) between subjects (111).

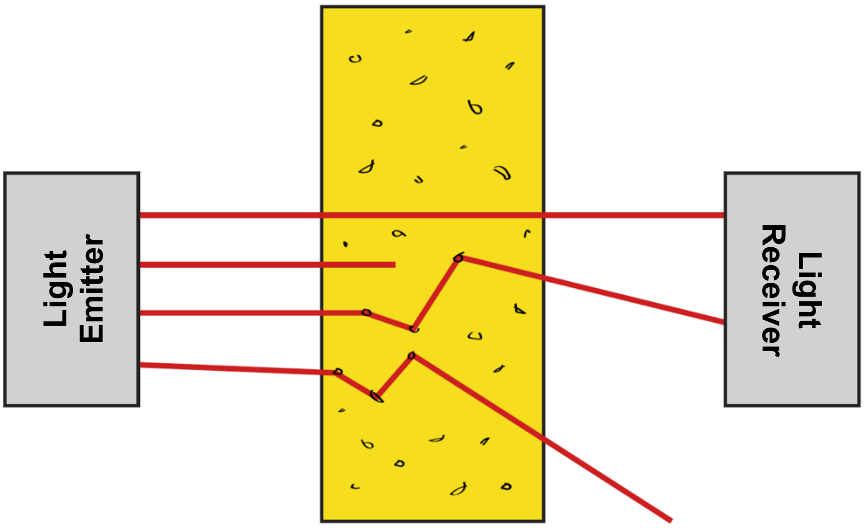


Figure 4. Diagram to illustrate the light scattering process accounted for by the modified Beer-Lambert Law (adapted from Barstow 2019 (97)).

As with any measurement tool it is important to understand the coefficient of variation, to ensure that any reported change seen within a measurement is as a result of the underlying measure and not merely device variation. With reliability measurements specific to the equipment, exercise and muscle groups used, a range of coefficients of variation are found in the literature. The measurement of NIRS can be obtained with good reproducibility at rest (112, 113), and across various intensities of exercise (114), with an accepted threshold of 10% (113). The coefficient of variation when using a PortaMon device has reported values of 4.7% in the *Vastus Lateralis* of clinical patients (115), and 1.8% - 2.5% in the *Vastus Lateralis* during supine rest in male participants (113). It is also common in NIRS literature to report change values, rather than absolute values, to eliminate any variation in measurement caused by differences in baseline absolute values. Previous research has stated no significant differences in NIRS output across multiple inter and intra-day analyses (116), indicating the NIRS measurement is reliable, highly reproducible, and therefore holds potential as a measurement tool to be used within training and competition.

3.3. NIRS use in sport

Portable NIRS technology has allowed for the monitoring of hemodynamics and muscle oxygenation into the applied environment. The ability to securely place the wearable devices onto the skin using simply non-restrictive tape or strapping allows for free movement and application in most sports (92, 117). NIRS devices can be placed on most muscle groups and allow for useful multi-site measurements, although currently the majority of NIRS research has observed the responses of the VL muscle (104). Although this technology has yet to be applied to full triathlon performance it has the potential to do so, and the following sections identify current NIRS research into isolated swimming, cycling and running.

NIRS has been used within triathlete populations in swimming, comparing muscular contributions to the exercise and the differentiation between triathletes and swimming specialists (118). Measurement of the extent of desaturation within the TSI (%) signal demonstrated that triathletes predominantly use their upper body musculature (*Latissimus Dorsi (LD)*) for propulsion when swimming, potentially saving the lower limb for later disciplines, whilst swimming specialists rely on a more even mix between upper and lower body (as evidenced by even desaturation in the upper and lower musculature). This research highlights how NIRS can identify individual muscular effort in a way global measures cannot. Swim research has also profiled hemodynamic responses to repeated sprint swim exercise, observing rapid drops in blood flow/volume (indicated by O_2Hb and tHb), and an immediate rise in oxygen extraction (indicated by HHb) upon the onset of maximal sprinting, with patterns reversing in recovery (119). These studies have demonstrated the relative ease of collecting peripheral muscle oxygenation data underwater in a way previously unattainable and hence support the use of NIRS in performance monitoring within swimming and therefore triathlon. When using NIRS for swim research it is important to consider the differences in device temperature when underwater, the additional pressure placed upon the device when wearing a wetsuit and the effect of tumble turns whilst swimming in a pool when analysing the NIRS measurement. Previous research has found a significant reduction in light intensity when NIRS devices are immersed in water, however changes are not significantly different across different water temperatures (30°C, 20°C, or 10°C), and this only results to a negligible change in TSI data (118). The influence of external pressure upon NIRS devices are presented by McManus et al (Figure 1a) (113), where the effects vary across manufacturers. It is reported that both MOXY and PortaMon are stable to pressure related changes when between 2-30 mmHg external pressure, although the effects in TSI signal of the MOXY device are magnified when pressure exceeds 20 mmHg. This study applied external pressure using an inflatable cuff and the exact pressure applied by a wetsuit or triathlon suit has yet to be reported in the literature. Researchers should be cautious of external pressure when using NIRS underneath a wetsuit, and select the PortaMon device which is less sensitive to pressure change, when conducting their studies. Finally, the large variation in swimming modalities used in the literature (see 2.4.1, Table 1), results in some methods involving continuous swimming (open water, flume pool, tethered swimming, swim ergometry) and other methods requiring turns (pool swimming). The effect of turning of NIRS data has been presented by Jones et al (Figure 4 and Figure 5) (119), and cause 'V' shapes in hemoglobin and muscle oxygenation traces as a result of momentary constriction during the turning action. Researchers must be aware of the changes to NIRS traces caused by turns when analysing NIRS data from pool swimming.

Recovery is a large topic within the NIRS literature. Close agreements have been found between NIRS derived O_2 recovery and magnetic resonance spectroscopy (MRS) derived phosphocreatine (PCr) recovery time (a process of adenosine triphosphate (ATP) resynthesis by oxidative phosphorlyation after musculature contraction) (120-122). The above study noted a signficant correlation between the recovery of muscle oxygenation (reoxygenation) rate and swim performance ($r = 0.70$, $p < 0.01$), suggesting that NIRS also has the potential to be used as a measurement tool to predict triathlon performance. Within triathlon the transition areas provide recovery periods (~1 - 2 minutes, dependent on ability level and layout of transition area (123, 124)) where this could be applied, alongside end recovery upon completion of the event. Although recovery at the end of an event cannot directly influence the performance that preceded it, it does provide an indicator of the underlying physiological processes such as replenishment of oxygen and the removal of exercise metabolites. An athletes ability to recover more quickly would suggest that they are more efficient in these physiological processes, which in turn would aid their performance during an event. Despite the transition areas being active and a key part of the race, each discipline requires different active musculature. Therefore the ability to observe recovery across multiple muscle sites during transition would provide insight into the recovery of active musculature in one discipline that are less prominent in the following stage of the race. The ability to observe this information may also allow researchers to gain a greater understanding of the physiological processes occuring in transition, an area currently not fully understood.

Research using NIRS in cycling has created a *Vastus Medialis* muscle oxygenation profile in response to maximal incremental exercise and a 20 km time trial (125), the same distance endured in a sprint triathlon. The incremental test showed an initial increase in oxygenation followed by a steady decline until $\dot{V}O_2max$ was reached. The time trial showed a rapid decrease in muscle oxygenation followed by a gradual decline, with $\dot{V}O_2$ data initially rising and then remaining stable throughout. This study shows that observations of specific muscle oxygenation are possible using NIRS, and that responses can be profiled to detail changes that occur in response to a particular exercise. The inclusion of a maximal incremental exercise test not only provided comparison to the time trial but was able to show the sensitivity of NIRS devices in response to changes in intensity which were enforced as participants progressed through stages of the test. Although intensity is not enforced upon athletes during a triathlon, it would be expected that intensity is consciously altered within and between each stage of the race, and therefore this capability could prove useful. This study also included a 3 week endurance training program, consisting of 60 minute sessions four times a week working at 85 - 90% of $\dot{V}O_2max$. Although the exact nature of the programme is not important, this study identified the ability of NIRS to monitor adaptations to training, where greater desaturation was seen when repeating these tests again. This would suggest that participants had a greater ability to utilise oxygen after training, and is a factor that could not be determined without the NIRS technique. With oxidative metabolism being the primary source of energy in skeletal muscle in endurance events, the ability to monitor this across time is of interest and from this it is inferred that NIRS has the potential to inform up training prescription and competition strategies within triathlon. Further cycle research utilising multi-site NIRS has shown that upon completion of exercise the working muscles (VL) re-saturate immediately whilst inactive muscles, such as those of the upper limb during cycling (*Biceps Brachii* (BB)), continue to desaturate for a short period of time after exercise. The recovery half-time is also significantly longer in these inactive muscles (126). If this pattern was observed during triathlon transition, it may be that the oxygenation response of inactive muscles is able to inform upon the inconclusive debate surrounding performance decrement following transition versus isolated exercise.

Differences between upper and lower limb contributions has also been examined within maximal running (93). Decreases in TSI were evident in VL and BB at the onset of maximal exercise, with a greater desaturation in upper body measurement (BB). This was paired with a decrease in blood flow (tHb) to the less active musculature of the upper body, indicating NIRS ability to show the physiological adjustments that are made to aid the active musculature (VL) in maintaining high intensity efforts. This biological adaptation is proposed to be a result of sympathetic flow and vasoconstriction of the less active muscle, directing the blood to the more active musculature (functional sympatholysis) (127). This NIRS technology is therefore an important measurement tool that also has the sensitivity and capability to provide insight into blood flow redistribution during exercise. This may hold value in understanding the effects of transition and how physiological response and overall performance in following stages are affected, an area currently limited in the literature. Further NIRS running research has been completed by examining muscle oxygenation during brick training (55), a training modality more applicable to triathlon. This single study highlighted how prior cycling effected subsequent running performance with large impacts seen in muscle oxygenation data. A higher muscle oxygenation in the brick trial suggested NIRS could inform on neuromuscular fatigue, proposing a failure to utilise intramuscular oxygen led to inability to increase intensity, causing this elevated value (i.e. reduced extraction). Perhaps more importantly, changes in kinematics, physiological or perceptual parameters were trivial suggesting NIRS could be a more effective measure of monitoring exercise intensity instead of HR, with HR not able to detect immediate changes in intensity or fatigue (128). The above review highlighted how the sprint triathlon is characterized by a requirement for short high intensity bursts due to variable race dynamics and the geographical nature of the course and therefore NIRS may be a useful tool of measurement here. Similar research has been conducted within running over varying hilly terrain, which suggested that NIRS is more effective at monitoring intensity than HR in this situation (129). Predominantly this study highlighted the sensitivity of the NIRS devices and the ability to respond instantaneously to changes in intensity, compared to the time delay seen in the physiological processes of HR and $\dot{V}O_2$ responses to the same change in intensity (130). This is also furthered possibly by the superior measurement frequency capabilities of NIRS technology, measuring multiple times per second. With HR currently being one of the most commonly used measurement tools in the field due to other measurement tools lacking portability, the idea that NIRS could provide a superior measurement warrants research attention. The more explicit change in intensity within a sprint triathlon occurs during transitions, and therefore NIRS may be able to provide greater insight into this area. Research has been conducted successfully to assess muscle oxygenation across multiple muscle groups within another multi-sport in the form of biathletes (131). This research was able to profile the hemodynamic response of both upper and lower limbs on a variety of terrains (flat, incline, downhill) endured within this discipline. It is highlighted here that although different individuals largely respond in the same fashion to a particular workload, specific individual differences may be present. This could either identify an outright difference to adequately utilise oxygen in the muscle, or perhaps outlines a variation in technique and muscle contributions to exercise. This biathlon research outlines how the NIRS information can be useful to coaches, where the NIRS devices were able to identify a lack of resaturation of the VL during a downhill rest period where this muscle should not be engaged. It was noticed that due to the particular technique of the participant, the VL was not relaxed and the expected resaturation could not occur, highlighting how NIRS can be used by coaches to give real time feedback to their athletes. The triathlon equivalent to this may come during the swim phase where triathletes typically aim to rest their legs for later stages (118), and therefore monitoring muscle oxygenation profiles of the legs during the swim stage may have the potential to improve overall triathlon performance. With the legs being rested or relatively unused during this stage a highly saturated and stable oxygenation profile should be displayed, however if athletes are contracting muscles in the legs this would be noticeable to the coach, who could reinforce coaching points to benefit the athlete in the cycle and run stages. The same could be suggested for monitoring the upper body muscle oxygenation profile during the cycling stage, where the upper body cannot have more than a minimal impact, if at all, on propulsion. Despite research into swimming, cycling, running, cycle-run brick training and other multi-sports, there has yet to be any studies using NIRS that have included swimming, cycling and running to profile changes in muscle oxygenation during a continuous triathlon effort.

From a performance perspective, the portability of new NIRS devices and the subsequent possibility to collect data in all environments gives NIRS great potential within training and competition load monitoring. Research into other disciplines has indicated how NIRS is able to combat some of the limitations provided by other commonly used measurement tools, and also can be used in aquatic environments with relative ease. The ability to observe swimming data and the sensitivity of technology possibly having implications within transition means that NIRS could inform on gaps in the current triathlon literature, whilst it may also be more effective at monitoring intensity throughout. From isolated sport and other multi-sport literature it is evident that it is feasible to use NIRS within triathlon, and other researchers have noticed the practicality of the measurement in this discipline with a single study previously using triathletes as participants in brick training. As sprint distance triathlon is recognized as an aerobic event, oxygen delivery and utilisation are key and NIRS is able to provide insight on this non-invasively at a peripheral level. The addition of this output provides a separate set of physiological responses to a given workload, and if combined simultaneously with the commonly used cardiorespiratory measures would enable a greater evaluation of the internal load placed upon the athlete. As of yet no research has used NIRS across all three disciplines.

4. Experimental Study

4.1. Abstract

Despite the knowledge that sprint triathlon is greater than the sum of its parts, the inclusion of all three disciplines in triathlon research is rare. Research into this sport is generally focussed around global physiological responses, although the literature has shown an interest in utilising Near-Infrared Spectroscopy (NIRS) within triathletes. A laboratory based study explored the utility of multi-site NIRS as a measurement tool within triathlon using recreational male triathletes (n=11). Participants were required to complete maximal incremental exercise tests on a treadmill, cycle ergometer and swim ergometer, before completing a simulated sprint distance triathlon. A comprehensive profile of global and peripheral responses across the triathlon simulation was created, recording oxygen consumption, heart rate, blood lactate concentration, RPE and multi-site NIRS (*Vastus Lateralis and Latissimus Dorsi).* As expected, the NIRS devices worked effectively and were able to inform upon muscle oxygenation status across the full triathlon simulation. NIRS yielded different oxygenation responses between upper and lower limbs throughout and identified a greater peripheral measurement variability between participants compared to global physiological measures. Furthermore, NIRS was also able to identify unique individual aspects, such as differing desaturation profiles within individuals, which were not evident from global physiological data alone. It is suggested this variability highlights differences in efficiency between athletes, although no direct correlations were found between NIRS data during swim, cycle and run and overall performance. As a measurement tool NIRS has the potential to increase the specificity of physiological information available when creating strategies to be applied in sprint triathlon training and competition. Similar observational research should be completed in the field to add further understanding to the application of NIRS to triathletes, increase ecological validity and compare the physiological responses seen in a competition environment.

Key words: Sprint triathlon, NIRS, Physiological Profile, Laboratory, Performance

4.2. Introduction

The sequential combination of high intensity swimming, cycling and running performed during a sprint triathlon places unique demands upon the triathlete, different to those experienced in isolated versions of the three disciplines. Despite the knowledge of the residual effects each discipline has on overall performance, much of the laboratory simulated literature has focused on swim-cycle (T1) or cycle-run (T2) transitions (1), with few studies simulating the event in its entirety (2). This area of research typically observes the global physiological responses (oxygen consumption, heart rate, blood lactate concentration among others) that occur during participation, with frequent calculations of intensity compared to maximal exercise. With each individual study comprising of a unique combination of measurement tools and outcome variables, it becomes complex when piecing together the findings into a larger picture of overall performance. Due to practical difficulties of testing in water, there is a gap in the literature when it comes to swimming research for both field and laboratory studies. The use of: open water, traditional pool swimming, flume pool swimming and swim ergometry techniques further complicates this area of research.

The literature has previously shown an interest in applying NIRS technology to triathletes, where a lone study reported the inability of the VL muscle to utilise oxygen following the bike to run transition (55). NIRS is a light-based technology that reports on the relationship between oxygen delivery and consumption or changes in tissue blood volume at the sight of gas exchange in the muscle (10, 11). As sprint distance triathlon is recognized as an aerobic event, oxygen delivery and utilisation should be a key indicator of performance. NIRS has already been successfully applied to each individual sport of a triathlon, including swimming (118, 119) which is a research area where it is complex to record the global physiological response. Swim research has compared muscular contributions to the exercise between triathletes and swimming specialists (118), whilst the use of multi-site NIRS has been further explored to investigate upper and lower limb contribution to maximal running (93). There is additional value to understanding muscular contributions to exercise in multi-sport events such as triathlon, where the primary muscles being used for propulsion change across each stage of the race. Cycle based research has created a muscle oxygenation profile of the *Vastus Medalis* muscle across a 20 km time trial (125), the same distance of the cycle stage in a sprint triathlon. NIRS has also been applied to other multi-sports in the form of biathletes, where the hemodynamic response was profiled across a variety of terrains endured within this discipline (131). This study highlighted specific individual differences in NIRS response, influenced by an outright difference to adequately utilise oxygen in the muscle, or underlying variation in technique and muscle contributions to exercise which could provide useful information to coaches.

