University of Physical Education

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Dual Master's Programme in International Spots Coaching

Effects of Cooling and Magnesium on the Regulation of Physiological Mechanisms After Acute Physical Exercise

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ABSTRACT

Background: It is known that body temperature and imbalanced ion conditions can limit physical performance. To date, no investigations have evaluated the beneficial effects of varying cryotherapy interventions modalities and the acute magnesium iron supplementation in enhancing physical performance. We hypothesized that cryotherapy and magnesium supplementation will increase the physical performance and mental attitude of athletes.

Methods: Twenty-four recreational athletes (doing regular exercises) were recruited (n=24, all male): age 25±4 years, weight 76±10 kg, height 175±5 cm, were respectively. The experiment consisted of two parts: pre-test part, and experimental parts. In pre-test part, all subjects did 400 m rowing as homogeneity analysis divided into four groups (6 subjects/group), each group participated only one intervention. In the experimental part, there were four different interventions (① control group ② hand cooling ③ whole body cooling and ④ magnesium supplementation.) Each intervention had three sections, which before exercise interventions, acute high intensity 1000 m open-end rowing (until exhaustion) and recovery intervention period. During rowing, mean watt, speed, were collected in every 200 m. Before and after rowing the following blood plasma parameters were collected: NH_3, lactate. Heart rate (HR), tympanic temperature (Tear), whole-body temperature (Tw), and rate of perceived exertion (RPE by standard questionnaire) were also collected. At the end of exercise, subjects were asked to rate their perception of thermal sensation (TS) on a scale of 1 (cold) to 7 (hot) in 1 increments. All subjects were independent sample.

Results: The measured physiological parameters (lactate, NH_3, mean watt and rowing speed) were not significantly different ($p > 0.05$) between interventions, but the whole body temperature (Tw) decreased ($p < .00$, 29.7±0.70), thermal sensation (TS) decreased and the rate of perceived exertion (RPE) decreased significantly ($p < .05$).

Conclusions: Based on the results we suggest that cooling and magnesium supplementation - although did not improved the physiological parameters - but significantly decreased the body temperature and thermal sensation and perceived difficulty of exercise, which could be important in the prevention of drop out cases in sport and continue the training program.

Keywords: cooling, magnesium, thermoregulatory, thermal sensation

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ACRONYMS AND ABBREVIATIONS

HR	Heart rate
La	Lactate
La_{max}	Maximal lactate acid
NH_3	Blood ammonia
mmol/L	Millimole per liter
µmol/L	Micromol per liter
min^{-1}	Beat per minute
m	Meters
T_c	Core body temperature
T_w	Whole body temperature
T_{ear}	Tympanic temperature
T_a	Ambient temperature
Ts	Temperature sensation
RPE	Rate of perceived exertion
C	Control
HC	Hand-cooling
WBC	Whole-body cooling
MgS	Magnesium supplementation
R	Rest
R2	Before exercise
E1-E15	First to fifteen minutes after exercise

LIST OF TABLES

Table 1 Rate of perceived exertion (RPE)

6	
7	Very, very light
8	
9	Very light
10	
11	Fairly light
12	
13	Somewhat hard
14	
15	Hard
16	
17	Very hard
18	
19	Very, very hard
20	

(Borg, 1982)

Table 2 Scale of thermal sensation (TS)

1	Cold
2	Cool
3	Slightly cool
4	Neutral
5	Slightly warm
6	Warm
7	Hot

(Gagge, Stolwijk, & Hardy, 1967)

CHAPTER 1: INTRODUCTION

1.1 Background and significance

Physical activity is essential to maintain health, and – when it is intensive – to achieve increased performance in athletes. There are many different types and modes of physical training to athletes and one of the main problem of exercise physiology is that how can be optimize and improve the effect of training in athletes by appropriate coaching. Physical preparation of sports science play an important role in training session especially in taper period, to understanding the physiological and performance elements of the taper combing research and practical experience of coaches and athletes, experimental and observational research, and elegant mathematical models (Pyne, Mujika, & Reilly, 2009). In addition to various training methods, there were several interventions used and tried to further enhancing the performance of athletes.

Different type of interventions shows different effect. The threshold-training model emerges improvements among untrained subjects training at their lactate threshold intensity. In contrast, a polarized-training model emerges from a limited number of published observations of some athletes (Stephen & Øvrevik, 2006). Neuromuscular training protocols can significantly improve biomechanics and neuromuscular performance and reduce anterior cruciate ligament injury risk in female athletes (Myer, Ford, Brent, & Hewett, 2006), etc. High intensity exercise due to increased muscle activity the cellular metabolism, such as glycolysis, terminal oxidation and oxidative phosphorylation increases eliciting heat production. Exercise induces a prothrombotic state, which might be partially relying on the magnitude of the exercise-induced core body temperature rise (Veltmeijer et al., 2017). To maintain an optimal core temperature dissipation of heat is an important issue.

On the basis of ideas, several research have been conducted with the use of a hand cooling device equipment (RTX CoreControl, AVAcore Inc., CA, USA) in order to lower core temperature. Adams et al., (Adams et al., 2016) used the vacuum seal as hand cooling showed that it may have been too great to exhibit a greater cooling effect. Another effort was the use of palm cooling, which are still unclear and future studies are needed to prove its beneficial effects (Bongers, Hopman, & Eijsvogels, 2017). Bongers et al., (Bongers et al., 2017) summarize that cold water immersion delayed the onset of muscle soreness and the subjective outcome (rate of perceived exertion) but did not impact on objective outcomes and physiological parameters. In contrast, it has been shown that cryotherapy can delay only onset of the muscle soreness in subjective outcomes, but importantly it increased the muscle strength and decreased the inflammatory response in exercise.

Unlike whole body cryotherapy, partial body cryotherapy (which is WBC in my research) cooling without subject’s head used primarily in studies of exercise recovery (Bouzigon, Grappe, Ravier, & Dugue, 2016). Athletes (all levels), active participants (moderate level of sport activity) as well as common people can use this therapy

(Bouzigon et al., 2016). The stimulation of the sympathetic nervous system, the release of noradrenalin and the vasoconstriction during and after the cold exposure may also have a significant impact on pain and joint and/or muscle soreness (Bouzigon et al., 2016; Leppäluoto et al., 2008).

Bouzigon et al., (Bouzigon et al., 2016) suggests that after exercise both single and repeated short-term treatments with whole body cryotherapy or partial body cryotherapy both have beneficial effects on recovery. Whole body cryotherapy or partial body cryotherapy affected inflammatory response, muscle damage recovery, autonomic nervous system, quality of sleep and oxidative stress. Others (Fonda & Sarabon, 2013) used a 3-min exposure at -140°C to -195°C partial body cryotherapy protocol, which elicited in athletes higher squat jump start power and maximal torque production and torque rate of development compared to control group. Power output was adjusted well before reaching the critical limiting core body temperature (Bongers et al., 2017), but retaining a large core to skin temperature gradient may be even more important than keeping Tc below the critical Tc to preserve exercise performance (Bongers et al., 2017). One of the most important issues in my thesis is to find the optimal cooling strategy combined with acute exercise.

The exact physiological mechanisms of pre-cooling underlying possible performance improvements, practical guidelines and their interaction are still not well -characterized (Marino, 2002; Wegmann et al., 2012). Wegmann et al., (Wegmann et al., 2012) review described that pre-cooling is barely affected in sprint exercise. Most studies are used endurance protocol in the past. Although (Wegmann et al., 2012) indicates that in sprint exercise protocols pre-cooling intervention does not showing positive effect, but their review did not refer to cryo-cabin treatment. However, others (Bongers et al., 2017; Tyler, Sunderland, & Cheung, 2015) pointed out that pre-cooling can significantly improve intermittent sprint exercise and endurance exercise performance but in single sprint performance. That might be due to the fact that sprint exercise is mainly influenced by muscle temperature and anaerobic metabolism, rather than thermoregulatory factors (Girard, Brocherie, & Bishop, 2015; Tyler et al., 2015). In the past, most literature selected a workload with long lasting training protocol. In this paper, the acute model as rarely used in experiment's training protocol, is aim to shorten the time for athletes to intervene during the competition and training so as to achieve greater economic benefits. To find not only the effects of cooling intervention on endurance exercise training, but also sprint paired with different interventions. Although it is not completely clear in specific sports, we previously hypothesized that different-trained athletes will also benefit from this. Bongers et al., (Bongers et al., 2017) pointed out longer durations do not affect thermal and cardiovascular responses, but increase thermal discomfort of the participants. The acute exercise-induced response during heavy resistance exercise also play an important role in the long-tern anabolic adaptation (Linnamo, Pakarinen, Komi, Kraemer, & Häkkinen, 2005). They actually have an an across effect. Due to the inhomogeneous methodology used in the respective studies (Wegmann et al., 2012). The blood lactate has been shown to increase more when the number of repetitions is high with high loads than when loads are lower (Linnamo et al., 2005).

Regardless of sprint or endurance exercise, if the interventions help to accelerate elimination of lactate in athletes's plasma accumulation. Whatever different kind of protocols use should depend on the goal of training.

Several studies used oral menthol application (T, 2010; Valente et al., 2015) as intervention, but there has been no study which used magnesium as potential intervention to increase the performance of athletes. In Nielsen's (Nielsen, 2006) paper was shown that exercise induces a redistribution of magnesium in the body to accommodate the metabolic needs. Notwithstanding some studies have found that only magnesium-deficient athletes can improve their performance after supplementation with magnesium, but normal magnesium supplementation with magnesium cannot improve performance. But still magnesium is involved in numerous processes that affect muscle function including oxygen uptake, energy production (ATP and phosphocreatine synthesis), and electrolyte (sodium, potassium, and calcium) balance (Nielsen, 2006). World health organization (World Health Organization. Butte, Lopez-Alarcon, & Garza) has also pointed out the nutritional essentiality and benefits from sufficient dietary intakes of calcium and magnesium are well established but quantitatively imprecise. According to Nielsen (Nielsen, 2006) paper, significant number of physically active people apparently would find it beneficial to increase their intake of magnesium not only athletes. Byrne et al., (Byrne, Owen, Cosnefroy, & Lee, 2011) observations show that during rest, ingestion of cold fluid over 35 to 5 minutes before exercise a greater decrease in rectal temperature than the control can be achieved beneficial. The problem of how many milligrams of daily magnesium intake are optimal for athletes in a day is a burning issue. Average intake that would result in magnesium-deficient status for the average person and especially, for athletes is not yet clear (Nielsen, 2006). Athletes who are doing lot of exercise cannot obtain sufficient amount of magnesium directly from the food, especially if they consume food with the lower levels of magnesium (such as protein and refined carbohydrates). Thus, it is effective and practical to take supplemental magnesium before exercise.

Interestingly, hot environment also reported to be connected with magnesium loss. Losses of magnesium via sweeting is about ten-to-one in some environment (Consolazio, Matoush, Nelson, Harding, & Canham, 1963). Combined with the magnesium above. Elite athletes have a better adapted thermoregulatory system compared with untrained subjects (Cheung & Mclellan, 1998; Edwards & Clark, 2006; Wegmann et al., 2012). Nunneley et al., (Nunneley, Jr, & Webb, 1971) previously hypothesized that cooling's will only effect at ambient temperatures reaching about 40°C. For the normal sport program, which cannot meet this temperatures requirement, whether cooling intervention can be still be beneficial worth to be noticed .

Thus, based on the conclusion above my research the aim is to figure out if take magnesium effect of acute exercise that interfering with body temperature and increase performance.

1.2 Hypothesis

On the basis of aforementioned, in this study, we hypothesized that during and after physical exercise NH_3 and core body temperature (Tc) would be alleviated by pre-cooling and magnesium supplementation. As a result, plasma lactate concentration will be reduced and the peak of lactate concentration delayed during exercise and these methods also increases the physical performance.

