EFFECTS OF SPORTS DRINK ON ENDURANCE PERFORMANCE IN HEALTHY THAI MALES AFTER GLYCOGEN DEPLETION

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ABSTRACT

The purpose of this study was to examine the effects of sports drink intake on endurance performance, in concomitant with cardiorespiratory, blood chemistry, and metabolic variables. Fourteen healthy males with ages ranged from 18-25 years old participated in 3 trials, with 1 week apart. Three consecutive occasions in each trial, including glycogen depletion, recovery period and endurance exercise (cycling at 70%VO_{2peak}), were employed. During 2 hours recovery period, subjects were randomly assigned to three kinds of fluid intake including sports drink (SD, 10%carbohydrate, 0.16%electrolytes, placebo (PL, 10%carbohydrate); and plain water (WT). The time to exhaustion and work done, cardiorespiratory, blood lactate and glucose concentrations and metabolic variables were recorded. The results showed longer time to exhaustion in SD (52.93 ± 6.98 min) than PL (45.05 ± 4.47 min) and WT (37.95 ± 4.92 min), but significant difference was only observed between SD and WT ($p<0.05$). Significantly higher work done under SD and PL compared with WT, significantly higher ejection fraction (EF) ($p<0.05$) in SD than WT. Moreover, significantly lower minute ventilation under SD than PL and WT ($p<0.05$). There was no difference in blood lactate, blood glucose among three groups. It can be concluded that carbohydrate with electrolytes did not affect cardiorespiratory function, as well as blood chemistry profiles. However, additional electrolytes in sports drink tend to exhibit further exhaustive time.

KEY WORDS: SPORTS DRINK / ENDURANCE PERFORMANCE /GLYCOGEN DEPLETION / CARBOHYDRATE / ELECTROLYTE

71 pages

ผลของเครื่องดื่มทางการกีฬาต่อสมรรถภาพทางกายแบบทนทานในชายไทยสุขภาพดีภายหลังการพร่องของไกลโคเจน

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บทคัดย่อ

การศึกษานี้มีวัตถุประสงค์เพื่อศึกษาผลของเครื่องดื่มทางการกีฬาต่อสมรรถภาพทางกายแบบทนทานร่วมกับการเปลี่ยนแปลงของระบบหัวใจและหายใจ สารเคมีในเลือด และระบบเผาผลาญในชายไทยสุขภาพดี โดยจะคัดเลือกผู้เข้าร่วมวิจัยเพศชาย ช่วงอายุ 18-25 ปี จำนวน 14 คนเพื่อเข้าร่วมการศึกษาทั้งหมด 3 ครั้ง และมีระยะเวลาห่างกันอย่างน้อย 1 สัปดาห์ โดยแต่ละครั้งจะประกอบด้วยการทดสอบ 3 ช่วงดังนี้ 1.การทำให้สารไกลโคเจนในร่างกายหมดไป 2.ช่วงพักฟื้นร่างกาย และ 3.การออกกำลังกายแบบทนทานที่ระดับความหนัก 70% ของอัตราการใช้ออกซิเจนสูงสุดของร่างกาย ผู้เข้าร่วมวิจัยจะได้รับการสุ่มให้เครื่องดื่ม 3 ชนิดได้แก่ เครื่องดื่มทางการกีฬา (สารคาร์โบไฮเดรต 10%, สารอิเล็กโทรไลต์ 0.16%), เครื่องดื่มหลอก (สารคาร์โบไฮเดรต 10%) และน้ำเปล่าในช่วงพักฟื้น 2 ชั่วโมง ระยะเวลาการออกกำลังกายจนล้า, ตัวแปรของระบบหัวใจและหายใจ, สารแลคเตทและกลูโคสในเลือด และตัวแปรของระบบเผาผลาญถูกวัดและบันทึกในระหว่างการทดสอบ จากผลการศึกษาพบว่า ระยะเวลาการออกกำลังกายในผู้ที่ได้รับเครื่องดื่มทางการกีฬา เครื่องดื่มหลอก และน้ำเปล่าคือ 52.93 ± 6.98 นาที, 45.05 ± 4.47 นาที และ 37.95 ± 4.92 นาทีตามลำดับ อย่างไรก็ตาม มีเพียงเครื่องดื่มทางการกีฬาและน้ำเปล่าที่พบความแตกต่างอย่างมีนัยสำคัญทางสถิติ ($p<0.05$) นอกจากนี้ยังพบว่างานที่ได้ (work done) ในเครื่องดื่มทางการกีฬาและเครื่องดื่มหลอกมีค่าสูงกว่าน้ำเปล่าอย่างมีนัยสำคัญทางสถิติ ($p<0.05$) ยิ่งไปกว่านั้นพบว่าค่าความสามารถของหัวใจในการบีบเลือดออกจากหัวใจ (Ejection fraction) และปริมาตรอากาศต่อนาที (minute ventilation)ของกลุ่มเครื่องดื่มทางการกีฬามีค่ามากกว่า และน้อยกว่ากลุ่มเครื่องดื่มหลอกและน้ำเปล่า ตามลำดับ ($p<0.05$) อย่างไรก็ตาม ไม่พบความแตกต่างระหว่าง 3 กลุ่มของสารแลคเตทและกลูโคสในเลือด สรุปผลการศึกษา เครื่องดื่มสารคาร์โบไฮเดรตและสารอิเล็กโทรไลต์ไม่มีผลต่อการเปลี่ยนแปลงของระบบหัวใจและหายใจ รวมทั้งสารเคมีในเลือด อย่างไรก็ตาม เครื่องดื่มสารคาร์โบไฮเดรตและสารอิเล็กโทรไลต์มีแนวโน้มต่อการยืดระยะเวลาการออกกำลังกายจนกว่าจะล้าได้ดีกว่าสารคาร์โบไฮเดรตเพียงอย่างเดียว

71หน้า

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CHAPTER I
INTRODUCTION

1.1 Background and justification

In general, athlete performance impaired after matches due to many factors, such as, depletion of energy sources, water and electrolytes lost, and muscle fatigue and minor damages. Thus, back-to-back sports matches obviously influence to recovery period and also athletes' performance. Various recovery strategies were suggested, such as, neuromuscular electrical stimulation, active recovery, massage, cold water immersion and sports drink supplementation (1-4). Sports drink is one of those strategies which can mainly replace fluid and electrolytes lost during exercise. Dehydration and water imbalance have been known to induce physiological impairment which leads to attenuation of performance (5).

Sports drink composed, normally, of water, carbohydrate, amino acid and electrolytes. It is well known that carbohydrate is an essential energy substance for vigorous and also prolonged exercises. The early depleted glycogen may represent inadequate endogenous glycogen storage, as the consequence, athlete may not able to maintain his performance throughout exercise duration. Thus, it is important to consume sports drink containing energy substrates and electrolytes to sustain exercise ability. Many studies investigated effect of sports drink based on different phases of consumption period. Some recent studies showed that sports drink consumed before exercise improved performance during 1 hour running (6), but other (Gomes RV, 2013) demonstrated no difference in specific tennis skill when compared with taste-matched placebo (7). On the other hand, Zachwieja JJ (1993) investigated carbohydrate supplement, given within 2 hours after exercise at 70%VO_{2max}, and demonstrated histologic finding of replenished glycogen storage in muscle and liver (8). In addition, 6.4% carbohydrate-electrolytes ingestion was reported to improve soccer skill performance (9). Moreover, Burke LM (2004) suggested that proper amount of carbohydrate is essential to replace glycogen lost during competition (10).

McLellan TM (2014) suggested that athletes should consume at least 1g of carbohydrate/kg body weight/ hour to replace glycogen between and after competition and also be as an ergogenic benefits for the subsequent exercise (11).

Glucose level must be precisely regulated to sustain long-term exercise (12). Some sports require short recovery period within a day, for instance, swimming, badminton and taekwondo. Thus, glycogen re-synthesis may be critical before the second bout (13). Recent studies found that exercise time trial performance was improved in subsequent exercise after consumed sports drink (14-16). Furthermore, carbohydrate combined with electrolytes can extend further time to exhaustion than placebo (17-20). In addition, greater blood lactate (14) and blood glucose (16, 21) were found under sports drink compared with placebo condition.

Evidence has been elucidated that electrolytes, mainly as sodium and potassium, play an important role to stimulate rehydration and fluid absorption (22). Another role is also, basically, as an essential substance for cardiac function (23). Therefore, it might be such an important factor to sustain higher exercise duration than beverage contained carbohydrate alone. Only Sun JM (2008) investigated effect of sports drink on hydration status and found that 0.72 ± 0.38% fluid loss in Gatorade sports drink group while 1.10 ± 0.52% fluid loss in water group (24). However, there are controversial results in cardiovascular function. Some studies showed lower heart rate (16, 25) which may lead to sustain prolonged exercise or competition under sports drink, while the others showed unaltered of cardiovascular function (19, 21). There is no any research differentiated the effect between carbohydrate-electrolyte and carbohydrate drink alone. Thus, the present study is aimed to examine the effect of sports drink which is mainly composed of carbohydrate and electrolytes on endurance performance compared with carbohydrate only.

1.2 Objectives

1. To examine the acute effects of sports drink ingestion on subsequent endurance performance following glycogen depletion exercise in healthy Thai males.

2. To compare the acute effects of water, placebo (carbohydrate alone) and sports drink (carbohydrate-electrolytes) on endurance performance, in concomitant with cardiorespiratory, blood chemistry and metabolic variables in healthy Thai males.

1.3 Hypothesis

It was hypothesized that intakes of carbohydrate-electrolytes drink during recovery period could improve subsequent endurance exercise performance, as well as on cardiorespiratory, blood chemistry, and metabolic variables more than sports drink containing carbohydrate alone and water.

1.4 Benefits of this study

It could be applied as: a) a guideline for athletes involving repeated prolonged competitions who need fluid and electrolytes and b) understanding on changes of physiologic variables including cardiorespiratory, blood chemistry and metabolic functions during subsequent exercises.

CHAPTER II
LITERATURE REVIEW

2.1 Physical performance

Physical performance or physical fitness is defined as well-being status that derives the body for all activities. Its effectiveness depends upon storages and mobilizing different types of energy substrates essential in either for emergency situation or long-term performance. Physical performance can be divided into two different categories (26). The details are as follow:

1. Health-related physical performance is defined as performance needed for general health promotion. It composes of body composition, muscular strength, muscular endurance, flexibility and cardiorespiratory functions.

2. Sport-related physical performance is composed of components of special skills needed for specific sports, for instance, balance, speed, power, coordination, reaction time, anaerobic and aerobic capacities.

Each physical performance component needs specific test. Main factors affecting on performance included age, gender, body composition and level of physical training, which involve with both health and sports-related functions. For example, resting heart rate (Table 2.1) and oxygen consumptions (represents body's endurance) (Table 2.2) vary with ages and fitness levels (27). Therefore, these can be used as indicators to distinguish between active group and sedentary group.

Table 2.1 Illustrations of males resting heart rates at different ages and levels of physical fitness (28)

Age / Level	18-25	26-35	36-45	46-55	56-65	65+
Athlete	49-55	49-54	50-56	50-57	51-56	50-55
Excellent	56-61	55-61	57-62	58-63	57-61	56-61
Good	62-65	62-65	63-66	64-67	62-67	62-65
Above Average	66-69	66-70	67-70	68-71	68-71	66-69
Average	70-73	71-74	71-75	72-76	72-75	70-73
Below Average	74-81	75-81	76-82	77-83	76-81	74-79
Poor	82+	82+	83+	84+	82+	80+

Table 2.2 Illustrations of maximal oxygen consumption of Thai males at different ages and levels of physical fitness (27)

Fitness level	**Age (year)**					
	17-19	20-29	30-39	40-49	50-59	60-72
Excellent	≥ 55.5	≥ 51.6	≥ 43.3	≥ 37.4	≥ 33.9	≥30.7
Good	50.6-55.4	47.1-51.5	39.4-43.2	34.1-37.3	30.7-33.8	27.9-30.6
Average	40.7-50.5	38.0-47.0	31.5-39.3	27.4-34.0	24.2-30.6	22.2-27.8
Below average	35.8-40.6	33.5-37.9	27.6-31.4	24.1-27.3	21.0-24.1	19.4-22.1
Poor	≤ 35.7	≤ 33.4	≤ 27.5	≤ 24.0	≤ 20.9	≤19.3

2.2 Energy systems

Basically, body movement makes possible via using of an essential energy source in our body, named as adenosine triphosphates (ATP). In fact, all cells and organs in the body require high amount of ATP to maintain their functions throughout life. In some circumstance, like strenuous exercise, where ATP demand is too high, its production may become limited. Rates of ATP being generated are generally dependent upon exercise intensity, duration and work-rest interval. Three energy systems which produce ATP within human body are as follows (29):

1. Phosphagen system (ATP-PC): A very basic energy substrate, which is used at initiations of all physical activities and exercises. Creatine phosphate (CrP or Phosphocreatine) is a highest energy substrate. Phosphate from the broken down of CrP is combined with adenosine diphosphate (ADP) to reproduce for ATP. Therefore, this system can synthesize ATP at fastest rate when compare with another systems, however, both CrP and ATP are stored in a very limited amount (Table 2.3). Some activities with high intensity in short duration, such as heavy weight lifting, pushing, and sprinting require this earlier production of this energy system.