As of yet no research has used NIRS technology to observe the physiological response across a full triathlon, although with the successful application of NIRS within swimming, cycling, running and other multi-sports there are no perceived issues in using NIRS in this sport. The majority of the triathlon physiology literature has observed the global physiological response to exercise applying commonly used measurement tools, and although widely utilised, useful and informative, the developments of new technology means it is important to investigate whether this gives us new and helpful insight.

4.3. Research Aims

The purpose of the present investigation was: to explore the utility of NIRS as a new measurement tool within triathlon; produce a full oxygenation profile, including swimming response, to triathlon; to create a profile of the physiological responses to triathlon; and to identify the physiological responses associated with triathlon performance. As this research is exploratory and is observing physiological response to a full sprint triathlon for the first time, no formal hypotheses are proposed.

4.4. Methods

4.4.1. Participants

Previous research has stated that in order to detect a 10% change in simulated triathlon performance parameters, an estimated sample size of 5 to 10 is required, whilst a sample size of 10 would allow for the detection of a 5% change in mean HR and $\dot{V}O_2$(12, 132). Eleven recreationally active male participants (age: 38 ± 5 years, stature 1.77 ± 0.05 m; mass: 78.9 ± 8.2 kg; skin fold thickness *Latissimus Dorsi* (LD) 14.1 ± 3.2 mm; skin fold thickness *Vastus Lateralis* (VL) 7.4 ± 1.4 mm) volunteered to participate in the study through responding to advertisements. Participants had completed sprint distance triathlon in a time of 82:14 ± 9:56 minutes in the past 2 years, split into 13:49 ± 1:49 minutes swim, 40:08 ± 4:45 minutes bike and 23:57 ± 3:33 minutes run. Participants had been training for the discipline for 6.2 ± 2.2 years and completed 7 ± 3 sprint distance triathlons in that time, amongst other triathlon distances (super-sprint, Olympic, half Ironman and Ironman). Participants average weekly training routines, regarding training hours and kilometres completed, are detailed in Table 5. All participants completed a physical activity readiness questionnaire (PAR-Q), were provided with written and verbal project information and signed an informed consent form before engagement. All participants reported to be injury free prior to participation. Approval by the School of Sport Rehabilitation and Exercise Sciences (SRES) Ethics Committee was obtained before this investigation.

Table 5. Mean ± SD weekly training duration and distances of participants (n=11).

	h/week	**km/week**
Average weekly training	10.8 ± 3.0	108.4 ± 46.5
Swimming	2.3 ± 0.8	5.0 ± 1.8
Cycling	4.9 ± 2.0	80.2 ± 38.0
Running	3.7 ± 1.1	26.2 ± 11.6

4.4.2. Design

Participants were required to attend four visits to the University of Essex Biomechanics laboratory, controlled to ~19°C. Three maximal incremental exercise tests were completed (swim, bike, run) before completing a simulated triathlon in the laboratory. Visits were separated by 7 days and testing commenced at the same time (± 1 hour) to remove possible effects of diurnal variation in PPO, $\dot{V}O_2$, HR and BL (133-136). A randomised crossover design was used for treadmill and bike maximal incremental exercise tests (Visit 1 and Visit 2) to counterbalance any order effects. The swim test was not included in randomisation due to the requirement of familiarisation of this equipment which took place across Visit 1 and Visit 2. The simulated Triathlon was an accumulation of the previous visits and hence had to take place in Visit 4.

4.4.3. Experimental Procedures

Participants were instructed to not partake in intense physical exercise or consume caffeine for 24 hours prior to testing session, and not to eat a heavy meal in the two hours prior to testing. Participants were asked to maintain a similar pattern of exercise and feeding before each visit. Stature and mass anthropometrics were recorded during visit 1 and all calculations were derived using these values.

4.4.3.1. Maximal Incremental Exercise Tests

Participants wore a heart rate strap (Polar RCX5, Polar Electro OY, Finland) located around their chest which was synced to a portable gas analyser (Cosmed, Cosmed K5, Italy). NIRS devices were placed onto the VL and LD (see 4.4.1). A five-minute seated baseline period allowed the recording of resting measures of heart rate and muscle oxygenation, upon conclusion of the baseline period a blood lactate sample was taken. The portable gas analyser was attached, and a five-minute warm up was completed. All warm up exercise was completed on the same equipment used for the maximal incremental exercise tests. After warming up the gas analyser was removed to allow participants to have a drink of water if desired. A five-minute passive rest period was conducted before starting the maximal incremental exercise test (see 4.4.3.2, 4.4.3.3, 4.4.3.4). Each test increased in intensity until volitional exhaustion. Respiratory gas data, muscle oxygenation and heart rate were recorded throughout the test, with blood lactate concentration and RPE recorded at the end of each stage. Upon completion of the maximal incremental exercise test, participants underwent a five-minute seated rest period with all equipment remaining on to measure recovery values. After five minutes a final blood lactate sample was taken. All equipment was then removed, and the participant was dismissed.

4.4.3.2. Maximal Incremental Exercise Test – Treadmill

All testing was carried out using a motorised treadmill (Saturn, HP Cosmos, Germany), with participants attached to a harness for safety purposes. The treadmill was set at 1% gradient at all times to mimic outdoor running (137). The five-minute warm up period was completed at 9 km·h^{-1}, where participants were familiarised with dismounting and remounting the treadmill whilst the belt was still moving. Due to the width of the treadmill, a straddle method was not possible, therefore participants were encouraged to move to the left side of the treadmill only, ensuring all parts of their shoes were off of the moving belt. The maximal incremental exercise test consisted of three-minute stages, followed by 30 seconds of passive rest where participants would move off of the belt. The test started at 9 km·h^{-1} and increased by 1.3 km·h^{-1} in each stage similar to that described by Jones et al (138). Participants remounted the treadmill at the new speed. RPE was recorded in the final 20 seconds of each stage with BL being taken immediately after each stage during the rest periods. The test finished when participants could no longer keep the speed of the treadmill, and hence dismounted. To determine achievement of $\dot{V}O_2$max the following criteria were used: relative plateauing of $\dot{V}O_2$ (<2.1 ml·kg^{-1}·min^{-1}decrease) despite an increase in workload (139-141); or, if a plateau was not observed: BL ≥ 8 mmol/L (139, 142, 143); HR within 10 bpm of age estimated HRmax (220 - age) (73, 141); and RPE >17 (141, 144).

4.4.3.3. Maximal Incremental Exercise Test – Bike

This testing session took place on a cycle ergometer (Lode, Excalibur Sport, Netherlands). Participants were free to set up the dimensions of the cycle ergometer, mimicking their own bikes as closely as possible. All participants used their choice of clipless pedals and shoes. Warm up intensity was set at 1 W·kg^{-1}, with participants instructed to maintain a cadence between 70 - 90 rpm. The maximal incremental test was continuous and consisted of two minute stages, starting at 1 W·kg^{-1} and increasing by 0.5 W·kg^{-1} in each stage (145). The test was completed between 70 - 90 rpm and was ceased when cadence dropped below 65 rpm. Participants were given one warning regarding a low cadence if it was perceived that it was a lapse in concentration, rather than exhaustion, that caused the decrease in cadence. RPE and BL were recorded in the final 20 seconds of each stage.

4.4.3.4. Maximal Incremental Exercise Test – Swim

All swim related exercise was performed using a swim ergometer (Vasa, SwimErg, USA). It is important to note there is no leg activity for this ergometer, and this is solely upper body exercise. Two familiarisation sessions for this equipment was completed in the two prior visits. The first was an unstructured 10 minutes, where participants were given guidance on technique and learnt how to maintain a set pace. The second session was used to identify the maximal pace that the participant could maintain for 2:30 minutes. This pace was used to calculate the starting swim pace for the maximal incremental test, with an aim of completing six stages. Warm up pace was set at the starting pace for the main test (e.g 2:30 min·100m^{-1}). The maximal incremental exercise test was discontinuous and consisted of 2:30 minutes stages that quickened by 10 sec·100m^{-1} each stage, followed by 30 second rest periods. RPE was recorded in the final 20 seconds of each stage with BL being taken immediately after each stage during the rest periods. The test was stopped when participants were 10 sec·100m^{-1} below the required pace for a constant period of 10 seconds.

4.4.3.5. Simulated Sprint Distance Triathlon

Participants completed a simulated sprint distance triathlon in the laboratory. The same treadmill and swim ergometer were used as maximal incremental exercise tests, with the introduction of an electromagnetically braked cycle ergometer (Racermate, Velotron Pro, USA) for the bike to allow for self-pacing. Participants completed a five-minute warm up on this cycle ergometer to ensure bike set up was the same as that of the maximal incremental exercise test, and to become familiar with the gear changing process. Participants used the same clipless pedals and shoes as previous. Following a five-minute rest period the test began. Participants completed 750 m swimming, 20 km cycling and 5 km running as if it were a triathlon competition. Transition areas were replicated using shuttle runs within the laboratory, totalling a distance of 100 m for both swim-bike and bike-run transitions. Transitions were controlled to 120 seconds for T1 and 150 seconds for T2. The additional time in comparison to competition (56) was required to record BLC at the start and end of each stage, whilst T2 had extra time to attach participants to the treadmill harness as quickly as was safely possible before starting up the treadmill. As the treadmill was motorised, and controls were out of participant reach, hand signals were used to indicate a desired change in speed. Participants were encouraged to change the speed frequently, by 0.1 $km \cdot h^{-1}$, to best replicate the variation in speed in outdoor running. No food or supplementation was allowed during the simulated triathlon in order to minimize factors which could affect the recorded data. $\dot{V}O_2$, HR and TSI% were recorded throughout, with RPE taken at regular intervals. BL was taken immediately before and after each individual discipline, with an additional measure five minutes post completion.

4.5. Measures

4.5.1. Near-Infrared Spectroscopy.

The PortaMon (Artinis, PortaMon, Netherlands) is a dual wavelength spatially resolved spectrophotometer, which provides information on haemoglobin concentration using the modified Beer-Lambert law and a quantitative measure of tissue oxygen saturation ((TSI) (%)). This device was placed on the belly of the VL muscle midway between the greater trochanter of the femur and the lateral femoral epicondyle; and the LD muscle at the midpoint between the midaxilla and the spinal column. The location was marked with ink and participants were instructed to keep this mark visible, ensuring the same location was used on each subsequent visit. The devices were secured to the skin with adhesive tape (Hypafix, BSN medical, Hamburg Deutschland). A further black neoprene strapping was wrapped around the device to block out outside light, with the cup shaping ensuring no extra pressure was placed on the device, two factors that could potentially interfere with the accuracy of the device (113, 146). Data was recorded at 10 Hz, with one second averages used for analysis.

4.5.2. Oxygen Consumption.

Respiratory gas was continuously recorded during each maximal incremental exercise test and simulated sprint distance triathlon using a portable gas analyser (Cosmed K5), this allowed the participants free movement around the laboratory. This is a valid measure of oxygen consumption (147). The gas analyser was calibrated to manufacturers recommendations before each individual test. Mask size was fitted to the individual in advance to the first maximal incremental test and was kept consistent across each following visit. Ten second averages of $\dot{V}O_2$ were taken at all time points used for analysis.

4.5.3. Physiological Responses.

HR, recorded continuously using a HR monitor, and Borg's scale of RPE (6 - 20) (148) were measured to determine the actual and perceived physiological responses to the maximal incremental exercise tests and simulated sprint triathlon. Five second averages of HR were taken at all time points used for analysis.

4.5.4. Blood Lactate Concentration.

All blood samples used left index fingertip capillary blood following a lancet (Unistik 3 Extra, Owen Mumford, Oxford, UK) puncture to initiate bleeding. Twenty µl of blood was collected into a capillary tube, immediately transferred to an Eppendorf tube (EKF diagnostic, Magdeburg, Germany) and shaken. Blood was analysed using the Biosen C line Clinic (EKF diagnostic, Magdeburg, Germany) on the day of testing, and stored in a medical refrigerator at -40ºC until analysis.

4.5.5. Preference and Perceived Performance.

Upon completion of all testing participants were given a short survey to provide additional exploratory details on their discipline preferences and perceptions of the testing equipment used in comparison to real world difficulty. The survey is displayed in Appendix G.

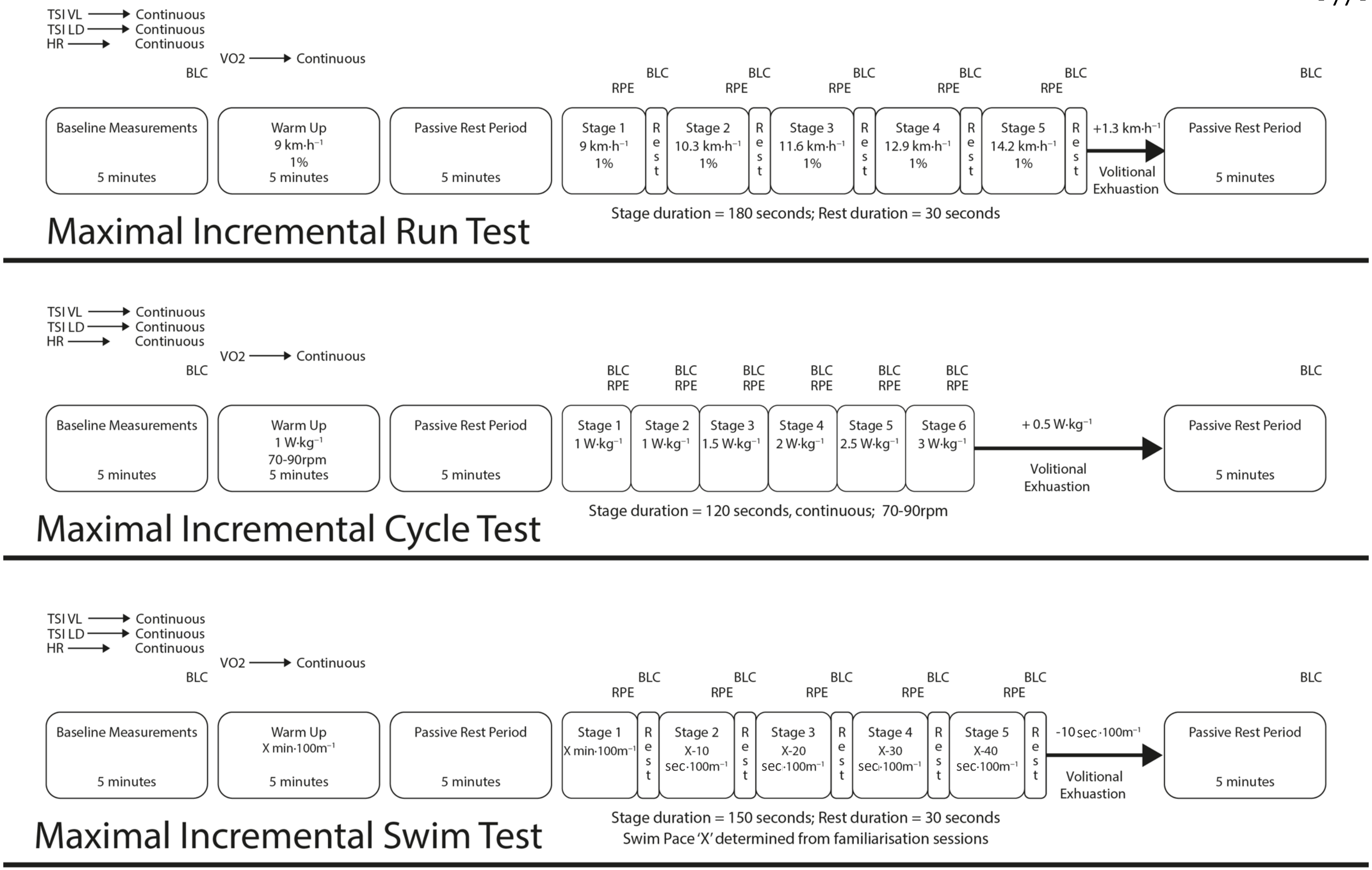


Figure 5. Schematic to illustrate the methodology used during maximal incremental exercise tests.