1.3 Aims

Thus the aims of our studies were:

1) To measure the blood plasma (lactate and NH_3) effects of the different cryotherapy and supplemental magnesium by acute exercise.

2) Investigate different recovery interventions like cryotherapy and supplemental magnesium applied on heart rate regulation and other psychological parameters.

1.4 Definitions

Core body temperature

Core body temperature measurements near the eardrum (called tympanic temperature) have been found to be a good estimate of the actual brain temperature (Powers & Howley, 2007). The whole body temperature can be calculated by assigning certain factors to each individual skin measurement in proportion to the fraction of the body's total surface area that each measurement represents. Estimated by the following formula:

$$T_w = (T_{neck} + T_{clavicle} + T_{abdomen} + T_{thigh}) \div 8$$

Where T_{neck}, $T_{clavicle}$, $T_{abdomen}$ and T_{thigh} represent skin temperatures measured on the nech, clavicle, abodmen and thigh respectively. Combine with temperature sensation, tympanic temperature and whole body temperature, can reflect actual trends of core temperature in before exercise and after.

Magnesium

In this context, magnesium is mineralstoffe product form German: Magnesium 400 + C + E Brausetabletten (Kneipp). Should be taken orally with 250ml water.

Rate of Perceived Exertion, RPE

RPE was evaluated using the 6 to 20 point Borg scale (Borg, 1982; Klionsky et al., 2016), to expressed their physical exertion.

Temperature sensation, TS

(Gagge, Stolwijk, & Saltin, 1969) used 7 scales indicate the temperature of body 1 - Cold, 2 - Cool, 3 - Slightly cool, 4 - Neutral, 5 - Slightly warm, 6 - Warm and 7 - Hot. Mark at rest time (R), before exercise (R2), E1, E3, E6, E7 and E15, separately.

CHAPTER 2: LITERATURE REVIEW

2.1 Human physiological response in thermal environment

Maintaining constant core body temperature is essential to maintaining life of humans. Therefore several multilayer mechanics are involved in this process. Among others the parasympathetic and sympathetic nervous system controls skin temperature and blood flow and perspiration, thereby controlling heat preservation and heat loss (Plowman & Smith, 2007). Similarly, brown fat (mitochondria rich) is also important in body temperature regulation just like the skin micro-vessels (Kozak, Koza, & Anunciado-Koza, 2010). During our life there are many temperature challenges are exerted to our body.

The thermoregulatory control loop is divided into two cooperating subsystems: a passive system representing the thermal properties of the body with a mean body temperature which is influenced by incidental exogenous and endogenous thermal disturbances; and an active one consisting of the controlled processes of cold and heat defense (metabolic heat production M, evaporative heat loss E, and by the control of heat flow by peripheral vasomotor tone, PVMT) (Braun, Brück, & Heldmaier, 2012).

Adaptation to higher environmental temperature elicited by: 1) an increase in plasma volume, 2) an earlier onset of sweating during exercise, 3) a higher sweat rate, 4) a reduction in the amount of electrolytes lost in sweat, and 5) higher in skin blood flow (Powers & Howley, 2007). Cooling down the body temperature is necessary in order to avoid the heat shock. Elevated body temperature causes hyperventilation leading to hypocapnia and consequently reduced cerebral blood flow. A lower oxygen partial pressure causes fatigue, cerebral dysfunction and muscular motor dysfunction (Wegmann et al., 2012)

2.2 Characteristics and energy metabolism during rowing

2.2.1 Energy metabolism and rowing patterns

It is now that exercise, such as rowing need high level of adenosine triphosphate (ATP) to supply the contractile activity of skeletal and cardiac muscles. ATP derives from both aerobic and anaerobic energy systems during the cellular/mitochondrial respiration (Laursen, 2010).

There are several rowing patters which can be measured by ergometers developed in an attempt to reproduce movement and resistance of on-water rowing (Mahony, 1999). Rowers during the general preparation phase, which is predominated by aerobic exercise and probably insufficient to induce changes in TNF-α concentration (Skarpańska-Stejnborn, Basta, Trzeciak, & Szcześniak-Pilaczyńska, 2015) .

2.2.2 Adaptation to acute physical exercise and regulation of physiological mechanism

Short-term, high-intensive exercise is mainly effect the cardiovascular and respiratory systems (Wegmann et al., 2012). For an intense exercise event that lasts beyond 75 seconds, total energy output is mostly aerobically driven (Laursen, 2010) and also improves the ability of the engaged skeletal muscle to generate ATP aerobically (Laursen, 2010). High-intensity training and high-volume training can elicit improvements in the aerobic power and capacity (Laursen, 2010). Whereas, the 30 seconds sprint (17%) had increased the speed endurance (Iaia et al., 2009).

When training does not have an appropriate blend of both high-intensity training and high-volume training inserted into the program, performance ability will not improve (Laursen, 2010). Coaches should avoid making every session the same intensity to avoid stagnation and staleness (Estevelanao, Foster, Seiler, & Lucia, 2007).

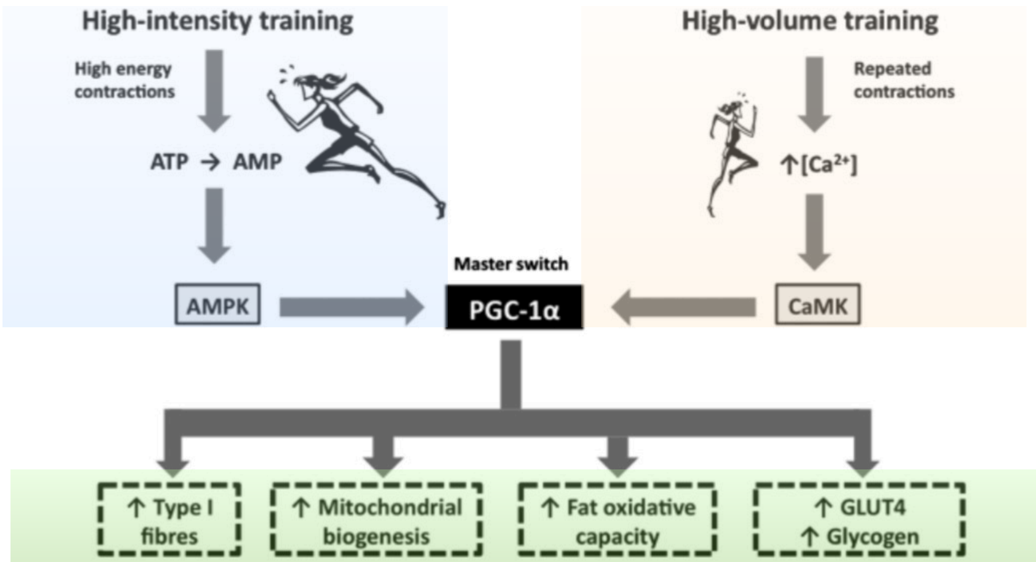

Figure 2-2-2-1 Effects of high intensity training on cellular signaling and fiber types, mitochondrial function, fat acid oxidation and glucose metabolism.

2.3 Discussion of two different cooling methods

In the past, several body cooling methods have been applied in order to reduce body temperature and improve performance (Wegmann et al., 2012).

2.3.1 Hand-cooling

Our hands have a large surface to mass ratio and low metabolic heat production that suit high rates of conductive heat transfer (Ruddock, Tew, & Purvis, 2017). It has been shown that pre-cooling with cold-water immersion facilitated post-exercise recovery enhanced the rate of heat storage, and caused less thermoregulatory strain-attenuated rise in skin and rectal temperatures and heart rates during physical activity (McArdle, Katch, & Katch, 2010).

Exercise in a cold environment enhances the athletes's ability to loose more heat and therefore greatly reduces the chance of heatstroke. Higher intermittent blood flow to the hands and feet is one of the adaptations to cold outside temperature. The overall goal of this adaptation is to increase heat production and maintain core temperature which will make the individual more comfortable during cold exposure (Powers & Howley, 2007). Most investigations have attempted to reduce the core temperature and so have cooled the torso and/or lower limbs because there is a larger surface area and therefore greater potential for heat exchange (Tyler et al., 2015). On the other hands, hand cooling may be a most convenient method to athletes, practicality and economy of the cooling way needs to be consider. The practical application at present is limited (Marino, 2002), hand-cooling device may provide additional benefits beyond passive rest. In halftime and intermission periods where full body immersion is too cumbersome, but hand cooling can be easily applied to further decrease body temperature. Additionally, in situations where multiple bouts of exercise are separated by rest, hand cooling could potentially mitigate a rise in body temperature (Adams et al., 2016). Hand cooling provide an easy way to achieve the aim. There is a latest US-developed hand cooling device on the market, and the results show that using this device before exercise is more effective for anaerobic exercise (Krishnan, Singh, Sharma, Upadhyay, & Singh, 2018), also expresses the reason why we apply this method during our investigation.

2.3.2 Body cryotherapy

Strenuous exercise induces the release of the C-reactive protein (CRP) and an increase in the pro-inflammatory cytokines TNF-α, and interleukins (IL), such as IL-1β, IL-2 and IL-8 and IL-6 in the circulatory system (Bouzigon et al., 2016).

Parasympathetic nervous system activity is associated with motor recovery and reduces the risk of cardiovascular events. The sympathetic nervous system accelerates the heart, (increasing heart rate) via neural and humoral pathways (catecholamine), while the parasympathetic branch decelerates the heart rate through neural pathways (Bouzigon et al., 2016).

In addition to the above mentioned biological changes, pre-cooling has been shown to effect the scales of thermal sensation and/or the ratings of perceived exertion (Wegmann et al., 2012). A review (Marino, 2002) reported that pre-cooling is only beneficial for endurance exercise of up to 30 – 40 minutes rather than intermittent or short duration exercise. Whole body pre-cooling provided no thermoregulatory benefit during a simulated triathlon or on the physiologic responses to a 90 minutes soccer-specific activities protocol under normal environmental conditions (McArdle et al., 2010). Tyler et al. (Tyler et al., 2015) summarized that cooling is effective on exercise of a long period time and not on short-duration, high-intensity exercise. For now, more and more coaches applies the high intensity programs into their training program. This is also a way to improve the anaerobic capacity of athletes, owing to the short duration of a sprint, performance is influenced by muscle temperature, contractile function and/or anaerobic metabolism efficiency rather than cardiovascular or thermoregulatory factors (Tyler et al., 2015). The difference is due to various demands of the protocols, the most effectiveness of cooling intervention for the enhancement of performance or capacity in a hot environment is dependent on the thermal strain experienced and the magnitude of cooling applied. Previous report (Duffield & Marino, 2007) showed that only high intensity exercise can reach thermal stress that pre-cooling can reaction.

At the present, a larger pre-cooling effect was found compared with sprint or intermittent sprint (Wegmann et al., 2012). Tyler et al., (Tyler et al., 2015) made an analysis on the pre-cooling data by dividing the data set into three groups: 1) cooling prior to a single sprint of < 70 seconds (primarily anaerobic in nature), 2) cooling prior to a test using intermittent sprints (mixed energy system) and 3) cooling prior to prolonged (> 6 minutes), primarily aerobic exercise. Castle et al., (Castle et al., 2006) investigated twelve male team-sport players completed four cycling intermittent sprint protocols. Obviously, pre-cooling has been shown to increase time to exhaustion or increase the distance run or cycled (Marino, 2002). Marsh et al., (Marsh & Sleivert, 1999) found that up to 30 minutes of pre-cooling improved performance by approximately 3.3% compared with control conditions. They speculated that the cold induced vasoconstriction of the skin and a concomitant increase in central blood volume, possibly increasing muscle blood flow and metabolite removal, thereby sustaining higher exercise intensity. Pre-cooling can increase in central blood volume lead to improved perfusion of the working skeletal muscles caused by peripheral vasoconstriction due to reduced skin temperature (Wegmann et al., 2012). Using cooling modalities with greater cooling rates will allow for greater reductions in body temperature after and between bouts of exercise (Adams et al., 2016). With such different experimental interventions it is important to find the most efficient protocol to apply in the future training, required to identify optimal cooling strategies for different events taking into account the level of thermal strain experienced (Tyler et al., 2015) and also be individualized for the specific athletes and sport activity.

