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Pulmonary responses to cold water endurance swimming

A quasi-experimental cross-sectional study on healthy, active adults

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1 Abstracts

1.1 English

Objectives: Long-distance, extreme triathlons and open water swimming (OWS) events have become popular leisure time activities over the last decades, attracting a broad range of participants. However, reports on high mortality rates and the incidence of life-threatening conditions like hypothermia, swimming-induced pulmonary edema (SIPE) and exercise-induced bronchoconstriction (EIB) have been reported in the OWS and triathlon communities. Facial-skin cooling and inhalation of cold air can induce acute bronchoconstriction, consequently making cold water swimmers susceptible of developing severely reduced lung function during their athletic performance. Our primary aim was thus to assess the pulmonary responses to an OWS in cold water. Secondly, we wanted correlate the potential pulmonary changes with body composition characteristics and weekly training volumes to examine if such relationships existed.

Methods: Nineteen (♀7/♂12) healthy individuals completed a 45-90 minutes OWS in cold water (10±0.9ºC), wearing a swim-specific wetsuit. Body composition was measured by dual-energy x-ray absorpiometry (DXA) at baseline. Lung function, determined by maximal expiratory flow-volume curves, whole-body plethysmography and alveolar gas diffusion measured by transfer factor for carbon monoxide ($T_{L,CO}$) were assessed before and 2.5-3 hours after the OWS. Concurrently, exhaled fractional nitric oxide (FE_{NO}) and oxygen saturation by pulse oximetry (SpO_2) were also assessed. In 12 participants, lung function was measured before and 3, 10, 20 and 45 minutes after the OWS. Results are given as median (25^{th} to 75^{th} percentile) and effect sizes as *r*. Level of statistical significance was set to $p \leq 0.05$.

Results: Four of 12 participants (33.3%) developed EIB, a ≥10% decline in forced expiratory volume in one second (FEV_1). FEV_1 was significantly reduced immediately after the OWS: -6.32% (-8.49, -5.41), ($p = 0.008$, $r = 0.568$). After 2.5-3 hours, significant declines were observed in $T_{L,CO}$: -5.47% (-8.36, -3.91), ($p < 0.001$, $r = 0.61$), SpO_2: -2.5% (-3.25, -2.00) ($p = 0.001$, $r = 0.56$) and FE_{NO}: -11.65% (-14.49, 1.44), ($p = 0.014$, $r = 0.40$). One male participant stood out, measuring severely reduced pulmonary function and indicating medical signs of SIPE. Three (16.6%) additional male participants were within the definition of mild hypoxemia (94-95%). No

significant correlations were found between body composition variables, weekly training volumes and changes in lung function.

Conclusion: EIB was observed in one third of the participants, while additional pulmonary impairments were found up to three hours after the OWS. Certain individuals, predominantly male participants, experienced severe pulmonary impairments during/after the OWS. Referring to the large effect sizes, these are all observations that could be of crucial practical importance for the swimming athlete. To increase the methodological specificity, we encourage future research to be performed in racelike settings as to increase the physiological stress levels experienced by the participants.

1.2 Norwegian

Bakgrunn: Oppslutningen rundt triatlonkonkurranser og open water-arrangementer har aldri vært høyere enn nå. Personer i alle aldre og med ulike fysiske forutsetninger ønsker å flytte personlige grenser ved å gjennomføre ekstreme konkurranser; konkurranser av både lang varighet og under ekstreme temperatur- og værforhold. De siste tiårene har fokuset likevel blitt rettet mot risikoene forbundet med langvarig svømming. Parallelt med høye dødelighetsrater i triatlon, er hypotermi, svømmeindusert pulmonalødem (SIPØ) samt anstrengelsesutløst bronikalkonstriksjon (exercise-induced bronchoconstriction; EIB) noen av de medisinske tilstandene rapportert i forbindelse aktivitet i kald luft og kaldt vann. Da inhalasjon av kald luft og nedkjøling av ansiktet vil kunne indusere bronkialkonstriksjon, vil trolig svømmende individer være særlig utsatt for redusert lungefunksjon under og/eller etter deres aktivitet dersom vannet er kaldt. Vårt hovedmål var derfor å undersøke lungefunksjon før og regelmessig etter langvarig svømming i åpent, kaldt vann (open water swim; OWS). Videre ønsket vi å undersøke korrelasjonene mellom maksimale endringer i lungefunksjon og ukentlige treningsvolumer samt antropometriske variabler.

Metode: 19 (♀7/♂12) friske personer gjennomførte 45-90 minutter svømming i kaldt vann (9-10°C) iført en svømmespesifikk, tettsittende våtdrakt. Antropometriske variabler ble innhentet gjennom dual-energy x-ray absorpiometry (DXA), mens lungefunksjon ble kartlagt gjennom maksimale, ekspiratoriske flow-volum-kurver, helkroppslig pletysmografi, alveolær gassdiffusjon for karbonmonoksid (*T*L,CO) og arteriell oksygenmetning ved fingermålt pulsoksimetri (SpO_2). Samtidig ble ekspirert

nitrogenmonoksid (FE_{NO}), et mål på eosinofil luftveisinflammasjon, også målt. Alle variabler ble innhentet før og 2.5-3 timer etter avsluttet OWS. På 12 av deltakerne ble flow-volum-kurver også innhentet før og 3, 10, 20 og 45 minutter etter avsluttet OWS. Ukentlige treningsvolumer ble rapportert gjennom et spørreskjema, mens aerob kapasitet ble definert som maksimalt oksygenopptak etter spesifikke kriterier under en tredemølletest. Resultatene og effektstørrelse er gitt som henholdsvis median (25.-75. persentil) og *r*. Statistisk signifikansnivå ble satt til $p \leq 0.05$.

Resultater: Fire av 12 deltakere (33.3%) utviklet anstrengelsesutløst bronkialkonstriksjon (EIB), definert som ≥10% reduksjon i forsert ekspiratorisk ett-sekundsvolum (FEV_1). Den største reduksjonen i FEV_1 ble målt 3 minutter etter avsluttet OWS: -6.32% (-8.49, -5.41), ($p = 0.008$, $r = 0.568$), og var fortsatt signifikant redusert etter 10 minutter. 2.5-3 timer etter avsluttet OWS ble det målt signifikant reduksjon i *T*L,CO: -5.47% (-8.36, -3.91), ($p < 0.001$, $r = 0.61$), SpO_2: -2.5% (-3.25, -2.00) ($p = 0.001$, $r = 0.56$) og FE_{NO}: -11.65% (-14.49, 1.44), ($p = 0.014$, $r = 0.40$). Én mannlig deltaker opplevde svært nedsatt lungefunksjon som følger av svømmingen, og viste indikasjoner på utvikling av SIPØ. Ytterligere tre mannlige deltakere (16.6%) målte oksygenmetning tilsvarende mild hypoksemi (SpO_2: 94-95%). Det ble ikke funnet noen signifikante korrelasjoner mellom kroppssammensetning, ukentlige treningsvolumer og målte endringer i lungefunksjon.

Konklusjon: Èn tredjedel av deltakerne utviklet EIB, og ytterligere signifikant reduksjoner ble funnet i *T*L,CO, FE_{NO} og SpO_2 opptil tre timer etter avsluttet OWS. Enkelte individer – hovedsakelig menn – opplevde svært reduserte lungefunksjonsvariabler under/etter svømmingen. Det ble derimot ikke funnet noen signifikante sammenhenger mellom de observerte endringene i lungefunksjon, kroppssammensetning og ukentlige treningsvolumer. Med bakgrunn i de høye effektstørrelsene kan de observerte endringene i lungefunksjon likevel være av avgjørende, praktisk betydning for den svømmende utøveren. For å heve den metodologiske spesifisiteten tilnærmet en reel konkurransesituasjon, oppfordrer vi til at videre studier gjennomføres i konkurranselignende situasjoner for å øke det fysiologiske stresset deltakerne utsettes for.

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3 Abbreviations

Table 3.1 The commonest abbreviations used in the present thesis, including applied units of measurement.

OWS	Open water swim
SIPE	Swimming-induced pulmonary edema
IRV	Inspiratory reserve volume (L)
ERV	Expiratory reserve volume (L)
TLC	Total lung capacity (L)
V	Alveolar ventilation ($L \cdot min^{-1}$)
Q	Pulmonary blood flow ($L \cdot min^{-1}$)
$\mathbf{VO_{2max}}$	Maximal oxygen uptake measured during exercise test ($mL \cdot kg^{-1} \cdot min^{-1}$)
%BF	Percentage body fat (%)
LBM	Lean body mass (kg)
FM	Fat mass (kg)
$\mathbf{FEV_1}$	Forced expiratory volume in one second (L)
FVC	Forced vital capacity (L)
EIB	Exercise-induced bronchoconstriction
BHR	Bronchial hyper-responsiveness
$\mathbf{FE_{NO}}$	Fractional orally exhaled nitric oxide (ppb)
$\boldsymbol{V_T}$	Tidal volume ($L \cdot min^{-1}$)
$\boldsymbol{T}$**L,CO**	Transfer factor for carbon monoxide ($mmol \cdot min^{-1} \cdot kPa^{-1}$)
$\boldsymbol{K}$**CO**	Diffusion constant for carbon monoxide ($mmol \cdot min^{-1} \cdot kPa^{-1} \cdot L^{-1}$)
VA	Alveolar volume (L)
$\mathbf{D_M}$	Diffusing properties of the alveolo-capillary membrane
$\boldsymbol{V}$**c**	Pulmonary capillary blood volume
θ	Carbon monoxide-hemoglobin chemical reaction rate
$\mathbf{SpO_2}$	Arterial oxygen saturation, measured by pulse oximetry (%)
DXA	Dual-energy x-ray absorptiometry
EIAH	Exercise-induced hypoxemia

4 Introduction

Over the last decades, extreme aquatic events, challenges and races have become popular leisure time activities among people of all ages, thus inducing a rapid growth in the number of extreme endurance events(6). Always on the lookout for new experiences and challenges to push one's limits, organizers continue to increase the extremeness of their events. This is done either by prolonging the performance duration and/or distance to be covered, creating rougher courses and/or arranging their events in areas with tough climates, varying temperatures and fast weather changes(6). Extreme triathlons, like Norseman and Celtman XTri, and open water swimming (OWS) events, like English Channel crossings, are only few of the many extreme aquatic events where the combination of long performance duration (1-19 hours), rapid weather changes and cold air and water are present(6-8). Not only are the physical and psychological requirements high, but nutritional needs, logistics, racing crew/assistants and equipment must also be coordinated in terms of a successful completion(7).

One of the many challenging aspects in the aforementioned events are the low water temperatures, exposing the participants to an increased risk of hypothermia and cardiac and cardiovascular abnormalities with death as the most severe consequence(9-11). Strict rules and regulations have therefore been provided by international sport unions as to maintain the athlete's safety during these events(12, 13). As an example, the International Triathlon Union (ITU) states that the use of wetsuit is mandatory in water temperatures below 16°C when the swimming distance is >1501 meters, but is to be cancelled if the temperature falls below 13°C(12). However, as many race organizers are ITU independent, and therefore not obliged to follow the cited regulations, the swim leg of an extreme triathlon may still be arranged in water temperatures below the minimum temperature limits dictated by ITU.

Harris, Henry, Rohman et al. (10) reported a death rate of 1.5 per 100 000 participants in triathlons; almost twice the numbers as in marathon running (0.8 per 100 000 participants). The primary cause of death was concluded to be drowning due to cardiac abnormalities, and the authors advice all race organizers and participants to perform a medical and cardiac screening prior to racing(10). Although the water temperature is an insignificant factor in this specific research letter, the physiological responses induced when swimming in cold water have been suggested to trigger potentially latent cardiac abnormalities in susceptible subjects(11). Facial skin-cooling, which is a natural

consequence of freestyle swimming in cold water, has also been found to cause acutely reduced lung function in healthy adults(14), and may also reduce the level of safety when swimming in cold water.

Moreover, an increasing number of case reports on the occurrence of swimming-induced pulmonary edema (SIPE) shed light on an additional risk connected to OWS(15-20). This medical condition, together with immersion-induced hypothermia, will thus be presented in two of the following paragraphs. Firstly, however, the basic anatomy of the respiratory tract and lungs will be presented together with the essential principles of pulmonary physiology. This will establish the theoretical basis for the further presentation of the physiological and pulmonary responses to swimming (in cold water) and the aforementioned risks connected to this extreme mode of exercise.

5 Lung anatomy and lung physiology

The lungs are intricate structures, anatomically divided into two subdivisions: the respiratory tract/airways (consisting of the nose cavity, pharynx, larynx, trachea, bronchi and bronchioles) and the gas-exchange tissue (the alveoli)(3, 21). Although clearly divided in anatomical literature, the subdivisions form a complex whole in performing one of the most crucial functions in living humans – respiration(3, 21). When referring to the lungs as a working system, one may use the terms ‘respiratory’ or ‘pulmonary’ system. To avoid any confusion with cellular respiration, the term ‘pulmonary’ will be used. This limits the extent of the current thesis to that of the working lungs and how they, as organs, are affected by cold water swimming. The description of anatomical structures, however, will be used according to discipline tradition (e.g. respiratory tract).

On its way down the respiratory tract towards the alveoli, the inhaled air is heated to near body temperature and fully humidified(21). It is also purified by cilia and mucus as to avoid contact between potentially harmful particles from ambient air and the airway epithelium(3, 21). Phagocytic cells are also located in the alveolar wall, hopefully to inactivate the remaining, potentially harmful, particles and substances(3). From trachea and down to the bronchi, the respiratory tract consists mainly of cartilage, connective tissue and a small amount of smooth muscle cells(21). However, as the bronchi get smaller further down the respiratory tree, smooth muscle cells gradually become the dominant material of the airway walls(21). Consequently, the lumen of the bronchioles

can be accurately regulated by the sympathetic and parasympathetic nervous systems, causing muscle relaxation and constriction, respectively(21).

A single layer of epithelial cells cover the inner walls of trachea, the bronchi, bronchioles and the alveoli(21). In addition to phagocytic cells, the alveolar walls also consist of two other types of cells: type I and type II cells, the latter lowering the alveolar surface tension by producing surfactant(3). This lipoprotein complex also keeps the lungs from collapsing after a full expiration, as well as maintaining a dry environment inside the alveoli(3). Together, the alveolar cells make up a permeable membrane through which oxygen molecules (O_2) can diffuse freely – driven by the differences in partial pressure in the airways, alveoli and pulmonary capillaries(3, 21). In reference to carbon dioxide (CO_2), the steps are carried out in the opposite direction, also driven by the differences in partial pressure(3).

The active process of inhalation, conducted by the respiratory muscles (e.g. diaphragm and mm. levatores costarum), causes the rib cage to expand and the elastic fibres of the lungs to stretch. This creates a negative intrapulmonary pressure (atmospheric pressure > intrapulmonary pressure), filling the lungs with ambient air. However, as the respiratory muscles relax, the rib cage and elastic fibres rapidly recoil and the intrapulmonary pressure increases, reversing the airflow towards the mouth(3, 22).

During resting conditions, the volume of air moving through the lungs during a respiratory cycle is termed the tidal volume (V_T), and measures ± 0.5 liter(22). The inspiratory and expiratory reserve volumes (IRV and ERV, respectively) represent the volume of air which can be inhaled and exhaled exceeding that of the V_T (3). However, even after a full expiration a small amount of gas still remains in the lungs; the residual volume, measuring approximately 1.2 litres. The total sum of these volumes make up the total lung capacity (TLC). The size of TLC, and it's subdivisions, are primarily determined by age, height, sex and ethnicity in healthy adults, and usually measures 5.5-6 liters(3, 22).

To maintain adequate O_2 delivery to vital organs and working muscles, it is essential that ambient air reaches the alveoli and that the alveolar membrane is permeable, as to enable gas diffusion. However, proper ventilation (V) and alveolar gas diffusion also depend on blood flow through pulmonary capillaries (perfusion, Q) which is strongly affected by the gravitation, making the ratio between the two (V/Q ratio) unequal across

the lungs(3, 22). The V/Q ratio is low at the base of the lung (high Q) and high at the apex (low Q, high V), but when pulmonary capillaries merge, the blood of various O_2 content is mixed and normal oxygen saturation can be measured during rest(22).