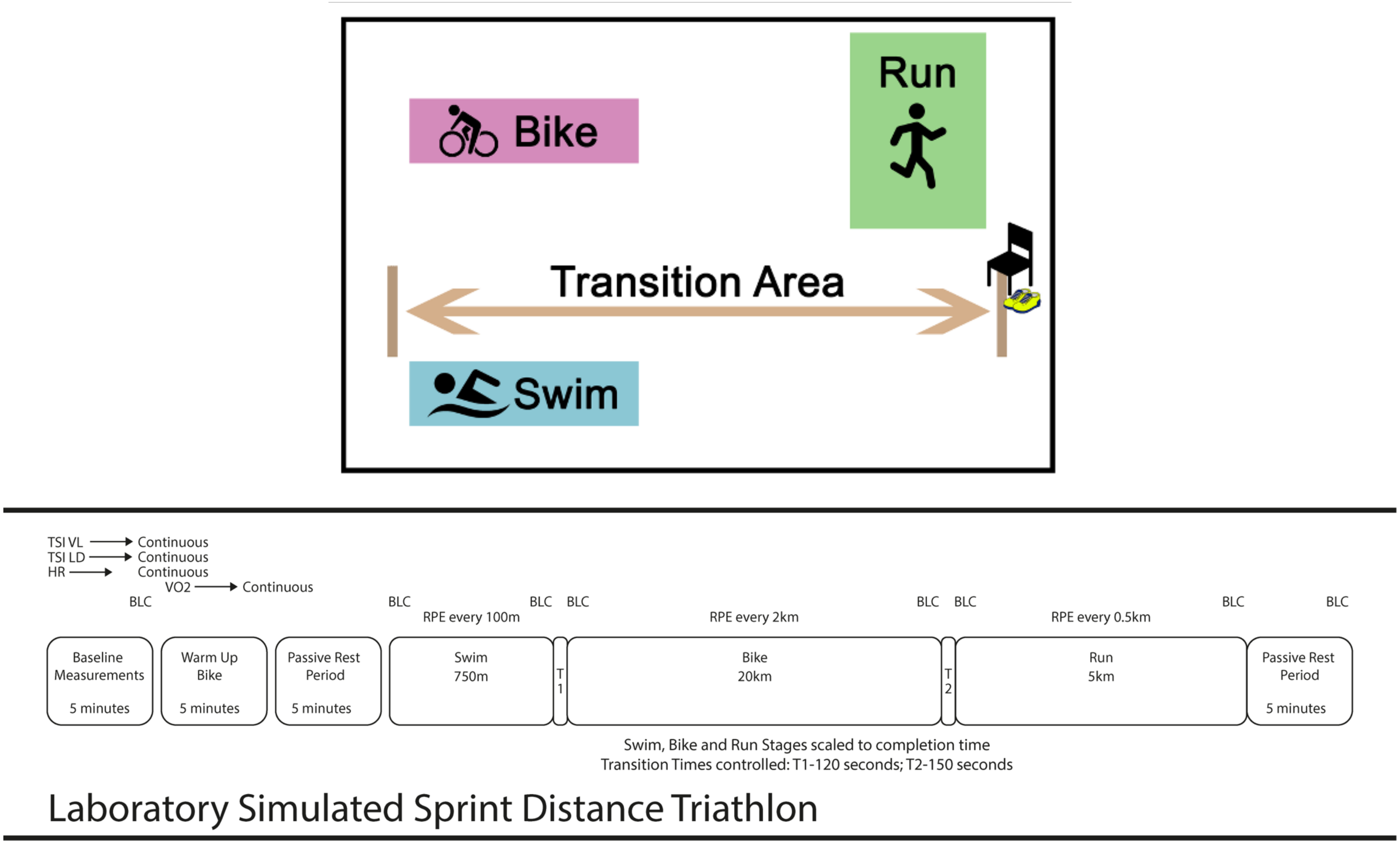


Figure 6. Schematic to illustrate the methodology and laboratory setup used during laboratory simulated sprint distance triathlon.

4.6 NIRS data analysis

TSI was selected as the sole NIRS measurement in this analysis. TSI is considered the most robust of the NIRS signals, least affected by surface interference and skin blood flow (149). With TSI informing upon muscle oxygen utilisation, and oxidative metabolism being the primary source of energy during sprint triathlon events, it was perceived as the major signal of interest for this primary observation. As TSI has previously been reported to be influenced by skin fold thickness (150), a primary correlation calculation was performed to understand the effects of skin fold thickness on the extent of desaturation between participants. No correlations were found in the VL ($r= -0.05$, $p= 0.871$) or LD ($r= -0.09$, $p= 0.808$) in the present study. Due to the NIRS measures recording in a higher time-resolution than that possible in $\dot{V}O_2$ and HR data, the measurements were filtered and averaged to one second averages. The recovery kinetics of TSI (%) are modelled as an exponential function, with end recovery from triathlon, and individual maximal incremental exercise tests, across both VL and LD sites, meeting the minimum fit criterion of data acceptance ($r^2> 0.80$). An exponential function was used for the curve fitting process. Recovery kinetics during transition did not meet this criteria $r^2< 0.60$, and therefore T1 and T2 response is reported as both ΔTSI from entering to leaving the transition area, and ΔTSI from baseline to maximal value achieved during transition. NIRS data is displayed as absolute values from a representative subject, or as group change (Δ) from baseline. NIRS data from all participants during T1, T2 and overall triathlon are provided in the Appendix B, C and D.

4.7 Statistical analysis

Descriptive statistics are presented as mean ± SD unless otherwise stated. Relative intensity was calculated for each discipline as:

$$\text{Relative Intensity} = \frac{\text{Averaged values across stage during simulated triathlon after plateau}}{\text{Maximal values achieved during maximal incremental exercise test}} \times 100\ (\%)$$

Equation 2. Relative Intensity (%) calculation.

A repeated measures ANOVA was used to analyse the differences between relative intensity $\dot{V}O_2$, HR, BLC, RPE and TSI responses across swim, cycle and run stages. A Bonferroni adjustment of alpha was applied to correct for the multiple comparisons in this analysis. The data was then split based on overall triathlon completion time into best three (fastest) versus worst three (slowest) performers to assess differences in the physiological response between these groups. A two-way mixed ANOVA was used to assess the interaction between $\dot{V}O_2$, HR and BLC and performance level, with comparisons of each stage of the triathlon between these groups. A partial eta squared (η_p^2) value >0.01 was considered a small effect, $\eta_p^2 > 0.06$ was considered a moderate effect and $\eta_p^2 > 0.14$ was considered a large effect size (151). It is recognised that this particular analysis is statistically underpowered. Pearson's product-moment correlation analysis was used to compare associations between $\dot{V}O_2$, HR, BLC, RPE and TSI responses across maximal incremental exercise tests and simulated triathlon, with discipline-specific and overall triathlon completion time. All analyses were performed using GraphPad Prism 8 (GraphPad Software, San Diego, CA, USA).

4.8. Results

4.8.1. Maximal incremental exercise tests

The physiologic criteria in attaining $\dot{V}O_2$max is shown in Table 6. Of those who did not achieve a plateau in $\dot{V}O_2$, two participants failed to achieve HR criteria during the swim ergometer, two separate participants failed to achieve either HR criteria or BLC criteria during the cycle ergometer and one participant failed to achieve BLC criteria during run. Of the 44 maximal incremental exercise tests conducted, 3+ achievement criteria were reached 39 times, with remaining tests having met achievement criteria in 2+ categories. Subsequently all data was used for analysis. The HR criterion was met by one participant during the swim ergometer test. With a posture change from seated and erect (cycling and running) to prone (swimming) the hydrostatic gradient is removed, resultant HR is significantly lower, and reaching predicted HRmax (220 - age) in this discipline is unlikely (152). Maximal group mean data for $\dot{V}O_2$, HR and TSI is displayed in Table 8. $\dot{V}O_2$, HR and TSI responses of all three tests from a representative participant is displayed in Appendix A.

Table 6. Summary of achievement criteria for three modes of maximal incremental exercise test (n=11).

Criteria	**Swim ergometer**	**Cycle ergometer**	**Treadmill**
$\dot{V}O_2$peak plateau (<2.1 $ml \cdot kg^{-1} \cdot min^{-1}$)	8 (73%)	9 (82%)	10 (91%)
HR (within 10 bpm of predicted max)	1 (9%)	11 (100%)	11 (100%)
BLC (≥ 8 mmol/L)	8 (73%)	10 (91%)	10 (91%)
RPE (>17)	11 (100%)	11 (100%)	11 (100%)

$\dot{V}O_2$ = Oxygen consumption ($ml \cdot kg^{-1} \cdot min^{-1}$), HR = Heart Rate (bpm), BLC = Blood Lactate Concentration (mmol/L), RPE = Rating of Perceived Exertion (6-20).

4.8.2. Simulated triathlon

The mean overall completion time, and discipline-specific split times, are presented in Table 7. The average group mean data for $\dot{V}O_2$, HR and TSI is displayed in Table 8. Mean pacing profiles for each discipline are displayed in Appendix E.

Table 7. Discipline and total completion time during simulated triathlon (minutes:seconds) (mean ± SD)

	Time
Triathlon	83:24 ± 5:12
Swim	14:56 ± 1:07
Transition 1	2:00 (Controlled)
Bike	36:39 ± 2:43
Transition 2	2:30 (Controlled)
Run	27:19 ± 3:11

Table 8. Maximal physiological responses to incremental exercise tests and mean responses during simulated triathlon (mean ± SD).

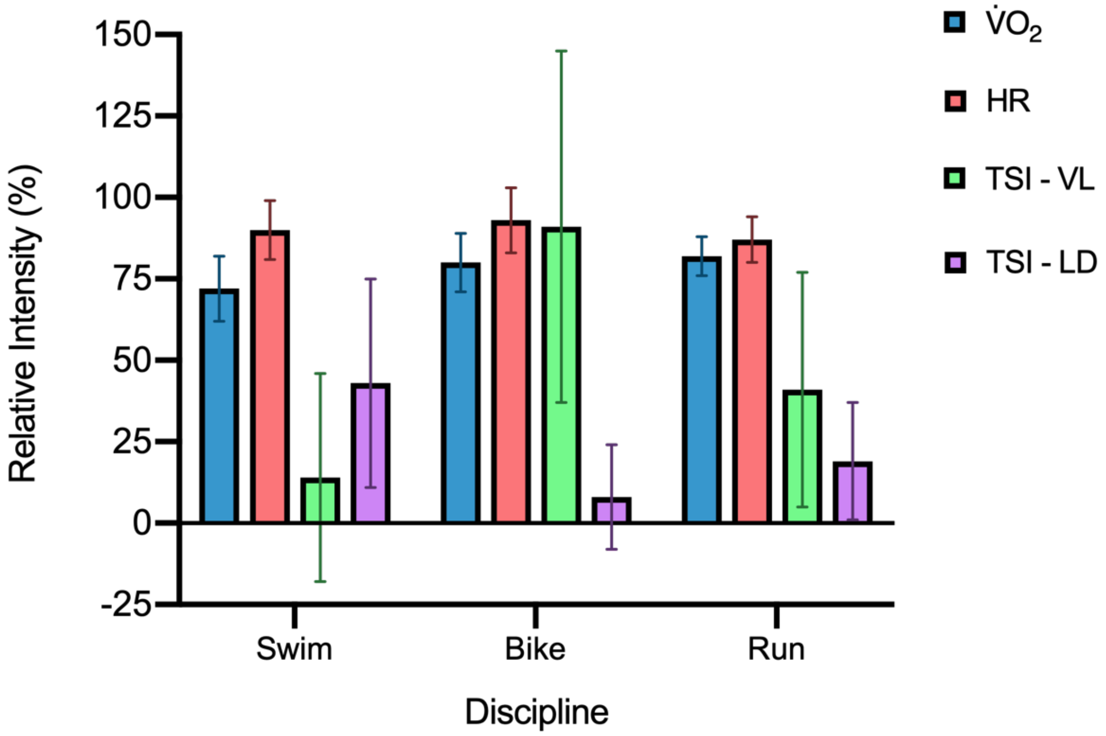
		$\dot{V}O_2$	**HR**	**TSI of the VL**	**TSI of the LD**
Swim	Max Test	32.72 ± 4.34	142.11 ± 17.83	-6.27 ± 3.85	-9.29 ± 3.65
	Triathlon	23.95 ± 5.51	127.85 ± 15.54	-0.72 ± 1.86	-4.17 ± 2.76
Bike	Max Test	54.43 ± 6.12	168.43 ± 14.44	-14.01 ± 2.69	-15.55 ± 4.65
	Triathlon	43.44 ± 7.30	155.48 ± 13.06	-12.74 ± 8.17	-1.13 ± 2.51
Run	Max Test	61.81 ± 4.84	179.84 ± 13.66	-16.13 ± 6.41	-15.00 ± 5.86
	Triathlon	50.56 ± 5.57	163.55 ± 14.95	-6.17 ± 5.43	-2.88 ± 3.18

Note. *TSI data is calculated as change from baseline.*

$\dot{V}O_2$ = Oxygen Consumption ($ml \cdot kg^{-1} \cdot min^{-1}$), HR = Heart Rate (bpm), TSI = Tissue Saturation Index (%).

4.8.3. Relative intensity

Relative intensity for $\dot{V}O_2$, HR and TSI is provided in Figure 7. Relative intensity of $\dot{V}O_2$ was comparable during swim, bike and run stages (72 ± 10%; 80 ± 9%; and 82 ± 6% respectively). Relative intensity of HR remained consistent throughout swim, bike and run stages (90 ± 9%; 93 ± 10%; 82 ± 6% respectively), with $\dot{V}O_2$ and HR portraying smaller standard deviations compared to TSI values. TSI of the VL was significantly different between swim and bike disciplines (14 ± 32% vs 91 ± 54%), but comparable between swim and run (14 ± 32% vs 41 ± 36%), and bike and run (91 ± 54% vs 41 ± 36%), disciplines with large standard deviations in this measure. TSI of the LD followed the same pattern, significantly different between swim and bike disciplines (43 ± 32% vs 8 ± 16%), but comparable between swim and run (43 ± 32% vs 19 ± 18%), and bike and run (8 ± 16% vs 19 ± 18%), with swim having the greatest intensity followed by run and bike stages. When comparing between variables, TSI of the VL was significantly lower than $\dot{V}O_2$ and HR across swim and run stages, whilst TSI of the LD differed to $\dot{V}O_2$ and HR in the bike and run stages. When comparing upper and lower limb intensities, TSI of the VL was higher than TSI of the LD during the bike stage but comparable in both swim and run disciplines. Statistically significant comparisons for $\dot{V}O_2$, HR and TSI within and between disciplines and measures are displayed in Table 9.



Note – See Table 9 for significance values within and between disciplines.

Figure 7. Relative intensity of $\dot{V}O_2$, HR and TSI shown during simulated triathlon in comparison to discipline maximal incremental tests.

Table 9. Repeated measures ANOVA statistical significance between relative intensity of $\dot{V}O_2$, HR and TSI during simulated triathlon.

		$\dot{V}O_2$			HR			TSI VL			TSI LD		
		Swim	Bike	Run	Swim	Bike	Run	Swim	Bike	Run	Swim	Bike	Run
$\dot{V}O_2$	Swim		1	1	**0.005**			**0.006**			0.423		
	Bike			1		0.260			1			**0.001**	
	Run						0.158			**0.048**			**0.001**
HR	Swim					1	1	**0.001**			0.086		
	Bike						1		1			**0.001**	
	Run									**0**			**0.001**
TSI VL	Swim								**0.039**	1	1		
	Bike									0.405		**0.040**	
	Run												1
TSI LD	Swim											**0.001**	1
	Bike												0.392
	Run												

Note. *Values presented with Bonferroni adjustment.*

*Values considered significant ($p< 0.05$) are denoted in **bold.***

4.8.4. Tissue Saturation Index response

During the initial seconds of the swim stage, a significant decrease in TSI of the LD is evident, paired with an increase in TSI of the VL. Although TSI is not designed to directly inform upon blood flow redistribution, this would suggest some initial constriction of the blood vessels in the LD, and dilation of the blood vessels in the VL. There is an immediate rise in TSI of the LD after this initial response back to baseline levels by 250 m, where this remains fairly stable throughout the entirety of the swim stage. TSI of the VL reverses this pattern, decreasing at a slower rate to ~500 m. After this point TSI of the VL is more variable but shows an increase until the cessation of the swim stage. Responses between participants were similar during the swim stage after the immediate stages. Notably, participant *b* showed continual desaturation of the LD muscle throughout, with participants *h* and *k* showing more pronounced desaturation in the VL muscle across the swim stage. Both TSI traces show an increase during T1, with TSI of the LD increasing significantly greater in the same time frame. From this point TSI of the LD decreases to a plateau ~6 km into the cycling stage, where it remains stable for the entirety of the duration of the bike stage. The reduction in TSI of the VL occurs at a significantly faster rate where a plateau is reached ~2 km. Differences in TSI of the VL and LD are most evident during the cycle stage in all participants. The extreme desaturation seen in TSI of the VL in participant *k* must be highlighted. Participants *e*, *f* and *j* show an increase in TSI of the VL in the final moments of the cycle stage, indicating a decrease in intensity. During T2 both traces display an ‘M’ shape, characterized by rapid increases in TSI upon the completion of the bike stage, interrupted by a drop in TSI, before a second spike is seen as participants move into the run stage of triathlon. During the run both TSI of the VL and TSI of the LD decrease steadily until ~3.5 km, where a plateau is reached, before a significant decrease in TSI is seen in the final stages. The run stage shows the most individual differences in TSI response across both sites, with participants showing increases, decreases or stability in TSI of the VL / LD. The extent of desaturation was not influenced by skin fold thickness in the corresponding sites of the VL (r = -0.05, p= 0.871) or LD (r = -0.09, p= 0.808). TSI response across the full simulated triathlon from a representative subject is displayed in Figure 8. TSI responses across the full simulated triathlon for all participants is seen in Appendix B.

An enhanced view of the responses of a representative subject during transition is seen in Figure 10, full T1 and T2 responses of all participants are seen in Appendix C and Appendix D.

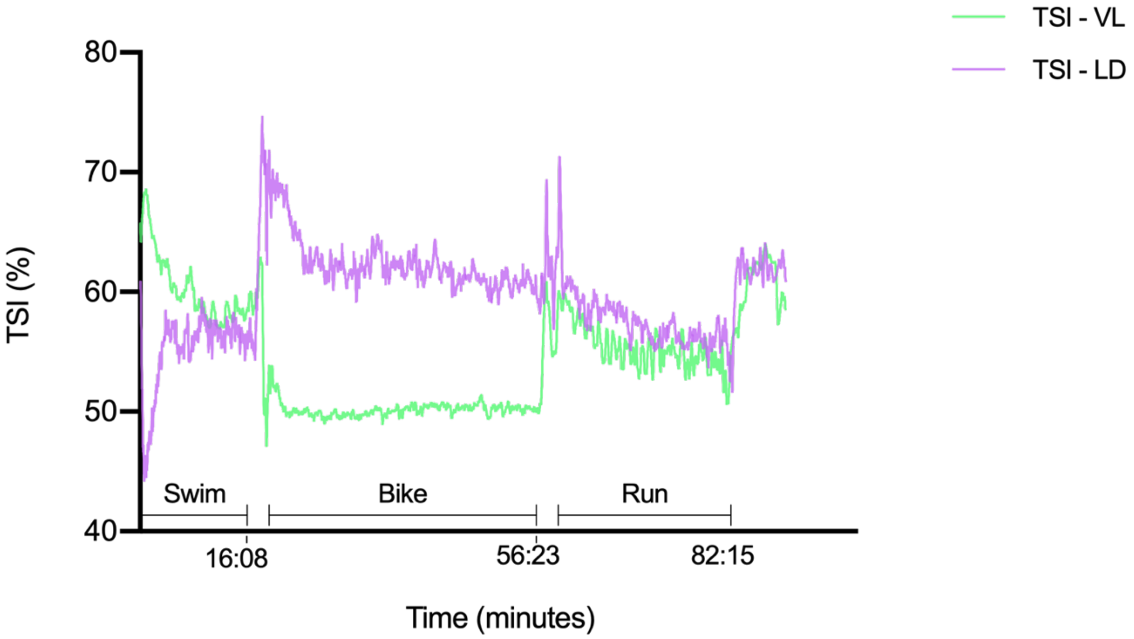


Figure 8. TSI response during simulated triathlon from a representative participant.