Post-cooling is defined as any opportunity to reduce the core, skin and/or muscle temperature directly after exercise, to enhance recovery from exercise and to reduce the exercise-induced muscle soreness (Bongers et al., 2017). The effects of post-cooling applied directly after exercise improves subjective recovery, since it lowers the symptoms of delayed onset muscle soreness after 24 and 96 hours of recovery (Bongers et al., 2017; Hohenauer, Taeymans, Baeyens, Clarys, & Clijsen, 2015).

Cold water immersion (5 – 15°C) appeared to be more effective compared to the other post-cooling strategies (cold air exposure, cooling packs and cryotherapy). (Hohenauer et al., 2015) pointed out similar benefits were observed across different post-cooling ways. Also, there was no evidence that post-cooling had an impact on objective recovery outcomes such as blood lactate, creatinine kinase and c-reactive protein concentration. Post-cooling may enhance recovery from strenuous exercise by reducing the intramuscular temperature and muscle metabolism to reduce the metabolic stress and reactive oxygen species (ROS) generation associated with muscle damage, while local vasoconstriction may reduce the formation of edema, the inflammatory response and the associated secondary muscle damage. In this process superoxide dismutase (SOD) a magnesium dependent enzyme can exert great protective effect. The subjective pain response to muscle soreness may be diminished by a cooling-induced decrease in nerve conduction velocity (Bongers et al., 2017).

2.4 Cardiovascular and metabolic mechanisms

Cooling reduces the stress on the metabolic and cardiovascular system (Reilly, Drust, & Gregson, 2006). Core temperature reductions may inhibit blood lactate accumulation and increase the lactate threshold (Bongers et al., 2017; James, Richardson, Watt, Gibson, & Maxwell, 2015).

Temperature changes can also influence cardiovascular function. Due to the redistribution of blood from the core to the periphery (increased skin perfusion due to vasodilatation of the blood vessels of the skin to improve heat dissipation), an adequate (higher) cardiac output is maintained by an increased heart rate. Cardiac stroke volume is decreased with increasing core body temperature, because the reduced central blood volume leads to a lower filling pressure, and diastolic filling time is reduced due to a higher heart rate (José Gonzálezalonso & Calbet, 2003). In this context, the arteriovenous anastomoses of the skin play an important role, as they are opened with increasing body temperature and create a short cut between arteries and veins. Blood flowing through these cross connections does not flow through capillaries and can thus not be used for exchange of nutrients and oxygen. With respect to metabolism, lactate and glycogen depletion do not appear to be a limiting factor for sub-maximal exercise in the heat (J Gonzálezalonso et al., 1999; Wegmann et al., 2012)

2.5 Effect of magnesium on sports

Magnesium is involved in numerous processes that affect muscle function including oxygen uptake, energy production and electrolyte balance (F. H. Nielsen & Lukaski, 2006). Exercise induces a redistribution of magnesium in the body to accommodate metabolic needs. Magnesium-deficient individuals can have beneficial effects on magnesium supplementation or increased dietary intake of magnesium also in exercise performance. Individuals engaged in both short-term high-intensity and long-term strenuous exercise may become deficient at 10 - 20% higher intakes (see Table 2-5-1) because of increased losses of magnesium through sweat and possibly urine (Nielsen, 2006). Lukaski (Lukaski, 2001) found that exercise resulted in a redistribution of magnesium in the body and the type of exercise and magnesium status influenced the nature of this redistribution. Setaro et al., (Setaro et al., 2014) reported that magnesium supplementation improved low lactate level anaerobic metabolism, even though the athletes were not magnesium-deficient. Magnesium is mobilized from bone, as well as soft tissues and muscle, the change in extracellular magnesium apparently reflects that the body is responding to exercise by redistributing magnesium to locations with increased metabolic need for example for energy production and counteracting oxidative stress (like SOD) (Nielsen, 2006). Rowing athletes participating in extended and intensive training loads also exhibited increased urinary magnesium excretion (Hartmann, Mader, Böhnert, Wagner, & Hollmann, 1996; Nielsen, 2006). Nine well-trained male and nine well-trained female athletes, a significant decrease in both ionization and total magnesium immediately after a single bout of exercise (Terink et al., 2017). We can known that an individual routinely participating in strenuous exercise has a magnesium demand 10 - 20% above the average inactive person of the same age and sex (Nielsen, 2006).

Elevated the lactic acid production leading to a metabolic acidosis which causes magnesuria also has been suggested to be responsible for decreased tubular reabsorption in the kidney (Nielsen, 2006). Rock et al., (Rock et al., 1995) show that free radical-mediated processes may also be involved in skeletal muscle lesions of mg-deficient rats as it s known that superoxide dismutase (SOD) can function properly only in the presence of Mg^{2+}. Thus mg-deficiency can induce an oxidative and inflammatory state (Rock et al., 1995). Also, exercise has been found to amplify the decrease in erythrocyte magnesium concentration in marginally magnesium-deficient rats (Lowney, Stern, Gershwin, & Keen, 1990). In animal experiments, magnesium-deficient weak at coupling between oxidation and phosphorylation, skeletal muscle magnesium and serum magnesium decreased was associated with a partial uncoupling of the respiratory chain and a reduced ADP to oxygen ratio in rat liver mitochondria (Heaton & Elie, 1984). The physiological processes involved in reactive oxygen species production during exercise has been reviewed by (Laires & Monteiro, 2001). The effect of exercise on immune function also has been reviewed (Konig, Weinstock, Keul, Northoff, & Berg, 1998; Laires & Monteiro, 2001).

Table 2-5-1 Recommended Dietary Allowances (RDAs) for Magnesium

Age	Male	Female
Birth to 6 months	30 mg*	30 mg*
7–12 months	75 mg*	75 mg*
1–3 years	80 mg	80 mg
4–8 years	130 mg	130 mg
9–13 years	240 mg	240 mg
14–18 years	410 mg	360 mg
19–30 years	400 mg	310 mg
31–50 years	420 mg	320 mg
51 + years	420 mg	320 mg

**Adequate Intake*

Early studied have shown that taking magnesium tablets can help reduce depression (Eby & Eby, 2006). People with magnesium-deficiency are associated with many pathological and psychological disorders (Medicine, Board, Research, & Committee on Optimization of Nutrient Composition of Military Rations for Short-Term, 2006).

Molina et al., (Molina-Lopez et al., 2012) resulted show that RPE correlated significantly with erythrocyte magnesium levels and also tended to preserve changes in mineral levels during training and competition. In the past studies (Veasey et al., 2015) investigated that took Guaraná (MVM + G) (containing 100 mg of magnesium) before moderate exercise increased the cognitive function as well as prevented damage of physical and psychological outcomes, and significant impact of reduced the RPE after the exercise. Although the mental fatigue can be alleviated in some way because the caffeine component of Guaraná, the authors also believe it is not excluded that the magnesium (100 mg) have an auxiliary effect.

On he basis of the aforementioned studies we hypothesized that cryotherapy and magnesium supplementation will increase the physical performance and mental attitude of athletes.

Thus the aims of our studies were:

1) To measure the blood plasma (lactate and NH_3) effects of the different cryotherapy and supplemental magnesium by acute exercise.

2) Investigate different recovery interventions like cryotherapy and supplemental magnesium applied on heart rate regulation and

On the basis of aforementioned, in this study, we hypothesized that during and after physical exercise NH_3 and core body temperature would be alleviated by pre-cooling and magnesium supplementation. As a result, plasma lactate concentration will be reduced and the peak of lactate concentration delayed during exercise and these methods also increases the physical performance.

CHAPTER 3: METHODS

3.1 Subjects

In the present study 24 recreational athletes were recruited from different backgrounds of exercise activities. The mean age 25±4 years, weight 76±10 kg, height 175±5 cm, were respectively. Based on the rowing power output participants were evenly assigned to controls (no intervention) (n = 6) group 1, hand cooling group 2 (n = 6), whole body cooling group 3 (n = 6), and magnesium supplementation group 4 (n = 6). All the subjects were informed regarding the experimental process and they signed the letter of consent. The ethical approves was given by the Fu Jen Catholic University Institutional Review Board

3.2 Experimental Procedure

The following chart illustrates the experimental procedure and logic.