Even during strenuous exercise, the lungs are rarely considered a limiting factor, although some highly trained individuals have been reported to systematically measure reduced oxygen saturation during maximal whole-body exercise(23). The latter observation could be explained by a large, O_2 consuming muscle mass and a large cardiac output, reducing pulmonary transit time and consequently causing a V/Q mismatch(23).

6 Swimming physiology and cold water swimming

Compared to vertically oriented exercise, such as running or cycling, swimming (freestyle) is affected by the gravitational differently than most other recreational activities(24). When adding the resistive forces created by the water, swimming is regarded as a whole-body exercise with a high energy cost and high pulmonary requirements(3, 24). Both at submaximal and maximal work intensity the swimmer's breathing pattern is restricted by the rhythm of movement; a restriction which is found to affect the pulmonary gas exchange significantly(25).

In a horizontal position, the altered gravitational forces increase the diastolic filling of the heart, and a decline in heart rate and cardiac output can be observed(24, 25). The positional change also causes a redistribution of blood from the extremities towards the pulmonary circulation, equalizing the regional perfusion differences of the lung(3). This equalization, increasing Q could thus be regarded as a counterbalancing response, maintaining an adequate pulmonary oxygen uptake and oxygen hemoglobin saturation, despite of the reduced heart rate – even during maximal exercise(3, 24, 25).

During full-body immersion, the swimmer experiences an increased external pressure on the thoracic cage and abdominal wall, consequently reducing IRV and TLC(3). Also affecting V_T, immersion increases the work of breathing and airway resistance substantially(3). This can be observed through declines in variables such as forced vital capacity (FVC) and forced expiratory volume in one second (FEV_1) during immersion(3, 26). Naturally, the variables renormalize as the healthy, normothermal subject exits the water.

An additional thermoregulatory vasoconstriction is induced when the water is cold and the skin is cooled(3, 11, 27). Cold sensitive thermoreceptors, located in the skin, stimulate the thermoregulatory centre of anterior hypothalamus, causing vasoconstriction in the arms, legs and skin to prevent heat loss(27). The peripheral vasoconstriction furthermore increases the arterial blood pressure, thus adding to the already elevated blood pressure occurring when swimming in a prone position(24, 25, 27). As a counterbalancing measure, a cold-induced diuresis follows(11)

6.1 Cold water swimming and the pulmonary system

Although swimming is a physically and hemodynamically challenging mode of exercise compared to vertical exercise, healthy humans are still capable of maintaining adequate ventilation, gas exchange and muscular oxygen uptake to sustain high exercise intensity over a long period of time in normal water temperatures(3). However, the general consensus regarding the physiological and pulmonary responses to cold water swimming and immersion in healthy adults seems to be universal, exemplified by a citation of Tipton and Bradford (11): "there is little evidence to indicate that the alterations associated with cold water immersion impair lung function to an extent that it interferes with oxygen uptake during exercise" (p4).

Although there is an apparent distinction between cold water immersion and breathing cold air, the lack of scientific research on pulmonary parameters after cold water swimming makes the comparison unavoidable. When swimming outside in cold, open water in Norway, the ambient temperature is likely to be below 15°C nevertheless, making the comparison not too farfetched to draw parallels between the two stimuli.

With the reference to this point, Cotes, Chinn and Miller (3) present the following statement:

> *"In healthy persons who are not atopic or exposed to polluted air, the airways resistance and forced expiratory volume are unaffected by breathing cold air through the nose, taking exercise that entails mouth breathing during exercising at subzero temperature or undergoing a cold water challenge test" (p501).*

This statement is also supported by the American College of Sports Medicine(28), stating that healthy exercising athletes rarely experience impaired lung function when breathing cold air.

Moreover, atopic(3, 29) and asthmatic(30) individuals have reported a susceptibility in experiancing bronchoconstriction and reduced lung function during exercise (exercise-induced asthma, EIA), especially when ventilating cold air simultaneously. Supposedly, approximately 50% of all atopic individuals experience some degree of bronchoconstriction when they are stimulated by cold, hence increasing airway resistance and the work of breathing(3). When studying the pulmonary responses to cold water swimming, one may presume that atopic, asthmatic and BHR affected individuals could be more susceptible to pulmonary changes compared to healthy individuals.

Studies examining the effect of skin-cooling have implied that a facial-cooling reflex induces bronchoconstriction in individuals with pulmonary disorders, such as asthma and chronic obstructive disease, but also in healthy subjects(14, 30, 31). Gradual facial-cooling is a natural consequence when swimming in a prone position in cold water(11, 28), hence activating the afferent and efferent arches of n. Trigeminus and n. Vagus, respectively(14, 30, 31). This reflexive response causes bronchoconstriction, impairing the subject's lung function - regardless if an asthmatic/atopic diagnosis is present or not(14).

In the following paragraphs, additional aspects of the risks connected to cold water swimming are presented. The two medical conditions in which pulmonary impairments are most likely to be found, hypothermia and SIPE, will be prioritized. When exposing a study sample to an OWS in cold water, one should be aware of these potential risks and furthermore to interpret the results bearing these conditions in mind.

6.2 Physiological risks connected to cold water swimming

Despite the long history of cold water and hypothermia research, no clear definition of *cold water* has been made(11). Temperatures as high as 18°C have been labelled *cold* in previous studies, the normal range used in scientific contexts is normally 10-15°C(11). In certain extreme endurance races, like the ones mentioned above, water temperatures can be as low as 9-10°C, and are thus within this definition(7). Military personnel, especially navy sailors and Special Forces, are regularly faced with open water challenges – both during exercise and real-life operations where the water temperatures

are uncontrollable(15, 32). When military operations are executed across the globe, it is likely that the water temperatures can be below 15°C – especially in European and North American seas(33).

International and national sports federations and unions have created clear sets of regulations in terms of protecting the sporting individual from cold-induced hazards during events and races(12, 13). As an example, ITU states that the use of wetsuit is mandatory when the water temperature is ≤15.9°C, and optional in temperatures between 15.9-20.0°C(12). In OWS events organized by FINA, wetsuits are always prohibited, but the swimming distance and/or maximum time allowed in the water should be systematically regulated according to the water temperature(13). National and local races, on the other hand, have more liberal sets of regulations where organizers are – to a certain extent – free to determine whether the use of wetsuit is mandatory, optional or prohibited when the water temperature ranges from 13-16.5°C(34).

6.2.1 Hypothermia

Hypothermia is likely to be the most profound risk connected to cold water activities(9, 11). Defined by a core temperature ≤ 35°C, mild hypothermia is followed by reflexive responses like shivering, amnesia, apathy and loss of muscular coordination(28). As the core temperature is further reduced, distinct signs such as unconsciousness, severe cardiac abnormalities (e.g. ventricular fibrillation, bradycardia and asystole) become apparent(28, 35). The outcome could hence be fatal(10, 11, 28).

Regarding the pulmonary responses to mild hypothermia, shivering causes an increase of muscular oxygen uptake, respiratory rate (breaths per minute) and V_T, increasing minute ventilation(35). At even lower core temperatures (≤32-34°C), both shivering and hyperventilation ceases, causing respiratory failure (either hypoxemia alone (type I) or with the concurrent retention of carbon dioxide and lowering blood pH (type II))(3, 35). As a compensatory response to maintain adequate alveolar gas exchange, a bronchodilation is induced by the inhibition of n. Vagus(35). However, as the mechanical properties of the pulmonary system are found to remain unaffected by hypothermia until ≤29°C, Datta and Tipton (35) suggest that “some aspects of respiratory control is abnormal during hypothermia”(p2062) and should thus be ascribed hypothermia-induced neural changes. If not removed from the cold water, the core temperature will continue to fall rapidly(11). In fact, studies have also shown that

the core temperature can continue to fall – even after removal from the water(3, 11). This would make triathletes vulnerable to the unfortunate responses induced by hypothermia, as they continue racing even after the swim leg is completed.

The use of a tight-fitted wetsuit is found to protect the immersed subject from developing hypothermia(28). This is one of the main reasons why ITU and FINA have developed clear regulations regarding the use of wetsuit during races and OWS events(9, 13, 34). To insulate the swimming body, it is essential that the wetsuit is tight-fitted as to 1) reduce the amount of circulating cold water and 2) to trap a small amount of water inside the suit(11, 28). The volume of water is then heated by the metabolic heat produced during exercise(3, 22). This may delay a decline in core and skin temperature, consequently delay the onset of hypothermia-related responses(28, 35).

According to Sawka, Castellani, Cheuvront et al. (28), the susceptibility for developing hypothermia is closely related to body composition. Not only is a high body fat percentage (%BF) advantageous in regards to swimming buoyancy, but also as insulation – protecting the swimmer against subnormal core temperatures during long-lasting endurance events in water(3, 11, 27). Consequently, one often find open water swimmers gaining excessive amounts of body fat as they prepare for racing season as to delay premature fatigue occurring when the core temperature is lowered(9, 28).

Aerobic fitness is not considered an important factor in maintaining a normal core temperature under extreme temperature conditions(28). A large lean body mass (LBM) is found to be beneficial due to the high metabolic heat production of working muscles(27, 28). Compared to untrained individuals, endurance-trained athletes with a large LBM and good swimming skills would be able to sustain high exercise intensity for a longer period of time, producing a significant amount of metabolic heat and hopefully delay a potential decline in core temperature(28). Therefore, well-trained athletes with a good work economy and aerobic fitness could – hypothetically – be protected by their own ability to produce metabolic heat and sustain a high exercise intensity(28).

Considering the importance of body composition in maintaining a normal core temperature, one could question if a combination of anthropometrical characteristics (e.g. low %BF and low LBM) would make certain individuals more susceptible to pulmonary changes following an OWS in cold water. On the other hand, when

acknowledging the thermal protection offered by a tight-fitted wetsuit, one could also question if the wetsuit would neutralize a potential effect of different body composition characteristics.

6.2.2 Swimming-induced pulmonary edema (SIPE)

In the beginning of the 21st century, medical communities worldwide began publishing case reports describing the occurrence of SIPE; a medical condition which was previously unnoted in healthy humans swimming at sea level(16, 18). From that time, SIPE has been reported among otherwise healthy military trainees(15, 16, 32), divers(3) and triathletes(17-20, 36), always associated with strenuous swimming in a range of different water temperatures.

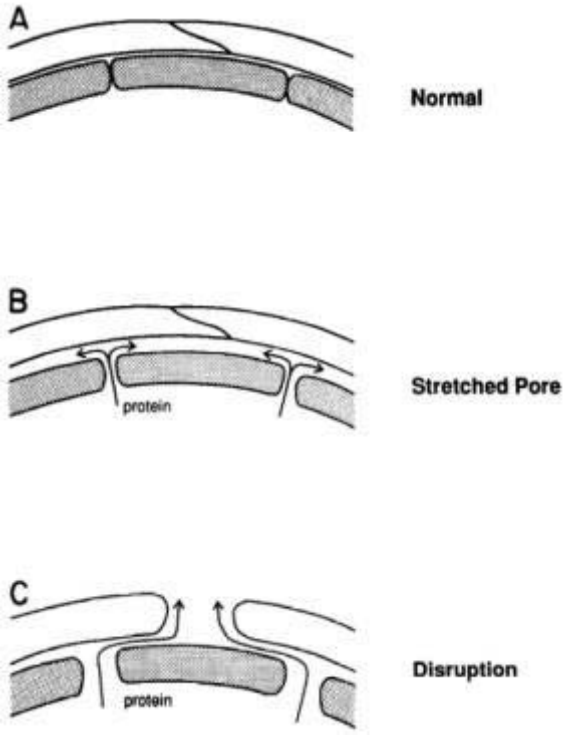

Figure 6.1 According to West, Tsukimoto, Mathieu-Costello et al. (1) pulmonary edema is caused by a rise in transmural capillary pressure, causing a stretch (1.A) in endothelial layer and a complete disruption can be observed as the pressure increases further (1.C). Consequently, this increases protein permeability and gas diffusion is impaired (5).The figure is located in West, Tsukimoto, Mathieu-Costello et al. (1), and reprinted with permission ("not required" for master's thesis') by the Copy Clearance Center.

In short, pulmonary edema is the accumulation of fluids in pulmonary interstitial areas and/or alveoli, caused by increased permeability of the alveolo-capillary membrane(3). West, Tsukimoto, Mathieu-Costello et al. (1) describe how even a moderate increase of transmural pressure in the pulmonary capillaries could cause a stress-induced failure of the blood-gas barrier. As illustrated in Figure 6.1.A, one first discovers a stretch in the endothelial layer of pulmonary capillaries, causing protein leakage into the capillary wall interstitium (6.1.B). A further increase of the transmural pressure results in a total disruption of the capillary endothelium, and proteins and fluids can be found within the alveolar space due to increased membrane permeability (6.1.C)(1, 3, 16, 22).

Couch, shortness of breath, crackles and bloody sputum are normal findings when examining liable subjects(16, 19, 36), and when pulmonary function tests are performed, reduced alveolar gas diffusion(5), oxygen saturation(16, 20, 32, 36) and lung function(16, 32) are usually observed. Premature, involuntary termination of exercise would thus be a natural outcome(17, 20).

In previous studies, the applied assessment methods and diagnostic criterions for SIPE have been inconsistent, ranging from the completion of an online survey(18) to self-reported shortness of breath and coughing(16) and radiographic imaging(19, 20, 36). The latter method is, however, considered the most valid method when assessing pulmonary edema, together with a thorough physical examination(5, 16)

Despite a clear physiological understanding of capillary stress failure and altered membrane permeability, one still questions why some, otherwise healthy adults develop this life-threatening condition, while others remain unaffected. As pulmonary edema can be observed in hypothermal individuals(28), some have suggested that the cold-induced peripheral vasoconstriction and central blood pooling could be precursors to the development of SIPE(16, 18, 32). As previously mentioned, swimming in a prone position alters the gravitational forces, also pooling blood centrally increasing both the mean arterial blood pressure and the capillary transmural pressure(1, 25).

Pre-existing hypertension and the consumption of omega-3 fatty acid supplements(18), overhydration prior to swimming(32) and the use of a tight-fitted wetsuit(18, 19) have also been suggested as potentially triggering factors. However, these are only hypotheses – waiting to be examined in well-controlled studies.

7 Expected responses in assessed variables

Altered lung function, alveolar gas diffusion and eosinophilic inflammatory markers have been reported after strenuous exercise(5, 37), water immersion(15, 16, 38) and long-lasting endurance events(18). However, there is a limited number of studies examining the pulmonary responses to a long-distance OWS in cold water. Aiming to give an acceptable account of the expected pulmonary responses to an OWS in cold water, one is therefore obliged to draw parallels to studies examining pulmonary responses to sedentary immersion, warm water swimming, cold air ventilation and in a variety of study samples.

7.1 Spirometry and flow-volume curves

Flow-volume curves, obtained by forced expiratory manoeuvres, are of essential importance when assessing dynamic lung volumes such as FVC and FEV_1. FVC is the total amount of air forcibly exhaled after a full inhalation and offers a good indication on airflow limitation and restriction(3). FEV_1, on the other hand, is the amount of air forcibly exhaled during the first second of exhalation(3). Subnormal levels of FEV_1 would indicate airflow obstruction, and could furthermore be expressed as percentage of FVC $\left(\frac{FEV1}{FVC}\ x\ 100\right)$. Unlike FEV_1 alone, this standardized ratio is nearly independent of lung and body size and can be used when interpreting the results from a broad range of respiratory patients(3, 26). To avoid misinterpretations, FEV_1/FVC should not be interpreted alone, but together with each variables individually (as FEV_1/FVC will remain within a normal range when both FEV_1 and FVC are reduced). By using standardized guidelines(26), these three variables are highly recommended in monitoring both acute and chronic changes in lung function(26).