2. Anaerobic glycolysis: It takes place as CrP-ATP becomes diminish and also implies the condition where glucose metabolism is being done without oxygen required. Glycogen, stored in the liver and muscle cells, are converted to glucose. It is known that 1 molecule of glucose can produces 2 ATP in this anaerobic glycolytic process. Even though this energy system rushes for an immediate ATP, however, the storage amount is limited (Table 2.3). This energy system will be used in some activities with short-term duration of less than 3 min, for instance, 200 and 400-meters running.

3. Aerobic system: It is the process where 3 main energy sources, carbohydrate, protein and fat, are metabolized with oxygen supplied. In this aerobic process, 1 molecule of carbohydrate and fat can approximately provide 38 and 44 ATPs respectively. Even though these processes take quite longer duration for ATP generation, however, in term of health promotion it is worth to sustain physical activity up to fat-burning outcome. In general, this is mostly used in daily life and also used in some sports with long duration, such as cycling and marathon. All three energy systems characteristics are shown in Table 2.3.

Table 2.3 Characteristic of the energy systems, conversion rates and quantities in human (30)

	ATP	Creatine phosphate	Anaerobic glycolysis	Aerobic glycolysis	Fatty acid oxidation
Amount	5 mmoles/kg	17 mmoles/kg	350 g muscle glycogen	440 g glycogen muscle/liver	9000-15000 g
Total phosphate available (mmoles)	220	660	6700	84000/19000	>4,000,000
Maximum rate of ATP synthesis	-	73 mmoles	40 mmoles	17/6 mmoles	7 mmoles
Duration	-	4 seconds	2-3 minutes	1-2 hours	Many hours

2.3 Cardiovascular system and exercise

Heart is a pump, which takes venous blood from vena cava and distributes oxygenated blood via aorta to supply the entire body. Heart composes of 4 separated chambers, with 2 chambers of right and left atriums and 2 chambers of right and left ventricles. Most cells in the chambers are specialized tissues named as cardiac myocytes, which can self-generate action potential. Heart chamber contains special cells named as sinoatrial (SA) node, which is located at right atrium and when it is stimulated signals or action potential of the heart will be sent to the second special node called arteriovenous (AV) node which is located at interventricular septum. Signals from these two nodes dictate electrical potentials of other myocardium cells. Similar to other excitable tissues, these cardiac myocytes function via two main electrolytes: sodium (Na^+) and potassium (K^+), which mobilize across its membrane, thus electrical potential is taken place in a “all or none fashion” and so called action potential. Calcium (Ca^{2+}) which is released from sarcoplasmic reticulum signals cardiac muscle contraction in the simultaneous pattern of atrium and ventrical (31). In general, venous blood return, which riches of carbon dioxide (CO_2) concentration, is filled through right atrium and then right ventricle, which then is pushed to the lungs for gas exchange at alveolar level. Arterial, oxygenated blood from the lungs is returned through left atrium and ventricle before pumps out via aorta to supply the entire body (23). During exercise, the body requires more oxygen supplied, particularly when increases workload. Cardiovascular function is responsible to that increasing such requirements (32). Main cardiovascular parameters involved will be altered as follow:

Heart rate: In general, heart rate is regulated by autonomic nervous system which is composed of sympathetic and parasympathetic components. During exercise, the mechanoreceptor in skeletal muscle is additionally stimulated according to mechanical changes. This, therefore, needs more oxygen to supply to the moving parts. Afferent inputs travels into the brain, where cardiac control center is located. After integration, efferent signals are sent via sympathetic nerve to increase heart rate frequency (33). In addition, exercise with high intensity or prolonged duration stimulates central command activation. It is the matter of fact that as the body recruits more skeletal muscle into its action, more oxygen requirements will be remarkably increased (34). This compensation is possible by the increase in cardiac frequency.

Stroke volume: It can be calculated from the differences between end-diastolic and systolic volume for any heartbeat. During exercise, sympathetic nervous activity will be stimulated and it can affect to myocardial contraction. This augments contractility of heart, allowing to decreasing end-systolic volume. In contrast, increase of end-diastolic volume is resulted from both pulmonary artery wedge pressure and increase of venous return to the heart. Therefore, increase of end-diastolic pressure and reduction of end-systolic volume result to be more stroke volume (32, 35). However, exercise with high intensity can be caused to be less progressive of stroke volume due to too fast heart rate (35).

Cardiac output: Normally, cardiac output is calculated from heart rate and stroke volume. It was found that both heart rate and stroke volume are increased during exercise, thus cardiac output will be increased. Moreover, there is more venous return to the heart resulted by skeletal muscle pump, respiratory muscle pump and more redistributed blood flow to active muscle (32). However, exercise at too high intensity diminishes cardiac output with faster heart rate and reduction in ventricular filling time (36).

Blood pressure: Baroreceptor regulates blood pressure via the stretched-vessels and increases its firing rate to cardiac-vasomotor center in the brain. Blood pressure is governed via balancing between sympathetic and parasympathetic activity which in turn influences vasodilation. Therefore, low blood pressure is the result of reduction in heart rate or vasodilation (37). During exercise, blood pressure is altered by central command activation and exercise pressor reflex. Central command will stimulate sympathetic nerve activity, which leads to increase cardiac contractility. As a result, mean arterial blood pressure will increase. Furthermore, exercise pressor reflex which is located in skeletal muscle will be stimulated via activation of chemical substances during exercise. It sends afferent signaling to the brain and blood pressure alteration will be occurred afterward (34, 38).

2.4 Respiratory system and exercise

Breathing is regulated by alteration of main chemical substance concentrations in arterial blood which are oxygen (O_2), carbon dioxide (CO_2) and hydrogen ion (H^+). These substances can affect both peripheral and central breathing controllers located in vessels around the neck and medulla oblongata of the brain stem respectively. Under low O_2 concentration, peripheral chemoreceptor at carotid and aortic bodies will be responsible and induce higher pulmonary ventilation (V_E). High concentration of CO_2 and H^+ also stimulate respiratory ventilation via central chemoreceptor located at ventral surface of the medulla oblongata (39). When both peripheral and central chemoreceptors are stimulated, ventilation might be increased from 5-6 L/min up to 100 L/min during exercise. Ventilation is normally determined by increasing in tidal volume (VT) and respiratory rate (RR). These changes are adjusted in order to control O_2 and CO_2 in the narrow ranges (40). At the beginning of exercise, pulmonary ventilation is proportionately adjusted due mainly to O_2 uptake and CO_2 production. However, during exercise between 40-60%VO_{2max}, pulmonary ventilation is more increased than O_2 uptake and CO_2 produced (41). Thus there are other governing factors on respiratory ventilation. For example, when exercise is progressively performed at the near maximum intensity, H^+ concentration in arterial blood remarkably increased, leading to decrease of blood pH which indicates acidosis(39). Therefore, stimulated ventilation is occurred to modulate that change.

2.5 Exercise metabolism

Exercise is caused of increased metabolism to generate energy supply for the active muscle. This metabolism is also regulated by some corresponding hormones during exercise. Metabolism rate during exercise depends on exercise type which is identified by intensity, and duration. Basically, there are four classes of energy substances consisted of compounds of high phosphoryl transfer potential, carbohydrate, lipids and protein are mainly required for increasing energy supply (42). Carbohydrate and lipids which are most essential sources, as well as associated hormones during exercise will be detailed as follow:

2.5.1 Effect of exercise on carbohydrate metabolism

Generally, glucose entered into human body is mainly stored in liver and muscles as glycogen form. Besides glycogen breakdown for substrate availability increases during exercise, glucose from the blood is up taken by exercising muscle. It is because of augmented blood flow and also enhanced glucose transporter called as GLUT4 (42). GLUT4 which is located in intracellular vesicles will be stimulated to be surrounding plasma membrane and responsible to allow glucose into active muscle. Considering amount of energy efficiency, oxygen is played an important role for glucose in aerobic pathway. However, high intensity of exercise can cause to be limited oxygen availability. Consequently, it will be shifted to lactate system to support the demand for high intensity. Lactate is, actually, one of substrates for gluconeogenesis which is process of glucose synthesis during exercise. It is transported to liver in Cori cycle for glucose generation. Once glycogen in muscle and liver is depleted, blood glucose levels will be falling, leading to decrease exercise performance. On the other hand, carbohydrate will be led to broken down aerobically during constantly moderate intensity exercise.

2.5.2 Effect of exercise on lipid metabolism

Fatty acid and triacylglycerol, types of lipid in human body, are most important during exercise. Triacylglycerol breakdown called lipolysis is augmented by effect of epinephrine from adrenals and norepinephrine sympathetic nerve activation (42). Moreover, rate of insulin secretion promoted triacylglycerol synthesis is decreased leading to be more lipolysis. In addition, oxidation of fatty acids in muscle resulted by increased fatty acid delivery in blood circulation and augmented lipolysis in adipose tissue is stimulated during exercise. Fatty acids availability serves to light and moderate intensity of exercise because its entry rate into muscle is low, as well as low rate of ATP synthesis from fatty acid oxidation. Therefore, high intensity exercise cannot be supported by this substrate.

2.5.3 Effect of hormones on glucose regulation during exercise

It is well known that glucose, the smallest substrate of carbohydrate, is essential energy substrate during exercise, thus, glucose concentration is regulated in order to maintain exercise performance. Hormones included epinephrine, glucagon, and insulin play an essential role to influence glucose regulation (42). During exercise, glycogen breakdown in muscle and liver will be increased by dramatically increased epinephrine action. Moreover, it stimulates more glucose synthesis in liver to blood circulation therefore exercising muscle will be derived sufficient glucose during exercise. Like epinephrine, glucagon acts to raise glucose concentration resulted from increased glycogen breakdown, as well as glucose synthesis in the liver. Insulin stimulates glucose uptake into muscle cells, liver and adipose tissue on the other hand. Therefore, it will be attenuated its secretion in order to permit glucose availability during exercise.

2.6 Energy drink

Many types of energy products have been introduced both for the public and athletes. The beverages contained stimulants claim to increase energy for performance. The substances contained in beverages composed of both macro- and micronutrients, but in varieties of compositions and amount. The term macronutrient is defined as the main energy sources for human living, including carbohydrates, protein and fat. Micronutrient, on the other hands, refers to other sources that play important roles as catalyst for biochemical reactions, the generating of energy metabolism, muscle protein-repaired and antioxidant. In general, exercise can influence to metabolic pathway, leading to more micronutrient requirement in the body (43, 44). Thus, vigorously trained-man may undergo insufficient micronutrient, and their performance would be reduced. Energy drinks might be affected to maintain the physical performance during longer exercise, it might be involved one or more physical performance components, such as speed, strength, and endurance.

2.7 Water, electrolytes and vitamins on performance

Supplements consumption is aimed to enhance performance in most sports. Because of complexity of ingredients, administration, dosage and effective duration, sometimes it may induce adverse effect to consumers (45). This means it requires knowledge and recommendations to improve understanding among athletes and those who need supplements. American College of Sport Medicine (ACSM) recommended that athletes should consume liquid before exercise or competition to administration balance between liquid and electrolytes, which can be influenced to performance. In addition, athletes should gradually consume liquid during exercise to eliminate sweat loss more than 2% of body weight. It was reported that sweat loss more than 2% of body weight related with impaired cardiovascular, thermoregulatory and central nervous functions (46) and possibly leads to attenuate performance. Moreover, liquid with electrolytes consumed after exercise is also emphasized to replace those liquid and electrolytes loss during exercise (5). It was found that some electrolytes loss, such as sodium (Na^+), potassium (K^+) and chloride (Cl^-), can negatively affect muscle function (47, 48). Within narrow ranges of these electrolytes on physiologic function, they should be provided at proper amount (48, 49) (Table 2.4). On the other hand, negative effects on performance will be found if they are insufficiently provided those essential electrolytes (Table 2.5). In addition, it was recommended that carbohydrate consumed at approximate rate of 30-60 g/h could maintain blood glucose level when exercise duration is more than 60-min (5). Furthermore, vitamin, which is categorized in micronutrient, can impair performance, especially in athletes who insufficiently consume some vitamins (Table 2.6) (50). Moreover, some study found that vitamin could reduce arrhythmia, which is an abnormal rhythm of heart (51). Thus, types and amounts of electrolytes and vitamins make them to be consumed with care.