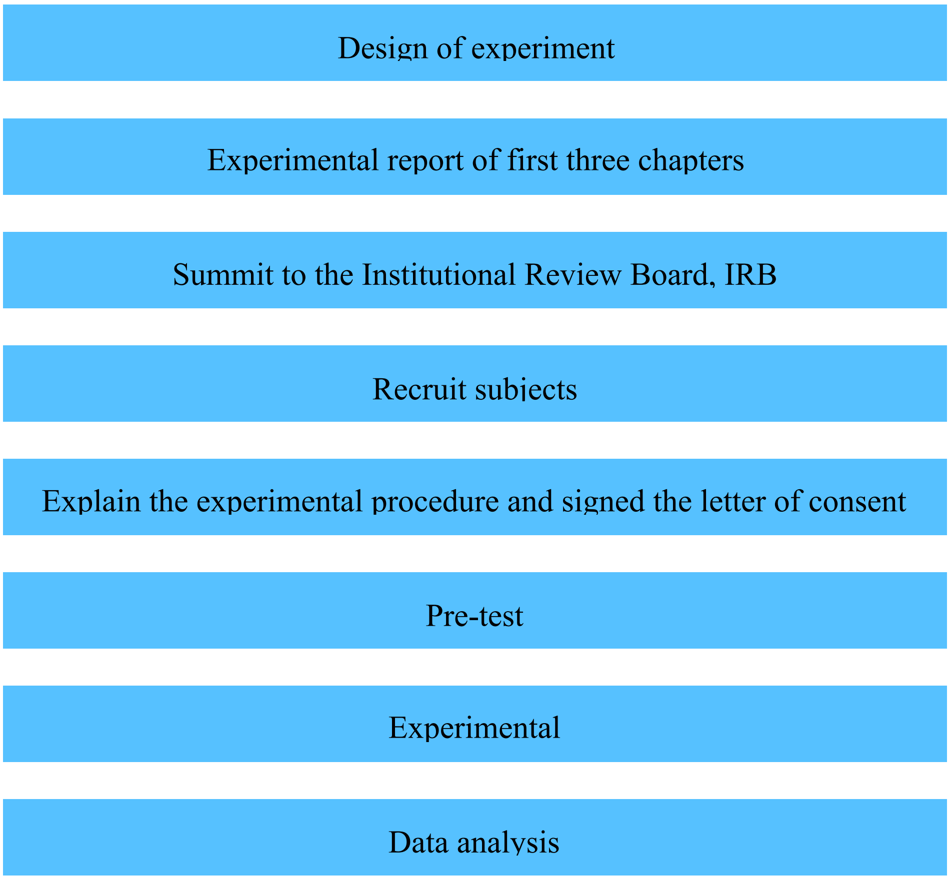

Figure 3-2-1 Experimental Procedure

1. Pre-test - 400 m rowing

2. Experiment

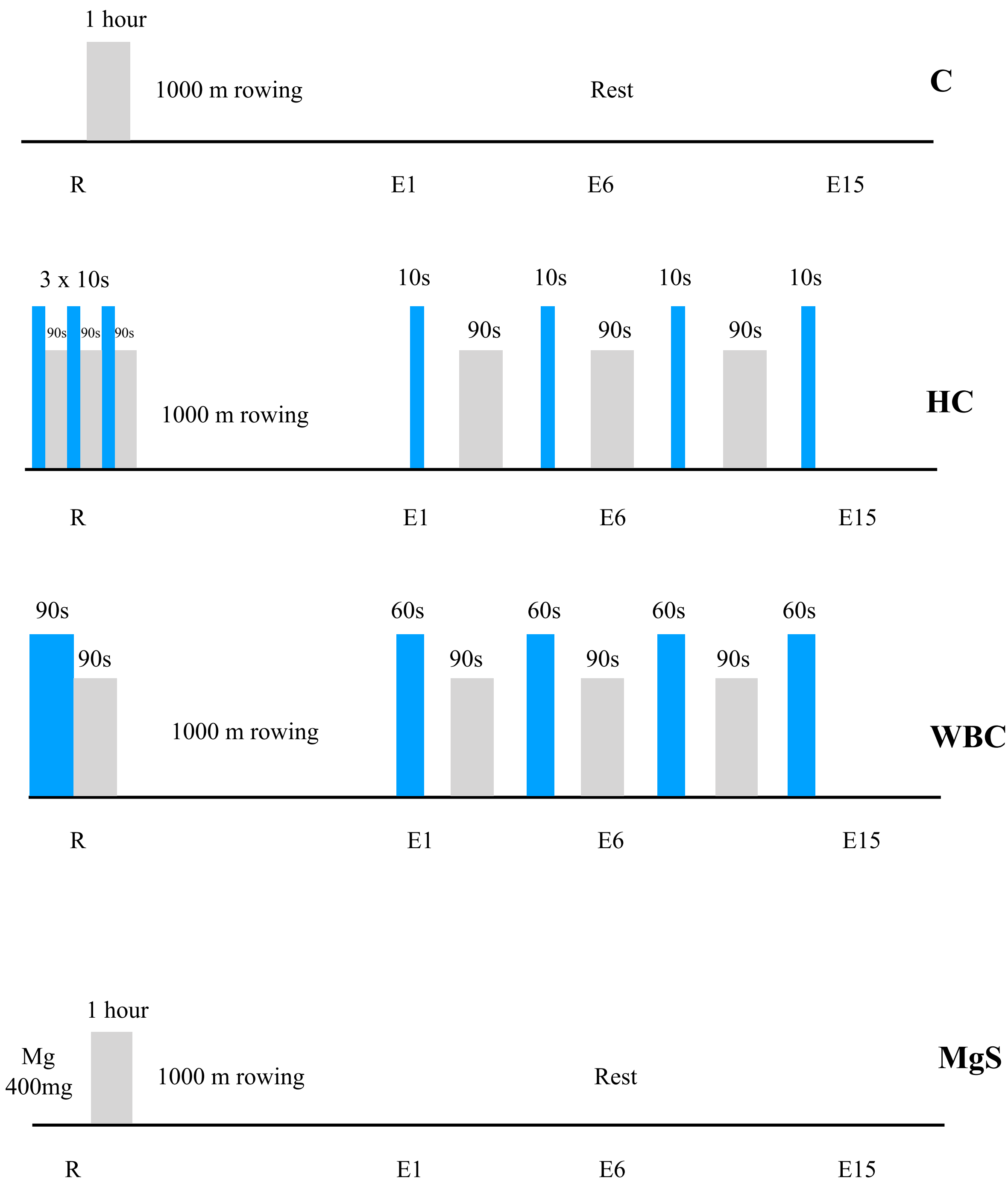

Figure 3-2-2 Illustration of the experimental protocol

The experiments consisted of two parts: pre-test (one day), and experiment (four days: 1 sedentary control group and 3 exercise groups). In pre-test part, all subjects did 400 m rowing as homogeneity analysis that divided into four groups (6 subjects/group), each group participated only one intervention. In the experimental part, there were four different interventions (① control group (C) ② hand cooling (HC) ③ whole body cooling (WBC) and ④ magnesium supplementation (MgS)). Each intervention had three sections, which were pre-exercise intervention, acute high intensity 1000 m open-end rowing (until exhaustion) and after-exercise (recovery period). There was a same recovery period interventions for 15 minutes respectively.

Rowing:

In pre-test part, all subjects did 400 m rowing as homogeneity analysis divided into 4 groups (6 subjects/group), each group participated only one intervention. 1000 m rowing as exercise was performed in each interventions.

Interventions:

Group 1 (C): The first group was control group (C). Subjects had to static rest (sited down) before 1 hour of the exercise and static rest (standing) 15 minutes after exercise in a quiet, peaceful room before all biological parameters were taken, to prevent any physical effort influencing baseline levels.

Group 2 and 3 (Cryotherapy methods):

Group 2 (HC): The second group was hand cooling (HC). Immersion of the hand for three times 10 seconds in ice water before the exercise. The same protocol but after exercise was used different time, the participants did four times 10 seconds ice water immersion by the hand, and rest for 90 seconds.

Group 3 (WBC): The third group was whole body cooling (WBC). Participants were subjected to hypothermia for 90 seconds by placing the body - except the head - into a cold chamber (air temperature: - 120 - 140 Celsius) before the exercise. During the recovery period four times 60 seconds cooling intervention were performed and rest for 90 seconds.

Group 4 (MgS): The fourth group was magnesium supplementation (MgS). The subjects were instructed to take 400 mg magnesium before an hour of the exercise.

Blood plasma and other parameters collected:

The following parameters were collected: lactate (La), NH_3, heart rate (HR), rate of perceived exertion (RPE), tympanic temperature (Tear), whole-body temperature (Tw), thermal sensation (TS). These measurements were performed at the rest time (R), before the exercise (R2), the first minute after exercise (E1), the third minutes after exercise (E3), the sixth minutes after exercise (E6), the seventh minutes after exercise (E7) and the fifteenth minutes after exercise (E15). As well as NH_3 and lactate was only measured in R1, E1, E6 and E15. As the exercise in each time, subjects all in the same 1000 m rowing at 100% intensity until exhausted. During exercise, mean watt, speed, were measured in every 200 m.

The subjects rated their thermal sensation (Ts) with a seven point standard scale (refs) by answering the question ‘How are you feeling now?’. They were informed to answer by naturally giving a number ranging from 1 to 7 (1 = cold, 2 = cool, 3 = slightly warm, 4 = neutral, 5 = slightly warm, 6 = warm, 7 = hot).

Statistical All subjects were used independent method.

3.3 Equipments

The following equipments were used in the present study:

Concept II Model D

The Concept II Model D delivers an effective cardiovascular workout and used to simulate 1000 m rowing exercise (Concept II Model D). This equipment will show the data regrading watt speed that automatically stores these informations in either its internal memory or on the monitor.

Figure 3-3-1 Concept II Model D

Cryotherapy

Luxe in bamboo, Space Cabin, CRYOMED Ltd ,Ukraine

This device ("Luxe in bamboo,") can cool the body to -110 ~ -195 °C in a cabin by using liquid nitrogen. The cabin allows body exposure to extremely cold temperature in a short time. The experimental temperature was set at -120 ~ -140 °C in this experiment and the athletes spent 3 x 90 second before and 4 x 60 seconds after exercise. The temperature and time were recorded at specific times.

Figure 3-3-2 Space Cabin

Measurements of blood plasma lactate

Diagnostic Biosen C-line; EKF-diagnostic, Germany

Blood lactate (mmol/L) was measured with the glucose enzymatic-amperometric method ("Biosen C-Line,") during the experiments.

Figure 3-3-3 EKF - diagnostic

Measurements of NH_3

PocketChemBA PA - 4130 ;ARKRAY, Japan and AMMONIA CHECKER II

The blood volume was collected 20 µL and placed in the ammonia test paper for three minutes after that analysis NH_3 (µmol/L).

Figure 3-3-4 PocketChemBA PA - 4130

Measurement of cardiovascular parameters

i-Heart system, Taiwan

The system monitors real-time heart rate, heart rate safety indicator and heart rate variability. i-Heart can also monitor the resting heart rate, exercise heart rate and recovery heart rate. i-Heart can display real-time heart rate in a graphical curve. The real-time heart rate of a target person can be displayed on an enlarged monitor screen and displayed independently.

Figure 3-3-5 i-Heart system to measure hemodynamic parameters

Measuring the core body temperature

Braun, Schutzkappen zu ThermoScan IRT4520

Tympanic temperature (T_{ear}) was measured with a tympanic thermometer (Braun, Schutzkappen zu ThermoScan IRT4520) in order to estimate core temperature.

Figure 3-3-6 RT4520 tympanic thermometer

Measuring body temperature

Terminal Camera P384-20

The thermal image is 384 x 288 pixels. A whole-body temperature report was automatically generated for each test session in Excel format.

Figure 3-3-7 Terminal Camera P384-20

Examples of images of various regions of whole-body temperature. Red indicate higher (36 °C - 37 °C) temperature. Green (and blue) indicates lower (28 °C - 30 °C) temperatures.

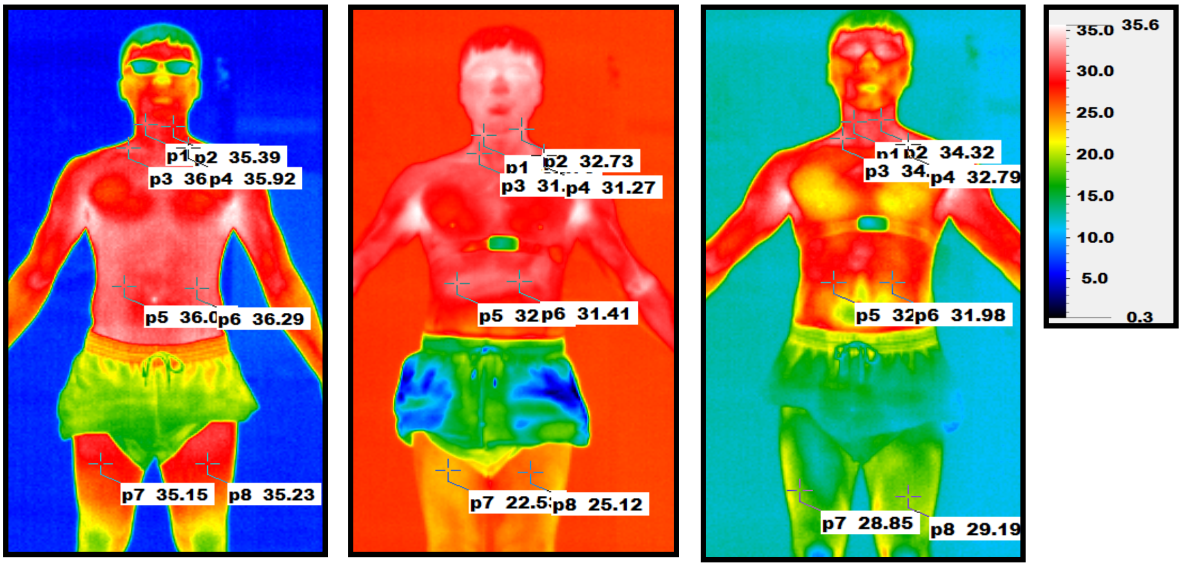

Figure 3-3-8 Examples of whole-body temperature

3.4 Statistical analysis

Two-way mixed (ANOVA) and Tukey' post hoc testing were used to analyze the data obtained in effects of different interventions. In this study, the correlative relationships between the biochemical hemodynamic parameters of subjects and the variables of their time were analyzed. The overall significance level was set at alpha = 0.05. Basic statistics of means and standard deviations (SD) in each group were computed using SPSS for Windows Version 22.0.

CHAPTER 4: RESULTS

The anthropometric of the subjects who participated in this study were similar for all performance, and body mass were also consequently highly homogeneous in this aspect.