Airway obstruction is usually observed in individuals experiencing bronchoconstriction: The narrowing of both small and large airways caused by smooth muscle contractions(39). Bronchial hyper-responsive and asthmatic individuals are found to be particularly susceptible for developing episodic bronchoconstriction after the exposure of polluted and/or cold air(3), but also after strenuous exercise(3, 37, 39). There are two established theories on this topic, explaining the physiological mechanisms behind this reflex response: The natural increase of ventilation when exercise is initiated, either cools and/or dehydrates the bronchial epithelium(3, 39). When cooled, bronchial venules constrict to conserve heat(40). As exercise, ventilation and cooling cease, this vasoconstriction is counterbalanced by a vasodilatation, inducing mucosal edema and reflex parasympathetic nerve stimulation – causing bronchoconstriction and increased airway resistance(39, 40). During bronchial "drying", on the other hand, extracellular movement of water causes intracellular ion concentration to increase, releasing inflammatory and bronchoconstricting mediators like eicosanoids derived from leukotrienes(40).

A high prevalence of exercise-induced bronchoconstriction (EIB) and bronchial hyper-responsiveness (BHR) have been observed among athletes reporting large training volumes in cold and/or polluted air(39, 41, 42). This phenomenon has been termed

"athlete's asthma" (p797)(39) due to the similarity of the responses oftentimes observed in asthmatic and atopic individuals during exercise-induced asthma. In addition to bronchoconstriction, elevated levels of inflammatory markers and epithelial shedding can be found(39, 41).

Reciting the established beliefs presented in the introduction, healthy, normothermal individuals are not expected to undergo any pulmonary changes during/after an OWS in cold water(3, 11). Nevertheless, Koskela and Tukiainen (14) observed that facial cooling, without inhaling cold air, could induce bronchoconstriction, hence reducing FEV_1 and FVC. This response has later been termed *the facial cooling-induced bronchoconstriction reflex*(30), and is suggested to be induced by the facial-innervating, pain-temperature sensitive trigeminal nerve (n. trigeminus). The succeeding stimulation from the parasympathetic, efferent arch of the vagus nerve (n. vagus) would cause a bronchoconstriction(14). Besides, both healthy and asthmatic individuals seem to be equally affected by this neural reflex response, making even healthy individuals susceptible to pulmonary impairments during exercise in a cold environment(14, 30). Bearing the observations by Koskela and Tukiainen (14) in mind, individuals swimming face-down (e.g. during freestyle swimming) could be liable subjects in experiencing facial cooling-induced bronchoconstriction.

7.2 Total lung capacity (TLC)

TLC is regarded as one of the most important lung function parameters in clinical practice, and is the sum of several lung subdivisions illustrated in Figure 7.1(43). Sitting in an airtight box, also termed a body plethysmograph, the patient's intrathoracic gas content is analysed during standardized breathing sequences, estimating the individual's static lung volumes(3, 43).

Changes in TLC (or any of the lung subdivisions) are usually explained by altered lung distensibility; the lung's ability to expand(3). Lung distensibility is again determined by 1) the compliance and 2) elastic recoil of the lung tissue – two inversely related factors(3). The elastic recoil is generated by the active process of contracting respiratory muscles as to create a rebounding, elastic effect of the lung tissue(3). The compliance, on the other hand, consists of two submeasurements: static (compliance without gas flow, e.g. during breath hold) and dynamic compliance (compliance during gas flow)(3). In other words, increased airway resistance and increased resistance of

movement of the chest wall affects the dynamic lung compliance(3). This would be the case during immersion, as the external pressure against the chest and abdominal increases from the water(3, 35). This would reduce pulmonary parameters such as V_T, IRV, FVC and FEV_1. Moreover, Datta and Tipton (35) claim that ERV decreases with as much as 66% (p2058). As a result, the work of breathing increases significantly, but ceases immediately when the subject exits the water.

Despite the abundant amount of literature describing static lung volumes, there are few scientific reports on acute changes in TLC induced by exercise, inhalation of cold air or cold water immersion in healthy adults. As previously mentioned in 6.2.1 Hypothermia, V_T decreases gradually as the core temperature declines, and the regulation of breathing is oftentimes described as "abnormal"(p2062) in hypothermal subjects(35). Although this affects lung function considerably, these impairments are assumed to be caused by neural alterations – not changes in lung distensibility or the mechanical properties of the lung(35).

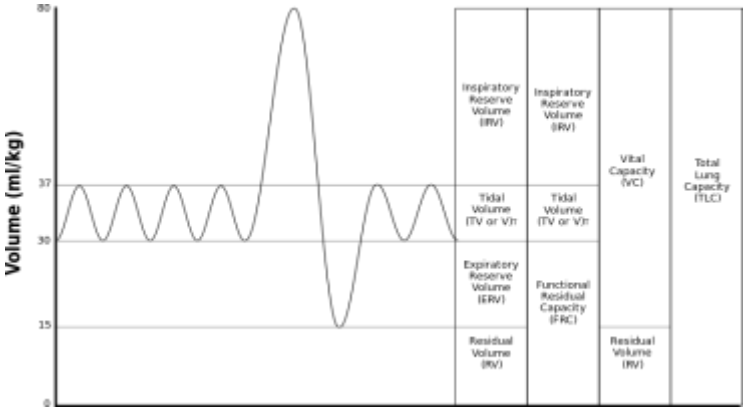

Figure 7.1 Total lung capacity (TLC) is the sum of inspiratory reserve volume (IRV), tidal volume (V_T), expiratory reserve volume (ERV) and the residual volume (RV), making the variable sensitive to changes in any of the subdivisions (3). Reprinting was permissioned according to the GNU Free Documentation Licence and Creative Commons Attributions-ShareAlike(4)

It is also debated whether or not the elastic recoil and/or lung compliance could be affected by subnormal core temperatures in living humans. Previous studies have been executed on animals(44) or anesthetized men(45), and are both of significant age, affecting the validity and reliability of the measuring devices and applied testing protocols.

In a state of pulmonary edema, the increase of interstitial fluids reduces lung compliance which restricts expansion of the lungs(3). Accordingly, this reduces TLC. Although the physiological explanation still remains unclear, some studies have also suggested that an asthma attack could increase TLC by an increase of lung

compliance(3). On the other hand, different baseline values of TLC in asthmatic and healthy individuals indicates that chronic inflammation could increase airway stiffness in asthmatic individuals, hence increasing elastic recoil and reducing TLC(3).

However; unless hypothermia or SIPE develop, there is little evidence to indicate that asthmatic, BHR affected or healthy individuals should experience any responses in TLC during and/or after an OWS in cold water.

7.3 Alveolar gas diffusion and arterial oxygen saturation (SpO_2)

The transfer factor for carbon monoxide ($T_{L,CO}$), also termed diffusing capacity for carbon monoxide ($D_{L,CO}$) in older literature, is a complex variable easily affected by methodological inconsistencies connected to the equipment, calibrations, test maneuvers and/or potential "noise" disturbing the results(46). However, when assessed in accordance to standardized guidelines(47) in the same laboratory under equal conditions, $T_{L,CO}$ can be a variable of high importance when monitoring and evaluating pulmonary status in both patients with respiratory diseases and healthy individuals(3, 46, 47).

$T_{L,CO}$ is the product of the permeability factor (kCO) and alveolar volume (VA). When kCO is adjusted for the pressure of dry gas present in the lungs (Equation 7.1), the quotient is the diffusion constant (KCO)(3, 46).

$$T_{L,CO} = \frac{kCO \; x \; VA}{(barometric\ pressure - water\ vapor\ pressure\ in\ 37°C)}$$

Equation 7.1 The relationship between transfer factor for carbon monoxide ($T_{L,CO}$), diffusion constant for carbon monoxide (kCO) and alveolar volume (VA). By subtracting the barometric pressure (101.3 kPa at sea level) from the pressure of water vapour at 37°C (6.3 kPa), one is left with the pressure of dry air, mainly consisting of oxygen, carbon dioxide and nitrogen(3).

Illustrated Figure 7.2, $T_{L,CO}$ is furthermore determined by a number of factors, briefly divided into three points:

- **D_M**: The anatomical diffusion properties of the alveoli and the membrane conductivity; the potential of gas diffusion across alveolar capillary membrane(3).
- **V_C**: The blood volume of the pulmonary capillaries.

- **θ**: The chemical reaction rate between CO and hemoblobin (Hb). To indicate the hematologic properties of the blood, V_C and θ are multiplied(47).

Consequently, *T*L,CO changes accordingly depending on the magnitude of the potential changes in D_M and/or θV_C(46, 47). By also assessing and interpreting *K*CO and/or VA, one may detect on what step of gas diffusion a potential change has occurred. Reduced gas diffusion can moreover be confirmed by examining oxygen saturation. This parameter is likely to be reduced if the alveolar gas diffusion ability is impaired(23).

In otherwise healthy adults, Cotes, Chinn and Miller (3) claim that immediate changes in *T*L,CO usually are caused by variations in V_C. Under normal conditions, V_C is increased during exercise, when in a lying position or when blood is pooled centrally due to peripheral vasoconstriction(3). By contrast, a standing posture or a post-exercise redistribution of blood to previously active muscles reduces V_C, and a decrease in *T*L,CO may be observed – even 2-8 hours after maximal exercise(3, 23, 48). Seeing that this is a normal circulatory response following strenuous exercise, no mechanical changes are usually observed in D_M(3, 23). Oxygen saturation is also found to be normal – even after maximal exercise(3, 23).

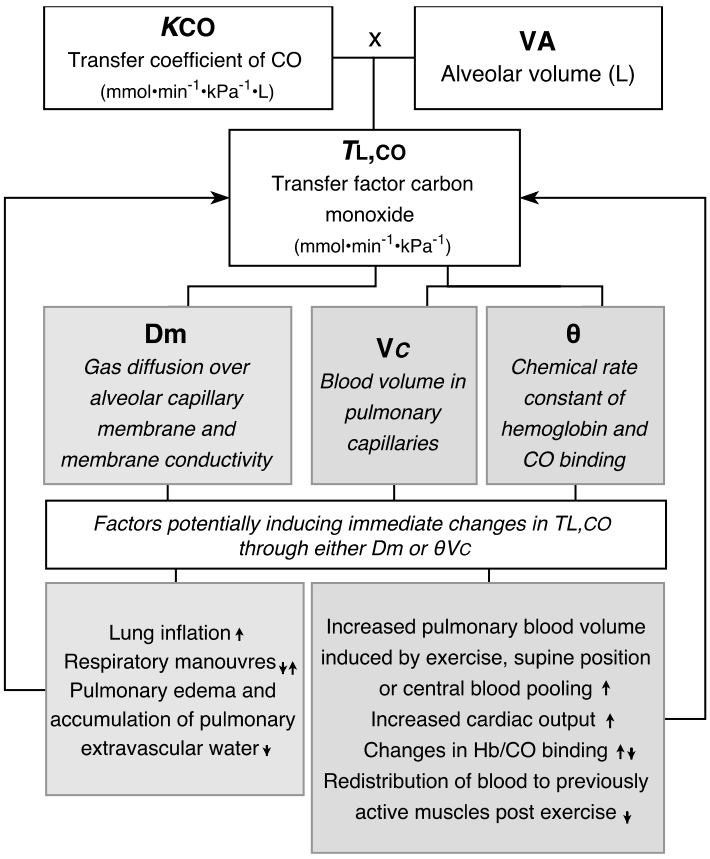

Figure 7.2 Transfer factor for carbon dioxide (*T*L,CO) can be affected by physiological responses altering the membrane component (Dm) and/or the blood/hemoglobin component (θVc). Included in the figure are factors known to induce immediate changes in these components, consequently altering *T*L,CO in the directions indicated with arrows.

A limited number of studies have examined the response in $T_{L,CO}$ following skin cooling. After an exercise bout until exhaustion on a cycle ergometer, Storebo, Hope, Vaagbo et al. (48) observed a significant post-exercise decline in both $T_{L,CO}$ and K_{CO}. Immediately after exercise, participants were requested to wait in room temperature (control) for 30 minutes or outside in 3-9°C (experiment) until the first muscle contraction (shivering) was observed. Neither room temperature nor post-exercise skin cooling seemed to induce further reductions in $T_{L,CO}$, as both parameters remained unaltered even after skin cooling. Corresponding with the statement of Cotes, Chinn and Miller (3), the post-exercise reduction in gas diffusion was ascribed the redistribution of blood from pulmonary circulation to the previously active muscles.

In regards to the arterial oxygen saturation measured by pulse oximetry (SpO_2), there are few indices to indicate that an endurance OWS in cold water would affect this parameter in healthy adults. When performing high-intensity exercise, the concentration of H+ in the blood increases. This would reduce blood pH, right-shifting the O_2-Hb dissociation curve and reducing the affinity for O_2 (metabolic acidosis)(22, 23). To maintain an adequate gas exchange, this right-shift is counterbalanced as the core temperature and ventilation are increased during continued exercise.

Although highly trained athletes seem susceptible in developing exercise-induced arterial hypoxemia (EIAH) when performing whole-body exercise (such as swimming), this would usually require (close-to) maximal exercise intensity(23). These athletes would have a large cardiac output, reducing the pulmonary transit time to an extent as to induce a V/Q mismatch as previously described(23). Additionally, a large LBM may require a large amount of O_2 to keep up high intensity exercise(23). During whole-body exercise, there may be a large extraction of O_2 from the blood, increasing the arteriovenous O_2 difference(22, 23). In combination, these factors could reduce SpO_2 in highly trained athletes, but should not be expected to occur during/after an OWS in cold water where the exercise intensity is to be low and/or moderate.

As previously mentioned in 6.2.2 Swimming-induced pulmonary edema (SIPE), this life-threatening syndrome is caused by stress-induced disruptions of the alveolo-capillary membrane. By altering D_M, both $T_{L,CO}$ and K_{CO} will be reduced(3, 5). Accompanied by a concurrent decline in oxygen saturation of varying degree(19, 20, 23, 36), this medical condition will impair pulmonary parameters and alveolar gas

diffusing properties significantly. Unless SIPE is developed, there are no current evidence to suggest that *T*L,CO, nor any of it's subcomponents, should be reduced by immersion in neither warm nor cold water(3).

7.4 Fractional orally exhaled nitric oxide (FE_{NO})

The physiological role of nitric oxide (NO) has been studied excessively since the beginning of the 1990'ies when it was first described how the molecule could be used as a marker of eosinophilic airway inflammation in asthmatic individuals(49). As standardized assessment protocols and official recommendations were made, one quickly discovered that FE_{NO} would differ between healthy individuals and individuals with lung diseases(50).

When used as a marker of eosinophilic inflammation, NO is produced by type II nitric oxide synthases (NOS-II; formerly known as inducible nitric oxide synthases, iNOS) located in the airway epithelial cells and originates from the lower and upper airways(50). Unlike other types of NOS' which are dominantly calcium-calmodulin dependent for NO synthesis, NOS-II's are stimulated by proinflammatory cytokines and are therefore regarded as an essential part of the eosinophilic inflammatory response of the airways(40, 50)

Raised levels of eosinophilic inflammatory markers – such as NO – can therefore be observed in asthmatic individuals(50). BHR affected athletes reporting large training volumes in polluted environments have also measured raised levels of eosinophilic inflammatory markers – presumably caused by repeated epithelial damages occurring during training and competitions(39, 41). On the other hand, levels of neutrophilic inflammation markers have been found to correlate with weekly training volume, regardless of environmental and/or temperature exposure(39, 41).

As mentioned in a previous paragraph, elite triathletes(42) and competitive swimmers(41) are at high risk of developing BHR and EIB. When knowing the link between repeated epithelial damages, BHR and increased airway inflammation, this subgroup of athletes may measure elevated levels of exhaled eosinophilic markers after strenuous and/or long-lasting exercise in polluted and/or cold environments(41) – such as an OWS in cold water. Studies have also shown how even an occasional exposure to chlorinated air could cause transient epithelial damages and a following development of

BHR(41), consequently making even recreational swimmers susceptible for this unfortunate development.