4.8.5. Oxygen Consumption and Heart Rate response

Upon the onset of the simulated triathlon HR increased rapidly as participants commence swimming, with a continued rise until 500 m before the rate of increase reduced in the final stages. $\dot{V}O_2$ had small fluctuations throughout the swim but did not largely deviate from the pre-exercise baseline value. From T1 onwards $\dot{V}O_2$ and HR followed a similar pattern. Upon transition both variables rapidly returned to that of the first seconds of exercise, before rising equally as quickly upon the onset of the bike stage. A greater rise in $\dot{V}O_2$ is seen when transitioning from swim to bike when compared to changes in HR. After ~6 km of cycling there is a slight decline in $\dot{V}O_2$ and

HR, before a plateau is reached and remains until the end of the bike stage. During T2 both $\dot{V}O_2$ and HR portrayed a ‘W’ shaped trace, characterised by a significant drop, interrupted by a brief spike, before returning to the levels of that seen at the end of the bike stage. Like the T1 transition although less pronounced, a greater rise in $\dot{V}O_2$ is seen when transitioning from bike to run when compared to changes in HR. A slight consistent rise in $\dot{V}O_2$ and HR is seen across the run stage, increasing further from ~3 km, with an accelerated rise in the final stages most evident in the $\dot{V}O_2$ trace. $\dot{V}O_2$ and HR response across the full simulated triathlon from a representative subject is displayed in Figure 9.

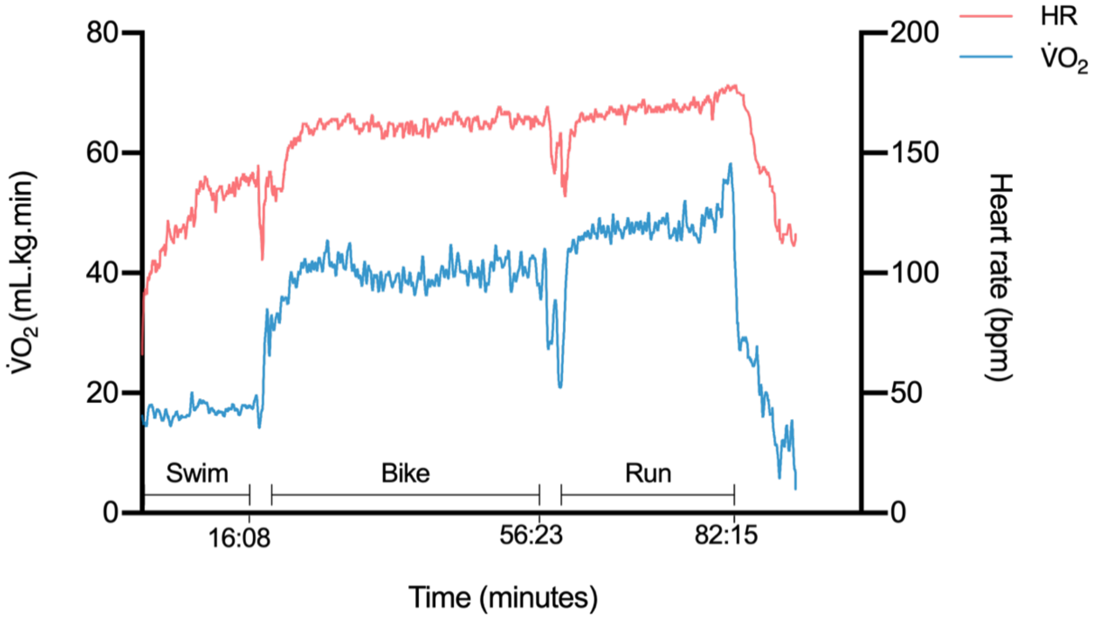


Figure 9. $\dot{V}O_2$ and HR response during simulated triathlon from a representative participant.

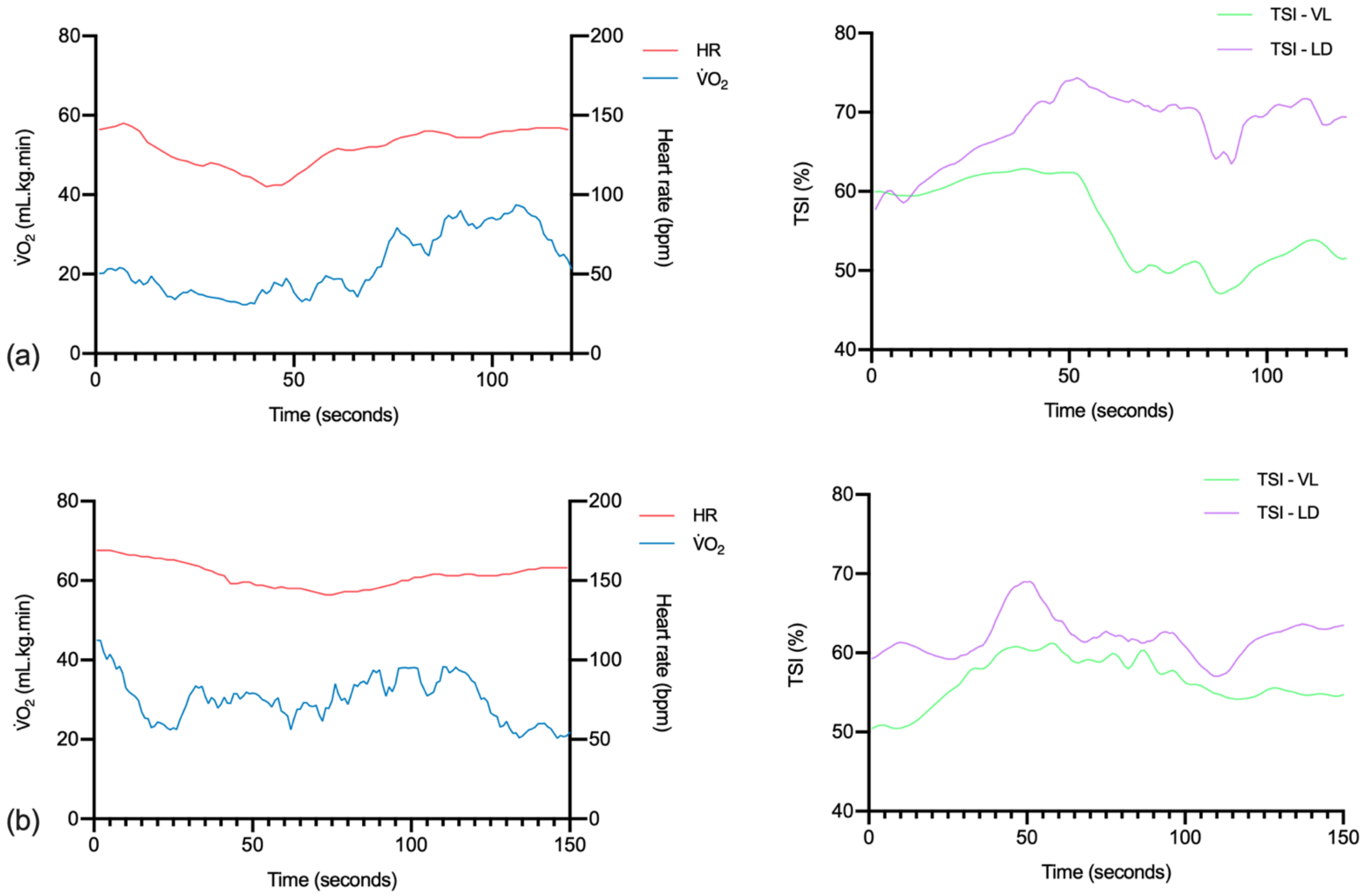


Figure 10. Representative participant V̇O_2, HR and TSI responses during T1 (a) and T2 (b) during simulated triathlon.

4.8.6. Blood Lactate Concentration and Rating of Perceived Exertion response

The responses of BLC and RPE can be characterized as increasing throughout with a resetting process occurring during transition. RPE rose from 12 to 15 during the swim, before reducing to 13 during the early stages of the bike stage. The bike stage rose 13 to 16, before reducing to 14 during the initial stages of the run. During the run, RPE continued to increase during the final stages finishing with a rating of 18 in the final seconds of the triathlon. The greatest variety of responses occur at the end of the bike stage, and the first measurement taken during the run. BLC was measured pre and post each discipline. BLC increased significantly from the onset of exercise to the end of the swim stage, before decreasing ~1.5 mmol/L during T1. During the bike stage BLC increased back to the level seen at the end of the swim stage, with the greatest variations between participants seen at this stage in the triathlon. BLC reduced ~2 mmol/L during T2, before reaching a peak value upon the completion of the run stage ~8.5 mmol/L BLC and RPE group response across full simulated triathlon are displayed in Figure 11 and Figure 12. Participants perception of difficulty of laboratory equipment in comparison to competing in the real world are displayed in Appendix F. Participants responses suggest the swim ergometer required more physiological effort to attain a given velocity in comparison to swimming in aquatic environments. Cycling and running responses are mixed, varying between more or less effort required, and comparable effort required.

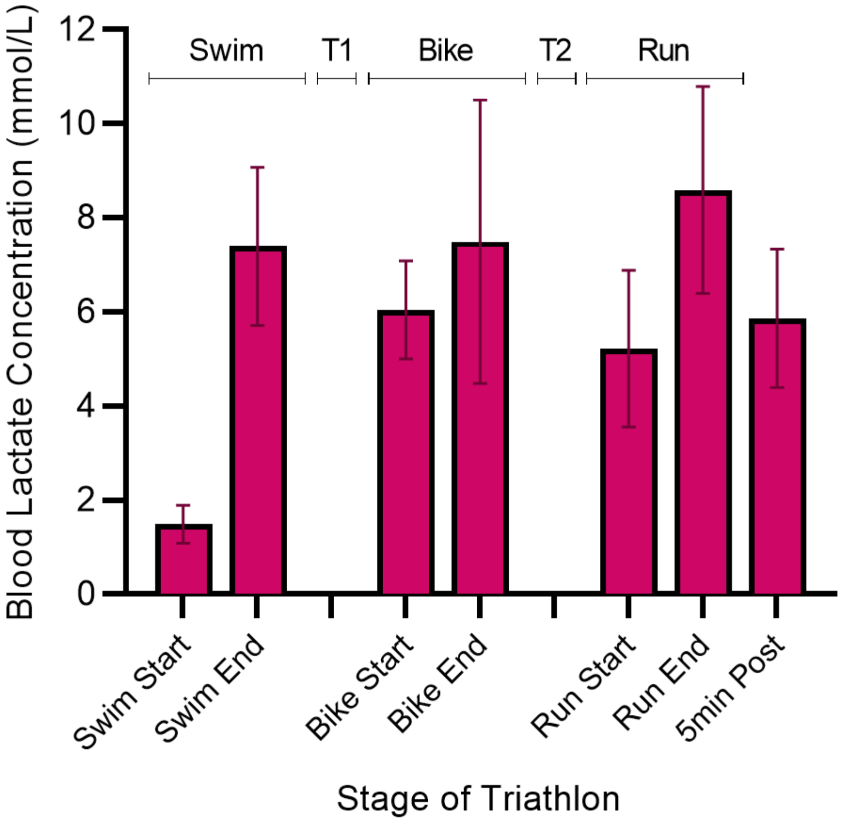


Figure 11. BLC response during simulated triathlon.

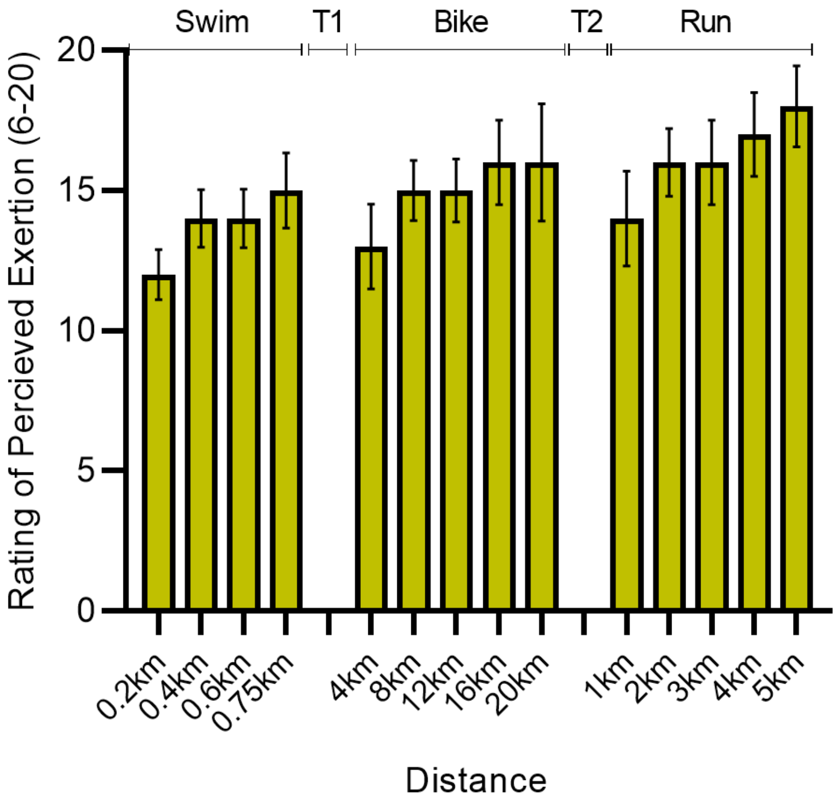


Figure 12. RPE response during simulated triathlon.

4.8.7. Associations to performance

When comparing discipline time to overall completion time, the bike stage accounted for the greatest proportion of the completion time (43%), followed by the run stage (33%), and the swim stage (18%) (Note – the sum of percentages do not total 100% due to the time spent in transition). Both bike time and run time produced significant relationships with overall completion time respectively (r= 0.902, *p*= 0.000; r= 0.788, *p*= 0.004), whilst swim time was not statistically significant (r= 0.209, *p*= 0.538). It must be noted that this analysis, correlating total time and components of this time, violates an assumption of parametric testing.

4.8.7.1. TSI response

No significant correlations were found between ∆TSI of the VL / LD from baseline and swim, cycle, run or overall completion time, $p > 0.05$.

4.8.7.1.1. Simulated triathlon

End recovery data from simulated triathlon provided: TSI of the VL half-life 42.59 ± 15.73 seconds, Tau 61.44 ± 22.69; TSI of the LD half-life 29.31 ± 11.72 seconds, Tau 38.82 ± 13.06. Simulated triathlon end recovery data from a representative participant is displayed in Figure 13. No significant correlations were found between triathlon end recovery data and discipline or overall completion time. After splitting the data in best versus worst performers it can be seen that best performers have longer half-time recovery in TSI of the VL and shorter half-time recovery in TSI of the LD, albeit not statistically significant, (VL $p = 0.64$, LD, $p = 0.25$; Figure 14). No curve-fit could be applied to either site during transition. Transition data was then analysed using ∆TSI across transition, and between baseline and peak transition values. Pearson correlational analysis showed no association between ∆TSI across transition, or from baseline, with performance in either TSI of the VL / LD during T1. When the data was split into best versus worst performers, no significant differences could be identified. During T2, no association was found between ∆TSI across transition, or from baseline, with performance in TSI of the VL. However, when split into best versus worst performers, differences could be found in TSI of the VL. Those who completed the triathlon more quickly reached a peak TSI 3% *lower* than pre-race baseline (∆TSI = -3%) during T2, compared to the worst performers who reached a peak TSI 2.2% *higher* than pre-race baseline (∆TSI = 2.2%), a 5.2% difference $p < 0.05$. No such differences between best and worst performers were evident in TSI of the LD during

T2, however there was an association between ∆TSI of the LD from pre-race baseline during T2 and swim time r= 0.679, *p*< 0.05.

4.8.7.1.2. Maximal incremental swim test

End recovery data from the maximal incremental swim test provided: TSI of the VL half-life 30.76 ± 12.93 seconds, Tau 44.38 ± 18.66; TSI of the LD half-life 20.65 ± 17.32 seconds, Tau 20.11 ± 6.74. No significant correlations were found between maximal incremental swim test end recovery data and discipline or overall completion time. No differences were seen between best and worst performers.

4.8.7.1.3. Maximal incremental bike test

End recovery data from the maximal incremental bike test provided: TSI of the VL half-life 18.72 ± 7.35 seconds, Tau 27.01 ± 10.60; TSI of the LD half-life 32.19 ± 17.36 seconds, Tau 43.02 ± 24.43. No significant correlations were found between maximal incremental bike test end recovery data and discipline or overall completion time. No differences were seen between best and worst performers.

