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EFFECTS OF WATER-BASED EXERCISE TRAINING ON PHYSIOLOGICAL ADAPTATIONS AND CUTANEOUS MICROVASCULAR REACTIVITY IN TYPE 2 DIABETIC PATIENTS.

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อภิวรรณ ณัฐมนวรกุล : ผลของการฝึกออกกำลังกายในน้ำต่อการปรับตัวทางสรีรวิทยาและการตอบสนองของหลอดเลือดจุลภาคที่ผิวหนังในผู้ป่วยโรคเบาหวานประเภทที่ 2. (EFFECTS OF WATER-BASED EXERCISE TRAINING ON PHYSIOLOGICAL ADAPTATIONS AND CUTANEOUS MICROVASCULAR REACTIVITY IN TYPE 2 DIABETIC PATIENTS.) อ.ที่ปรึกษาวิทยานิพนธ์หลัก: รศ. ดร.ดรุณวรรณ สุขสม, อ.ที่ปรึกษาวิทยานิพนธ์ร่วม: รศ. ดร. สุภัทรา อมาตยกุล, , หน้า.

งานวิจัยนี้มีวัตถุประสงค์เพื่อศึกษาผลของการฝึกออกกำลังกายในน้ำที่มีต่อการปรับตัวทางสรีรวิทยาและการตอบสนองของหลอดเลือดจุลภาคที่ผิวหนังโดยเปรียบเทียบกับการฝึกออกกำลังกายบนบกในผู้ป่วยโรคเบาหวานประเภทที่ 2 กลุ่มตัวอย่างเป็นผู้ป่วยสูงอายุโรคเบาหวานประเภทที่ 2 จำนวน 53 คน (อายุ 60-70 ปี) ทั้งหมดถูกสุ่มเลือกเข้ากลุ่ม 4 กลุ่ม ได้แก่ กลุ่มออกกำลังกายในน้ำ (จำนวน 13 คน), กลุ่มควบคุมด้วยการแช่ในน้ำ (จำนวน 15 คน), กลุ่มออกกำลังกายบนบก (จำนวน 10 คน), และกลุ่มควบคุมที่ให้คำแนะนำในการออกกำลังกาย (จำนวน 15 คน) กลุ่มออกกำลังกายทั้งสองกลุ่มได้รับการฝึกออกกำลังกายที่ความหนัก 70 เปอร์เซ็นต์ของอัตราการเต้นหัวใจสูงสุด วันละ 30 นาที 3 วันต่อสัปดาห์ เป็นเวลา 12 สัปดาห์ ก่อนและหลังการได้รับโปรแกรมฝึกออกกำลังกายอาสาสมัครทุกคนได้รับการทดสอบสมรรถภาพร่างกาย เก็บตัวอย่างเลือดเพื่อวิเคราะห์สารชีวเคมี และประเมินการตอบสนองของหลอดเลือดจุลภาคที่ผิวหนัง

ผลการศึกษาพบว่า ภายหลังการฝึกออกกำลังกายตามโปรแกรมเป็นระยะเวลา 12 สัปดาห์ กลุ่มออกกำลังกายในน้ำมีน้ำหนักตัว อัตราการเต้นของหัวใจขณะพัก และความดันโลหิตตัวบนลดลงอย่างมีนัยสำคัญทางสถิติที่ระดับ 0.05 ขณะที่กลุ่มออกกำลังกายบนบกมีเปอร์เซ็นต์ของไขมันในร่างกายและความดันโลหิตตัวล่างลดลงอย่างมีนัยสำคัญทางสถิติ (ระดับ 0.05) สำหรับความแข็งแรงของกล้ามเนื้อแขนเพิ่มขึ้นทั้งกลุ่มออกกำลังกายในน้ำและบนบก แต่อัตราการใช้ออกซิเจนสูงสุดดีขึ้นอย่างมีนัยสำคัญทางสถิติที่ระดับ 0.05 ในกลุ่มออกกำลังกายในน้ำเท่านั้น ขณะที่ผลของระดับน้ำตาลในเลือดฮีโมโกลบินเอวันซี และระดับไขมันคอเลสเตอรอล ทั้งกลุ่มออกกำลังกายในน้ำและบนบกพบมีการเปลี่ยนแปลงที่ดีขึ้นอย่างมีนัยสำคัญทางสถิติ (ระดับ 0.05) ส่วนภาวะดื้ออินซูลิน ซี-รีแอ็คทีฟโปรตีน และระดับมาลอนไดอัลดีไฮด์ในพลาสมาของกลุ่มออกกำลังกายในน้ำ มีระดับต่ำลงอย่างมีนัยสำคัญทางสถิติที่ระดับ 0.05 โดยเฉพาะอย่างยิ่งพบระดับซี-รีแอ็คทีฟโปรตีนลดลงอย่างมีนัยสำคัญทางสถิติที่ระดับ 0.05 เมื่อเปรียบเทียบกับกลุ่มที่ออกกำลังกายบนบก นอกจากนี้พบว่า ค่าการไหลของเลือดชั้นคิวทาเนียสสูงสุด และค่าระยะเวลาการฟื้นตัวของการไหลของกลุ่มออกกำลังกายในน้ำมีค่าดีขึ้นภายหลังการฝึก 12 สัปดาห์ ทั้งกลุ่มออกกำลังกายในน้ำและบนบก อย่างไรก็ตามไม่พบความแตกต่างของตัวแปรทั้งหมดในกลุ่มควบคุม

ผลการวิจัยในครั้งนี้แสดงให้เห็นว่าการออกกำลังกายในน้ำมีผลต่อสมรรถภาพทางกาย การควบคุมระดับน้ำตาลและไขมันในเลือด และการตอบสนองของหลอดเลือดจุลภาคในผู้ป่วยสูงอายุโรคเบาหวานประเภทที่ 2 และมีประสิทธิภาพมากกว่าการออกกำลังกายบนบก จากการที่สามารถลดภาวะการอักเสบในร่างกายของผู้สูงอายุลงได้

สาขาวิชา ชีวเวชศาสตร์ ลายมือชื่อนิสิต .....

ปีการศึกษา 2556 ลายมือชื่อ อ.ที่ปรึกษาวิทยานิพนธ์หลัก .....

ลายมือชื่อ อ.ที่ปรึกษาวิทยานิพนธ์ร่วม .....

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APIWAN NUTTAMONWARAKUL: EFFECTS OF WATER-BASED EXERCISE TRAINING ON PHYSIOLOGICAL ADAPTATIONS AND CUTANEOUS MICROVASCULAR REACTIVITY IN TYPE 2 DIABETIC PATIENTS.. ADVISOR: ASSOC. PROF. DAROONWAN SUKSOM, Ph.D., CO-ADVISOR: ASSOC. PROF. SUPATHRA AMATYAKUL, Ph.D., pp.

The purpose of this study was to investigate the effect of water-based exercise training on physiological adaptations and cutaneous microvascular reactivity and to compare with land-based exercise training in type 2 diabetic patients.

Fifty-three elderly patients with type 2 diabetes mellitus were randomly allocated to the one of four intervention groups: the water-based exercise (n=13) group received aqua-aerobic exercise; the water-based control (n=15) group received only water immersion; the land-based exercise (n=10) group received general aerobic exercise; and the land-based control (n=15) group received behavior change advice. Both training groups performed aerobic exercise protocol which consisted of 70% of maximum heart rate, 30 minutes/day, and 3 days/wk for 12 weeks. Physical fitness, biochemical variables and cutaneous microvascular reactivity were measured at baseline and week 12.

Body weight, resting heart rate, systolic blood pressure were significantly decreased (all $P<0.05$) only in the water-based exercise training group. As percentage of body fat and diastolic blood pressure were significantly decreased ($P<0.05$) only in the land-based exercise training group. Handgrip strength was also increased significantly ($P<0.05$) in both groups. In addition, maximum oxygen consumption (VO2max) was significantly increased in only the water-based exercise training group. Significantly decreased of fasting blood glucose, glycosylated hemoglobin (HbA1c), total cholesterol, low density lipoprotein cholesterol, and High density lipoprotein cholesterol increased (all $P<0.05$) in both exercise groups. Only the water-based exercise training group had significantly reduction ($P<0.05$) in insulin, HOMA-IR, C-reactive protein (CRP), and malondialdehyde (MDA). Moreover, CRP was significant difference between water-based and land-based group. The greater in amplitude of peak flux during hyperemia (PORHpeak) was significantly increased when compared to baseline in only the water-based exercise training group. Time to peak and recovery time of post-occlusive reactive hyperemia were significantly improved after 12 weeks in both groups ($P<0.05$).

The present finding demonstrated that the water-based exercise training effect in the elderly with type 2 diabetic patients by improving physical fitness, hemodynamic, glycemic control, and cutaneous microvascular reactivity. Interestingly, water-based training higher improves inflammation than land-based exercise training.

Field of Study: Biomedical Sciences Student's Signature .....

Academic Year: 2013 Advisor's Signature .....

Co-Advisor's Signature .....

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CHAPTER I
INTRODUCTION

Background and Rationale

Diabetes mellitus affects approximately 100 million persons worldwide [1]. Five to ten percent have type 1 (insulin-dependent) and 90% to 95% have type 2 (non-insulin-dependent) diabetes mellitus [2]. Prevalence of type 2 diabetes characterized by insulin resistance, which leads to morbidity and mortality associated with atherosclerotic cardiovascular disease and late complications, has been increasing in Thailand. In these pathological processes, cutaneous microvascular dysfunction, were thought to be crucial early events [3]. Mechanisms contributing to insulin resistance and cutaneous microvascular dysfunction include glucotoxicity, lipotoxicity and inflammation [4]. Previous reports proposed that oxidative stress or the increase production of reactive oxygen species (ROS) plays a key role in those deleterious effects of diabetes [5].

Exercise training was an essential component in both medical management of patients with type 2 diabetes and in preventing the development of diabetic complications. A meta-analysis of 14 trials found that exercise training reduced HbA1C levels by 0.66% and whole body exercise was associated with improvement of NO vasodilator function in subjects with type 2 diabetes [6]. These data showed that exercise training act as antioxidant treatment was effective in reducing cutaneous microvascular dysfunction and vascular disease. However, most of them have studied only in the effect of land-based exercise regimens. Whereas, effect of water-based exercise training and cutaneous microvascular vasodilator function in patients with type 2 diabetes mellitus were still limited.

Water-based exercise was defined as activities performed in the water that promote and enhance physical and mental fitness. The principle of water-based exercise is an immersion of the body in thermal water for therapeutic purposes, based on the composition and temperature of the water [7]. Although the scientist evidence for overall benefits of aquatic exercise is extensive, the dose-response relationship and the effects of this exercise on people with diabetes were not clearly understood. It should be recommended to patients with type 2 diabetes provides more benefits, including the improved muscle strength and endurance, reduction of edema, and increased circulation. However, the remaining unexplained that it is suitable for type 2 diabetes patients. The effectiveness comparison between land-

based exercise training and water-based exercise training still remains inconclusive. Thus, the purpose of the present study was to investigate the effect of water-based exercise training on physiological adaptations and cutaneous microvascular reactivity in type 2 diabetic patients as well as compare with land-based exercise training. Fasting plasma glucose, glycosylated hemoglobin (HbA1c), health related physical fitness, and cutaneous microvascular reactivity were evaluated at baseline and after 12 weeks of training with three training sessions per week.

The results from the present study were established the suitable exercise training which safe and effective for patients with type 2 diabetes and may explained mechanisms which responsible for the beneficial effects of land-based and water-based exercise training on diabetic physiological adaptations and cutaneous microvascular reactivity.

Research questions

1. Whether physiological adaptations and cutaneous microvascular reactivity in type 2 diabetic patients can be changed by water-based exercise training.

2. How difference of the effects of water-based exercise training on physiological adaptations and cutaneous microvascular reactivity when compared with land-based exercise training.

The purpose of this study

1. To determine the effects of water-based exercise training on physiological adaptations and cutaneous microvascular reactivity in type 2 diabetic patients.

2. To compare the effect of water-based and land-based exercise training on physiological adaptations and cutaneous microvascular reactivity in type 2 diabetic patients.

Research Hypotheses

Water-based exercise training affects physiologic and cutaneous microvascular reactivity in type 2 diabetic patients in a greater extent than land-based exercise training.

Scope of research

1. The participants were the fifty-three elderly with type 2 diabetes, aged 60-70 years, were recruited from the supreme patriarch center on aging and

Yanasungwararam hospital, Chon-buri province, Thailand. The participants were randomly allocated into equal numbers, ages and gender ratios into 4 groups;

- Water-based exercise group (Wex; n=20) (Continuous water-based exercise training program)

- Water-based control group (Wco; n=20) (water immersion)

- Land-based exercise group (Lex; n=20) (Continuous land-based exercise training program)

- Land-based control group (Lco; n=20) (non-exercise)

2. The variables used in the study included:

2.1 Independent variables were water-based exercise training and land-based exercise training.

2.2 Dependent variables were

- Biological variables as to body weight, body mass index, resting heart rate, systolic blood pressure and diastolic blood pressure.

- Health-related physical fitness variables as to percentage of body fat, percentage of total body water, body mass index, waist-to-hip ratio, muscular strength, maximal oxygen consumption and body flexibility.

- Blood biochemistry variables as to fasting blood glucose, glycosylated hemoglobin, insulin, total cholesterol, high density lipoprotein cholesterol, low density lipoprotein cholesterol, triglyceride, C - reactive protein, malondialdehyde.

- Post-occlusive reactive hyperemia variables as to time to peak, peak perfusion flux and recovery time.

Operational definition

Water-based exercise training is an activity in the water that raises the body's demand for oxygen. In this study, the water-based exercise training is performed in swimming pool (water temperature ~34 to 36°C) at moderate intensity of oxygen consumption (70% of maximum heart rate).

Land-based exercise training is an activity on land that raises the body's demand of oxygen, resulting in a temporary increase in rate of respiration and heart rate. In this study, the land-based exercise training is performed aerobic exercise on stable ground at moderate intensity of oxygen consumption (70% of maximum heart rate).

Maximum oxygen consumption (VO_2max) is the maximum oxygen uptake or the maximum volume of oxygen that can be utilized in one minute during maximal or exhaustive exercise. It is measured as milliliters of oxygen used in one minute per kilogram of body weight.

Glycemic control is an ability of cells to control blood sugar and glycosylated hemoglobin.

Endothelial function is an ability of endothelial cells to control vascular tone (Vasoconstriction/ Vasodilatation) and coagulation process.

Endothelial dysfunction is dysregulation of endothelial cells to control vascular tone (Vasoconstriction/ Vasodilatation) and coagulation process.

Endothelial dependent vasodilatation is an ability of blood vessels to dilate vascular tone through endothelial function which can measure by the change of vascular diameter.

Expected benefits and applications

1. To understand the effects of water-based exercise training on physiological adaptations and cutaneous microvascular reactivity in patients with type 2 diabetes mellitus.

2. To understand the comparative effects between water-based exercise training and land-based exercise training on physiological adaptations and cutaneous microvascular reactivity in type 2 diabetic patients.

3. To provide appropriate exercise training program for safe and effective in patients with type 2 diabetes mellitus.

4. To provide fundamental information which essential for planning treatment in type 2 diabetic patients.

5. To provide physiological knowledge for further studies.

Conceptual Framework

This study was outlined possible courses of action in figure 1.1

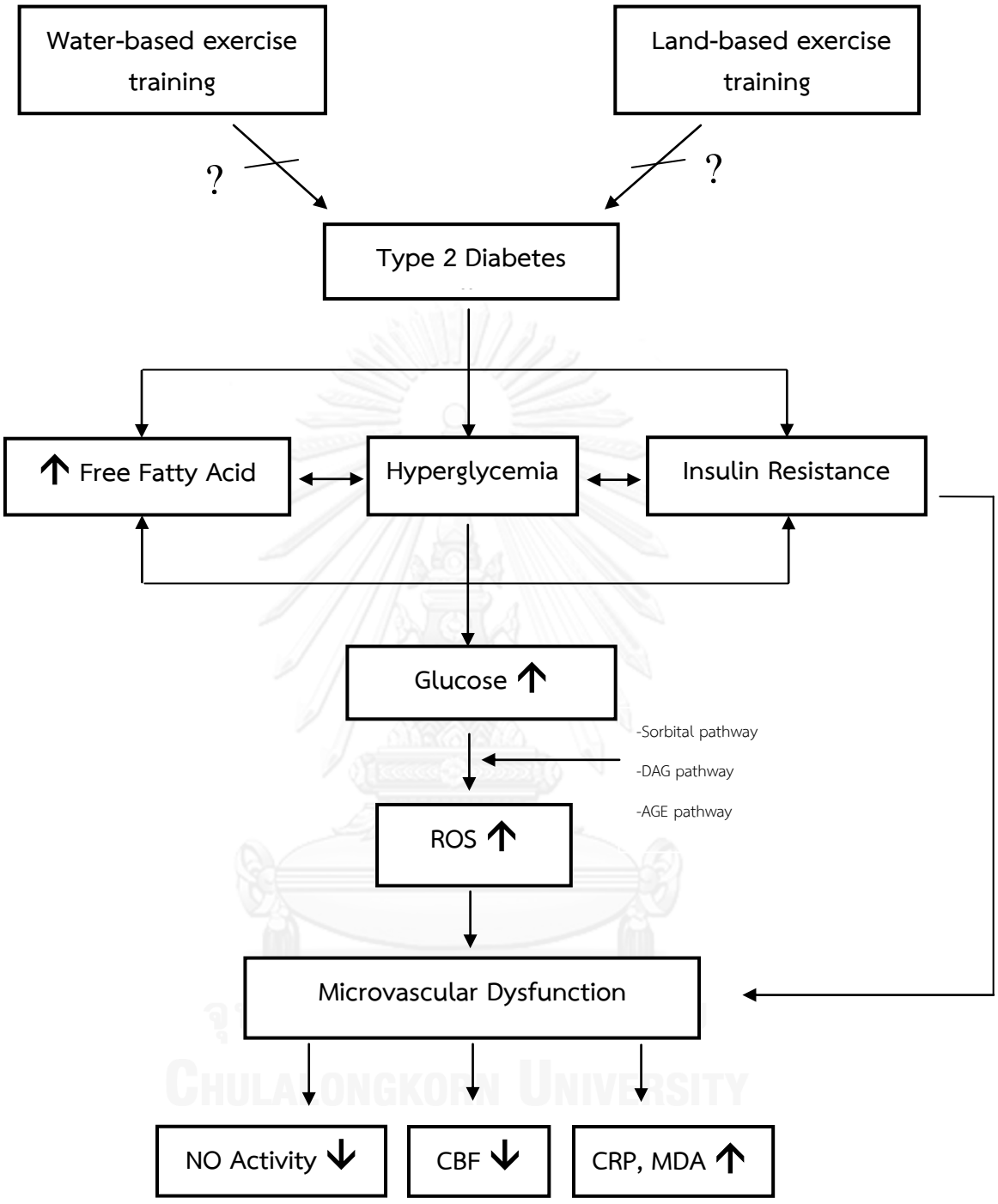


Figure 1 Conceptual framework. The underlying mechanisms in water-based and land-based exercise training are currently unknown in patients with type 2 diabetes.

CHAPTER II

LITERATURE REVIEW

The aim of the present study was to determine the effect of water-based exercise training on physiological adaptations and cutaneous microvascular reactivity in patients with type 2 diabetes mellitus. This chapter will explore the literature were listed as followed:-

1. Type 2 diabetes mellitus
    - 1.1 Definition and diagnosis of type 2 diabetes mellitus
    - 1.2 Prevalence and epidemiology of type 2 diabetes mellitus
    - 1.3 Pathophysiological of type 2 diabetes mellitus
    - 1.4 Clinical symptoms of type 2 diabetes mellitus
    - 1.5 Treatment of type 2 diabetes mellitus
2. Oxidant and antioxidant
    - 2.1 Free radicals and oxidative stress
    - 2.2 Antioxidant
    - 2.3 Free radicals, antioxidant and exercise
    - 2.4 Oxidative stress and type 2 diabetes mellitus
3. Exercise
    - 3.1 Exercise and type 2 diabetes mellitus
    - 3.2 Exercise recommendation for type 2 diabetes mellitus
    - 3.3 Water-based exercise

1. Type 2 diabetes mellitus

1.1 Definition and diagnosis of type 2 diabetes mellitus

Type 2 diabetes mellitus (known as non-insulin-dependent diabetes mellitus, NIDDM) is accounting for 85-95% of all cases of diabetes. As well as often remains undiagnosed until complications become symptomatic [8]. Thus, particularly in patients with type 2 diabetes, early detection of the disease allows them to make lifestyle changes and initiate medical therapy, which can reduce the appearance of complications. Considering the higher and constantly increasing prevalence of type 2 diabetic patients and the great potential of non-pharmacological measures

associated with small modifications in lifestyle for primary and secondary disease prevention, the medical community has granted much attention to this type of diabetes mellitus and has defined a set of rules for early diagnosis of the disease. Tests to detect the occurrence of type 2 diabetes should always be considered in subjects who have one or more of the risk factors for the development of the disease, as shown in Table 1 [9].

Table 1 Criteria to check for the existence of type 2 diabetes mellitus in adults (adapted from the American Diabetes Association [9])

1	Subjects aged more than 45 years (When normal, the checkup should be repeated every 1–3 years)
2	Occurrence of symptoms like polyuria, polydipsia, and unexplained weight loss
3	Asymptomatic subjects carrying at least one of the following risk factors:
	3.1 Have a first-degree parent with diabetes
	3.2 Overweight or with a BMI >25 kg/m^2
	3.3 History of impaired glucose tolerance or impaired fasting glucose in a previous test
	3.4 Hypertension (>140/90 mmHg)
	3.5 A plasma high density lipoprotein cholesterol level <35 mg/dl and/or a plasmatic triglyceride level >250 mg/dl
	3.6 History of gestational DM or of a newborn child weighing more than 4.08 kg
	3.7 Sedentary lifestyle
	3.8 Other clinical conditions associated with insulin resistance, such as polycystic ovary syndrome and acanthosis nigricans

1.2 Prevalence and epidemiology of type 2 diabetes mellitus

The World Health Organization has estimated that, globally, the number of people with diabetes in 2000 was 177 million, with 33 million in Europe. By 2030, this will increase to 370 million worldwide and much of the anticipated increase will occur in economically developing countries including Thailand [10].

Diabetes already accounts for a large proportion of spending on health care and the anticipated increase in cases has significant implications for economic productivity and resource allocation. Because the long-term complications of diabetes are an important determinant of the total cost of care, there is the prospect of major additional demands on health services in the years to come.

For several years that everyone was facing a worldwide epidemic of type 2 diabetes. It causes both social, economic, and health consequences to the nation. Disability-adjusted life years lost (DALYs) due to diabetes ranked the 3rd and the 5th for Thai women and men adult populations respectively [11]. The type 2 diabetes prevalence increases progressively due to population growth, aging, urbanization, and increasing prevalence of obesity and physical inactivity, the diabetes epidemic will be worse in the future. It was estimated that the number of people with diabetes in adults aged 20 years and over (especially after 35 years old) in Thailand will increase from 1,017,000 in 2000 to 1,923,000 in 2025 [12]. Early interventions could be cut down on the disease development and their complications among this population subgroup are then expected to have tremendous positive health and other impacts on the individual and the country as a whole.

The interactions of environmental and behavioral factors, such as physical inactivity, obesity, and stress, with genetic factors are responsible for the alteration in glucose homeostasis by impaired glucose tolerance, which can result from pancreatic β cell dysfunction and/or from the increase of insulin resistance in target tissues [13, 14]. Eighty-five to 95% of all diabetes mellitus cases are characterized by glucose intolerance induced by insulin resistance. Independent of its origin, the resultant chronic hyperglycemia may negatively influence the structure and function of many organs, particularly the cardiovascular, nervous, and renal systems [8, 15].

1.3 Pathophysiological of type 2 diabetes mellitus

Type 2 diabetes is characterized by hyperglycemia, increase free fatty acids, and insulin resistance provokes molecular mechanisms that alter the function and structure of blood vessels [16]. These include increased oxidative stress, disturbances of intracellular signal transduction such as activation of Protein Kinase C (PKC), and activation of receptor for advanced glycation end product (RAGE) lead to cutaneous microvascular dysfunction in diabetic patients.

Hyperglycemia induces a series of cellular events that increase the production of reactive oxygen species (ROS) then promotes a cascade of cutaneous microvascular processes that activate increasing inflammation and decreases endothelium-derived NO. Hyperglycemia also increases the production of the lipid second messenger diacylglycerol, which causes the membrane translocation and activation of PKC. Activation of PKC inhibits the activity of the phosphatidylinositol 3 kinase pathway, thereby limiting activation of Akt kinase and subsequent phosphorylation of NOS, which results in less NO production [17].

In addition, hyperglycemia and free fatty acids induced oxidative stress leads to the activation of stress-sensitive signaling pathways. This, in turn, worsens both insulin secretion and action, leading to overt type 2 diabetes. Furthermore, oxidative stress induced by elevations in glucose and FFA plays a key role in causing insulin resistance and β-cell dysfunction. Thus, treatment aimed at reducing the degree of oxidative stress and activation of stress-sensitive signaling pathways would appear to warrant consideration for inclusion as part of the treatment program for patients with type 2 diabetes [5].

Circulating levels of free fatty acids are elevated in diabetes because of their excess liberation from adipose tissue and diminished uptake by skeletal muscle. Free fatty acids may impair cutaneous microvascular function through several mechanisms, including increased production of ROS, activation of PKC, and exacerbation of dyslipidemia [18]. Elevation of free fatty acid concentrations activate PKC and decrease insulin receptor substrate-1 associated phosphatidylinosital-3 kinase activity. These effects on signal transduction may decrease NOS activity as results in decrease NO production.

Insulin resistance most often precedes the onset of type 2 diabetes may be explained by alterations in intracellular signaling that reduce the production of NO. Also, insulin resistance is associated with elevations in free fatty acid levels. Abdominal adipose tissue, the type found prominently in type 2 diabetes, is more insulin resistant and releases more free fatty acids compared with the type of adipose in other locations. Activating lipoprotein lipase to metabolize these free fatty acids increases insulin sensitivity. Thus, free fatty acid induced alterations in intracellular signaling may also contribute to decreased NOS activity and reduced production of NO in insulin-resistant states of type 2 diabetes [19].

There is a large amount of experimental and clinical data indicating that cutaneous microvascular dysfunction is involved in disease states such as

atherosclerosis, hypertension, heart failure and diabetes. Decreased endothelium-dependent vasodilation of the forearm and coronary circulation has been associated with human aging [20], but less information about microcirculatory function and its relationship with exercise in type 2 diabetes has been reported to date. Previously reported experimental data have shown that chronic exercise training is associated with enhanced endothelium-dependent vasodilation which is accompanied by increased NO production in large coronary vessels and micro-vessels in animals [21]. Urinary excretion of NO metabolites increases with increasing levels of chronic physical activity and, in patients with cardiovascular disease, levels of urinary NO metabolites increase in direct relationship with the gain in functional capacity, suggesting that increased NO production may be a major adaptive mechanism by which aerobic exercise training benefits the cardiovascular system [22].

Exercise induces a complex integrated physiological response that involves activation of circulating hormones and local autacoids, such as catecholamine, adenosine and ATP, which could exert an influence in the long-term regulation of NO biosynthesis. In addition, exercise increases myocardial oxygen consumption and reduces vascular resistance at the coronary level. This effect will elicit cutaneous microvascular shear stress, which may increase cutaneous microvascular NO synthase (eNOS) expression and activity [23]. Other mechanisms implicated in the increased NO facilitated by exercise-induced expression of antioxidant enzymes [24]. It is possible that the beneficial effects of regular exercise training could be mediated by the increase in NO availability and action, and this could be improving cutaneous microvascular function in some clinical conditions associated with type 2 diabetes.

In diabetes, cutaneous microvascular cell dysfunction is characterized not only by decreased NO but also by increased synthesis of inflammatory cytokine. The metabolic abnormalities that characterize type 2 diabetes associated with chronic inflammation [25]. Peterson and co-worker study recently indicates that physical inactivity can increase pro-inflammatory independently of obesity, and exercise may induce anti-inflammatory mediators [26]. Although exercise training has been proven beneficial in treatment of type 2 diabetes, this preventive and therapeutic modality remains underused [27]. Previous studies show that moderate exercise enhances T-cell function and decreases respiratory infections, which suggests that the volume of exercise is a critical element of inducing a positive or negative immune response in diabetic patients [28].

In the reference paper, the decrease in TNFα was related to the decrease in FFM [29], although, this may not be apply to all situations as unchanged and increased values of FFM after endurance training were also reported [30]. However, previous studies showed that long-term gentle jogging increased insulin action despite no influence on BMI or Vo_2 max [31].

1.4 Clinical Symptoms of type 2 diabetes mellitus

Type 2 diabetes may be present for many years before a diagnosis is made. The development of this disease is a gradual process. It is quite common for a diagnosis to be made by chance, with the patient having a routine blood test or being in hospital for another reason. The common symptoms to be aware of include:

Polydipsia: chronic thirst and craving for fluids. A person with diabetes who has high blood glucose concentrations will constantly crave water.

Polyuria: frequent urine excretion. In addition to being thirsty the patient will make frequent visit to the bathroom, even throughout the night.

Weight loss may be an additional symptom, especially weight loss over a short period of time.

Blurred vision. In some cases, where blood glucose concentrations are very high, vision may become blurred.

Many who develop type 1 diabetes have some or all of these symptoms, but those with type 2 diabetes may remain asymptomatic. The fact that one third of the population with diabetes do not know they have the disease underscores the lack of symptoms experienced by many.

1.5 Treatment of type 2 diabetes mellitus

The goal of diabetes mellitus treatment is directed toward glycemic control, the correction of all metabolic and functional abnormalities such as dyslipidemia, hypertension, and obesity, and the control of morbidity and mortality related with the macro- and microvascular complications [9]. Apart from medication, the standard therapeutic procedures for the treatment of type 2 diabetes mellitus include correct dietary control and regular exercise [32, 33]. According to these authors, the management of each patient should be individualized and adjusted to age, medical history, earlier physical activity, eating habits, and sociocultural factors. Especially before starting an exercise program, a medical examination is necessary to

detect other diseases or diabetic complications that might represent contraindications.

Glycemic control

Glycemic control is a main objective in diabetic patients. Several studies have shown that improved glycemic control is associated with decreased rates of chronic complications, such as retinopathy, nephropathy, neuropathy, and cardiovascular diseases [34-37]. Capillary blood glucose monitored by the patient himself represents a useful procedure to control glycaemia and to adjust medications or physical activity/exercise prescriptions. Usually, capillary blood glucose monitored once a day is important for the patient to check to prevent asymptomatic hypoglycemia. Glycated hemoglobin (HbA1c) is required to estimate the mean glycemia over the preceding 2–3 months and must be measured at least twice a year to determine whether metabolic control is maintained within a normal range [38]. Thus, HbA1c is useful in assessing treatment efficiency and in comparing the result of the patients self-reported testing (Table 2).

Table 2 Parameters used for a glycemic control in type 2 diabetic patients (adapted from the American Diabetes Association [9])

Blood samples	Glucose plasma concentrations	HbA1c plasma levels
Fasting condition	90–130 mg/dl (5.0–7.2 mmol/l)	<7.0%
Postprandial peak level	<180 mg/dl (<10.0 mmol/l)	

Therapeutic exercise

Exercise is one of the main therapeutic measures to deal with high blood glucose levels in diabetic patients [9, 39-41]. It is believed to control hyperglycemia through the improvement of peripheral insulin sensitivity particularly in skeletal muscle [42, 43]. In addition, many studies have also described exercise as a countermeasure against many abnormalities observed in diabetic patients, such as hyperlipidemia, hypertension, and a tendency for hyper coagulation [44-46], which are considered to increase the risk of macro- and microvascular complications [47, 48]. These and many other chronic complications of diabetes mellitus seem to be attenuated by the regular exercise.

2. Oxidant and antioxidant

2.1 Free radicals and oxidative stress

A number of complications arise as a consequence of macro and microvascular complications that result from diabetes; these deficits have a central role in the tissue-damaging effects of chronic hyperglycemia [6]. Since endothelial cells (as well as renal mesangial and Schwann cells) are unable to limit glucose transport as well as other cells do, they are more vulnerable to the toxic effects of hyperglycemia. In fact, from a cardiovascular medicine perspective, diabetes can also be classified as a cardiovascular disease [7]. Several studies have shown that diabetes mellitus (types I and II) is accompanied by increased formation of free radicals and decreased antioxidant capacity, leading to oxidative damage of cell components [8]. There are multiple sources of reactive oxygen species (ROSs) production in diabetes including those of mitochondrial and nonmitochondrial origins; ROS accelerates the four importantmolecular mechanisms involved in hyperglycemia-induced oxidative tissue damage. These four pathways are activation of protein kinase C (PKC),

increased hexosamine pathway flux, increased advanced glycation end product (AGE), and increased polyol pathway flux [9].

