 doi: 10.1111/cpf.12404

Metabolic responses to exercise on land and in water following glucose ingestion

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Accepted for publication
Received 8 September 2016;
accepted 1 November 2016

Key words
aquatic exercise; carbohydrate metabolism; exercise prescription; glycaemic control; postprandial hyperglycaemia

Summary

Although aerobic exercise after a meal decreases postprandial blood glucose, the differences in glucose response between land and aquatic exercise are unclear. Thus, we examined the effect of different modes of exercise with same energy expenditure following glucose ingestion on carbohydrate metabolism. Ten healthy sedentary men (age, 22 ± 1 years) participated in this study. All subjects performed each of three exercise modes (cycling, walking and aquatic exercise) for 30 min after ingestion of a 75-g glucose solution with 1–2 weeks between trials. The exercise intensity was set at 40% of the maximum oxygen uptake that occurred during cycling. The velocity during walking and the target heart rate during aquatic exercise were predetermined in a pretest. The plasma glucose concentration at 30 min after exercise was significantly lower with aquatic exercise compared to that with cycling and walking ($P<0{\cdot}05$). However, there were no significant differences among the three exercise modes in respiratory exchange ratio. On the other hand, serum free fatty acid concentration with aquatic exercise was significantly higher at 120 min after exercise compared with that after walking ($P<0{\cdot}05$). These results suggest that aquatic exercise reduces postprandial blood glucose compared with both cycling and walking with the same energy expenditure. Aquatic exercise shows potential as an exercise prescription to prevent postprandial hyperglycaemia.

Introduction

In addition to a proper diet, physical exercise is an effective way to prevent and control type 2 diabetes mellitus (T2DM) as it maintains blood pressure, glycaemia and lipidaemia in the appropriate values (Lakka & Laaksonen, 2007; Colberg *et al.*, 2010). Several studies have reported that the postprandial blood glucose level in newly detected T2DM is a more important predictor for cardiovascular disease and death rate compared with fasting blood glucose (Hanefeld *et al.*, 1996; Decode Study Group the European Diabetes Epidemiology Group, 2001). Aerobic exercise performed in the postprandial condition decreases glucose concentration (de Lima *et al.*, 2015). Thus, exercise after a meal may be useful for the prevention and improvement in T2DM (Haxhi *et al.*, 2013).

Aerobic exercise is one of the exercises that help improve glycemic control, and walking, running, cycling and aquatic exercise are the most commonly used types of aerobic exercise (Lakka & Laaksonen, 2007; Colberg *et al.*, 2010; Cugusi *et al.*, 2015). However, the recruited muscle fibre type differs depending on the mode of aerobic exercise. Carter *et al.* (2000) reported that cycling may recruit a greater number of type II fibres than running at the same relative intensity possibly due to ischaemia of the legs caused by a higher intramuscular tension in cycling compared with running. Indeed, blood flow in the leg during cycling is lower compared with that during running (Matsui *et al.*, 1978). However, it is well known that as type I fibres possess a high degree of glucose transporter-4 (GLUT-4), the ability for glucose uptake is higher in type I than in type II fibres (Marette *et al.*, 1992; Gaster *et al.*, 2000, 2001). Hence, a greater glucose disposal has been observed during running compared with that during cycling at equal oxygen uptake ($\dot{V}O_2$) (Derman *et al.*, 1996; Tsintzas *et al.*, 2003).

Moreover, differences in land and aquatic exercise exist. As aquatic exercise receives buoyancy, surface electromyography (EMG) activities are smaller in water than exercise on land with equal $\dot{V}O_2$ (Wiesner *et al.*, 2010). The magnitude of the surface EMG is related to the force generation during muscle contraction (Moritani & Muro, 1987). Furthermore, the EMG of the tibialis anterior and gastrocnemius during running, which have a predominance of type II fibres, are clearly lower in water than on land at the same ratings of perceived exertion (RPE) (Masumoto *et al.*, 2009). Therefore, there is a possibility that more type I fibres are recruited during aquatic exercise

compared with exercise on land and, thus, may induce more glucose transport. We hypothesized that aquatic exercise would more effectively decrease postprandial glucose compared with land exercise. As aquatic exercise has a lower risk of orthopaedic injuries, it is frequently used as an exercise prescription for patients with obesity, low back pain, osteoarthritis, etc. (Hinman *et al.*, 2007; Wang *et al.*, 2007; Irandoust & Taheri, 2015).