4.8.7.1.4. Maximal incremental run test

End recovery data from the maximal incremental bike test provided: TSI of the VL half-life 29.04 ± 16.79 seconds, Tau 31.99 ± 18.64; TSI of the LD half-life 12.85 ± 7.35 seconds, Tau 13.01 ± 9.02. No significant correlations were found between maximal incremental run test end recovery data and discipline or overall completion time. No differences were seen between best and worst performers.

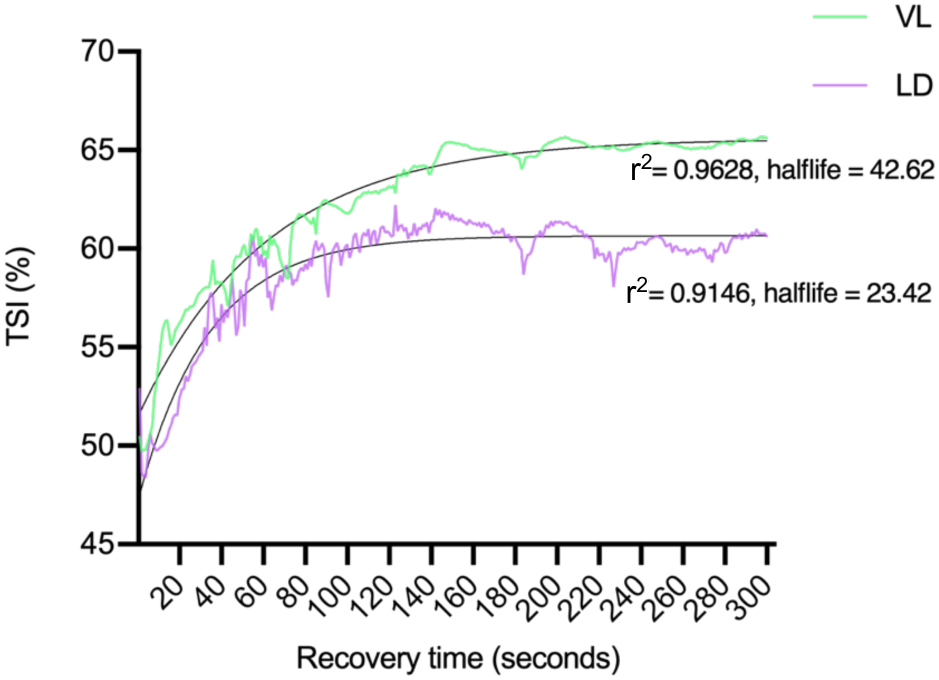


Figure 13. Exponential curve fit of muscle oxygen recovery following the simulated triathlon (representative participant shown).

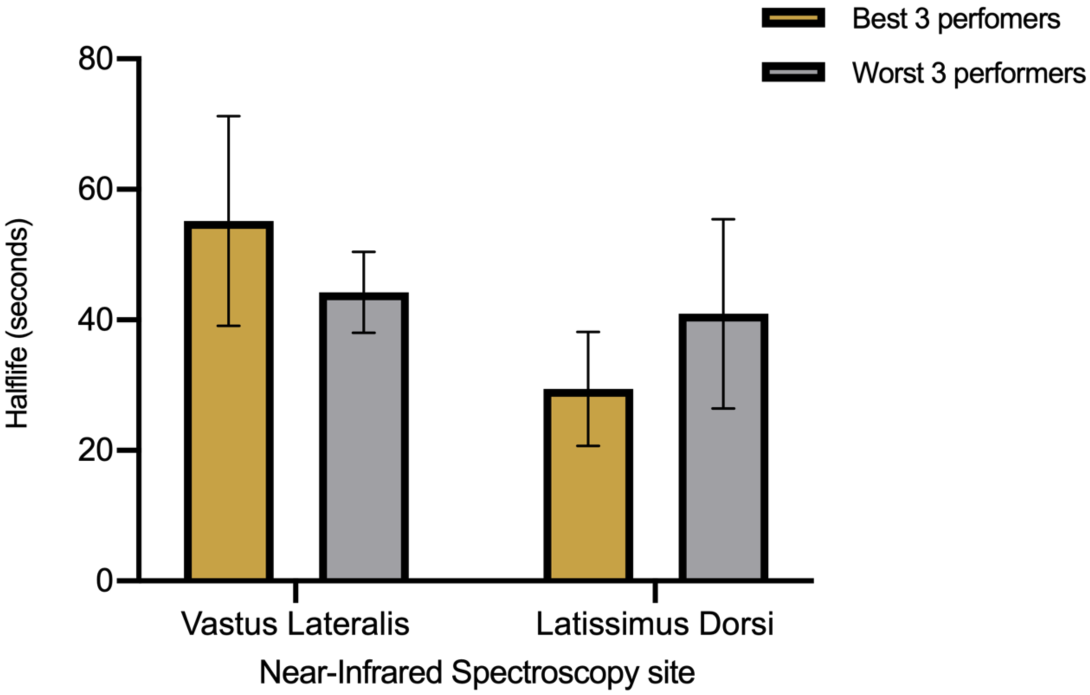


Figure 14. Simulated triathlon half-life end recovery time (seconds) of TSI in best vs worst performers.

4.8.7.2. Oxygen Consumption

Participants who achieved a high $\dot{V}O_2max$ during the maximal incremental swim test, were able to produce faster swim times $r= -0.686$, $p= 0.029$, likewise those who could sustain a high $\dot{V}O_2$ during the triathlon swim also swam faster times $r= -0.768$, $p= 0.010$. Neither $\dot{V}O_2max$ nor $\dot{V}O_2$ during the swim stage of the triathlon correlated to overall triathlon completion time, $p< 0.05$.

$\dot{V}O_2max$ achieved during the maximal incremental bike test correlated to bike time and overall triathlon completion time, $r= -0.760$, $p= 0.007$, $r= -0.810$, $p= 0.003$ respectively. Likewise, average $\dot{V}O_2$ sustained during the bike stage of the simulated triathlon also correlated to bike time, $r= -0.622$, $p= 0.041$, and overall triathlon completion time $r= -0.810$, $p= 0.003$.

$\dot{V}O_2max$ achieved during the maximal incremental run test, and the average $\dot{V}O_2$ sustained during the run stage of the simulated triathlon did not significantly correlate to run time, $r= -0.461$, $p= 0.154$, $r= -0.474$, $p= 0.141$ respectively. However, these metrics did significantly correlate to overall triathlon completion time, $r= -0.666$, $p= 0.025$, $r= -0.844$, $p= 0.001$ respectively.

When comparing best versus worst performers, there was no significant interaction between discipline and performance level, $F(2,8) = 0.341$, $p = 0.596$, $\eta_p^2 = 0.079$. Despite being not statistically significant, $p> 0.05$, the best performers were able to sustain a higher $\dot{V}O_2$ to a large effect size (Swim +9.67 $ml.kg^{-1}.min^{-1}$, $p= 0.06$, $\eta_p^2 = 0.629$; Bike +15 $ml.kg^{-1}.min^{-1}$, $p= 0.10$, $\eta_p^2 = 0.532$; Run +11.67 $ml.kg^{-1}.min^{-1}$, $p= 0.06$, $\eta_p^2 = 0.612$), Figure 15.

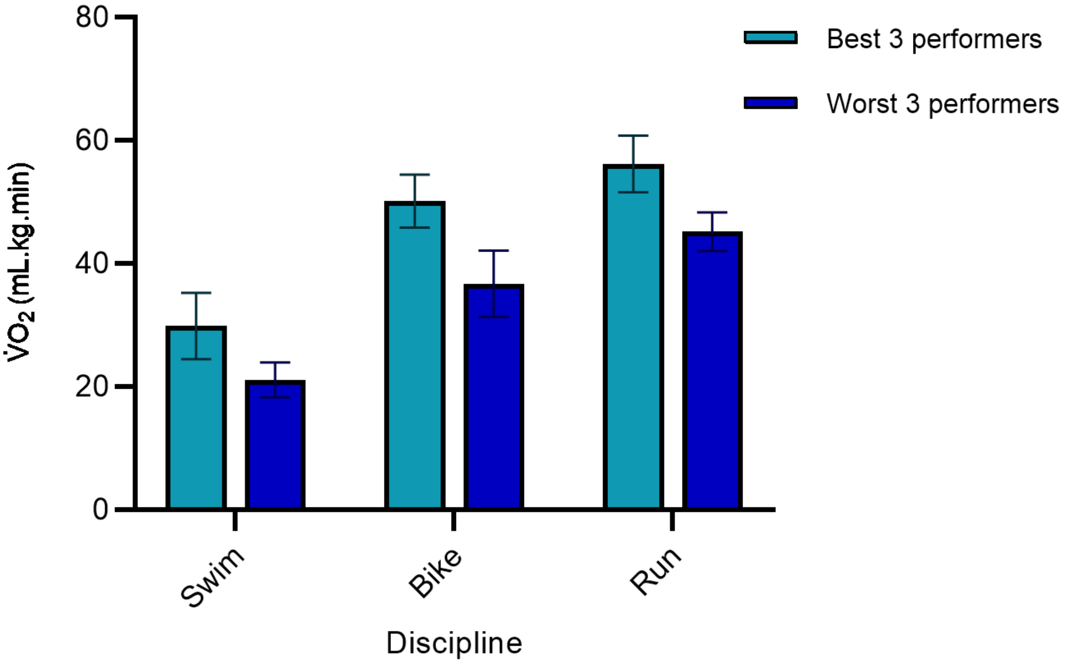


Figure 15. $\dot{V}O_2$ sustained during simulated triathlon effort in best vs worst performers.

4.8.7.3. Heart Rate

HRmax during the maximal incremental swim test did not correlate to swim time or overall triathlon completion time, $p< 0.05$. Average HR sustained during the swim stage of triathlon was significantly correlated with an inverse relationship to swim time r= -0.730, $p= 0.011$, and in the opposite direction in overall triathlon completion time r= 0.672, $p= 0.024$.

No correlations were observed between HRmax or average HR during the bike stage of the triathlon, with bike time or overall triathlon completion time. The same patterns were seen with respective values within the run. When comparing best versus worst performers, there was no significant interaction between discipline and performance level, F(2,8) = 1.013, $p = 0.373$, $\eta_p^2 = 0.202$. Despite being not statistically significant,

$p > 0.05$, the best performers were able to operate at a lower HR in the bike and run stages to a large effect size (Bike -11.33 bpm, $p = 0.283$, $\eta_p^2 = 0.277$; Run -19.67 bpm, $p = 0.135$, $\eta_p^2 = 0.466$), Figure 16.

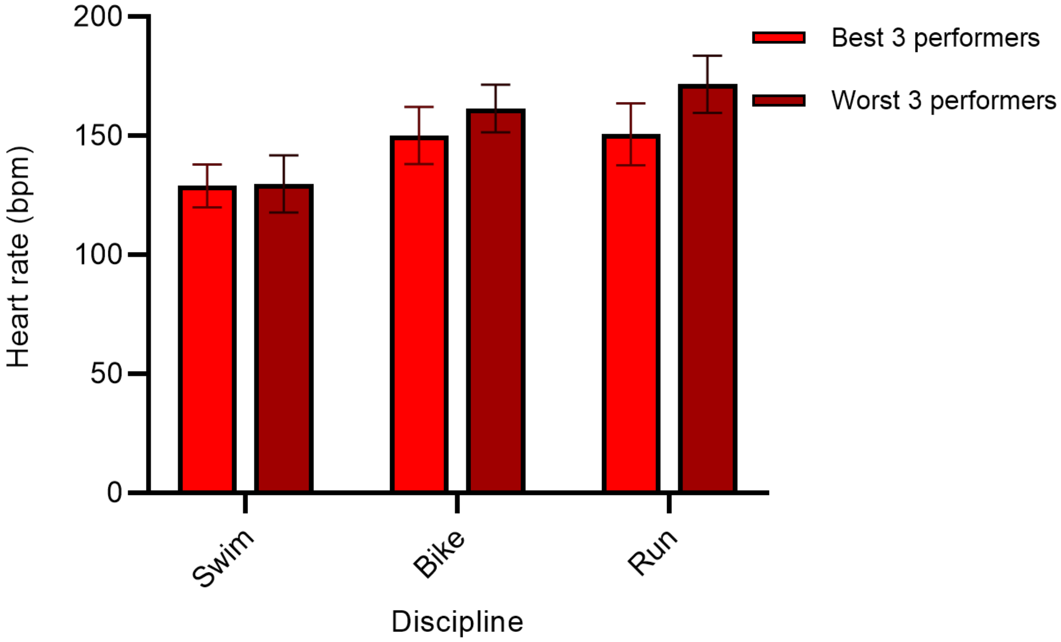
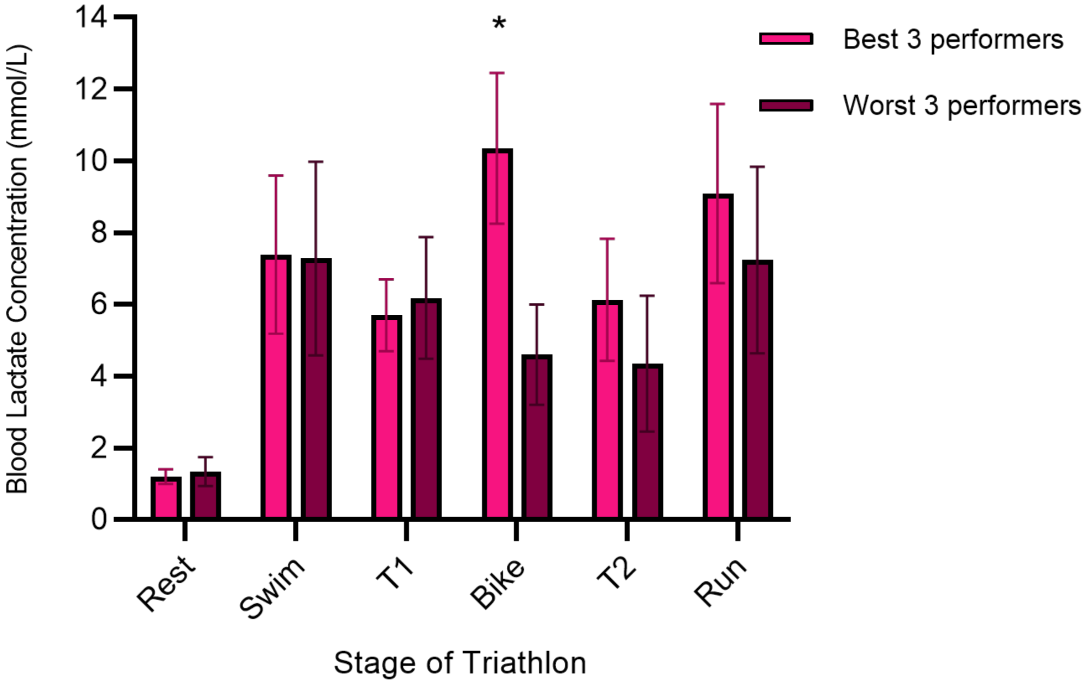


Figure 16. HR sustained during simulated triathlon effort in best vs worst performers.

4.8.7.4. BLC and RPE response

Swim time was significantly correlated to multiple BLC time points, including: BLC at completion of bike stage r= -0.611, $p = 0.046$; BLC at the end of T2 r= -0.666, $p = 0.025$; and BLC at completion of the run stage r= -0.787, $p = 0.004$, where faster swim times equated to higher BLC. Conversely those who achieved faster run times, had lower BLC at the completion of the swim stage r= 0.645, $p = 0.032$ and at the end of T1 r= 0.777, $p = 0.005$. When comparing best versus worst performers, there was a significant interaction between discipline and performance level, there was a significant interaction between time point and performance level, F(6,24) = 5.463, p =

0.001, $\eta_p^2 = 0.577$*.* Pairwise comparisons report that BLC trends were similar across the majority of the triathlon, however best performers had a significantly greater BLC at the end of the bike stage, $p = 0.018$, $\eta_p^2 = 0.788$, Figure 17. No correlations were found when analysing RPE response.



*Indicates statistically significant difference to worst performers, $p < 0.05$.

Figure 17. BLC across all time points during simulated triathlon effort in best vs worst performers.

4.9. Discussion

The primary purpose of this study was to assess the utility of NIRS devices as a new measurement tool to assess physiological responses to triathlon. As expected, the NIRS devices worked effectively during the experimental study and the equipment was able to inform upon muscle oxygenation status of both upper and lower limbs across the full triathlon simulation. NIRS devices were attached to both upper and lower limbs due in order to account for the predominant muscle used within the different disciplines during triathlon. Muscle oxygenation of the VL and LD yielded different responses between sites and disciplines as expected due to these muscle groups working to a different extent throughout. The information provided by the NIRS device is different to that of global physiological data. This is due to global physiological measures of oxygenation measuring total oxygen uptake at tissue level, whereas NIRS informs upon peripheral oxygen delivery and utilisation within the site of gas exchange in the muscle. Although it is necessary to understand the differences in these measures, they do not need to be mutually exclusive when it comes to research observations. Therefore, a secondary aim of this study was able to observe both peripheral and global physiological data simultaneously, which produced a full profile of the physiological responses to a full sprint triathlon simulation. The NIRS data complemented the other measures of $\dot{V}O_2$, HR, BLC and RPE and the created profile provides useful insight into the physiological responses of each stage of the sprint triathlon, including the responses of the transition stage. The profile provides an understanding of the oxygenation response during the swim stage, which is a new addition to the literature in this area. The importance of swim data cannot go unnoticed, as swim performance had great impact on athlete performance in the following stages of the race, with those performing best in the swim typically unable to sustain the high intensity throughout. Further associations to performance were found within the $\dot{V}O_2$ and HR responses, where the best performers are able to operate at a higher $\dot{V}O_2$ throughout the entirety of the triathlon, paired with a lower HR, to large effect sizes. This study was able to profile the physiological response, both peripheral and global, across a full sprint triathlon simulation whilst identifying associations to performance, and therefore successfully achieved all four aims that were set out.