Table 2.4 Illustration of types and functions of essential electrolytes (48).

Electrolyte	Primary roles	Target dose per 250 ml of fluid	Performance daily intake
Sodium	Muscle contraction		
	Nerve transmission	150-250 mg	1,500-4,500 mg
Chloride	Peak muscle function	45-75 mg	-
Potassium	Muscle contraction		
	Nerve transmission	50-80 mg	2,500-4,000 mg
	Glycogen formation		
Magnesium	Muscle relaxation		
	ATP (energy) production	20-30 mg	400-800 mg

Table 2.5 Illustration essential electrolytes and deficiency effects on performance (44).

Exercise-related functions of selected minerals		
	Functions	Deficiency sign or symptom
Magnesium	Energy metabolism, nerve conduction, muscle contraction	Muscle weakness, nausea, irritability
Iron	Hemoglobin synthesis	Anemia, cognitive impairment, immune abnormalities
Zinc	Nucleic acid synthesis, glycolysis, carbon dioxide removal	Loss of appetite, growth retardation
Chromium	Glucose metabolism	Glucose intolerance

Table 2.6 Illustration of essential vitamins and deficiency effects (44).

	Function	Deficiency sign or symptom
Thiamin (B1)	Carbohydrate and amino acid metabolism	Weakness, decreased endurance
Riboflavin (B2)	Oxidative metabolism, electron transport system	Nervous system function
Niacin	Oxidative metabolism, electron transport system	Irritability, diarrhea
Pyridoxine (B6)	Gluconeogenesis,	Dermatitis, convulsion
Cyanocobalamin (B12)	Hemoglobin formation	Neurologic symptoms
Folic acid	Hemoglobin and nucleic formation	Anemia, fatigue
Ascobic acid (vitamin C)	Antioxidant	Fatigue, loss of appetite
Retinol (vitamin A)	Antioxidant	Loss of appetite
Tocopherol (vitamin E)	Antioxidant	Nerve and muscle damage

2.8 Effect of sports drink on performance

Sports drink, kinds of beverage used to rehydrate or protect fluid loss, is typically contained carbohydrate and essential electrolytes which may influence to performance during exercise or sport competition (52). Normally, carbohydrate which is one of macronutrients is mainly used as essential energy during exercise or sports competition, especially during intense and prolonged exercise (53). The early depleted glycogen may represent inadequate endogenous glycogen storage, as the consequence, it may not maintain throughout exercise duration. One study reported that muscle glycogen concentration was nearly empty in the first half time during soccer match (53). After muscle biopsy, 3 folds muscle glycogen concentration was depleted when compared with baseline values (pre-competition: 96 mmol/kg wet weight, half time: 32 mmol/kg wet weight). Moreover, it was found 10 folds of muscle glycogen depletion after the end of match (end of match: 9 mmol/kg). Thus, it is important to be replaced glycogen lost during exercise. McLellan TM (2014) (11) suggested that athletes should consume at least 1g of carbohydrate/kg body weight/ hour to replace glycogen between (10) and after competition. It may be an optimal rate to enhance the next endurance performance.

Recent studies reported effects of sports drink on performance before, during or after exercise. It was suggested that sports drink can improve performance by 6.5% and also lower exercise heart rate (22). Similarly, Zachweija (1993)(8) stated that carbohydrate ingestion can induce muscle glycogen re-synthesis after exercise. This study found that the rate of glycogen repletion during the first 2 hours of recovery period after consuming 24% of carbohydrate was increased on male cyclist. Moreover, sports drink contained 6% carbohydrate improved 80-km time trial performance on trained male cyclist and also found higher heart rate after ingested drink (14). In addition, consuming carbohydrate solution, which is contained 1.1g of carbohydrate/kg body weight, 1 hour before 45-min exercise at 70%VO_{2max} found improved power output and time trial of exercise were + 13.1% and -12.5% respectively when compared with placebo (54). This study also showed increased blood glucose concentration; so, it may be caused from exogenous carbohydrate oxidation that improved performance. Rollo I (2009) (6) also showed the results of 6.4 % carbohydrate plus electrolyte solution consuming before test that running distance was greater during 1 hour performed on treadmill. However, there is some study showed controversial effect of sports drink on performance. It was reported that after ingested drink contained 6% carbohydrate did not enhance gastric emptying and time trial performance during performed cycling exercise at 60 % VO_{2peak} for 85-min (55). Factors which influence to gastric emptying during prolonged exercise are carbohydrate sources and concentration (56). These may affect affected on performance differently as shown in Table 2.7. Commercial drink, which was contained 100% maltrodextrin plus electrolyte, could extend time to exhaustion during intermittent high intensity running in younger players (19). However, it was not found to affect sprint time performance. In addition, commercial sports drink contained mixed maltodextrin and fructose was shown to enhance exercise duration (57). It may indicate to be higher total and exogenous carbohydrate rates during steady state of exercise performing. Furthermore, the effect was compared effect between 2 commercial sport drinks which were composed of different carbohydrates on exercise performance (15).

Not all sports drinks provide good results on performance. Some evidence showed the negative result on decreased time to exhaustion after ingested sports drink (58). This sports drink was mentioned to contain galactose, which may be lower glucose and insulin concentration when compared with placebo. Many researches have been investigated how sports drink affected to their specific skill. One study conducted in mimic soccer match (59) and it was found that sports drink contained 7% of carbohydrate did not significantly improved agility and time to exhaustion. Additionally, Gomes RV (2013) (7) found similar effect of sports drink to tennis-specific skills. It was reported that number of stroke per rally, first and second service in and first and second return in were not different between sports drink and placebo. Controversy, one study (60) demonstrated the positive outcome after consumed sports drink on dribbling precision, and also increased concentration in soccer players.

Table 2.7 Details of sport drinks containing different amounts of carbohydrate and electrolytes, phases of supplement and changes in performance (* commercial sports drink)

Compositions	%CHO	Phase of supplement	Subjects	Type of testing	Result on performance	References
Maltodextrin, dextrose, Na^+ (*)	8%	Prior to subsequent exercise	Recreational male cyclists	Cycling	↑Performance times	Roberts JD et al., 2012
Maltodextrin, Na^+, K^+, Mg^{2+}, Ca^{2+}	6%	Prior to subsequent exercise	Recreational players	Running	↑Time to exhaustion	Phillips SM et al., 2010
Galactose	8%	Before exercise	Recreational male cyclists	Cycling	↓Time trial power output	Stannard SR et al., 2009
CHO, sugar, Na^+ (*)	6.4%	Before exercise	Endurance-trained male runners	Running	↑Running distance (on 1 hr)	Rollo I et al., 2009

Table 2.7 Details of sport drinks containing different amounts of carbohydrate and electrolytes, phases of supplement and changes in performance (* commercial sports drink) (cont.)

Compositions	%CHO	Phase of supplement	Subjects	Type of testing	Result on performance	References
Dextrose, fructose, maltose, Na^+, K^+	6.5%	Prior to subsequent exercise	Recreational male runners	Running	↑Running time trial	K. Tsintzas et al., 2003
CHO, Na^+, K^+	6.9%	Prior to subsequent exercise	Endurance-trained males	Running	↑Time to exhaustion	Wong SH et al., 2000
CHO, Na^+, K^+	6.4%	After exercise	Healthy male university soccer players	Running and soccer skill test	↑Soccer skill and sprint performance	Ali A et al., 2007
Maltodextrin	6%	Before exercise	Tennis male athletes	Simulated tennis match play	↔ specific tennis skill (number of stroke, service in and return in)	Gomes R. V. et al., 2013

CHAPTER III
MATERIALS AND METHODS

The aim of this study was to examine acute effects of three different drinks composed of sports drink, placebo and water on subsequent physical performance following glycogen depletion exercise on cardiorespiratory and biochemistry functions. This study was preceded by randomized controlled trial with cross-over study designed. The process of this study was done as follows:

3.1 Subjects

Fifteen healthy Thai males, with age-ranged between 18-25 years old, were recruited. Subjects were asked to complete the questionnaire about their health conditions (61) and physical activity (62). Subjects were screened by and interview to ensure they were in a good condition and ready for exercise test. Sample size was determined using time to exhaustion data of previous research (63). It was calculated using G*power with power and significant level setting at 0.80 and 0.05, respectively. This experiment was approved by Ethical Committee on Human Study of Mahidol University (COA No. MU-CIRB 2016/ 111.2408). All subjects were instructed about experimental protocol before signing their informed consent forms. Subjects were recruited according to the following criteria:

Inclusion criteria: Subjects were recruited according to these criteria

-Male with age-ranged 18-25 years old

-Healthy with no exercise restriction

-Physically active lifestyle determined by averages peak oxygen consumption (40.7-50.5 ml/kg/min for age-ranged between 17-19 years old; 38.0-47.0 ml/kg/min for age-ranged between 20-29 years old) (27)

-Able to complete all 3 testing trials

Exclusion criteria: Subjects were excluded according to one of these criteria.

-Had signs and symptoms of infection/inflammation or cardiopulmonary diseases, musculoskeletal disorders or metabolic diseases.

-Had taken any drugs or supplements which may affect cardiorespiratory, metabolic system and level of consciousness e.g. beta-blocker, caffeine- containing supplement, conventional antihistamines etc.

3.2 Equipment

Data composed of energy expenditure, cardiorespiratory, anthropometry variables, performance and urine specific gravity were collected at rest, during and after exercise using the following sets of equipment:

3.2.1 Anthropometry

Basic anthropometric variables were collected using bioimpedance analysis machine (Omron, HBF362, Japan) to evaluate body compositions including weight, height, fat mass and muscle mass. (Figure 3.1)

Figure 3.1 Bioimpedance machine (Omron, HBF362, Japan)

3.2.2 Blood pressure and heart rate

Blood pressure and heart rate were monitored using a sphygmomanometer and telemetry heart rate monitor (Polar, Finland) (Figure 3.2 and 3.3).

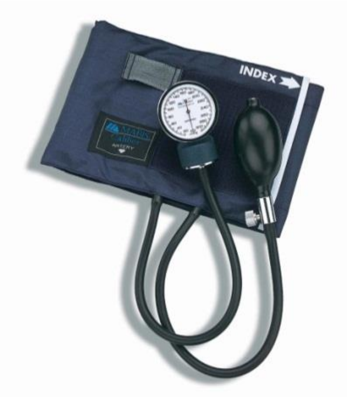

Figure 3.2 Sphygmomanometer (Blue Nylon Cuff, China)

Figure 3.3 Heart rate monitor (Polar T31, England)

3.2.3 Hydration status

Urine specific gravity, an indicator for hydration status, was assessed using refractometer (ANTAGO manual, Figure 3.4). It was drop the distilled water on the oval dark area and close the cover to calibrate before using. The shadow must be seen at a zero point. It was adjusted scale in order to correction the zero point. Open the cover and dry the dark area using soft tissue paper.

Figure 3.4 Refractometer (ATAGO manual, Japan)

3.2.4 Indirect calorimetry

The present study used a telemetry gas analyser (Oxycon Mobile, Jager, Germany) attached around subject's chest to measure respiratory function (Figure 3.5). Main cardiorespiratory variables including oxygen consumption, carbon dioxide production, tidal volume, pulmonary ventilation and respiratory exchange ratio were collected. There were steps to calibrate this system. First of all, setting up equipment and followed by 15 min program warm-up. Secondly, volume, gas analyzer and gas flow calibration were done, respectively.

Figure 3.5 Telemetry Gas Analyzer (Jaeger Oxycon Mobile, Germany)

3.2.5 Cardiac function

Cardiovascular system was evaluated using a telemetry impedance cardiography (Physioflow, France) (Figure 3.6). According to the standard procedure, six electrodes were attached on body surfaces, around the chest, to synchronously monitor cardiac rhythm and electrical signals then convert to cardiovascular variables including stroke volume (SV), heart rate (HR), cardiac output (CO), end-diastolic volume (EDV), ejection fraction (EF), and systemic vascular resistance (SVR).