Thus, we examined the effect of cycling, walking on land and aquatic exercise following glucose ingestion on metabolic responses with equal $\dot{V}O_2$ to find the most effective exercise mode that decreases postprandial hyperglycaemia.

Methods

Subjects

Ten young, healthy men (mean age, 22 ± 1 years; height, 173·4 ± 2·2 cm; body mass, 63·7 ± 2·2 kg; body mass index, 21·2 ± 1·3 kg m^{-2}) participated in this study. They were sedentary and were not taking any medications or supplements. The subjects were informed of the experimental procedure and possible risks, and their written informed consent was obtained. Furthermore, we abstained from explaining the physiological effect of the exercise protocol to the subjects so as not to influence their performance. The protocol complied with the Helsinki declaration and was approved by our local ethics committee.

Experimental procedures

Subjects visited the laboratory six times throughout the experimental period. During the first visit, the subject's maximal oxygen uptake ($\dot{V}O_{2max}$) was assessed using a graded exercise test on a cycle ergometer (Well Bike BE-360; Fukuda Denshi, Tokyo, Japan). Heart rate (HR) was assessed using a wireless monitor (FT1; Polar Electro, Kempele, Finland). The test began at 70 W and the load was increased progressively at 30-W increments every 2 min until exhaustion. The test was terminated when the subject failed to maintain the prescribed pedalling frequency of 70 rpm or reached the levelling off of $\dot{V}O_2$. The collected data were averaged over the last 30 s in each load. The exercise intensity at 40% $\dot{V}O_{2max}$ during cycling was determined using linear regression. During the second and third visits, the subjects performed preliminary experiments to determine the speed of walking and HR during aquatic exercise that matched the $\dot{V}O_2$ to 40% $\dot{V}O_{2max}$ during cycling.

During visits 4–6, all subjects participated in the following three trials, with each trial separated by at least 1 week: cycling, walking on a treadmill (WELL ROAD 200 E; Takei Scientific Instruments, Niigata, Japan) and aquatic exercise (a combination of stepping, kicking, jumping and punching) in a swimming pool. The cycling and walking on land were undertaken randomly in the first or second trial, and aquatic exercise was carried out in the third trial for all subjects due to the limited availability of the swimming pool. However, results were unlikely to be affected by the order of exercise mode, because all subjects preliminarily had experience with all three modes. We instructed the subjects to refrain from intense exercise and crapulous on the day before each trial. In addition, the subjects ate identical meals (breakfast, lunch and dinner) at the same time on the day before each trial. All trials were performed between 8:00 a.m. and noon following a 12-h overnight fast. The subjects visited the laboratory in the morning and rested for 30 min on a chair prior to the collection of baseline respiratory gas and blood samples. They then consumed a 75-g glucose solution (225 ml) and rested again for 30 min prior to the collection of the same samples, after which the exercise was performed for 30 min. Respiratory gas, HR and RPE were assessed prior to the end of exercise. Immediately after exercise, a blood sample was taken, and the subjects rested for 2 h during which respiratory gases and additional blood samples were collected at 30, 60, 90 and 120 min after exercise. They performed the aquatic exercise while looking at a wireless monitor in order to maintain the predetermined HR. The water level in the swimming pool was set at the xiphisternal level for all subjects. The room temperature was 24·7 ± 0·0°C and the water temperature in the swimming pool was 30·2 ± 0·2°C throughout the experimental period.

Respiratory gas analysis

Respiratory gases were collected and analysed using an automatic gas analyser (Aeromonitor AE310s; Minato Medical Science, Tokyo, Japan). We applied two different methods of gas sampling: the breath-by-breath method during the graded exercise test and the Douglas bag method during the three exercise trials. The Douglas bag method was used because for aquatic exercise in a swimming pool, it is difficult to measure $\dot{V}O_2$ using the breath-by-breath respiratory system. $\dot{V}O_2$, carbon dioxide production ($\dot{V}CO_2$) and expired minute ventilation ($\dot{V}E$) were averaged in each 5-min period (2-min period only during exercise) throughout the three trials. The respiratory exchange ratio (RER) was determined from the $\dot{V}O_2$ and $\dot{V}CO_2$ measurements. The O_2 and CO_2 sensors were calibrated appropriately before the start of the exercise. The coefficient of variation (CV) for $\dot{V}O_2$ between the breath-by-breath and Douglas bag methods during exercise was 3·6%.