2.2 Antioxidant

Cells have evolved highly complex enzymatic and nonenzymatic antioxidant systems which work synergistically, and in combination with each other, to protect the body against free radical-induced damage. The most efficient enzymatic antioxidants involve glutathione peroxidase, catalase, superoxide dismutase, heme oxygenase-1 (HO-1), NAD(P)H quinone oxidoreductase-1 (NQO-1), and thioredoxin [63]. Non-enzymatic antioxidants include vitamins E and C, thiol antioxidants (glutathione, thioredoxin) [64]. These antioxidants are capable of combining with reactive oxidants to produce other less reactive species. SOD promotes the dismutation of the superoxide radical to form hydrogen peroxide (H_2O_2) and oxygen. Glutathione peroxidase (GPx) uses reduced glutathione (GSH) as a reducing equivalent to reduce H_2O_2 to form oxidized glutathione and water. Catalase converts H_2O_2 to water and oxygen. Further, GSH can remove selected oxygen radicals directly and assist in the recycling of vitamins C and E. The newly identified peroxiredoxin family is also a group of peroxidases that catalyze the reduction of H_2O_2 and so far at least six isoforms have been identified in mammalian cells [65]. Among them, peroxiredoxin III is synthesized with a mitochondrial targeting sequence (as is MnSOD) so that when it is transferred to mitochondria, its targeting residues are cleaved during maturation. Some studies suggest that peroxiredoxin III is a critical regulator of mitochondrial H_2O_2 concentrations, which promotes apoptosis in cooperation with other mediators of apoptotic signaling [66]. The specific localization of peroxiredoxin III within the mitochondria is thought to provide a primary line of defense against H_2O_2 produced by the mitochondrial respiratory chain [67].

Exercise training results in an upregulation of antioxidant defense mechanisms in various tissues, presumably due to increased levels of oxidative stress that occurs during exercise. Low/moderate amounts of ROS produced during regular skeletalmuscle work are a part of “hormesis”, which describes the generally favorable biological responses to low exposures to toxins and other stressors. A pollutant or toxin showing hormesis has opposite effects in small versus large doses.

Hormesis is characterized by stimulation at low doses and inhibition at higher doses, resulting in an inverted U-shaped dose response effect [68]. For example, exercise-induced increased production of ROS can be beneficial by evoking specific

adaptations, such as increased antioxidant/oxidative damage repairing enzyme activity, increased resistance to oxidative stress and lower levels of oxidative damage. On the other hand, excessive production of ROS is usually associated with detrimental effects.

2.3 Free radicals, antioxidant and exercise

Inflammation has a prominent role in the pathogenesis of several cardiovascular diseases. Atherosclerosis is an inflammatory disease that is mediated by monocyte derived macrophages which accumulate in arterial plaques and become activated to release cytokines that cause tissue damage [88]. Atherosclerotic plaques in type II diabetic patients have increased inflammatory properties and worse cardiovascular outcomes than plaques observed in non-diabetic subjects [89]. We reported that systemic inflammation precedes either hyperglycemia or oxidative stress in db/db mice [90]. As evidence accumulates favoring the role of inflammation during the different phases of atherosclerosis, it is likely that markers of inflammation such as high-sensitivity C-reactive protein (hs-CRP)may be increasingly used to provide additional insights on the biological status of atherosclerotic lesions. Several studies have shown that CRP and proinflammatory cytokines, including interleukin-6 (IL- 6) and tumor necrosis-α (TNF-α), are elevated in type II diabetic patients [89, 91]. CRP is considered to be an inde pendent predictor of cardiovascular events and of the outcome of acute coronary syndromes [92]. Diabetic patients can be grouped as being at low, intermediate, and high risk for cardiovascular disease based on their levels of hs-CRP [93]. Besides its role as a marker of systemic inflammation and a predictor of cardiovascular risk, CRP and other inflammatory cytokines also directly trigger vascular dysfunction [94], possibly via altering calcium channel expression and activity [95], upregulation of Rho-kinase expression and function [96], increasing the production of ROS [97], and/or enhancing cyclooxygenase expression [98]. In turn, cyclooxygenase enzymes cause vascular hypercontractility by increasing the synthesis of constrictor prostanoid(s) [99, 100] and excessive formation of ROS [101]. Cyclooxygenase inhibitors alleviate the augmented contractile responses in several animal models of diabetes [102–106]. These findings may partially explain the inconsistent and mostly disappointing results with antioxidant use in diabetic patients [64], since inflammation rather than oxidative stress may be the principle contributor to diabetic vascular dysfunction. In agreement with this concept is the finding that endothelial function improves in type 2 diabetic patients treated with rosiglitazone, an agent that reduces inflammation but not oxidative stress [107].

Exercise produces a short-term inflammatory response that is accompanied by leukocytosis, increases in oxidative stress, and plasma levels of CRP. This pro-inflammatory response is followed by a long term anti-inflammatory effect [108]. Regular exercise reduces CRP, IL-6, and TNF-α levels and also increases anti-inflammatory substances such as IL-4 and IL-10 [109, 110]. In healthy young adults, a 12-week, high-intensity aerobic training program down regulates cytokine release from monocytes [110]. In fact, even leisure time physical activity (e.g., walking, jogging, or running, etc.) reduces hs-CRP concentration in a graded manner [111]. Table 1 summarizes the findings of clinical studies on the effects of exercise on anti-inflammatory and antioxidant markers in diabetic patients.

2.4 Oxidative stress and type 2 diabetes mellitus

Oxidative stress plays an important role in the pathogenesis of cardiovascular disease, including atherosclerosis, hypertension, and the macro- and micro-vascular diseases associated with diabetes. Oxidative stress is defined as an increase in ROS and/or a decrease in the antioxidant defense mechanisms such as catalase, glutathione, superoxide dismutase (SOD) [49].

Recently, Melikoglu et al [50] also reported that regular long term training can induce antioxidant response to the oxidative stress. These results support the possibility that the beneficial effect of physical exercise on oxidative stress might be associated with increased antioxidant defenses.

Oxidative stress has been proposed to be a potential pathogenic mechanism linking obesity and insulin resistance with cutaneous microvascular dysfunction [51] and may explain the presence of inflammation in all of these conditions. Oxidative stress resulting from increased production of ROS (or their inadequate removal) plays a key role in the pathogenesis of late diabetic complications [52, 53]. Although our understanding of how hyperglycemia-induced oxidative stress ultimately leads to tissue damage has advanced considerably in recent years [54], effective therapeutic strategies to prevent or delay the development of this damage remain limited.

3. Exercise

3.1 Exercise and type 2 diabetes mellitus

Regular exercise is a recommended treatment for patients with type 2 diabetes because it enhances the glucose transporter 4 (GLUT4) expressions in trained skeletal muscle with a consequent increase in the glucose transport capacity

[55]. This mechanism may explain the enhanced sensitivity to insulin observed in physically active diabetic patients [56-59]. Increased mass in skeletal muscle, enhanced blood flow in muscle, greater density of insulin receptors, enhanced disposal of glucose in skeletal muscle, and a reduction in body fat could also contribute to this insulin sensitivity and improved glucose tolerance induced by exercise training in diabetic patients [40, 60].

As mentioned above, the aim of exercise in type 2 diabetic patients is to induce acute and chronic physiological changes, which improve insulin sensitivity and glucose tolerance. To maintain glucose homeostasis, glucagon and adrenalin are released during acute exercise, which increases hepatic glucose production; simultaneously, the insulin secretion by pancreatic β cells is reduced [61-63]. Thus, an increase in glucose utilization during acute exercise is accompanied by an increase in hepatic glucose production, an improved insulin sensitivity enhancing the binding of insulin to sarcolemmal receptors, and an increased GLUT4 expression [39]. From a chronic perspective, the improvement in insulin sensitivity is positively correlated with the intensity of aerobic endurance training, which is in accordance with the fact that insulin resistance in type 2 diabetic patients is associated with diminished physical fitness. This association of regular exercise leads to changes in body composition, a reduction in body fat and an increase in muscle mass.

When patients with type 2 diabetes participate in exercise training programs, metabolic control is improved, as evidenced for example by a decrease in serum triglyceride (TG) and very low density lipoprotein (VLDL) cholesterol and an increase in high density lipoprotein cholesterol at rest [64]. However, some studies have found that middle-aged subjects (age 40–54 years) who exercise regularly have a better response in metabolic control when compared to older subjects (age 57–61 years), perhaps due to different pre-training levels of metabolic control or to differences in the intensity of the training program or magnitude of chronic complications with advanced age [55].

The purpose of exercise training for the primary prevention and the treatment of lifestyle-related diseases are to improve insulin resistance. As in acute effects of exercise, during exercise the glucose uptake by the working muscles rises 7 to 20 times over the basal level depending on the intensity of the work perform. Intense exercise provokes the release of insulin-counter regulatory hormones such as glucagons and catecholamine, which ultimately cause a reduction in the insulin action [65]. As effects of long-term exercise training, continue exercise training

improves reduced peripheral tissue sensitivity to insulin in IGT and type 2 diabetes mellitus [66], and also improves abnormal lipid metabolism. Further, the water-based and land-based exercise training should be improving cutaneous microvascular function in type 2 diabetic patients.

Evidence for the benefit of exercise from previous studies shown that individuals who maintain a physical exercise and active lifestyle are less likely to develop insulin resistance, impaired glucose tolerance, or type 2 diabetes [67]. The effects of exercise training on glucose control and related physiological parameters have also been extensively studied in patients with type 2 diabetes. In 2001, Boule et al [68] published a meta-analysis showing beneficial effects of exercise training on one aspect of glucose control and percent of HbA1c in blood of diabetic patients. They also found reductions in two measures of abdominal obesity and little effect on the only other parameter they meta-analyzed: body mass. Fourteen studies in the meta-analysis, 12 studies used aerobic training and 2 studies used resistance training. Boule et al. found some physiological adaptations difference between effects of aerobic and resistance training, but there were insufficient studies of resistance training for this finding to be anything more than tentative. Aerobic exercise is widely recognized to reduce the risk of coronary heart disease, so much so that consensus panels routinely recommend physical activity as part of a cardioprotective regimen for healthy people. Surprisingly, despite the wealth of clinical and epidemiological data indicating benefit, the physiological or mechanistic basis of this protection is unknown. Some recent studies have suggested that exercise may promote cardioprotection through anti-inflammatory effects and that these effects may be dose dependent [6, 69, 70].

The effects of exercise training on the improvement of the metabolic profile have been controversial, but water-based and land-based exercise training might be effective in delaying or preventing type 2 diabetes mellitus. As for exercise intensity, the majority of studies in which the effects of exercise training were analyzed used high-intensity exercise such as bicycle ergometer or running (land-based) graded up to 70% to 90% of the Vo_2 max. McAuley et al. [71] showed that modest levels of dietary and exercise recommendations did not improve significantly the insulin sensitivity, but a more intensive program did. Therefore, they emphasized that intensive lifestyle changes were necessary to improve insulin sensitivity. Similarly to the evidence, Marek and co-worker [72] found that regular physical exercise decreases tumor necrosis factor-α (TNFα) system activity and that decrease may be

responsible for the concurrent increase in insulin sensitivity. Theirs result could be explain that changes in the TNF system are related to the improvement in insulin sensitivity during prolonged physical activity. There are also other changes resulting in enhanced insulin action following training, such as increased amount of the glucose transporter GLUT4 [73], increased capillarization of the muscle tissue [74] and enhanced insulin signal transduction, for example at the level of phosphatidylinositol 3-kinase activity [75]. Therefore, a decrease in TNFα expression is only one among many other proposed mechanisms.

3.2 Exercise recommendation for type 2 diabetic patients

Exercise programs for diabetic patients should consist of aerobic endurance exercise, which enhances cardiorespiratory fitness, muscular strength, and endurance, and modifies body composition. However, in patients with type 2 diabetes, this type of training together with resistance training, as opposed to aerobic training alone, seems to induce better positive adaptations of glucose control, insulin function, muscular strength, and exercise tolerance [76, 77]. Nevertheless, resistance exercise or exercise performed at high intensities should only be carried out by individuals without any diabetes mellitus associated chronic complications, such as proliferative retinopathy or hypertension [44, 63, 65, 78].

Table 3 describes the exercise components recommended for type 2 diabetic patients. To achieve cardiorespiratory and metabolic improvements, the exercise intensity must be individually adjusted from low to moderate, in correspondence to 40–70% of maximum oxygen consumption [35]. The use of heart rate for monitoring exercise intensity may not be suitable in patients with type 2 diabetes because they may develop autonomic neuropathy, which affects the heart rate response to exercise [63]. Intensity should rather be estimated using ratings of perceived exertion (RPE), with recommended RPE of 10–16 (very light to hard). The duration of exercise is directly related to the caloric expenditure required and is inversely related to the intensity. At least 30 min of continuous and moderate exercise should be performed in each training session. If weight loss is the main target of exercise, its intensity needs to be set from low to moderate (50% VO2 max), and the duration needs to be increased up to 60 min.

Table 3 Exercise characteristics recommended for type 2 diabetic patients (based on data from the American Diabetes Association [79], Eriksson [39], Leutholtz and Ripoll [41], and Peirce [80])

Type	Duration	Frequency	Intensity
Aerobic exercise such as walking,	At least 30 min/session	3–5 days a week	40–60% VO_{2max} or RPE 10–16 or
running, bicycling,	or 150 min/week		50–70% of MHR
swimming	90 min/week		>60% VO2 max or >70% of MHR
Resistance	10–15 repetitions	2 days a week	40–50% of 1 RM
Warm up/cool down	5–10 min	Before and after exercise	Low intensity

To improve cardiovascular performance, the recommended frequency of exercise is between three and five sessions per week. Moreover, obese diabetic patients may need to participate in daily physical activity to maximize caloric expenditure for effective weight management. Normally, patients with type 2 diabtes are elderly, showing a decline in muscle mass associated with a reduction in metabolic function. Resistance training programs increase muscle strength and muscle mass. Patients, however, with blood glucose higher than 300 mg/dl, congestive heart failure, uncontrolled arrhythmias, severe valvular malfunction, or an aerobic capacity lower than five MET, are not allowed to perform this type of training [63, 64].

Resistance training should be performed at low intensity [40–50% of 1 repetition maximum (RM)], with 10–15 repetitions for each muscle group and a 3:1 ratio of endurance/resistance sessions. Low intensity is suggested to avoid exaggerated blood pressure. In the same way, regular breathing during exertion can also prevent a severe rise in cardiac afterload. The type of weightlifting equipment that could be used depends on the patients' preferences and abilities [39, 70].

There are several safe forms of exercise such as walking, jogging, bicycling, and swimming, which allow patients to maximize their caloric expenditure and to improve physical fitness enjoyably and effectively. Long-term exercise programs are usually accompanied by a dropout rate of 90% of the initial

participants after 1 year, while in short-term exercise programs, dropout rates range from 35–80% during the first 3–6 months [69]. Many reasons could contribute to refraining from maintenance of exercise, including lack of time, travel problems, expenses, and injury, but waning motivation seems to be the main cause [33]. Setting of personal goals for individuals is necessary, both for the diabetic patients and staff involved in the program, because both are equally important in providing encouragement to continue exercising. Even though participation in regular exercise programs may be low, a good strategy to maintain patients' involvement in such programs can be enhanced by routine activities of daily living. Lifestyle modifications such as stair climbing instead of using an elevator, household tasks, and gardening can provide appropriate physical activity with increased calorie burning [39].

Aerobic exercise has consistently been shown to improve glucose control, enhance insulin sensitivity, and improve cardiovascular risk factors such as visceral adiposity, lipid profile [44], arterial stiffness, and cutaneous microvascular function [81]. Consistent with this evidence, the American Diabetes Association (ADA) recommends in individuals with type 2 diabetes perform at least 150 min of moderate-intensity aerobic exercise and/or at least 90 min of vigorous aerobic exercise per week [82]. Although a lifestyle modification of this nature could have substantial impact on the metabolic and cardiovascular health of this population, it is often difficult for those who have been habitually sedentary to adhere to these guidelines. Indeed, a recent population-based study found that only 28% of individuals with type 2 diabetes achieve these recommendations. Unfortunately, it is frequently those who would benefit the most from aerobic exercise that have the greatest difficulty performing it. For individuals with severe obesity, arthritis, physical disabilities, and/or diabetes complications, even walking for 20–30 min may be challenging, uncomfortable, and/or painful to perform. With the continued increase in the prevalence of type 2 diabetes [83], it is evident that alternate forms of physical activity that produce similar metabolic improvements to aerobic exercise may be beneficial in the management of this disease.

However, because of complications or coexisting conditions such as peripheral neuropathy or degenerative arthritis, those with type 2 diabetes may require alternative modes that are non-weight bearing activities such as stationary cycling, swimming, and aquatic exercise activities. In addition, most of studies only in the effect of land-based exercise regimens. Whereas, study characteristics and magnitude effects of water-based exercise training on cutaneous microvascular vasodilator function, HbA1C and other measures of glucose control included

physiological parameters related to complications of diabetes in type 2 diabetic patients are still limited.

3.3 Water-based exercise

Water-based exercise or aquatic exercise is defined as therapeutic and exercise activities performed in the water carried out in heated pools by a variety of providers that promote and enhance physical and mental fitness. It is said to incorporate the use of the "healing powers" of water. Many practitioners of 'aquatic exercise' feel water has significant curative properties and that, unlike other medicinal agents, is not harmful or potentially toxic [63]. The water's unique properties allow the pool to provide an environment for people of all abilities. Aquatic exercise training incorporates individual assessment, evidence-based practice, and clinical reasoning skills to devise treatment plans based on the principles of hydrostatics, hydrodynamics, physiologic effects of immersion and sometimes its temperature [84]. Water-based exercise training offers several benefits over land-based exercise training for people with type 2 diabetes. Buoyancy reduces loading across joints affected by pain and allows the performance of functional closed-chain exercises that otherwise may be too difficult on land. Water turbulence can be used as a method of increasing resistance, and percentage of body weight borne across the lower limbs can be decreased or progressed in proportion to the depth of immersion [85]. The warmth and pressure of the water may further assist with pain relief**,** swelling reduction, and ease of movement.

Water lends itself to a well-balanced workout that improves all major components of physical fitness-aerobic training, muscular strength and endurance, flexibility and body composition. Shallow water programming is performed in waist to chest depth. The feet remain in contact with the pool bottom during most of the workout providing a low impact training option. Currently, hydrotherapy is applied in many rehabilitative programs. Examples of its use include improving muscular and cardiopulmonary endurance in the elderly, osteoarthritis and rheumatoid arthritis, various dermatological conditions, chronic heart failure, reducing spasticity in traumatic brain injury patients, burns and wound healing, pressure ulcer [86]. Although many studies have reported positive effects of hydrotherapy interventions in randomize controlled trial with various conditions. Most studies have employed outcome measures for impairments such as pain, strength, range of motion, cardiovascular fitness, or inflammation [87] that can impede function. There is little evidence attesting to its efficacy on cutaneous microvascular function in type 2

diabetic patients. Furthermore, most water-based exercise programs demonstrate little consideration of hydrostatic or hydrodynamic principles in their choice of exercises, thus reducing the potential for benefit from the overall program. The present study is design to address the limitations of previous studies through the use of an adequately powered randomized controlled trial and a functional progressive intervention that maximized the unique properties of water to optimize outcomes.

Aquatic exercise has been shown to improve impairments and functional limitations in individuals with various diseases and to improve the fitness of older women [88]. Elderly women participating in a 12-week water exercise class demonstrated significant improvements in oxygen consumption, muscular strength, agility, skin-fold thickness, and cholesterol as compared with control group. Although most studies have reported positive effects of water-based exercise measures including in osteoarthritis, cardiovascular disease, and inflammation. There is little evidence attesting to its efficacy on cutaneous microvascular function in type 2 diabetic patients.

Water cools more efficiently than air, so when exercising in the water the body is able to eliminate excess heat more effectively. This is not to say that you will not sweat during a workout in the pool, but water helps prevent overheating and washes away the perspiration as you exercise. Because the water cools the body quickly, it is imperative that you begin every workout with a “thermal warm up” designed to elevate the body’s core temperature, warm the muscles and prepare the joints for the increased workload to come. Even at the recommended temperature of 83-86 degrees Fahrenheit (28-30 degrees Celsius), a proper warm up is necessary to prevent injury and provide comfort. (NOTE: Special populations and specialty training may require deviations from this recommended range). With type 2 diabetes are over 40 years of ages and many are obese, the body does not produce adequate insulin. Exercising vigorously just once a week reduces your risk of type 2 diabetes by 23%; exercising five or more times per week reduces the risk by 42%. A regular exercise program is considered to be a cornerstone to diabetic care. Before initiating an exercise program, should consult physician as modifications in medications or exercise scheduling may be in order. Shoes should always be worn in and around the pool area and the diabetic should be aware that might be at a higher risk for heat related illness and should avoid exercising in excessive heat. [References: AEA Aquatic Fitness Professional Manual and Fitness Empowerment of Active Adults-FEOAA, 2003]

Water temperature that is too warm or too cold can add a significant thermal load to the cardiovascular system. Choukroun and Varene [89] found cardiac output to be unchanged from 25°C to 37°C (77°F to 93°F) but significantly increased at 40°C (104°F); oxygen consumption was significantly increased at 25°C (77°F). Several studies have found a decreased HR in subjects exercising in cold water and exercising in very warm water can increase HR. Most pool temperatures range from 27°C to 35°C (81°F to 95°F) and thermoneutral temperature is suggested to be approximately 34°C. Thus, the current pool temperature in the place of present study is in range and potential effects on the patient.

Heart rate responses differ when exercising in the water than when exercising on land. Typically, aquatic exercisers experience a reduced heart rates response such as lowered pulse rate, but the water should not be considered less effective. Studies have shown that oxygen consumption, the true measure of the cardiovascular benefits, is comparable to a similar program on land, although the heart rate response is lower. Several factors, some of which have been previously mentioned, influence the exercising heart rate when submerged in the water to mid-chest: Lessened gravity allows a more efficient return of blood to the heart from the extremities. The cooling effect of water reduces the workload on the heart. (One function of the heart is to keep the body cool during sustained exercise.) Hydrostatic pressure, the pressure that the water exerts on the body while submerged, assists in blood flow and improves the exchange of oxygen into the blood.

When training in the pool, the increase in blood flow to the working muscle and the increase cardiac load resulting from hydrostatic pressure still occurs. Most studies have found the HR to be lower or unchanged compared with similar cardiovascular activity on land [90]. When performing resistive exercise in the pool, be sure to realize that most muscle contractions are concentric because of the negation of gravity. The strength training principles and progressions used in water-based activities are the same as those used on land. These include variables such as frequency, intensity, and duration. As with techniques to increase mobility, progress traditional strength and endurance training exercises to address functional limitations and disabilities as quickly as possible.

Thus, land-based exercise regimen exerts positive improvements for diabetes group. As water-based exercise becomes more and more popular in Thailand, we are based on the unclear understanding on effectiveness of this type of exercise on diabetes patients. It is believed that if this water-based exercise gives rise

in positive outcomes in other disease-specific patients. This exercise should exhibit similar outcome in type 2 diabetic patients. The second aim of this study is to compare the effectiveness of two forms of exercise during the 12-week training on physiological and cutaneous microvascular reactivity in a group with type 2 diabetic patients, and further, to determine which method of exercise is more effective in improving fasting blood glucose, glycosylated hemoglobin A1c (HbA1c), health related physical fitness, and cutaneous microvascular dysfunction in type 2 diabetic patients. The primary hypothesis is that water-based exercise training can improve cutaneous microvascular dysfunction in type 2 diabetes. The secondary hypothesis was that water-based exercise training would result in greater improvements on cutaneous microvascular dysfunction in type 2 diabetes patients than land-based exercise training.

CHAPTER III

METHODOLOGY

This study aims to determine the effects of water-based exercise training and land-based exercise training on physiological adaptations and cutaneous microvascular reactivity in type 2 diabetic patients. The experimental protocols were divided into 4 parts. First, physiological characteristics and physical fitness in all subjects were examined. Second, blood chemical data i.e. fasting blood glucose, glycosylated hemoglobin (HbA1c), lipid profile, insulin, C-reactive protein (CRP), and malondialdehyde (MDA) levels were analyzed. Third, exercise intervention i.e. mode, frequency, duration were performed. And Fourth, cutaneous microvascular reactivity were assessed. All protocol and procedures employed in this study were reviewed and approved by the supreme patriarch center on aging research and ethics committee, Department of Medical Service, Ministry of Public Health, NO 14/2009 (Appendix A).

Sample group

Fifty-three elderly subjects with diagnosis of type 2 diabetes mellitus aged 60 to 70 years were recruited from the supreme patriarch center on aging and Yanasungwararam hospital, Chon-buri province, Thailand. The participants were randomly allocated into equal numbers, ages and gender ratios into 4 groups with an equal number of subjects; Wex; Water-based exercise training, Wco; Water immersion, Lex; Land-based exercise training, Lco; non exercise control.

Potential participants received information about the study, screen for eligibility, and invite to participate. All subjects have duration of type 2 diabetes at least 3 years and regularly physical activity before the test. Physiotherapist assessed the patients' capability of starting exercise in the gym or pool safely, and obtain written inform consent before baseline assessment.

Inclusion criteria

1. All participants were patients with type 2 diabetes as defined according to criteria of the American Diabetes Association, 2007 and a baseline glycosylate hemoglobin (HbA1c) value of 7% to 9% (moderate severity) (James et al. 2002).

2. All participants were patients with type 2 diabetes at least 3 years, but not more than 10 years and non-previous (6 months) exercise training.

3. All participants were screened by physical activity readiness questionnaire (PAR-Q) (Appendix B) and general health questionnaire (Appendix E). They were free from diabetic nephropathy, diabetic retinopathy, severe diabetic neurophathy, severe cardiovascular disease and severe cerebrovascular disease under approval of attending physicians.

4. All participants were encouraged to follow their habitual life style including medication throughout the whole investigation period.

5. All participant received information sheet for research participant (Appendix C) and signed the inform consent (Appendix D).

Exclusion criteria

1. The participants were illness or infection.

2. The participants were not willing to continue in the experiment.

3. The participants completed less than 80% of the training schedule.

Data collection

A 12-week intervention took place at gymnasium, the supreme patriarch center on aging, Chon-buri province, Thailand. All participants (Wex, Wco, Lex, Lco) reported to the laboratories 2 days prior to the intervention. On the first day, after 8-hour of overnight fasting, the venous blood sample was collected from the ante-cubital vein. After having breakfast for 2 hours, the participants were asked to perform health-related physical fitness assessment**.** Post-occlusive reactive hyperemia data were performed on the same day. These investigations were repeated in all participants after the finish of exercise intervention.

Instruments

Instrument used in the selection of the sample

1. The patient/ Participant Information Sheet

2. The Informed Consent Form

3. The Physical Activity Readiness Questionnaire (PAR-Q)

4. The general health history questionnaire

5. Medical assessment record form

Instrument for exercise training program

1. Treadmill (Quinton, USA)

2. Heart rate monitors (Sport tester PE 3000, Finland)

Instrument for measuring physiological data variables

1. Bioelectrical impedance analysis (BIA-101 impedance analyzers, USA)

2. Digital blood pressure (Omron, Japan)

3. Weight and height digital scale (Detector, USA

4. Heart rate monitors (Sport tester PE 3000, Finland)

Instrument for measuring blood chemical data variables

1. Centrifugator

2. Freezer -40 $^{\circ}$C

Instrument for measuring physical fitness variables

1. Cardiopulmonary gas exchange system (Vmax 29, USA)

2. Treadmill (Quinton, USA)

3. Grip dynamometer (D-type No.1857, Tokyo Japan).

4. Back and leg dynamometer (No.1281 Takei kiki & Company, Tokyo Japan).

5. Sit and reach box (No.1229 Takei kiki & Company, Tokyo Japan).

7. Heart rate monitor (Sport tester PE 3000, Finland)

Instrument for measuring cutaneous microvascular variables

1. Laser Doppler flow meter (DRT4 Moor LAB, Moor instrument, England)

Places of study

Each subject was informed of the purposes, methods, procedures, the possible risks and benefits of the study. Written informed consent forms document were obtained from all subjects in this study prior to giving the test. The experiments are conducted in the same period of time at swimming pool and gymnasium of the supreme patriarch center on aging, ministry of public health, Chonburi province. Ambient temperatures at the swimming pools and room are 29.5 ± 5.5 ℃ and 31.5 ± 6.0 ℃ with 86.0 ± 10.0 % relative humidity.

Methodology

1. All volunteers have been aware of the details to perform the testing and data collection and signed the informed consent form.

General history of each subject were obtained from questionnaire (Appendix F) in which duration of type 2 diabetes, medical history, injury, daily physical activities, past illness and health status were included. Subjects with history of kidney, liver, and other diseases which could be aggravated by water-based and land-based exercise were excluded.

2. The sample account for 4 groups by using interventions data.

Group I: Subjects were performed water-based exercise by aqua-aerobic exercise in the indoor swimming pool with control temperature at 30-34 degree celsius at moderate intensity or 70% maximum heart rate, 13 persons. The target heart rate used this following formula;

HRmax = 220 – age (year)

Target heart rate = % Intensity: (70%) x HRmax

Group II: Subjects were performed water-based control by immersion in the indoor swimming pool with control temperature at 30-34 degree celsius, 15 persons.

Group III: Subjects were performed land-based exercise by land-aerobic exercise in the gymnasium at moderate intensity or 70% maximum heart rate, 10 persons.

Group IV: Subjects could do daily life as usual but no exercise program, 15 persons.

Exercise training protocol

Subjects were separately attended water-based exercise training in the indoor swimming pool with control temperature at 30-34 degree celsius or land-based exercise training in the indoor gymnasium at the same temperature and humidity. One classes for 50 minutes, three times a week for 12 weeks with similarly pattern in both exercise groups. Aqua-aerobic exercise program was designed by the multidisciplinary team of the supreme patriarch center on aging and professional team from Mahidol and Chiangmai University which was approved by Division of Medical Service, Ministry of Public Health.

Mode: land-based or water-based exercise training

Frequency: 3 times per week, 12 weeks

Duration: 50 min; 10 min warm-up, 30 min exercise, and 10 min cool-down

The warm up in the water-based exercise group consist of head, neck, upper and lower limbs stretching. The aquatic exercises is aerobic exercise which is the whole body exercise in all directions, for example shoulder adduction and abduction, shoulder horizontal adduction and horizontal abduction, trunk flexion and extension, trunk rotation, hip flexion and extension, hip adduction and abduction, knee flexion and extension, ankle dorsiflexion and plantar flexion. Cool down involved walking forwards, sideways, and backwards through the water, and knee cycling.

The warm up in the land-based exercise group involved about ten minutes of stretching. The gym exercise is aerobic exercise and cool down consist of upper and lower limbs stretches. The same protocol for increasing exercise intensity in the water was also applied in the gym.

Intensity: 70% HR_{max}; HR_{max} = 220 – age (year)

3. The data collection protocol that is defined (Figure 2). Subjects all group have been tested parameters before and after experiment

Parameter Assessment

Hemodynamic data, health-related physical fitness data, biochemistry data and cutaneous blood flow data were collected at baseline and after 12 weeks.

Physiological characteristics

1. Hemodynamic data (Appendix G)

Resting heart rate

The participants sat at least 5 minutes for resting period prior to the measurement. The resting heart rate was measured with heart rate monitor (Sport tester PE 3000, Finland)

Resting blood pressure

The participants sat at least 5 minutes for resting period before the measurement. The blood pressure was measured with digital blood pressure (Omron, Japan). The systolic blood pressure and diastolic blood pressure were recorded in unit of millimeters of mercury (mmHg).