Figure 3.6 Telemetry impedance cardiography (Physioflow, France)

3.2.6 Exercise equipment

A cycle ergometer (Monark 828E, Sweden) was used as performance testing device to evaluate work done, power output, endurance time (Figure 3.7). It was calibrated using 0.5 kg of weight applied. It was needed to ensure that there is no load on the device. Weight which was beside equipment must be at zero scale. It should be moved to 0.5 scales after 0.5 kg of weight applied.

Figure 3.7 Stationary cycle ergometer (Monark 828E, Sweden)

3.2.7 Blood glucose and lactate

Blood samples were collected via finger tips which then analyze for glucose (Glu) (ACCU-CHECK) and lactate (LA) (LACTATE SCOUT) levels using set of devices (Figure 3.8) strips and needle puncture under aseptic techniques. Cotton with 70%alcohol was applied to finger then use needle puncture on finger. Initial small amount of blood was wiped away to eliminate contamination of alcohol. After that, the second blood dropped was obtained again and used the strip sensor to analyze.

Figure 3.8 Hand-held glucose and lactate analyzer (ACCU-CHECK and LACTATE SCOUT)

3.2.8 Miscellaneous

Supporting items including 70%alchohol, cotton, stop watch and rating perceived exertion chart (Borg scale 6-20) were employed.

3.3 Protocol

This protocol (Figure 3.9) was conducted at 9 a.m. in the air-conditioned laboratory where temperature and humidity were controlled within the ranges of 24 ±0.9 ° C and 56.7 ±2.9% relative humidity. All subjects were instructed to refrain from heavy exercise and caffeine-containing beverage within 48 hours before the test, adequately slept at least 8 hours, and consumed similar meals within 4 individual's trials. Subjected were asked to recall for 3 days diet before the testing date. All visits were separated with at least 1 week apart.

3.3.1 Maximum oxygen consumption test

On the first visit (Figure 3.9), data collections for anthropometry, including weight, height, body composition, urine specific gravity (USG), resting blood pressure (BP) and heart rate (HR) were initially evaluated. It was compulsory to test for peak oxygen consumption (VO_{2peak}) using progressive graded exercise test on cycle ergometer. After 5-min warm-up at 0.5kp, pedal cadence of 60 rpm, incrementally workload of 0.5 kp, 60 revolutions per min, for every 3 min interval until reaching their maximal ability (61). Termination was judged according to one of the following: a) irregular pedal cadence of less than 60 rpm over 30-second despite the encouragement; b) subject requests to stop; c) approaching targeted HR (it was determined by 220-age) and RPE ($\geq$ 17 using Borg's scale). VO_{2peak} was either predicted from last 30-second or when the amount of oxygen uptake was increased less than 150 ml/min (VO_2 plateau) despite an additional workload (64). Maximum power output (P_{max}) was calculated at the end of the test to indicate individual workload used in the next for glycogen depletion session.

Termination of incrementally graded exercise test (61):

1. Chest pain during testing
2. Systolic BP decreases > 10 mmHg with increased workload
3. Systolic BP> 250 mmHg or diastolic BP > 115 mmHg during exercise
4. Abnormal breathing, leg cramps
5. Poor perfusion (e.g. ataxia, dizziness or nausea)
6. No changes of HR although increasing of workload
7. Abnormal signs of heart rhythm
8. Severely physical or verbal fatigue
9. Participants request to terminate

3.3.2 Visit 2-4

The design of this study was a randomized double-blinded where each subject was provided either sports drink (Sponsor™), placebo or water for 3 separated trials. Each trial was divided into 3 consecutive components; glycogen depletion, 2 hours for recovery period and endurance exercise.

3.3.2.1 Glycogen depletion phase (Figure 3.9, 3.10): Prior to this test, subjects were re-assessed for body compositions, USG, BP, HR, LA and Glu levels as baselines. After 2-min warm up at 0.5kp, pedal cadence of 60 rpm, intensities were initially adjusted at 90%P_{max} for 2-min, with 50%P_{max} (80 rpm, 2-min interval). Followed by workloads between 60-90% P_{max} for 2-min, with 50% P_{max} (80 rpm, 2-min interval) intersperse between stages (63). If the cadences were experienced to be below than 70 rpm more than 30 sec, the power output was considerably reduced to 80%P_{max}, 70%P_{max} and 60%P_{max} respectively. If they were still experiencing to be below than 70 rpm more than 30 sec even when performing at 60%P_{max} which is the last stage, they were terminated or they request to stop themselves. During testing, cardiorespiratory and metabolic parameters were collected. LA and Glu were immediately collected after test. In addition, USG and weight loss were measured to evaluate hydration status.

3.3.2.2 Recovery period phase: Subjects were allowed to rest for 2 hours in the temperature-controlled room and randomly provided different drinks (Table 3.1). Amount of energy consumed from the drink was calculated at 1 gram of carbohydrate (CHO)/kilogram (kg) body weight (BW) (65). For example, subject whose weight was 70 kg consumed 603.45 ml (calculated from (70 kg BW * 1 gm CHO/kg BW * 250 ml/ 29 g of CHO). Within 2 hrs. recovery period, the amount of energy drink given was divided into 3 parts: first at 50%body weight (BW) at immediately after finished glycogen depletion, then at two 25%BW at 30 minutes and 60 minutes respectively (63) (Table 3.1).

Table 3.1 Composition of 3 different drinks

Compositions	Sports drink	Placebo	Water
Volume per bottle (ml)	250	250	250
Total energy (kcal)	120	120	-
CHO (%, gm)	10, 29	10, 29	-
Sucrose (%, gm)	7.0, 17.5	7.0, 17.5	-
Dextrose (%, gm)	4.0, 10	4.0, 10	-
NaCl (%, gm)	0.13, 0.33	-	-
KCl (%, gm)	0.03, 0.075	-	-
Osmolarity (m.Osm/L)	168.5	155.7	-

* Manufactured products from T.C. Pharmaceutical Industries CO., LTD.

3.3.2.3 Endurance exercise phase (Figure 3.9, 3.11): Similar baseline parameters, as shown in glycogen depletion session, were re-collected prior to this exercise session. Subjects then performed endurance exercise on cycle ergometer. This test composed of 5-min stretching, followed by 2-min warm up on cycle ergometer at 0.5 kp at 60 rpm (61) and cycling at steady workload of 70%VO_{2peak} at 60 rpm until volitional exhaustion (63)**.** Exercise termination was considered if their revolutions either less than 60 rpm more than 30 second or voluntarily stop. Total FAT-CHO oxidation was calculated using equations as followed (66).

$$CHO_{TOT} = 4.585(VCO_2) - 3.226(VO_2)$$

$$FAT_{TOT} = 1.695\ (VO_2) - 1.701\ (VCO_2)$$

Time to exhaustion was assessed using stop watch, and work done was calculated after completed this exercise session.

3.4 Statistical analyses

Data was analyzed by using SPSS software (version 17.0; SPSS Inc., Chicago, IL, USA). All data was calculated from these statistical analyses as follows:

- Shapiro wilk test was used to test normal distribution of all variables before choosing the appropriate statistics.

- One-way repeated measures analyses of variance was used to compare the mean values of SV, RR, V_E, VO_2, VCO_2, RER, FAT-CHO oxidation, TTE and WD among three trials, while Friedman test was used to compare difference of HR, CO, SVR, EDV, EF, VT, LA and Glu among three trials.

- Post-hoc analysis of significant difference of SV, RR, V_E, VO_2, VCO_2, RER, FAT-CHO oxidation, TTE and WD was expressed using Bonferroni, while HR, CO, SVR, EDV, EF, VT, LA and Glu was expressed using Wilcoxon Signed-Rank Test.

- The difference between pre- and post-exercise of SV, RR, V_E, VO_2, VCO_2 and RER within each group was performed using Paired sample t-test, while HR, CO, SVR, EDV, EF, VT, LA and Glu was performed using Wilcoxon Signed-Rank Test.

- Data analyzed from one-way repeated measures analyses of variance and Paired sample t-test are expressed as mean ± SEMs, while data analyzed from Friedman test and Wilcoxon Signed-Rank Test are expressed as median ± interquartile. The probability < 0.05 was used to determine the significant difference.

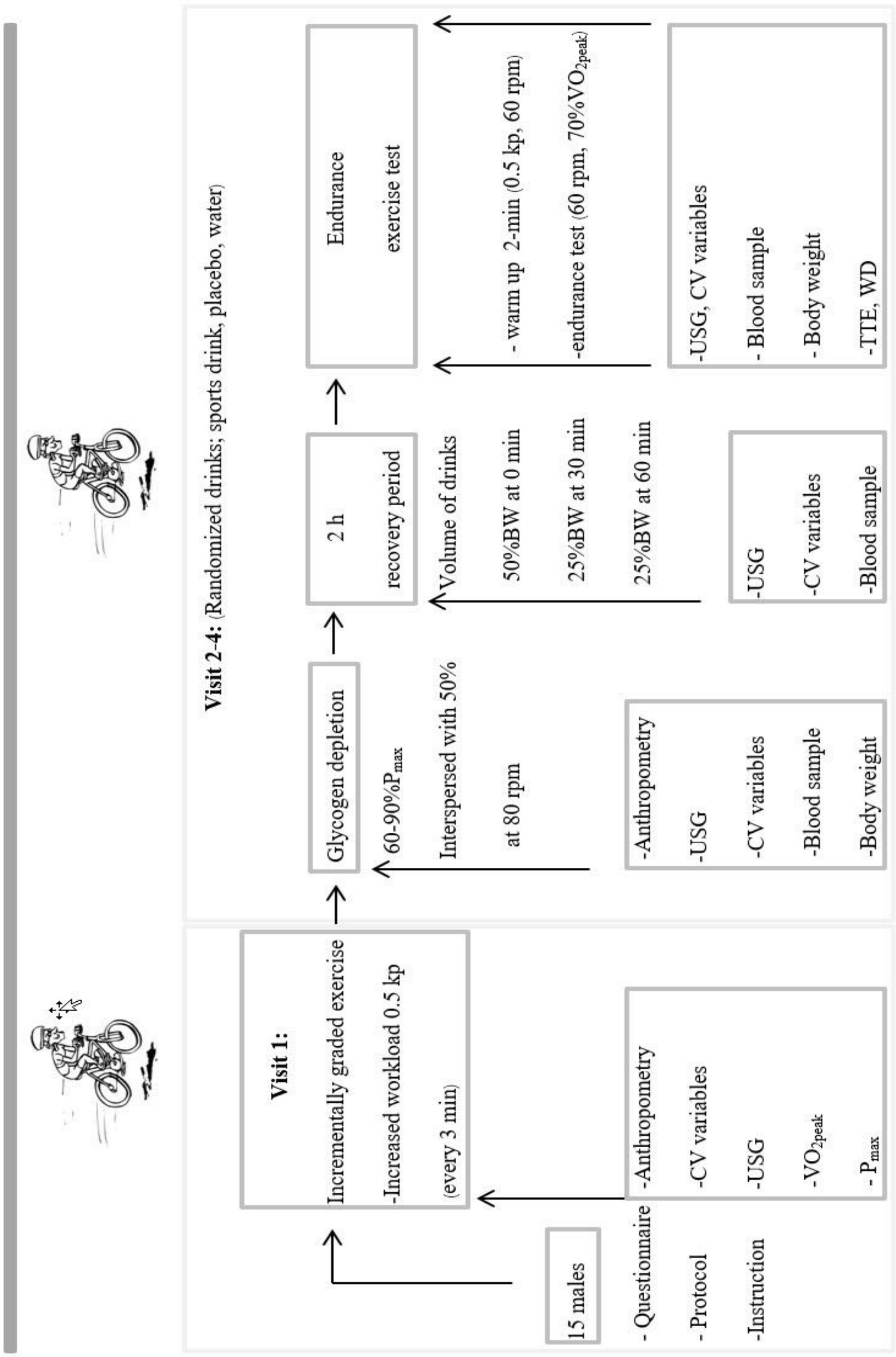

Figure 3.9 Experimental protocol diagrams

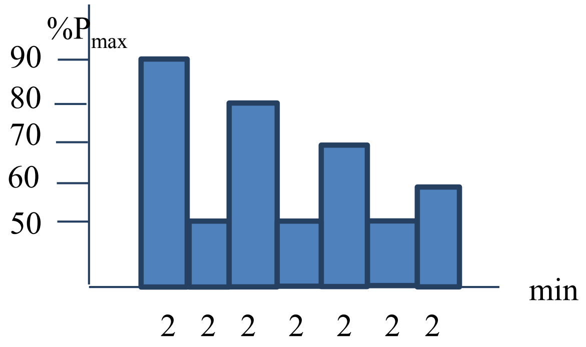

Figure 3.10 Glycogen depletion protocol

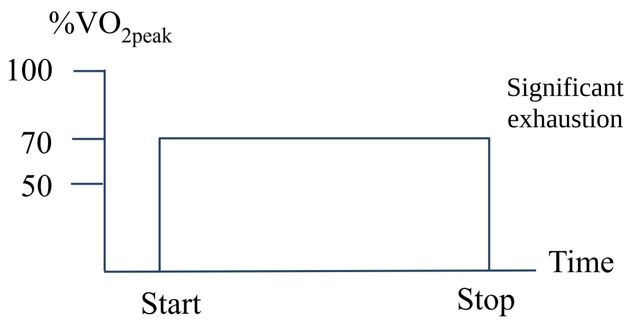

Figure 3.11 Endurance exercise protocol

CHAPTER IV
RESULTS

4.1 General characteristics

Fifteen Thai males successfully completed health screenings and physical activity questionnaires to meet inclusion criteria previously determined. One of them was excluded from this study due to unable to complete 3 trials of previously determined experimental protocol. Finally, fourteen healthy Thai males were recruited in this study. Ages, systolic and diastolic blood pressures, height, weight, %body fat, %muscle mass, body mass index and VO_{2peak} were shown in Table 4.1.