The NIRS responses provided far greater variability (standard deviation) between participants than global physiological measures. Furthermore, NIRS was also able to identify unique individual aspects (differing desaturation profiles, within individuals and muscle groups), with such differences not evident from global physiological data alone. It is difficult to determine whether this new measure, and the unique capabilities of the technology, provides a truly useful tool to aid sports performance whilst used in isolation. NIRS provided new information on the peripheral response to exercise during the triathlon event which was added to previously known global physiology measurements, enabling a more comprehensive oxygenation profile during sprint distance triathlon. Based on the TSI response alone, it was determined that oxygenation of the VL generally remains stable during swim and cycle disciplines, and deoxygenates in the latter stages of the run, where oxygenation profile of the LD is more variable. As expected, the responses are dependent on the muscle groups being worked.

NIRS aids the researcher in understanding the differences between global and peripheral responses, identifying disparities in relative intensity within VL oxygenation during swim and run disciplines, with significant differences between LD oxygenation and global measures across all stages of the triathlon simulation. Also, the NIRS data established differences in relative intensity of muscle oxygenation across each stage, where little to no differences were found within the $\dot{V}O_2$ and HR data between swim, cycle and run disciplines. Most notably perhaps is the magnitude of the standard deviations in the TSI data, most pronounced in TSI of the VL, in comparison to the global physiological data, and needs to be examined further. Whether this data is useful, or simply different will be discussed further in below sections. Inter-individual variations in NIRS response is a common theme in the literature (125, 153, 154) and has been proposed to identify differences in participant's ability to utilise oxygen in the muscle. However, it has also been suggested that such differences could be attributed to a variety of factors, such as the participant's technique for a particular movement, or underlying blood flow differences identified when analysing other NIRS derived data such as tHb (not carried out in this analysis). Differences in NIRS response in this study were not associated to performance level using correlational analysis, indeed no NIRS derived oxygenation responses during swim, cycle or run disciplines were positively associated with discipline-specific or overall triathlon performance. Some of the explanation may be attributed to different anthropometrics between subjects as NIRS data has been suggested to provide different values dependent on skinfold thickness (150). To minimise this the TSI signal was selected as the NIRS measure least affected by this issue (149) and no correlations were found between extent of desaturation and skin fold thickness in VL (r= -0.05, *p*= 0.871) or LD (r= -0.09, *p*= 0.808) in the present study. Further reasoning arises when assessing the apparent heterogeneity of muscle deoxygenation between participants (155), where pronounced differences between participants could be reasoned by participants' musculature naturally de-saturating to different extents. Further to this issue, despite NIRS device placement being rigorously controlled during this study, it may be that individual techniques and the way participants use their leg muscles could result in certain areas of the VL being more active than others (155). It is suggested that these factors could attribute to the lack of correlations found between NIRS response and triathlon performance. Similarly, authors have evidenced changes in NIRS responses following training, with no obvious correlations between oxygenation change and performance (156, 157). It may be that muscle oxygen profile is not associated to triathlon performance in a laboratory setting.

In the following sections the findings will be split into swim, cycle, run and transition, and discussed further.

4.9.1. Swim

The literature review identified a gap in research understanding surrounding the swim stage of the triathlon race, as a result of the limited measurement tools available in aquatic environments. The swim stage in this study was completed using swim ergometry, to enable replication within a laboratory environment. This modality of swimming does not incur the methodological complexities of aquatic environments, and despite it being the best non-aquatic swimming option, it does lack of ecological validity. Nonetheless, the NIRS was able to inform on muscle oxygenation during this stage of the race. The initial stages show contrasting TSI responses for the VL and LD, proposed to be a result of blood flow redistribution, however after reaching a plateau no significant differences were evident between the sites. Interestingly, two participants (*h* and *k*) demonstrated deoxygenation of the VL during the swim stage of the triathlon simulation. This is surprising given the swim bench used in this study did not give the participants the ability to create propulsion using the lower body. Previous studies have shown desaturation of non-active muscles during exercise (BB during running (93)), however with only two participants in the present study showing this response, this highlights the ability of NIRS to provide muscle oxygenation differences not possible via global measures. Explanations for this may be attributed to differences in swim technique between these participants versus the rest, such as the contraction of the lower limbs that may not be beneficial during this part of the race. Although not evident in the individual NIRS traces, when calculating relative intensity TSI of the VL was comparable between swim and run stages, adding further evidence to the use of the VL by participants on the swim ergometer. As a measurement tool NIRS can therefore aid coaches in examining the techniques of their athletes, in order to refine movement patterns and to save their legs for the latter disciplines. With economy being an important determinant of triathlon performance (9), and swimming economy in particular an area where triathletes can make sizeable improvements (3), NIRS could be useful in providing insight into this measure. Given the lack of ecological validity of the swim bench, observation is needed within a swim environment to confirm these findings.

There is currently no available research that has assessed the NIRS response to swim ergometry for comparison. Triathletes have previously been used in swim based NIRS research, and the TSI patterns do not match that of the current study, with much larger differences in muscle oxygenation response between VL and LD seen in the literature (118). However, this research also observed club level swimmers, and the TSI responses seen in this population more closely match the responses seen by triathlete participants in the present study. The disparities here were attributed to swim specialists using the lower body to a greater extent for propulsion in comparison to triathletes, which again questions why such patterns were seen in the present study. Researcher observation showed that participants did not display a kicking motion during the swim stage of the triathlon simulation, however it may be possible that participants were engaging the VL muscle to aid balance on the ergometer.

Due to logistical complexities no study has yet reported on the $\dot{V}O_2$ response during the swim stage of a triathlon, and therefore the use of a non-aquatic swim ergometer in the present study at least provides some insight into the physiology of this stage of the race. It can be seen that triathletes operate at 72 ± 10% of $\dot{V}O_2$max, lower than any other time during the race (Bike 80 ± 9% , Run 82 ± 6%), although the differences are not statistically significant. The free breathing available in this study, versus in water, may affect the $\dot{V}O_2$ response in a different way to triathlon training and competition, as previously stated above the swim ergometer lacks ecological validity. Triathletes operated at 90 ± 9% of HRmax across the swim stage, higher than the 85% previously reported in the literature (42). It must be noted that this HRmax in the literature was calculated from a treadmill test, and therefore expected to be a lower percentage due to athletes recording higher HRmax whilst running. During maximal exercise testing participants largely failed to meet the HRmax criteria, although this is attributed to the prone position of the swim testing, and change in hydrostatic gradient (152). When in aquatic environments, individuals will be subject to the diving response, a natural physiological mechanism that initiates bradycardia reducing HR among other processes (158). Therefore, comparing the HR response from swim ergometry to simulated triathlon must be done so with caution. Similar levels of BLC, 7.39 ± 1.6 mmol/L versus 5.7 ± 1.8 - 6.5 ± 1.1mmol/L in the literature, and identical RPE, 15 ± 1, suggest this simulation closely matched that of triathlon competition in this regard (42, 43).

As seen in the literature (51), participants who achieved high $\dot{V}O_2$max scores were able to produce faster swim times, however this did not transfer into overall triathlon performance. Most interestingly it appears that there is an opposite relationship between HR during the swim stage and swim time versus triathlon time, with those having a high HR performing best in swim stage, and those with low HR performing best in overall triathlon. This is furthered by BLC responses, where faster swim times equated to higher BLC in the later stages in the race, and those who had lower BLC after the swim stage were able to complete the run stage the fastest.

Previous research has suggested the swim stage is crucial in triathlon competition for tactical and drafting reasons (24, 39). Although it appears the fastest swimmers in this study ultimately failed to be the best performers overall, in real world competition they may have benefitted from being in the leading group of athletes exiting the water (1, 38, 40, 41). Due to the equipment used to simulate the sprint triathlon it is also a possibility that the fastest swimmers in this study were participants who were able to perform better on the swim ergometer than others. However, this study does add further support to suggestions that athletes should avoid being influenced by the race dynamics, and complete the swim at lower intensity to create an energy reserve that can be used in bike and run stages (2, 23).

4.9.2. Bike

The bike stage of the triathlon simulation is where oxygen saturation differences between the muscles are most evident. This is as expected, with this being the only discipline where one half of the body, in this case the upper body, is able to rest somewhat. The relatively stable nature of the VL oxygenation trace highlights that a constant effort was executed by triathletes, as triathletes largely operated within a ~1.5 km/h window of variation throughout. The variation in TSI of the LD across the bike stage is likely to be explained by the participants position on the bike, and the activation of the LD as a result in changes in position. It is known that when cycling athletes are able to use their upper body for extra power generation (159, 160). Although not pronounced during this 0% grade time trial, it was evident from the live NIRS trace during maximal incremental testing that when participants started to consciously pull on the bars in order to create more power, TSI of the LD started to reduce (potentially a result mostly due to local vasoconstriction - rather than a redistribution of oxygenation). The TSI of the VL during the bike stage of the triathlon matched the HR and $\dot{V}O_2$ intensity in which was a rare occurrence for peripheral intensity to match global intensity. Due to the leg muscles providing all of the propulsion whilst cycling, and the upper body required to do minimal work, if at all, it can be understood why these values are comparable. The large inter-individual variations in desaturation are most pronounced in VL muscle during the bike stage. For example, participant *d* experienced considerable desaturation (Participant *d* 35.5% versus 16.6% Group mean excluding participant *d*) without displaying any noticeable signs of extra-ordinary effort or struggle to the researcher. This large disparity between participant *d* and the rest of the participants was only noticed by using the NIRS devices, and as previously suggested, would give a coach reasons to examine technique used during this stage in order to refine movement patterns.

Upon reaching a plateau in $\dot{V}O_2$, participants in this study operated at 80 ± 9% of $\dot{V}O_2$max with little variations across the entirety of the bike stage, matching that seen in previous literature (74 - 82%) (12, 14). Also similar to prior research was participants operating at 93 ± 10% of HRmax (versus 86 - 90%) throughout the bike stage, and finishing this stage with BLC of 7.5 ± 3 mmol/L (versus 7 mmol/L) (12, 14, 42). It would be expected that the $\dot{V}O_2$, HR and NIRS traces may differ in triathlon competition from the relatively stable oxygenation seen, due to the requirements of short high intensity bursts, and variations in speed and power output as a result of the geographical nature of the course (58).

The completion time of the bike stage produced significant associations to overall performance ($r= 0.902$, $p< 0.001$), and accounts for the most variance in overall completion time out of any stage in the race. $\dot{V}O_2max$ during maximal testing, and sustained $\dot{V}O_2$ during the triathlon could be associated to completion time of the bike stage and overall triathlon, with high $\dot{V}O_2$ scores resulting in superior performance. This is further evidenced when the data was split into performance levels, with best performers sustaining a higher $\dot{V}O_2$ ($+15\ ml\cdot kg^{-1}\cdot min^{-1}$) during the triathlon compared to worst performers to a large effect size. This was paired with best performers producing this with a lower HR ~10 bpm, again to a large effect size. No correlations were observed between HRmax or average HR during the bike stage of the triathlon, with bike time or overall triathlon completion time. BLC at the conclusion of the cycle stage has previously been associated with overall triathlon performance ($r = 0.83$, $p< 0.05$) (80), although no such trends were observed in the present study. The bike stage did however display a significant difference in BLC scores between best and worst performers (Best 10.4 mmol/L versus 4.6 mmol/L Worst), heightened by both performance levels leaving T1 with comparable levels of BLC (Best 5.7 mmol/L versus 6.2 mmol/L Worst) as seen in Figure 17. It would be expected that these high levels of BLC would be unsustainable for best performers, however during T2 BLC levels reduced back to a similar level seen by worst performers (Best 6.1 mmol/L versus 4.4 mmol/L Worst), perhaps suggesting greater recovery ability.

4.9.3. Run

The NIRS trace during the run stage of the race can be characterised as a steady decrease in oxygen saturation throughout, with more dramatic reductions in muscle oxygenation as participants made their final burst to the finish line. The NIRS devices were able to inform upon TSI response of the LD whilst running, an area not covered in the literature, which remained similar to TSI of the VL throughout. The run stage gave the greatest variation in oxygenation responses across all participants, with increases, decreases or stability in TSI of the VL / LD. The run stage of the race is the only area where NIRS has previously been applied to triathletes; where muscle oxygenation in VL was observed during a 12 minute run effort preceded by a 20-minute cycle time trial (the swim stage was not included in this study) (55). The study used a different NIRS device (Moxy monitor) and therefore reported %SmO_2, although this measurement is comparable to TSI reported in the present study. The traces across both studies follow a similar pattern, ~60% (present study 61 ± 6%) muscle oxygen saturation at the onset of the run, although lower levels of desaturation were seen in the present study (minimum muscle oxygen saturation 52 ± 8% versus <40%). The differences may be explained by the nature of the bike stage, with +20 bpm differences in HR in favour of the previous research. Furthermore, the participants in the present study had to run a greater distance (5 km versus 3 km), and therefore maintained a slower speed and intensity throughout, which is likely to give greater values of muscle oxygenation.

During this final stage of the simulation, triathletes operated at 82 ± 6% $\dot{V}O_2$max and 87 ± 7% HRmax, similar to that previously reported in the literature (83 – 90% $\dot{V}O_2$peak, 92% HR max (12, 14)). Both $\dot{V}O_2$ and HR traces appeared largely stable, rising gently throughout, before noticeable increases in the final stages as participants put in a final burst. BLC concentration was marginally higher than that previously reported 8.6 ± 2.2 mmol/L versus 7.6 ± 0.9 mmol/L (12), where methodological differences in site of BLC collection (fingertip versus earlobe) may explain the small variations between studies with similar differences ~1 mmol/L previously observed during submaximal exercise (161).

Completion time during the run stage could be correlated to overall triathlon completion time ($r = 0.78$, $p = 0.004$), suggesting this stage is important to achievement within the sport. However, completion time of this discipline was not associated to $\dot{V}O2max$ or average $\dot{V}O2$ sustained during the run stage $p > 0.05$. These metrics however could be significantly associated with overall triathlon completion time $p < 0.05$, as seen previously in the literature ($r = -0.80$, $p < 0.001$) (14). No correlations were observed between HRmax or average HR during the run stage of the triathlon, with run time or overall triathlon completion time. Identical to the patterns seen within the bike stage, best performers were able to sustain a higher $\dot{V}O_2$ (+11 $ml \cdot kg^{-1} \cdot min^{-1}$) whilst operating at a lower HR (-20 bpm), to a large effect, when compared to worst performers.

These continuing patterns of higher $\dot{V}O_2$ and lower HR suggest that the best performers had superior cardio-respiratory endurance capabilities than worst performers, as would be expected. However, this did not carry over into the triathlon end recovery TSI data, where best performers had elongated half-life recovery times in the VL muscle. Previously recovery time had been correlated with repeated sprint swim performance $r = 0.70$, $p < 0.01$ (119), but no such performance associations were evident in the present study. With previous researchers utilising differing analysis approaches for recovery data this has led to differing methods of calculation within recovery (e.g expontial curve fit, recovery half-time, linear modelling and percent per second reoxygenation), and difficulties when comparing between studies. It seems that muscle oxygenation recovery rates are generally applied to repeated sprint research (119, 162), and it might be this difference in duration of exercise which caused inconclusive results. However, other researchers have also failed to find associations between reoxygenation rate and performance, which perhaps suggests that muscle oxygenation recovery rates are not an associated or limiting factor to endurance performance (162).

4.9.4. Transition

Recovery calculations were initially proposed to be used during triathlon transition, aiming to add to the lack of research understanding in this area, however it was not possible to curve-fit the data during transition. The studies influencing this idea all used passive recovery periods, but it must be highlighted that the transition areas during triathlon are not merely a passive recovery zone, but an important part of the race (3, 14). The ‘M’ and ‘W’ traces seen in TSI and $\dot{V}O_2$ and HR responses respectively during transition can be explained by enforced rest periods implemented to record BLC, before running the transition, then stopping to change to the appropriate footwear required for the next stage of the race, before running out of the transition area. The dramatic changes that are portrayed during transition on the full triathlon graph become less severe when transition areas are graphed separately (Figure 10). Where the NIRS does provide effective monitoring during transition is profiling the different oxygenation responses of the muscles, most pronounced during the T1 transition.

Here the VL muscle has been less active during the swim stage, and hence deoxygenates at the onset of running in transition, whereas the LD muscle has been heavily used during swimming but is less relied upon during running, and therefore can reoxygenate.

Section 2 identified that cycling preceded by swimming has different physiological demands to that of isolated cycling, including reduced efficiency, increased BLC and increased $\dot{V}O_2$ (1, 2, 49), although the exact causes of these effects are unknown. The NIRS observations here, showing reduced muscle oxygenation of the VL throughout T1 adds to the understanding, and might influence the physiological differences observed. Such information cannot be acquired from solely analysing the $\dot{V}O_2$ and HR graphs, and these patterns also provide support for the literature proposing NIRS is able to react more quickly to sudden changes in intensity (129). It is important to note that although T1 is where athletes transition from swim to bike, neither modalities are performed during transition, and all physiological changes seen are influenced by athletes running through transition. Within triathlon competition wetsuits are mandatory during the swim stage in water temperatures below 14 °C, optional in water temperatures up to 22 °C, and become forbidden when water temperature exceeds 22 °C (163). The use of a wetsuit may therefore alter the physiological response seen during transition, either due to athletes arriving at transition in a reduced physiological condition (44, 45, 47), or due to the physiological cost of wetsuit removal during T1 (57).