Table 4-0-1 The anthropometric of all subjects

	Age (years)	Height (cm)	Mass (kg)
Mean±SD	25±4	175±5	76±10

n = 24

Table 4-0-2 Anthropometric, pre-test and experiment output

n = 6	C	HC	WBC	MgS
Age,yr	25±4	27±4	25±4	25±4
Height, cm	174±7	177±3	175±7	175±3
Body mass, kg	73±10	80±10	77±13	75±7
400 m (watts)	334±41	333±57	334±71	336±82

n = 6
C = Control
HC = Hand-cooling
WBC = Whole-body cooling
MgS = Magnesium supplementation

4.1 Effect of different interventions in plasma lactate and NH_3

The results shows that after 1 min of rowing exercise the blood plasma lactate concentration was 11.5±2.7 mmol/L in C, 11.1±2.7 mmol/L in HC, 10.9±1.4 mmol/L I WBC, and 11.9±1.0 mmol/L in MgS. There was no significant difference ($p > .05$) among the four groups (Figure 4-1-1). Interestingly, the lactate level tended to be lower with 2.6 mmol/L at E6 in WBC group than in HC group (not significant however). Also, at E15 the lactate level were less with 3.3 mmol/L in WBC than HC group (not significant). In addition, HC is the only group that at E15 the lactate level (13.31 ± 4.53) was greater than at E1 (11.55 ± 2.49). No statistically important difference is found among the other interventions.

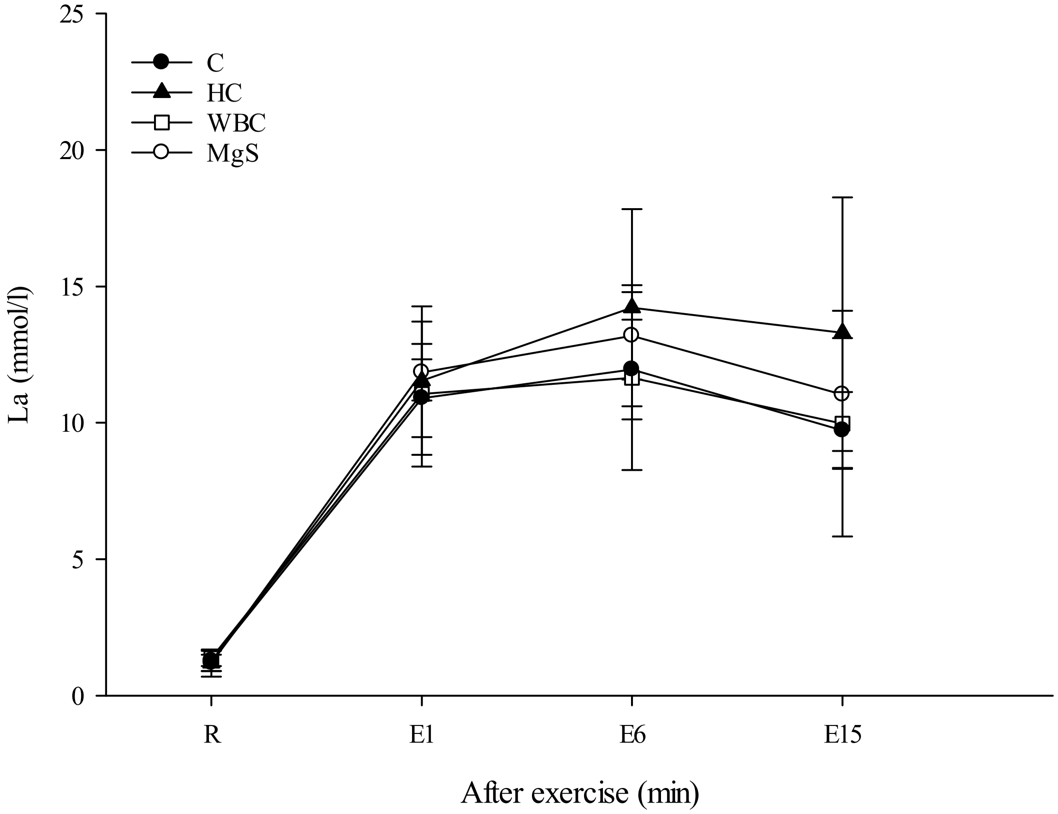

Figure 4-1-1 Plasma lactate concentration in different the time periods after 1000 m in four groups.

There were no significant differences in plasma NH_3 levels between four groups. At the E1, NH_3 in MgS 104±32 µmol/L, WBC 114±41 µmol/L, HC and C 130±56, 90±34 µmol/L respectively. Despite, HC 40 µmol/L was generally higher in the E1 compared with the WBC was 24 µmol/L. The data shows WBL as intervention during exercise can lower level of NH_3.

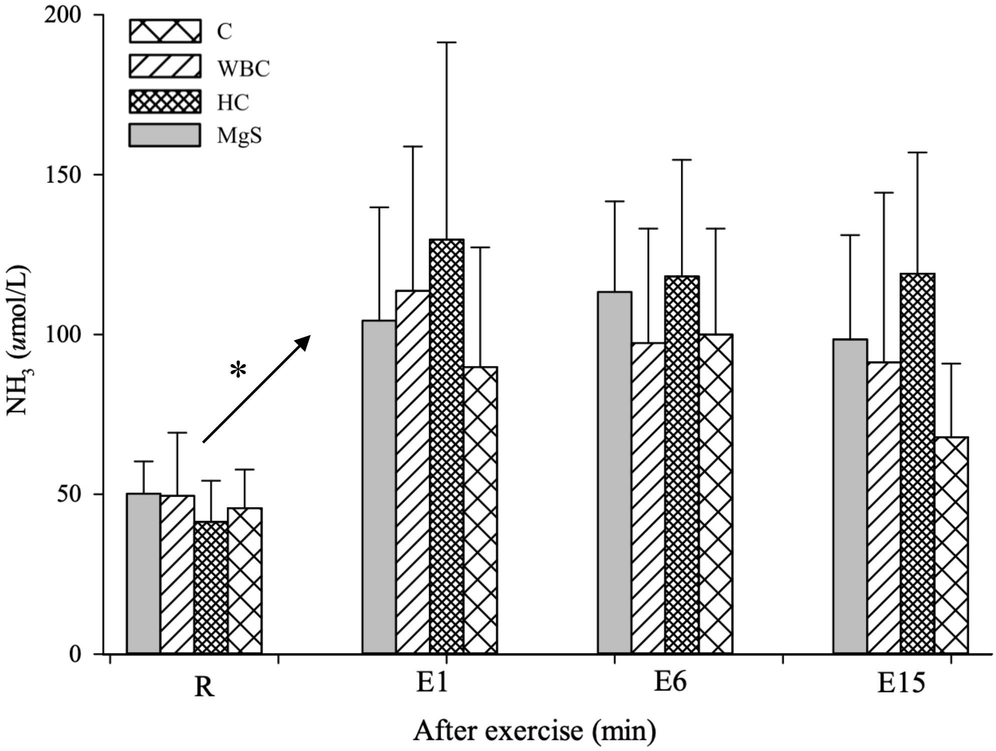

Figure 4-1-2 Plasma NH_3 level at rest and after different time period after rowing 1000 m in four groups. Asterisk (*) indicates significant between two set of columns.

Table 4-1-3 Plasma lactate and plasma NH_3 levels in different groups at different time points.

		R	E1	E6	E15
C	La	1.3±0.3	10.9±1.3	12±1.7	9.7±1.3
	NH_3	46±11	90±34	100±30	68±21
HC	La	1.2±0.5	11.5±2.5	14.2±3.3	13.3±4.5
	NH_3	41±12	130±56	118±33	119±35
WBC	La	1.4±0.3	11.1±2.4	11.7±3.1	10±3.8
	NH_3	50±18	114±41	97±33	91±48
MgS	La	1.2±0.3	11.9±1	13.2±1.5	11±1.9
	NH_3	50±9	104±32	113±26	99±30

C = Control
HC = Hand-cooling
WBC = Whole-body cooling
MgS = Magnesium supplementation

4.2 Effect of different interventions in HR

Changes in the heart rate response during the recovery periods is shown in Figure 4-2-1. Heart rate was not significantly different between trials. HC is lowest one, the heart rate decreased over time from 172±15 min^{-1}, 133±15 min^{-1}, 126±18 min^{-1}, 117±15 min^{-1} to 107±19 min^{-1} respectively at E1. Compare with trails at the first minute after exercise while MgS 180±12 min^{-1}, WBC 182±9 min^{-1} and C 176±10 min^{-1}. At E3, E6, E7 and E15, WBC is obviously higher than the intervention groups. The heart rates were 147±18, 135±15, 130±14 and 116±10 beats per minute for the E3, E6, E7 and E15 period, respectively. HC which was progressively decreased in 107±19 min^{-1} at the E15.

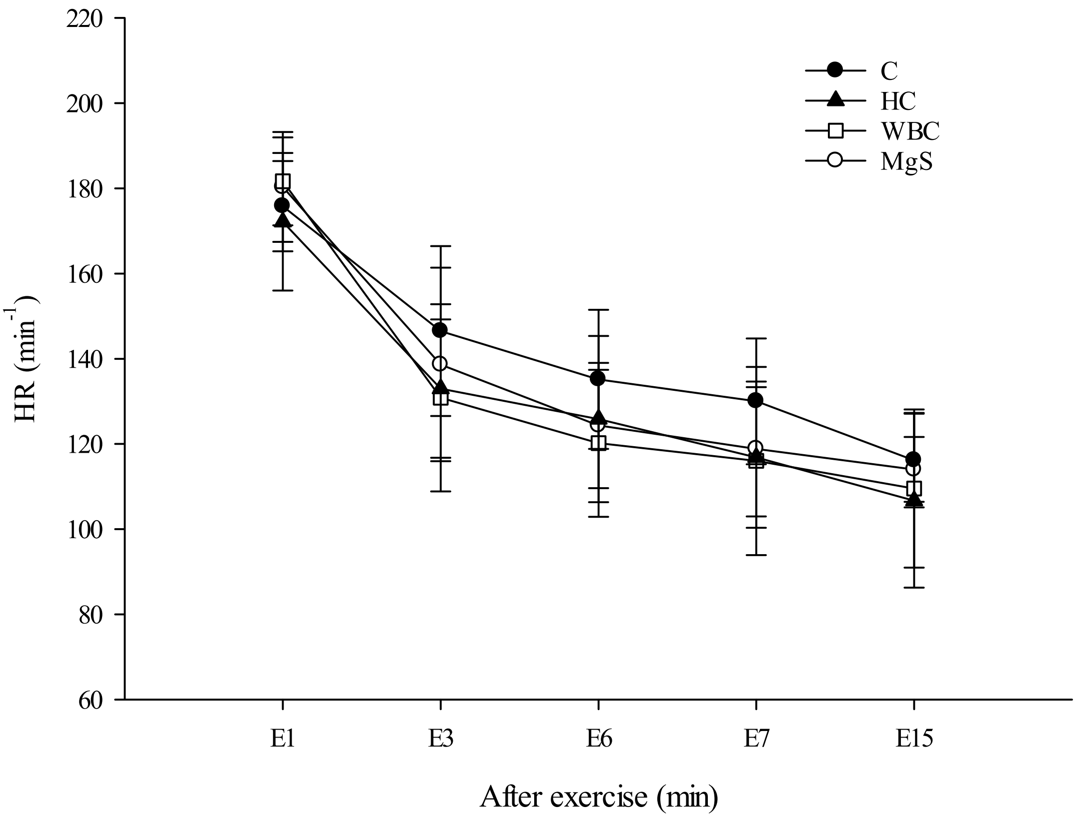

Figure 4-2-1 Changes in heart rate after rowing 1000 m in different time periods in the four groups.

Table 4-2-2 Heart rate in different groups after rowing 1000 m at different time points.

	E1	E3	E6	E7	E15	ER
C	176±10	147±18	135±15	130±14	116±10	4.29
HC	172±15	133±15	126±18	117±15	107±19	4.64
WBC	182±9	131±20	120±16	116±20	110±17	5.14
MgS	180±12	139±21	124±13	119±14	114±7	4.71

C = Control
HC = Hand-cooling
WBC = Whole-body cooling
MgS = Magnesium supplementation
ER = elimination rate

4.3 Effect of different interventions in rowing performance

The power out put (mean watts) were not significantly different (p>.05) among the groups. The large SD indicate that more data are needed to draw conclusion. Nevertheless, interestingly, the power out put was maintained on a similar level during the 1000 m rowing.