Pendergast, Krasney and DeRoberts (51), on the other hand, found reduced levels of FE_{NO} after gradually increasing exercise intensity (cycle ergometer) in gradually decreasing water temperatures in a group of healthy men. It was hypothesized that iNOS could be temperature-sensitive and thus impaired by the inhalation of cold air(51). It was furthermore suggested that desaturated hemoglobin could have a higher affinity for NO than oxygenated hemoglobin, increasing diffusion of NO across the alveolar-capillary membrane and thereby reducing the amount of NO exhaled by breath(51). Reduced levels of FE_{NO} after strenuous exercise have later become a well-documented phenomenon in healthy adults(52-54), and have furthermore been observed during resting conditions in individuals suffering from pulmonary edema(49, 50), hypothermia(50) and exercise-induced arterial hypoxemia (EIAH)(38).

The need for future research on the exact role of NO is eminent, as the results are inconsistent – clearly affected by the study sample's health status(49, 50, 52), ambient temperature and humidity(39, 50), altitude(49, 53) and the settings in which measurements were performed (during exercise or resting conditions)(3, 50).

8 Methods and materials

8.1 Overview

The current study was classified as a quasi-experimental cross-sectional study with pre/post measurements, and was carried out and completed in the period from October to December 2015.

All variables were collected as a component part of a larger study, aiming to examine potential changes in skin and core temperatures during and after cold water swimming in tight-fitted, swim-specific neoprene wetsuits (Ph.D dissertation by Jørgen Melau). Jonny Hisdal (PhD) was appointed project leader, whereas Trine Stensrud (PhD) at the Norwegian School of Sport Sciences (NIH) was responsible for the organization and collection of the data used in this Master's thesis.

The practical execution of testing on Day 1 was divided equally between Julie Stang, Jon W. Rødland (Bachelor's degree student), Øyvind Rossvoll (Bachelor's degree

student) and Camilla R. Illidi (author and Master's degree student). Under the guidance from Stensrud and Stang, the practical execution of testing on Day 2 was carried out by Rossvoll, Rødland and Illidi. After all the variables were collected, the raw data were plotted and processed by Stensrud and Illidi.

Hisdal and Melau applied to the Regional Committees for Medical and Health Research Ethics (REK South East) for research approval. The decision was that the present study was not obligated to be evaluated by REK according to the Act on medical and health research (the Health Research Act) from 2008. The project was commenced immediately after approval was granted.

Financial support was given by the Norwegian Triathlon Federation and Hardangervidda Triathlon Club for the specific purpose of purchasing thermometric equipment. No conflicts of interest have been made apparent.

8.2 Recruitment and inclusion

8.2.1 Sample size calculations

A priori sample size calculations (based on the main aim of the project) resulted in a minimum requirement of eight men and eight women (n=16) to detect a statistically and clinically significant change in core temperature of 0,1°C. As a result of NIH's late involvement, no 'a priori' sample size calculations were performed to find the adequate sample size to detect a clinically significant change in lung function. However, post-hoc calculations were performed to examine the achieved effect size in the significant changes observed. The process and calculations are described in detail in 8.4.2 'Post hoc' calculations.

8.2.2 Recruitment and inclusion criteria

Twenty-five individuals were recruited through triathlon-related websites, social media and spoken communication, and only 19 (♀7/♂12) appeared at scheduled time. All potential participants were required to pass a mandatory resting electrocardiography screening and blood pressure monitoring before embarking on the OWS challenge. There were no criteria regarding the style of swimming other than the ability to complete a continuous 3800 meter swim with a maximum time limit of 90 minutes. They were also obliged to bring their own swim-specific, neoprene wetsuits.

Participants were given a detailed written account of the research protocol and potential risks connected to project completion. They were furthermore informed about privacy regulations, the voluntariness of participation and that they were free to withdraw from the project at any given time (see 12.2 Appendix 2 – Participant information and written informed consent (Norwegian). This was first sent by e-mail during project recruitment, and once again presented at project start as the written informed consent was to be signed.

8.3 Project coordination and data collection

Figure 8.1[1] illustrates the project timeline, including the assessed variables and times of measurement. The study sample was divided into three equally sized groups, each group scheduled for Day 1 on one of three consecutive days.

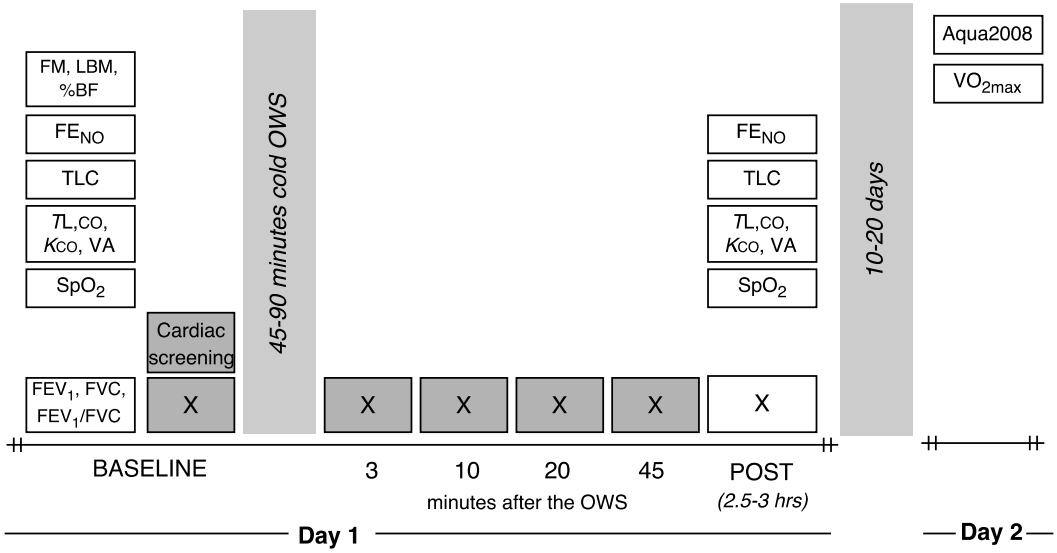

Figure 8.1 Timeline of test procedure, including assessed variables and their specific times of measurement. Body composition by DXA was assessed at baseline, together with FE_{NO} as a marker of eosinophilic airway inflammation, total lung capacity, TLC), alveolar gas diffusion parameters for carbon monoxide (TL,CO, KCO and VA), SpO_2 and maximally expired flow-volume curves (FEV_1, FVC and FEV_1/FVC x 100). The tests were repeated 2.5-3 hours after the OWS (labelled POST) on Day 1 (white boxes). Cardiac screening was performed and lung function by maximally expired flow-volume curves were again collected directly prior to the OWS (grey boxes), and repeated 3, 10, 20 and 45 minutes after the OWS. On Day 2, 10-20 days after Day 1, participants completed the Aqua2008 questionnaire and maximal oxygen uptake (VO_{2max}) was obtained[1].

[1] Figure 8.1. FM, fat mass; LBM, lean body mass; %BF, percentage body fat; FE_{NO}, fractional exhaled nitric oxide; TLC, total lung capacity; TL,CO, transfer factor for carbon monoxide; KCO, transfer coefficient for carbon monoxide; VA, alveolar volume; SpO_2, peripheral arterial oxygen; FEV_1, forced expiratory volume in one second; FVC, forced vital capacity.

Day 2 was appointed according to laboratory availibility and personal schedules. As a measure of precaution during the OWS, both Day 1 and Day 2 were performed and completed individually by each participant.

8.3.1 Day 1

During baseline testing, body composition characteristics were obtained by dual-energy X-ray absorptiometry (DXA; Lunar Prodigy densitometer, GE Medical Systems, US) and digital weight and height measurements (Seca 877, Seca, Hamb., Ger.). The participants were instructed to abstain from food and to avoid ingestion of any liquids the last 8 hours prior to the DXA, but were permitted to eat directly after the scan was completed. Caffeine was not permitted until the OWS and post measurements were completed.

Figure 8.2 Standardized lung function measurements were performed at the laboratory at NIH (baseline and post, n = 19) and at the seaside directly prior to the OWS (illustrated by the pictures). To examine the immediate responses in lung function, the measurements were repeated 3, 10, 20 and 45 minutes after the OWS (n = 12). Reprinting was permissioned by photographer Jørgen Melau on February 3rd 2017. Written permission is attached as 12.3 Appendix 3 – Picture permission.

Lung function was determined by the use of maximal expiratory flow-volume curves (Masterscreen PFT System, Carefusion/BD, San Diego, US), performed according to standardized guidelines(26). Calibration was performed using a calibration syringe of 3 liters (3L Calibration Syringe Series 5530, Hans Rudolph Inc., Kansas City, US), and adjusted to ambient conditions. TLC was assessed by standardized(43) whole-body plethysmography (Vmax Autobox V62J, Carefusion/BD, San Diego, US). Alveolar gas diffusion was assessed by the single-breath determination of carbon monoxide uptake (47) with a breath-hold time of 8 seconds (MasterScreen PFT System, Carefusion/BD,

San Diego, US). The inhaled gas composition was 0.3% CO, 0.3% acetylene, 0.3% methane, 20.9% O_2 and 72.8% nitrogen (Lung test gas, AGA, The Linde Group, Oslo, NO). Gas diffusion parameters were all adjusted for hemoglobin levels of 14.6 g/dL and 13.4 g/dL for men and women, respectively(46).

FE_{NO} was measured as a marker of eosinophilic airway inflammation by standardized(49) online measurements with a chemiluminescence analyzer (CLD88sp, Eco Medics, Dürnten, CH). The analyzer was calibrated with a syringe of 100 mL (100ml Calibration Syringe Series 5510, Hans Rudolph Inc., Kansas City, US). Peripheral oxygen saturation of arterial blood (SpO_2) was registered using a finger-mounted pulse oximeter (Spot Vital Signs, LXi, Welch Allyn, New York, US).

After baseline testing, participants were transported from the laboratory to the seaside (a drive of approximately 30 minutes) where a mandatory cardiac screening(55) was performed by authorized medical personnel, as to exclude potentially hypertensive individuals (Cardiovit AT-102 Plus, Schiller, Baar, CH). Directly prior to the OWS, lung function was again determined by maximal expiratory flow-volume curves (Masterscreen Pneumo spirometer (Carefusion/BD, San Diego, US).

After the completion of the OWS, lung function was measured regularly after 3, 10, 20 and 45 minutes. Following the last assessment at 45th minute, participants were transported back to the laboratory to repeat baseline tests (with the exception of DXA, which was only performed during baseline testing).

Figure 8.3 Participants were instructed to swim alongside a quay in laps of approximately 100 meters (red line), during which medical personnel retained oral communication with the participants. Reprinting was permissioned by ©Kartverket (GEOVEKST, Rambøll Norge AS)(2).

8.3.2 Open water swim

Wearing a wetsuit, specifically designed for triathlons and swimming, participants were required to complete a 3800 meter OWS with a maximum time limit of 90 minutes alongside a quay in laps of 100 meters (see Figure 8.3). Style of swimming and exercise intensity were self-selected. Ambient

temperature was 5-10ºC, and water temperature measured 10±0.9ºC (Reed ST-612, Reed Instruments, NC, US).

Medical personnel were placed along the quay, maintaining oral communication with the participants regularly during the OWS and asking control questions (e.g. day of the week, name of the Norwegian king) to monitor cognitive state and alertness.

After the first of three days of swimming/testing, a large decline in the participants' core temperature was observed. Consequently, it was rendered ethically necessary to shorten the OWS. The remaining two-thirds of the study sample were thus required to swim for only 45 minutes, regardless of distance covered in the given time.

8.3.3 Day 2

Approximately 10-20 days after Day 1, maximal oxygen uptake (VO_{2max}) was measured (Oxygon Pro, Jaeger Instrument, Carefusion/BD, San Diego, US) using a graded (5,3%) step test on treadmill (Bari-Mill, Woodway, Wisconsin, US) with gradually increasing running speed each minute until a level of great exhaustion(56). Calibration was performed according to equipment manuals and guidelines(26) using a volume calibration syringe of 3 liters (3L Calibration Syringe Series 5530, Hans Rudolph Inc., Kansas City, USA). Values were obtained as an average of 30 second measurements.

All participants were allowed to complete warm-up of easy running (10 minutes) on the treadmill before test start. During the test, all subjects wore a nose clip (9015 Reusable Series, Hans Rudolph Inc., Kansas City, US) and used a silicone rubber mouth-piece (9060 Reusable Series, Hans Rudolph Inc., KS, USA). VO_{2max} was identified when a plateau (a rise of less than 2 $mL \cdot kg^{-1}min$ in VO_2, despite increasing running speed) was observed by the testing personnel(56). Additionally, two more criteria of VO_{2max} were applied: A respiratory exchange ratio ($RER = \frac{VCO_2}{VO_2}$) of ≥1.10 and heart rate of ≥95% of HF_{max} (based on personal maximum heart rate or 220 $beats \cdot min^{-1}$ – age). Heart rate was monitored by a chest strapped heart rate monitor which was synchronized with a sports watch (Polar RS400 Endurance Sports Watch, Polar Electro OY, Kempele, Finland). Borg Scale of Perceived Exertion$_{6\text{-}20}$(57) and blood lactate levels (Lactate Analyzer 1500, YSI Inc., Xylem Analytics Inc., New York, US) were also assessed immediately after test termination(56), but were not used as criteria as the VO_2 plateaued in all of the participants. A modified version of AQUA: Allergy Questionnaire for Athletes (see

Bonini, Braido, Baiardini et al. (58) and Appendix 1 – Modified Aqua2008) was completed, reporting weekly training volumes (in total and in chlorinated swimming pools).

8.4 Data processing and statistical analyses

Due to skewed data distributions and small sample size, we decided to present the results as median together with the 25^{th}-75^{th} percentile. Non-parametric tests were used to perform the hypothesis tests. The matters were discussed, and agreed upon, with both study counsellors (Stensrud and Stang) and the statistician at NIH, Morten W. Fagerland.

The statistical work presented in the included paper was performed using IBM SPSS Statistics (IBM Corporation, NY) and Microsoft Excel 2016 (Microsoft Corporation,WA). Level of significance (α) was set to $p \leq 0.05$.

8.4.1 Hypothesis tests

Wilcoxon Signed Rank Tests were used to examine the changes between the repeated measures at baseline and post assessment. To analyse for immediate changes in lung function measured at the seaside, Friedman Tests were performed. When significant changes were found using Friedman Tests, Wilcoxon Signed Rank Tests were applied to study differences between two repeated measures.

Mann-Whitney *U* tests were used to examine potential differences between two independent samples (men/women), both in demographic and anthropometrical characteristics, but also when analysing for pulmonary responses to the OWS.

When significant changes in pulmonary variables were found, the participants' percentage changes were correlated with anthropometrical variables (LBM; fat mass, FM; %BF) and weekly training volume (total and in swimming pool) using Kendall's τ. Rank correlation coefficients were calculated as to study the potential relationship between lung function impairments and body composition characteristics and weekly training volumes.

8.4.2 'Post hoc' calculations

As mentioned in a previous paragraph, Melau and Hisdal (59) performed 'a priori' sample size calculations based on the main variable in the study; the absolute change in core temperature of 0.1ºC. Consequently, we were not certain if the included number of

participants would be adequate as to avoid making a statistical type II error(60). By the school statistician, it was strongly discouraged to perform a 'post hoc' power analyses, as these results often are misleading. Instead, effect sizes should be calculated and presented to indicate the substantive value of statistically significant results(60).

Effect size (*r*) calculations were performed by using the equation suggested by Pallant (61), utilizing the Z value associated with the specific Wilcoxon Signed Ranks Tests in IBM SPSS Statistics $\left(r = \frac{Z}{\sqrt{n_x+n_y}}\right)$. $n_{x/y}$ = the number of observations on the two points of time. Pallant (61) furthermore encourages the use of Cohen's criteria when interpreting the estimated effect sizes, grading the effect sizes (0.1 = small, 0.3 = medium, 0.5 = large) to describe the strengths of the significant changes observed (cited by Pallant (61)).

8.5 Methodological considerations

Like in most other research projects, there are a number of deliberations to be made regarding our choice of research design and preferred measuring devices. In the following paragraphs there are both methodological strengths and limitations to be accounted for.