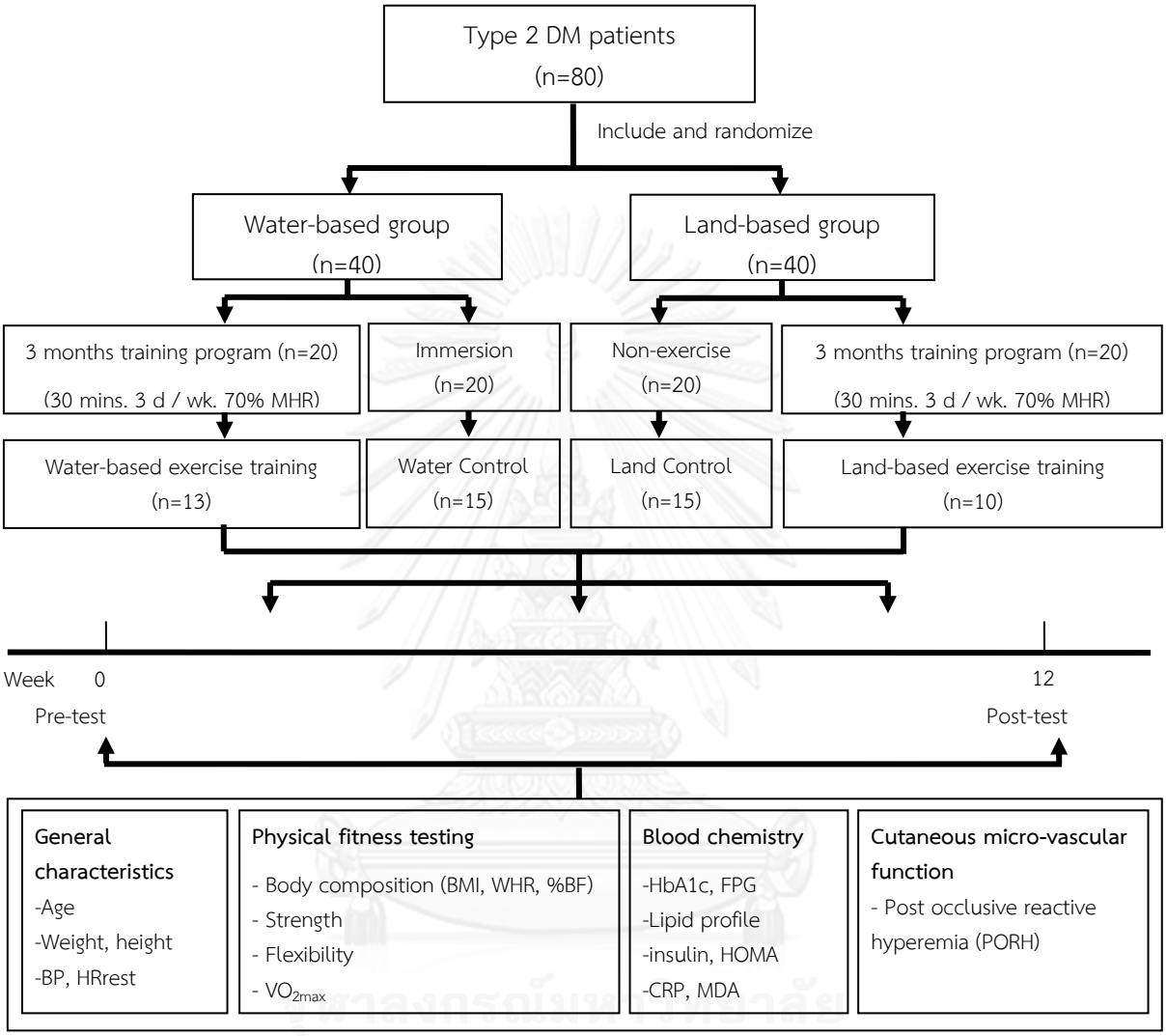


Figure 2 Data collection protocol

2. Health related physical fitness testing (Appendix G)

Body composition assessment

Direct segmental multi-frequency bioelectric impedance analysis method is used to measure percentage body fat, a body composition analysis device (BIA-101 impedance analyzers, USA) Subjects take off their shoes and socks before measuring. BMI is calculated as body weight x height^{-2} and expressed in kg/m^2. The waist circumference is measured at the smallest circumference between the rib cage and the iliac crest, with the subject in the standing position.

$$\text{BMI} = \frac{\text{Body weight (kg)}}{\text{Height (m}^2\text{)}}$$

Body weight and height of each subject are measured using weight and height digital scale (Detector, USA). Per cent body fat is determined using the bioelectrical impedance analysis (BIA-101 impedance analyzers, USA). Subject's per cent body fat is measured after 12 hours fasting. Bio-impedance measurement is taken on the right side of the body with the client lying supine on a nonconductive surface in a room with ambient temperature. The skin of each subject at the electrode sites is cleaned with alcohol pad. Then, the sensor (proximal) electrodes are placed on the dorsal surface of the wrist so that the upper border of the electrode bisected the head of the ulna and the dorsal surface of the ankle so that the upper border of the electrode bisects the medial and lateral malleoli. A measuring tape and surgical marking pen could be used to mark these points for electrode placement. After that the source (distal) electrodes are placed at the base of the second or third metacarpo-phalangeal joints of the hand and foot. The distance between the proximal and distal electrodes is at least 5 cm. The lead wires are then attached to the appropriate electrodes. Red leads are attached to the wrist and ankle, and black leads are attached to the hand and foot. The subject's legs and arms are abducted approximately 45°. Therefore, there are contacts between the thighs and between the arms and the trunk. Resistance and reactance are measured. The test repeats three times and the best value is recorded.

The complementary computer software allows to estimate subject's body composition (per cent body fat, body fat, per cent lean body mass, lean body mass, per cent total body water, total body water, and body mass index) based on physical characteristics i.e., age, gender, race, physical activity level, and level of body fatness.

Isometric strength test

Isometric leg strengths of knee extensors (quadriceps muscle) and knee flexors (hamstrings, gastronemius muscle, etc.) were measured with a back and leg dynamometer (No.1281 Takei kiki & Company, Tokyo Japan). Each subject was performed a maximum exertion force while standing with flexed knee approximately at 130°-140° for leg strength measurement.

Isometric hand grip strengths were tested by using a digital grip dynamometer (D-type No.1857 Takei kiki & Company, Tokyo Japan). Each subject was performed the test while standing relaxed with hand extended, forming a 35 degree angle with the forearm and then squeezed vigorously, exerting the maximum force of which subject is capable.

Both tests were repeated three times and the best value was recorded. Before measuring, every strength dynamometers were taken calibration procedures. All subjects were asking to perform with maximum efforts. Verbal command was encouraged throughout the experiment to ensure each subject's maximal effort.

Flexibility assessment

Flexibility instrument was designed to test the stretching ability of the hamstrings and lower back muscles. The indirect methods were performed in each subject by using the most popular techniques being the sitting toe-touch tests.

Sit and reach box: In order to consider hip and trunk flexibility, trunk forward bending was use. The subject was suggested to keep both legs in a fully extended posture and place both feet, in the totally contact fashion, against sit and reach box. Like a scoop position, subject sit on the floor and gradually bend forward as far as possible and both hands slide along the measuring scale. The final position is hold for 2 seconds and the distance in centimeters is note. The experiment repeats three times and the highest value is record.

Cardiovascular and respiratory fitness

Maximal O_2 uptake (Vo_2max) is a valid mean of assessing a person's cardiorespiratory fitness. Various form of exercise could be used for the test such as treadmill running, cycling, or bench stepping. In any case, the exercise load should be increased stepwise until exhaustion. During the exercise test, Vo_2 is increased with increasing workload until it reaches a peak or plateau despite further increases in the work rate. Vo_2max is also referred to as maximal aerobic power.

Treadmills (Quinton, USA) were used to test Vo_2max in this study. After a medical exam, start the test by walk on treadmill at low speed. Then, depending on the Bruce's treadmill protocol, belt speed and grade will increase at regular intervals of Bruce's treadmill protocol. While running, subjects will be breathing through a 2-way valve system. Air will come in from the room, but will be expired through sensors that measure both volume and oxygen concentration. Oxygen uptake will be calculated by a computer at each stage. With each increase in speed or incline, more muscle mass will be employed at a greater intensity. Oxygen consumption will increase linearly with increasing workload. However, at some point, an increase in intensity will not result in an "appropriate" increase in oxygen consumption. Ideally, the oxygen consumption will completely flatten out despite ever-increasing workload. This is the true indication of achieving VO_2 max.

3. Cutaneous microvascular reactivity assessment (Appendix H)

Microvascular reactivity analysis

Cutaneous blood flow study was performed on all participants on the right wrist with a laser Doppler flowmetry (DRT4 MoorLAB, Moor Instrument, UK), using the post-occlusive reactive hyperemic method. All participants rested in the supine position for 20 minutes. Baseline data was monitored for 1 min and then placed the cuff around the right upper arm, inflated rapidly to 200 mmHg for 5 minutes and deflated for 5 minutes of recovery (Betik et al. 2004). Blood flow data at baseline and after deflated cuff at maximal blood flow were collected.

Dual-channel DRT4 laser Doppler fluximetry (Moor Instruments, UK) used in this study is a noninvasive device that permits real-time continuous measurement of microvascular perfusion. In the present study, this instrument was used together with DPIT-V2 skin laser probe (Moor Instruments), which was stably held by PHI-V2 probe holder (Moor Instruments). Laser Doppler fluximetry generates a low-intensity beam of monochromatic coherent 780-nm light through an inbuilt infrared semiconductor laser diode. This light was delivered to the site of measurement by a flexible fiber optic probe. The laser light usually penetrates to a depth of 1-2 mm of skin (Anderson and Parrish, 1981), and the measurement is therefore dominantly a reflection of perfusion in arterioles, capillaries, postcapillary venules and venules of the superficial dermal plexus (Ryan, 1992).

Laser Doppler fluximetry uses laser Doppler shift principle to measure perfusion of blood cells, mainly erythrocytes, in the skin. Photons of laser light scattered in moving blood cells produce a Doppler shift on the reflect light. This

reflected light is detected by a photo-detector, and the signal is processed to determine the amount of the frequency shift. Theoretically, the blood perfusion measured by laser Doppler fluximetry is determined by the product of blood flow velocity and the number of moving red cell corpuscles with the surface microvessels of skin. The blood perfusion recorded is generally termed as "flux" and is expressed in perfusion arbitrary units (PU). Besides measuring skin perfusion, the DPIT-V2 probe used also monitors skin temperature of the measurement site.

Prior to each study session, subjects fasted overnight and avoided from consuming high salt food for at least 12 hours. They were also told to refrain from any heavy activities or vigorous exercises for 12 hours to avoid short-term exercise effects on microvascular reactivity (Bircher, De-Borer, Agner, Wahlberg, and Serup, 1994; Smolander, Kolari, Korhonen, and Ilmarinen, 1987). Alcohol and caffeine containing beverages were withheld for at least 48 hours. Subjects have familiarization visits with the instruments and study environment before the actual study.

All subjects are studied in room controlled for both temperature and humidity (room temperature is 24°C with 70% relative humidity). The laser Doppler probe is fixing to the skin surface of the third digit of the left hand and of the first digit of the left foot. SBF is measure under basal conditions, after local heating at 44°C and during reactive hyperemia following a 3-min brachial artery occlusion, at the beginning of the study, 6 week and at the end of 12 week. The output signal is linearly related to red cell flow as predicted by theoretical and experimental models.

The laser Doppler apparatus is connected to a computer which install program allows the storage and analysis of the recordings by used Moor Soft for Windows/DRT4 software package version 1.2 (Moor Instruments). Changes in skin blood perfusion flux are expressed in perfusion arbitrary perfusion units (PU) Coefficient of variation (three measurements) for baseline SBF is less than 5%. The following laser-Doppler-derived parameters of post-occlusive reactive hyperemia measurement are studied (Figure 4):

1. PORHmax, the maximum increase in hyperemia perfusion (peak flow above minimum rest flow), expressed in AU as the difference between maximal perfusion flux during post-occlusive reactive hyperemia and minimal baseline perfusion flux;

2. Tp, time-to-peak, time after occlusion cuff decompression until the post-occlusion peak perfusion flux is reached;

3. PORHpeak, amplitude of peak perfusion flux during hyperemia;

4. PORHmax/Tp, mean velocity of the postocclusive hyperemia increase, expressed as a ratio between PORHmax and Tp;

5. PORH%, the percentage of hyperemic response, is calculated as follows:

$$\frac{PORH_{peak} - \text{Mean baseline perfusion flux}}{\text{Mean baseline perfusion flux}} \times 100\%$$

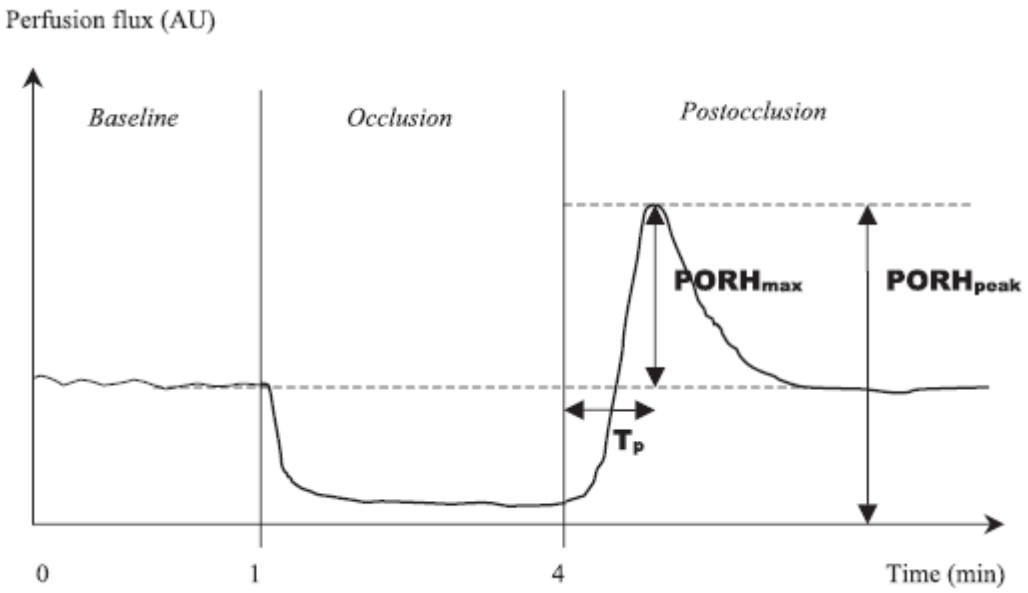


Figure 3 Schematic illustration of some studied reactive hyperemia parameters. Abbreviations: $PORH_{max}$, maximum increase in hyperemia perfusion flux; T_p, time-to-peak; $PORH_{peak}$, amplitude of peak perfusion flux; PU, arbitrary perfusion units.

Blood sample analysis

All participants, who fasted for at least 12 h before the start of the study, are requested to adhere to a low nitrate diet (e.g. exclusion of foodstuffs containing a high concentration of nitrates, such as cured meat, fruits and, in particular, leafy green vegetables) for 72 h before collection of blood samples. Blood samples are taken before and at the end of interventions. Plasma samples are frozen at -85°C until analyzed.

Blood samples were collected and centrifuged at 3500 rpm for 10 min at 4 °C for separation of erythrocyte and plasma. Malondialdehyde (MDA) was measured in erythrocyte with thiobarbituric acid reactive substances (TBAR) method (Appendix I). For fasting blood glucose (FBG), glycosylated hemoglobin (HbA1c), lipid profiles, insulin and C-reactive protein (CRP) studies were measured with standard procedures (enzymatic reference method with hexokinase, immunoturbidity, enzymatic colorimetric test, enhanced chemiluminescence and linked immuno assay method respectively) at NHS Lab, Bangkok, Thailand. Homeostasis model assessment (HOMA), the parameter for insulin resistance, was calculated by using equation of [FBG (mg/dL) × Insulin level (uU/ml) / 405] (Turner et al., 1993).

Data analysis

The data including biological data, health-related physical fitness data, biochemistry data, cutaneous blood flow data were expressed as means ± standard error of mean (SEM). Statistical comparisons between baseline and after 12 weeks were conducted using the paired student's t-test. Two-way analysis of variance (group x time), followed by tukey's-b multiple comparison was used to determine the significant differences among groups. $P<0.05$ was considered to be a significant difference.

CHAPTER IV
RESULTS

1. General physical characteristic

Fifty-three subjects with diagnosis of type 2 diabetes mellitus whose age ranges are 60-75 years old voluntarily participated in this study. After promoting in the senile club of local community, more twenty participants were included because they interested in this program and could be participating in the twelve-week period. All subjects had no known serious health problems and participated until the end study session. They had undergone a detailed medical examination focusing on inclusion criteria. All volunteers in this experiment had history of type 2 diabetes at least 3 years, but not more than 10 years before this test. In addition, they were trained under the similar intervention protocol.

The program of training began with warm-up, exercise training and cool down. The warm-up exercise consisted of 10 minute isometric contractions in combination with stretching the upper and lower extremities. Training session consisted of a standardized set of aerobic exercises program 30 minute and cool down period of 10 minute. This training was regularly performed in the afternoon (13.30 – 14.30 PM.). All of them trained approximately 50 minute per day and 3 days per week within therapeutic pool temperature of ~34 to 36°C continuously for 12-wk. The subjects in the water-based and land-based exercise training program performed at 70% of maximum heart rate (MHR). They were monitored by a HR monitoring device or rating of perceived exertion scale (RPE) of 10-16 in accordance with standard recommendations by ACSM for diabetic patients [65]. The subjects understood that at any time they experienced uncomfortable feeling they could stop exercising.

General physical characteristics at baseline among four groups of subjects with type 2 diabetes mellitus: water-based exercise training subjects (Wex), water immersion subjects (Wco), land-based exercise training subjects (Lex), non-exercise control subjects (Lco) are shown in Table 4. Average body weights (BW) and the body mass index (BMI) were in the obesity range. Their mean age, heights, body weights, body mass index, resting heart rate (HR rest), and systolic blood pressure (SBP) were similar between and among groups at baseline. Whereas, diastolic blood pressure (DBP) was observed significantly difference among four groups. There were also no significant difference in fasting blood glucose (FBG) and glycosylated hemoglobin (HbA1c) in all four groups at pre-training.

Table 4 General physical characteristics data among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects at baseline (n = 53).

Variables	Baseline				P-value
	Wex (n = 13)	Wco (n = 15)	Lex (n = 10)	Lco (n = 15)	
Age (years)	66.3 ± 0.7	65.7 ± 1.1	64.4 ± 0.9	63.6 ± 0.9	0.18
Height (cm)	156.0 ± 1.9	154.5 ± 1.9	154.1 ± 2.7	154.8 ± 1.6	0.92
BW (kg)	64.7 ± 2.4	60.3 ± 2.6	59.1 ± 3.5	65.7 ± 2.1	0.21
BMI ($kg \cdot m^{-2}$)	26.1 ± 0.9	25.2 ± 0.8	25.2 ± 0.9	27.4 ± 0.7	0.18
HR rest (bpm)	82.6 ± 3.1	77.9 ± 2.2	74.6 ± 1.9	77.8 ± 3.0	0.27
SBP (mm Hg)	116.5 ± 3.7	111.7 ± 2.2	117.9 ± 5.3	116.7 ± 2.3	0.99
DBP (mm Hg)	72.2 ± 3.1	69.5 ± 1.8†	72.8 ± 2.0	78.1 ± 1.8	0.04
FBG ($mg \cdot dL^{-1}$)	157.7 ± 6.5	151.4 ± 8.3	151.1 ± 12.1	145.5 ± 5.9	0.74
HbA1c (%)	8.0 ± 0.4	7.6 ± 0.1	7.7 ± 0.1	7.6 ± 0.1	0.44

Values are mean ± SEM.

n; number of subjects, BW; body weight, BMI; body mass index, HR rest; heart rate at rest, SBP; systolic blood pressure, DBP; diastolic blood pressure, FBG; fasting blood glucose, HbA1c; glycosylated hemoglobin. †represents statistical difference among four group, P<0.05.

2. Effect of exercise training on hemodynamic changes

Hemodynamic data in the four group subjects of type 2 diabetic before and after 12 weeks are shown in Table 5 to 9. After 12 weeks of exercise training, resting heart rate (Figure 4) and systolic blood pressure (Figure 5) were significantly reduction as compared with pre-exercise training (P<0.05) only observed in water-based exercise group. However, there was no significant difference in these variables in the other groups. Nevertheless, heart rate at rest represented statistical difference among four groups (P<0.05). In addition, diastolic blood pressure was significantly decreased (P<0.05) only in land-based exercise group (Figure 6).

Table 5 Effects of exercise training on hemodynamic data among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects at baseline.

Variables	Baseline				P-value
	Wex (n = 13)	Wco (n = 15)	Lex (n = 10)	Lco (n = 15)	
HR rest (bpm)	82.6 ± 3.1	77.9 ± 2.2	74.6 ± 1.9	77.8 ± 3.0	0.27
SBP (mm Hg)	116.5 ± 3.7	111.7 ± 2.2	117.9 ± 5.3	116.7 ± 2.3	0.99
DBP (mm Hg)	72.2 ± 3.1	69.5 ± 1.8†	72.8 ± 2.0	78.1 ± 1.8	0.04

Values are mean ± SEM.

n; number of subjects, HR rest; heart rate at rest, SBP; systolic blood pressure, DBP; diastolic blood pressure. †represents statistical difference among four groups, P<0.05.

Table 6 Effects of exercise training on hemodynamic data among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Variables	After 12 weeks				P-value
	Wex (n = 13)	Wco (n = 15)	Lex (n = 10)	Lco (n = 15)	
HR rest (bpm)	74.2 ± 2.0	81.5 ± 1.6	72.4 ± 1.7†	79.6 ± 2.0	0.004
SBP (mm Hg)	114.2 ± 3.5	117.7 ± 2.2	112.7 ± 3.2	116.0 ± 2.7	0.71
DBP (mm Hg)	71.0 ± 2.2	73.4 ± 1.7	65.8 ± 1.8†	79.0 ± 1.5	0.000

Values are mean ± SEM.

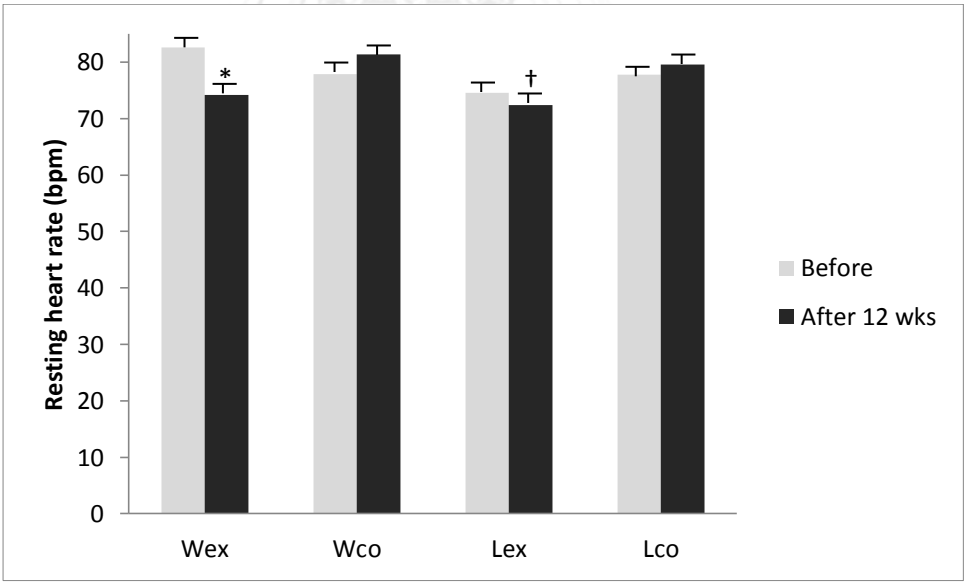
n; number of subjects, HR rest; heart rate at rest, SBP; systolic blood pressure, DBP; diastolic blood pressure. †represents statistical difference among four groups, P<0.05.

Table 7 The comparison of resting heart rate (bpm) between pre- and post- training of four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Resting heart rate (bpm)		P-value
	Pre-test	Post-test	
Wex (n = 13)	82.6 + 3.1	74.2 + 2.0*	0.03
Wco (n = 15)	77.9 + 2.2	81.5 + 1.6	0.20
Lex (n = 10)	74.6 + 1.9	72.4 + 1.7	0.40
Lco (n = 15)	77.8 + 3.0	79.6 + 2.0	0.62

Values are mean ± SEM.

*represents statistical difference from pre-test, P<0.05.



*represents significant difference from baseline, P<0.05. †represents statistical difference among four groups, P<0.05.

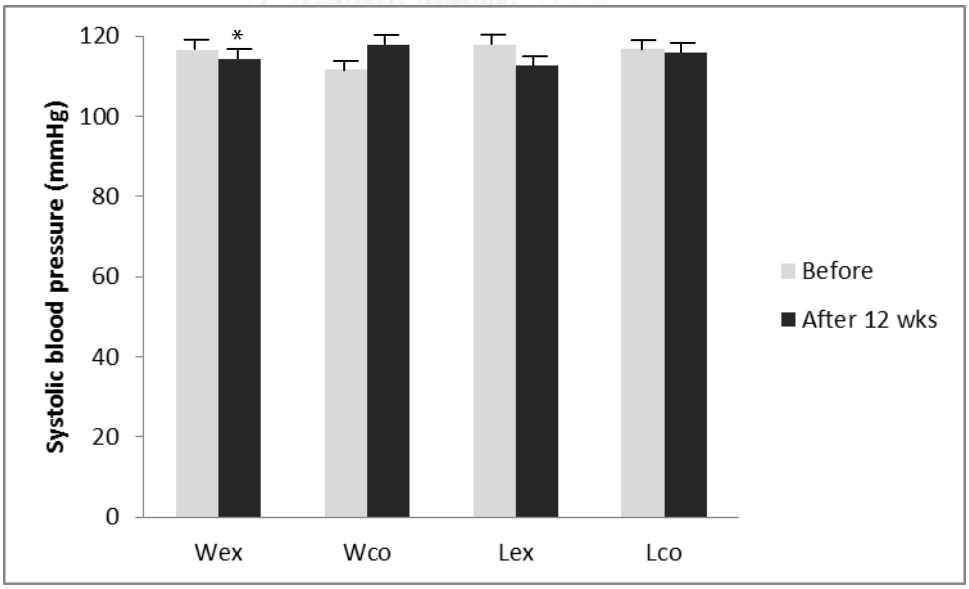
Figure 4 The comparison of resting heart rate (bpm) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 8 The comparison of systolic blood pressure (mmHg) between pre- and post-training of four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Systolic blood pressure (mmHg)		P-value
	Pre-test	Post-test	
Wex (n = 13)	116.5 + 3.7	114.2 + 3.5*	0.04
Wco (n = 15)	111.7 + 2.2	117.7 + 2.2	0.11
Lex (n = 10)	117.9 + 5.3	112.7 + 3.2	0.41
Lco (n = 15)	116.7 + 2.3	116.0 + 2.7	0.85

Values are mean ± SEM.

*represents significant difference from baseline, P<0.05



*represents significant difference from baseline, P<0.05

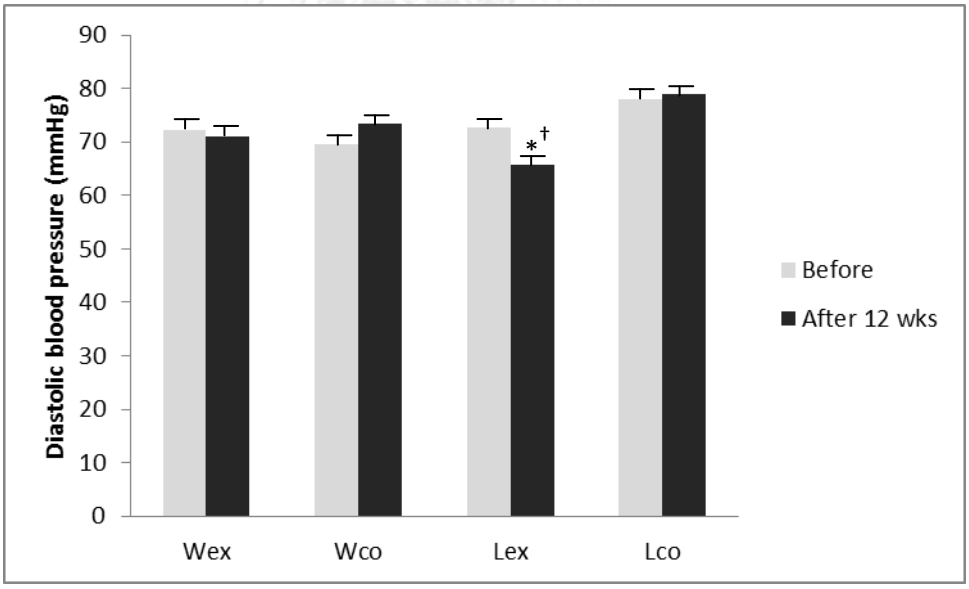
Figure 5 The comparison of systolic blood pressure (mmHg) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 9 The comparison of diastolic blood pressure (mmHg) between pre- and post-training of four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Diastolic blood pressure (mmHg)		P-value
	Pre-test	Post-test	
Wex (n = 13)	72.2 ± 3.1	71.0 ± 2.2	0.74
Wco (n = 15)	69.5 ± 1.8	73.4 ± 1.7	0.12
Lex (n = 10)	72.8 ± 2.0	65.8 ± 1.8*	0.01
Lco (n = 15)	78.1 ± 1.8	79.0 ± 1.5	0.68

Values are mean ± SEM.

*represents statistical difference from pre-test, P<0.05



*represents significant difference from baseline, P<0.05. †represents statistical difference among four groups, P<0.05.

Figure 6 The comparison of diastolic blood pressure (mmHg) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

3. Effect of exercise training on body composition

The body compositions data before and after 12 weeks of water-based and among four groups of subjects with type 2 diabetes mellitus; Water-based exercise training subjects (Wex), Water immersion subjects (Wco), Land-based exercise training subjects (Lex), non-exercise control subjects (Lco) are presented in Table 10 to 13. There was no significant difference in body weight (BW), percentage body fat (BF), body mass index (BMI), percentage of total body water (TBW) and waist hip ratio (WHR) in the four groups (Figure 8).

Table 10 Effects of exercise training on body composition data among four groups of subjects with T2DM at baseline.

	Baseline				
Variables	Wex (n = 13)	Wco (n = 15)	Lex (n = 10)	Lco (n = 15)	P-value
BW (kg)	64.7 ± 2.4	60.3 ± 2.6	59.1 ± 3.5	65.7 ± 2.1	0.21
BF (%)	38.8 ± 1.6	36.2 ± 1.3	39.9 ± 1.0	39.6 ± 0.8	0.12
TBW (%)	48.0 ± 1.9	49.5 ± 1.5	45.7 ± 0.9	45.3 ± 0.8	0.09
BMI ($kg{\cdot}m^{-2}$)	26.1 ± 0.9	25.2 ± 0.8	25.2 ± 0.9	27.4 ± 0.7	0.18
WHR	0.87 ± 0.02	0.85 ± 0.01	0.86 ± 0.02	0.89 ± 0.02	0.49

Values are mean ± SEM.

Table 11 Effects of exercise training on body composition data among four groups of subjects with type 2 diabetes mellitus after 12 weeks.

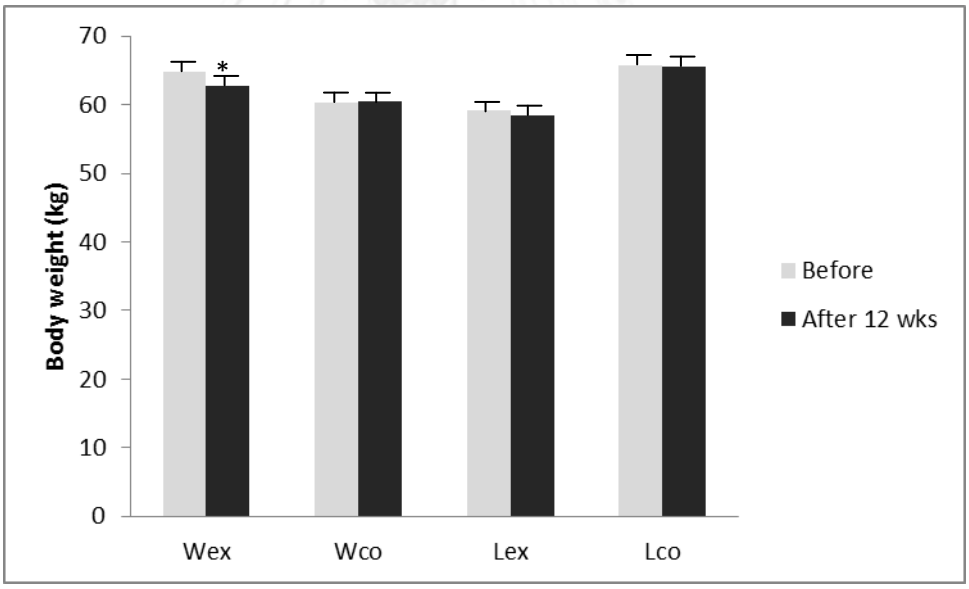
	After 12 weeks				
Variables	Wex (n = 13)	Wco (n = 15)	Lex (n = 10)	Lco (n = 15)	P-value
BW (kg)	62.7 ± 2.3	60.5 ± 2.6	58.4 ± 3.4	65.6 ± 2.0	0.25
BF (%)	37.9 ± 1.6	36.9 ± 1.4	37.7 ± 1.0	40.2 ± 1.5	0.46
TBW (%)	47.6 ± 2.0	48.9 ± 1.1	47.3 ± 1.2	45.2 ± 1.6	0.34
BMI ($kg{\cdot}m^{-2}$)	26.2 ± 1.1	25.3 ± 0.8	24.6 ± 1.0	27.4 ± 0.7	0.16
WHR	0.88 ± 0.02	0.86 ± 0.01	0.87 ± 0.02	0.89 ± 0.02	0.62

Values are mean ± SEM.

Table 12 The comparison of body weight (kg) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Body weight (kg)		P-value
	Pre-test	Post-test	
Wex (n = 13)	64.7 ± 2.4	62.7 ± 2.3*	0.045
Wco (n = 15)	60.3 ± 2.6	60.5 ± 2.6	0.94
Lex (n = 10)	59.1 ± 3.5	58.4 ± 3.4	0.89
Lco (n = 15)	65.7 ± 2.1	65.6 ± 2.0	0.98

Values are mean ± SEM.