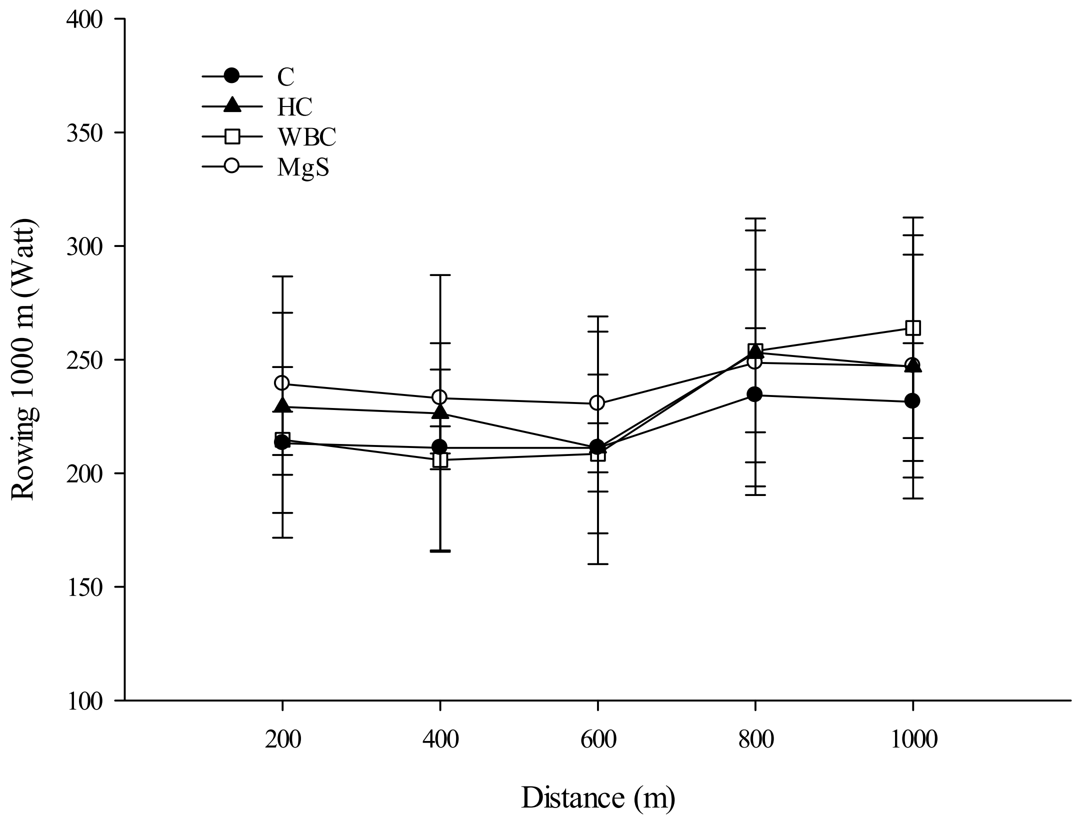

Figure 4-3-1 Average watts of rowing 1000 m produced by the athletes at different distances in the four groups.

In the 4 groups the watts produced were not significantly different interventional groups ($p > .05$).

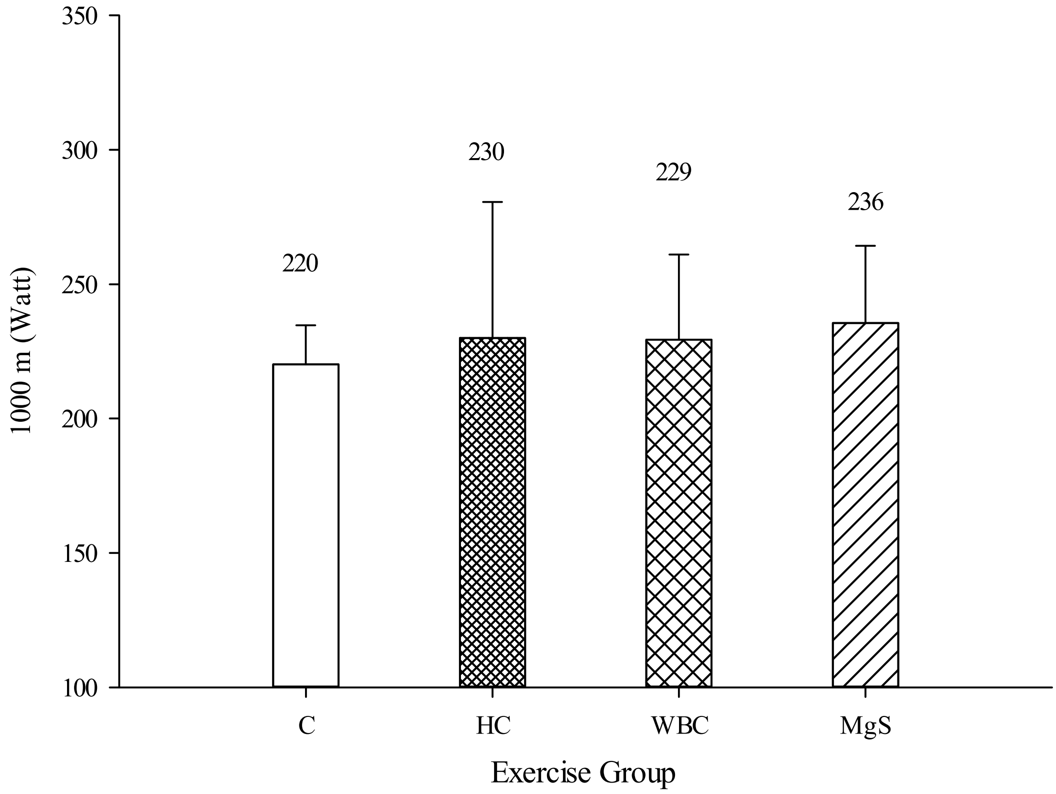

Figure 4-3-2 The total work (watts) of rowing 1000 m in different interventional groups.

The speed (m/s) of rowing in each group were recorded from the ergometer's monitor (total speed and segment speed). In the different groups there was no significant difference in the rowing speeds C (33±2), HC (34±3), WBC (31±3) and MgS (32±4).

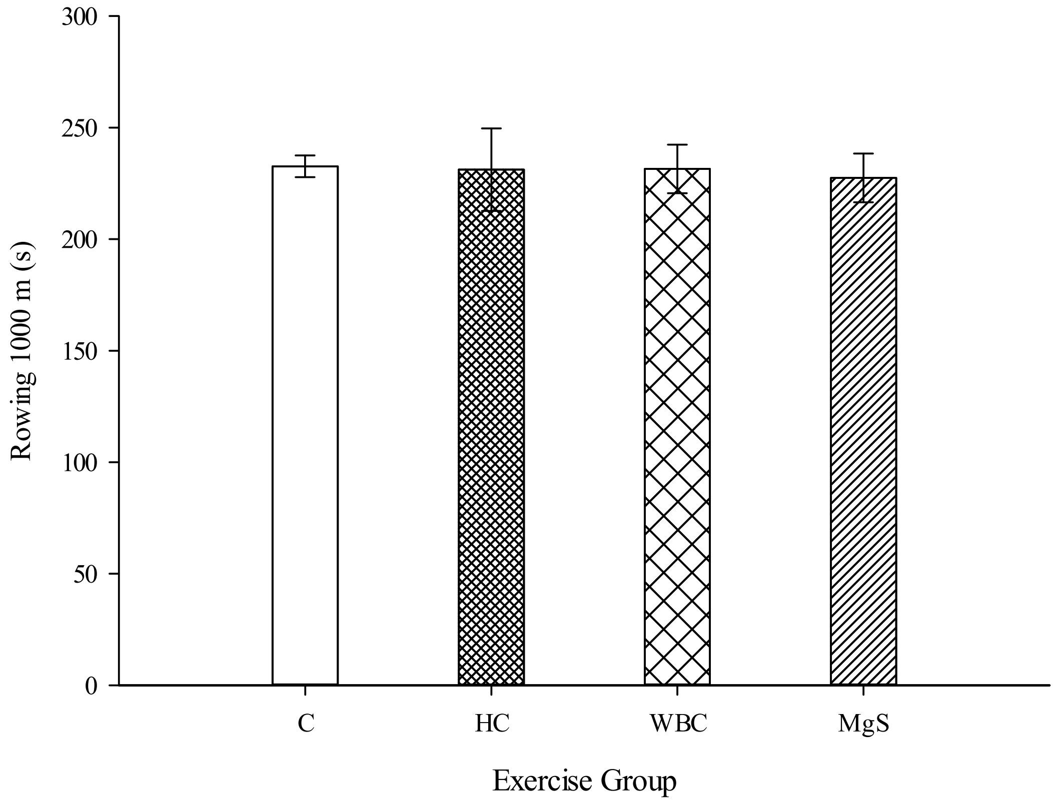

Figure 4-3-3 Separate speed in different interventional groups.

4.4 Effect of different interventions on whole-body temperature and sensations

Before exercise, at R2, TS scale was significantly lower in WBC group. Also, after exercise at time periods of E3 ($p < .00$, 2.67±0.47), E6 ($p < .00$, 2.33±0.47) and E7 ($p < .00$, 1.67±0.47) the TS scales were in the WBC groups than in the other are significantly lower than in the other groups. No other differences were found among the groups.

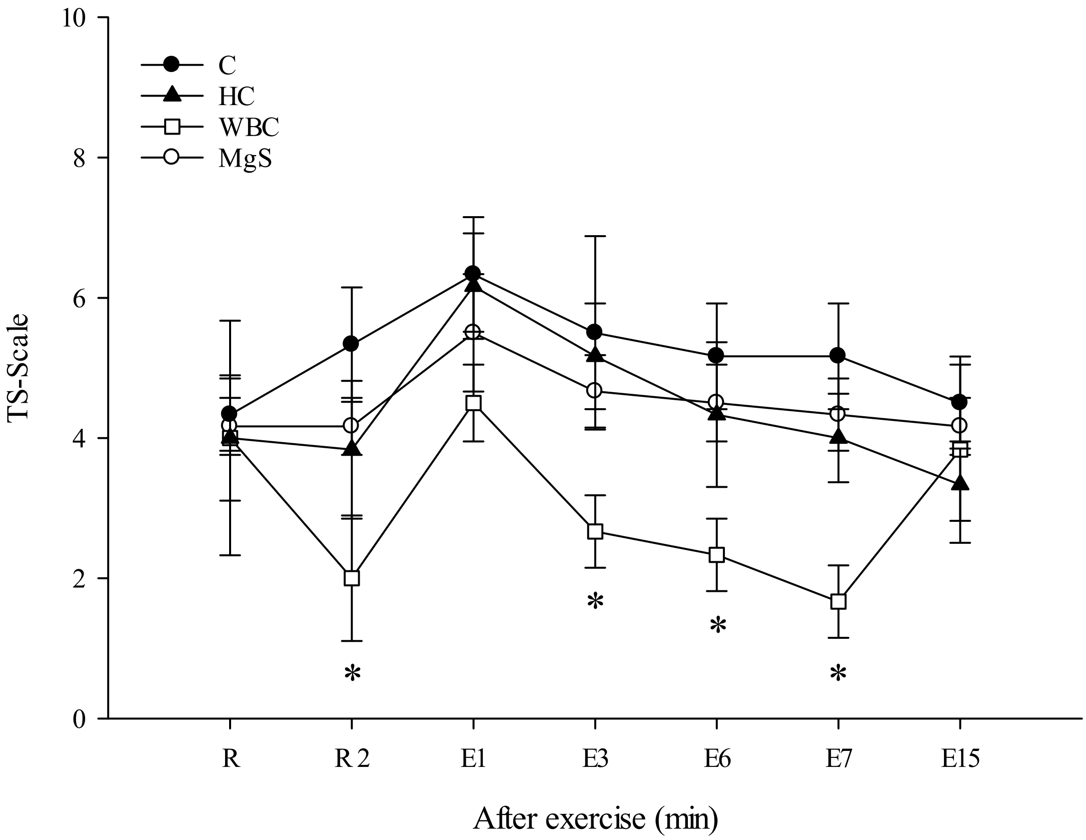

Figure 4-4-1 Terminal sensation (Ts) scale in different interventional groups at rest (R) and after exercise (E). Asterisks (*) indicated significant differences.