8.5.1 Study design

During the last decades, authors have suggested a causal connection between physical contact during chaotic mass starts in triathlons and the occurrence of circulatory abnormalities, such as ventricular fibrillation or a supranormal rise in pulmonary artery pressure in susceptible athletes(10, 11, 18). As described in a previous paragraph, increased pulmonary artery pressure may cause SIPE, potentially impairing lung function and inducing great physical and mental distress to the affected individual. Because our OWS challenge was performed individually (without the physical and psychological aspects of a mass start) in a self-selected pace and exercise intensity, it is likely that our results could differ from what may have been found if the OWS challenge was more similar to a race situation.

Although our study sample (n = 19) was too small as to use the mean as our measure of central tendency, standard deviation as the measure of dispersion and statistical tests of higher validity (such as Pearson's r for correlation analyses and Student's t-tests for time/group differences), our study sample was larger than what is normally observed in

similar studies. A great majority of the literature on SIPE is based on smaller case reports(19, 20, 36), whereas the largest research projects on pulmonary responses to a given stimuli rarely count more than 30 subjects(5, 14, 31, 37, 48, 51). In this case, a small sample size made it possible to assess a great number of parameters regularly over a fairly long period of time (0-3 hours). Moreover, we were able to use time-consuming and/or costly assessment methods, such as DXA, whole-body plethysmography and the measurement of maximal oxygen uptake(43, 62). However, precise conclusions are difficult to obtain when sample sizes are small, thus making it evident that there are both strengths and limitations with a study sample of 19.

8.5.2 Measuring devices

One of our concerns regarding the measurement of arterial oxygen saturation was the use a finger-mounted pulse oximeter. As poor peripheral perfusion is found to reduce the accuracy of the oximetry analysis, there is a chance that measurement errors were made(63). After completing the OWS challenge, most of the participants reported a varying degree of coldness, especially in their hands, feet and face (in a written post-project report, not published). Although there were approximately 2.5-3 hours from the cold water exit to the post assessment of SpO_2, it is likely that some of the participants still were affected by cold extremities even after this amount of time. Consequently, SpO_2 could have been underrated in some participants.

An alternative to noninvasive pulse oximetry is an arterial blood gas sampling test, in which levels of arterial gas tension (PaO_2 and $PaCO_2$), pH, lactate and hemoglobin can be obtained(3, 64). The blood gas analyzer must be temperature adjusted according to the exact blood temperature as to report valid results(64). If not, arterial blood gases may be misinterpreted in individuals measuring abnormal core temperature. Unlike a finger-mounted pulse oximeter, low skin temperature is not found to reduce the measurement validity of a blood gas analyzer(64). Instead, this would require the appropriate, qualified laboratory personnel who were not available for the present study.

Core and skin temperatures were also registered during the OWS challenge. Knowing how even mild hypothermia can affect the pulmonary and circulatory system(3, 11, 28), it would have raised the quality of our statistical analyses if these variables were included. In that case, one may have examined the statistical correlations between potential temperature changes and changes in our pulmonary and inflammatory

parameters. Owing to the fact that the assessment of skin and core temperatures was the primary objective of this project, the variables were not available for this thesis.

When detecting changes in lung function, alveolar gas diffusion and oxygen saturation after strenuous swimming, case reports on SIPE usually report chest x-rays as a tool for diagnostic confirmation(5, 17, 19, 36). There would be several advantages in including chest radiography (either x-rays or computerized tomography) to a future study. Firstly, one might confirm/reject the presence of SIPE in the individuals reporting larges, pulmonary declines(3, 5). Secondly, this would increase the validity of our results, increasing the over-all quality of our study(16). However, this would require qualified personnel, the access to proper equipment and more logistic arrangements in terms of executing a similar-looking protocol.

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11 Paper with tables and figures

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Reduced lung function, oxygen saturation and eosinophilic inflammation markers following an endurance swim in cold water

ABSTRACT

Objectives: We aimed to study the pulmonary responses to an organized open water swim (OWS) in cold water (10±0.9ºC), and secondarily to investigate the correlation between potential pulmonary responses and body composition and weekly training volumes.

Methods: Nineteen (♀7/♂12) healthy individuals completed a 45-90 minutes OWS wearing a wetsuit. Maximal expiratory flow-volume curves, whole-body plethysmography, transfer factor for carbon monoxide (TL,CO), exhaled fractional nitric oxide (FE_{NO}), oxygen saturation by pulse oximetry (SpO_2) and body composition by DXA measurements were obtained before and 2.5-3 hours after the OWS. Additionally, maximal expiratory flow-volume curves were measured in 12 participants before and 3, 10, 20 and 45 minutes following the OWS. Results and effect sizes are given as median (25^{th}, 75^{th} percentile) and *r*, respectively.

Results: Four out of 12 participants (33.3%) developed exercise-induced bronchoconstriction (EIB), a ≥10% decline in forced expiratory volume in one second (FEV_1). FEV_1 was significantly reduced immediately after the OWS: -6.32% (-8.49, -5.41), ($p = 0.008$, $r = 0.568$). After 2.5-3 hours, significant declines were observed in TL,CO: -5.47% (-8.36, -3.91), ($p < 0.001$, $r = 0.61$), SpO_2: -2.5% (-3.25, -2.00) ($p = 0.001$, $r = 0.56$) and FE_{NO}: -11.65% (-14.49, 1.44), ($p = 0.014$, $r = 0.40$). Three men (16.6%) were within the definition of exercise-induced hypoxemia, measuring SpO_2 of 94-95%. No significant correlations were found between body composition variables, weekly training volume and changes in pulmonary function.

Conclusion: One third developed EIB. Pulmonary function, the level of eosinophilic inflammatory markers and arterial oxygen saturation were reduced up to three hours following the OWS. Although we cannot indicate why, certain individuals – predominantly men – experienced severe pulmonary impairments during/after the swim.

Considering the large effect sizes, these impairments could be of crucial practical significance to the swimming athlete.

WHAT ARE NEW FINDINGS?

- Lung function was significantly reduced immediately after an OWS in cold water. Four out of 12 participants (33.3%) furthermore experienced exercise-induced bronchoconstriction (≥10% decline in FEV_1) during the first 10 minutes after the OWS.
- Alveolar gas diffusion for carbon monoxide, oxygen saturation and fractional exhaled nitric oxide were significantly reduced up to three hours after the OWS. Weekly training volumes and body composition did not seem to influence the observed responses in the present study.
- Certain individuals, predominantly men, developed severe pulmonary impairments during/after the OWS, and the occurrence of swimming-induced pulmonary edema (SIPE) may be suggested.

KEY WORDS

- Facial-cooling induced bronchoconstriction reflex
- Exercise-induced bronchoconstriction
- Exercise-induced hypoxemia
- Swimming-induced pulmonary edema
- Cold water swimming
- Triathlon

INTRODUCTION

Triathlon competitions and open water swimming (OWS) events have become popular leisure time activities over the last decades, attracting a broad range of individuals of different ages and fitness levels(1, 2). The increasing number of extreme long-distance races, arranged in rapidly shifting weather conditions and cold temperatures is also evidence of the growing interest(3, 4).

There seems to be a consensus of opinion regarding the pulmonary responses to cold water swimming. Although there are initial reflex responses to cold water immersion, such as gasping and hyperventilation(5), the pulmonary system itself should remain unaffected by cold air ventilation(6) and cold water immersion(5, 7) – given that the individual is healthy and normothermal.

However, a growing body of literature shed new light on the risks associated with OWS events(8). Hypothermia(6, 9), swimming-induced pulmonary edema (SIPE)(10-16) and exercise-induced bronchoconstriction (EIB) have been reported during and after exercise and swimming in cold conditions. These are all medical conditions known to impair lung function (forced expiratory volume in one second, FEV_1; forced vital capacity, FVC)(5, 6, 10, 11, 17). Additionally, reduced alveolar gas transfer for carbon monoxide (TL,CO) has been observed in well-trained triathletes after a triathlon – presumably to be explained by the occurrence of SIPE(16, 18). Affected by SIPE, the accumulation of interstitial fluids and reduced lung compliance would furthermore reduce the total lung capacity (TLC)(5). By contrast, there is little evidence to indicate that TLC should be affected by neither hypothermia(19) nor cold water swimming in healthy adults(6, 7, 17).

Cold water temperatures, pre-race overhydration and strenuous exercise have been suggested to trigger the abnormal pulmonary responses observed during and after OWS events(12-14). A medical history of hypertension and long swimming distances(20) or intense mass starts(8) have also been suggested to be potentially triggering factors. Asthmatic and bronchial hyperresponsive individuals in particular, could be negatively affected by ventilating cold air during exercise, causing a reflex parasympathetic-stimulated bronchoconstriction(5, 21, 22). In otherwise healthy adults, Koskela and Tukiainen (23) also observed how facial cooling alone, without the inhalation of cold air, could induce a reflex bronchoconstriction through the afferent and efferent arches of n. trigeminus and n. vagus, respectively(24). These are all phenomena increasing airway resistance, potentially causing dysponoea when swimming in cold water.

High incidences of bronchial hyperresponsiveness (BHR) and EIB have been found among elite triathletes(25), but also among competitive swimmers and cold-air athletes(16, 21, 26). Repeated exposure to airway irritants during training, such as chlorine and cold air, have thus been associated with the development of BHR, EIB and eosinophilic airway inflammation(16, 21, 26). Large training volumes, regardless of environmental, ambient factors, have been found to raise levels of neutrosinophilic inflammation markers(26, 27). How an OWS in cold water can affect lung function and the levels of fractional exhaled nitric oxide, a commonly used marker of eosinophilic airway inflammation(27), is therefore an intriguing query.

A high percentage body fat (%BF) is important in terms of thermal insulation and to delay the onset of cold-induced reflex responses(6). The risk of developing hypothermia during immersion is furthermore strongly related to body composition characteristics(6, 7). If lean individuals would be more susceptible to pulmonary impairments during/after an OWS in cold water, compared to individuals with a high %BF, is thus another interesting query waiting to be assessed.

Well-controlled, experimental studies regarding the risks associated with OWS, especially in cold water, are lacking. Our primary aim was therefore to assess pulmonary responses from before to after (3 minutes – 2.5-3 hours) an OWS in cold water, and secondarily to investigate the correlations between possible pulmonary changes and body composition (%BF; lean body mass, LBM; fat mass, FM) and weekly training volumes.

METHODS

Recruitment and inclusion

Active, nonsmoking men and women (n = 19, ♀7/♂12) were recruited through spoken communication and social media advertisements on triathlon related websites. Before inclusion, all participants passed a cardiac screening, excluding potentially hypertensive individuals (Cardiovit AT-102 Plus, Schiller, Switzerland; protocol described in detail by Maron, Friedman, Kligfield et al. (28)). None of the participants reported any doctor-diagnosed allergic or pulmonary diseases, and were thus regarded as a healthy study sample.

We applied the regional ethical committee (REK), and the decision was that the present study was not obligated to be evaluated by REK according to the Act on medical and health research (the Health Research Act) from 2008. The study was performed according to the declaration of Helsinki and written informed consent was granted at project start.

Study design and methods

The current project was a quasi-experimental cross-sectional study with pre/post measurements, and the presented data were collected as part of a larger study aiming to assess core temperature during and after an OWS (study in progress by Melau and Hisdal (29)). Figure 1 illustrates the executed protocol as a timeline, including the

assessed variables and times of measurements. Day 1 was performed individually by each participant on one of three consecutive days, whereas Day 2 was scheduled 10-20 days after Day 1.

Day 1

Laboratory assessment

On first attendance, dual-energy x-ray absorptiometry (DXA) was measured (Lunar Prodigy densitometer, GE Medical Systems, US) followed by the assessment of pulmonary variables in the laboratory. Lung function was measured by maximal expiratory flow-volume curves and expressed as FEV_1, FVC and their ratio (FEV_1/FVC) (MasterScreen PFT System, CareFusion, US).

Transfer factor for carbon monoxide (TL,CO) by the single-breath testing technique (MasterScreen PFT System, CareFusion), total lung capacity (TLC) by whole-body plethysmography (Vmax Autobox V62J, CareFusion/BD, US), fractional expired nitric oxide (FE_{NO})(CLD88sp, Eco Medics, Dürnten, CH) and arterial oxygen saturation (SpO_2) registered by a finger-mounted pulse oximeter (Spot Vital Signs, LXi, Welch Allyn, N.Y, US) were assessed at baseline and 2.5-3 hours after the OWS. All parameters were collected according to standardized guidelines(30-34).

Field assessment and open water swim

Participants were transported to the seaside and lung function was again assessed by flow-volume curves (MasterScreen Pneumo spirometer, CareFusion/BD, US) preceding the OWS. Owing to an equipment failure on the second day of testing, we were only capable of assessing field spirometry in 12 of our 19 participants (63.2%).

Wearing tight-fitted neoprene wetsuits, participants performed a 3800 m swim in cold seawater with a maximum time limit of 90 min. Swimming stroke and exercise intensity were both self-selected. Due to a rapid decline in core temperature on the first of three days of testing, the remaining two-thirds were only required to swim for a maximum of 45 min, regardless of swimming distance covered in the given time. Water temperature was 10±0.9ºC and ambient temperature was 8.0-10.0 °C.

Lung function was measured immediately after (3, 10, 20 and 45 minutes) the OWS, and then transported back to the laboratory to repeat baseline tests, 2.5-3 hours after the OWS was completed.

Day 2

Maximal oxygen uptake (VO_{2max}) was obtained on a treadmill (Bari-mill, Woodway, Wis, US) by using a graded step test (Oxycon Pro, Jaeger Instrument, Germany), starting at an inclination of 5.3% and gradually increasing speed (described in detail by Astrand, Rodahl, Dahl et al. (35)). Training volumes per week (in total and in chlorinated swimming pools) were reported in the Modified Aqua2008 questionnaire(36) on the same day.

Statistical Analyses

Due to skewed distributions and a small sample size, median and interquartile range (25^{th}, 75^{th} percentiles) were used to indicate central tendency and statistical dispersion, respectively.

Non-parametric tests were performed throughout, using IBM SPSS Statistics (IBM Corporation, NY): Friedman and Wilcoxon signed ranks tests to examine significant changes over time and Mann-Whitney U test to test for sex differences. Kendall's τ was used to study the correlation between the maximal percentage changes in pulmonary parameters, anthropometrical characteristics (LBM, FM, %BF) and weekly training volumes. Effect sizes (r) were calculated and interpreted according to the guidelines of Pallant (37): r = 0.1 (small), r = 0.3 (medium), r = 0.5 (large). Level of significance was set to $p \leq 0.05$.

Sample size calculations were performed prior to study start, based on the main parameter of the study; an absolute change in core temperature. The calculations resulted in a sample size of n = 16 (♀8/♂8).

RESULTS

Pulmonary parameters

Selected anthropometrical, physiological and training characteristics are presented in Table 1. Weight, LBM, %BF and VO_{2max} differed significantly between men and women.

FEV_1 and FVC were significantly reduced 3 minutes after the OWS (FEV_1, p = 0.008, r = 0.568; FVC, p = 0.005, r = 0.597). FVC was still reduced after 10 minutes (p = 0.041, r = 0.436) (Figure 2). A total of four participants (33.3%) experienced EIB during the first 10 minutes after the OWS.

In table 2, significant declines were found in *T*L,CO: -5.47% (-8.36, -3.90), FE_{NO}: -11.65% (-14.49, 1.44) and SpO_2: -2.5% (-3.25, -2.0) between baseline ands post measurements 2.5-3 hours after the OWS. No significant changes were found in FEV_1, FVC, FEV_1/FVC and TLC. The change in *T*L,CO was primarily caused by a decline in the diffusion constant for CO (*K*CO): -4.71% (-9.01, -3.46), as alveolar volume (VA) remained unchanged.

Three out of 18 participants (16.6%) measured SpO_2 of 94-95%, which are levels regarded as mild hypoxemia. Additionally, one male measured reductions in *T*L,CO and *K*CO of -17.39% and -12.72%, respectively, and experienced reduced TLC (-5.46%) and VA (-5.59%), indicating medical signs of SIPE. There were no significant differences between men and women in either of the pulmonary responses.

Body composition and training volume correlations

In figure 3, no significant correlations were found between the maximum percentage changes in FEV_1 and *T*L,CO and anthropometrical parameters (LBM, %BF and FM) and weekly training volumes. Likewise, maximum percentage change in FE_{NO} showed no significant relationship with weekly training volume and training volume in swimming pools (Figure 4).