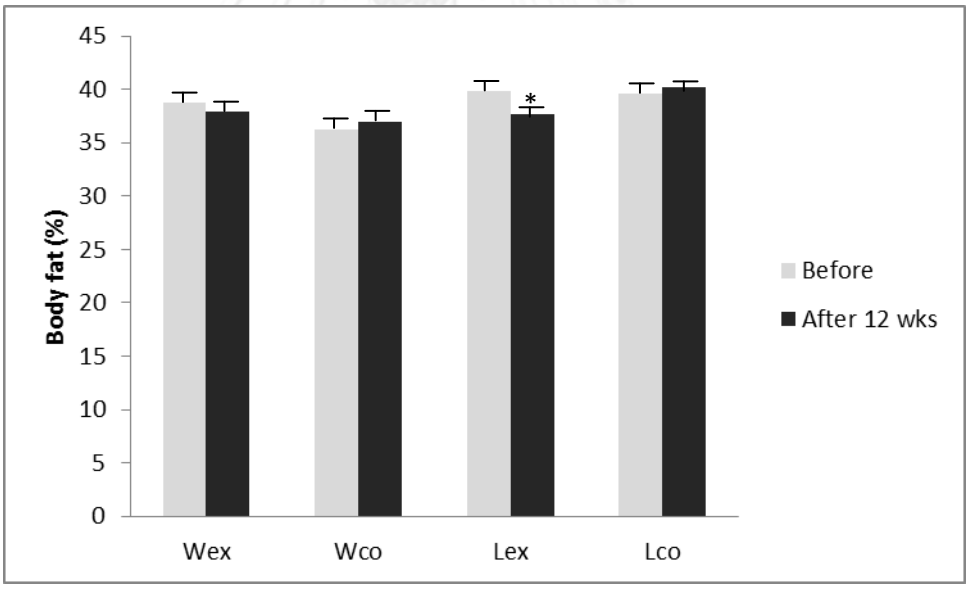
*represents significant difference from baseline, P<0.05

Figure 7 The comparison of body weight (kg) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 13 The comparison of body fat (%) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Body fat (%)		P-value
	Pre-test	Post-test	
Wex (n = 13)	38.8 ± 1.6	37.9 ± 1.6	0.71
Wco (n = 15)	36.9 ± 1.4	36.9 ± 1.4	0.69
Lex (n = 10)	39.9 ± 1.0	37.7 ± 1.0*	0.03
Lco (n = 15)	40.2 ± 1.5	40.2 ± 1.5	0.81

Values are mean ± SEM.



*represents significant difference from baseline, P<0.05

Figure 8 The comparison of body fat (%) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 14 The comparison of percentage of total body water (%) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Total body water (%)		P-value
	Pre-test	Post-test	
Wex (n = 13)	48.0 ± 1.9	47.6 ± 2.0	0.88
Wco (n = 15)	49.5 ± 1.5	48.9 ± 1.1	0.73
Lex (n = 10)	45.7 ± 0.9	47.3 ± 1.2	0.27
Lco (n = 15)	45.3 ± 0.8	45.2 ± 1.6	0.97

Values are mean ± SEM.

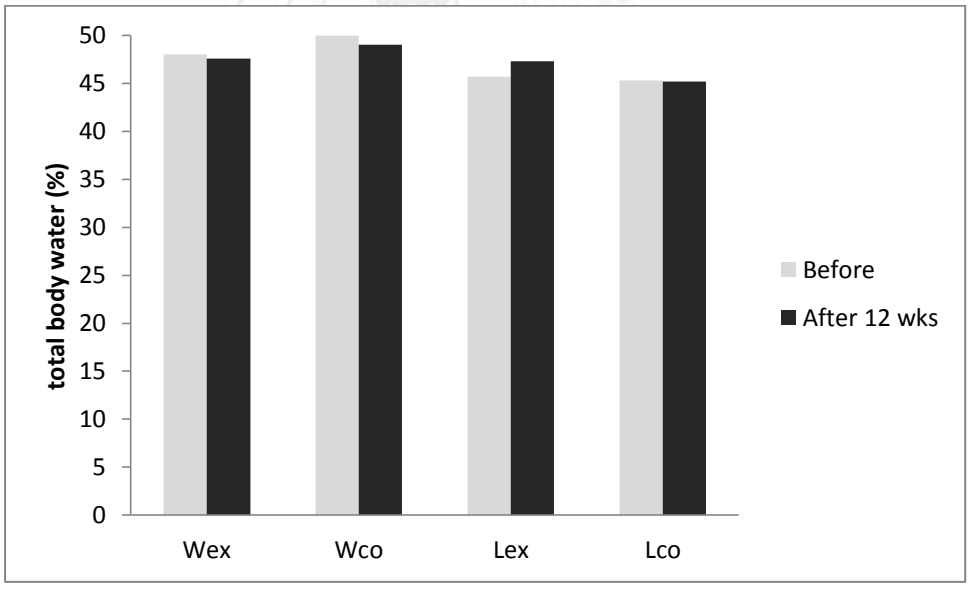


Figure 9 The comparison of total body water (%) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 15 The comparison of body mass index ($kg \cdot m^{-2}$) between pre- and post-training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Body mass index ($kg \cdot m^{-2}$)		P-value
	Pre-test	Post-test	
Wex (n = 13)	26.1 ± 0.9	26.2 ± 1.1	0.90
Wco (n = 15)	25.2 ± 0.8	25.3 ± 0.8	0.91
Lex (n = 10)	25.2 ± 0.9	24.6 ± 1.0	0.63
Lco (n = 15)	27.4 ± 0.7	27.4 ± 0.7	0.99

Values are mean ± SEM.

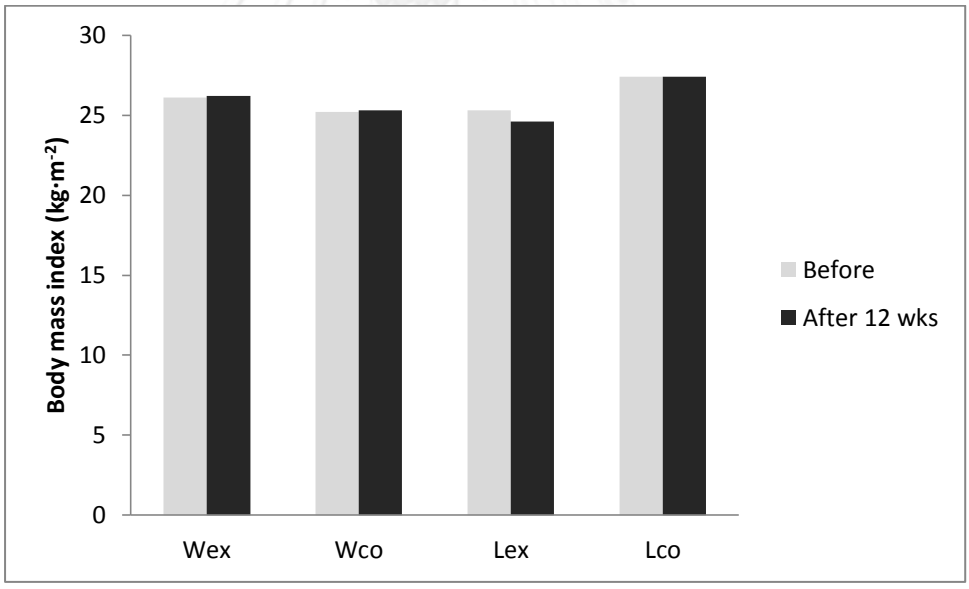


Figure 10 The comparison of body mass index ($kg \cdot m^{-2}$) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 16 The comparison of waist hip ratio between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Waist hip ratio		P-value
	Pre-test	Post-test	
Wex (n = 13)	0.87 ± 0.02	0.88 ± 0.02	0.78
Wco (n = 15)	0.85 ± 0.01	0.86 ± 0.01	0.65
Lex (n = 10)	0.86 ± 0.02	0.87 ± 0.02	0.71
Lco (n = 15)	0.89 ± 0.02	0.89 ± 0.02	0.88

Values are mean ± SEM.

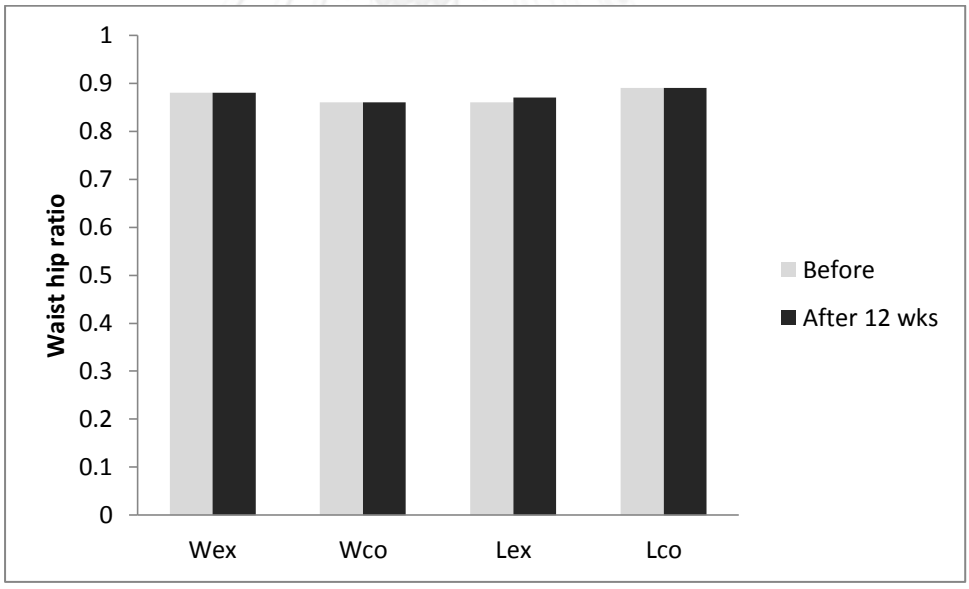


Figure 11 The comparison of waist hip ratio between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

4. Effect of exercise training on physical fitness characteristic

Physical fitness parameters data at baseline and after 12 weeks training of type 2 diabetic subjects in the four groups are shown in Table 17 to 19. Handgrip strength was significantly increased ($P<0.05$) in both exercise training group (Figure 9). In addition, changes of maximal oxygen consumption (VO_2max) was significantly higher than baseline ($P<0.05$) in only the water-based exercise training group (Figure 10). There were no significant differences in leg strength and flexibility for four groups.

Table 17 Effects of exercise training on physical fitness data among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects at baseline.

Variables	Baseline				P-value
	Wex (n = 13)	Wco (n = 15)	Lex (n = 10)	Lco (n = 15)	
GS ($kg \cdot BW^{-1}$)	0.32 ± 0.04	0.33 ± 0.02	0.40 ± 0.04	0.31 ± 0.02	0.20
LS ($kg \cdot BW^{-1}$)	0.79 ± 0.08	0.80 ± 0.11	0.90 ± 0.17	0.74 ± 0.07	0.70
Flexibility (cm)	10.2 ± 1.7	9.8 ± 1.2	9.2 ± 1.1	5.8 ± 2.7	0.32
VO_2max ($mL \cdot kg^{-1} \cdot min^{-1}$)	23.5 ± 0.2	22.8 ± 0.1	23.0 ± 0.4	22.2 ± 0.2†	0.001

Values are mean ± SEM.

n; number of subjects, GS; grip strength, LS; leg strength, VO_2max; maximal oxygen consumption.
†represents statistical difference among four groups, $P<0.05$.

Table 18 Effects of exercise training on physical fitness data among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Variables	After 12 weeks				P-value
	Wex (n = 13)	Wco (n = 15)	Lex (n = 10)	Lco (n = 15)	
GS ($kg \cdot BW^{-1}$)	0.40 ± 0.03	0.32 ± 0.02†	0.46 ± 0.04	0.32 ± 0.02	0.001
LS ($kg \cdot BW^{-1}$)	0.77 ± 0.08	0.80 ± 0.10	0.80 ± 0.13	0.78 ± 0.06	0.99
Flexibility (cm)	11.7 ± 2.6	9.9 ± 1.2	14.2 ± 2.7	5.5 ± 2.8	0.08
VO_2max ($mL \cdot kg^{-1} \cdot min^{-1}$)	23.8 ± 0.1	22.6 ± 0.1	23.6 ± 0.3	22.1 ± 0.1†	0.00

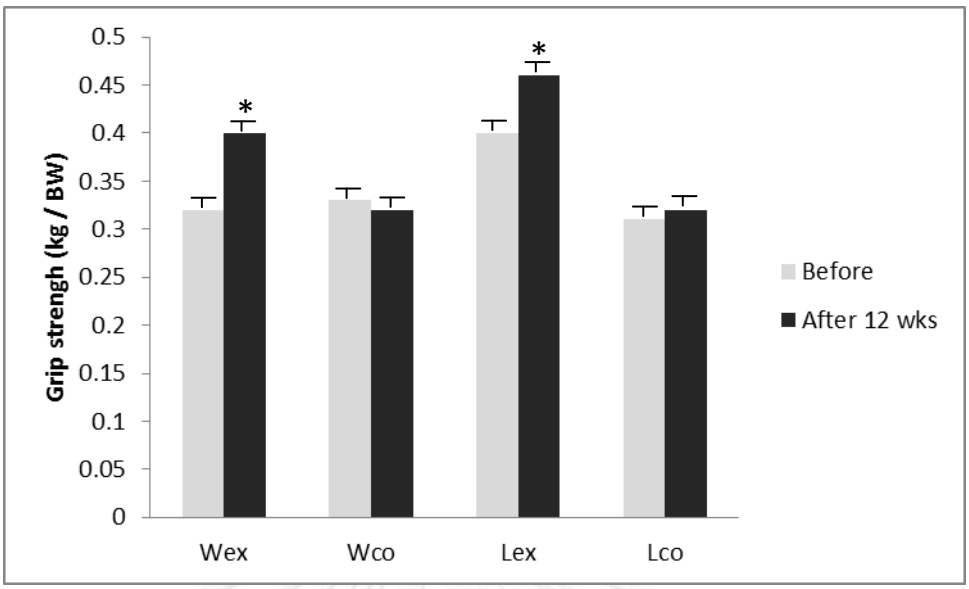
Values are mean ± SEM.

n; number of subjects, GS; grip strength, LS; leg strength, VO_2max; maximal oxygen consumption. †represents statistical difference among four groups, P<0.05.

Table 19 The comparison of grip strength ($kg \cdot BW^{-1}$) between pre- and post- training of four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Grip strength ($kg \cdot BW^{-1}$)		P-value
	Pre-test	Post-test	
Wex (n = 13)	0.32 ± 0.04	0.40 ± 0.03*	0.03
Wco (n = 15)	0.33 ± 0.02	0.32 ± 0.02	0.87
Lex (n = 10)	0.40 ± 0.04	0.46 ± 0.04*	0.03
Lco (n = 15)	0.31 ± 0.02	0.32 ± 0.02	0.79

Values are mean ± SEM.



*represents significant difference from baseline, P<0.05

Figure 12 The comparison of grip strength (kg·BW^{-1}) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 20 The comparison of leg strength (kg·BW^{-1}) between pre- and post- training of four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Leg strength (kg·BW^{-1})		P-value
	Pre-test	Post-test	
Wex (n = 13)	0.79 ± 0.08	0.77 ± 0.08	0.88
Wco (n = 15)	0.80 ± 0.11	0.80 ± 0.10	0.94
Lex (n = 10)	0.91 ± 0.17	0.80 ± 0.13	0.61
Lco (n = 15)	0.74 ± 0.07	0.78 ± 0.06	0.64

Values are mean ± SEM.

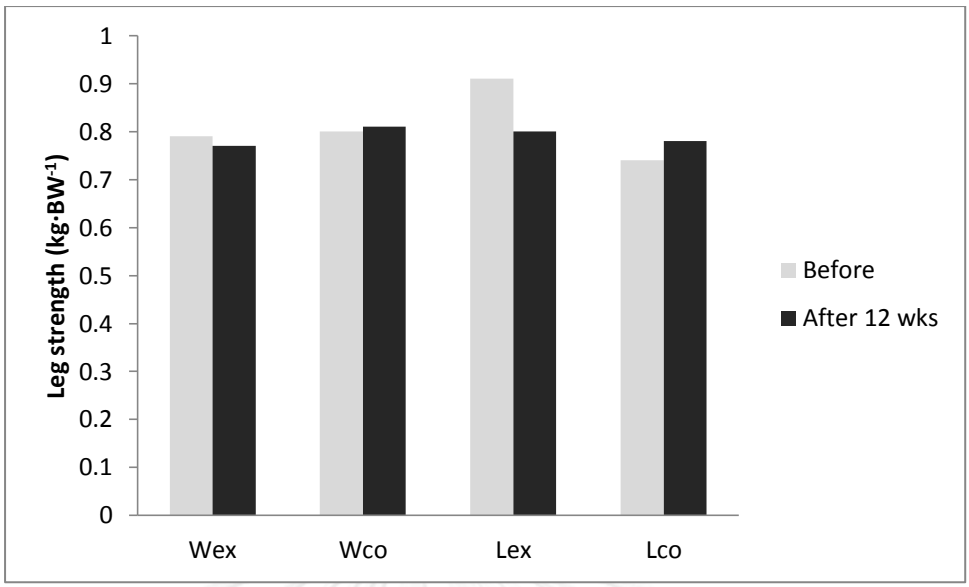


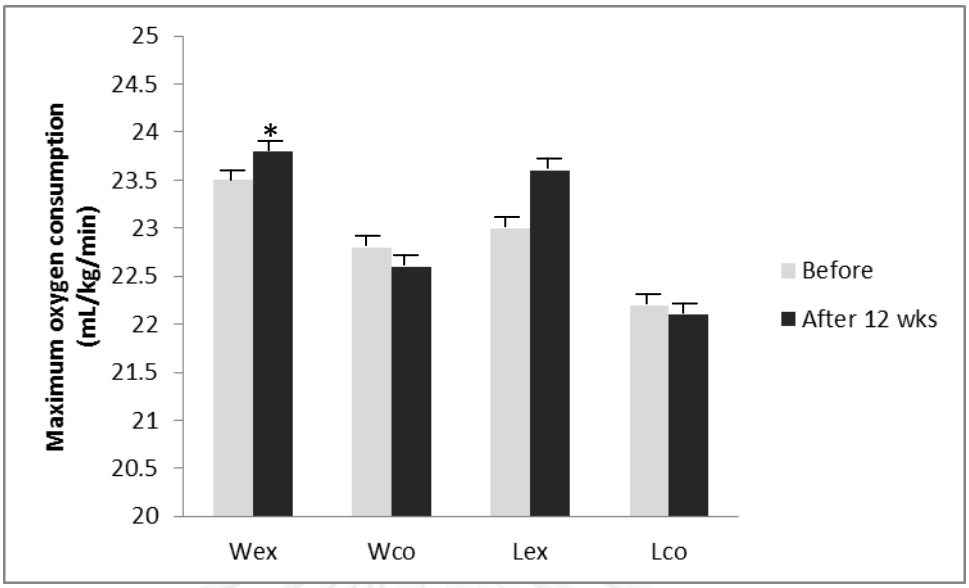
Figure 13 The comparison of grip strength ($kg \cdot BW^{-1}$) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 21 The comparison of maximal oxygen consumption ($mL \cdot kg^{-1} \cdot min^{-1}$) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Maximal oxygen consumption ($mL \cdot kg^{-1} \cdot min^{-1}$)		P-value
	Pre-test	Post-test	
Wex (n = 13)	23.5 ± 0.2	23.8 ± 0.1*	0.047
Wco (n = 15)	22.8 ± 0.1	22.6 ± 0.1	0.32
Lex (n = 10)	23.0 ± 0.4	23.6 ± 0.3	0.32
Lco (n = 15)	22.2 ± 0.2	22.1 ± 0.1	0.65

Values are mean ± SEM.

*represents statistical difference from pre-test, P<0.05.



*represents statistical difference from pre-test, P<0.05

Figure 14 The comparison of maximal oxygen consumption ($mL \cdot kg^{-1} \cdot BW^{-1}$) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

5. Effect of exercise training on blood chemistry

Blood chemical data before and after 12 weeks of four group subjects are shown in Table 18 to 12. There were significant differences from pre-test (P<0.05) in fasting blood glucose (FBG), glycosylated hemoglobin (HbA1c), total cholesterol, high density lipoprotein cholesterol (HDL-chol) and low density lipoprotein cholesterol (LDL-chol) in both water-based and land-based exercise groups. Similarly, fasting blood glucose was significantly decreased (P<0.05) in the group of water-based control (Figure 11). However, a significant decrease (P<0.05) in triglyceride (TG), insulin, HOMA-IR, C-reactive protein (CRP) and malondialdehyde (MDA) level was found only in water-based exercise group. In addition, the change of CRP in water-based exercise training group was significantly lower than those of the water-based control group, land-based exercise training group and land-based control group.

Table 22 Effects of exercise training on blood chemical parameters data among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects at baseline.

Variables	Baseline				P-value
	Wex (n = 13)	Wco (n = 15)	Lex (n = 10)	Lco (n = 15)	
FBG ($mg \cdot dL^{-1}$)	157.7 ± 6.5	151.4 ± 8.3	151.1 ± 12.1	145.5 ± 5.9	0.74
HbA1c (%)	8.0 ± 0.4	7.6 ± 0.1	7.7 ± 0.1	7.6 ± 0.1	0.11
Insulin ($\mu U \cdot mL^{-1}$)	23.2 ± 1.3	21.4 ± 1.4	23.8 ± 1.5	21.8 ± 1.2	0.58
HOMA-IR	3.32 ± 0.2	3.03 ± 0.2	3.37 ± 0.2	3.06 ± 0.2	0.61
Cholesterol ($mg \cdot dL^{-1}$)	230.2 ± 9.0	226.5 ± 13.1	213.6 ± 9.9†	226.4 ± 14.6	0.000
TG ($mg \cdot dL^{-1}$)	186.4 ± 27.2	181.6 ± 16.6	149.7 ± 11.6	147.7 ± 11.0	0.28
HDL-chol ($mg \cdot dL^{-1}$)	51.8 ± 3.6†	54.9 ± 3.3	60.4 ± 3.3	64.7 ± 2.6	0.02
LDL-chol ($mg \cdot dL^{-1}$)	137.1 ± 7.0	135.7 ± 10.9	136.2 ± 8.2	149.6 ± 9.1	0.64
CRP ($mg \cdot dL^{-1}$)	5.5 ± 0.2	5.4 ± 0.1	5.8 ± 0.2	6.0 ± 0.3	0.14
MDA ($mg \cdot dL^{-1}$)	1312.7 ± 52.5	1202.7 ± 42.9	1197.5 ± 65.6	1228.9 ± 76.7	0.54

Values are mean ± SEM.

†represents statistical difference among four groups of subjects, $P<0.05$.

Table 23 Effects of exercise training on blood chemical parameters data among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Variables	After 12 weeks				P-value
	Wex (n = 13)	Wco (n = 15)	Lex (n = 10)	Lco (n = 15)	
FBG ($mg \cdot dL^{-1}$)	133.8 ± 4.1	148.5 ± 7.7	145.3 ± 11.5	147.2 ± 6.4	0.47
HbA1c (%)	6.8 ± 0.2	7.6 ± 0.1	6.8 ± 0.1†	7.6 ± 0.1	0.00
Insulin ($\mu U \cdot mL^{-1}$)	21.9 ± 1.2	22.1 ± 1.5	22.6 ± 1.3	22.0 ± 1.1	0.98
HOMA-IR	3.03 ± 0.2	3.11 ± 0.2	3.16 ± 0.2	3.10 ± 0.1	0.97
Cholesterol ($mg \cdot dL^{-1}$)	220.2 ± 9.4	226.2 ± 12.9	206.3 ± 9.5	227.2 ± 14.6	0.68
TG ($mg \cdot dL^{-1}$)	173.5 ± 24.2	180.4 ± 16.5	145.5 ± 11.5	145.8 ± 10.6	0.32
HDL-chol ($mg \cdot dL^{-1}$)	58.2 ± 3.3	54.7 ± 3.2	62.9 ± 3.2	63.9 ± 3.0	0.13
LDL-chol ($mg \cdot dL^{-1}$)	128.5 ± 6.7	136.3 ± 9.7	125.7 ± 7.8	150.2 ± 8.9	0.19
CRP ($mg \cdot dL^{-1}$)	5.0 ± 0.1†	5.3 ± 0.1	5.7 ± 0.2	6.0 ± 0.3	0.003
MDA ($mg \cdot dL^{-1}$)	1161.4 ± 42.1	1224.6 ± 36.2	1152.7 ± 73.2	1234.1 ± 82.6	0.71

Values are mean ± SEM.

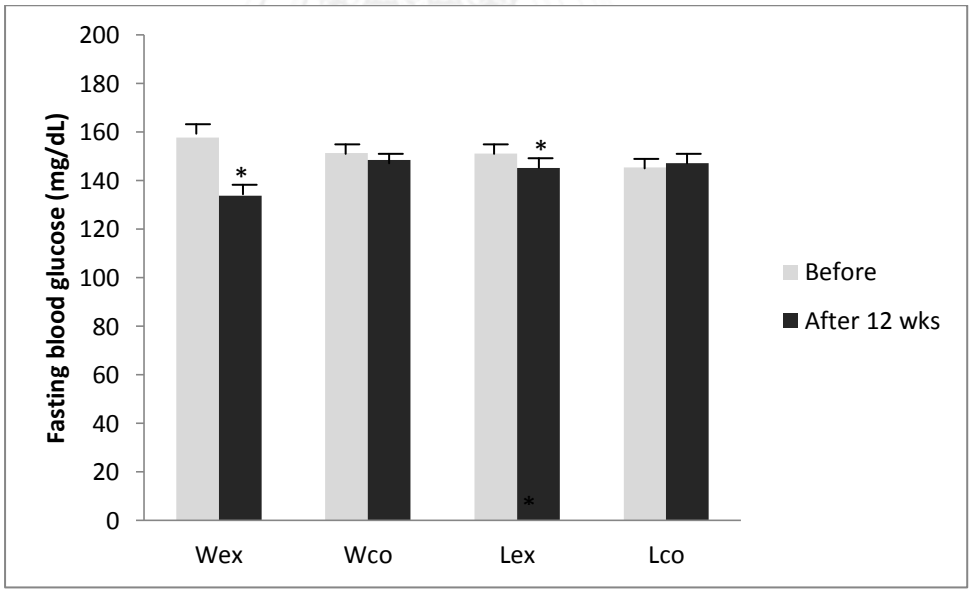
†represents statistical difference among four groups of subjects, $P<0.05$.

Table 24 The comparison of fasting blood glucose (mg·dL^{-1}) between pre- and post-training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Fasting blood glucose (mg·dL^{-1})		P-value
	Pre-test	Post-test	
Wex (n = 13)	157.7 ± 6.5	133.8 ± 4.1*	0.004
Wco (n = 15)	151.4 ± 8.3	148.5 ± 7.7	0.79
Lex (n = 10)	151.1 ± 12.1	145.3 ± 11.5*	0.03
Lco (n = 15)	145.5 ± 5.9	147.2 ± 6.4	0.84

Values are mean ± SEM.

*represents statistical difference from pre-test, P<0.05



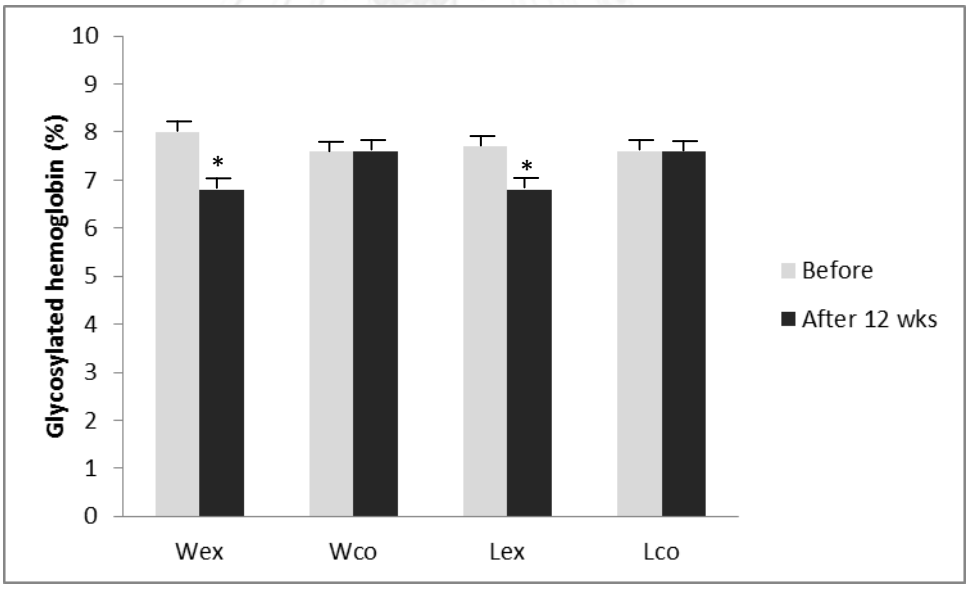
*represents significant difference from baseline, P<0.05.

Figure 15 The comparison of fasting blood glucose (mg·dL^{-1}) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 25 The comparison of glycosylated hemoglobin (%) between pre- and post-training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Glycosylated hemoglobin (%)		P-value
	Pre-test	Post-test	
Wex (n = 13)	8.0 ± 0.4	6.8 ± 0.2*	0.006
Wco (n = 15)	7.6 ± 0.1	7.6 ± 0.1	0.72
Lex (n = 10)	7.7 ± 0.1	6.8 ± 0.1*	0.0004
Lco (n = 15)	7.6 ± 0.1	7.6 ± 0.1	0.87

Values are mean ± SEM.



*represents significant difference from baseline, P<0.05.

Figure 16 The comparison of glycosylated hemoglobin (%) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 26 The comparison of insulin ($\mu U \cdot mL^{-1}$) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Insulin ($\mu U \cdot mL^{-1}$)		P-value
	Pre-test	Post-test	
Wex (n = 13)	23.2 ± 1.3	21.9 ± 1.2*	0.04
Wco (n = 15)	21.4 ± 1.4	22.1 ± 1.5	0.74
Lex (n = 10)	23.8 ± 1.5	22.6 ± 1.3	0.53
Lco (n = 15)	21.8 ± 1.2	22.0 ± 1.1	0.88

Values are mean ± SEM.

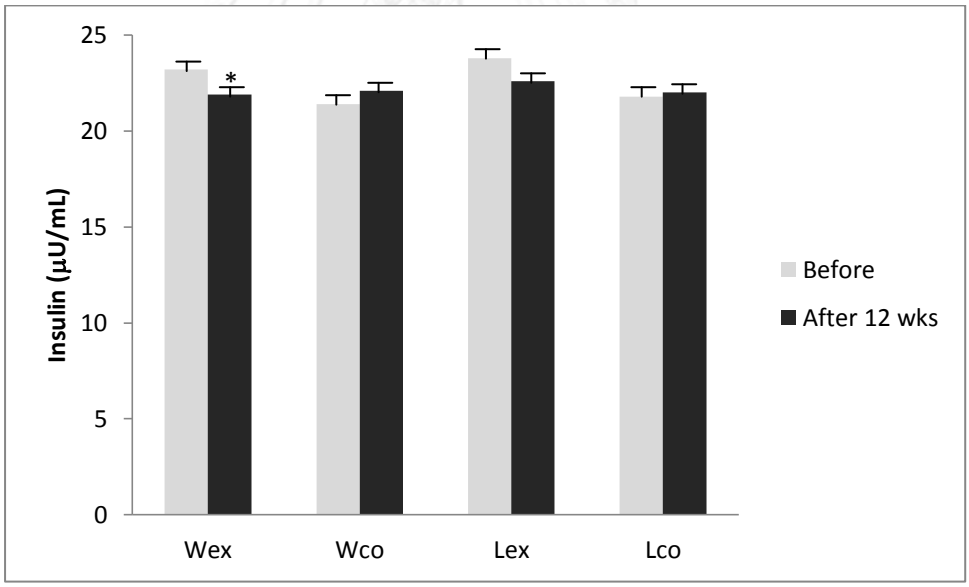
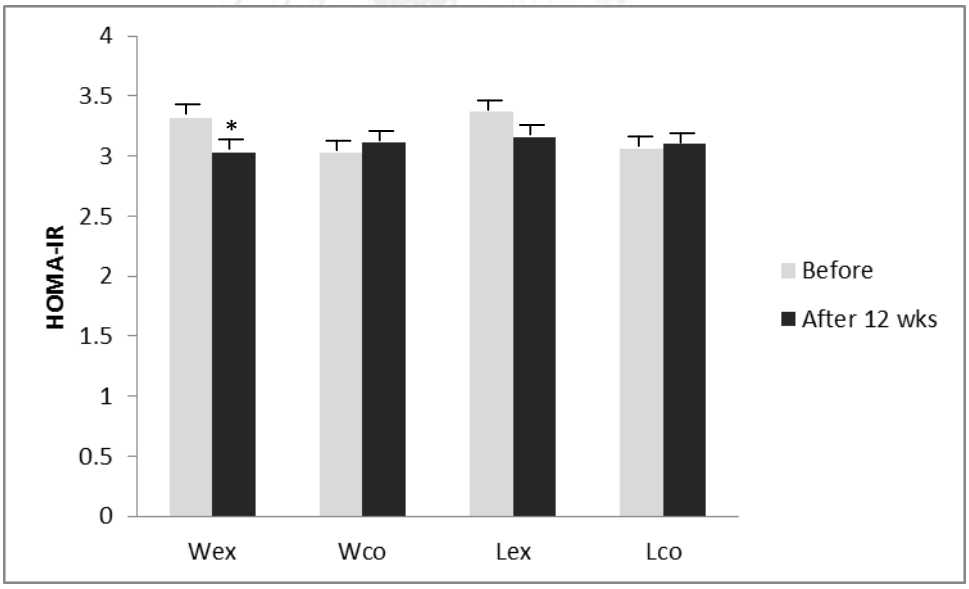


Figure 17 The comparison of insulin ($\mu U \cdot mL^{-1}$) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 27 The comparison of HOMA-IR between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	HOMA-IR		P-value
	Pre-test	Post-test	
Wex (n = 13)	3.32 ± 0.2	3.03 ± 0.2*	0.027
Wco (n = 15)	3.03 ± 0.2	3.11 ± 0.2	0.77
Lex (n = 10)	3.37 ± 0.2	3.16 ± 0.2	0.45
Lco (n = 15)	3.06 ± 0.2	3.10 ± 0.1	0.86

Values are mean ± SEM.