Table 4.1 General characteristics of Thai males subjects participated in this study (n=14). Values are mean ± SD.

	VALUES
Age (yr)	21.50 ± 1.65
SBP (mmHg)	116.14 ± 7.32
DBP (mmHg)	67.21 ± 3.76
Height (cm)	174.00 ± 5.30
Weight (kg)	68.37 ± 6.74
Body fat (%)	14.09 ± 3.61
Muscle (%)	37.05 ± 1.36
Body mass index ($kg.m^{-2}$)	22.54 ± 1.96
VO_{2peak} ($ml.kg^{-1}.min^{-1}$)	40.71 ± 2.72

To determine effects of sports drink on physiologic and metabolic function, data were serially measured at four intervals, before and after two consecutive exercises including glycogen depletion (GD) and endurance exercise (EX). To prove the effects of sports drink, three interventions including water (WT), placebo (PL) and sports drink (SD) were randomly employed. The above fluids was produced and blinded by the manufacturer (T.C. Pharmaceutical Industries CO., LTD.). During recovery period, these fluids were randomly intervened by investigator and provided to subjects in an opaque container with lid. Thus, the subjects did not realize which kind of drinks to be taken. This study was then a double-blinded designed.

4.2 Changes in cardiorespiratory function during glycogen depletion exercise

All cardiorespiratory variables at rest in all three conditions were no difference among conditions (Table 4.2). Exercise-induced glycogen depletion (GD) remarkably increased in Heart rate (HR), stroke volume (SV), cardiac output (CO), end-diastolic volume (EDV), ejection fraction (EF), respiratory rate (RR), tidal volume (VT), and minute ventilation (V_E) compared to the corresponding resting values ($p < 0.05$). On the other hand, lower significant differences of systemic vascular resistance (SVR) in all three conditions compared to the corresponding resting values were observed ($p < 0.05$). After completed GD, there were no differences of all cardiorespiratory variables among three conditions.

Table 4.2 Comparisons of pre- and post-exercise cardiorespiratory variables during glycogen depletion exercise in Water (WT), Placebo (PL) and Sports drink (SD) conditions (n=14). SV, RR and V_E are means and SEMs; HR, CO, EDV, SVR, EF and VT are medians and interquartile.

Variables		Conditions		
		WT	PL	SD
HR (bpm)	Pre-GD	73.50 ± 9.25	75.50 ± 13.75	74.00 ± 13.50
	Post-GD	170.50 ± 24.50 *	173.50 ± 18.00 *	174.00 ± 12.00 *
SV (ml)	Pre-GD	82.8 ± 4.1	88.1 ± 3.9	85.9 ± 3.8
	Post-GD	98.0 ± 6.6*	97.0 ± 6.6*	103.6 ± 6.5*
CO (L/min)	Pre-GD	5.87 ± 1.70	6.73 ± 2.48	6.00 ± 1.38
	Post-GD	15.63 ± 3.43 *	16.35 ± 5.93 *	17.23 ± 5.16 *
EDV (ml)	Pre-GD	147.7 ± 19.35	139.7 ± 16.20	141.9 ± 29.63
	Post-GD	157.0 ± 10.62*	148.8 ± 28.22*	156.1 ±66.79*
SVR	Pre-GD	1092.5 ± 386.50	1013.5 ± 159.75	1108 ± 389.75
(dyne./s/cm^3)	Post-GD	397.83 ± 134.99 *	372.16 ± 107.33 *	380.5 ± 141.16 *
EF (%)	Pre-GD	59.5 ± 19.03	65.6 ± 24.78	59.9 ± 15.10
	Post-GD	65.9 ± 14.36*	63.1 ± 26.82*	62.5 ± 18.77*
RR	Pre-GD	18.92 ± 0.71	20.14 ± 0.76	18.78 ± 0.72
(breaths/min)	Post-GD	48.71 ± 2.51*	50.5 ± 2.61 *	50.28 ± 2.43 *
VT (L/breath)	Pre-GD	0.76 ± 0.19	0.73 ± 0.16	0.84 ± 0.26
	Post-GD	1.85 ± 0.53*	1.74 ± 0.52 *	1.76 ± 0.58 *
V_E (L/min)	Pre-GD	15.42 ± 0.86	15 ± 0.59	15.85 ± 0.67
	Post-GD	85.97 ± 5.64*	89.42 ± 4.14 *	90.26 ± 5.48 *

Symbols

[a]: significant difference between WT-PL; [b]: significant difference between WT-SD; [c]: significant difference between PL-SD; [*]: significant difference within group; All values are compared at $p < 0.05$.

4.3 Changes in metabolic and blood chemistry variables during glycogen depletion

All blood chemistry including blood lactate (LA) and blood glucose (Glu), and metabolic variables including oxygen consumption (VO_2) , carbon dioxide production (VCO_2) and respiratory exchange ratio (RER) at rest in all three conditions were no difference among conditions (Table 4.3) . Exercise-induced glycogen depletion (GD) significantly increased in LA, VO_2, VCO_2 and RER compared to the corresponding resting values ($p < 0.05$). Although lower Glu was shown in all three conditions compared to the corresponding resting values, but the differences were not observed. There were no differences of all blood chemistry and metabolic variables among three conditions after completed GD.

Table 4.3 Comparisons of pre- and post-exercise blood chemistry and metabolic variables during glycogen depletion exercise in Water (WT), Placebo (PL) and Sports drink (SD) conditions (n=14). VO_2, VCO_2 and RER are means and SEMs; LA and Glu are medians and interquartile.

Variables		**Conditions**		
		WT	PL	SD
LA (mMol/L)	Pre-GD	1.45 ± 0.98	1.35 ± 1.07	1.45 ± 0.87
	Post-GD	12 ± 3.90*	11.75 ± 5.03*	11.35 ± 1.92*
Glu (mg/dL)	Pre-GD	98.00 ± 13.50	96.50 ± 14.25	94.50 ± 18.50
	Post-GD	90.00 ± 27.75	87.50 ± 19.75	91.50 ± 20.00
VO_2 (L/min/kg)	Pre-GD	0.30 ± 0.01	0.32 ± 0.01	0.31 ± 0.01
	Post-GD	2.32 ± 0.10*	2.38 ± 0.10 *	2.40 ± 0.12*
VCO_2 (L/min/kg)	Pre-GD	0.24 ± 0.01	0.27 ± 0.01	0.26 ± 0.01
	Post-GD	2.44 ± 0.17*	2.55 ± 0.17 *	2.61 ± 0.18*
RER	Pre-GD	0.81 ± 0.02	0.83 ± 0.01	0.83 ± 0.02
	Post-GD	1.04 ± 0.03*	1.06 ± 0.03*	1.07 ± 0.03*

Symbols

[a]: significant difference between WT-PL; [b]: significant difference between WT-SD; [c]: significant difference between PL-SD; [*]: significant difference within conditions; All values are compared at $p < 0.05$.

4.4 Changes in hydration status

At baseline, there were no differences of urine specific gravity (USG) among three conditions during glycogen depletion (GD) and endurance exercise (EX) (Table 4.4). At the end of GD and EX, there were also no differences among three conditions. Similarly, no differences of body weight changes during GD and EX were observed.

Table 4.4 Comparisons of pre- and post-exercise urine specific gravity (USG) and body weight changes (ΔBW) during glycogen depletion and endurance exercise in Water (WT), Placebo (PL) and Sports drink (SD) conditions (n=14). All values are means and SEMs.

Variables			**Conditions**		
			WT	PL	SD
Glycogen depletion (GD)					
	USG	Pre-GD	1.00 ± 0.0	1.00 ± 0.0	1.00 ± 0.0
		Post-GD	1.01 ± 0.0*	1.01 ± 0.0*	1.01 ± 0.0*
	% ΔBW		0.71 ± 0.15	0.70 ± 0.11	0.62 ± 0.08
Endurance exercise (EX)					
	USG	Pre-EX	1.00 ± 0.0	1.00 ± 0.0	1.00 ± 0.0
		Post-EX	1.01 ± 0.0*	1.01 ± 0.0*	1.01 ± 0.0*
	% ΔBW		1.06 ± 0.15	1.07 ± 0.16	1.37 ± 0.16

Symbols

[a]: significant difference between WT-PL; [b]: significant difference between WT-SD; [c]: significant difference between PL-SD; *: significant difference within conditions; All values are compared at $p < 0.05$.

4.5 Accumulated total work done during glycogen depletion exercise

After completed glycogen depletion exercise, there were no differences of total work done among three conditions (Figure 4.1).

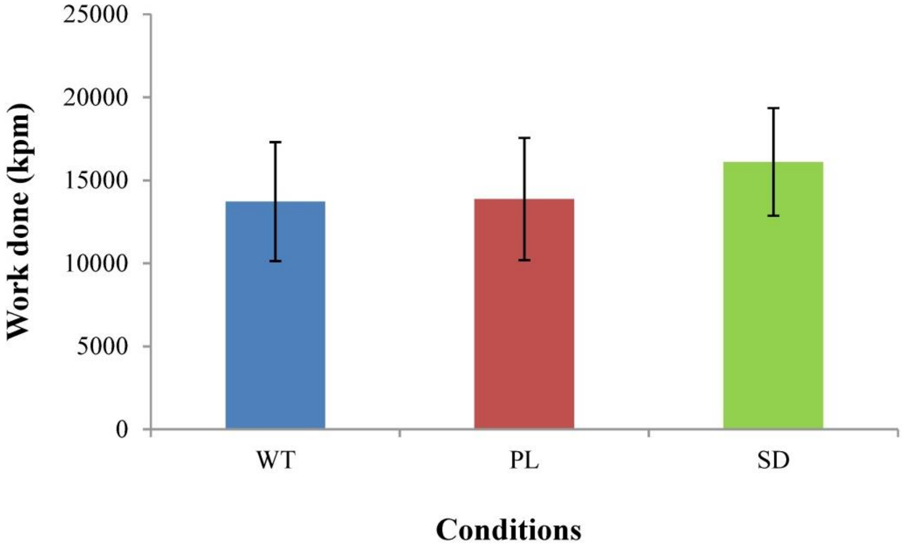

Figure 4.1 Comparisons of Accumulated total work done during glycogen depletion exercise in Water (WT, blue), Placebo (PL, red) and Sports drink (SD, green) (n=14). All values are means and SEMs.

Symbols

^{a}: significant difference between WT-PL; ^{b}: significant difference between WT-SD; ^{c}: significant difference between PL-SD; *: significant difference within conditions; All values are compared at $p < 0.05$.

4.6 Changes in cardiorespiratory function during endurance exercise

All cardiorespiratory variables at rest in all three conditions were no differences among conditions (Table 4.5). After completed endurance exercise, HR, CO, SVR, RR, VT, and V_E were significant differences in all three conditions compared with the corresponding values ($p < 0.05$). In addition, SV in SD condition, and EF in PL and SD conditions were significantly higher compared with corresponding values ($p < 0.05$). Moreover, EF in SD condition was significantly higher than WT condition ($p < 0.05$), significantly lower V_E in SD condition than WT and PL conditions were also observed ($p < 0.05$).