DISCUSSION

The main findings of the present study were that 33.3% of the participants developed EIB after the OWS, and that *T*L,CO, SpO_2 and FE_{NO} were significantly reduced 2.5-3 hours after the OWS. Moreover, medium to large effect sizes indicate that the results could be of moderate to crucial practical importance to the swimming athlete.

Although our results contradict the established scientific beliefs claiming that healthy individuals should remain unaffected by cold water swimming and immersion(5-7), they are in accordance with case reports describing changes in lung function after endurance swimming(10, 11, 23, 24, 38) and exercise in cold environments(39, 40).

Maximal expiratory flow-volume curves

A decline of ≥10% in FEV_1 is defined as EIB(41, 42), and was found in 33.3% of our participants either 3 or 10 minutes after the OWS. Additionally, both FEV_1 and FVC were significantly reduced up to 10 and 20 minutes, respectively, after the OWS.

According to Carlsen (21), a parasympathetic reflex bronchoconstriction could be

induced by ventilating cold air while exercising. The exercise-induced increase of ventilation would cool the airways, causing a heat-conserving vasoconstriction of bronchial venules. As exercise ceases and ventilation renormalizes, the vasoconstriction is rebounded by a vasodilation, inducing mucosal edema and parasympathetic-stimulated smooth muscle constriction. Consequently, airway calibre and dynamic lung function decline(5, 21). However, this reflex response is usually observed in asthmatic or BHR affected individuals after termination of exercise; not in otherwise healthy, recreationally active adults(5, 7, 17)..

Seeing that the lung function rapidly returned to baseline values in the present study, without the use of bronchodilatating treatment, one might suggest that the bronchoconstriction was caused by the "facial cooling-provoked reflex"(24)(p96) reported by Koskela et al (23, 24, 38). Unlike the theory proposed by Carlsen (21), this reflex response has been observed in healthy individuals, as well as in individuals with other respiratory disorders(23, 24).

During facial cooling, the afferent and efferent arches of n. trigeminus and n. vagus, respectively, cause a bronchoconstriction which usually ceases as the facial skin temperature is renormalized(23, 24, 38). This would imply that the peak declines in lung function occurred *during* the OWS as the participants' faces were in direct contact with the cold water. Immediately after the OWS, the subjects were brought indoors for recovery which would increase their facial skin temperature substantially, suggesting a parallel increase of facial skin temperature and lung function. Therefore, our measurements during the 3rd and 10th minute could barely have been able to register the already reversing bronchoconstriction(23). Considering the large effect sizes, this would imply that the practical significance of the bronchoconstriction could be of crucial importance to the athlete swimming in cold water.

A relationship was proposed between the maximal percentage changes in lung function and weekly training volumes, due to the high prevalence of BHR and EIB among athletes reporting large training volumes(16, 21, 25, 26). However, this was not the case in the present study. Despite the fact that our participants were regarded as *active* individuals, their training volumes were substantially lower than the ones reported in the cited studies; assessing pulmonary responses in elite, aquatic athletes. Compared to recreationally active individuals, well-trained athletes would also be able to sustain

higher exercise intensity over longer durations, maintaining a large minute ventilation(5). In a chlorinated swimming pool, elite athletes doing high intensity training would therefore inhale larger amounts of airway irritants than their less trained counterparts, potentially making the latter less susceptible to epithelial damages and BHR(26). Although chlorine-induced epithelial damages and BHR have been found in recreational swimmers and pool workers with only limited chlorine exposure(16, 21), this did not seem to affect the levels of eosinpophilic inflammatory markers in the present study sample.

Alveolar gas diffusion

Seeing that the decline in *T*L,CO corresponded with a decline in both *K*CO and SpO_2, a blood-gas barrier failure may be suspected(5, 43-45). This could be caused by an increase of capillary transmural pressure, induced by the centralization of blood when swimming in a prone position(10, 46, 47). Elevated mechanical stress on capillary endothelium and alveolar epithelium would potentially cause alveolar and/or microvascular disruptions, increasing the alveolo-capillary membrane permeability. Consequently, proteins and fluid from capillary lumen leak into alveolo-capillary interstitium and alveolar space, causing pulmonary edema and reducing *T*L,CO and *K*CO (10, 43, 48).

As in the present study, Caillaud, Serre-Cousine, Anselme et al. (18) found declines of 5.0% and 6.6% in *T*L,CO and *K*CO, respectively, after the completion of an Olympic distance triathlon. Due to the accumulation of pulmonary interstitial fluid, extravascular water and increased lung density, the occurrence of pulmonary edema was suggested. However, it was not established if the declines were caused by the high exercise intensity, long duration or a specific mode of exercise (e.g. the swim leg).

Cotes, Chinn and Miller (5) claim that "short-term variations in transfer factor are due to changes in pulmonary capillary blood volume"(p253). Reduced pulmonary blood perfusion would reduce the amount of blood available for pulmonary gas diffusion. As *K*CO reflects the quality of the alveolo-capillary gas uptake, both *T*L,CO and *K*CO are highly sensitive to blood volume changes(45, 48). Besides, a post-exercise redistribution of blood from the pulmonary circulation to previously active skeletal muscles has been found to reduce *T*L,CO with as much as 10% - even several hours after exercise

termination(48, 49), and may be logical explanation to the declines observed in the present study.

Eosinophilic airway inflammation

Based on observations reporting raised levels of eosinophilic inflammatory markers and a high prevalence of BHR among elite swimmers(26) and EIB among elite triathletes(25), it was suggested that the levels of FE_{NO} may increase after our OWS in cold water; especially in those reporting large training volumes in chlorinated swimming pools(16). Significantly reduced FE_{NO} after the OWS, together with insignificant correlations between training volumes and maximal declines in FE_{NO}, demonstrated otherwise in the present study.

Supporting our results, Pendergast, Krasney and DeRoberts (40) observed reduced levels of exhaled NO after strenuous exercise in cold water in a group of healthy men. As the exercise intensity gradually increased and the water temperature decreased, levels of NO declined correspondingly. Nitric oxide synthases, produced by epithelial cells of the central and lower airways, were therefore suggested to be temperature-sensitive, potentially inhibited by the inhalation of cold air(40). Increased pulmonary blood volume when swimming in a prone position in cold water may also increase the alveolar diffusion of NO, consequently reducing the amount of NO exhaled by breath(40). Compared to the levels of FE_{NO} measured in temperate conditions, Stensrud, Stang, Thorsen et al. (39) also found lower levels of FE_{NO} after moderate and high intensity exercise on a treadmill in cold environment. Deoxyhemoglobin has been found to have a high affinity for NO(40), suggesting that increased levels of deoxyhemoglobin, induced by a high exercise intensity and/or pulmonary impairments, may also reduce the levels of NO exhaled by breath(40).

The insignificant correlations between training volumes and exhaled levels of FE_{NO} could be explained by the small training volumes reported by our study sample. Previous reports have been made on study samples with large training volumes in chlorinated swimming pool(26), potentially increasing their susceptibility for BHR and elevated levels of inflammatory markers in exhaled breath(16, 21, 25). Although this did not seem to affect the present results, a newer review by Del Giacco, Firinu, Bjermer et al. (16) shed light on the high incidence of BHR and asthma in individuals exposed to chlorine only occasionally.

Nevertheless, studies examining the response in exhaled NO after exercise are inconsistent, clearly affected by the exercise intensity and duration, ambient temperature and the altitude on which the exercise is performed(39, 40, 50). Levels are also affected by the presence of BHR or any lung diseases in the study sample(21, 26, 30). A clear understanding of the responses in FE_{NO} to cold water activities is therefore difficult to attain and should be implemented in future research.

Body composition

Considering the importance of %BF and FM in maintaining a normal core temperature during immersion(5-7, 9), we proposed that potential changes in pulmonary variables might be influenced by body composition. In line with our insignificant correlations, it does not seem like lean individuals would be more susceptible to pulmonary changes during cold water swimming, nor that fatter individuals are protected by the insulative effect of adipose tissue(5, 7, 17).

A tight-fitted neoprene wetsuit could be of thermal protection to the leanest individuals, delaying or inhibiting a more severe response which could have occurred without a wetsuit(6). Well-trained individuals with a high level of swimming skills, measuring low %BF and high LBM may be protected by the ability to produce a substantial amount of metabolic heat during exercise(7, 9). This combination could potentially counterbalance the insulating benefit of a high %BF, consequently explaining the insignificant correlations.

STRENGTHS AND LIMITATIONS

We acknowledge that the arranged OWS might not have been adequately similar to a real race situation. As it was performed individually, in a self-selected exercise intensity, we suspect that the participants were not exposed to the psychological and physiological stress levels which can be experienced during a real race. Mass start procedure, standardized pre-race feeding/hydration and high/maximal exercise intensity should therefore be included in future research for methodological improvements.

The current project was carried out in late October; a month which is usually regarded as “off-season” among Scandinavian triathletes. It is therefore reasonable to assume that the reported training volumes were markedly lower than the volumes before/during competitive season. Acknowledging the potential relationship between weekly training volumes, elevated levels of eosinophilic inflammatory markers and the incidence of

BHR and EIB following strenuous exercise, one may question if the pulmonary responses would have been larger during a period of larger training volumes.

Facial cooling was described by most participants during and after the swim, consequently impairing facial sensations and causing involuntary swallowing of seawater. Not only does this increase the risk of developing pulmonary edema(5), but severe facial cooling also reduces the contractile properties of facial skeletal muscles(7). Methodologically acceptable spirometry measurements were therefore difficult to obtain immediately after the OWS, and could have affected the measurement validity.

To our knowledge, no previous experimental study has performed a well-controlled OWS with the collection of anthropometrical, demographic and pulmonary variables in a study sample of this size. The diversity within our sample is also a good reflection of the heterogeneity seen among participants in extreme triathlons.

CONCLUSION

One third developed EIB. Pulmonary function, the level of eosinophilic inflammatory markers and arterial oxygen saturation were reduced up to three hours following the OWS. Although we cannot indicate why, certain individuals – predominantly men – experienced severe pulmonary impairments during/after the swim. Medium to large effect sizes suggest that the impairments could be of crucial importance to the swimming athlete. However, none of the observed changes correlated significantly with weekly training volumes or body composition characteristics in the present study sample.

COMPETING INTERESTS

None.

FUNDING

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TABLES

Table 1

Table 1 Demographic, anthropometric and physiological characteristics of the study sample; as a total and for both women and men separately. Values are given as median (25th to 75th percentile)[2].

	Total	Women	Men
Number (n)	19	7	12
Age (yrs.)	35.4 (31.1, 45.2)	35.4 (33.3, 38.0)	35.4 (31.3, 45.2)
Body composition			
Weight (kg)	73.2 (68.6, 84.5)	65.5 (62.9, 72.7)*	73.2 (68.6, 84.5)
Height (cm)	179 (173, 180)	174 (172, 177)	179 (173, 180)
LBM (kg)	57.3 (49.5, 66.1)	20.0 (14.6, 23.5)†	57.3 (49.5, 66.1)
%BF (%)	20.4 (16.0, 32.0)	30.8 (20.5, 33.6)*	20.4 (16.0, 32.0)
FM (kg)	15.2 (10.9, 23.5)	20.0 (14.6, 23.5)	15.2 (10.9, 23.5)
VO_{2max}			
Relative ($ml \cdot kg^{-1} \cdot min^{-1}$)	56.2 (52.3, 65.3)	49.5 (45.5, 54.7)*	56.2 (52.3, 65.3)
Absolute ($ml \cdot min^{-1}$)	4554 (3608, 5338)	3459 (3326, 3595)†	4554 (3608, 5338)
Hours training pr. week			
Total (hrs.)	8.5 (6.0, 12.5)	7.2 (5.5, 8.5)	8.5 (6.0, 12.5)
Indoor swimming pool (hrs.)	1.0 (0.5, 2.0)	1.0 (0.7, 2.1)	1.0 (0.5, 2.0)

[2] Table 1.*$p < 0.05$; † $p < 0.001$, value significantly lower in women (Mann-Whitney *U* test).

LBM, lean body mass; %BF, percentage body fat; FM, fat mass; VO_{2max}, maximal oxygen uptake.

Table 2

Table 2. Forced expiratory volume in one second (FEV_1), forced vital capacity (FVC), fractional exhaled nitric oxide (FE_{NO}), total lung capacity (TLC), transfer factor for CO (TL,CO), diffusion constant for CO (KCO) and arterial oxygen saturation (SpO2) before (baseline) and 2.5-3 hours (post) OWS in cold water (n = 18). Results are given as median (25^{th} to 75^{th} percentile).[4]

	Baseline	Post	P value	Effect size (r)
FEV_1 (L)	4.11 (3.74, 4.52)	4.08 (3.84, 4.43)	0.631	0.07
FVC (L)	5.22(4.52, 5.96)	5.16 (4.5, 5.84)	0.433	0.12
FEV_1/FVC	0.77 (0.75, 0.79)	0.77 (0.76, 0.80)	0.631	0.13
TLC (L)	6.27 (5.71, 7.51)	6.39 (5.66, 7.21)	0.76	0.05
***T*L,CO** ($mmol \cdot min^{-1} \cdot kPa^{-1)}$	10.03 (8.34, 12.81)	9.49 (7.6, 11.0)	< 0.001†	0.61
***K*CO** ($mL \cdot min^{-1} \cdot kPa^{-1} \cdot L^{-1}$)	1.56 (1.49, 1.64)	1.49 (1.42, 1.51)	< 0.001†	0.61
VA (L)	6.11(5.53, 7.35)	6.24 (5.51, 7.05)	0.744	0.53
SpO_2 (%)	99 (98, 100)	96 (96, 97)	0.001†	0.56
FE_{NO} (ppb)	17.05 (14.05, 21.85)	15.85 (13.7, 20.05)	0.014*	0.40

[4]Table 2. * p < 0,05; † p < 0,001, significantly different from baseline measurement.

FIGURES

Figure 1

[5]

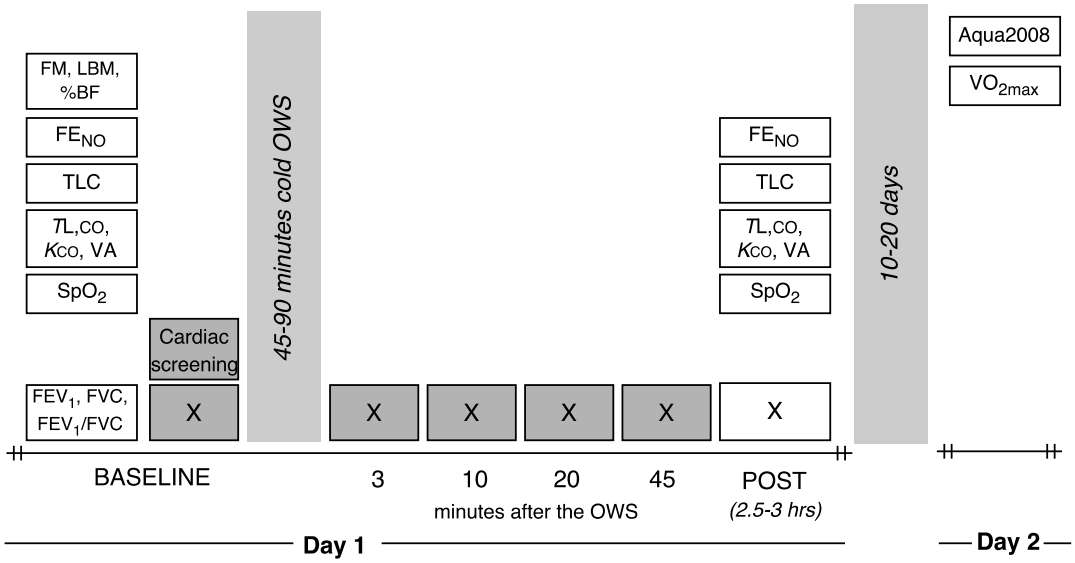

Figure 1 Timeline of test procedure, including assessed variables. Spirometry (FEV_1, FVC), pulse oximetry (SpO_2), alveolar gas diffusion for carbon monoxide (TL,CO, KCO, VA), whole-body plethysmography (TLC), online measurement of fractional nitric oxide (FE_{NO}) and dual-energy absorptiometry (DXA) were assessed at baseline and 2.5-3 hrs after the OWS (white boxes). Field spirometry was performed directly prior to, and repeated 3, 10, 20 and 45 minutes after, the OWS (grey boxes). On Day 2, the Aqua2008 questionnaire was completed and VO_{2max} was obtained.