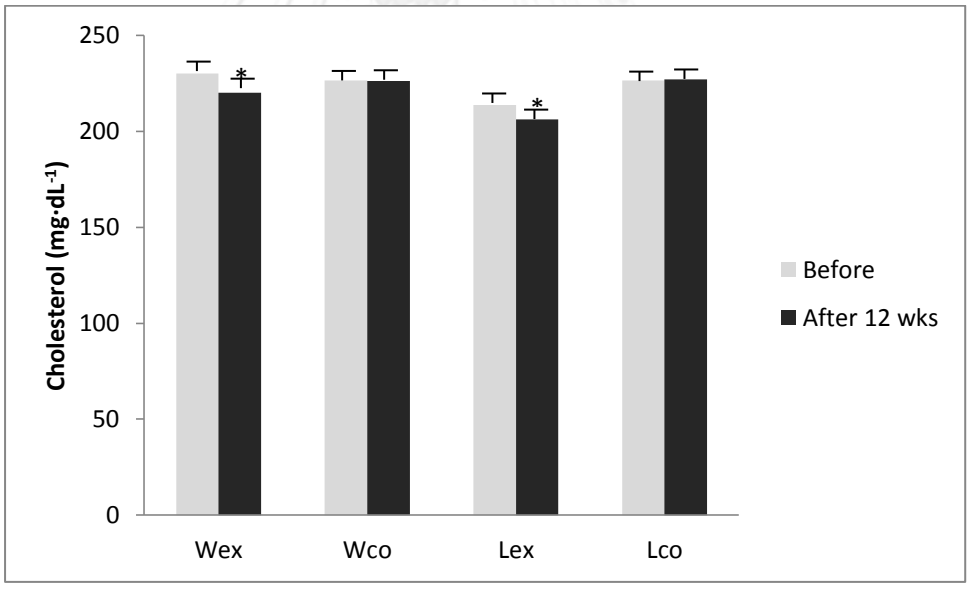
*represents significant difference from baseline, P<0.05.

Figure 18 The comparison of HOMA-IR between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 28 The comparison of cholesterol ($mg{\cdot}dL^{-1}$) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Cholesterol ($mg{\cdot}dL^{-1}$)		P-value
	Pre-test	Post-test	
Wex (n = 13)	230.2 ± 9.0	220.2 ± 9.4*	0.040
Wco (n = 15)	226.5 ± 13.1	226.2 ± 12.9	0.98
Lex (n = 10)	213.6 ± 9.9	206.3 ± 9.5*	0.047
Lco (n = 15)	226.4 ± 14.6	227.2 ± 14.6	0.96

Values are mean ± SEM.



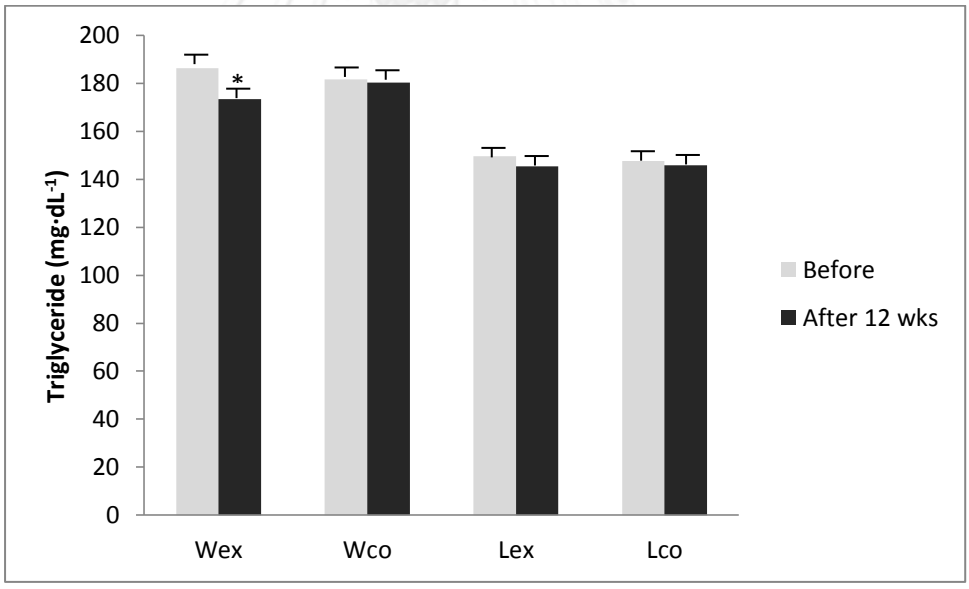
*represents significant difference from baseline, P<0.05.

Figure 19 The comparison of cholesterol ($mg{\cdot}dL^{-1}$) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 29 The comparison of triglyceride (mg·dL^{-1}) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Triglyceride (mg·dL^{-1})		P-value
	Pre-test	Post-test	
Wex (n = 13)	186.4 ± 27.2	173.5 ± 24.2*	0.03
Wco (n = 15)	181.6 ± 16.6	180.4 ± 16.5	0.95
Lex (n = 10)	149.7 ± 11.6	145.5 ± 11.5	0.79
Lco (n = 15)	147.7 ± 11.0	145.8 ± 10.6	0.96

Values are mean ± SEM.



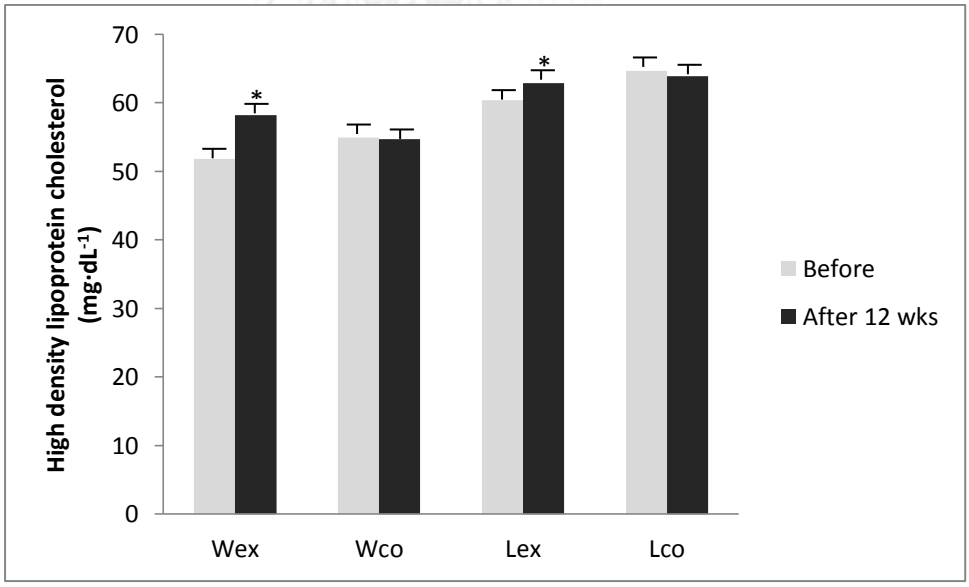
*represents significant difference from baseline, P<0.05.

Figure 20 The comparison of triglyceride (mg·dL^{-1}) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 30 The comparison of high density lipoprotein cholesterol (mg·dL^{-1}) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	High density lipoprotein cholesterol (mg·dL^{-1})		P-value
	Pre-test	Post-test	
Wex (n = 13)	51.8 ± 3.6	58.2 ± 3.3*	0.019
Wco (n = 15)	54.9 ± 3.3	54.7 ± 3.2	0.96
Lex (n = 10)	60.4 ± 3.3	62.9 ± 3.2*	0.039
Lco (n = 15)	64.7 ± 2.6	63.9 ± 3.0	0.84

Values are mean ± SEM.



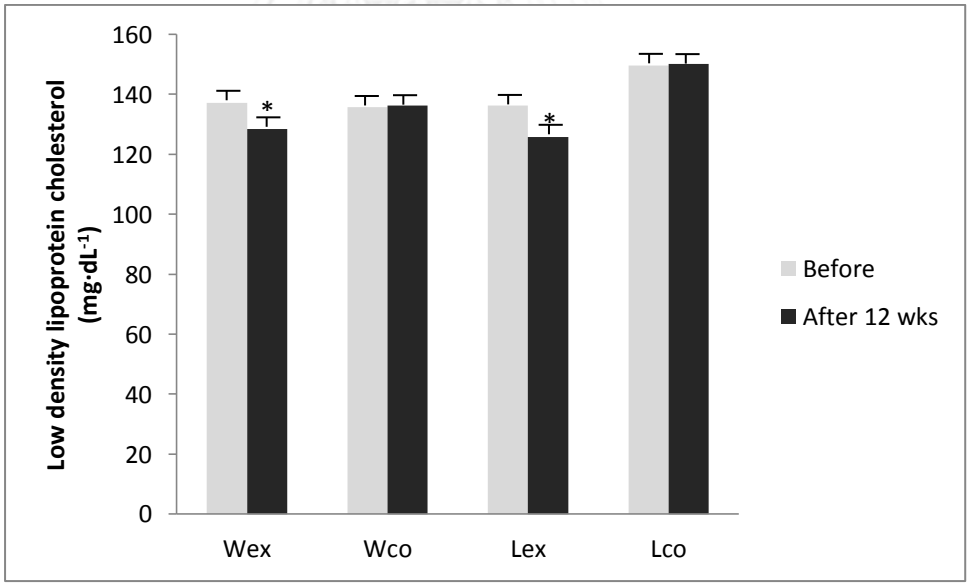
*represents significant difference from baseline, P<0.05.

Figure 21 The comparison of high density lipoprotein cholesterol (mg·dL^{-1}) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 31 The comparison of low density lipoprotein cholesterol (mg·dL^{-1}) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Low density lipoprotein cholesterol (mg·dL^{-1})		P-value
	Pre-test	Post-test	
Wex (n = 13)	137.1 ± 7.0	128.5 ± 6.7*	0.038
Wco (n = 15)	135.7 ± 10.9	136.3 ± 9.7	0.97
Lex (n = 10)	136.2 ± 8.2	125.7 ± 7.8*	0.036
Lco (n = 15)	149.6 ± 9.1	150.2 ± 8.9	0.96

Values are mean ± SEM.



*represents significant difference from baseline, P<0.05.

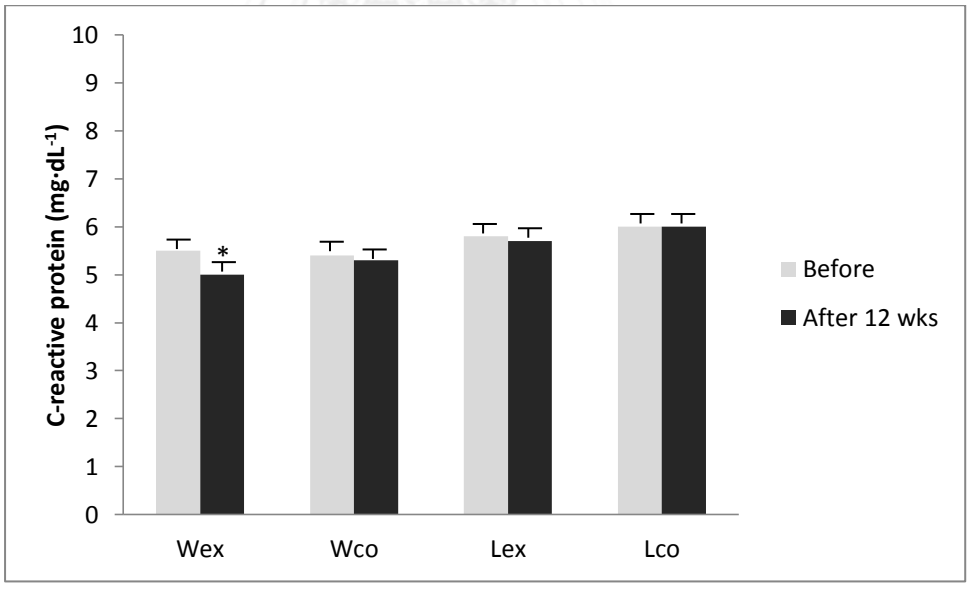
Figure 22 The comparison of low density lipoprotein cholesterol (mg·dL^{-1}) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 32 The comparison of C-reactive protein (mg·dL^{-1}) between pre- and post-training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	C-reactive protein (mg·dL^{-1})		P-value
	Pre-test	Post-test	
Wex (n = 13)	5.5 ± 0.2	5.0 ± 0.1*	0.01
Wco (n = 15)	5.4 ± 0.1	5.3 ± 0.1	0.61
Lex (n = 10)	5.8 ± 0.2	5.7 ± 0.2	0.68
Lco (n = 15)	6.0 ± 0.3	6.0 ± 0.3	0.97

Values are mean ± SEM.

*represents significant difference from baseline, P<0.05.



*represents significant difference from baseline, P<0.05.

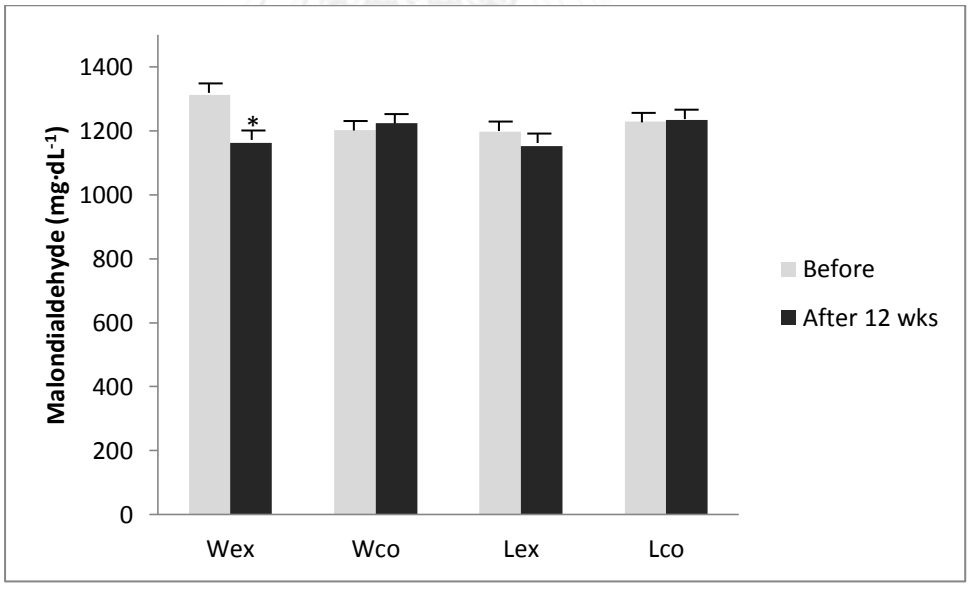
Figure 23 The comparison of C-reactive protein (mg·dL^{-1}) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 33 The comparison of malondialdehyde (mg·dL^{-1}) between pre- and post-training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Malondialdehyde (mg·dL^{-1})		P-value
	Pre-test	Post-test	
Wex (n = 13)	1312.7 ± 52.5	1161.4 ± 42.1*	0.03
Wco (n = 15)	1202.7 ± 42.9	1224.6 ± 36.2	0.69
Lex (n = 10)	1197.5 ± 65.6	1152.7 ± 73.2	0.65
Lco (n = 15)	1228.9 ± 76.7	1234.1 ± 82.6	0.96

Values are mean ± SEM.

*represents significant difference from baseline, P<0.05.



*represents significant difference from baseline, P<0.05.

Figure 24 The comparison of malondialdehyde (mg·dL^{-1}) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

6. Effect of exercise training on post-occlusive reactive hyperemia (PORH)

Post-occlusive reactive hyperemia (PORH) of type 2 diabetic subjects in water-based group and land-based group are shown in Table 9. The significantly higher (P<0.05) of peak perfusion (PORHpeak) were observed at the post-test training only in water-based exercise group. Time-to-peak (Tp) was also significantly increased from pre-test (P<0.05) in the both exercise groups. However, there was no significant difference in these variables in the water-based and land-based control group. Furthermore, recovery time was significantly lower (P<0.05) in the water-based and land-based exercise groups compared with before exercise training.

Table 34 Effects of exercise training on post-occlusive reactive hyperemia (PORH) among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects at baseline.

	Baseline				
Variables	Wex (n = 13)	Wco (n = 15)	Lex (n = 10)	Lco (n = 15)	P-value
Time to peak (s)	12.66 ± 0.5	12.44 ± 0.5	12.00 ± 0.7	11.83 ± 0.6	0.73
PORHpeak (PU)	71.85 ± 2.6	70.93 ± 2.7	65.60 ± 3.1	70.53 ± 3.2	0.53
Recovery time (s)	188.37±2.4	186.14±2.9	191.74±3.1	190.32 ± 2.4	0.50

Values are mean ± SEM.

n = number of subjects. PORHpeak; amplitude of peak perfusion flux, PU; arbitrary perfusion units.

Table 35 Effects of exercise training on post-occlusive reactive hyperemia (PORH) among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Variables	After 12 weeks				P-value
	Wex (n = 13)	Wco (n = 15)	Lex (n = 10)	Lco (n = 15)	
Time to peak (s)	13.80 ± 0.4	12.52 ± 0.7	13.78 ± 0.9	10.77 ± 0.8†	0.009
PORHpeak (PU)	79.69 ± 2.5	75.60 ± 2.4	73.30 ± 3.3	73.93 ± 2.7	0.36
Recovery time (s)	170.87±2.6†	179.71±2.2	177.36±4.4	190.75 ± 2.1	0.000

Values are mean ± SEM.

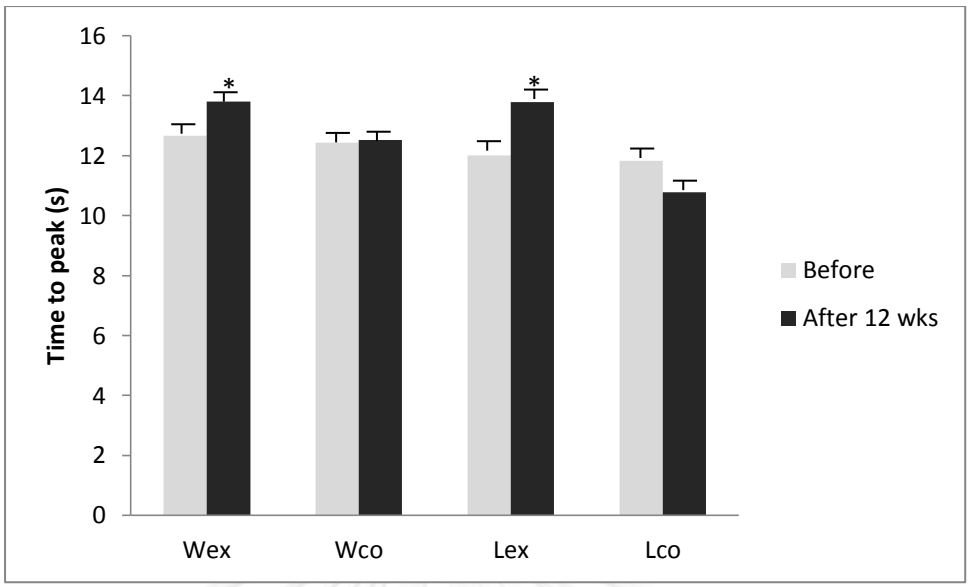
n = number of subjects. PORHpeak; amplitude of peak perfusion flux, PU; arbitrary perfusion units. †represents statistical difference between Wex versus Wco, P<0.05.

Table 36 The comparison of time to peak (s) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	**Time to peak (s)**		**P-value**
	Pre-test	**Post-test**	
Wex (n = 13)	12.66 ± 0.5	13.80 ± 0.4*	0.03
Wco (n = 15)	12.44 ± 0.5	12.52 ± 0.7	0.93
Lex (n = 10)	12.00 ± 0.7	13.78 ± 0.9*	0.04
Lco (n = 15)	11.83 ± 0.6	10.77 ± 0.8	0.27

Values are mean ± SEM.

*represents significant difference from baseline, P<0.05.



*represents significant difference from baseline, P<0.05.

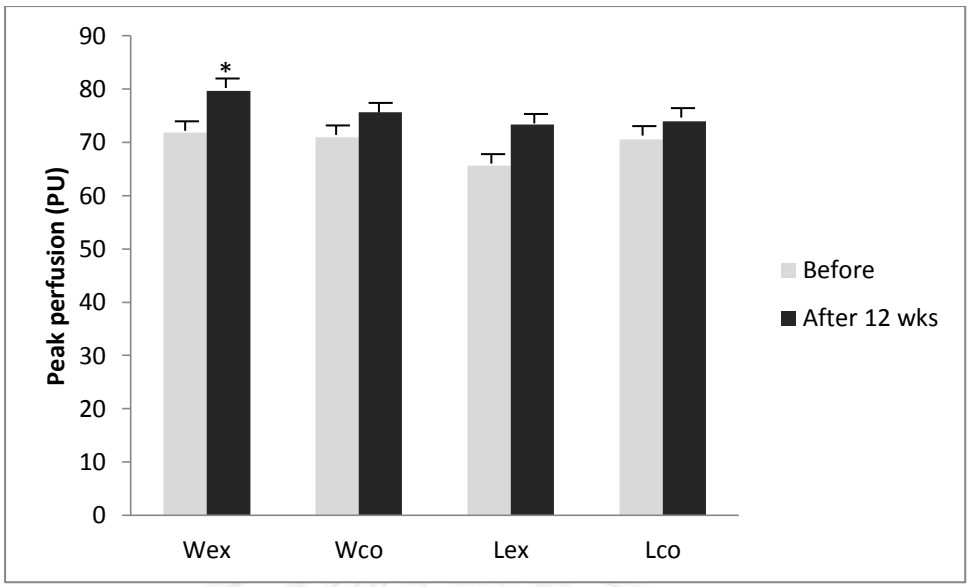
Figure 25 The comparison of time to peak (s) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 37 The comparison of peak perfusion (PU) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Peak perfusion (PU)		P-value
	Pre-test	Post-test	
Wex (n = 13)	71.85 ± 2.6	79.69 ± 2.5*	0.04
Wco (n = 15)	70.93 ± 2.7	75.60 ± 2.4	0.21
Lex (n = 10)	65.60 ± 3.1	73.30 ± 3.3	0.10
Lco (n = 15)	70.53 ± 3.2	73.93 ± 2.7	0.42

Values are mean ± SEM.

*represents significant difference from baseline, P<0.05



*represents significant difference from baseline, P<0.05.

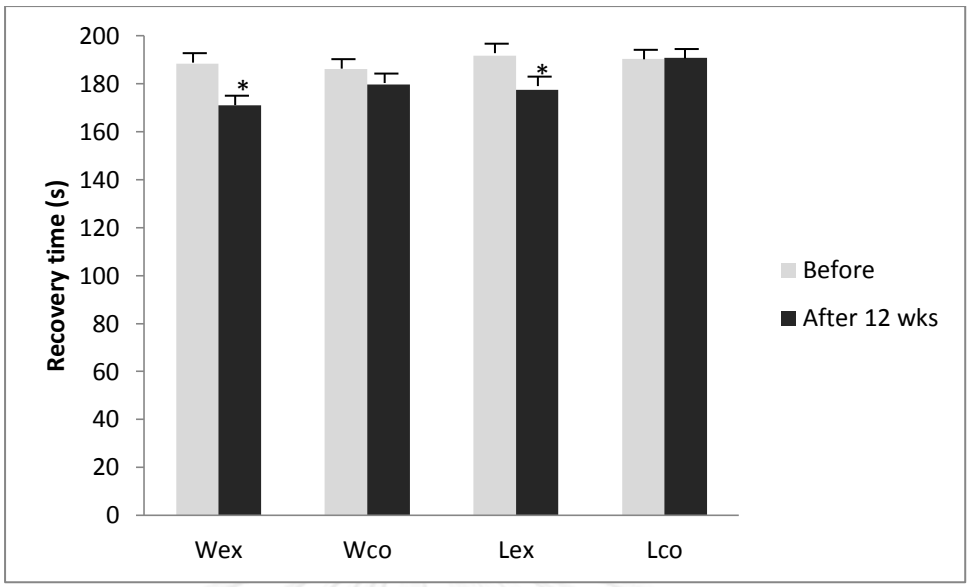
Figure 26 The comparison of peak perfusion (PU) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

Table 38 The comparison of recovery time (s) between pre- and post- training and among four groups of subjects with type 2 diabetes mellitus: Wex; Water-based exercise training subjects, Wco; Water immersion subjects, Lex; Land-based exercise training subjects, Lco; non exercise control subjects after 12 weeks.

Group	Recovery time (s)		P-value
	Pre-test	Post-test	
Wex (n = 13)	188.37±2.4	170.87±2.6*	0.0005
Wco (n = 15)	186.14±2.9	179.71±2.2	0.09
Lex (n = 10)	191.74±3.1	177.36±4.4*	0.01
Lco (n = 15)	190.32 ± 2.4	190.75 ± 2.1	0.89

Values are mean ± SEM.

*represents significant difference from baseline, P<0.05.



*represents significant difference from baseline, P<0.05.

Figure 27 The comparison of recovery time (s) between before and after 12 weeks training and among four groups: water-based exercise training (Wex), water immersion (Wco), land-based exercise training (Lex), non-exercise control (Lco).

CHAPTER V
DISCUSSION

The present study investigated the effect of water-based exercise training for 12 weeks, compare to land-based exercise, on physiological adaptations and cutaneous microvascular function in type 2 diabetic patients. A significant decrease in body weight was only observed in the Wex group meanwhile percentage body fat was significantly decreased only in the Lex group. However, there was no significant change in BMI and WHR for all groups. Resting heart rate and SBP decreased were significantly only observed in the Wex group. In addition, there was a significant difference in heart rest at rest between the Wex and Wco group. Only in the Lex group, DBP was significantly decreased. In contrast, DBP increased in Wco group. Handgrip strength was also increased in both Wex and Lex group. Moreover, maximal oxygen consumption significantly increased only in the Wex group. In this study, FBG, HbA1c, total cholesterol, LDL was decreased and HDL was increased in both exercise groups. Also, FBG was decreased significant difference in the Wco group. Only the Wex group had reduction in triglyceride, insulin, HOMA-IR, CRP and MDA. Furthermore, there was a significant difference in CRP between the Wex and Lex group. The greater in PORHpeak of Wex group were also observed significantly. In addition, time-to-peak was significantly increased and the recovery time was also significantly decreased in both exercise groups. However, there were no significant changes in PORH were obtained in the both control group. Therefore, this is clear that water-based exercise training could be used as an effective program to improve physical functions and microvascular function in patients with T2DM.

Subjects characteristic

In this study, the subjects were similar in age, height and initial fitness level. Some of their physical fitness variables were far beyond the normal range of the population. The body mass index found in our subjects was in the high range, it is interesting to note that this value was higher than that in the sedentary [91]. Therefore, it seems likely that type 2 diabetic patients in this study have overweight. Correspond that aging is generally followed by physiological changes in body composition. Essentially, elderly patients with T2DM are showing a decline in muscle mass and increase of body fat. Thus, age is considered a primary risk factor for the development and progression in chronic disease.

The number of patients with diabetes mellitus is expected to increase by 46% around the world, between 2000 and 2010, the incidence of diabetes mellitus is increasing rapidly as a result of aging population. Also, type 2 diabetes in older person is an age-prevalent metabolic disorder, characterized by insulin resistance with relative insulin deficiency [92, 93] with the highest prevalence. In T2DM, microvascular function is impaired from the onset of the disease and is closely associated with hyperglycemia and may contribute to insulin resistance. The way of cutaneous microvascular function is altered in diabetic patients is not yet fully understood, but the loss of normal microvascular function could be involved in the pathogenesis of diabetic angiopathy, as microvascular dysfunction is associated with diabetic microangiopathy and macroangiopathy [94, 95]. Under conditions of hyperglycemia, recent reports indicate that an improved metabolic control in diabetic patients, whatever the treatment used, is associated with near normalization or restoration of normal microvascular function [6]. Consistent with this study supports that these physical characteristic of subjects are influenced the aging process in older adults type 2 diabetic patients. Therefore, effective exercise protocols, at both the individual and population levels, are desperately needed to slow the diabetes epidemic and reduce diabetes related complications.

Long-term effects of exercise training in type 2 diabetes mellitus

In the present study, the subjects were attended either to the water-based exercise training or the land-based exercises training for 30 minute of continuous aerobic exercise performed at 70% of maximum heart rate 3 days/wk. There were effective in reducing body weight, SBP, heart rate at rest, fasting blood glucose, HbA1c, lipid profile, insulin, HOMA-IR, CRP, MDA and increasing grip strength, VO_2max as well as improving post-occlusive reactive hyperemia (PORH). Consistent with previous studies [96-98] have shown that both aerobic and resistance training improve metabolic control (blood glucose level and insulin resistant), fat oxidation and muscle storage. Exercise training also enhances skeletal muscles responsive to increased insulin expression or activity of proteins involved in glucose metabolism and insulin signaling in T2DM. Fat oxidation is also a key aspect of improved insulin action, and training increases lipid storage in muscle and fat oxidation capacity [97]. In addition, the findings of earlier reported that deep water running is also maintaining and improving cardiovascular fitness and resistance-type shallow water exercise reduces the impact stress to the joints and often allows greater mobility and improves strength [96, 98].

Exercise program is an important goal of treatment in T2DM to achieve or maintain optimal blood glucose, lipid, and blood pressure levels to prevent or delay chronic complications of diabetes [99]. Aerobic exercise has been the mode traditionally recommended for T2DM patients that are strongly associated with weight loss, improved fasting blood glucose, and increased cardiovascular fitness [100]. In addition, the American Diabetes Association (ADA) and American College of Sports Medicine (ACSM) also recommend resistance training as one part of exercise program for diabetes prevention and management [91]. ACSM endorses exercise as a treatment method for people with type 2 diabetes and currently recommends expending a minimum cumulative total of 1000 kcal/wk of energy from aerobic activities [101]. ADA has similar recommendations for at least 150 min per week of moderate intensity aerobic physical activity and/or 90 min per week of vigorous aerobic exercise [63]. Accordingly, the water-aerobic exercise program of this study was conducted in an indoor swimming pool within a range of water temperature pool (~34 to 36°C). This protocol has been the major focus for exercise-training studies due to consistent findings of improved glucose control [102, 103]. Confirm to the results of our study with warm water training in the elderly, supporting that this training is safe for patients with T2DM.

Comparison of the effects of water-based and land-based exercise training on hemodynamic changes

After 12 weeks of Wex and Lex training program, SBP and HR_{rest} were significantly reduction as compared with pre-exercise training ($P<0.05$). However, there was no significant difference in these variables in the Wco and Lco group. Beside DBP was decreased only in Lex group. In contrast, DBP increased in Wco group. In this study, the subjects practiced a simplified of aqua-aerobic form same as modification of aerobic form in the land-based exercise group by following instructions provided by a qualified trainer. Both training groups performed aerobic exercise protocol at 70% of MHR by used a HR monitoring device to ensure that the training intensity was maintained as prescribed. Subjective estimation of working load was carried out using RPE scale.

In the present study, subjects were required to attend exercise training program for 30 min, 3 days/wk for 12 weeks. It has been recommended that frequency of exercise is between three and five sessions per week can improve cardiovascular performance [65]. In general, increasing activity levels have proven to be effective in reducing blood pressure [44]. Therefore, changes in resting heart rate

and blood pressure (systolic and diastolic blood pressures) at 12 weeks of training in the water-based exercise training group are consistent with the findings of earlier studies [104]. This finding is very interesting considering aerobic exercise, traditionally thought to be better type of exercise to improve blood pressure. Aqua-aerobic exercise training, when properly supervised, is an excellent type of exercise to improve the hemodynamic responses of T2DM subjects. This was demonstrated with the decrease in the subjects' HR and the improvement in the sensitivity of the aortic baroreceptors, which contributed to a more efficient regulation of blood pressure [105-107]. Furthermore, the hydrostatic pressure of being in the water helped to increase the venous return to the heart and, thus improve the subjects' blood circulation [108].

Comparison of the effects of water-based and land-based exercise training on body composition

In the current study, weight loss and decreasing percentage of body fat after 12 weeks exercise training was observed in water-based and land-based exercise training groups, respectively. Clearly that water-based exercise as effective as exercise on land for weight control although no significant change in body mass index (BMI) after training in this study. It may be participants were still have physical activity and did not strong diet restriction. Similar results were seen by Taunton and colleagues (1996) following 12 weeks of shallow water aquatic training [109]. But, some investigations has been shown insufficient for weight loss after aquatic training [6, 96, 110], because of obese and older people frequently have difficulty performing sufficient exercise to create a large energy deficit and can easily counterbalance expenditures by eating more. However, a reduction in body fat [98, 111] was found in higher exercise ~2,000 to 2500 kcal/wk sustainable more weight loss than 1,000 kcal/wk of exercise.