The decrease in whole-body temperature (Tw) as a function of time was significantly reduced in the WBC intervention at all time period, except in MgS, HC and C groups. Compared with WBC, another interventions during the recovery time (E1 - E15) were not statistically different but no other differences were found. The Tw of WBC cooling ($p < .00$, 29.7±0.70) was significantly lower ($p < .05$) compared to the HC (34.8±1.2) and MgS (34.5±0.5) intervention before the exercise (R2), E1 ($p < .02$, 31.37±1.15), E3 ($p < .00$, 29.05±1.52), E6 ($p < .00$, 27.13±1.46), E7 ($p < .00$, 26.41±1.35) and E15 ($p < .00$, 29.99±1.48).

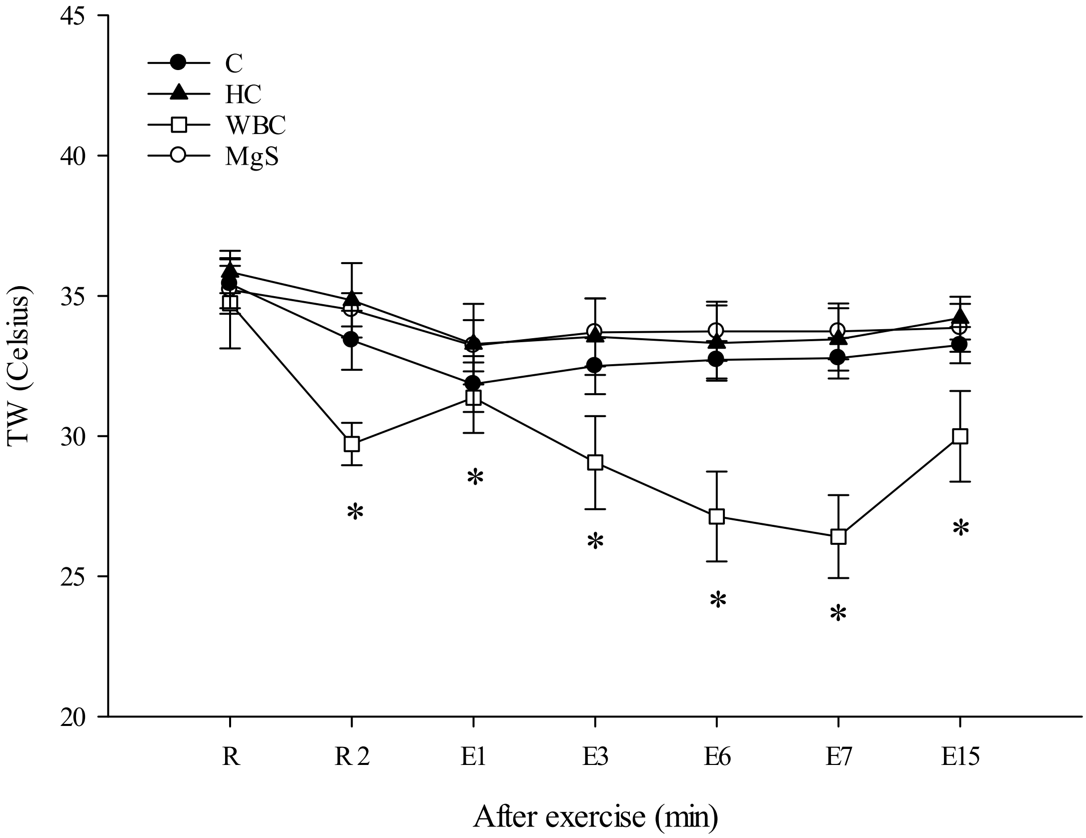

Figure 4-4-2 Whole-body temperature (°C) at different time points in four interventional groups. Asterisks indicated significant differences.

The RPE values of rowing 1000 m are shown in Figure 4-4-3. After exercise RPE were significantly reduced as a function of time in each group ($p < .05$). Also, in the first minute after exercise the RPE value was significantly lower in magnesium group than in the other interventional groups.

Where it was 8±1 ($p < .05$) at E15 in WBC, which was the pre-exercise level. In HC there was no difference.

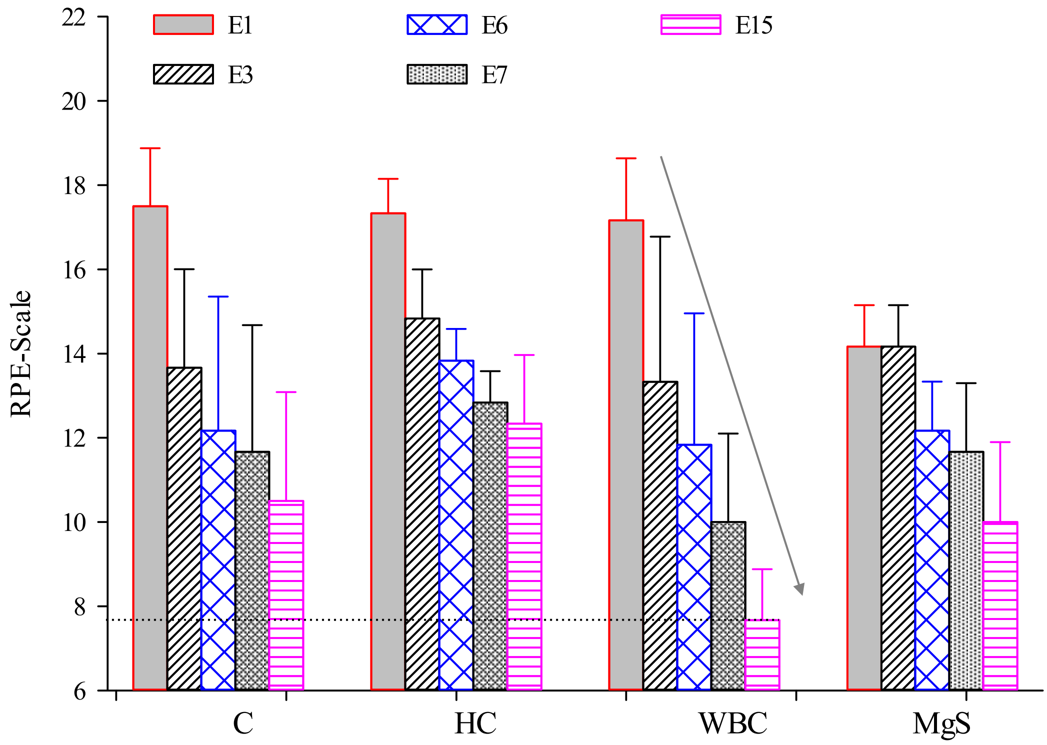

Figure 4-4-3 Rate of perceived exertion (RPE) after rowing 1000 m in four interventional groups at different time periods

CHAPTER 5: CONCLUSIONS

There were no significant difference ($p > .05$) among the four groups in plasma lactate, plasma NH_3, heart rate, total work (watts), speed (m/s).

The TS scales were significantly lower in WBC group than in the other groups at time periods of E3 ($p < .00$, 2.67±0.47), E6 ($p < .00$, 2.33±0.47) and E7 ($p < .00$, 1.67±0.47).

The whole-body temperature of WBC cooling ($p < .00$, 29.7±0.70) were significantly lower ($p < .05$) compared to other groups before the exercise (R2), E1 ($p < .02$, 31.37±1.15), E3 ($p < .00$, 29.05±1.52), E6 ($p < .00$, 27.13±1.46), E7 ($p < .00$, 26.41±1.35) and E15 ($p < .00$, 29.99±1.48).

The RPE values were significantly reduced as a function of time in each group ($p < .05$). Also, in the first minute after exercise the RPE value was significantly lower in magnesium group than in the other interventional groups. Where it was 8±1 ($p < .05$) at E15 in WBC, which was the pre-exercise level.

The main findings of this study are:

1) Compared to control group (C) hand-cooling (HC) did not affect the plasma NH_3.

2) Utilizing whole-body cooling (WBC) not only felt pleasurable cooling sensation during the therapy session, but also were able to maintain the high-dynamics performance.

3) Acute supplementation of magnesium (MgS) did not improved the physiological parameters but significantly decreased the whole-body temperature (Tw) and thermal sensation (Ts).

Comparing our finds to those in the literature and based on our results we suggest that cooling and magnesium supplementation - although did not improved the physiological parameters on the average - but significantly decreased the body temperature and thermal sensation and perceived difficulty of exercise, which could be important in the prevention of drop out cases in sport and likely facilitate the continuation of the training program.

Potential role of magnesium supplementation and cooling on cellular functions.

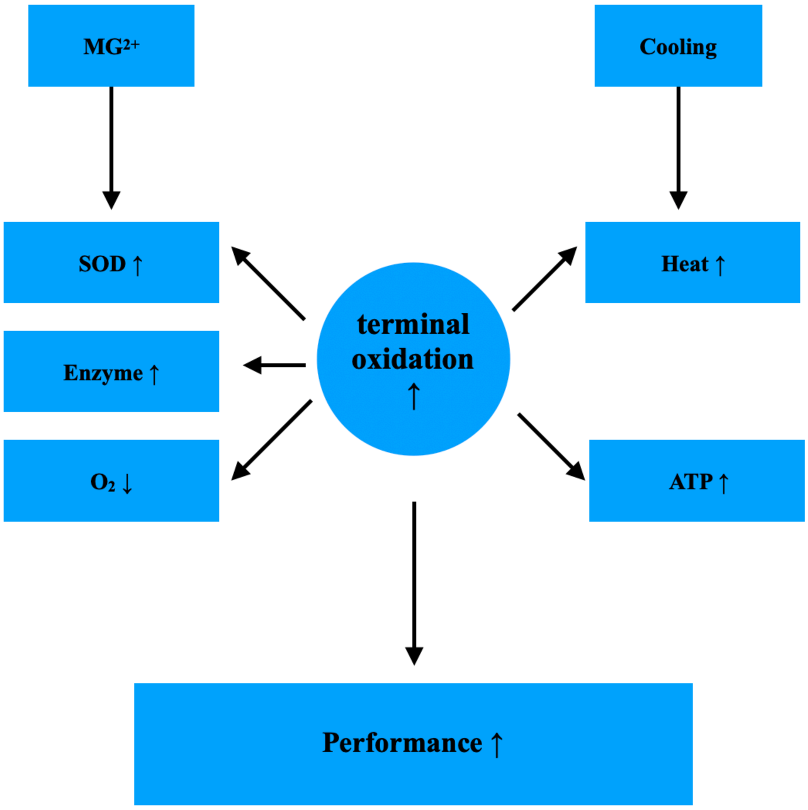

Figure 5-0-1 A possible mechanisms for the effectiveness of magnesium and whole-body cooling. Several metabolic mechanisms such as citric acid cycle, glycolytic cycle, terminal oxidation are affected by magnesium-dependent enzymes (Cowan, 2002) and cooling can promote the elimination of heat produced during the terminal oxidation, giving theoretical base for their use in exercise

5.1 Recovery of biological parameters after exercise in the 4 interventional groups

Lactate:

Current literature (Liesen, 1983; (張嘉澤，運動能力診斷與訓練調整，2010) suggests that lactate concentration exceed 12 mmol/L will effect sport performance. None of the intervention effected the physical performance of athletes in this study. The maximum lactate concentration was highest in E6 and the lowest in E15. C (Control: (E15 - E6) / 9 = 0.26 mmol/L) compare to the other of groups (MgS: 0.25 mmol/L, WBL: 0.19 mmol/L, HC: 0.10 mmol/L) has the highest elimination rate of lactate. It might because in the 1000 m rowing, C cannot reached the greatest power output which is only 219±13 watts, compared to highest one MgS 239±34 watts, WBL 232±49 watts and HC 227±29 watts. The lactate concentration is a combined result of many factors. Accumulating production of lactic acid would lead to an unbalanced biochemical reaction in the physiological system (Ahmad, 2019). The rowing power output also related to the speed of activity. Nevil et al., (Nevill, Beech, Holder, & Wyon, 2010) advised that the presentation of the results are more accurate and symmetric if rowing performance times were converted to average speeds (m/s).