Blood analysis

Serum and plasma samples were obtained by centrifugation and were stored at −80°C prior to analysis. Plasma glucose concentrations were analysed using an enzymatic method (Adams Glucose GA-1170; ARKRAY, Kyoto, Japan). The interassay and intraassay CVs were 0·6% and 1·0%, respectively. Serum insulin concentrations were measured using a chemiluminescent immunoassay method (Architect Insulin kit; Abbott Japan, Chiba, Japan). The interassay CV was 1·9%. Serum free fatty acid (FFA) concentrations were measured using an enzymatic method (NEFA-HRII; Wako Pure Chemical

Industries, Osaka, Japan). The interassay and intraassay CVs were 1·5% and 2·2%, respectively.

Statistical analysis

Data are expressed as mean ± SE. A two-way (time × mode) analysis of variance (ANOVA) with repeated measures was initially applied to determine interaction and main effects. When the ANOVA revealed a significant interaction or main effect, a Tukey test was performed for *post hoc* analyses to assess differences among the exercise modes and the temporal changes. A one-way ANOVA with repeated measures was applied to compare the HR and RPE. For all tests, a $P<0·05$ was considered to be significant.

Results

Baseline parameters

The average value of $\dot{V}O_{2max}$ across all subjects was 2·59 ± 0·09 l min^{-1} (41·1 ± 2·0 ml kg^{-1} min^{-1}). The average exercise intensity for each exercise mode was as follows: cycling, 71 ± 3 W and 41 ± 3% $\dot{V}O_2$max; walking, 5·5 ± 0·2 km h^{-1} and 40 ± 2% $\dot{V}O_2$max; and aquatic exercise, 108 ± 3 beats min^{-1} and 40 ± 3% $\dot{V}O_2$max. There were no significant differences among the modes at baseline.

Respiratory parameters, heart rate and ratings of perceived exertion

The respiratory parameters throughout the entire experimental period for each of the three exercise modes are given in Table 1. The $\dot{V}O_2$, $\dot{V}CO_2$ and $\dot{V}E$ during exercise were significantly increased in all modes compared to baseline ($P<0·05$). As expected, the $\dot{V}O_2$ during exercise in the three exercise modes was approximately the same (cycling, 1·05 ± 0·04 l min^{-1}; walking, 1·03 ± 0·03 l min^{-1}; aquatic exercise, 1·02 ± 0·05 l min^{-1}). In the cycling condition, the $\dot{V}O_2$ was higher at 30 and 120 min during recovery compared with the walking condition ($P<0·05$). For the $\dot{V}CO_2$ responses, the smallest values were observed in the walking condition at 30 min during recovery ($P<0·05$, versus cycling and aquatic exercise), and in aquatic exercise at 90 min during recovery ($P<0·05$, versus both land exercises). The $\dot{V}E$ during exercise was significantly higher in aquatic exercise than in cycling and walking ($P<0·05$). The RER significantly increased from baseline only at 60 min during recovery in the cycling condition ($P<0·05$), but no significant differences were observed among the three exercise modes. In addition, HR during waking (107 ± 2 beats min^{-1}) and aquatic exercise (108 ± 3 beats min^{-1}) was significantly lower than that during cycling (114 ± 3 beats min^{-1}, $P<0·05$). There were no significant differences in RPE among exercise modes (cycling, 12·3 ± 0·6; walking, 11·6 ± 0·4; aquatic exercise, 10·9 ± 0·6); however, the RPE in aquatic exercise tended to be lower than that in cycling ($P = 0·054$).

Blood parameters

The time course of plasma glucose concentration is depicted in Fig. 1a. The plasma glucose concentration in all exercise modes was significantly higher at 30 min after a glucose load and at 30 min after exercise than at baseline ($P<0·05$). Further, the plasma glucose concentration in both land exercise

Table 1 Changes in respiratory parameters throughout the experimental period.