Favorable changes in subjects with T2DM found in this study are demonstrate that will help type 2 diabetic patients associated with obesity, dislipidemia, and hypertension, which are important risk factors for micro- and macrovascular diseases. In fact, regular exercise seems to be effective in reducing the level of triglycerides and lipoprotein cholesterol in patients with T2DM relates to weight loss [112]. Previous study shown that low intensity exercise, 5 times per week, 1 h per session, and performed continuously during 1 year, can reduce body weight [111]. Weight loss also leads to a decrease in insulin resistance [44].

Comparison of the effects of water-based and land-based exercise training on physical fitness

In fact, the elderly patients with T2DM are showing a decline in muscle mass associated with a reduction in metabolic function. Therefore, the results of the current study were found a significant increase in VO_2 max (i.e., even a small increase) and grip strength after 12-wk water-based exercise training program should be viewed as a positive physiological response (in terms of cardiovascular disease). These finding is supported by previous reports that concluded deep water training is effective to maintain and improve cardiovascular fitness, muscular strength as well as functional mobility [113, 114]. Scientific reports [95, 110] are also available to induce the same energy expenditure in water as on land at a speed of 3.5 $km \cdot h^{-1}$ and to maintenance aerobic performance. Clearly, aqua-aerobic exercise programs are the one of save forms for diabetic patients that consist of aerobic endurance exercise combine to resistance exercise, which enhances cardiorespiratory fitness, muscular strength and endurance.

However, there was no significant difference in VO_2max between water-based and land-based exercise. As expected, low VO_2max values have been found mostly in patients with T2DM when compared with healthy age-matched controls [104]. Thus, a main target of any intervention should be to increase this clinical component of disease as showing in the result. Increasing VO_2max at 12 weeks of training in both groups is useful to prevent cardiovascular disease (CVD), hypertension, neuropathy, or micro-vascular changes. Since, a recommend clinical trials evaluating exercise interventions in T2DM have used a frequency of three times per week, should be performed at least 10 min bouts of exercise, corresponding approximately to 40%–60% of VO_2max [44]. It is clear that a greater benefit in the water-based exercise training is suitable for subjects with T2DM as same as the land-based exercise training. In addition, this study demonstrated that a significant improvement in grip strength increased with VO_2max. The reason for this finding may be resistance is provided by the water that surrounds the exercise participant. This multi-directional resistance helps the individual to maintain or enhance muscular strength and endurance even with gentle movements [115].

Comparison of the effects of water-based and land-based exercise training on blood chemical

This present finding found more changes of glycemic control (FBG, HbA1c) and lipid profile (total cholesterol, HDL, LDL) were observed in water-based and

land-based exercise training at 12 weeks. In addition, there were improvements in triglyceride, and insulin resistance only in water-based exercise training. During exercise, contracting muscles increase uptake of BG to supplement intramuscular glycogenolysis. As glycogen stores become depleted, muscles increase their uptake and use of circulating BG, along with FFA released from adipose tissue. Intramuscular lipid stores are more readily used during longer-duration activities and recovery. Glucose production also shifts from hepatic glycogenolysis to enhanced gluconeogenesis as duration increases [97]. Similarly, a meta-analysis of 15 papers suggested that regular exercise in those with T2DM lowers weight and improves HbA1c values [6]. Previous studies of small RCTs involving T2DM have reported that aerobic training decreases total cholesterol and LDL including raises HDL [116].

Several studies [81, 117, 118] have shown that improved glycemic control is associated with decreased rates of chronic complication and cardiovascular diseases. Aquatic exercise improved the subjects' glycemic control, which has been reported in a meta-analysis of 15 papers that suggested regular exercise in patients with T2DM can lose weight and improve HbA1c values [6]. In the present study, the water-based exercise group showed a significant difference in HbA1c when compared to baseline. It is clear that the water-based exercise training protocol represents a useful procedure to control glycaemia of older subjects with T2DM. Although, exercise in water resulted in a non-significant correlation between VO_2 max and HbA1c. In this study, participants in the water-based control group were immersed in the same water temperature of training period. Water temperatures around 30-34°C in the present study seems to be appropriate for microvascular function improvement. This particular range of water temperature is selected according to the comfortable feeling of population in tropical zone. Previous study [119] confirmed that pure immersion in a heated pool from 37 to 40 degrees was resulted in reduction in weight and plasma blood glucose. It may be due to increasing skeletal muscle blood flow facilitates insulin mediated glucose uptake from immersion in a heated pool [120].

In the present study, there were shown in a significant reduction of CRP and MDA level only in the Wex group. In addition, the change of CRP in Wex group was significantly lower than those of the Wco, Lex and Lco group as well as significantly difference between the Wex and Lex groups. These finding indicated that the Wex training was effective in the attenuating ROS may be one of the reason to improve cutaneous vascular function. Since the diminished values of ROS could be related to an enhancement of cutaneous microvascular function, or due to an increase in

antioxidant activity therefore facilitating the removal of ROS [121]. It could be described that aqua-aerobic training protocol was sufficient to enhance vascular inflammatory markers significantly [122].

Comparison of the effects of water-based and land-based exercise training on micro-vascular reactivity

In the current study, $PORH_{peak}$ was significantly higher only in water-based exercise training compare to baseline while, no significant difference between the other groups. Microcirculation is known to be disturbed in T2DM [123]; a reduction of capillary skin blood flow during reactive hyperemia may cause a regional ischemia and an impaired tissue exchange during stress situations [124]. Several non-invasive techniques have been developed to investigate the microcirculation of the skin. The increase in micro-vascular blood flow following arterial occlusion, PORH, can be assessed using the non-invasive laser Doppler technique [125]. Following release of the arterial occlusion there is a marked transient increase in micro-vascular blood flow to the ischemic tissues. Laser Doppler derived PORH measurements have been shown to be highly reproducible [126]. The mechanisms behind PORH are believed to be a combination of: myogenic relaxation of the vessels [127], release of local mediators and metabolites from the ischemic tissue [128], and more recently the involvement of sensory nerves [129]. This non-invasive assessment may reflect micro-vascular cutaneous microvascular function, which depends on nitric oxide (NO) production in cutaneous microvascular cells.

This study has shown that the recovery time was significantly lower in Wex and Lex training group at 12 weeks training. Time-to-peak was also significantly increased from pre-test ($P<0.05$) in the both exercise groups. There are only a few studies using the laser-Doppler method have been performed on skin microcirculation in older subjects with T2DM and the results still unclear. Some investigation reported that diabetes was associated with decreased in skin blood flow and found that microvascular flow was worse in the lower limbs [124, 130]. In contrast that Edmonds et al., 1982 [94] and Archer et al., 1984 [131] had reported increase foot blood flow in subjects with diabetes compared with control subjects. Reason for disturbances of the cutaneous microcirculation may change the course of diabetes. At the onset of the disease abnormalities of the microvascular hemodynamics are reversible and may be due to alterations in smooth muscle cell function [132], probably partly caused by altered cutaneous microvascular cell function resulting in increased endothelium-derived relaxing factor [133] and

decreased prostacyclin production [134]. This finding suggested that water-based exercise training was sufficient to improved cutaneous microvascular function in the elderly with type 2 diabetic patients.

CHAPTER VI

CONCLUSION

The present study investigated the effect of water-based exercise training for 12 weeks on physiological adaptation and cutaneous microvascular function in type 2 diabetic patients using protocol for a randomized controlled trial. The results conclude that water-based exercise training significantly improved body composition and hemodynamic better than land-based exercise training when compared to baseline. Both water-based and land-based exercise training significantly enhanced grip strength although the leg strength and flexibility were not changed. In addition, water-based exercise training induced improvement in microvascular function possible through improved VO_2max, glycemic control and lipid profiles as similar pathway as exercise on land (Figure 4 and 5). Furthermore, only water-based exercise training that contributed to recovered insulin resistance, decreased CRP and improved amplitude of peak flux during post-occlusive reactive hyperemia. Similarly, both water-based and land-based exercise training affects recovery time of PORH. These resulted in the better benefits in the water-based exercise training on micro- and macro-vascular function.

It is suggested that a long term effect of aqua-aerobic exercise training improves hemodynamic, glycemic control, lipid profile, insulin resistance and health-related physical fitness in older adults with T2DM. Important finding of water-based exercise training was more effective exercise to improved antioxidant and increased microvascular function than land-based exercise training. Moreover, due to the design and monitoring of training protocol no subjects were injured during the training program and no significant muscle soreness. Therefore, it is more than reasonable to conclude that aquatic exercise is a beneficial mode of exercise for elderly patients with type 2 diabetes.

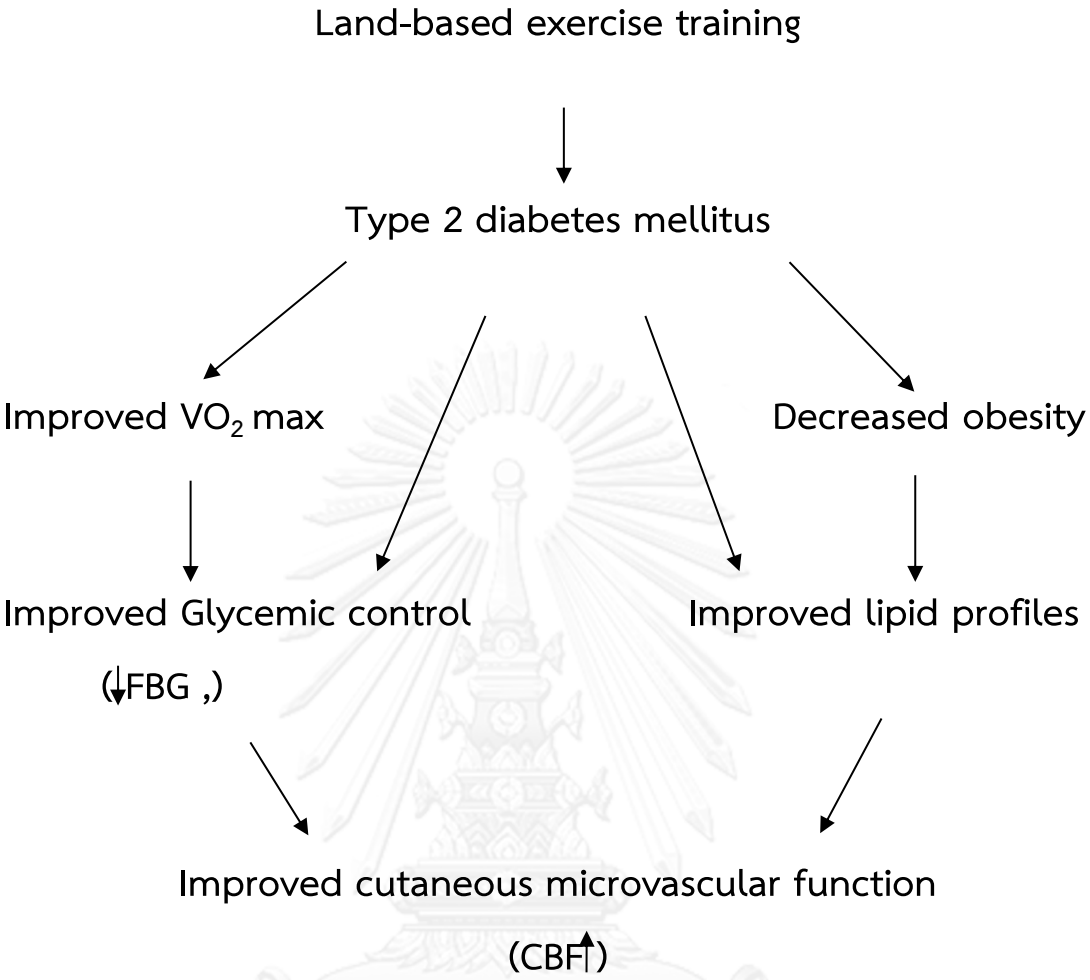


Figure 28 The mechanism of land-based exercise training on cutaneous microvascular function.

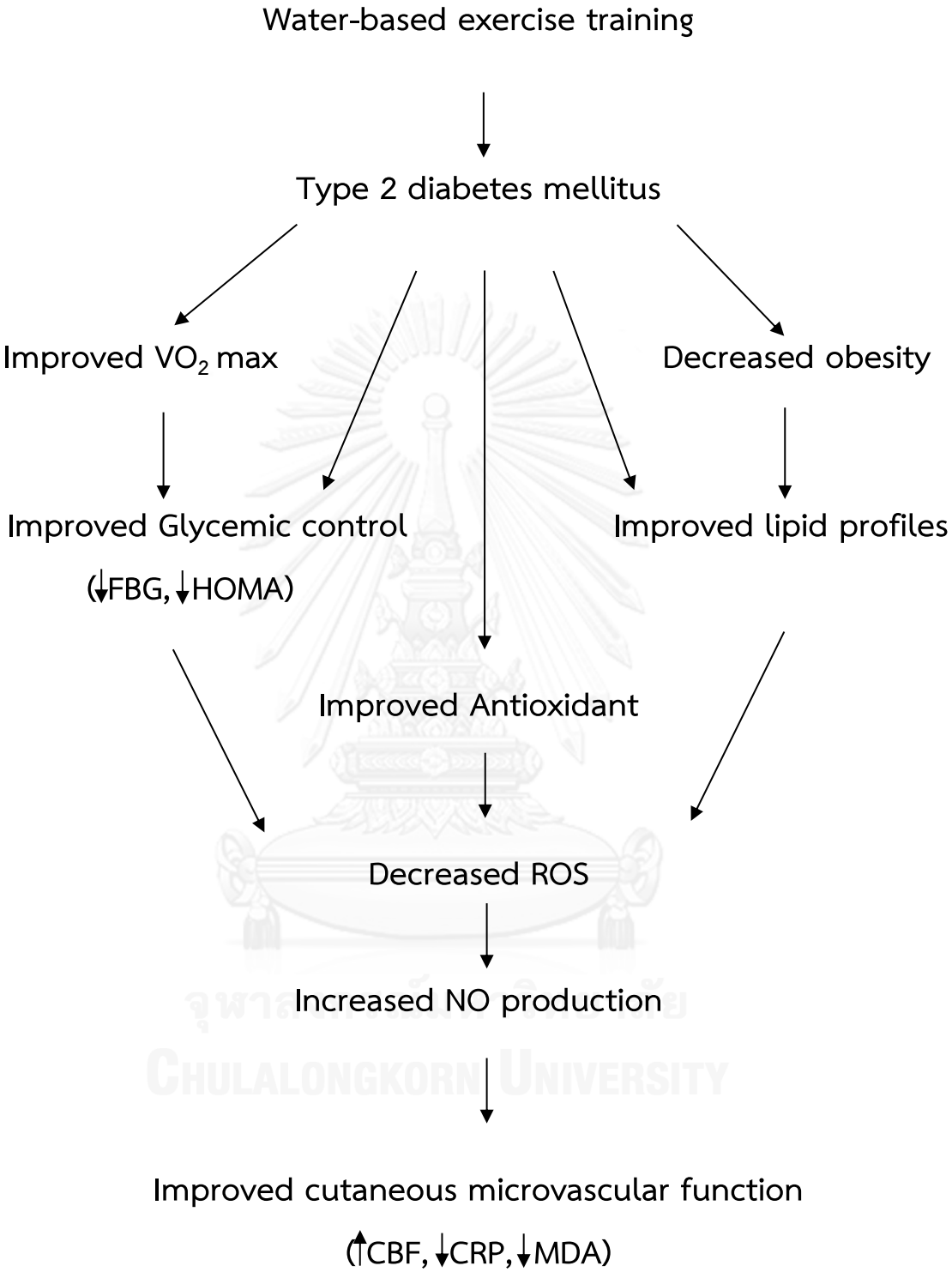


Figure 29 The mechanism of water-based exercise training on cutaneous microvascular function.

Limitation of this study

1. Due to included only 7-9 % of HbA1c (moderate severity), finding participants is difficult.

2. In this study, we could not control daily life behaviors of our subjects which may affect the results of biological data, blood chemistry and blood flow data.

Suggestion for further research

1. To explain the mechanism of water-based exercise training deeply, other parameters i.e. NO, GLUT4 and insulin signaling should be included.

2. The difference between duration of the recovery between bouts and a progression in water-based exercise training intensity should be compared.

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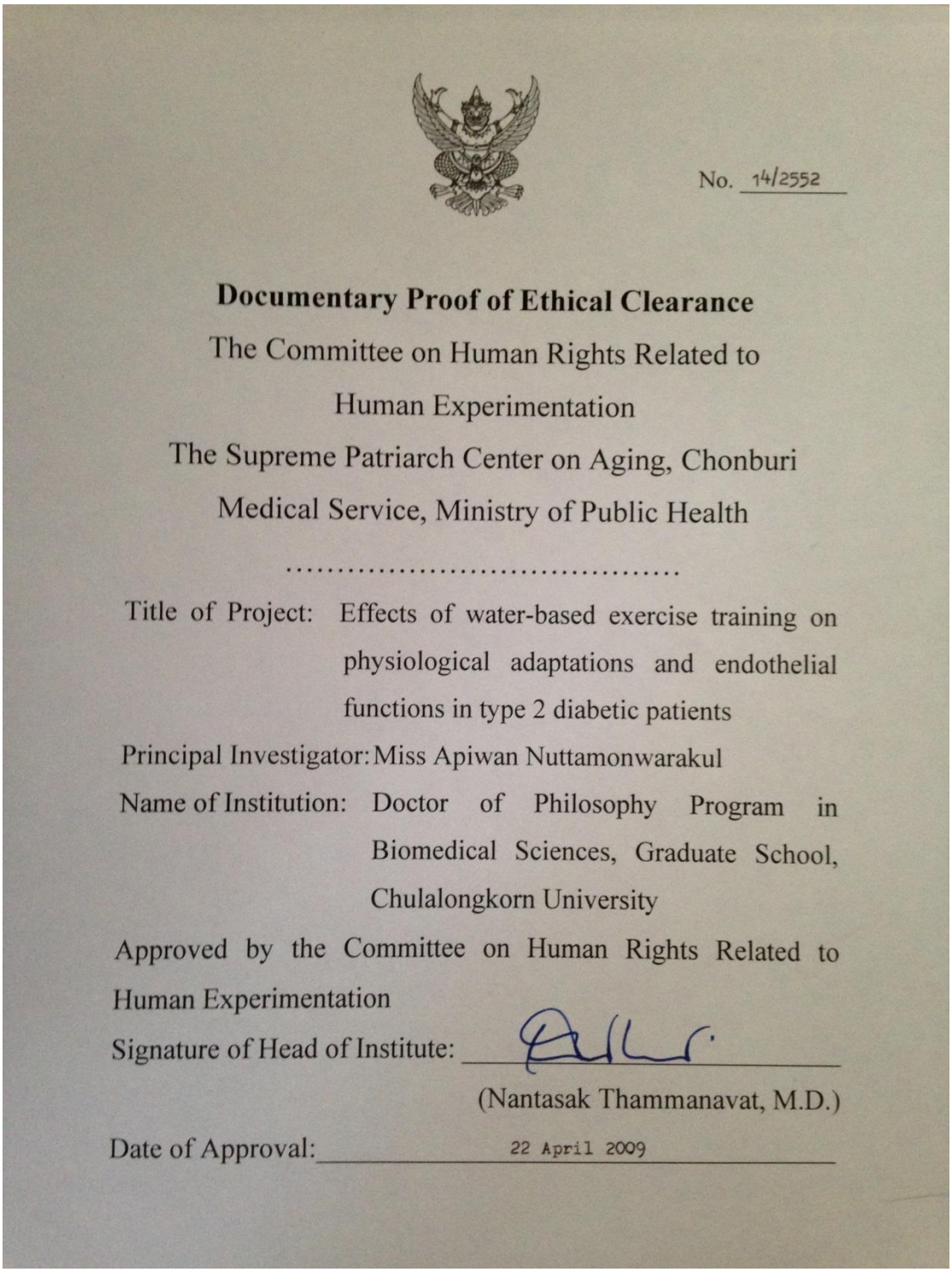
APPENDIX

APPENDIX A

The Institutional Certificate of Approval

APPENDIX A

The Institutional Certificate of Approval



No. 14/2552

Documentary Proof of Ethical Clearance

The Committee on Human Rights Related to Human Experimentation

The Supreme Patriarch Center on Aging, Chonburi

Medical Service, Ministry of Public Health

.....

Title of Project: Effects of water-based exercise training on physiological adaptations and endothelial functions in type 2 diabetic patients

Principal Investigator: Miss Apiwan Nuttamonwarakul

Name of Institution: Doctor of Philosophy Program in Biomedical Sciences, Graduate School, Chulalongkorn University

Approved by the Committee on Human Rights Related to Human Experimentation

Signature of Head of Institute: _____

(Nantasak Thammanavat, M.D.)

Date of Approval: 22 April 2009

APPENDIX B

Physical Activity Readiness Questionnaire; PAR-Q

APPENDIX B

Physical Activity Readiness Questionnaire; PAR-Q

แบบสอบถามที่ใช้ในการวิจัย

รหัส.....

แบบประเมินความพร้อมก่อนออกกำลังกาย

(Physical activity readiness questionnaire; PAR-Q)

การออกกำลังกายอย่างสม่ำเสมอเป็นผลดีต่อสุขภาพและมีความสนุกสนาน ประชาชนจำนวนมากเริ่มสนใจที่จะเข้าร่วมออกกำลังกายมากขึ้นทุกวัน โดยทั่วไปการออกกำลังกายหนักปานกลางค่อนข้างปลอดภัยสำหรับคนส่วนใหญ่ อย่างไรก็ตาม อาจมีบางคนที่จำเป็นต้องได้รับการตรวจร่างกายจากแพทย์ก่อนที่จะเข้าร่วมการออกกำลังกายที่หนักขึ้น

ถ้าท่านมีแผนการที่จะออกกำลังกายหนักปานกลางมากกว่าที่เป็นอยู่ในปัจจุบัน กรุณาตอบคำถามทั้ง 7 ข้อข้างล่างนี้ ถ้าท่านมีอายุระหว่าง 15-69 ปี การตอบคำถามในแบบประเมินจะช่วยบอกว่าท่านสมควรเข้ารับการตรวจร่างกายจากแพทย์ก่อนที่ท่านจะเริ่มต้นออกกำลังกายหรือไม่

โปรดอ่านอย่างละเอียดและตอบคำถามเหล่านี้ตามความเป็นจริงว่า มี / เคย หรือ ไม่มี / ไม่เคย ในช่วง 6 เดือนที่ผ่านมา

☐ เคย	☐ ไม่เคย	1. แพทย์ที่ตรวจรักษาท่าน เคยบอกหรือไม่ว่า ท่านมีความผิดปกติของหัวใจ และควรออกกำลังกาย ภายใต้คำแนะนำของแพทย์เท่านั้น
☐ เคย	☐ ไม่เคย	2. ท่านมีความรู้สึกเจ็บปวดหรือแน่นบริเวณหน้าอก ขณะที่ท่านออกกำลังกาย หรือไม่ ?
☐ เคย	☐ ไม่เคย	3. ในรอบเดือนที่ผ่านมา ท่านเคยมีอาการเจ็บแน่นหน้าอก ในขณะที่อยู่เฉยๆ โดยไม่ได้ออกกำลังกายหรือไม่ ?
☐ เคย	☐ ไม่เคย	4. ท่านมีอาการสูญเสียการทรงตัว (ยืนหรือเดินแย) เนื่องมาจาก อาการวิงเวียนศีรษะหรือไม่ ? หรือท่านเคยเป็นลมหมดสติหรือไม่ ?
☐ เคย	☐ ไม่เคย	5. ท่านมีปัญหาที่กระดูกหรือข้อต่อ ซึ่งจะมีอาการแย่ลง ถ้าออกกำลังกาย หรือไม่ ?
☐ เคย	☐ ไม่เคย	6. แพทย์ที่ตรวจรักษาท่าน มีการสั่งยารักษาโรคความดันโลหิตสูง หรือความผิดปกติของหัวใจให้ท่านหรือไม่ ?
☐ เคย	☐ ไม่เคย	7. เท่าที่ท่านทราบ ยังมีเหตุผลอื่นๆ อีก ที่ทำให้ท่านไม่สามารถออกกำลังกายได้ หรือไม่ ?

ที่มา : ACSM, 2000.

ข้าพเจ้าได้อ่านได้ทำความเข้าใจและกรอกแบบ PAR-Q ทุกคำถามด้วยความเต็มใจ

ลงชื่อ.....ผู้เข้าร่วมกิจกรรม

(.....)

วันที่...../...../.....

ลงชื่อ.....ผู้ทำการวิจัย

(.....)

วันที่...../...../.....

APPENDIX C

Information sheet for research participants

APPENDIX C

Information sheet for research participants

เอกสารข้อมูลสำหรับผู้เข้าร่วมโครงการวิจัย

ชื่อโครงการวิจัย: ผลของการฝึกออกกำลังกายในน้ำต่อการปรับตัวทางสรีรวิทยาและหน้าที่ของเซลล์บุผนังหลอดเลือดในผู้ป่วยโรคเบาหวานประเภทที่ 2

ผู้ทำการวิจัย: นส.อภิวรรณ ณัฐมนวรกุล

ที่อยู่: ศูนย์สมเด็จพระสังฆราชญาณสังวรเพื่อผู้สูงอายุ 444 หมู่ 11 ต.ห้วยใหญ่ อ.บางละมุง จ.ชลบุรี 20260 เบอร์โทรศัพท์ 038 238484

ที่ปรึกษาโครงการวิจัย: ผศ.ดร.ดรุณวรรณ สุขสม, รศ.ดร.สุภัทรา อมาตยกุล

เรียน ท่านผู้เข้าร่วมโครงการวิจัยทุกท่าน

เอกสารนี้เป็นเอกสารที่แสดงข้อมูลเพื่อใช้ประกอบการตัดสินใจของท่านในการเข้าร่วมการศึกษาวิจัย อย่างไรก็ตามก่อนที่ท่านจะตกลงเข้าร่วมโครงการนี้ ขอให้ท่านอ่านเอกสารฉบับนี้อย่างละเอียดเพื่อให้ท่านได้ทราบถึงเหตุผลและรายละเอียดของโครงการศึกษาวิจัยนี้ หากท่านมีข้อสงสัยใดๆเพิ่มเติม กรุณาซักถามรายละเอียดจากผู้ทำการวิจัยโดยตรง

ท่านสามารถขอคำแนะนำในการเข้าร่วมโครงการศึกษาวิจัยจากสมาชิกในครอบครัว เพื่อน หรือแพทย์ประจำตัวของท่านได้ ถ้าท่านตัดสินใจแล้วว่าต้องการเข้าร่วมโครงการศึกษาวิจัยนี้ ขอให้ท่านลงชื่อยินยอมในเอกสารแสดงความยินยอมในเอกสารยินยอมของโครงการวิจัยนี้

<u>วัตถุประสงค์ของการศึกษา</u>

วัตถุประสงค์ของการศึกษาในครั้งนี้คือ เพื่อศึกษาผลของการฝึกออกกำลังกายในน้ำต่อการปรับตัวทางสรีรวิทยาและหน้าที่ของเซลล์บุผนังหลอดเลือดในผู้ป่วยโรคเบาหวานประเภทที่ 2

<u>คุณสมบัติของผู้วิจัย</u>

เป็นผู้ที่มีองค์ความรู้เรื่องการจัดทำโครงการวิจัย โดยครอบคลุมถึงระเบียบวิธีวิจัย วรรณกรรมที่เกี่ยวข้อง ผู้เชี่ยวชาญที่ต้องประสานองค์ความรู้และสถิติที่ใช้ในการวิเคราะห์ข้อมูล

<u>วิธีการที่เกี่ยวข้องกับการวิจัย</u>

หากท่านมีคุณสมบัติเหมาะสมและยินยอมที่จะเข้าร่วมการวิจัยครั้งนี้ ท่านจะได้รับเชิญเข้าเข้าร่วมโครงการวิจัย เป็นระยะเวลา 3 เดือน ณ ศูนย์สมเด็จพระสังฆราชญาณสังวรเพื่อผู้สูงอายุ และทำการวัดข้อมูลทางสุขสมรรถนะ ประกอบด้วย น้ำหนัก ส่วนสูง เส้นรอบเอว ค่าดัชนีมวลกาย ความ

ดันโลหิต ชีพจร วัดค่าการใช้ออกซิเจนสูงสุดขณะออกกำลังกาย ความแข็งแรงของกล้ามเนื้อแขน ขา และตรวจทางห้องปฏิบัติการ 1 ครั้งในวันก่อนเข้าปฏิบัติตามโปรแกรมการฝึกออกกำลังกาย หลังจากครบกำหนดท่านจะได้รับการวัดผลข้อมูลทางสุขสมรรถนะ ประกอบด้วย น้ำหนัก ส่วนสูง เส้นรอบเอว ค่าดัชนีมวลกาย ความดันโลหิต ชีพจร วัดค่าการใช้ออกซิเจนสูงสุดขณะออกกำลังกาย ความแข็งแรงของกล้ามเนื้อแขน ขาและตรวจทางห้องปฏิบัติการอีก 1 ครั้ง ในวันที่สิ้นสุดการปฏิบัติตามโปรแกรมการฝึก เพื่อประเมินผลของการฝึกออกกำลังกาย โดยระยะเวลาที่ท่านจะเข้าร่วมในการวิจัยคือ ระหว่างเดือนพฤษภาคม 2552 ถึง พฤษภาคม 2553

<u>คุณสมบัติของผู้เข้าร่วมวิจัย</u>

ผู้ที่เข้าร่วมโครงการวิจัยที่มีอายุ 60 ปีขึ้นไป ทั้งเพศชายและเพศหญิง จำนวน 80 คน เป็นประชากรในเขตภาคตะวันออกที่ได้รับการวินิจฉัยจากแพทย์ว่าเป็นผู้ป่วยเบาหวานประเภทที่ 2 และไม่แสดงอาการเจ็บป่วยที่เป็นอุปสรรคต่อการเข้าโปรแกรมการออกกำลังกาย เช่น โรคหัวใจ ไทรอยด์ โรคสมองและระบบประสาท อัมพาต เป็นต้น มีความสมัครใจยินยอมเข้าร่วมการศึกษาครั้งนี้ โดยมีหนังสือยินยอมเป็นลายลักษณ์อักษรด้วยตนเองของผู้เข้าร่วมโครงการวิจัย

<u>ความรับผิดชอบของอาสาสมัครผู้เข้าร่วมโครงการวิจัย</u>

เพื่อให้งานวิจัยนี้ประสบความสำเร็จ ผู้วิจัยจึงขอความร่วมมือจากผู้ร่วมโครงการวิจัยทุกท่าน ซึ่งมีความจำเป็นอย่างยิ่งต่อความสำเร็จของงานวิจัย ดังนั้นผู้วิจัยจึงต้องขอให้ท่านปฏิบัติตามคำแนะนำของผู้วิจัย รวมทั้งแจ้งถึงความเปลี่ยนแปลงต่างๆที่เกิดขึ้นกับตัวท่านเองให้ผู้วิจัยได้รับทราบอย่างชัดเจนและตรงตามความเป็นจริง

<u>ความเสี่ยงที่อาจได้รับ</u>

หากท่านเกิดอาการผิดปกติ ไม่สบาย รู้สึกไม่พร้อม รู้สึกไม่ปลอดภัย หรือ ไม่ต้องการเข้าร่วมกิจกรรมใดที่ผู้วิจัยจัดเตรียมไว้ให้ ท่านควรแจ้งให้ผู้วิจัยทราบทันที และหากท่านมีข้อสงสัยใดๆ เกี่ยวกับความเสี่ยงที่อาจได้รับจากการเข้าร่วมโครงการวิจัยท่านสามารถซักถามผู้ทำวิจัยได้ตลอดเวลา

<u>ความเสี่ยงที่ได้รับจากการตรวจเลือด</u>

ท่านมีโอกาสที่จะเกิดอาการเจ็บ เลือดออก ช้ำจากการตรวจเลือด อาการบวมบริเวณที่ตรวจเลือดหรือหน้ามืด แต่โอกาสที่จะเกิดการติดเชื้อบริเวณที่ตรวจเลือดพบได้น้อยมาก

<u>ความเสี่ยงที่ไม่ทราบแน่นอน</u>

ท่านอาจเกิดอาการข้างเคียง หรือความไม่สบาย นอกเหนือจากที่ได้แสดงในเอกสารฉบับนี้ ซึ่งอาการข้างเคียงเหล่านี้เป็นอาการที่ไม่เคยพบมาก่อน เพื่อความปลอดภัยของท่าน ควรแจ้งผู้ทำวิจัยให้ทราบทันทีเมื่อเกิดความผิดปกติใด ๆ เกิดขึ้น

หากท่านมีข้อสงสัยใด ๆ เกี่ยวกับความเสี่ยงที่อาจได้รับจากการเข้าร่วมในโครงการวิจัย ท่านสามารถสอบถามจากผู้วิจัยได้ตลอดเวลา

หากมีการค้นพบข้อมูลใหม่ ๆ ที่อาจมีผลต่อความปลอดภัยของท่านในระหว่างที่ท่านเข้าร่วมในโครงการวิจัย ผู้ทำวิจัยจะแจ้งให้ท่านทราบทันที เพื่อให้ท่านตัดสินใจว่าจะอยู่ในโครงการวิจัยต่อไป หรือจะขอถอนตัวออกจากการวิจัย

ประโยชน์ที่อาจได้รับ

การเข้าร่วมโครงการวิจัยนี้อาจจะทำให้ท่านมีสุขภาพดีขึ้น หรืออาจลดความรุนแรงของโรคได้ แต่ไม่ได้รับรองว่าสุขภาพของท่านจะต้องดีขึ้นหรือความรุนแรงของโรคจะลดลงอย่างแน่นอน