Table 4.5 Comparisons of pre- and post-exercise cardiorespiratory variables during endurance exercise in Water (WT), Placebo (PL) and Sports drink (SD) conditions (n=14). SV, RR and V_E are means and SEMs; HR, CO, EDV, SVR, EF and VT are medians and interquartile.

Variables		Conditions		
		WT	PL	SD
HR (bpm)	Pre-EX	77.50 ± 14.50	75.00 ± 18.25	80.50 ± 18.75
	Post-EX	154.50 ± 18.25 *	159.50 ± 20.75 *	159.00 ± 15.50 *
SV (ml)	Pre-EX	81.70 ± 3.15	82.13 ± 2.28	79.94 ± 4.06
	Post-EX	90.86 ± 5.31	92.08 ± 5.13	95.61 ± 4.85 *
CO (L/min)	Pre-EX	6.57 ± 0.93	5.88 ± 2.33	5.80 ± 1.56
	Post-EX	15.48 ± 4.66 *	15.28 ± 4.80 *	14.66 ± 3.67 *
EDV (ml)	Pre-EX	132.00 ± 25.28	130.05 ± 39.40	130.55 ± 24.60
	Post-EX	144.50 ± 23.46	131.73 ± 31.56	140.13 ±37.98
SVR (dyne./s/cm^3)	Pre-EX	1025.00 ± 184.75	1092.50 ± 259.25	1061.50 ± 403.00
	Post-EX	431.83 ± 157.41 *	418.83 ± 119.00 *	425.00 ± 148.00 *
EF (%)	Pre-EX	60.50 ± 14.95	64.00 ± 18.20	62.40 ± 16.03
	Post-EX	64.75 ± 20.87	64.78 ± 19.73*	72.00 ± 13.31*,b
RR (breaths/min)	Pre-EX	20.85 ± 0.8	18.85 ± 0.95	19.28 ± 0.86
	Post-EX	41.64 ± 2.13 *	40.07 ± 1.16 *	39.21 ± 1.69 *
VT (L/breath)	Pre-EX	0.77 ± 0.24	0.76 ± 0.21	0.78 ± 0.25
	Post-EX	1.39 ± 0.42*	1.43 ± 0.37 *	1.27 ± 0.30 *
V_E (L/min)	Pre-EX	15.57 ± 2.95	14.5 ± 0.82	14.92 ± 0.68
	Post-EX	60.02 ± 2.95 *	57.9 ± 2.27 *	52.52 ± 2.36 *,b,c

Symbols

[a]: significant difference between WT-PL; [b]: significant difference between WT-SD; [c]: significant difference between PL-SD; [*]: significant difference within conditions; All values are compared at $p < 0.05$.

4.7 Changes in metabolic and blood chemistry variables during endurance exercise

4.7.1 Changes in LA, Glu, VO_2, VCO_2, and RER

All blood chemistry including LA, Glu, VO_2, VCO_2, and RER at rest in all three conditions were no differences among conditions (Table 4.6). After completed endurance exercise, all parameters were higher significant differences ($p < 0.05$) in all three conditions compared with corresponding values except Glu. However, there were no differences of all parameters among conditions.

Table 4.6 Comparisons of pre- and post-exercise blood chemistry and metabolic variables during endurance exercise in Water (WT), Placebo (PL) and Sports drink (SD) conditions (n=14). VO_2, VCO_2 and RER are means and SEMs; LA and Glu are medians and interquartile.

Variables		Conditions		
		WT	PL	SD
LA (mMol/L)	Pre-EX	1.55 ± 1.75	1.80 ± 1.13	1.95 ± 1.22
	Post-EX	2.70 ± 3.10*	3.15 ± 3.45*	3.20 ± 2.42*
Glu (mg/dL)	Pre-EX	91.00 ± 12.25	86.00 ± 15.25	90.00 ± 14.00
	Post-EX	88.00 ± 11.50	84.50 ± 11.50	87.50 ± 11.75
VO_2 (L/min/kg)	Pre-EX	0.31 ± 0.01	0.31 ± 0.01	0.32 ± 0.01
	Post-EX	2.03 ± 0.08*	2.01 ± 0.08 *	1.92 ± 0.07*
VCO_2 (L/min/kg)	Pre-EX	0.24 ± 0.01	0.25 ± 0.01	0.26 ± 0.01
	Post-EX	1.86 ± 0.08*	1.90 ± 0.08 *	1.79 ± 0.08*
RER	Pre-EX	0.81 ± 0.02	0.83 ± 0.01	0.83 ± 0.02
	Post-EX	1.04 ± 0.03*	1.06 ± 0.03*	1.07 ± 0.03*

Symbols

[a]: significant difference between WT-PL; [b]: significant difference between WT-SD; [c]: significant difference between PL-SD; [*]: significant difference within conditions; All values are compared at $p < 0.05$.

4.7.2 Changes in FAT-CHO oxidation rate

There were no differences in fat and carbohydrate (CHO) oxidation at the beginning of endurance (Figure 4.2). Fat oxidation was higher under WT than PL and SD after completed endurance exercise, but it was only significantly different between WT and PL ($p < 0.05$). CHO oxidation was remarkably higher in PL and SD than WT at the end of endurance exercise ($p < 0.05$).

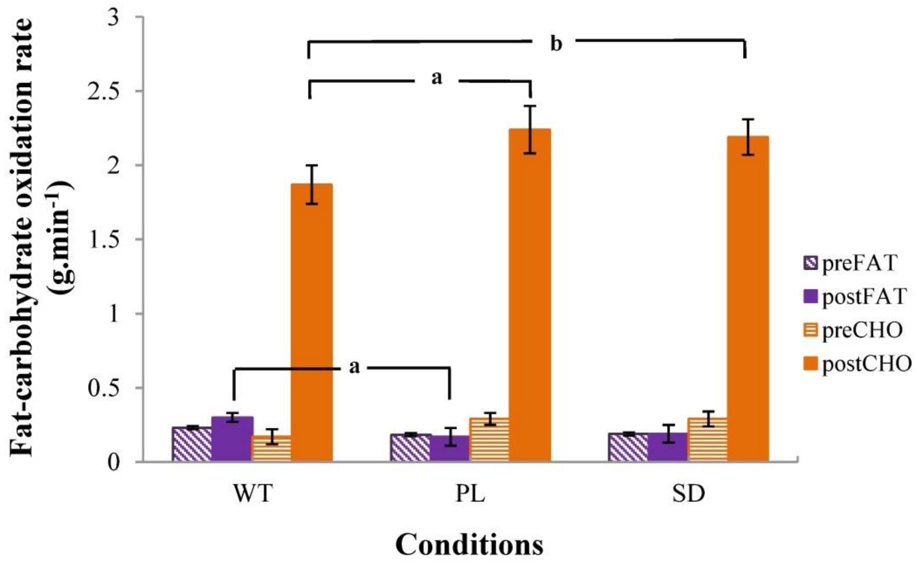

Figure 4.2 Comparisons pre- and post-fat oxidation rate and carbohydrate oxidation rate during endurance exercise in Water (WT), Placebo (PL) and Sports drink (SD) (n=14). All values are means and SEMs.

Symbols

[a]: significant difference between WT-PL; [b]: significant difference between WT-SD; [c]: significant difference between PL-SD. All values are compared at $p < 0.05$.

4.8 Changes in exercise performance during endurance exercise

4.8.1 Time to exhaustion

Time to exhaustion was measured at beginning of each 70%VO_{2peak} until reached criteria of exhaustion. Longer exercise duration in sports drink and placebo conditions compared with water condition were 40% and 20% respectively (Figure 4.3). However, statistics only showed that exercise duration in sports drink condition was significantly longer than water group ($p = 0.002$). Differences neither water-placebo nor placebo-sports drink were observed.

4.8.2 Accumulated total work done

During endurance exercise, three time points which were included work done (WD) up to 70%VO_{2peak}, work done up to exhaustion, and total work done were calculated (Figure 4.4). The result showed that there was no difference among three conditions during WD up to 70%VO_{2peak}. However, the significant differences among conditions were found in WD up to exhaustion and total WD ($p < 0.01$).

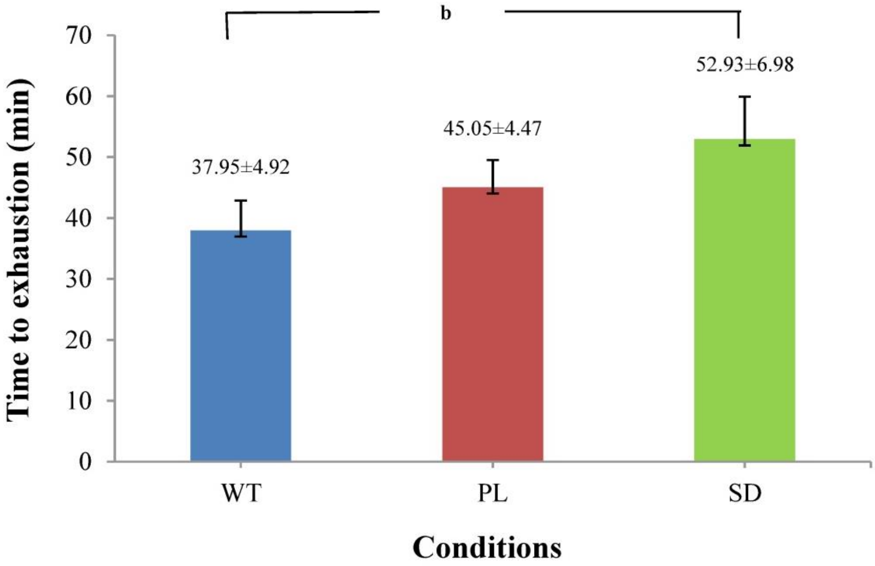

Figure 4.3 Comparisons of time to exhaustion during endurance exercise in Water (WT, blue), Placebo (PL, red) and Sports drink (SD, green) (n=14). All values are means and SEMs.

Symbols

[a]: significant difference between WT-PL; [b]: significant difference between WT-SD; [c]: significant difference between PL-SD. All values are compared at $p < 0.05$.

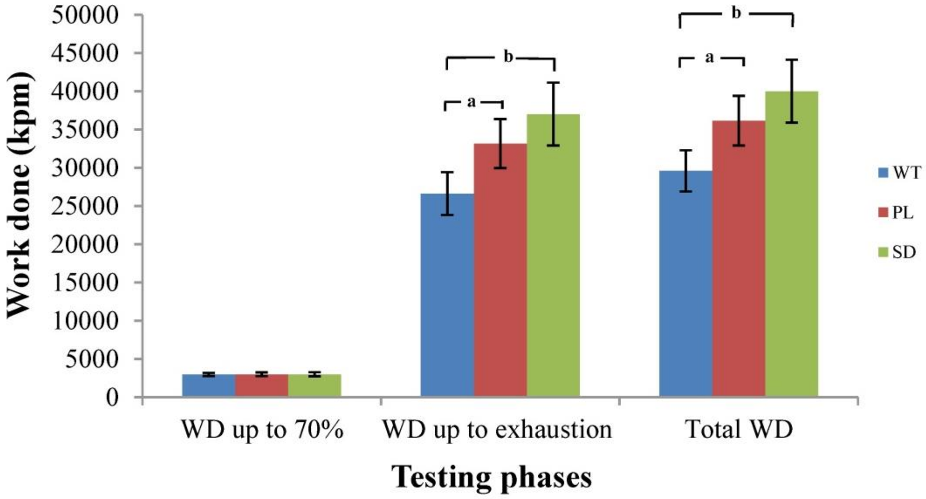

Figure 4.4 Comparisons of work done up to 70%VO_{2peak}, work done up to exhaustion and total work done during endurance exercise in Water (Blue), Placebo (Red) and Sports drink (Green) (n=14). All values are means and SEMs.

Symbols

[a]: significant difference between WT-PL; [b]: significant difference between WT-SD; [c]: significant difference between PL-SD. All values are compared at $p < 0.05$.

CHAPTER V
DISCUSSION

The aim of this study was to investigate the effect of sports drink on subsequent endurance performance following glycogen depletion exercise in healthy Thai males, and also compare among drinks. Fourteen subjects were randomly received three different beverages during 2 hours recovery period after glycogen depletion session, then, exercise on cycling ergometer at 70%VO_{2peak} until unable to sustain the testing protocol with signs of exhaustion. All parameters, comprised of endurance performance, cardiorespiratory, metabolism and blood chemistry parameters, were analyzed. The main finding in the current study was that time to exhaustion in sports drink group was significantly longer than water group. Moreover, sports drink group showed significantly greater % ejection fraction than water group, and lower minute ventilation than placebo and water groups. It was probably because exogenous carbohydrate supplementation, accompanied with electrolyte promoting improves endurance performance.