		Rest			Recovery			
Variables	**Trials**	**Baseline**	**−30 min**	**Exercise**	**30 min**	**60 min**	**90 min**	**120 min**
$\dot{V}O_2$ (l min^{-1})	Cycling	0·23 ± 0·01	0·23 ± 0·01	1·05 ± 0·04*	0·24 ± 0·01‡	0·24 ± 0·01	0·23 ± 0·00	0·23 ± 0·01‡
	Walking	0·21 ± 0·00	0·22 ± 0·01	1·03 ± 0·03*	0·22 ± 0·01	0·23 ± 0·00	0·23 ± 0·00	0·21 ± 0·00
	Aquatic exercise	0·22 ± 0·00	0·23 ± 0·00	1·02 ± 0·05*	0·23 ± 0·01	0·23 ± 0·00	0·22 ± 0·00	0·22 ± 0·01
$\dot{V}CO_2$ (l min^{-1})	Cycling	0·19 ± 0·01	0·21 ± 0·01	0·99 ± 0·04*	0·22 ± 0·01	0·23 ± 0·02	0·22 ± 0·01	0·21 ± 0·01
	Walking	0·19 ± 0·01	0·19 ± 0·01	0·94 ± 0·03*	0·20 ± 0·01†,§	0·21 ± 0·01	0·22 ± 0·01	0·20 ± 0·01
	Aquatic exercise	0·19 ± 0·01	0·21 ± 0·01	0·96 ± 0·06*	0·22 ± 0·01	0·20 ± 0·01	0·20 ± 0·01†,‡	0·19 ± 0·01
$\dot{V}E$ (l min^{-1})	Cycling	6·76 ± 0·56	7·23 ± 0·61	22·30 ± 1·35*	7·53 ± 0·53	7·91 ± 0·71	7·94 ± 0·57	7·57 ± 0·58
	Walking	6·75 ± 0·60	7·05 ± 0·50	23·10 ± 1·10*	7·05 ± 0·47	7·67 ± 0·67	7·87 ± 0·87	7·47 ± 0·54
	Aquatic exercise	7·06 ± 0·75	7·26 ± 0·56	27·28 ± 2·36*,†,‡	7·87 ± 0·59	7·25 ± 0·47	7·24 ± 0·54	7·21 ± 0·45
RER	Cycling	0·83 ± 0·03	0·91 ± 0·05	0·94 ± 0·02	0·94 ± 0·04	0·97 ± 0·05*	0·93 ± 0·05	0·91 ± 0·05
	Walking	0·87 ± 0·05	0·88 ± 0·05	0·92 ± 0·01	0·91 ± 0·03	0·93 ± 0·05	0·95 ± 0·06	0·92 ± 0·03
	Aquatic exercise	0·88 ± 0·05	0·89 ± 0·04	0·94 ± 0·03	0·95 ± 0·04	0·89 ± 0·03	0·89 ± 0·04	0·87 ± 0·04

$\dot{V}O_2$, oxygen uptake; $\dot{V}CO_2$, carbon dioxide production; $\dot{V}E$, expired minute ventilation; RER, respiratory exchange ratio.
Values are mean ± SE.
*$P<0·05$ versus baseline, $^{†}P<0·05$ versus cycling, $^{‡}P<0·05$ versus walking, $^{§}P<0·05$ versus aquatic exercise.