Data gathered during T2 shows a more similar pattern in oxygenation of the VL and LD, with values staying fairly stable throughout. This would suggest there is comparable use of the LD and VL muscles in the final stages of the cycle and the running performed during transition. With the LD muscle being used to a greater extent during the final kilometres of the cycle stage, and VL being used continuously whilst cycling, neither muscle had an absolute rest during when running through transition and is the likely reason behind the stability of TSI through T2. These findings are unable to provide any further understanding of the physiological complications that occur following T2, the greatest unknown identified during the literature review. Neither transitional graph shows obvious changes in TSI, $\dot{V}O_2$ or HR halfway through transition where triathletes stopped running, changed their footwear for the next discipline, before running the final stages of the transition. When comparing ∆TSI (maximum TSI reached during T2 minus pre-race baseline) of the VL, differences were found between best versus worst overall triathlon performers. Best performers reached a maximum of -3% ∆TSI from pre-race baseline, compared to the 2.2% of worst performers, a 5.2% difference. This would suggest that those who completed the triathlon more quickly arrived at T2 at a lower oxygen saturation, perhaps exerting greater effort across the cycle stage. It was previously described that best performers complete the cycle stage more quickly, and at a greater intensity, so the NIRS data fits the narrative of the other recorded measures during T2. A second possible explanation, although speculative, is that worst performers were less time efficient during the transition period, spending more time stationary when changing attire for the run stage, and this elongated passive rest would allow for greater reoxygenation. A positive association between ∆TSI of the LD from baseline and swim time $r = 0.679$, $p < 0.05$ was identified during T2, suggesting lasting effects of the fatigue created from the swim stage, again fitting the narrative of the other global physiological measures. Although the LD is not a primary factor in running performance, this outcome highlights the importance of not completing the swim stage at maximal intensity.

As seen in the literature, RPE appears to ‘reset’ between each stage of the race (73). This has previously been proposed due to athletes misunderstanding of the RPE measurement, using local muscle discomfort as an influencer factor in their judgement (164). With different muscular contributions between stages, it can therefore be understood why this RPE resetting pattern is seen during triathlon. With the recent development of a rating of fatigue scale (165), it may be useful in future study to use this measurement tool, rating upper and lower body fatigue separately to compliment the physiological data recorded.

4.9.5. Limitations

It is important to recognize that this study has its limitations and despite efforts to replicate a sprint triathlon the study was completed solely within the laboratory. (The original intention of this testing series was to complete a follow up experimental study in the field, however this was not possible due to Covid-19 restrictions. This caused the structure of the thesis to shift towards a review of the literature, and a single laboratory based experimental study. The importance of recreating this methodology in the field has not gone unnoticed.) Testing within a laboratory does have its benefits, allowing for a greater amount of control, access to a wider range of specialist measurement tools and the ability to create the same conditions for each participant.

The laboratory environment allowed for the swim stage to be replicated using swim ergometry, providing ease of measurement and the opportunity to measure $\dot{V}O_2$ during this stage which is limited in the current literature. This modality of swimming does not incur the methodological complexities of aquatic environments, and despite it being the best non-aquatic swimming option, it does lack of ecological validity. A swim ergometer is rarely used by athletes, who typically opt for pool or open water swimming for their training, and despite multiple familiarisation sessions built into the methodology it is unlikely that participants were truly comfortable on this equipment. Swim ergometry does not replicate the body temperature of being in water, the external pressures and performance benefits of wearing a wetsuit, or the potential requirement of turns during pool swimming. These factors all contribute to the physiological response of the athlete, therefore the wide variety of swimming modalities used in the literature and the combinations of these influencing factors makes it difficult to compare responses between studies.

Triathlon is fundamentally a racing sport, subjecting athletes to react to competitors' actions, of particular importance during draft-legal racing, rather than solely selecting a pacing strategy that is the most effective (23, 24). It may be that the results of this study are more similar to that of a fully drafted triathlon, due to the participants not being subject to environmental factors that would slow them down in this study. Secondly, this study did not account for geographical variations in typical triathlon courses, particularly evident in the NIRS response to the bike stage, where the trace remained stable as participants stayed at a set pace throughout. This did not allow for the exploration of NIRS as a measurement tool to react to these varying conditions.

Variation in pace and intensity was also limited during the run stage of the triathlon simulation, partly due the continuous gradient of the treadmill (1%) creating a flat course that would be highly unlikely in competition. Secondly, the run stage was not entirely self-paced as the use of a motorised treadmill in this study meant that participants had to communicate to the researcher when they wanted to alter the pace of the treadmill, and although frequent changes of treadmill speed were encouraged this would not match the variability in pace observed in the field. Although curved non-motorised treadmills were available to the researcher, which provide the self-pacing element missing in this stage of the triathlon, they provide unique physiological responses that cannot be compared to motorised treadmill or over ground running. Physiological responses, including $\dot{V}O_2$, HR, BL and RPE, are elevated when using curved non-motorised treadmills (166-168), with responses equivalent to motorised treadmills set to 6-8% gradient (169), therefore a motorised treadmill was selected for this triathlon simulation.

4.9.6. Future Study

Although this initial research was necessary, ultimately the methodology must be replicated in the field to understand the physiological responses to sprint triathlon in the real world. It is advised that this research closely reproduces the conditions of triathlon competition and may need to omit BLC measurement in order to increase the validity of transition data. There are no concerns in repeating the NIRS protocols in the field. Previous research has stated the change in temperature in an aquatic environment has negligible effects on temperature (118), whilst NIRS devices are stable to external pressures, that could be created by a wetsuit, up to 30mmHg (113). Prior NIRS swim research has been completed using tight-fitting swim attire with no recorded complications (118, 119). It may be that future study looks to add further NIRS devices, gaining multiple measurements on both upper and lower body to increase the information and understanding of muscle oxygenation during triathlon. The need for $\dot{V}O_2$ data in such studies must be carefully considered, with the complexities of recording $\dot{V}O_2$ during swimming, an enforced stop would need to be implemented to attach a respiratory gas analyser in T1. With the $\dot{V}O_2$ findings of bike and run stages in this study being similar to that of the literature, it may be concluded that there is an adequate understanding of this physiological response. The variations in the TSI, $\dot{V}O_2$ and HR data across each stage of the triathlon highlight the importance of displaying such traces in research, with a single average value characterising the whole stage not portraying the full story. Following this, studies should look to incorporate a racing element, either physical or virtual, to see how the physiological responses differ in a race situation where the pressures of competition cause athletes to adopt strategies that are not energetically optimal (23, 24).

5. Conclusions

The aim of this study was to observe the physiological responses to sprint triathlon, primarily utilising NIRS technology to inform on responses at a peripheral level, whilst simultaneously measuring commonly used global physiological data. The physiological profile created combines peripheral and global responses in a way not currently available in the literature, providing further understanding of the demands placed upon athletes when competing in this discipline.

Although previous interest had been shown, NIRS had yet to be applied within the sport of triathlon, where in the present study no complications arose when applying the technology to this new setting. This study has shown that NIRS technology can be successfully implemented into the sport of triathlon as a tool to inform upon multi-site muscle oxygenation, identifying differences between sites and to global physiological measures, as well as informing upon participant technique and highlighting the possibility for improvements in efficiency. These findings have potential implication for training and competition strategies, as it highlights the differences in physiological demand between the upper and lower body across each discipline.

The previous review highlighted the swim discipline as an area of little research, due to the logistical complexities of completing physiological testing in an aquatic environment. As a result, a secondary aim of this study was to produce an oxygenation profile of the swim stage, a response that had not been recorded to this point. The use of a laboratory-based swim-ergometer in this study allowed for ease of $\dot{V}O_2$ measurement and provided new insight into the physiological demands of this part of the discipline. Although this methodology could not be replicated to record swim $\dot{V}O_2$ in the field, there would be no forecasted difficulties in replicating NIRS application in triathlon field research in the future.

The NIRS technology also provided new information about the physiological responses during transition, namely identifying differences between oxygenation of the VL and LD during T1. This new insight could be combined with previous understanding to help understand the differences between triathlon cycling and isolated cycling. This study did not see differences between oxygenation of the VL and LD during T2 and was therefore unable to provide reasoning for difficulties experienced after T2, and this remains an area with a lack of research understanding. The ΔTSI during T2 (maximum TSI reached during T2 minus pre-race baseline) suggest that best performers have exerted themselves to a greater extent in swim and cycle disciplines, and that worst performers may need to revise their competition strategy. This observation using NIRS data fits the narrative provided using other global physiological measures during T2. Responses of $\dot{V}O_2$, HR, BLC, and RPE across the full triathlon match that previously observed in the literature, including field research, validating this laboratory simulation as similar in physiological demand as that of triathlon competition. This study was able to provide relevant intensities in comparison to discipline-specific maximal testing, where other work has used maximal running tests for intensities across all disciplines which would not be totally accurate.

A final aim of this research was to identify the physiological responses associated with triathlon performance, where both the literature review and the experimental study identified significant associations between measures of $\dot{V}O_2$ and BLC and performance. It appears that best performers are able to operate at a higher $\dot{V}O_2$ throughout the triathlon, paired with a lower HR. Also, it was identified that athletes who “go off” fast in the swim have high BLC later on in the triathlon and subsequently struggle to maintain intensity, whilst those who perform better in the later stages have lower BLC after the swim stage. Large inter-individual differences were seen within the NIRS data, with no evident differences between TSI response in best and worst performers. With no apparent associations between NIRS measures during swim, cycle and run and completion time during these disciplines, at this point the usefulness of such data in aiding performance is yet to be determined. The key NIRS findings from this research are the disparities between TSI of the VL / LD and measures of global physiology, and the inter-individual differences that are evident between participants which could not be determined using solely global physiological measures. The inter-individual differences must be explored further and understanding this is likely to be the key to effectively utilising NIRS as a measurement tool.

It is recognised that this research was simulated in a laboratory environment, and although the findings of this study are useful and informative, the methodology must ultimately be replicated in the field. The literature review identified that in any future work researchers must consider: sex, age, ability, competition strategies and drafting when analysing the data, all of which were controlled in this study. The varying topography of triathlon courses in the field, and the varying workloads and intensities that arise as a result, are likely to give extra opportunities for the NIRS devices to be used to their full potential.

Previous work has utilised a wide variety of measurement tools to provide insight into the physiological responses of triathlon, but due to variations in methodologies and measures recorded difficulties arise when attempting to piece together the findings into a larger picture of overall performance. This thesis observed the relevant physiological measurements, including a new peripheral measure using NIRS, in a single piece of work that can be referred to more simply in the future. Whilst providing a new measurement of muscle oxygenation, NIRS was able to inform upon gaps in the current literature, and no difficulties are predicted in utilising this technology in the real world. Although the exact importance of this data is not fully understood in the current moment, this primary observation highlighted areas of inter-individual differences that would not have been determined in the NIRS was not used. It is concluded that NIRS can be used within triathlon and is able to effectively inform upon the relationship between oxygen delivery and consumption across multiple specific muscle groups in real time. As a physiological measurement tool NIRS has potential to be useful within the sport of triathlon and its implementation would increase the specificity of information available to athletes and coaches when creating strategies to be applied in training and competition.

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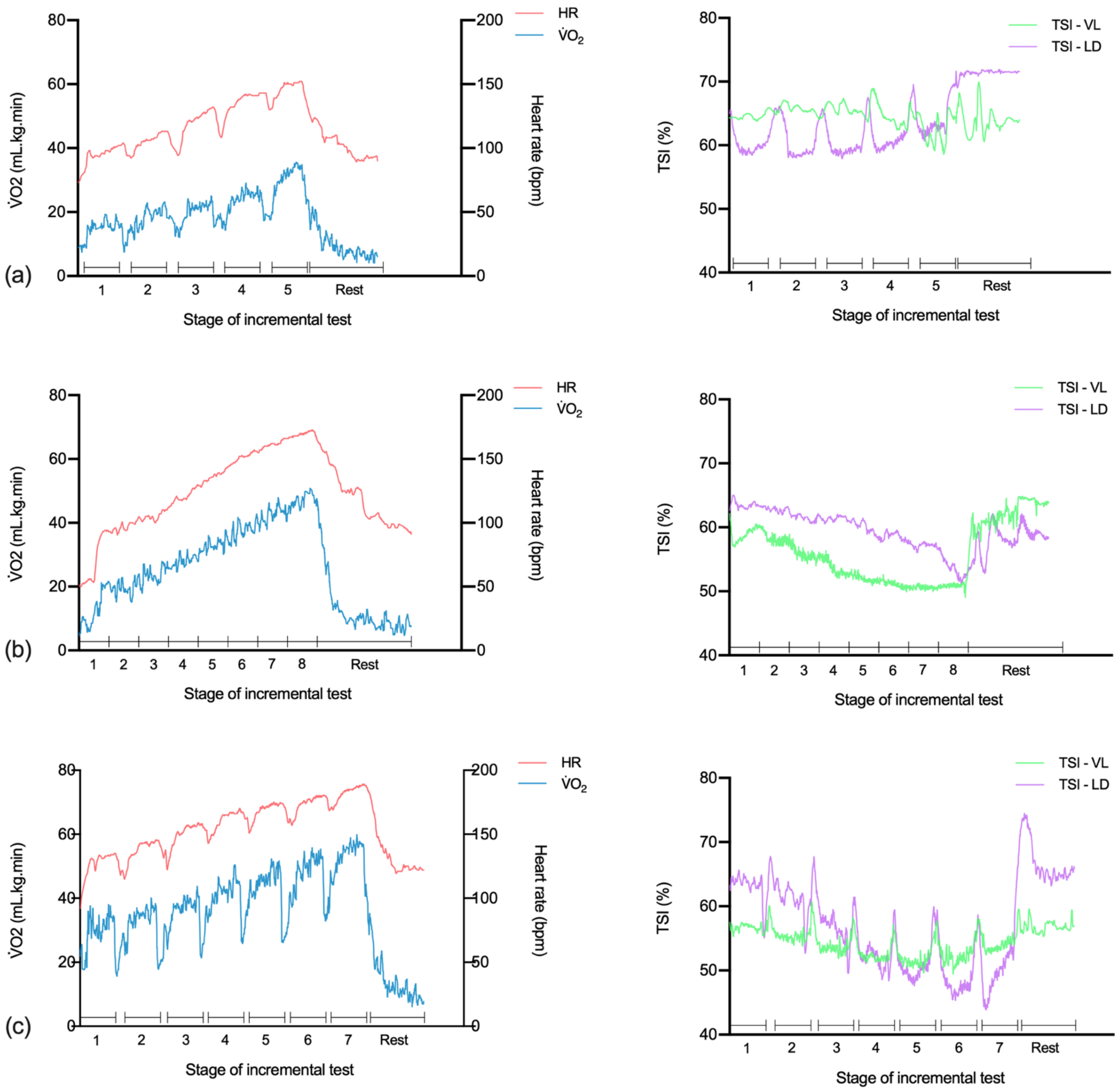
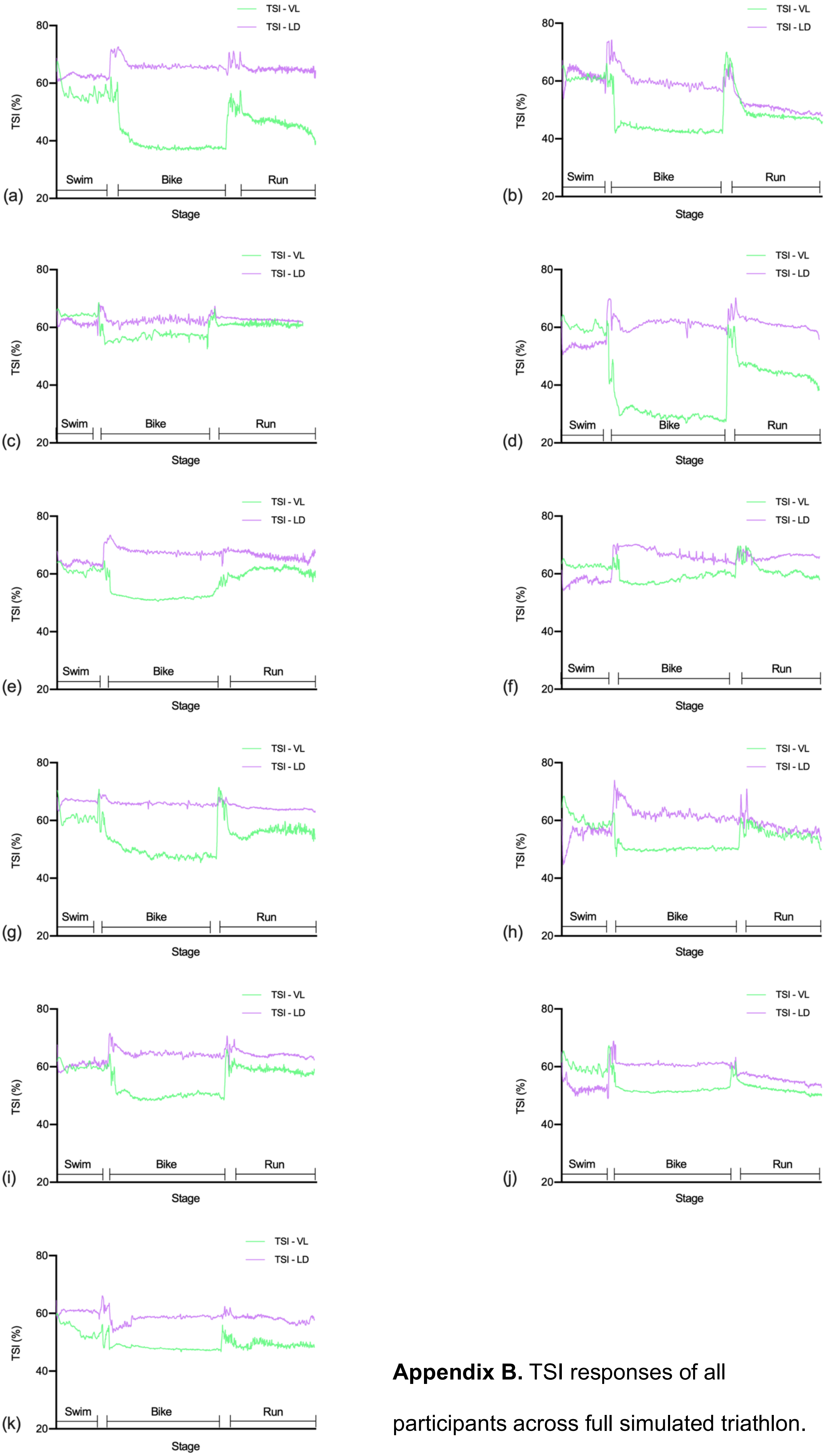
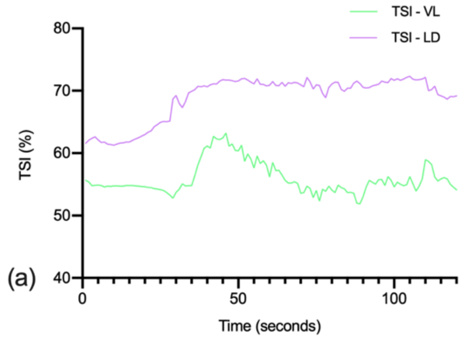
7. Appendices