5.1 General characteristics

All physical characteristics of subjects in the present study (Table 1) were in the normal values in Thai male population at this age-ranged (67). In addition, their VO_{2peak} was in the average to good levels of young Thai populations (27). Therefore, investigator successfully recruited normal healthy subjects for this study.

5.2 Stresses during 2 exercises

In this study, exercise-induced glycogen depletion causes higher physical stress conducted by non-invasive determination techniques than endurance exercise protocol since at the end of GD heart rate was induced up to 90% of predicted maximum heart rate, whereas it was 80% of predicted maximum heart rate in EX. Describe in, moreover, other parameters including overshooting ventilation (40), metabolic and lactate concentration (68). Lactate concentration at the end of GD was induced nearly to 12 mMol due to both highest generation and elimination abilities. Based on previous studies (69, 70), intramuscular glycogen depletion from physiologic findings conducted by muscle biopsy were correlated with this GD protocol. Therefore, this study confirms that GD protocol used is appropriately effective. In addition, glycogen depletion was similar among three groups shown as no differences of work done at the end of GD. Thus, physiologic changes in EX are solely derived from types of supplement.

5.3 Changes in cardiorespiratory function

Increasing in all cardiovascular parameters, including heart rate, stroke volume, cardiac output, end-diastolic volume, and ejection fraction were increased except systemic vascular resistance, during to exercise session are in the similar fashion no matter which kind of fluids are being intervened. These responses are solely dependent upon exercise stimuli. According to mechanical changes during exercise, mechanoreceptor in skeletal muscle is activated. Afferent input travels into the brain to adjust cardiovascular control center (33). Cardiac muscle is more activated via sympathetic activation; consequently, greater contractility and rhythm were shown at the beginning till the end of exercise. Moreover, greater venous return resulted by skeletal muscle pump and respiratory muscle pump (32) travels to the heart to promote sufficient blood volume from the heart to exercise muscle. All the above mechanisms activate for the readiness of physical activity and become more vigorous when exercise stimuli is more intense.

Physiologically, systemic vascular resistance was declined. It is explained via a selective activation of blood vessels at various parts throughout the body (32).

This means while blood vessels in some part of the body become dilate, the others may be shut. Because active muscle required more blood supply during exercise, it was compensated by lessen blood from some parts via higher peripheral resistance, which allow more blood to supply the target working muscles.

In the same time, respiratory function was changed during exercise. Minute ventilation in this study which was resulted by higher respiratory rate and tidal volume was increased nearly 100 L/min after completed glycogen depletion, it was due to peripheral and central chemoreceptor stimulation (40). The major aim in GD exercise was to stress intramuscular storage pool rather than cardiorespiratory storage pool. However, fluctuation of cardiorespiratory responses may be derived from exercise intensity concept. Similar to exercise-induced glycogen depletion, endurance exercise induced greater ventilation. During metabolic process, carbon dioxide and hydrogen ion were more produced, and the physiological regulation tried to remove them out as early as possible to retard acidosis status via respiratory pathway. Therefore, more ventilation was occurred during glycogen depletion and endurance exercise.

In this study, sports drink contained 10%carbohydrate and 0.16%electrolytes did not affect to cardiovascular function. Similarly, previous studies with 6%carbohydrate and electrolyte solution did not change cardiovascular function (19, 21). In contrast, some recent researches elucidated that heart rate was more elevated in low (6 %) carbohydrate sports drink compared with high (8%) (16). It was indicated that, during exercise, cardiac responses mainly rely on carbohydrate energy substrate rather than electrolytes (16, 25). Although high CHO concentration was recommended, but 10%CHO (SD and PL conditions) conducted in this study did not difference, compared with non-CHO supplement drinks (WT). Myocardium function under normal condition depends on oxidative processes of either lipid or carbohydrate. This excitable tissue contains various physiological functions due to specialization, for example intrinsic and extrinsic properties, autoregulation and self-generated firing or pacemaker. This cell autorhythmicity depends on movement of sodium (Na^+) and potassium (K^+) ions across myocardial cell membrane. The other electrolyte involves this excitable tissue is calcium (Ca^{2+}), which binds with troponin-C and finally causes transformational changes. Thus, myocardial contraction depends on Na^+, K^+ and Ca^{2+}

ions where Na^+ and K^+ are related to firing rate and Ca^{2+} is related to contraction (31). Sports drink used in this study contain only Na^+ and K^+, but no Ca^{2+}, in the hypotonic solution. Therefore, sports drink less likely causes physiologic changes of cardiac function. Exercise intervention is solely responsible for cardiac responses. Similarly, sports drink seemed to be less affecting to respiratory function, only changes in minute ventilation, but others were not.

5.4 Changes in metabolism

In general, aerobic glycolysis pathway produces more ATP but slower than anaerobic glycolysis pathway (29). According to glycogen depletion designed to be as high intensity exercise, active muscle needed more oxygen consumption for metabolic process. However, the energy system changed to utilize anaerobic glycolysis because it could not be able to extract oxygen supply sufficiently. Whenever carbon dioxide and hydrogen ion which were the last products of anaerobic glycolysis pathway were produced and could not be removed properly, the body would be experienced to acidosis status. Skeletal muscle needed more oxygen supply itself, while carbon dioxide was being produced during high intensity shown as higher VO_2, VCO_2 after completed glycogen depletion in this study. However, carbon dioxide was higher produced than oxygen uptake, accompanied with higher respiratory exchange ratio (RER) at the end of exercise. Therefore, anaerobic glycolysis was occurred. According to carbohydrate oxidation equation, that is $C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O$, CO_2 is produced equally to O_2 consumption by 1:1 ratio. This phenomenon supported over respiratory ratio (RER) value which was indicated plenty of carbohydrate substance was utilized during exercise and also been over normal ranges at the end of glycogen depletion exercise.

Because subjects in all groups were forced up to $70\%VO_{2peak}$; therefore, input and output, in term of gases at $70\%VO_{2peak}$ were no difference during energy liberation of either glycolysis or lipolysis, none of electrolytes played role. This was similar to previous research which investigated effect of commercial sport drink on short-term repeated exercise performance, and also performed with determined adherence of exercise intensity (64). Although there were no differences among three beverages, sports drink and placebo were showed higher RER than water. It was probably resulted from high carbohydrate oxidation during exercise.

As the results of this study, sports drink and placebo demonstrated that there was significantly utilized of carbohydrate substance under sports drink and placebo than water group during endurance exercise. Although the significant difference in fat oxidation between water and sports drink was not observed, but there was tendency to less utilize fat substance under sports drink than water, similar with placebo group. Recent research (71) investigated the rate of glycogen re-synthesis in vastus lateralis muscle by biopsy method. Significant higher rate of glycogen re-synthesis in carbohydrate ingestion group was resulted. In addition, other previous studies (25, 64, 72) suggested that attenuated endogenous glycogen could cause to utilize total fat oxidation and endogenous carbohydrate oxidation during exercise in solution without carbohydrate substance.

5.5 Changes in blood chemistry

Vigorous exercise, normally, transitions of energy system in order to sustain exercise duration. Unable to remove lactate is caused to be accumulation of blood lactate. Whenever it could not extract adequate oxygen for metabolic process, it would be changed to consider anaerobic glycolysis pathway which was not required oxygen. Lactate was a product of this energy system and could influence to attenuate function of muscle cell. The mechanism was explained that hydrogen ion could be caused to block Ca^{2+} function in excitation-contraction coupling (73). This may lead to be lower effectiveness of muscle contraction. Another possible cause was declined glycogen in both muscle and liver leading to accumulate of lactate in blood stream and could not be changed to be glycogen. Therefore, higher lactate concentration was occurred as shown at the end of glycogen depletion session. The highest blood lactate was suggested that this protocol could induce to deplete glycogen due to overshooting its normal ranges (68). All blood lactate values independent on any drinks were returned to be in normal ranges after 2 hours recovery periods in this study. Similar to glycogen depletion, endurance exercise induced lactate accumulation in all three groups, but it was in normal ranges. Therefore, it was indicated that the body could remove lactate produced properly.

During exercise, some hormones will regulate blood glucose concentration in order to sustain exercise duration (42). In this study, blood glucose in all three groups greatly declined in the same way after completed glycogen depletion. It was because of enormously required energy leading to breakdown and be utilized more glucose; consequently, depleted glucose was occurred and insufficient glucose broken down into blood stream was displayed.

5.6 Changes in exercise performance

The main findings of this study showed longer sustained cycling time during subsequent exercise under sports drink than placebo and water. However, it was only significantly different between sports drink and water ($p<0.05$). Furthermore, it was also found significantly greater work done in sports drink group compared with water, and also significantly greater in placebo than water. One study reported that muscle glycogen concentration was nearly empty in the first half time during soccer (53). They found 3 folds depleted muscle glycogen concentration by using muscle biopsy technique. It was indicated that endogenous carbohydrate was declined after prolonged exercise and/ or vigorous exercise. Thus, resynthesized muscle glycogen is important, especially in subsequent exercise (10). Carbohydrate ingestion could be able to elevate the rate of glycogen re-synthesis during short-term recovery (71). After biopsy method had employed, muscle glycogen re-synthesis under with and without carbohydrate ingestion were 5.2 and 0.5 µmol.g wet $wt^{-1}.h^{-1}$ respectively. In this study, an absent endogenous carbohydrate was performed during glycogen depletion session. After consumed beverage with or with exogenous carbohydrate contained during recovery period, it was surely that exogenous carbohydrate played an important role to sustain longer time under sports drink in the second exercise as longer time to exhaustion was demonstrated.

The current finding supported previous study which was also demonstrated longer time to exhaustion after thirteen recreational subjects consumed sports drink contained 6%CHO and underwent prolonged exercise then (19). They suggested that exogenous carbohydrate utilization played important role to spare and resynthesize muscle glycogen during recovery period, therefore, it led to perform greater duration during subsequent endurance exercise. Similarly, previous researches resulted greater performance times under commercial sports drink (64) and carbohydrate drinks (15). In addition, ingestion of a commercial sports drink with 6.9%CHO during 4 hours recovery period improved endurance performance (18). It was performed on treadmill at 70%VO_{2max}, and found 62 ± 6.2 min under sports drink VS 39.8 ± 6.1 min under placebo on subsequent exercise. Although carbohydrate with (SD) and without (PL) electrolytes drinks did not meet statistically significant, but SD could extend longer exercise duration, as well as higher total work done than PL. Besides carbohydrate substance, electrolyte especially Na^+ is possibly played role to support longer endurance exercise duration under SD. In part of muscle function, electrolytes, mainly as Na^+ and K^+, promote neurotransmitter function, as well as adequate actin-myosin function. Theoretically, Na^+ can stimulate more fluid absorption from stomach into small intestine (22). Na^+ gradient causes sodium dependent glucose transporter (SGLT1) generating, as well as GLUT2 generated by glucose concentration (74, 75); therefore, the combination of SGLT1 and GLUT2 in small intestine enhances carbohydrate availability. In addition, K^+ is contributed in CHO metabolism; it can support changes of glucose to glycogen and store in the liver for future fuel (76). It is required in the final step of glycolytic pathway to help pyruvate kinase which is a main enzyme in this process (77). This pathway is shown as follow;

$$\text{Phosphoenolpyruvate} + \text{ADP} \xrightarrow[K^+]{Mg^{2+}} \text{Pyruvate} + \text{ATP}$$

Therefore, electrolyte replenishment possibly supports more proper function of muscle and nerve in CHO-electrolytes than CHO alone.

5.7 Limitation and suggestion

1. Electrolyte losses via sweating could not be estimated in this study; therefore, investigator did not know the amount of electrolytes supplement via sport drink is appropriate or not. In particular when exercises are performed under extreme conditions of hot and humid, the amount of electrolytes supplement is remarkably in doubt.

2. The present study failed to get rid of psychological involvement since taste of water was unable to be adjusted.

3. There are various kinds of commercial sports drink which are available, some may contain ingredient which may affect physiologic function or even give-rise in risk. Therefore, our further investigations are aimed that to identify for safety, as well as giving the warning sign for some products.