[5] Figure 1. FEV_1, forced exhaled volume in one second; FVC, forced vital capacity; TLC, total lung capacity; SpO_2, percentage oxygen saturation of arterial blood; TL,CO, transfer factor for carbon monoxide; KCO, transfer constant for carbon monoxide; VA, alveolar volume; VO_{2max}, maximal oxygen uptake.

Figure 2

[6]

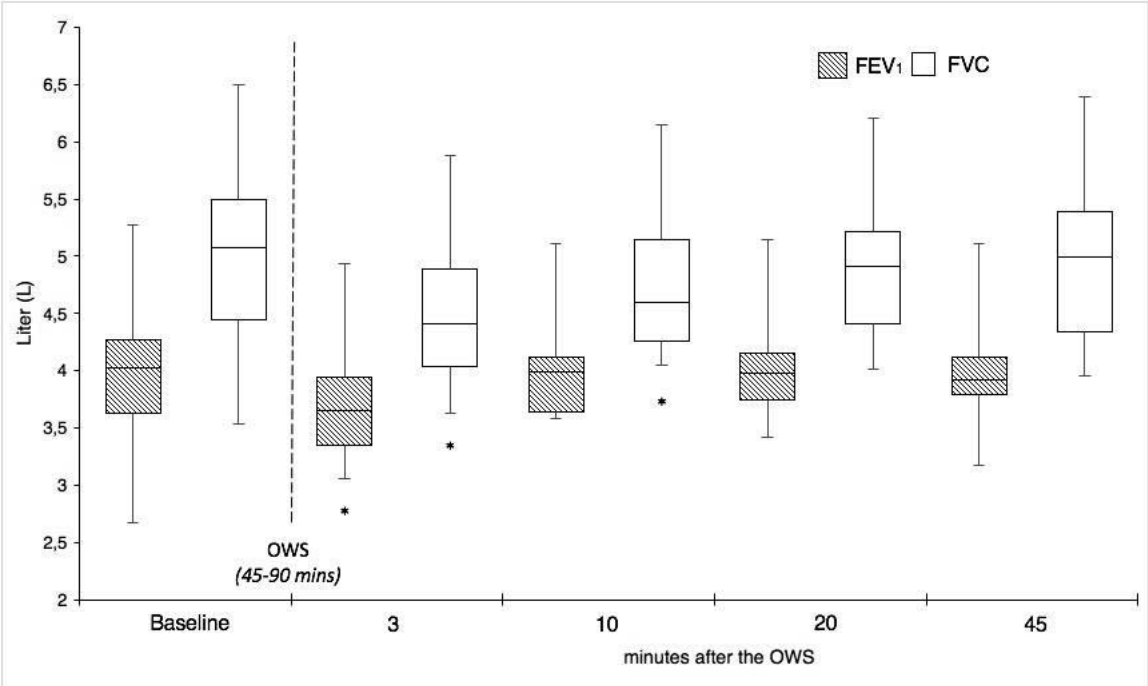

Figure 2 Immediate pulmonary responses in forced expiratory volume in one second (FEV_1; striped boxes) and forced vital capacity (FVC; white boxes)(n = 12). Boxes indicate median (25^{th}, 75^{th} percentiles) and whiskers indicate maximum and minimum values. Dotted line illustrating the OWS.

[6] Figure 2. *p <0.05, measurements significantly lower than baseline measurements in the respective variables. OWS, open water swim.

Figure 3a-f

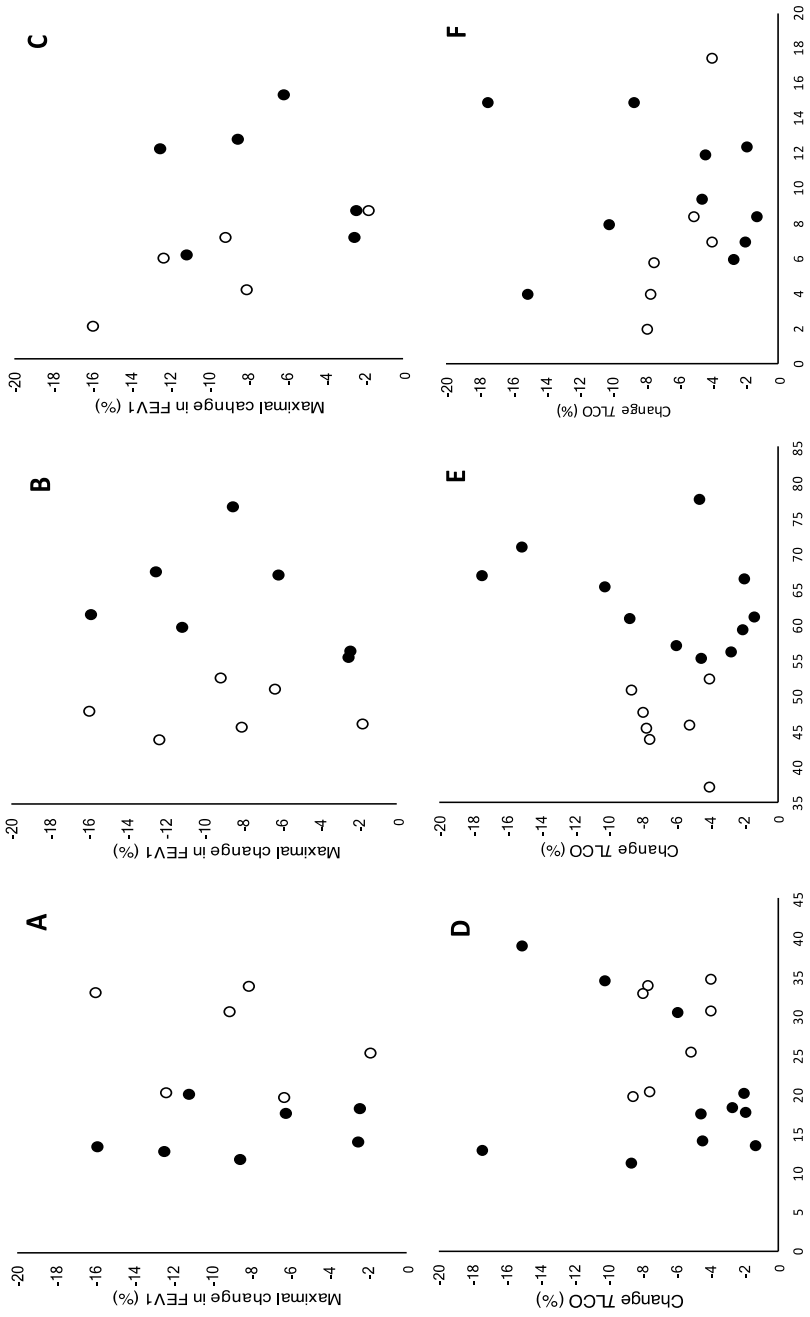

Figure 3. Scatter plots illustrating the relationship between maximal percentage decline in forced expiratory volume in one second (FEV_1; A, B and C, n = 12) and transfer factor for carbon monoxide ($TL_{,}CO$; D, E and F; n = 19) and percentage body fat (%BF), lean body mass (LBM) and weekly training volume. Black and white circles represent male and female participants, respectively. None of the variables were significantly correlated.

Figure 4

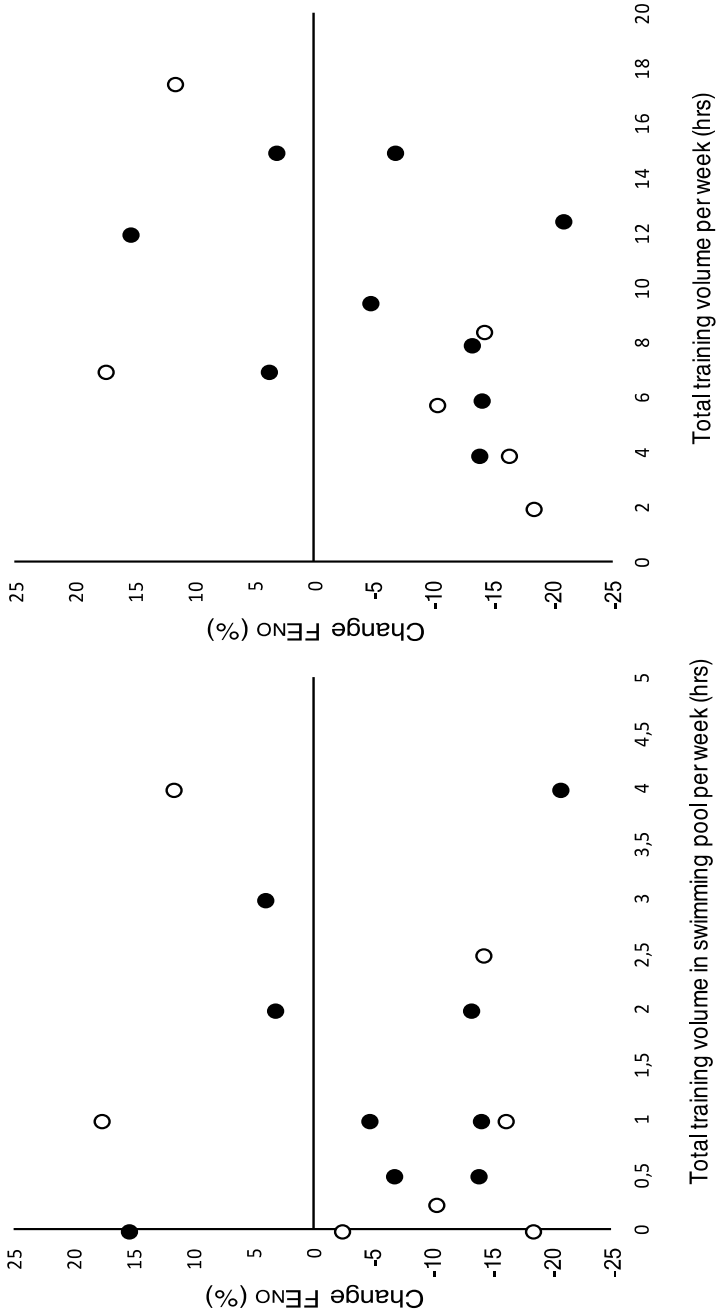

Figure 4. The relationship between percentage change in the fractional exhaled nitric oxide (FE_{NO}), used as a marker of eosinophilic airway inflammation, and total training volume in swimming pool per week (left) and total training volume per week (right). Black and white circles indicating men and women, respectively. None of the variables were significantly correlated.

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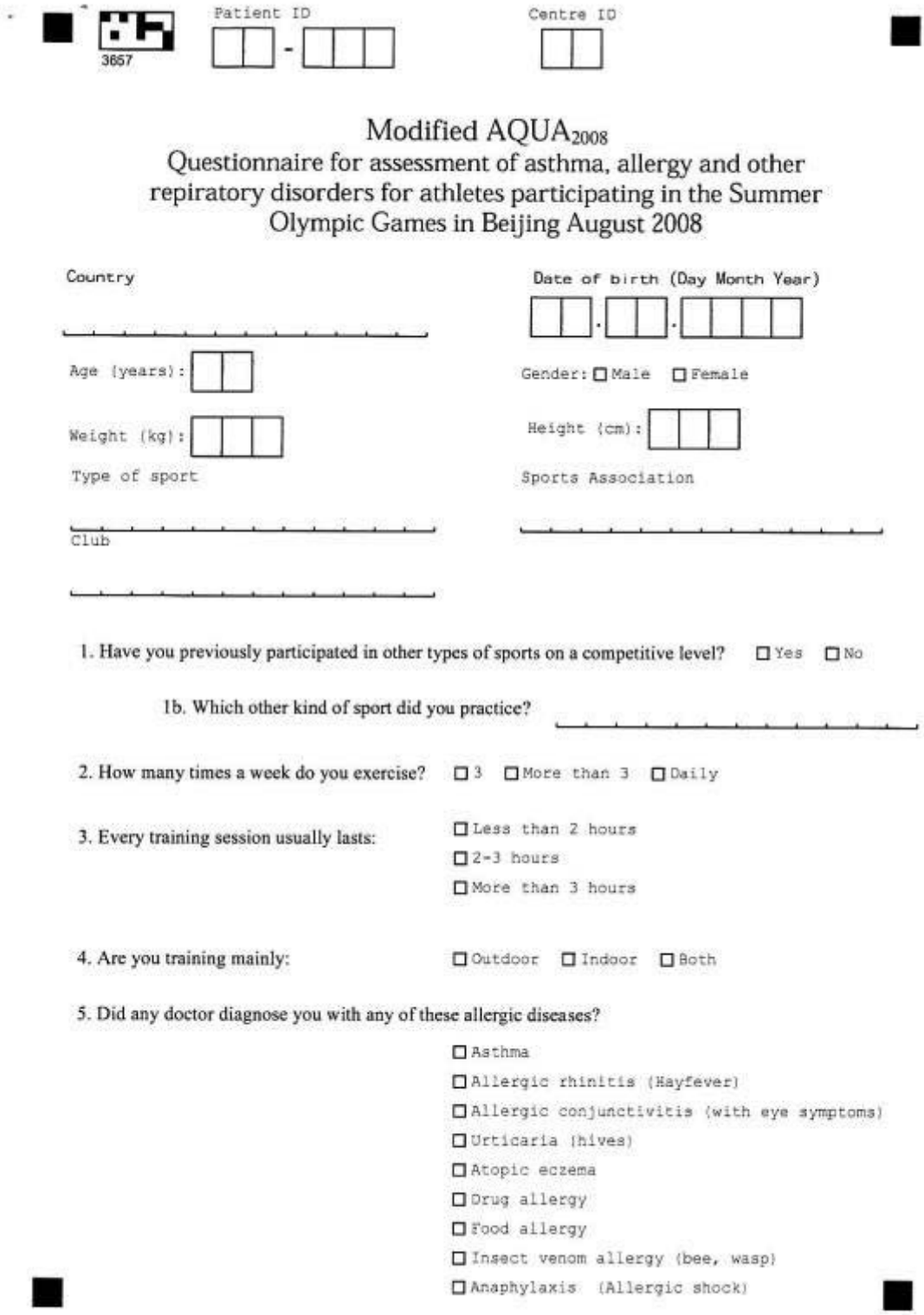
12 Appendices

12.1 Appendix 1 – Modified Aqua2008



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Patient ID ☐☐ - ☐☐☐

Centre ID ☐☐

Modified AQUA$_{2008}$

Questionnaire for assessment of asthma, allergy and other repiratory disorders for athletes participating in the Summer Olympic Games in Beijing August 2008

Country _____

Date of birth (Day Month Year) ☐☐.☐☐.☐☐☐☐

Age (years): ☐☐

Gender: ☐ Male ☐ Female

Weight (kg): ☐☐☐

Height (cm): ☐☐☐

Type of sport _____

Sports Association _____

Club _____

1. Have you previously participated in other types of sports on a competitive level? ☐ Yes ☐ No

1b. Which other kind of sport did you practice? _____

2. How many times a week do you exercise? ☐ 3 ☐ More than 3 ☐ Daily

3. Every training session usually lasts:
☐ Less than 2 hours
☐ 2-3 hours
☐ More than 3 hours

4. Are you training mainly: ☐ Outdoor ☐ Indoor ☐ Both

5. Did any doctor diagnose you with any of these allergic diseases?
☐ Asthma
☐ Allergic rhinitis (Hayfever)
☐ Allergic conjunctivitis (with eye symptoms)
☐ Urticaria (hives)
☐ Atopic eczema
☐ Drug allergy
☐ Food allergy
☐ Insect venom allergy (bee, wasp)
☐ Anaphylaxis (Allergic shock)