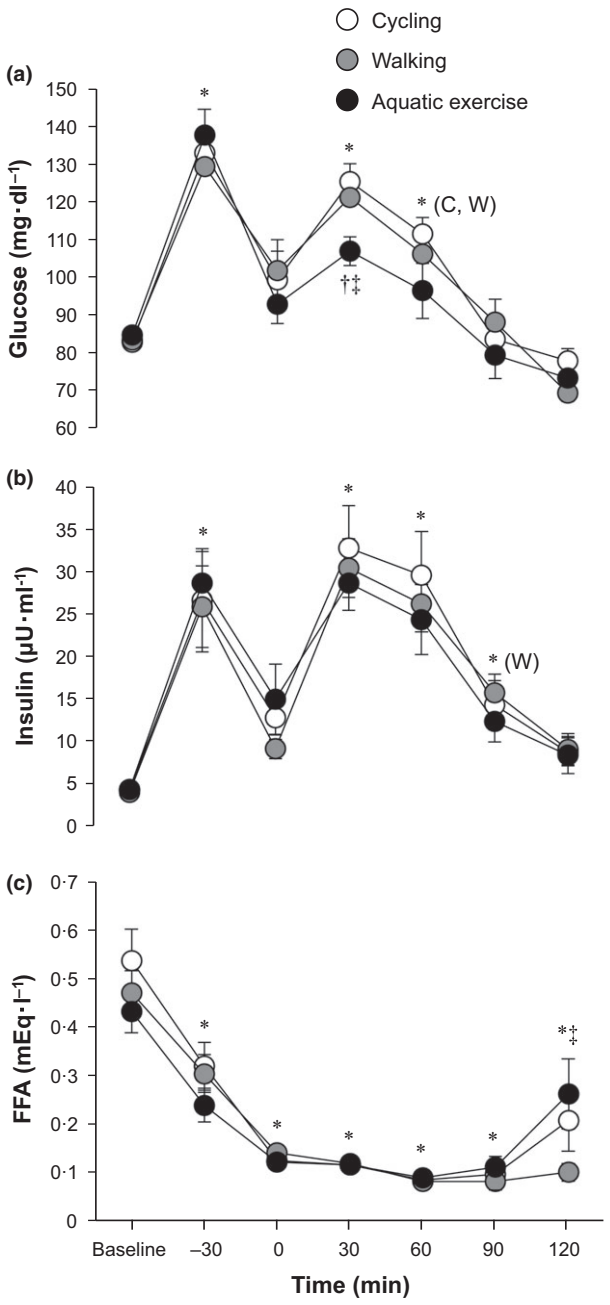

Figure 1 Plasma glucose (a), serum insulin (b) and free fatty acids (FFA) (c) concentrations (mean ± SE) during the three exercise conditions following the glucose load. C, cycling; W, walking. *P<0·05 versus baseline, †P<0·05 versus cycling, ‡P<0·05 versus walking.

modes maintained a high value at 60 min after exercise (P<0·05). Compared with land exercise modes, the plasma glucose concentration in aquatic exercise was significantly lower at 30 min after exercise (P<0·05). Therefore, aquatic exercise seems to restrain hyperglycaemia after a glucose load compared with both land exercises.

Similar to the pattern seen with plasma glucose, the serum insulin concentration at 30 min after a glucose load, and at 30 and 60 min after exercise was increased in all exercise modes compared with baseline (Fig. 1b, P<0·05). Moreover, this significantly high concentration was maintained until 90 min after exercise only in the walking condition (P<0·05). However, serum insulin responses did not differ significantly among exercise modes.

The serum FFA concentration during the period of −30 to 120 min after exercise was significantly decreased from baseline for all exercise modes (Fig. 1c, P<0·05). At 120 min after exercise, the serum FFA concentration was significantly higher in the aquatic exercise condition compared with the walking condition (P<0·05).

Discussion

The goal of the present study was to determine the most effective exercise mode to prevent postprandial hyperglycaemia. We investigated the effect of different modes of exercise after glucose ingestion on metabolic responses in healthy men and found that plasma glucose level at 30 min after exercise was significantly lower with aquatic exercise compared with both land exercise modes (walking and cycling). In addition, aquatic exercise promoted better lipolysis (higher FFA) at 120 min after exercise compared with both land exercises.

The control of glycaemia is dependent on the activities of the neuroendocrine system. In resting conditions, glucose uptake by cells is mainly insulin dependent, in which GLUT-4 is translocated to the cell membrane (Ryder *et al.*, 2001). Exercise increases the concentration of GLUT-4 in the cell membrane, which leads to an increase in glucose uptake, even with low insulin levels (insulin-independent) (Krook *et al.*, 2004). In the present study, serum insulin concentration decreased immediately after postprandial exercise as previously reported (Morishima *et al.*, 2014). Thus, the decrease in plasma glucose immediately after exercise shown in this study may be attributed to the translocation of GLUT-4 through insulin-independent pathways. However, there were no significant differences among exercise modes in plasma glucose immediately after exercise. Previous studies have shown that glucose uptake during exercise depends on the exercise mode (Tsintzas *et al.*, 2003); however, hepatic glucose output is increased to maintain blood glucose within the normal range during exercise (Marmy-Conus *et al.*, 1996; Suh *et al.*, 2007). As plasma glucose rapid declined immediately after three types of exercise, homeostatic system might have affected to maintain the glycemic level. Moreover, difference in surface EMG activities between exercise on land and in water come out with increase in workload (Wiesner *et al.*, 2010), therefore set workload in this study may not have been sufficient to decrease glucose level.

On the other hand, plasma glucose was lower at 30 min after aquatic exercise compared with the land exercises. Differences in body temperature during the three exercise modes should be considered as one reason for this result. A rise in body temperature closely relates to an increase in catecholamine (Febbraio *et al.*, 1996; Gagnon *et al.*, 2013). It has

been reported that body temperature and catecholamine release are greater during exercise on land than that in water of a similar temperature to that of the current study (Fujishima *et al.*, 2001). Increases in catecholamine promote hepatic glycogenolysis, which increases blood glucose levels (Hargreaves *et al.*, 1996; Morris *et al.*, 2005). In fact, it has been shown that catecholamine secretion during land exercise is greater than that during water exercise and accompanies an increase in blood glucose (Wiesner *et al.*, 2010). Therefore, the increase in blood glucose during land exercise at 30 min after exercise found in the current study is assumed to be due to glycogenolysis induced by catecholamine. Furthermore, exercise-induced hyperthermia augments glucagon, cortisol and growth hormone secretions, which could contribute to a greater increase in plasma glucose after cycling and walking (Hargreaves *et al.*, 1996).