Mg supplementation:

Strenuous physical exercise may lead to oxidative stress which is in part eliminated by Mg-dependent superoxide dismutase (MgSOD). Dimitrasonovic et al., (Dmitrašinović et al., 2016) reported that rugby players took four-weeklong Mg supplementation will effect ACTH and cortisol of body in recovery period after the competition day, also the levels of hormones, such as serum cortisol levels most significantly to see before the game. Kass et al., (Kass & Poeira, 2015) described that acute Mg supplementation have beneficial effects on peak torque and that physiological-regulatory functions of Mg within muscle contraction and relaxation, also in longer loading strategy (4 weeks) would improve these results, giving a more profitable and preeminent feedback. Even have some benefits for taking an acute Mg, especially before intense exercise. Others (Ç?nar, Nizaml?o?lu, & Mo?ulkoc, 2006) investigated effect of MG supplementation on lactate level in 4 week-period with 30 Tae-Kwan-do sportsmen, it was found that MG supplement may positively affect performance of sportsmen by decreasing their lactate levels. As the experimental hypothesis, MgS almost has the same rate as control group.

Temperature:

Morrissey et al., (Morrissey et al., 2019) compared three different group: 1) thermoneutral temperature (20°C, NEU); 2) cold with no active warm up (CNWU) and 3) cold with 25 minutes warm up (CWU) shown that even lactate threshold is likely higher in CNWU but the time to exhaustion and peak power output were also greater than CWU. Thus cooling environment is more benefit in performance as same as in our experiments. One study (Kozyreva, Tkachenko, Kozaruk, Latysheva, & Gilinsky, 1999) compared two different cooling, one of which is demonstrated a faster cooling whereas the other slower. The results show that plasma catecholamines extensive rise was increased by rapid cooling but slow cooling. External cold stimulation and local cooling of hypothalamus and spinal cord will stimulate sympathetic nerve activity in skin and reduce visceral sympathetic nerve activity. Cutaneous fibers generally showed inhibitory responses and visceral excitatory effects heat elicited reverse actions. The skin catecholamines may in connection with constriction of the skin blood vessels, which is an adaptive response. The dynamic activity of the skin cold receptors during rapid cooling presumably provided conditions for an earlier activation of the sympathetic system and, as a consequence, made feasible a more rapid initiation of the metabolic response to a sudden decrease in temperature. The more gradual reduction in lactate demonstrated by WBC may relate to thermoregulatory responses of body.

It is likely that MgS and WBC are the best interventions affecting both the speed and output watt, but it should be noted that HC is the lowest one in speed, which is contrary to actual watt output. Goosey et al., (Goosey-Tolfrey, Swainson, Boyd, Atkinson, & Tolfrey, 2008) founds that HC could lower the psychological stresses of exercising in the heat, but otherwise, hand sensitivity will effect of performance and technique in sports as well. Subject feel too cold of their hands to accurately hold the handle reach the maximum speed. The thermoregulatory and physiological of HC group is not elicited a positive effect compared with other groups. It might be the experiment environment, and not in the extremely hot temperature that HC cannot stimulate the heat exchange mechanism occurred at our body. Beside, as aforementioned studies, most of previous studies tested with endurance exercise in normal temperature and acute anaerobic exercise. Goosey et al., (Goosey-Tolfrey et al., 2008) also reported that pre-cooling techniques affect endurance performance and also can be effective in much shorter distances (3 km), but not such short acute exercise.

Though not completely clear, this acute high intensity exercise cannot reach an optimal effect. Alternatively, in this context we used cool water, replacing the negative pressure cooling device for economic consideration. Amorim et al., (Amorim, Yamada, Robergs, & Schneider, 2010) reported that used four heat stress trials included hand immersed in water bath (WB) results shows cooling surface area is too small to provide the necessary heat exchange under the heat accumulation. The device is used negative pressure cooling device but not like the non-pressure cooling situation that may result in different vasoconstriction, heat transfer and physiological responses.

NH_3:

Interestingly, plasma NH3 shows the highest level in the first minute after exercise (E1) of WBC (114±41 µmol/L), HC (130±56 µmol/L) but another interventions in E6 of MgS (113±26 µmol/L) and C (100±30 µmol/L). The lowest plasma NH3 concentration in all interventional groups were in E15 except HC in E6 (118±33 µmol/L). In HC group, the plasma NH3 level did not decreased, but increased. In C group showed the most decreased rate in 3.6 µmol/L (E6 - E15 / 9 = 3.6). Combined with the exercise output watt it is not difficult to see the results that C had the lowest NH3. As indicated above, the plasma NH3 in this test are not in high level because the exercise type 1000 m rowing tends to be a medium intensity exercise, it might not produce a highly amount of plasma NH3 in our body. This data was based on finding from a previous study that the NH3 was in range of 162 ± 82 µmol/L to 213 ± 95 µmol/L by different types of acute high intensity sprint exercise (Chen, 2010).

5.2 Recovery of perceptual measures after exercise in the 4 interventional groups

Whole-body temperature (Tw), HR combined with mental situation (RPE and Ts)

Humans can regulate their body temperature because of the very effective thermoregulatory system. Our body produces heat while doing physical exercise because muscle contraction needs ATP which is produced primarily in the mitochondria during the terminal oxidation when CO_2, heat and also water are produced (Ahmad, 2019).

Our results with whole-body temperature (Tw) show the highest thermal reading were in the resting condition (R1) in each groups except HC, it is only small differences in the R1 (34.7±1.5) and R2 (34.8±1.2) that may not have physiological importance. The lowest temperature of MgS and C groups both in the first minute after the exercise that this same result can be compared to Ahmad's past research. (Ahmad, 2019). But HC group present various result in temperature that the lowest temperature occur during E7 which MgS and C groups both in E1. An interesting point is that HC group show an extremely late and lowest temperature compared to other groups.

Previous findings (Ruddock et al., 2017) reported that while hand immersion in three different degree of cold water can augmented heat loss at the palm-water surface, enable cooler blood to return to the body and limit physiological strain. Hand cooling in wheelchair athletes (Goosey-Tolfrey et al., 2008) has been also reported to have positive effect. Which was different from this experiment, HC restrict thermoregulatory system of our body at the first minute of exercise but follows by different mechanisms compared with WBC. Even HC is not optimal choice, but while physical exercise metabolic heat production can raises by 10 to 20 fold, but lower than 30% of the heat generated is transformed in to mechanical energy (Lim, Byrne, & Lee, 2008; Sawka, 1988). Except as an end-product of metabolism, heat may act as a self-limiting signal that produce central inhibition during exercise (Ahmad, 2019).

In the present study Ts and Tw resulted in significant difference ($p < .05$) between- which groups??? differences in WBC intervention. This may be due to a strong low temperature effect in whole body cryostimulation. This data suggest that athletes could use the WBC as intervention to limit the rise in Tw in pre-exercise and post-exercise. The reductions in Ts also indicated that regardless of physical or psychological causes, WBC can make athletes feels better and thus the mental part can elicit an optimal option to athletes without changing their performance. The interactions between peripheral and central thermoregulatory mechanisms are complex, the protocol of WBC in this study that combination of pre-exercise and during exercise may have activate hidden mechanisms resulting in beneficial effects on physical exercise, although the exact mechanisms are still unclear.

Heffernan et al., (Heffernan, Horner, De Vito, & Conway, 2019) reviewed that changes in neuromuscular strength may be influenced by duration of magnesium supplementation and training intensity. Although magnesium supplementation increased the elimination of lactate , it but it did not had a positive effect on Ts and Tw. Studies of animal experiment in calcium and magnesium deficiencies suggest that any adjustment in the magnesium level will directly regulating the blood pressure and body temperature. Also the body temperature decreased in magnesium deficient rat (Itokawa, Tanaka, & Fujiwara, 1974). We suggest that magnesium supplementation used in our study may have no effect on physical performance when serum concentrations of lactate are in the normal range (serum Mg 0.8-1.2 mmol/L). As a limitation of this study, there had been previous reports showing that magnesium deficiencies are mostly present in female athletes and not in males. Newhouse et al., (Newhouse & Finstad, 2000) studies have found significantly improved resting magnesium levels but no effect on performance or recovery in a group of physically active females. Current literature (Siquier-Coll et al., 2019) compared in different temperature (normothermic and hyperthermic) on exhaustion exercise, how does it effect body concentrations of magnesium (Mg) and phosphorus (P). The results showed that exercise in extremely hot conditions induces a tissue redistribution of magnesium in the body, which is not observed in normothermic conditions. The ambient environment temperature of the present study which was the same as the result of above studies.

Hausswirth et al., (Hausswirth et al., 2013) compared that if partial-body cryostimulation (excepted head and neck) has the same effect as whole-body cryostimulation (included head and neck). They have found that whole-body cryostimulation (included head) can induce a larger increase in plasma catecholamines, and a larger increase in the parasympathetic modulation of the heart rate. Although WBC tended to be more effective than partial-body cryostimulation (except head and neck) the results showed that acute 3 minutes cryostimulation induced a strong autonomic response of both cryotherapy. It is known that heart is under the influence of the parasympathetic system controlling heart rate, especially in trained athletes. Seiler et al., (Seiler, Haugen, & Kuffel, 2007) monitored acute disturbances in autonomic balance using heart rate variability following different types of exercise in highly trained Norwegian orienteers. Heart rate variability is an easy way to monitor the intensity (aerobic and anaerobic) of exercise and the contribution of sympathetic and parasympathetic nervous system. We found that heart rate results are basically consistent with the observed results and that WBC has the most elimination?? rate and heart rate almost certainly??? decreased.

5.3 Thesis summary

This is the first study, applying different interventions to effect the recovery of biological and mental parameters after acute exercise. Training is a complex procedure that athletes of different sport fields results in changes various physiological pathway and responses. To know which recovery way is the best to bring back athletes to the competition can help us if we use scientific monitoring several post-training recovery interventions. More controlled studies are needed to clarify that the mechanism of magnesium supplemention, hand-cooling and whole body cooling. The supplementation of magnesium may benefit the performance of acute exercise by improving the output of exercising power, and also increased lactate elimination rates. The subjects utilizing the partial body cryotherapy, not only felt pleasurable cooling sensation during the therapy session, but also were able to maintain the high-dynamics performance. On the other hand, we found no positive effects in the procedure of hand-cooling, except for the NH_3 elimination rate.

It should be noted that changes in our body depends on the training conditions, which however can be influenced by magnesium supplementation and interventions. Further studies are needed to determine the influence and practical significance of magnesium supplementation and cryotherapy on different exercise intensities as well as regarding the ideal post-exercise recovery protocols. The results were based on laboratory experiments and we should use the post-exercise protocols during actual training secession in the future.

5.3.1 Limitations of our studies

The ambient temperature were lower than 25 °C on each day in our experiments, which does not present an extreme hot conditions, thus we assumed that subjects were not heat acclimatized. Also, the sample size was too low in some conditions, thus meaningful statistical analysis could not be applied.

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