CHAPTER VI
CONCLUSION

This study was hypothesized that carbohydrate with electrolytes drink could improve subsequent endurance exercise performance, as well as on cardiorespiratory, blood chemistry, and metabolic variables, than carbohydrate drink alone and water. It was concluded that carbohydrate with electrolytes drink similarly affected cardiorespiratory and blood chemistry function as carbohydrate alone and water. However, carbohydrate with electrolytes drink demonstrated higher utilization of carbohydrate substrate than water, leading to improve subsequent endurance performance as shown by higher work done, as well as longer time to exhaustion. It is recommended that carbohydrate supplement drink provided during recovery period is essential for the following exhaustive exercise. Although there was no difference of time to exhaustion between carbohydrate with (SD) and without (PL) electrolytes drinks, but additional electrolytes in carbohydrate drink tended to express further enhance exhaustive time.

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APPENDICES

APPENDIX A
DATA RECORING FORM

ID…………………………………………

DATE……………………………………..

Glycogen depletion test

Age……………………yrs USG………………… Height………………………cm

Heart rate ………… bpm BMI (Pre)……………kg/m^2 Predicted 90%MHR ………..bpm

Body mass (Pre)………………% Seat height…………… Body fat (Pre)………………%

Weight (Pre)………………………...kg Weight (Post)……………………......kg

Blood Lactate (Pre) ………………………mMol/L Blood Lactate (Post) …………………mMol/L

Blood glucose (Pre)…………………….mg/dL Blood glucose (Post)……………………mg/dL

Temperature……………………°C Humidity………………………%

Note:………
…….

ID…………………………………………

DATE……………………………………..

Glycogen depletion test

Time (min)	Work (kp)	%P_{max}	RPM	Oxycon					Physioflow				BP	RPE
				VO_2 (L/min)	VCO_2 (L/min)	VO_2 (ml/kg/min)	RR (beats/min)	RER	HR	SV	CO	CI		

ID…………………………………………

DATE……………………………………..

Endurance performance test

Age……………………yrs USG………………… Height………………………cm

Heart rate ………… bpm BMI (Pre)……………kg/m^2 Predicted 90%MHR ………..bpm

Body mass (Pre)………………% Seat height…………… Body fat (Pre)………………%

Weight (Pre)………………………...kg Weight (Post)…………………......kg

Blood Lactate (Pre) ………………………mmol/L Blood Lactate (Post) …………………mmol/L

Blood glucose (Pre)…………………….mg/dL Blood glucose (Post)……………………mg/dL

Temperature……………………°C Humidity………………………%

Note:……
…………………………………………………………………………………………………….

ID…………………………………………

DATE……………………………………..

Endurance performance test

Time (min)	Work (kp)	%Pmax	RPM	Oxycon					Physioflow				BP	RPE
				VO_2 (L/min)	VCO_2 (L/min)	VO_2 (ml/kg/min)	RR (beats/min)	RER	HR	SV	CO	CI		

ID…………………………………………

DATE……………………………………..

Maximal oxygen consumption test

Age………………yrs Height………………….cm Weight…………………kg

Body fat……………….% Body mass………………..kg BMI………………….kg/m2

Heart rate……………bpm Predicted 90%MHR………………..bpm

Time (min)	Work (kp)	HR (bpm)	RPM	Oxycon					BP	RPE
				VO_2 (L/min)	VCO_2 (L/min)	VO_2 (ml/kg/min)	RR (beats/min)	RER		

Note:………
……

ID…………………………………………

DATE……………………………………..

แบบบันทึกอาหาร

วัน/เดือน/ปี	มื้ออาหาร	รายการอาหาร

Note:……
……
………….

APPENDIX B
HEALTH STATUS ASSESSMENT

รหัสประจำตัว.......................... วันที่...........................

แบบประเมินความพร้อมก่อนการออกกำลังกาย (สำหรับบุคคลทั่วไปอายุ 18-25 ปี)

แบบประเมินนี้มีวัตถุประสงค์เพื่อประเมินสภาวะทางสุขภาพก่อนการออกกำลังกาย และสามารถประเมินได้ว่าท่านต้องเข้าการตรวจร่างกายจากแพทย์ก่อนการออกกำลังกายหรือไม่ กรุณาอ่านข้อคำถามอย่างระมัดระวังและตอบคำถามด้วยความสัตย์จริงทุกประการ

1. แพทย์ที่ตรวจรักษาเคยบอกหรือไม่ว่า ท่านมีความผิดปติของหัวใจ และควรออกกำลังกายภายใต้คำแนะนำของแพทย์เท่านั้น ☐ ไม่เคย ☐ เคย

2. ท่านมีความรู้สึกเจ็บปวดหรือแน่นบริเวณหน้าอก ขณะที่ออกกำลังกายหรือไม่ ☐ ไม่มี ☐ มี

3. ในเดือนที่ผ่านมา ท่านเคยมีอาการเจ็บหน้าอกขณะอยู่เฉยๆ โดยไม่ได้ออกกำลังกายหรือไม่ ☐ ไม่เคย ☐ เคย

4. ท่านมีอาการสูญเสียการทรงตัว (ยืนหรือเดินเซ) เนื่องจากอาการวิงเวียนศีรษะหรือไม่ หรือท่านเคยเป็นลมหมดสติหรือไม่ ☐ ไม่มี ☐ มี

5. ท่านมีปัญหาเกี่ยวกับกระดูกและข้อต่อ ซึ่งจะมีอาการแย่ลงถ้าออกกำลังกายหรือไม่ ☐ ไม่มี ☐ มี

6. แพทย์ที่ตรวจรักษามีการสั่งยารักษาความดันโลหิต หรือความผิดปกติของหัวใจท่านหรือไม่ ☐ ไม่มี ☐ มี

7. เท่าที่ท่านทราบ ยังมีเหตุผลอื่นๆอีกหรือไม่ ที่ทำให้ท่านไม่สามารถออกกำลังกายได้ ☐ ไม่มี ☐ มี

ถ้าท่านตอบว่ามีหรือเคย เพียงข้อหนึ่งข้อใด

ขอให้ท่านปรึกษากับแพทย์ **ก่อน** ที่ท่านจะเริ่มออกกำลังอย่างกระฉับกระเฉงหรือ **ก่อน** เข้าร่วมการทดสอบสมรรถภาพทางกาย โดยบอกแพทย์ว่า ท่านได้ตอบแบบประเมินความพร้อมก่อนการออกกาลังกาย และมีคำถามข้อใดที่ท่านตอบว่า **มีหรือเคย**

- ท่านอาจทำกิจกรรมใดตามที่ต้องการ ตราบเท่าที่ท่านเริ่มต้นทำอย่างช้า ๆ และค่อย ๆ เพิ่มเวลา หรือความหนักขึ้น หรือ**เลือกทำเฉพาะ** กิจกรรมออกกำลังกายที่ปลอดภัยสำหรับตนเอง ปรึกษาและปฏิบัติตามคำแนะนำของแพทย์เกี่ยวกับชนิดของการออกกำลังกายที่ท่านอยากเข้าร่วม

- หากิจกรรมออกกำลังกายที่ปลอดภัยและเป็นประโยชน์กับท่าน

ไม่มี/ไม่เคยทุกข้อ

ถ้าคำตอบของท่าน คือ ไม่มี/ไม่เคยทุกข้อ ด้วยความสัตย์จริง ค่อนข้างมั่นใจว่าท่านสามารถทำได้:

-เข้าร่วมออกกำลังอย่างกระฉับกระเฉงได้ โดยเริ่มต้นทำอย่างช้าๆ และค่อยๆ เพิ่มเวลา หรือความหนักขึ้น นี่เป็นวิธีการที่ปลอดภัยและง่ายที่สุดสำหรับท่าน

- เข้าร่วมการทดสอบสมรรถภาพทางกายได้ ซึ่งจะทำให้ท่านทราบระดับสมรรถภาพพื้นฐาน และวางแผนการใช้ชีวิตที่กระฉับกระเฉงเหมาะกับตนเอง แต่มีข้อเสนอแนะว่าท่านควรตรวจวัดความดันโลหิตก่อน ถ้าความดันโลหิตมากกว่า 144 / 94 มิลลิเมตรปรอท ท่านควรจะปรึกษาแพทย์ ก่อนที่จะเริ่มออกกำลังอย่างกระฉับกระเฉง

กรุณาหยุดพัก หรือเลื่อนการออกกำลังกาย อย่างกระฉับกระเฉงไปก่อน

- ถ้าท่านรู้สึกไม่ค่อยสบาย ครั่นเนื้อครั่นตัว เช่น เป็นหวัดหรือมีไข้ ควรหยุดพักผ่อนจนกระทั่งรู้สึกดีขึ้น หรือ

- ถ้าท่านกำลังตั้งครรภ์ หรืออาจจะตั้งครรภ์ โปรดปรึกษาแพทย์ก่อนที่ท่านจะเข้าร่วมออกกำลังอย่างกระฉับกระเฉง

หมายเหตุ : ถ้าสุขภาพของท่านมีการเปลี่ยนแปลงไปจนทำให้ท่านต้องตอบคำถามว่า **มีหรือเคย** ในข้อหนึ่ง ข้อใด กรุณาปรึกษาแพทย์หรือผู้เชี่ยวชาญด้านการ ออกกำลังกาย ว่าท่านควรจะเปลี่ยนหรือปรับแผนการ การออกกำลังกายหรือไม่อย่างไร

.

รหัสประจำตัว.......................................วันที่...

แบบสอบถามสำหรับผู้เข้าร่วมวิจัย

ชื่อ-สกุล..วัน เดือน ปีเกิด...อายุ.................ปี

ที่อยู่.. อาชีพ..

ประวัติสุขภาพ

โรคประจำตัว ☐ ไม่มี ☐ มี โปรดระบุ.......................................

ประวัติการผ่าตัด..

เคยมีประวัติการบาดเจ็บของกระดูกและกล้ามเนื้อหรือไม่?

☐ ไม่เคย ☐ เคย โปรดระบุ

กิจกรรมทางกาย

1. คุณทำกิจกรรมดังต่อไปนี้ด้วยระยะเวลาอย่างน้อย 20 นาที บ่อยแค่ไหนใน 1 สัปดาห์? (ระบุเป็นจำนวนครั้ง)

กิจกรรม	ระยะเวลา/ครั้ง	จำนวนครั้ง/สัปดาห์
1.1 ออกกำลังกายระดับหนัก; ยกของหนัก, ทำสวน, วิ่ง, ปั่นจักรยาน หรือกิจกรรมที่ต้องใช้แรงมาก		
1.2 ออกกำลังกายระดับปานกลาง; ถือของขนาดเล็ก, ปั่นจักรยาน หรือกิจกรรรมที่ใช้แรงปานกลาง		
1.3 ออกกำลังกายระดับเบา; เล่นโยคะ, ไท-ชิ, โบว์ลิ่ง หรือกิจกรรมที่ใช้แรงน้อย		
1.4 เดินด้วยความเร็วปกติ		

คะแนน.......... ผลการประเมิน...........

APPENDIX C
HUMAN SUBJECTS APPROVAL CERTIFICATE

COA No. MU-CIRB 2016/111.2408

Mahidol University Central Institutional Review Board (MU-CIRB)

Certificate of Approval

Protocol No.: 2016/117.1207

Title of Project: Sports drink on repeated performance in healthy Thai males after glycogen depletion.

Approval includes:

1) Principle Investigator: Miss Sothida Nantakool
2) Affiliation: College of Sports Science and Technology, Mahidol University
   Research Site: College of Sports Science and Technology, Mahidol University
3) Submission Form version date 19 August 2016
4) Protocol version date 12 July 2016
5) Questionnaire version date 12 July 2016
6) Data Collection Form version date 12 July 2016
7) Informed Consent Form version date 12 July 2016
8) Participant Information Sheet version date 19 August 2016
9) Recruitment Material version date 12 July 2016

MU-CIRB is in full compliance with International Guidelines for Human Research Protection such as Declaration of Helsinki, The Belmont Report, CIOMS Guidelines and the International Conference on Harmonization in Good Clinical Practice (ICH-GCP)

Date of Approval: *24 / August / 2016*

Date of Expiration: *23 / August / 2017*

Signature of Chairperson: ....

(Professor Dr.Rutja Phuphaibul)

MU-CIRB Chair

Signature of Institute Representative: ....

(Professor Dr.Sansanee Chaiyaroj)

Vice President for Research

** See list of Co-Investigators at the back page*

Page 1 of 2

BIOGRAPHY

NAME	Sothida Nantakool
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