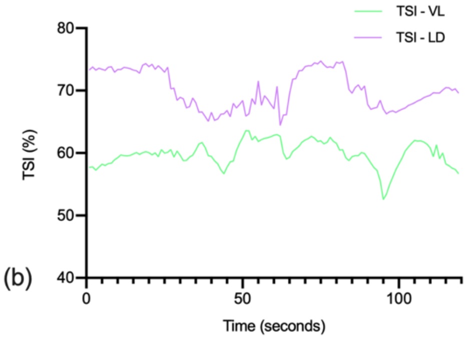


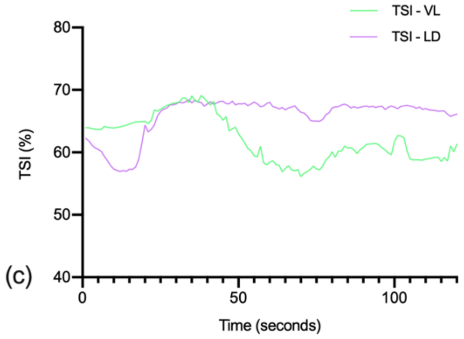
Appendix A. Representative participant V̇O₂, HR and TSI responses during swim ergometer (a), cycle ergometer (b), and treadmill (c) maximal incremental exercise tests.

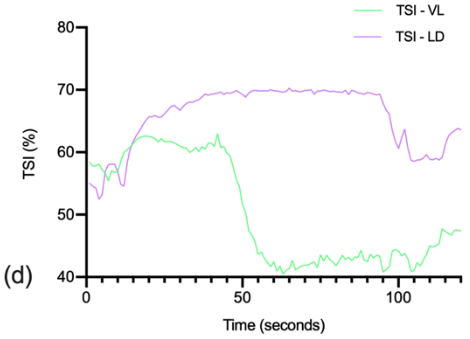


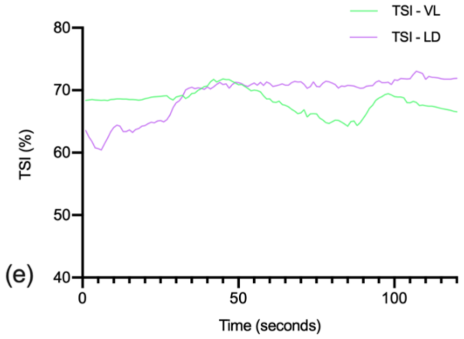
Appendix B. TSI responses of all participants across full simulated triathlon.

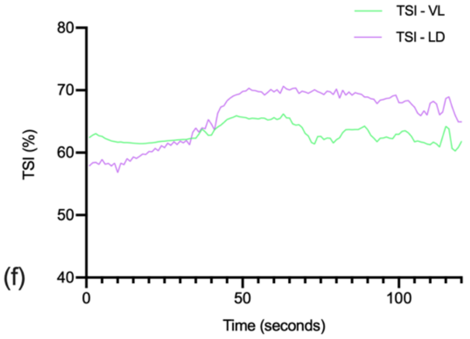


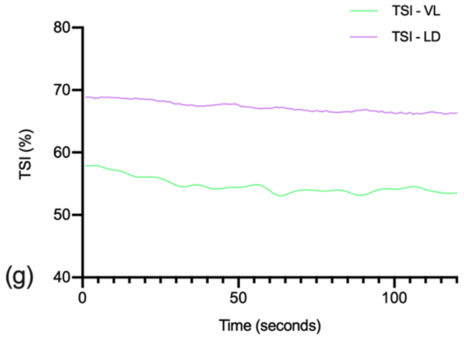


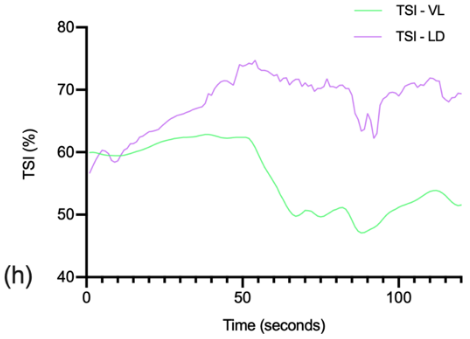


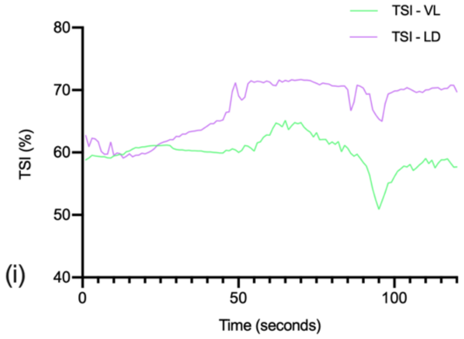


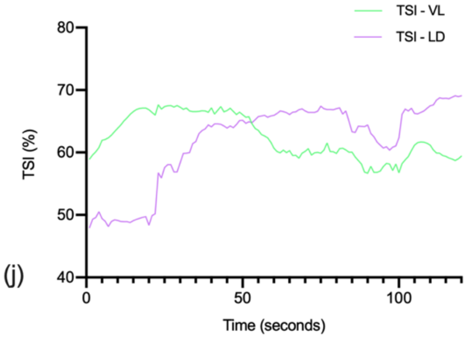


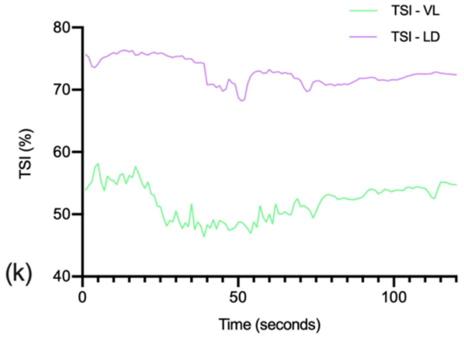




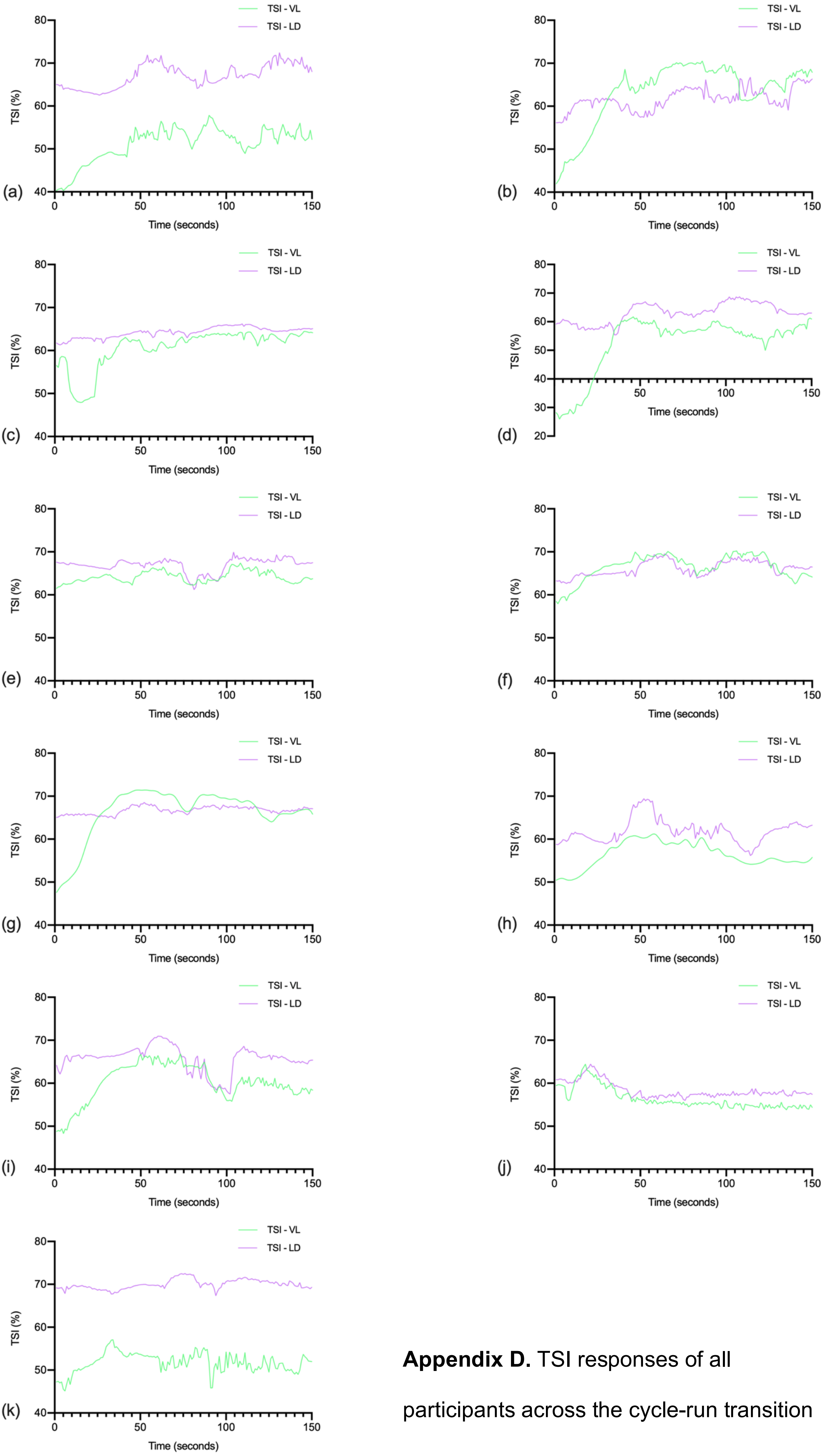




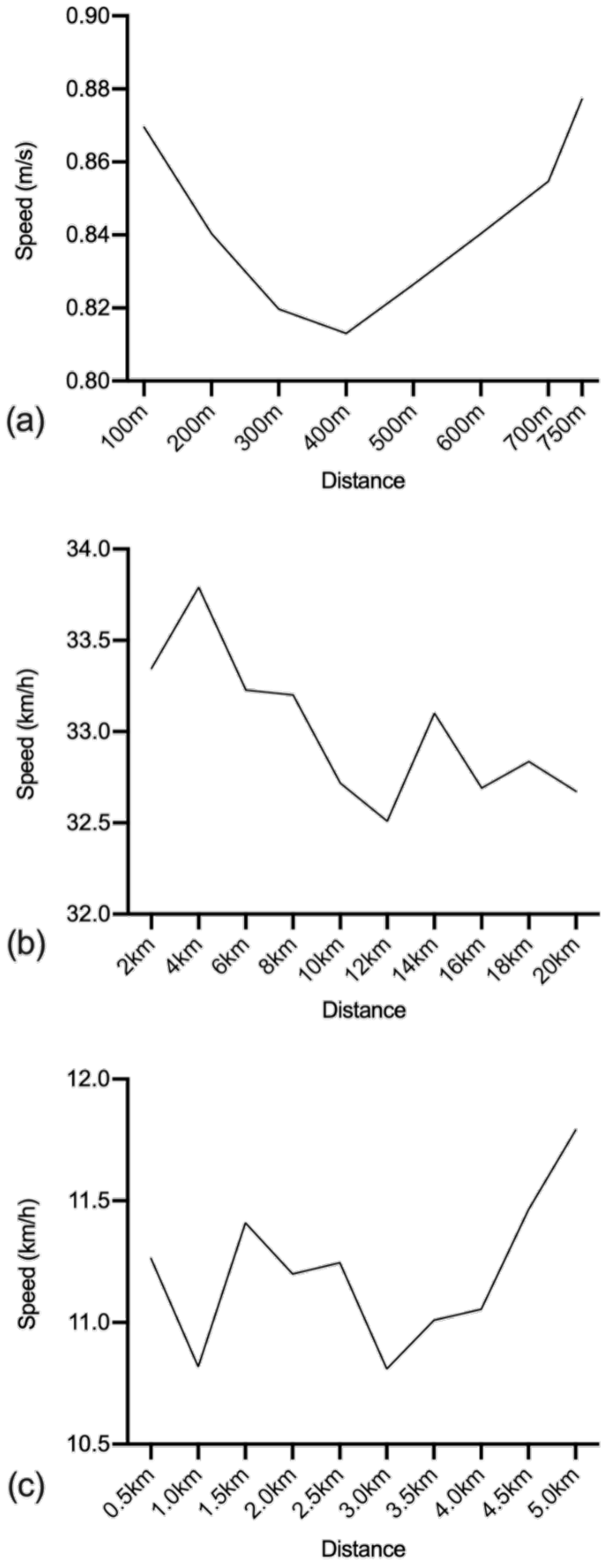




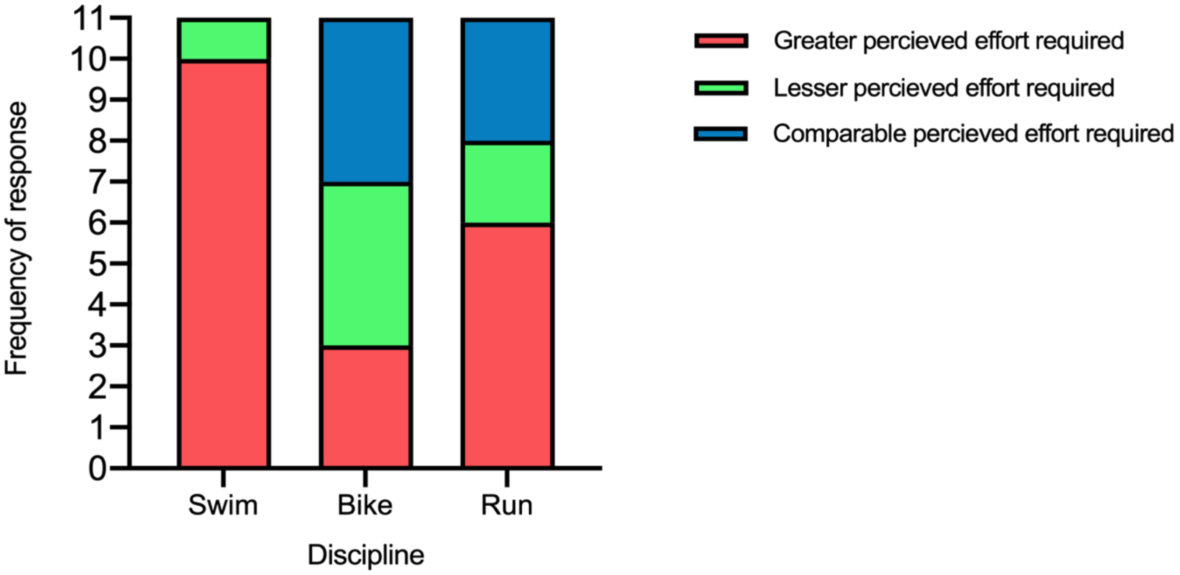
Appendix C. TSI responses of all participants across the swim-cycle transition



Appendix D. TSI responses of all participants across the cycle-run transition



Appendix E. Pacing profiles during swim (a), bike (b), and run (c) stages of simulated triathlon.



Appendix F. Perception of exertion required for laboratory apparatus in comparison to completing triathlon in the field.

The next section will ask for your opinions on the different pieces of equipment we used in the maximal exercise tests in comparison to the real world. Please circle if you believe the laboratory equipment was easier, harder or no difference in regards to workload at a given intensity.

The treadmill was **Easier** **Harder** **Not significantly different** than running outdoors

Why?..

The bike (max) was **Easier** **Harder** **Not significantly different** than cycling outdoors

Why?..

The swim bench was **Easier** **Harder** **Not significantly different** than swimming

Why?..

Rate your perceived performance in the maximal Swim, Bike, Run tests. *1=Best, 3=Worst*

1. ………………. 2. ………………. 3. ……………….

What are the reasons for rating the disciplines in this way?

Swim..

Bike..

Run...

The bike (tri) was **Easier** **Harder** **Not significantly different** than cycling outdoors

Why?..

Briefly explain how the simulated triathlon compared to a ‘real world’ sprint triathlon. Consider speed, exertion, breathing, motivation or any other factors effecting performance.

Swim..

....

Bike..

....

Run...

....

Overall...

Appendix G. Preferences Survey given to participants upon completion of simulated triathlon.