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6. Do you suspect that you suffer from allergy, independently of any medical diagnosis ? ☐ Yes ☐ No

7. Have you ever used anti-allergic or anti-asthma drugs ? ☐ Yes ☐ No

7b. If yes, which?
- ☐ Antihistamins
- ☐ Corticosteroids
- ☐ Bronchodilators
- ☐ Laukotrien antagonists (singulair)
- ☐ Allergy vaccines

8. Is there any allergic subject in your family? ☐ Yes ☐ No

8b. If yes, who?
- ☐ Mother
- ☐ Father
- ☐ Sibling(s) including half siblings
- ☐ Other relatives
- ☐ Children

9. Do you often have red eyes with tears and itching? ☐ Yes ☐ No

10. Do you often have runny, itchy nose (apart from colds): ☐ Yes ☐ No

11. Have you ever felt tightness in your chest and/or wheeze? ☐ Yes ☐ No

12. Have you ever had itchy skin eruptions? ☐ Yes ☐ No

13. Have you ever had severe allergic or anaphylactic reactions? ☐ Yes ☐ No

14. Have you ever had shortness of breath, cough and/or itching of the throat during or following exercise? ☐ Yes ☐ No

14b. If yes, you have more difficulties:
- ☐ At the beginning of the training session
- ☐ At the end of the training session
- ☐ During the whole training session

15. If you have suffered from any of the above, did these symptoms occur:
- ☐ Mainly outdoor
- ☐ Mainly indoor
- ☐ Indoor and outdoor equally
- ☐ Mainly in spring
- ☐ Mainly in cold or humid conditions
- ☐ All year around
- ☐ Independently of any environmental conditions

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16. Have you ever had allergic reactions to foods? ☐ Yes ☐ No

16b. If yes, do you remember to which food? _____

17. Have you ever had allergic reactions to drugs? ☐ Yes ☐ No

17b. If yes, do you remember to which drug? _____

18. Do you know that some drugs for allergic and respiratory diseases are prohibited or under restrictions by the World Anti-Doping Agency (WADA)? ☐ Yes ☐ No

18b. If yes, tick which substances, you think are included in this category:

- ☐ Antihistamines
- ☐ Bronchodilators
- ☐ Vasoconstrictors
- ☐ Topical corticosteroids (Nasal inhalers, eye droplets, dermatological preparations)
- ☐ Inhaled corticosteroids
- ☐ Injected or oral corticosteroids

19a. Do you think that anti-allergic and/or respiratory drugs may:

☐ Reduce performance ☐ Improve performance ☐ Don't affect performance

19b. Do you think that anti-allergic and/or respiratory drugs may be in conflict with anti-doping regulations? ☐ Yes ☐ No

20. Have you used more than three courses of any of these drugs during the last year? ☐ Yes ☐ No

20.b.. If yes, tick which category of drugs you did use:

- ☐ Antibiotics
- ☐ Anti inflammatory drugs
- ☐ Pain reducing drugs
- ☐ Drugs for reducing fever
- ☐ Others, which....

Patient ID ☐☐ - ☐☐☐

21. Have you used any other (except anti-asthma/anti-allergic) drug during the last week?

☐ Yes ☐ No

21 b. If yes, which drug? _____

22. Do you frequently suffer from upper respiratory infections (pharyngitis, colds, otitis media, tonsillitis, laryngitis) or fever?

☐ Yes ☐ No

22 b. If yes, are these infections more frequent during periods when you train more often than usual or during overtraining periods?

☐ Yes ☐ No

23. Have you suffered from recurrent labial herpes?

☐ Never
☐ 1-3 times
☐ More than 3 times

24. How many times during the last year were you unable to train because of infections?

☐ Never
☐ 1-3 times
☐ More than 3 times

25. If you have respiratory symptoms, which?

☐ Episodes of heavy breathing
☐ Wheeze
☐ Cough
☐ Phlegm, expectorate

26.Does this occur?

a. During exercise / training / competition: ☐ Yes ☐ No

b. During colds ☐ Yes ☐ No

c, After contact with animals, pollens, others: ☐ Yes ☐ No

27. With respiratory symptoms and dyspnoea related to exercise, when and how?

a. During maximum exercise ☐ Yes ☐ No

b. After the exercise: ☐ Yes ☐ No

c. In the afternoon, after training and/or competition: ☐ Yes ☐ No

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Patient ID

Additional questions for the modified AQUA 2008
(From the ECRHS II)

To compare the prevalence of symptoms among top athletes to figures of the general population, we would like to ask you some general questions about your respiratory health.

1. Have you had wheezing or whistling in your chest at any time in the last 12 months? IF 'NO' GO TO QUESTION 2, IF 'YES' GO TO QUESTION 1.1: ☐ No ☐ Yes

1.1. Have you been at all breathless when the wheezing noise was present? ☐ No ☐ Yes

1.2. Have you had this wheezing or whistling when you did not have a cold? ☐ No ☐ Yes

5. Have you been woken by an attack of shortness of breath at any time in the last 12 months? ☐ No ☐ Yes

14. Have you ever had asthma? ☐ No ☐ Yes

14.1 Was this confirmed by a doctor? ☐ No ☐ Yes

14.2 How old were you when you had your first attack of asthma? ☐ No ☐ Yes

14.5 Have you had an attack of asthma in the last 12 months? ☐ No ☐ Yes

14.10 Are you currently taking any medicines including inhalers, aerosols or tablets for asthma? ☐ No ☐ Yes

15. Do you have any nasal allergies, including hay fever? ☐ No ☐ Yes

74. Have you ever smoked for as long as a year? [‚Yes' means at least 20 packs of cigarettes or 12 oz (360 grams) of tobacco in a lifetime, or at least on cigarette per day or one cigar per week for one year.] ☐ No ☐ Yes

74.2 Do you now smoke, as of one month ago? ☐ No ☐ Yes

12.2 Appendix 2 – Participant information and written informed consent (Norwegian).

Forespørsel om deltakelse i forskningsprosjekt

”Hvordan påvirkes kjernetemperatur av 3800 m svømming med våtdrakt i 10 °C kaldt vann?”

Bakgrunn

Dette er et spørsmål til deg om å delta i et forskningsprosjekt som er et samarbeidsprosjekt mellom forskere ved Oslo Universitetssykehus, Olmpiatoppen og Norges Idrettshøgskole. Forskningsprosjektet støttes økonomisk fra Norges Triatlonforbund og Norseman Xtreme Triathlon. Triathlon er en idrett i sterk vekst. I Norge er det en utfordring for både utøvere og arrangører, at vannet i flere konkurranser kan være relativt kaldt. Vi ønsker å være med på å skape trygghet rundt arrangementene som arrangerer i områder med kaldt vann.

Hensikt

Hensikten med dette prosjektet er å måle kjernetemperatur hos utøvere som svømmer en lengde tilsvarende svømmeetappen på en langdistansetriathlon i 6°C - 12°C kald vann med våtdrakt godkjent for triathlon.

Hva innebærer studien?

Deltagelse i forskningsprosjektet vil innebære oppmøte på to forskjellige dager med inntil tre timer hver dag. Den ene dagen vil være medisinske og fysiologiske tester, samt en svømmetest i kaldt vann med våtdrakt. Tidspunkter for oppmøte vil i noen grad være mulig å tilpasse individuelt. Den andre dagen vil være en VO_2max test. Denne avtaler du helt som det passer for deg. Det er et mål for studien at vi har utøvere med variert bakgrunn innen triathlon. Vi krever av deg at du har en egen våtdrakt beregnet for svømming og at du kan gjennomføre en 3800 meter svømmetest i Songsvann.

Gjennomføring testing

Den medisinske og fysiologiske testen vil foregå på Idrettshøgskolens testlaboratorium ved Songsvann i Oslo. Her vil vi blant annet måle maksimalt oksygenopptak(VO_2max), muskel- og fettmasse og flere andre tester. Du vil også få et spørreskjema der vi vil kartlegge litt om din bakgrunn og erfaring innen triathlon.

Mulige fordeler og ulemper

- Du vil få en grundig gjennomgang av dine fysiologiske kapasiteter ved Idrettshøgskolen. Du vil også være med på å bidra til å gjøre triathlon til en tryggere idrett.
- Svømming i kaldt vann kan være ubehagelig. Vi har allerede gjort et pilotprosjekt, der vi ikke observerte noe nedkjøling på utøveren.
- Du vil være godt sikret under hele svømmetesten, med lege og/eller sykepleier med erfaring fra akuttmedisin og nedkjøling.

Avidentifisering og lagring av resultater

Informasjonen som registreres om deg skal brukes som beskrevet i hensikten med studien. Alle opplysninger vil bli behandlet uten navn og fødselsnummer eller andre direkte gjenkjennende opplysninger. Dine resultater vil bli avidentifisert og en kode vil bli benyttet til å knytte dine resultater og opplysninger til deg. Det er kun autorisert personell knyttet til prosjektet som har adgang til kode og navneliste. Resultatene vil bli lagret og bearbeidet på sykehusets forsknings-PC. Ferdigbehandlete data vil lagres i 10 år på Oslo Universitetssykehus sin forskningsserver og deretter slettes. Navnelisten med koblingsnøkkel oppbevares ved Oslo Universitetssykehus og vil bli slettet ved prosjektets avslutning. Det vil ikke være mulig å identifisere deg i resultatene av studien når disse publiseres.

Frivillig deltakelse

Det er frivillig å delta i studien. Du kan når som helst og uten å oppgi noen grunn trekke ditt samtykke til å delta i studien. Dersom du ønsker å delta, undertegner du samtykkeerklæringen på siste side. Om du nå sier ja til å delta, kan du senere trekke tilbake ditt samtykke uten at det påvirker din eventuelle øvrige behandling. Dersom du har spørsmål til studien, kan du kontakte oss.

Kontaktinformasjon

Dersom du er interessert, eller ønsker å vite mer om dette prosjektet kan du når som helst kontakte ph.d.-student/anestesisykepleier Jørgen Melau på e-post: jorgen@melau.no eller på telefon: 911 73 629 eller prosjektleder fysiolog dr. philos. Jonny Hisdal på e-mail: jonny.hisdal@medisin.uio.no eller på telefon: 92281977.

Vi ser frem til å høre fra deg!

Ytterligere informasjon om studien finnes i kapittel *A* *– utdypende forklaring av hva studien innebærer.* **Ytterligere informasjon om personvern og forsikring finnes i kapittel B** *– Personvern, økonomi og forsikring.*

Samtykkeerklæring følger etter kapittel B.

Kapittel A- utdypende forklaring av hva studien innebærer

Kriterier for deltakelse

- alder ≥ 18 år
- Du har en våtdrakt som er i god stand og beregnet for svømming
- Du er i stand til å svømme 3800 meter utendørs.

Prosjektet vil foregå i to omganger.

1. **Medisinsk og Fysiologisk test:** I forkant av svømmetesten vil du gjennomgå en legeundersøkelse, testing av fysisk kapasitet og kroppsammensetning. Disse testene vil bli utført på Idrettshøgskolen. Legeundersøkelse vil blir foretatt av en av de to

legene tilknyttet prosjektet, og de fysiske testene og måling av kroppsammensettning vil bli foretatt av kvalifisert personell på Idrettshøgskolen. Kroppsscanning gjennomføres på Idrettshøgskolen med en Dxa Scan maskin.

2. **Svømmetest:** Kjernetemperaturen måles kontinuerlig fra ca. 10 minutter før start og til 60 minutter etter at du har kommet opp av vannet. Det benyttes temperaturprobe som settes inn rektalt. Det måles også hudtemperatur på ulike steder på kroppen. Temperaturene lagres i en minnebrikke i ett belte som du har festet til brystet på innsiden av våtdrakten. Måleutstyret registrerer også klokkeslett, hudtemperatur og hjertefrekvens, slag for slag under hele forsøksperioden.
3. **Andre målinger:** Vanntemperaturen måles etter FINA´s retningslinjer i midten av svømmeløypa på 40 cm dyp. Vi benytter kalibrert thermometer for å male vanntemperatur.

PROSJEKTPROTOKOLL

Detaljert beskrivelse Del 1:

Du vil i forkant av Medisinsk, Fysiologisk og svømmetest bli bedt om å gjøre følgende forberedelser:

1. fastende minimum siste 8 timer før oppmøte
2. ingen trening eller koffein (kaffe, te, energidrikker) siste 24 timer
3. ingen bruk av tobakk (snus, røyk) ≥12 timer før testing
4. ingen hard styrke- eller utholdenhetstrening ≥24 timer før testing

Studien innebærer at du kommer til Idrettshøgskolen, Songsveien 228, Oslo om morgenen/formiddagen/ettermiddag/kveld (dette avtales nærmere). Hele testen er stipulert til å ta ~120 min. Ved oppmøte vil du bli forklart prosedyrene og bedt om å signere et dokument på at du har fulgt instruksene du har mottatt skriftlig i forkant av deltakelse i studien .

Dersom du aksepterer dette, vil vi ved oppstart og i etterkant av studien foreta målinger som følger:

1. Måling av DXA (Kroppsscanning)
2. Måling av lungefunksjon før svømming
3. Medisinsk sjekk
4. Oppkobling av puls- og temperaturmålere
5. Svømme 3800 m i Sognsvann
6. Testing av lungefunksjon like etter svømming
7. Avslutter målinger (60 min etter du har kommet opp av vannet)
8. Gjør avtale om når du skal teste VO_2 maks.

Kapittel B - Personvern, økonomi og forsikring

Personvern

Opplysninger som registreres om deg er demografiske data som kjønn, alder, medisinsk diagnose, medisiner, høyde, vekt, kjønn og fysiologiske data. I tillegg registreres kontinuerlig under svømmingen en rekke fysiologiske data (hjerteslag, pustefrekvens, temperatur) fra måleinstrumentene. Dataene kodes og lagres på forsknings-PC og forskningsserveren ved Oslo Universitetssykehus. Nøkkelen for å koble ditt navn til dine data oppbevares nedlåst og er kun tilgjengelig for medarbeidere i studien. Oslo Universitetssykehus ved administrerende direktør er databehandlingsansvarlig.

Utlevering av materiale og opplysninger til andre

Hvis du sier ja til å delta i studien, gir du også ditt samtykke til at avidentifiserte opplysninger utleveres til Oslo Universitetssykehus, Aker.

Rett til innsyn og sletting av opplysninger om deg og sletting av prøver

Hvis du sier ja til å delta i studien, har du rett til å få innsyn i hvilke opplysninger som er registrert om deg. Du har videre rett til å få korrigert eventuelle feil i de opplysningene vi har registrert. Dersom du trekker deg fra studien, kan du kreve å få slettet innsamlede prøver og opplysninger, med mindre opplysningene allerede er inngått i analyser eller brukt i vitenskapelige publikasjoner.

Økonomi og rolle

Studien er ikke finansiert. Det gis ingen kompensasjon for deltakelse.

Informasjon om utfallet av studien

Ved ønske, kan du som deltaker få tilsendt resultater av egne målinger. Dersom du ønsker informasjon om dine data fra studien kan du kontakte ph.d.-student/anestesisykepleier Jørgen Melau på e-post: jorgen@melau.no eller på telefon: 911 73 629 eller prosjektleder fysiolog dr. philos. Jonny Hisdal på e-mail: jonny.hisdal@medisin.uio.no eller på telefon: 92281977.

Samtykke til deltakelse i studien

Jeg er villig til å delta i studien

(Signert av prosjektdeltaker, dato)

Jeg bekrefter å ha gitt informasjon om studien

(Signert, rolle i studien, dato)

12.3 Appendix 3 – Picture permission

Til den det måtte angå.

Jeg gir med dette tillatelse til Camilla Illidi for å bruke bilder fotografert av meg i forbindelse med forskningsprosjekt på Høvik 27 til 29 oktober 2015. Jeg bekrefter at bildene er mine, og at Illidi kan benytte disse i forbindelse med sin Masteroppgave, til forelesninger og andre aktiviteter relatert til hennes deltagelse i forskningsprosjektet på Høvik.

Med vennlig hilsen

Jørgen Melau
Forsker Sykehuset i Vestfold
Tlf 91173629