วิธีการและรูปแบบการรักษาอื่นๆที่มีอยู่สำหรับอาสาสมัคร

ท่านไม่จำเป็นต้องเข้าร่วมโครงการวิจัยเพื่อประโยชน์ในการรักษาโรคที่ท่านเป็นอยู่เนื่องจากมีแนวทางการรักษาอื่นๆหลายแบบสำหรับรักษาโรคของท่านได้ ดังนั้นจึงควรปรึกษากับแพทย์ผู้ให้การรักษาท่านก่อนการตัดสินใจ

ข้อปฏิบัติของท่านขณะร่วมโครงการวิจัย

สิ่งที่ท่านควรปฏิบัติ คือ

- ท่านต้องให้ข้อมูลทางการแพทย์ของท่านทั้งในอดีตและปัจจุบัน แก่ผู้วิจัยด้วยความสัตย์จริง
- ท่านต้องแจ้งให้ผู้วิจัยทราบถึงความเปลี่ยนแปลงที่เกิดขึ้นระหว่างที่ท่านร่วมโครงการ

อันตรายที่อาจเกิดขึ้นในโครงการวิจัย

หากพบอันตรายที่เกิดขึ้นจากการวิจัย และพิสูจน์ได้ว่าท่านปฏิบัติตามคำแนะนำของทีมผู้ทำวิจัยแล้ว ผู้สนับสนุนโครงการวิจัยยินดีจะรับผิดชอบต่อค่าใช้จ่ายในการรักษาพยาบาลของท่าน การเซ็นชื่อในเอกสารยินยอมไม่ได้หมายความว่าท่านสละสิทธ์ทางกฎหมาย ตามปกติที่ท่านพึงมี

ในกรณีที่ท่านได้รับอันตรายใด ๆ หรือต้องการข้อมูลเพิ่มเติมเกี่ยวกับโครงการวิจัย ท่านสามารถติดต่อกับผู้ทำวิจัยได้ที่ โทร.038 238484 ในเวลาราชการ หรือ 086 6046519 ตลอด 24 ชั่วโมง

ค่าใช้จ่ายสำหรับอาสาสมัคร

ท่านจะได้รับสิทธ์ในการเข้าร่วมโปรแกรมการฝึกออกกำลังกายสำหรับผู้ป่วยเบาหวานประเภทที่ 2 ที่ผู้ทำวิจัยเป็นผู้จัดเตรียมให้โดยไม่ต้องเสียค่าใช้จ่ายใด ๆ ทั้งสิ้น

ค่าตอบแทนสำหรับผู้เข้าร่วมวิจัย

ท่านจะได้รับค่าตอบแทนในการเดินทางและค่าชดเชยในความไม่สะดวกสบายในการมาเข้าโครงการวิจัย รวมเป็นเงินจำนวน 175 บาทต่อคน

การเข้าร่วมและการสิ้นสุดโครงการวิจัย

การเข้าร่วมโครงการวิจัยในครั้งนี้เป็นไปโดยความสมัครใจ หากท่านไม่สมัครใจจะเข้าร่วมการศึกษาแล้ว ท่านสามารถถอนตัวได้ตลอดเวลา การขอถอนตัวจากโครงการวิจัยจะไม่มีผลต่อการ

ดูแลรักษาโรคของท่าน ตลอดจนความดูแลที่ท่านพึงได้รับจากศูนย์สมเด็จพระสังฆราชญาณสังวรเพื่อผู้สูงอายุ แต่อย่างใด

ผู้วิจัยอาจขอถอนท่านออกจากการเป็นอาสาสมัครในโครงการ เพื่อเหตุผลด้านความปลอดภัยของท่าน หรือเมื่อโครงการวิจัยนี้ยุติลงก่อนกำหนด หรือในกรณีต่อไปนี้

- ท่านไม่ให้ความร่วมมือ หรือ ไม่ปฏิบัติตามคำแนะนำของผู้ทำวิจัย

- ภาวะสุขภาพของท่านไม่เอื้อต่อการเป็นอาสาสมัครในโครงการ

<u>การปกป้องข้อมูลของอาสาสมัคร</u>

ข้อมูลที่อาจนำไปสู่การเปิดเผยตัวของท่าน จะได้รับการปกปิดและจะไม่เปิดเผยแก่สาธารณชน ในกรณีที่ผลวิจัยได้รับการตีพิมพ์ ชื่อและที่อยู่ของท่านจะได้รับการปกปิดอยู่เสมอ โดยจะใช้เฉพาะรหัสประจำโครงการวิจัยของท่าน

จากการลงนามยินยอมของท่าน ผู้วิจัย ที่ปรึกษาโครงการและผู้สนับสนุนการวิจัยมีสิทธิ์สามารถเข้าไปตรวจสอบบันทึกข้อมูลของท่านได้ตลอดเวลาแม้สิ้นสุดโครงการวิจัยแล้วก็ตาม หากท่านต้องการยกเลิกการให้สิทธิ์ดังกล่าว ท่านสามารถเขียนบันทึกขอยกเลิกการให้คำยินยอม โดยส่งไปที่ศูนย์สมเด็จพระสังฆราชญาณสังวรเพื่อผู้สูงอายุ 444 หมู่ 11 ต.ห้วยใหญ่ อ.บางละมุง จ.ชลบุรี 20260

หากท่านขอยกเลิกการให้คำยินยอมหลังจากที่ท่านได้เข้าร่วมโครงการวิจัยแล้ว ข้อมูลส่วนตัวของท่านจะไม่ถูกบันทึกเพิ่มเติม อย่างไรก็ตามข้อมูลอื่นๆของท่านอาจถูกนำมาใช้เพื่อประเมินผลการวิจัย และท่านจะไม่สามารถกลับเข้าร่วมโครงการนี้ได้อีก ทั้งนี้เนื่องจากข้อมูลของท่านที่จำเป็นต่อการวิจัยไม่ได้ถูกบันทึก

จากการลงนามยินยอมของท่าน ผู้ทำวิจัยสามารถบอกรายละเอียดของท่านเกี่ยวกับการเข้าร่วมโครงการนี้ให้ผู้แทนโดยชอบธรรมหรือแพทย์ผู้รักษาท่านทราบได้

<u>สิทธิ์ของผู้เข้าร่วมโครงการวิจัย</u>

ในฐานะอาสาสมัครผู้เข้าร่วมโครงการวิจัยท่านจะมีสิทธิ์ดังต่อไปนี้

- ท่านจะได้รับทราบข้อมูลเกี่ยวกับลักษณะและวัตถุประสงค์ของการวิจัยครั้งนี้

- ท่านจะได้รับการอธิบายเกี่ยวกับระเบียบวิธีการของการวิจัยรวมทั้งรูปแบบ วิธีการและกิจกรรมที่ใช้ในการวิจัยครั้งนี้

- ท่านจะได้รับการอธิบายถึงความเสี่ยงที่อาจได้รับจากการเข้าร่วมโครงการ

- ท่านจะได้รับการอธิบายถึงประโยชน์ที่อาจได้รับจากการเข้าร่วมโครงการ

- ท่านจะได้รับการเปิดเผยทางเลือกในการรักษาด้วยวิธีอื่น ซึ่งมีผลดีต่อท่านรวมทั้งประโยชน์และความเสี่ยงที่ท่านอาจได้รับ

- ท่านจะได้รับทราบแนวทางการรักษา ในกรณีที่พบโรคแทรกซ้อนภายหลังการเข้าร่วมโครงการ

- ท่านจะได้รับโอกาสในการซักถามเกี่ยวกับงานวิจัยหรือขั้นตอนที่เกี่ยวข้องกับงานวิจัย

- ท่านจะได้รับทราบว่าการยินยอมเข้าร่วมการวิจัยนี้ ท่านสามารถขอถอนตัวจากโครงการได้ทุกเมื่อ โดยผู้เข้าร่วมโครงการสามารถขอถอนตัวจากโครงการโดยไม่ได้รับผลเสียใดๆทั้งสิ้น

- ท่านจะได้รับสำเนาเอกสารใบยินยอมที่มีทั้งลายเซ็นและวันที่

- ท่านจะได้โอกาสในการตัดสินใจว่าจะเข้าร่วมโครงการหรือไม่ก็ได้ โดยปราศจากการใช้อิทธิพลบังคับ ข่มขู่ หรือหลอกลวง

หากมีข้อร้องเรียนทางด้านจริยธรรมการวิจัย กรุณาติดต่อสำนักงานคณะกรรมการพิจารณาจริยธรรมการวิจัย โทร. 038 238484 ในเวลาราชการ

ขอขอบคุณในความร่วมมือของท่านมา ณ ที่นี้

.....ผู้วิจัย

(นางสาวอภิวรรณ ณัฐมนวรกุล)

APPENDIX D

Informed consent form

APPENDIX D

Informed consent form

หนังสือยินยอม

(INFORMED CONSENT FORM)

การศึกษาเรื่อง ผลของการฝึกออกกำลังกายในน้ำต่อการปรับตัวทางสรีรวิทยาและหน้าที่ของเซลล์บุผนังหลอดเลือดในผู้ป่วยโรคเบาหวานประเภทที่ 2

วันให้คำยินยอม วันที่.....เดือน ..... พ.ศ. .....

ชื่อผู้ทำการศึกษา อภิวรรณ ณัฐมนวรกุล (นิสิตวิทยาศาสตร์ดุษฎีบัณฑิต สาขาชีวเวชศาสตร์)

ชื่อผู้ให้คำยินยอม ..... อายุ .....ปี

ก่อนที่จะลงนามในใบยินยอมให้ทำการศึกษานี้ ข้าพเจ้าได้รับการอธิบายจากผู้ทำการศึกษาวิจัยถึงวัตถุประสงค์ของการศึกษา วิธีการ ประโยชน์ที่จะเกิดจากการศึกษาในครั้งนี้อย่างละเอียด และมีความเข้าใจดีแล้ว

ผู้ทำการศึกษาวิจัยรับรองว่าจะตอบคำถามต่าง ๆ ที่ข้าพเจ้าสงสัยด้วยความเต็มใจ ไม่ปิดบังซ่อนเร้น จนข้าพเจ้าพอใจ

ข้าพเจ้ามีสิทธิที่จะบอกเลิกการเข้าร่วมในโครงการวิจัยนี้เมื่อใดก็ได้ และเข้าร่วมโครงการครั้งนี้โดยสมัครใจและการบอกเลิกการเข้าร่วมการวิจัยนี้ จะไม่มีผลต่อการรักษาโรคที่ข้าพเจ้าจะพึงได้รับต่อไป

คณะผู้ทำการศึกษารับรองว่าจะเก็บข้อมูลเฉพาะเกี่ยวกับข้าพเจ้าเป็นความลับ และจะเปิดเผยได้เฉพาะในรูปที่เป็นสรุปผลการวิจัย การเปิดเผยข้อมูลเกี่ยวกับข้าพเจ้าต่อหน่วยงานต่าง ๆ ที่เกี่ยวข้อง กระทำได้เฉพาะกรณีจำเป็นด้วยเหตุผลทางวิชาการเท่านั้น

ผู้ทำการศึกษาวิจัยรับรองว่าหากมีข้อมูลเพิ่มเติมที่ส่งผลกระทบต่อการวิจัย ข้าพเจ้าจะได้รับการแจ้งให้ทราบโดยไม่ปิดบังซ่อนเร้น

ข้าพเจ้าได้อ่านข้อความข้างต้นแล้ว และมีความเข้าใจดีทุกประการ และได้ลงนามในใบยินยอมนี้ด้วยความเต็มใจ

ลงนาม.....ผู้ให้คำยินยอม

ลงนาม.....พยาน

ลงนาม.....พยาน

APPENDIX E

General health questionnaire

APPENDIX E

General health questionnaire

แบบสอบถามและบันทึกข้อมูล

เลขที่.....

แบบสอบถาม

ชื่อผู้ให้ข้อมูล.....นามสกุล.....

บ้านเลขที่.....หมู่ที่.....บ้าน.....

ตำบล.....อำเภอ.....จังหวัด.....

ผู้สัมภาษณ์.....

วันที่..... / ..... / 2552

เวลาเริ่มสัมภาษณ์.....

เวลาสิ้นสุดสัมภาษณ์.....

ชุดแบบสอบถาม ประกอบด้วย

หมวดที่ 1 ข้อมูลส่วนบุคคล

หมวดที่ 2 การวัดภาวะพึ่งพาในกิจวัตรประจำวัน

หมวดที่ 3 สถานะสุขภาพ

หมวดที่ 4 พฤติกรรมเสี่ยง

หมวดที่ 1 ข้อมูลส่วนบุคคล

ส่วนที่ 1 ข้อมูลส่วนบุคคล

1. ชื่อ .....นามสกุล.....

2. วันเดือนปีเกิดของท่าน .....

3. อายุเต็มปีของผู้สูงอายุนับถึงวันที่เก็บข้อมูล ..... ปี

4. เพศ ☐ 1. ชาย ☐ 2. หญิง

5. สถานที่อยู่อาศัย ☐ 1. เขตเทศบาล ☐ 2. นอกเขตเทศบาล

6. สถานภาพสมรส ☐ 1. สมรส ☐ 2. โสด ☐ 3. หม้าย ☐ 4. หย่า/แยก

7. ศาสนา ☐ 1. พุทธ ☐ 2. คริสต์ ☐ 3. อิสลาม ☐4. อื่นๆ ระบุ.....

8. ระดับการศึกษา

☐ 1. ไม่ได้เรียน ☐ 2. ประถมศึกษา

☐ 3. มัธยมศึกษาตอนต้น ☐ 4. มัธยมศึกษาตอนปลาย / ปวช.

☐ 5. อนุปริญญา / ปวส. ☐ 6. ปริญญาตรี / สูงกว่า

ส่วนที่ 2 การทำงานและรายได้

1. ในระหว่าง 7 วันก่อนวันสัมภาษณ์ ท่านยังทำงานอยู่ใช่หรือไม่ (งานที่มีรายได้)

☐ 1. ใช่ ☐ 2. ไม่ใช่ (ข้ามไปถามข้อ 4)

2. <u>สำหรับผู้ที่ตอบว่าใช่ในข้อ 1</u> ท่านทำงานอะไร (อาชีพหรือตำแหน่งที่มีชั่วโมงการทำงานสูงสุด)

☐ 1. ค้าขาย/ธุรกิจส่วนตัว ☐ 2. รับจ้าง/ผู้ใช้แรงงาน

☐ 3. เกษตรกร/ทำสวน/ทำไร่ ☐ 4. ข้าราชการ/รัฐวิสาหกิจ

☐ 5. อื่นๆ (ระบุ).....

3. <u>สำหรับผู้ที่ตอบว่าใช่ในข้อ 1</u> เหตุผลสำคัญที่ท่านยังทำงานอยู่

3.1 ต้องการรายได้เลี้ยงครอบครัว/ตนเอง	☐1. ใช่	☐ 2. ไม่ใช่
3.2 สุขภาพแข็งแรง/ยังมีแรงทำงาน	☐1. ใช่	☐ 2. ไม่ใช่
3.3 เป็นอาชีพประจำ ไม่มีผู้ดูแลแทน	☐1. ใช่	☐ 2. ไม่ใช่
3.4 ใช้เวลาว่างให้เป็นประโยชน์	☐1. ใช่	☐ 2. ไม่ใช่
3.5 ช่วยบุตรและสมาชิกในครอบครัว	☐1. ใช่	☐ 2. ไม่ใช่
3.6 อื่นๆ (ระบุ).....	☐1. ใช่	☐ 2. ไม่ใช่

4. ใน 12 เดือนก่อนวันสัมภาษณ์ ท่านมีรายได้หรือทรัพย์สินในการเลี้ยงชีพตนเอง/ครอบครัวจากแหล่งต่อไปนี้ใช่หรือไม่ (ตอบได้มากกว่า 1 ข้อ)

4.1 งานประจำในปัจจุบัน	☐1. ใช่	☐ 2. ไม่ใช่
4.2 บำเหน็จ บำนาญ	☐1. ใช่	☐ 2. ไม่ใช่
4.3 เงินออม/ดอกเบี้ย/ทรัพย์สิน	☐1. ใช่	☐ 2. ไม่ใช่
4.4 เงินกองทุนเลี้ยงชีพ/เบี้ยยังชีพ	☐1. ใช่	☐ 2. ไม่ใช่
4.5 บุคคลอื่นให้ (บุตร/หลาน)	☐1. ใช่	☐ 2. ไม่ใช่
4.6 เงินสงเคราะห์	☐1. ใช่	☐ 2. ไม่ใช่
4.7 คู่สมรส	☐1. ใช่	☐ 2. ไม่ใช่
4.8 อื่นๆ (ระบุ)	☐1. ใช่	☐ 2. ไม่ใช่

5. ท่านมีรายได้จากทุกแหล่งในการเลี้ยงชีพตนเองหรือไม่

☐1. มีรายได้ ☐ 2. ไม่มีรายได้ (ข้ามไปถามข้อ 9)

6. สำหรับผู้ที่ตอบว่ามีรายได้ในข้อ 5 ท่านมีรายได้จากทุกแหล่งรวมกันประมาณปีละ.....บาท

7. ท่านคิดว่ารายได้ทั้งหมดที่ท่านได้รับจากทุกแหล่งพอเพียงหรือไม่

☐ 1. เกินพอเพียง ☐ 2. พอเพียง

☐ 3. เพียงพอบางครั้ง ☐ 4. ไม่เพียงพอ

8. ท่านหรือสมาชิกในครอบครัวมีหนี้สินหรือไม่

☐ 1. ตนเองมีหนี้ ☐ 2. สมาชิกในครอบครัวมีหนี้

☐ 3. ตนเองและสมาชิกในครอบครัวไม่มีหนี้

9. นอกจากเรื่องรายได้ ท่านมีความขัดสนดังต่อไปนี้ใช่หรือไม่ (ตอบได้มากกว่า 1 ข้อ)

9.1 อาหารการกิน	☐1. ใช่	☐ 2. ไม่ใช่
9.2 เสื้อผ้า เครื่องนุ่งห่ม	☐1. ใช่	☐ 2. ไม่ใช่
9.3 ที่อยู่อาศัย	☐1. ใช่	☐ 2. ไม่ใช่
9.4 การรักษาพยาบาลเมื่อเจ็บป่วย	☐1. ใช่	☐ 2. ไม่ใช่
9.5 อื่น ๆ (ระบุ)	☐1. ใช่	☐ 2. ไม่ใช่

หมวดที่ 2 การวัดภาวะพึ่งพาในกิจวัตรประจำวัน

คำชี้แจง : การวัดภาวะพึ่งพาในกิจวัตรประจำวัน การทำหน้าที่ ความจำกัดในการทำหน้าที่และกิจกรรมทางสังคมเป็นการวัดว่า ผู้ให้สัมภาษณ์ทำอะไรได้บ้าง ที่ทำได้จริง ๆ ในระยะเวลา 24-48 ชั่วโมงก่อนวันสัมภาษณ์

1. ท่านสามารถทำกิจกรรมต่อไปนี้ได้หรือไม่ (Activities of daily livings-ADL)

การดูแลตนเอง	ทำได้ด้วยตนเอง (2)	ต้องมีผู้อื่นช่วย (1)	ทำไม่ได้เลย (0)
1.1 อาบน้ำ/ล้างหน้า	☐	☐	☐
1.2 แต่งตัว	☐	☐	☐
1.3 กินอาหาร	☐	☐	☐
1.4 ลุกนั่งจากที่นอนหรือเตียง	☐	☐	☐
1.5 การใช้ห้องน้ำ/ส้วม	☐	☐	☐
1.6 เดินไปเดินมาภายในบ้าน	☐	☐	☐
1.7 กลั้นปัสสาวะ	☐	☐	☐
1.8 กลั้นอุจจาระได้	☐	☐	☐

2. ท่านสามารถทำกิจกรรมต่อไปนี้ด้วยตนเองได้หรือไม่ (Instrumental Activities of daily livings-ADL)

การทำงานบ้าน	ทำได้ด้วยตนเอง (2)	ต้องมีผู้อื่นช่วย (1)	ทำไม่ได้เลย (0)
2.1 ใช้เงิน นับเงิน ทอนเงิน	☐	☐	☐
2.2 จัดยากินเอง	☐	☐	☐
2.3 งานบ้านอย่างเบา (กวาดบ้าน/เก็บของ/ทำครัว)	☐	☐	☐
2.4 งานบ้านอย่างหนัก (ถูบ้าน/ตักน้ำ/ยกของ)	☐	☐	☐
2.5 ตัดเล็บเท้า	☐	☐	☐

3. ท่านมีปัญหาหรือความลำบากในการทำกิจกรรมต่อไปนี้เพียงใด (Functional limitations)

ความจำกัดในการทำหน้าที่	ไม่มีปัญหา (2)	มีปัญหาบ้าง (1)	ทำไม่ได้เลย (0)
3.1 เดินออกนอกบ้าน >15 นาทีในแต่ละครั้ง	☐	☐	☐
3.2 หิ้วของหนัก เช่น ไปตลาดได้ไกล 100 เมตร	☐	☐	☐
3.3 เดินขึ้นลงบันไดประมาณ 10 ขั้นโดยไม่พัก	☐	☐	☐
3.4 เดินไกลอย่างน้อย 400 เมตร	☐	☐	☐
3.5 ขึ้นรถโดยสารสาธารณะ	☐	☐	☐

4. ท่านมีปัญหาหรือความลำบากในการทำกิจกรรมต่อไปนี้หรือไม่ (Social communications)

การสื่อสารกับภายนอก	ไม่มีปัญหา (2)	มีปัญหาบ้าง (1)	ทำไม่ได้เลย (0)
4.1 การสื่อสารกับคนในครอบครัว	☐	☐	☐
4.2 การติดต่อสื่อสารกับเพื่อนบ้านในกรณีจำเป็น	☐	☐	☐
4.3 ใช้โทรศัพท์	☐	☐	☐

หมวดที่ 3 สถานะสุขภาพ

ส่วนที่ 1 สุขภาพโดยรวม

1. โดยทั่วไปท่านประเมินภาวะสุขภาพโดยรวมในวันนี้ของท่าน ให้อยู่ในระดับใด ขอให้ท่านพิจารณาทั้งภาวะสุขภาพร่างกายและภาวะสุขภาพจิตใจ

☐ 1. ดีมาก ☐ 2. ดี ☐ 3. ปานกลาง

☐ 4. ไม่ดี ☐ 5. ไม่ดีมาก

2. โดยรวมแล้วในช่วง 30 วันที่ผ่านมา ท่านคิดว่าภาวะสุขภาพของท่านดีกว่า หรือแย่กว่า เพื่อนในวัยเดียวกันขอให้ท่านพิจารณาทั้งภาวะสุขภาพร่างกายและภาวะสุขภาพจิตใจ

☐ 1. ดีมาก ☐ 2. ดี ☐ 3. ปานกลาง

☐ 4. ไม่ดี ☐ 5. ไม่ดีมาก

ส่วนที่ 2 สิทธิและสวัสดิการทางสุขภาพ

1. ในปัจจุบันนี้ท่านมีสวัสดิการหรือมีสิทธิในการรักษาพยาบาลหรือไม่ (ตอบได้มากกว่า 1 ข้อ)

1.1 บัตรทอง	☐1. ใช่	☐ 2. ไม่มี
1.2 ประกันสังคม	☐1. ใช่	☐ 2. ไม่มี
1.3 บัตรผู้สูงอายุ	☐1. ใช่	☐ 2. ไม่มี
1.4 สวัสดิการข้าราชการบำนาญ	☐1. ใช่	☐ 2. ไม่มี
1.5 สวัสดิการพนักงานรัฐวิสาหกิจ	☐1. ใช่	☐ 2. ไม่มี
1.6 ประกันเอกชน	☐1. ใช่ ระบุ	☐ 2. ไม่มี
1.7 กองทุน/สวัสดิการชุมชน	☐1. ใช่ ระบุ	☐ 2. ไม่มี
1.8 อื่น ๆ (ระบุ)	☐1. ใช่ ระบุ	☐ 2. ไม่มี

2. ในกรณีที่ท่านไม่มีสิทธิ์การักษาพยาบาลใด ๆ เมื่อท่านเจ็บป่วย ท่านใช้เงินเพื่อการรักษาพยาบาลจากแหล่งใด

☐ 1. เงินตนเอง ☐ 2. บุตร/หลาน/ญาติ ☐ 3. เพื่อน/เพื่อนบ้าน

☐ 4. การสงเคราะห์ ☐ 5. อื่น ๆ (ระบุ) .....

ส่วนที่ 3 สุขภาพจิต

1.ภาวะเครียด

1.1 ในช่วง 1 ปี ที่ผ่านมา ท่านมีความรู้สึก กระวนกระวาย หงุดหงิด รำคาญใจ ตึงเครียด นอนหลับยาก

	ไม่เคยเลย	บางครั้ง	หลายครั้ง	ตลอดเวลา
1.1 เนื่องจากปัญหาในที่ทำงาน หรือที่บ้านหรือไม่	☐	☐	☐	☐
1.2 ท่านมีความรู้สึกเครียดเช่นนี้จากที่ทำงานหรือไม่	☐	☐	☐	☐
1.3 ท่านมีความรู้สึกเครียดเช่นนี้จากที่บ้านหรือไม่	☐	☐	☐	☐

1.2 ท่านมีปัญหาความเครียดเกี่ยวกับเรื่องการเงินของท่านหรือไม่

☐ 1. ไม่มีเลย ☐ 2. มีบ้าง ☐ 3. มีความเครียดมาก

2.ภาวะซึมเศร้า

2.1 ในช่วง 2สัปดาห์ที่ผ่านมา ท่านมีอาการเหล่านี้บ่อยแค่ไหน

	ไม่มีเลย	เป็นบางวัน 1-7 วัน	เป็นบ่อย >7 วัน	เป็นทุกวัน
1.1 เบื่อ ไม่สนใจอยากทำอะไร	☐	☐	☐	☐
1.2 ไม่สบายใจ ซึมเศร้า ท้อแท้	☐	☐	☐	☐
1.3 หลับยาก หรือหลับ ๆ ตื่น ๆ หรือหลับมากไป	☐	☐	☐	☐
1.4 เหนื่อยง่าย หรือไม่ค่อยมีแรง	☐	☐	☐	☐
1.5 เบื่ออาหาร หรือกินมากเกินไป	☐	☐	☐	☐
1.6 รู้สึกไม่ดีกับตัวเอง คิดว่า ตัวเองล้มเหลว หรือทำให้ตนเองหรือครอบครัวผิดหวัง	☐	☐	☐	☐
1.7 สมาธิไม่ดีเวลาทำอะไร เช่น ดูโทรทัศน์ ฟังวิทยุ หรือทำงานที่ต้องใช้ความตั้งใจ	☐	☐	☐	☐
1.8 พูดช้า ทำอะไรช้าลงจนคนอื่นสังเกตเห็นได้ หรือกระสับกระส่าย ไม่สามารถอยู่นิ่งได้เหมือนเคยเป็น	☐	☐	☐	☐
1.9 คิดทำร้ายตนเอง หรือคิดว่าถ้าตายไปคงจะดี	☐	☐	☐	☐

ส่วนที่ 4 โรคและภาวะสุขภาพ

1. การตรวจสุขภาพประจำปี

☐ 1. ไม่เคยตรวจ ☐ 2. เคยตรวจ 2-3 ปีที่แล้ว

☐ 3. เคยตรวจแต่นานเกิน 5 ปี ☐ 4. ตรวจทุกปี

2. หากไม่สบายท่านมักจะ (ตอบได้มากกว่า 1 ข้อ)

☐ 1. ซื้อยากินเอง ☐ 2. ไปสถานบริการใกล้บ้าน/สถานีอนามัย

☐ 3. ไปคลินิก ☐ 4. ไปโรงพยาบาลของรัฐ

☐ 5. อื่นๆ ระบุ.....

ถ้าท่านมีโรคประจำตัว โปรดตอบคำถามต่อไปนี้

1. เบาหวาน

1.1 ในช่วง 12 เดือนที่ผ่านมา ท่านเคยได้รับการตรวจวัดระดับน้ำตาลในเลือด รวมทั้งตรวจด้วยตนเอง หรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

1.2 ท่านทราบค่าระดับน้ำตาลในเลือดที่ตรวจครั้งสุดท้ายของท่านหรือไม่

☐ 1. ทราบ ระบุ ..... มก/ดล. ☐ 1. ไม่ทราบ

1.3 ในช่วง 12 เดือนที่ผ่านมา ท่านเคยได้รับการบอกกล่าวจากบุคลากรทางด้านสาธารณสุข/แพทย์ว่าท่านเป็นเบาหวานใช่หรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่ (ข้ามไปถามข้อ 2.1)

1.4 ท่านทรายว่าตนเองเป็นโรคเบาหวานเมื่ออายุเท่าไร.....ปี

1.5 ขณะนี้ท่านได้รับการรักษาโรคเบาหวานจากบุคลากรทางด้านสาธารณสุขหรือไม่

☐ 1. ได้รับ ☐ 2. ไม่ได้รับ

1.6 ท่านรักษาโรคเบาหวานด้วยยาแผนปัจจุบันมานานกี่ปีแล้ว.....ปี

1.7 ท่านได้รับยารักษาโรคเบาหวาน โดยได้ยา Insulin ใน 2 สัปดาห์ที่ผ่านมาหรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

1.8 ท่านได้กินยารักษาโรคเบาหวานใน 2 สัปดาห์ที่ผ่านมาหรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

1.9 ในช่วง 2 สัปดาห์ที่ผ่านมา ท่านได้รับยารักษาโรคเบาหวานสม่ำเสมอหรือไม่ (รวมการรักษาเบาหวานทุกชนิด)

☐1. ใช่ ☐ 2. ไม่ใช่

1.10 ในรอบ 1 ปีที่ผ่านมา ท่านรับยาจากที่ใด

1.10.1 โรงพยาบาลรัฐ	☐1. ได้รับ	☐ 2. ไม่ได้รับ
1.10.2 โรงพยาบาลเอกชน/คลินิก/โพลีคลินิก	☐1. ได้รับ	☐ 2. ไม่ได้รับ
1.10.3 สถานีอนามัย/ศูนย์บริการสาธารณสุขชุมชน	☐1. ได้รับ	☐ 2. ไม่ได้รับ
1.10.4 ร้านยา	☐1. ได้รับ	☐ 2. ไม่ได้รับ
1.10.5 เพื่อน ญาติ คนรู้จัก สามี/ภรรยา	☐1. ได้รับ	☐ 2. ไม่ได้รับ

2. ความดันโลหิต

2.1 ท่านได้รับการวัดความดันโลหิตโดยบุคลากรทางด้านสาธารณสุขครั้งสุดท้ายเมื่อใด

☐ 1. ในช่วง 12 เดือน ☐ 2. ระหว่าง 1- 5 ปี

☐ 3. มากกว่า 5 ปี ☐ 4. ไม่เคยได้รับการวัด

2.2 ท่านทราบค่าความดันโลหิตของท่านหรือไม่

☐ 1. ทราบระบุ ..... มม.ปรอท ☐ 2. ไม่ทราบ

2.3 <u>ในช่วง 12 เดือนที่ผ่านมา</u> ท่านเคยได้รับการบอกกล่าวจากบุคลากรทางด้านสาธารณสุข/แพทย์ว่าท่านเป็นโรคความดันโลหิตสูงใช่หรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่ (ข้ามไปถามข้อ 2.1)

ขณะนี้ท่านได้รับการรักษาโรคความดันโลหิตสูงจากบุคลากรทางด้านสาธารณสุขตามวิธีการต่อไปนี้หรือไม่

2.4 ยารักษาโรคความดันโลหิตสูงใน 2 สัปดาห์ที่ผ่านมา

☐1. ใช่ ☐ 2. ไม่ใช่

2.5 ในช่วง 2 สัปดาห์ที่ผ่านมา ท่านรับประทานยาสม่ำเสมอหรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

2.6 ท่านรักษาโรคความดันโลหิตสูงด้วยยาลดความดันโลหิตมานานเท่าไหร่.....ปี.....เดือน

2.7 ในรอบ 1 ปีที่ผ่านมา ท่านรับยาจากที่ใด

2.7.1 โรงพยาบาลรัฐ	☐1. ได้รับ	☐ 2. ไม่ได้รับ
2.7.2 โรงพยาบาลเอกชน/คลินิก/โพลีคลินิก	☐1. ได้รับ	☐ 2. ไม่ได้รับ
2.7.3 สถานีอนามัย/ศูนย์บริการสาธารณสุขชุมชน	☐1. ได้รับ	☐ 2. ไม่ได้รับ
2.7.4 ร้านยา	☐1. ได้รับ	☐ 2. ไม่ได้รับ
2.7.5 เพื่อน ญาติ คนรู้จัก สามี/ภรรยา	☐1. ได้รับ	☐ 2. ไม่ได้รับ

3. ไขมันในเลือดสูง (คอเลสเตอรอล)

3.1 ท่านได้รับการตรวจวัดระดับไขมันในเลือดโดยบุคลากรทางด้านสาธารณสุขครั้งสุดท้ายเมื่อใด

☐ 1. ในช่วง 12 เดือน ☐ 2. ระหว่าง 1- 5 ปี

☐ 3. มากกว่า 5 ปี ☐ 4. ไม่เคยได้รับการวัด

3.2 ในช่วง 12 เดือนที่ผ่านมา ท่านเคยได้รับการบอกกล่าวจากบุคลากรทางด้านสาธารณสุข/แพทย์ว่าท่านมีปัญหาเกี่ยวกับระดับไขมันในเลือดใช่หรือไม่ (ไขมันตัวใดตัวหนึ่ง)

☐1. ใช่ ☐ 2. ไม่ใช่

3.3 <u>ในช่วง 30 วันที่ผ่านมา</u> ท่านรับประทานยาเพื่อรักษาระดับไขมันในเลือด หรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

3.4 ในรอบ 1 ปีที่ผ่านมา ท่านรับยาจากที่ใด

3.4.1 โรงพยาบาลรัฐ	☐1. ได้รับ	☐ 2. ไม่ได้รับ
3.4.2 โรงพยาบาลเอกชน/คลินิก/โพลีคลินิก	☐1. ได้รับ	☐ 2. ไม่ได้รับ
3.4.3 สถานีอนามัย/ศูนย์บริการสาธารณสุขชุมชน	☐1. ได้รับ	☐ 2. ไม่ได้รับ
3.4.4 ร้านยา	☐1. ได้รับ	☐ 2. ไม่ได้รับ
3.4.5 เพื่อน ญาติ คนรู้จัก สามี/ภรรยา	☐1. ได้รับ	☐ 2. ไม่ได้รับ

4. โรคหลอดเลือดหัวใจ

4.1 ท่านเคยได้รับการวินิจฉัยจากแพทย์ว่าเป็นโรคหัวใจหรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

4.2 ท่านเคยนอนโรงพยาบาลเพราะเจ็บหน้าอกหรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

4.3 แพทย์เคยวินิจฉัยว่าท่านเป็นโรคหลอดเลือดหัวใจ, กล้ามเนื้อหัวใจตาย ใช่หรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

4.4 ท่านเคยได้รับการฉีดสีเข้าหลอดเลือดหัวใจหรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

4.5 ท่านเคยได้รับการถ่างหลอดเลือดหัวใจด้วย Balloon/ขดลวด/ผ่าตัด หรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

4.6 ขณะนี้ท่านยังได้รับการรักษาโรคหลอดเลือดหัวใจหรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

5. โรคข้อเสื่อม

5.1 ท่านเคยมีปัญหาเรื่องข้อ หรือปวดข้อ ใช่หรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

5.2 <u>ในช่วง 6 เดือนที่ผ่านมา</u> ท่านมีอาการปวดที่ข้อใด

☐1. ข้อเข่า ☐ 2. ข้อนิ้วมือ และ/หรือมือ

☐ 3. อื่น ๆ (ระบุ).....