Although the plasma glucose level after exercise was the lowest in the aquatic exercise condition, carbohydrate utilization may not have necessarily been elevated (Table 1, RER). Rather, carbohydrate utilization tended to be higher after cycling. Elevation of body temperature with exercise leads to an increase in degradation and utilization of glycogen by the augmentation of catecholamine (Febbraio *et al.*, 1996; Morris *et al.*, 2005). Further, the disappearance of muscle glycogen is greater in cycling than in running because cycling recruits more Type II fibres (Arkinstall *et al.*, 2001). These factors are thought to be the reason carbohydrate utilization is elevated after cycling. However, it will be necessary to determine the body temperature, catecholamine and muscle glycogen contents at the same time to establish the above predictions in additional research.

Serum FFA at 120 min after exercise was higher in the aquatic exercise condition compared with both land exercise conditions. FFA is produced through the hydrolysis of triglycerides by hormone sensitive lipase (HSL) in adipocyte cells, but insulin inhibits the activity of HSL (Holm, 2003). Although serum FFA after glucose ingestion decreased in all modes of exercise, serum FFA after aquatic exercise tended to return sharply towards the baseline at 120 min in the recovery period. However, because there were no significant differences among exercise modes in serum insulin, it is necessary to consider other reasons why the serum FFA after the aquatic exercise was elevated. Wiesner *et al.* (Wiesner *et al.*, 2010) has reported that atrial natriuretic peptide (ANP) in a water exercise condition was significantly increased compared with that during a land exercise condition under the same $\dot{V}O_2$. ANP has a strong stimulatory effect on adipose tissue lipolysis and is released from the atrial wall by increased blood volume in the atria (venous return) during exercise in water than on land (Fenzl *et al.*, 2013). On the other hand, because catecholamine stimulates FFA production (Mora-Rodriguez & Coyle, 2000) as well as ANP, enhanced catecholamine on land exercise has the potential to elicit serum FFA production compared with water exercise. However, maximal ANP release has been observed at 30–120 min after water immersion (Ogihara *et al.*, 1986; Epstein *et al.*, 1989), whereas augmentation of catecholamine disappears within 30–60 min after exercise (Stich *et al.*, 2000; Ronsen *et al.*, 2001). Therefore, it is thought that increase in ANP after exercise lasts longer than catecholamine, and the lipolysis action induced by persistent secretion of ANP could be influenced more strongly after aquatic exercise than after both land exercises. In fact, it has been demonstrated that FFA is increased by sustained augmentation of ANP despite the lower concentration of catecholamine during exercise in water (Wiesner *et al.*, 2010). Thus, the same mechanisms could contribute to the increase in serum FFA at 120 min after aquatic exercise demonstrated in this study.

We understand that some limitations should be carefully considered in the interpretation of the present study. We suggested that supposed differences in catecholamine level and body temperature as well as in ANP secretion are reasons for dampened hyperglycaemia and augmented lipolysis after aquatic exercise. However, the endocrine data to support these speculations was lacking, thereby further studies are necessary to extend our understanding. Moreover, the use of a glucose clamp technique in addition to the assessment of glucose utilization via RER would allow for the determination of detailed glucose kinetics. Blood glucose responses during exercise are difference between healthy and T2DM subjects (Musi *et al.*, 2001); therefore, we have to show the availability of aquatic exercise for symptomatic improvement with T2DM subjects.

Conclusions

These results suggest that aquatic exercise after glucose ingestion decreased postprandial blood glucose compared with cycling and walking with same energy expenditure. Aquatic exercise may be an effective exercise prescription for preventing postprandial hyperglycaemia, but future studies are required to verify the postprandial metabolic and endocrine responses, including a body temperature measurement, with aquatic exercise in T2DM patients.

Acknowledgments

We are grateful to the subjects and members of the laboratory staff who helped with the implementation of this study. We especially thank Masahito Ohta with great sincerity for introducing many subjects to us.

Conflict of interest

The authors have no conflicts of interest.

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