5.3 ท่านมีอาการเคลื่อนไหวข้อลำบากในกรณีเหล่านี้ ใช่หรือไม่

5.3.1 หลังตื่นนอนใหม่ ๆ (เป็นเวลาประมาณ 30 นาที)

☐1. ใช่ ☐ 2. ไม่ใช่

5.3.2 เมื่อเวลาเคลื่อนไหวหลังจากอยู่ในท่าใดท่าหนึ่งนาน ๆ

☐1. ใช่ ☐ 2. ไม่ใช่

5.4 ท่านเคยได้รับการวินิจฉัยจากแพทย์ว่าเป็นโรคข้อเสื่อม ใช่หรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

5.5 ท่านต้องกินยาเป็นประจำ ใช่หรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

6. โรคไตเรื้อรัง

6.1 ท่านเคยได้รับการตรวจหรือรักษาโรคไตมากกว่า 2 ครั้งต่อปี ติดต่อกันอย่างน้อย 2 ปี ใช่หรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

6.2 ท่านเคยได้รับการวินิจฉัยจากแพทย์ว่าเป็นโรคไตเรื้อรัง ใช่หรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

หมวดที่ 4 พฤติกรรมเสี่ยง

ส่วนที่ 1 กิจกรรมทางกาย

1. ในช่วง 6 เดือนที่ผ่านมา ท่านออกกำลังกายหรือมีกิจกรรมทางกายหรือไม่

☐1. ใช่ ☐ 2. ไม่ใช่

2. การออกกำลังกาย/กิจกรรมทางกาย ที่ท่านปฏิบัติประจำ

☐ 2.1 วิ่งเหยาะๆ ☐ 2.2 ไทเก็ก มวยจีน
☐ 2.3 เดินเร็วๆ หรือเดินไกลๆ ☐ 2.4 เต้นแอโรบิก เต้นรำ
☐ 2.5 บริหารร่างกาย โดยวิธี (ระบุ).....
☐ 2.6 เล่นกีฬา (ระบุ).....
☐ 2.7 อื่น ๆ.....

3. ท่านออกกำลังกายอย่างน้อยสัปดาห์ละกี่วัน

☐ 1. ไม่ออกกำลังกายหรือออกกำลังกายน้อยกว่า 1 ครั้งต่อเดือน
☐ 2. ออกกำลังกาย 1-3 ครั้ง/เดือน
☐ 3. ออกกำลังกายสัปดาห์ละ 1 ครั้ง
☐ 4. ออกกำลังกายสัปดาห์ละ 3 ครั้ง

4. เวลาที่ใช้ในการออกกำลังกายแบบต่อเนื่องนานครั้งละ ประมาณเท่าไร

☐ 1. น้อยกว่า 15 นาที ☐ 2. 15-30 นาที ☐ 3. มากกว่า 30 นาที

ส่วนที่ 2 การสูบบุหรี่และดื่มแอลกอฮอล์

1. การสูบบุหรี่

☐ 1. ไม่สูบบุหรี่เลย
☐ 2. เคยสูบ แต่หยุดแล้วประมาณ.....ปี
☐ 3. ยังสูบบุหรี่ ประมาณ.....มวน/วัน สูบมาแล้ว.....ปี ชนิดของบุหรี่(ระบุ).....

2. พฤติกรรมการดื่มสุรา, เบียร์, ยาดองเหล้า

☐ 1. ไม่เคยดื่ม
☐ 2. เคยดื่ม แต่หยุดแล้วประมาณ.....ปี
☐ 3. ดื่มเป็นบางครั้งจนถึงปัจจุบัน ดื่มมาแล้ว.....ปี
☐ 4. ดื่มเป็นประจำประมาณ.....แก้ว/วัน ดื่มมาแล้ว.....ปี

3. การได้รับคาเฟอีน การดื่มชา ชาเขียว กาแฟ น้ำอัดลมสีดำ เครื่องดื่มชูกำลัง

☐ 1. ไม่เคยดื่ม

☐ 2. ดื่มเป็นบางครั้งจนถึงปัจจุบัน ดื่มมาแล้ว.....ปี

☐ 3. ดื่มเป็นประจำประมาณ.....แก้ว/วัน ดื่มมาแล้ว.....ปี

ส่วนที่ 3 การใช้ยาและอาหารเสริม

1. ใน 1 เดือนที่ผ่านมา ท่านได้ใช้ยาเหล่านี้หรือไม่ (ตอบได้มากกว่า 1 ข้อ)

☐ 1. ยาคลายเครียด/ยานอนหลับ

☐ 2. ยาปฏิชีวนะหรือยาฆ่าเชื้อแก้อักเสบ

☐ 3. ยาถ่าย /ระบาย

☐ 4. ยาขับปัสสาวะ

☐ 5. ยาระงับปวด

☐ 6. ยาแก้แพ้

☐ 7. ยาลดความดันโลหิต

☐ 8. ยาทางด้านจิตเวช

☐ 9. ยาลูกกลอน

☐ 10. ยาลดน้ำหนักหรือยาลดความอ้วน

☐ 11. ยาอื่นๆ(ระบุ).....

2. อาหารเสริม (ผลิตภัณฑ์เสริมอาหาร) และยาบำรุงที่รับประทานเป็นประจำ

☐ 1. ไม่มี

☐ 2. มี (ระบุ).....รับประทานมานาน.....ปี

3. ท่านได้รับยาและอาหารเสริม มาจากที่ใด

☐ 1. โรงพยาบาล/คลินิก

☐ 2. สถานีอนามัย/ศูนย์บริการสาธารณสุขชุมชน

☐ 3. ร้านยา ☐ 4. ร้านค้า

☐ 5. เพื่อน ญาติ คนรู้จัก ☐ 6. วัด

☐ 7. การขายตรง ☐ 8. อื่น ๆ (ระบุ).....

ส่วนที่ 4 อาหารและโภชนาการ

1. ท่านรับประทานอาหารหลัก วันละกี่มื้อ

☐ 1. มื้อเดียว ☐ 2. สองมื้อ ☐3. สามมื้อ

2. อาหารที่กินส่วนใหญ่

☐ 1.ทำกินเองที่บ้าน ☐ 2. ซื้อเข้ามากิน ☐ 3. กินนอกบ้าน

3. ที่บ้านท่านใช้น้ำมันชนิดไหนในการปรุงประกอบอาหารเป็นประจำ

☐ 1. น้ำมันปาล์ม

☐ 2. น้ำมันมะพร้าว

☐ 3. น้ำมันสัตว์ เช่น น้ำมันหมู

☐ 4. น้ำมันพืชประเภท น้ำมันถั่วเหลือง น้ำมันรำข้าว น้ำมันมะกอก น้ำมันดอกทานตะวัน น้ำมันดอกคำฝอย น้ำมันข้าวโพด

☐ 5. อื่นๆ ระบุ.....

4. ท่านชอบรับประทานอาหารรสใด

☐ 1. รสเค็ม ☐ 2. รสหวาน ☐ 3. รสเปรี้ยว

☐ 4. รสเผ็ด ☐ 5. อื่นๆ ระบุ.....

โดยเฉลี่ยท่านรับประทานอาหารประเภทต่าง ๆ มากน้อยเพียงใด

มากที่สุด	หมายถึง	มากกว่า 50%
มากพอประมาณ	หมายถึง	25-50%
ทานบ้างแต่น้อย	หมายถึง	น้อยกว่า 25%
ไม่ทาน	หมายถึง	หมายถึงไม่ทานเลย หรือน้อยกว่าเดือนละครั้ง

1. ประเภทเนื้อสัตว์ ได้แก่ หมู ไก่ เนื้อ กุ้ง ปลาหมึก (ยกเว้นปลา)

☐ 1. มากที่สุด ☐ 2. มากพอประมาณ ☐ 3. ทานบ้างแต่น้อย ☐ 4. ไม่ทาน

2. ข้าว ข้าวเหนียว อาหารที่ทำจากแป้ง ขนมหวาน

☐ 1. มากที่สุด ☐ 2. มากพอประมาณ ☐ 3. ทานบ้างแต่น้อย ☐ 4. ไม่ทาน

3. อาหารประเภทใส่กะทิ เช่นแกงเขียวหวาน กล้วยบวชชี ฯลฯ หรือ อาหารที่ทอดน้ำมันเยิ้ม เช่น หมูทอดกะเทียม กล้วยทอด ไก่ทอด ข้าวมัน ข้าวขาหมู

☐ 1. มากที่สุด ☐ 2. มากพอประมาณ ☐ 3. ทานบ้างแต่น้อย ☐ 4. ไม่ทาน

4. ประเภทผักสด ผลไม้สด น้ำผัก น้ำผลไม้

☐ 1. มากที่สุด ☐ 2. มากพอประมาณ ☐ 3. ทานบ้างแต่น้อย ☐ 4. ไม่ทาน

ผู้วิจัยขอขอบคุณท่านเป็นอย่างยิ่ง ที่ให้ความร่วมมือ

APPENDIX F

Medical assessment record form

APPENDIX F

Medical assessment record form

แบบบันทึกข้อมูล

เลขที่.....

แบบบันทึกข้อมูลการตรวจร่างกาย

ชื่อผู้ถูกตรวจร่างกาย.....นามสกุล.....

บ้านเลขที่.....หมู่ที่..... บ้าน.....

ตำบล.....อำเภอ.....จังหวัด.....

ผู้ตรวจร่างกาย.....

วันที่..... / ...../ 2552

เวลาเริ่มตรวจร่างกาย.....

เวลาสิ้นสุดตรวจร่างกาย.....

แบบบันทึกข้อมูล ประกอบด้วย

หมวดที่ 1 การตรวจร่างกาย

หมวดที่ 2 การตรวจทางห้องปฏิบัติการ

หมวดที่ 3 การตรวจสมรรถภาพร่างกาย

(ข้อมูลเหล่านี้จะถูกเก็บเป็นความลับ และใช้เฉพาะในโครงการวิจัยครั้งนี้เท่านั้น)

คำชี้แจง: ให้บันทึกข้อมูลที่ได้จากการทดสอบอย่างละเอียด ลงในแบบบันทึกข้อมูล

หมวดที่ 1 การตรวจร่างกาย

เลขที่.....
วันที่.....

บันทึกการตรวจร่างกายโดยแพทย์

(MEDICAL ASSESSMENT)

ชื่อ-นามสกุล..... อายุ.....ปี

ไม่เป็นโรคหรือมีอาการดังต่อไปนี้

- ☐ โรคหัวใจรุนแรงที่ต้องอาศัยยาควบคุม
- ☐ โรคเบาหวานชนิดที่ต้องพึ่งอินสุลิน
- ☐ มีความดันโลหิตในขณะพักสูงกว่า 140/90 มิลลิเมตรปรอท
- ☐ โรคผิวหนังที่สามารถติดต่อสู่ผู้อื่นได้
- ☐ ผู้พิการที่ไม่สามารถออกกำลังกายในน้ำได้
- ☐ ผู้ที่ไม่สามารถกลั้นปัสสาวะ/อุจจาระได้
- ☐ ผู้เป็นโรคปอดเรื้อรัง

ประวัติการเจ็บป่วยในอดีต (Past history)

.....

.....

.....

การตรวจร่างกายโดยแพทย์

General Appearance.....

Head, Ear, Nose, Throat.....

Heart/Lung.....

Abdominal cavity.....

Extremities.....

Nervous system.....

คำแนะนำ/ข้อเสนอแนะ (Comment) .....

.....

แพทย์ผู้ตรวจ

เกณฑ์วินิจฉัยโรคเบาหวาน (สำหรับแพทย์ตอบ)

ลำดับที่.....

วันที่ ..... เดือน ..... พ.ศ. .....

ผู้ป่วยชื่อ ..... นามสกุล .....

H.N. .....

1. เกณฑ์การวินิจฉัยโรคเบาหวานประเภทที่ 2 (ดัดแปลงจาก WHO & ADA)

ลักษณะทางคลินิกและผลตรวจทางห้องปฏิบัติการ (ค่าที่ตรวจได้ผิดปกติ จะต้องตรวจยืนยันซ้ำอีกครั้งด้วยวิธีใดวิธีหนึ่งในวันถัดมา ถ้าผิดปกติอีกจึงถือว่าเป็นเบาหวาน)

☐ 1. มีอาการของเบาหวาน (ปัสสาวะมาก, กินจุ และน้ำหนักตัวลดโดยไม่ทราบสาเหตุ) และตรวจกลูโคสในเวลาใดก็ได้ มีค่าตั้งแต่ 200 มิลลิกรัม/เดซิลิตร ขึ้นไป หรือ

☐ 2. ตรวจกลูโคสในพลาสมาขณะอดอาหารอย่างน้อย 8 ชั่วโมง (FPG) ได้ค่าตั้งแต่ 126 มิลลิกรัม/เดซิลิตร ขึ้นไป หรือ

☐ 3. ตรวจกลูโคสในพลาสมาหลังตรวจความทนของกลูโคส (OGTT; กลูโคส 75 กรัม [anhydrous glucose] ละลายน้ำแล้วดื่ม) 2 ชั่วโมง ได้ค่าตั้งแต่ 200 มิลลิกรัม/เดซิลิตร ขึ้นไป

2. การจำแนกชนิดของโรคเบาหวาน

☐ 1. ประเภทที่ 1 ☐ 2. ประเภทที่ 2 ☐ 3. ประเภทอื่นๆ (ระบุ).....

3. สาเหตุจาก

☐ A. Predominantly insulin resistance with relative insulin deficiency

☐ B. Predominantly insulin secretory defect with insulin resistance

☐ C. Diseases of the exocrine pancreas ☐ D. Endrocrinopathies

☐ E. Drug- or chemical-induced ☐ F. Infections

☐ G. Uncommon forms of immune-mediated diabetes

☐ H. Other genetic syndromes sometimes associated with diabetes

REFERENCE

1. The Expert Committee on the Diagnosis and Classification of Diabetes Mellitus. Report of the expert committee on the diagnosis and classification of diabetes mellitus. Diabetes Care 1998: 21 Suppl 1; S5-S19.

2. American Diabetes Association. Tests of glycemia in diabetes. Diabetes Care 1998: 21 Suppl 1; S69-S71.

3. Diagnosis and Classification of Diabetes Mellitus: New Criteria [บรรณาธิการ]. วารสารแพทย์หลังปริญญา 2542: 13; 19-24, 38-43.

แบบบันทึกข้อมูล

(นาย/นาง/นางสาว) ชื่อ..... นามสกุล.....อายุ.....ปี

เพศ ☐ ชาย ☐ หญิง

การวัดสัดส่วนร่างกาย (Body composition)

1. น้ำหนัก.....กิโลกรัม ส่วนสูง.....เซนติเมตร

2. ดัชนีมวลกาย (Body mass index; BMI) ..... (กิโลกรัม/เมตร^2)

☐ 1. น้ำหนักน้อย (น้อยกว่า 17.9 กก./ตรม.) ☐ 2. ปกติ (18-22.9 กก./ตรม.)
☐ 3. น้ำหนักเกิน (23-24.9 กก./ตรม.) ☐ 4. อ้วน (25-34.9 กก./ตรม.)
☐ 5. อ้วนรุนแรง (35 กก./ตรม.ขึ้นไป)

3. เส้นรอบเอว (Waist circumference) .....เซนติเมตร

เพศหญิง ☐ 1. ปกติ ☐ 2. ตั้งแต่ 80 เซนติเมตรขึ้นไป อ้วนลงพุง

เพศชาย ☐ 1. ปกติ ☐ 2. ตั้งแต่ 90 เซนติเมตรขึ้นไป อ้วนลงพุง

4. รอบเอว/สะโพก (Waist hip ratio) .....

☐ 1. ปกติ (0.7-0.8) ☐ 2. น้ำหนักเกิน (0.9-1.0) ☐ 3. อ้วน (> 1.0)

5. อัตราการเผาผลาญพลังงานขณะพัก (Basal metabolic rate; BMR) =.....Kcal

6. เปอร์เซ็นต์ไขมันในร่างกาย (% Body fat) =.....%

7. ไขมันในร่างกาย (Body fat mass) =..... kg.

8. เปอร์เซ็นต์น้ำในร่างกาย (% Total body water) =.....%

9. มวลกล้ามเนื้อ (Muscle mass) =..... kg.

10. มวลกล้ามเนื้อที่ปราศจากไขมัน (Fat free mass) =..... kg.

การวัดสัญญาณชีพ ขณะพัก (Vital sign: rest at sitting position 5 minute)

1. ชีพจร.....ครั้ง/นาที ☐ 1. ปกติ ☐ 2. ไม่สม่ำเสมอ

2. ความดันโลหิต.....มิลลิเมตร/ปรอท

☐ 1. ปกติ ☐ 2. ความดันโลหิตสูง ☐ 3. ความดันโลหิตต่ำ

3. อัตราการหายใจใน 1 นาที.....ครั้ง

☐ 1. ปกติ ☐ 2. เร็วกว่าปกติ ☐ 3. ช้ากว่าปกติ

4. ลักษณะการหายใจ

☐ 1.ปกติ ☐ 2.ค่อนข้างเหนื่อยเล็กน้อย ☐ 3.ค่อนข้างเหนื่อยมาก

หมวดที่ 2 การตรวจทางห้องปฏิบัติการ

ระดับน้ำตาลในเลือดและอินซูลิน

Fasting blood sugar = ... mg /dl

HbA1c = ...%

Insulin = ...%

ระดับไขมันในเลือด

Total cholesterol (TC) = ... mg %

Triglycerides (TG) = ... mg %

High-density lipoprotein cholesterol (HDL-C) = ... mg %

Low-density lipoprotein cholesterol (LDL-C) = ... mg %

Very low-density lipoprotein cholesterol (VLDL) = ... mg %

ตัวชี้วัดภาวะอักเสบและการทำหน้าที่ของเซลล์บุผนังหลอดเลือดขนาดเล็ก

MDA = ...

CRP = ...

Maximal postocclusive reactive hyperemia ($PORH_{max}$)= .....

Peak postocclusive reactive hyperemia ($PORH_{peak}$) = ...

Percent postocclusive reactive hyperemia $PORH_{\%}$ = ...

หมวดที่ 3 การทดสอบสมรรถภาพร่างกาย

1. แรงบีบมือ (กิโลกรัม) มือข้างที่ถนัด ☐ 1. ซ้าย ☐ 2. ขวา

ครั้งที่	แรงบีบมือข้างขวา (กิโลกรัม)	แรงบีบมือข้างซ้าย (กิโลกรัม)
1		
2		
3		

เพศหญิง (อายุ 50-59 ปี) ☐ 29 ขึ้นไป ดีมาก ☐ 27-29 ดี ☐ 23-27 ปานกลาง ☐ 21-23 ต่ำ ☐ 20 ลงมา ต่ำมาก

เพศหญิง (อายุ 60-72 ปี) ☐ 26 ขึ้นไป ดีมาก ☐ 24-26 ดี ☐ 19-24 ปานกลาง ☐ 16-19 ต่ำ ☐ 16 ลงมา ต่ำมาก

เพศชาย (อายุ 50-59 ปี) ☐ 47 ขึ้นไป ดีมาก ☐ 43-46 ดี ☐ 37-43 ปานกลาง ☐ 34-37 ต่ำ ☐ 34 ลงมา ต่ำมาก

เพศชาย (อายุ 60-72 ปี) ☐ 40 ขึ้นไป ดีมาก ☐ 37-40 ดี ☐ 30-37 ปานกลาง ☐ 26-30 ต่ำ ☐ 26 ลงมา ต่ำมาก

2. แรงเหยียดขาและหลัง (กิโลกรัม)

ครั้งที่	แรงเหยียดขา (กิโลกรัม)	แรงเหยียดหลัง (กิโลกรัม)
1		
2		
3		

เพศหญิง (อายุ 50-59 ปี) ☐ 73 ขึ้นไป ดีมาก ☐ 66-72 ดี ☐ 50-65 ปานกลาง ☐ 43-49 ต่ำ ☐ 42 ลงมา ต่ำมาก

เพศหญิง (อายุ 60-72 ปี) ☐ 57 ขึ้นไป ดีมาก ☐ 50-56 ดี ☐ 35-49 ปานกลาง ☐ 28-34 ต่ำ ☐ 27 ลงมา ต่ำมาก

เพศชาย (อายุ 50-59 ปี) ☐ 144 ขึ้นไป ดีมาก ☐ 132-143 ดี ☐ 107-131 ปานกลาง ☐ 95-106 ต่ำ ☐ 94 ลงมา ต่ำมาก

เพศชาย (อายุ 60-72 ปี) ☐ 106 ขึ้นไป ดีมาก ☐ 94-105 ดี ☐ 69-93 ปานกลาง ☐ 57-68 ต่ำ ☐ 56 ลงมา ต่ำมาก

3. ความอ่อนตัว (เซนติเมตร)

ครั้งที่	ระยะทางที่ทำได้ (เซนติเมตร)
1	
2	
3	

เพศหญิง (อายุ 50-59 ปี)	☐ 18 ขึ้นไป ดีมาก	☐ 15-17 ดี	☐ 8-14 ปานกลาง
	☐ 5-7 ต่ำ	☐ 4 ลงมา ต่ำมาก	
เพศหญิง (อายุ 60-72 ปี)	☐ 18 ขึ้นไป ดีมาก	☐ 15-17 ดี	☐ 8-14 ปานกลาง
	☐ 5-7 ต่ำ	☐ 4 ลงมา ต่ำมาก	
เพศชาย (อายุ 50-59 ปี)	☐ 17 ขึ้นไป ดีมาก	☐ 13-16 ดี	☐ 4-12 ปานกลาง
	☐ 0-3 ต่ำ	☐ (-1) ลงมา ต่ำมาก	
เพศชาย (อายุ 60-72 ปี)	☐ 14 ขึ้นไป ดีมาก	☐ 10-13 ดี	☐ 2-9 ปานกลาง
	☐ (-2) - 1 ต่ำ	☐ (-3) ลงมา ต่ำมาก	

4. อัตราการเต้นของชีพจรขณะออกกำลังกาย โดยเครื่องวัดและติดตามการเต้นของหัวใจ (Polar monitor heart rate)

ประเมินครั้งที่ 1 2 3 4 5

ช่วงอุ่นเครื่อง (5 นาที หลังจากเริ่มออกกำลังกาย).....ครั้ง / นาที

ช่วงออกกำลัง (20 นาที หลังจากเริ่มออกกำลังกาย).....ครั้ง / นาที

ช่วงเบาเครื่อง (หลังจากออกกำลังกายเสร็จทันที) .....ครั้ง / นาที

5. อัตราการใช้ออกซิเจนสูงสุด (VO_{2max}) .....ml / kg / min

APPENDIX G

Hemodynamic and health-related physical fitness assessment

APPENDIX G

Hemodynamic and health-related physical fitness assessment

Hemodynamic assessment

-Resting heart rate

The participants were sitting at least 5 minutes for resting period prior to the measurement. The resting heart rate was measured with heart rate monitor (Sport tester PE 3000, Finland)

-Resting blood pressure

The participants were sitting at least 5 minutes for resting period prior to the measurement. The blood pressure was measured with digital blood pressure (Omron, Japan). The systolic blood pressure and diastolic blood pressure were recorded in unit of millimeters of mercury (mmHg).

Health-related physical fitness assessment

-Body composition

Fat mass, body fat, muscle mass and waist-to-hip ratio, was performed by using bioelectrical impedance analysis (BIA-101 impedance analyzers, USA).

-Muscle strength

Muscle strength was performed with isometric strength test of leg strength and hand grip strength (Takei kiki, Japan), both tests were repeated three times and the best value was recorded. Before measuring, every strength dynamometers were taken calibration procedures. All subjects were asking to perform with maximum efforts. Verbal command was encouraged throughout the experiment to ensure each subject's maximal effort.

-Flexibility

The sit and reach test is a common measure of flexibility, and specifically measures the flexibility of the lower back and hamstring muscles. The measurement is to use the level of the feet as recording zero, so that any measure that does not reach the toes is negative and any reach past the toes is positive.

-Cardiovascular and respiratory fitness

All participants performed walking on treadmill (Quinton, USA). Maximal O_2 consumption was assess by Modified Bruce protocol in which the grade and intensity

were increased every 3 minutes until exhaustion. Oxygen consumption was measured with the cardiopulmonary gas exchange system (Vmax 29, USA) throughout the exercise test. Respiratory exchange ratio (RER) values 1.05 were used to determine maximal O_2 consumption.

End point criteria to stop testing: the attainment of VO2 max was validated if two of the following four criteria were satisfied: (1) oxygen uptake plateau despite increasing exercise intensity (≤120 mL·min-1); (2) respiratory exchange ratio ≥1.15; (3) maximal heart rate within 10 beats·min-1 of the age-predicted maximal value; and (4) a Borg scale value ≥17.

Cardiopulmonary gas exchange system

Stage	Time	Speed	Elevation	METS
1	3	1.7	0%	1.7
2	3	1.7	5%	2.8
3	3	1.7	10%	5.4
4	3	2.5	12%	7
5	3	3.4	14%	10
6	3	4.2	16%	13
7	3	5.0	18%	17

Modified Bruce protocol

APPENDIX H

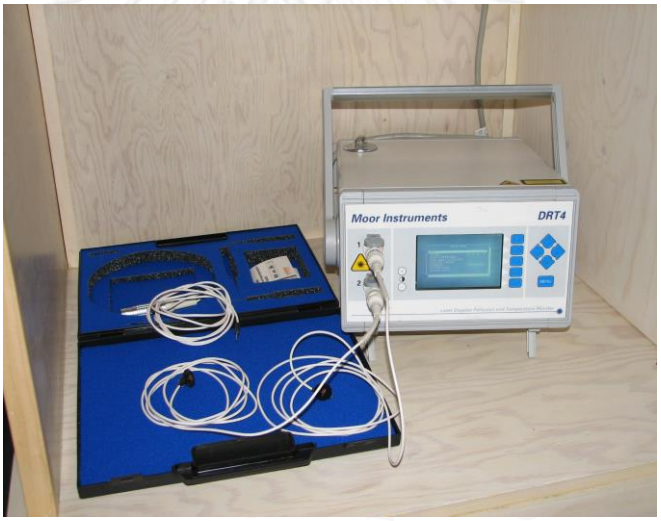
Cutaneous microvascular reactivity assessment

APPENDIX H

Cutaneous microvascular reactivity assessment

Cutaneous blood flow assessment

Cutaneous blood flow study was performed on all participants on the right wrist with a laser Doppler flowmetry (DRT4 MoorLAB, Moor Instrument, UK), using the post-occlusive reactive hyperemic method. All participants rested in the supine position for 20 minutes. Baseline data was monitored for 1 min and then placed the cuff around the right upper arm, inflated rapidly to 200 mmHg for 5 minutes and deflated for 5 minutes of recovery (Betik et al. 2004). Blood flow data at baseline and after deflated cuff at maximal blood flow were collected.



Laser Doppler flowmetry

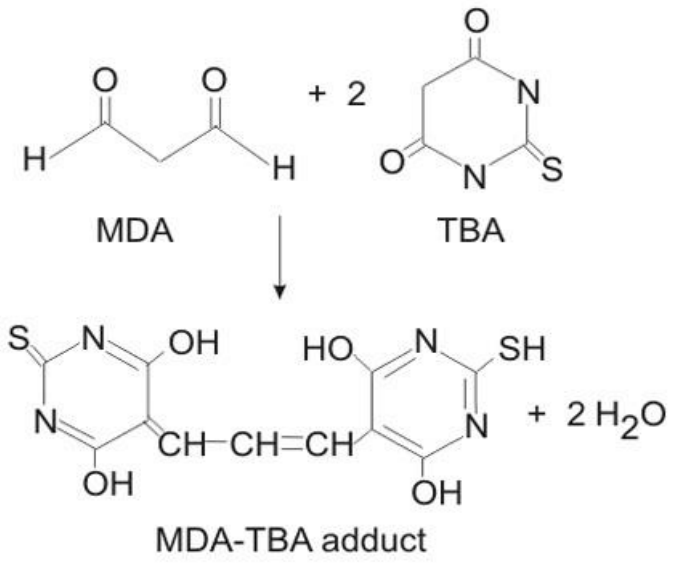
APPENDIX I

Malondialdehyde

APPENDIX I

Malondialdehyde

Malondiadehyde (MDA) is a ROS which is derived from the lipid oxidation of cell membrane polyunsaturated fatty acid and its formular is CH_2 $(CHO)_2$. This compound is highly reactive and toxic and is used as a biomarker to measure the level of oxidative stress. Thiobarbituric acid reactive substances method is used for analyzing level of MDA (Nanhini TA and Anuradha CV, 2003), by the concept that one molecule of MDA can react with 2 molecules of thiobarbituric acid (TBA). The chemical reaction and the method of measurement are described as follows:



MDA-TBA adduct

Reagents

1. Phosphate buffer saline (PBS) pH 7.4

- Na_2HPO_4 2.27 g, NaH_2PO_4 0.12 g and NaCl 8, 18 g are dissolved in distilled water until total volume of solution is 1,000 ml. The solution is adjusted to pH 7.4.

2. 30% Trichloroacetic acid (TCA)

- Dissolve tricholoacetic acid 30 g in distilled water until total volume of solution is 100 ml

3. Butylated hydroxyl toluene (BHT)

- Dissolve Butylated hydroxyl toluene 88 mg in ethanol 10 ml

4. 1% thiobarbituric acid (TBA)

- One g of 2-thiobarbituric acid in 100 ml distilled water

5. Malondialdehyde bis (diethylaceta) is used as external standard.

Samples

Use EDTA whole blood samples.

Sample preparation

Centrifuge whole blood for 10 minutes at 4 °C, 3500 rpm and then separate the plasma and erythrocytes. The erythrocytes are washed four times with 3 ml of 0.85% normal saline; centrifuging for 10 minutes at 4 °C, 3000 rpm after each wash.

Measurement of MDA; the sequences of the test are:

1. Draw 200 μl of each of washed erythrocytes, plasma, and diethylacetal (external standard) in three separate eppendorf tubes.

2. In each tube, add 800 μl of PBS, mix, and then add 30% TCA for erythrocyte lysis and protein precipitation

3. Add 25μl of BHT, mix, and then put in -20 °C refrigerator for 2 hours for the completeness of erythrocyte lysis and protein precipitation

4. Centrifuge at 12,000 rpm for 10 minutes.

5. Separate the supernatant into the new eppendorf tubes.

6. Add 250 μl of 1% TBA in each tube, mix, and boil in heat box (ACCUBLOCK™ Digital Dry Bath, Labnet International) at 103 °C for 15 minutes.

7. Stop the reaction by using cold water, draw 300μl of the solution and put in the 96-well-plate ELISA.

8. Measure light absorbance at 532 nm by using ELISA Reader (Enspire multilabel plate, Perkin-Elmer, USA)

9. Construct the concentration graft of the external standard, and then calculate the lipid oxidation of the samples from the standard curve.

VITA

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