นิพนธ์ต้นฉบับ • Original Article

ผลของการออกกำลังกายแบบแอโรบิคต่ออัลฟาอะไมเลสในน้ำลาย และจำนวนเม็ดเลือดขาวในประชากรไทยที่ออกกำลังกายไม่เพียงพอ

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Effect of Aerobic Exercise on Salivary Alpha-amylase and White Blood Cell Count among Sedentary Thais

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หลักการและวัตถุประสงค์ : การออกกำลังกาย อาจจะทำให้เกิดความเครียดทางร่างกายและทางจิตใจ กระตุ้นให้มีการทำงานของระบบประสาทซิมพาเธติกและมีการคัดหลั่งแคทธีโคลามีนเพิ่มขึ้น การศึกษานี้มีวัตถุประสงค์ที่จะตรวจวัดการทำงานของระบบประสาทซิมพาเธติกและทำงานของภูมิคุ้มกันในระหว่างที่มีการออกกำลังกายแบบแอโรบิคในประชากรไทยที่ออกกำลังกายไม่เพียงพอ

วิธีการศึกษา : ทำการศึกษาในกลุ่มอาสาสมัครที่ออกกำลังกายไม่เพียงพอจำนวน 50 ราย โดยแบ่งเป็น 2 กลุ่ม คือ กลุ่มควบคุม (25 ราย) และกลุ่มออกกำลังกาย (25 ราย) อาสาสมัครทุกคนเป็นผู้ที่มีสุขภาพดี ไม่มีโรคที่เกี่ยวข้องกับหัวใจและหลอดเลือด ในกลุ่มออกกำลังกาย ได้ทำการออกกำลังกายโดยวิธีการเดินเร็วแบบยกแขนสูงที่ระดับความหนักปานกลาง (ร้อยละ 75 ถึง 80 of HRmax) วันละ 30 นาที 3-5 วัน/สัปดาห์ ติดต่อกัน 12 สัปดาห์ ทำการประเมินความเครียดโดยการตรวจวัดปริมาณอัลฟาอะไมเลสในน้ำลาย (sAA) เพื่อประเมินการทำงานของระบบประสาทซิมพาเธติก รวมถึงทำงานของภูมิคุ้มกันโดยการตรวจนับเม็ดเลือดขาว (WBC) ชนิดนิวโทรฟิล (NE) ลิมโฟไซต์ (LY) โมโนไซต์ (MO) อิโอซิโนฟิล (EO) และแบโซฟิล (BA) วิเคราะห์ข้อมูลโดยใช้ค่าสถิติ ค่าเฉลี่ย ส่วนเบี่ยงเบนมาตรฐาน สถิติแบบจับคู่ สถิติวิเคราะห์ความแปรปรวนแบบวัดซ้ำ

Background and objectives : Physically and mentally stressful situations may arise during exercise, resulting in sympathetic activation and an increase in catecholamine secretion. This study aimed to examine sympathetic activation and immune function during aerobic exercise in sedentary Thais.

Methods : Fifty sedentary subjects were divided into 2 groups: control (n = 25) and exercise groups (n = 25). All subjects were healthy, having no clinical evidence of cardiopulmonary disease. The exercise group was engaged in brisk marching (75–to 80% HRmax) with 30 min/session, 3 to 5 sessions/week for 12 consecutive weeks. Stressful condition was evaluated by measuring salivary alpha amylase (sAA) levels in order to investigate sympathetic activation. Additionally, immune function was evaluated by white blood cell (WBC) count: neutrophil (NE), lymphocyte (LY), monocyte (MO), eosinophil (EO) and basophil (BA). The mean, standard deviation, paired t-test; repeated ANOVA and ANCOVA were employed for data analysis.

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ผลการศึกษา : เมื่อเปรียบเทียบกับก่อนการออกกำลังกายพบว่าระดับอัลฟาอะไมเลสในน้ำลาย (sAA) เพิ่มขึ้นในสัปดาห์ที่ 4 และ 8 ของการออกกำลังกายโดยการเดินเร็วแบบยกแขนสูง โดยมีค่า 125.90 ± 13.31 kIU/l เปรียบเทียบกับ 196.46 ± 19.19 kIU/l ($p < 0.005$) และ 177.69 ± 17.45 kIU/l ($p < 0.05$) ตามลำดับ และยังพบการเปลี่ยนแปลงอย่างมีนัยสำคัญของจำนวนเม็ดเลือดขาว ได้แก่ NE, LY, MO, EO และ BA ในช่วงก่อนเทียบกับช่วงหลังการออกกำลังกายแบบแอโรบิค เท่ากับ 6.48 ± 2.40 เทียบกับ 6.14 ± 1.78 103/uL, 57.58 ± 10.12 เทียบกับ 55.92 ± 8.42%, 31.94 ± 8.90 เทียบกับ 33.74 ± 7.64%, 6.62 ± 1.93 เทียบกับ 6.36 ± 1.35%, 3.36 ± 2.66 เทียบกับ 3.41 ± 1.97% และ 0.48 ± 0.18 เทียบกับ 0.53 ± 0.17% ตามลำดับ

สรุป : การศึกษานี้พบว่า การออกกำลังกายโดยวิธีการเดินเร็วแบบยกแขนสูงติดต่อกันนาน 12 สัปดาห์ อาจจะมีผลต่อภาวะเครียดที่อาจจะเพิ่มขึ้นในระยะเริ่มแรกของการออกกำลังกายแบบแอโรบิคแต่ไม่ส่งผลต่อการทำงานของภูมิคุ้มกัน ดังนั้นควรส่งเสริมให้มีการออกกำลังกายอย่างต่อเนื่องในประชากรที่ออกกำลังกายไม่เพียงพอ

คำสำคัญ : อัลฟาอะไมเลสในน้ำลาย, เม็ดเลือดขาว,ประชากรไทยที่ออกกำลังกายไม่เพียงพอ

Results : In comparison to the pre-exercise, sAA levels were significantly increased following 4 and 8 weeks of brisk marching, 125.90 ± 13.31 kIU/l *vs* 196.46 ± 19.19 kIU/l ($p < 0.05$) and 177.69 ± 17.45 kIU/l ($p < 0.05$), respectively. Nevertheless, they were restored by week 12 (127.87 ± 16.89 kIU/l). Moreover, significant differences in NE, LY, MO, EO and BA were not observed post-compared to pre-aerobic exercise (6.48 ± 2.40 vs 6.14 ± 1.78 10^3/uL, 57.58 ± 10.12 vs 55.92 ± 8.42%, 31.94 ± 8.90 vs 33.74 ± 7.64%, 6.62 ± 1.93 vs 6.36 ± 1.35%, 3.36 ± 2.66 vs 3.41 ± 1.97% and 0.48 ± 0.18 vs 0.53 ± 0.17%, respectively.

Conclusions : The present study suggests that aerobic exercise by brisk walking for 12 consecutive weeks may compromise a stressful situation induced earlier by aerobic exercise and maintain immune function. Therefore, prolonged exercise training should be recommended to sedentary individuals.

Keywords : salivary alpha amylase, white blood cell, sedentary Thais

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Introduction

Currently, Thais becomes to have a sedentary lifestyle[1]. A sedentary behaviour is commonly defined as an activity with metabolic equivalent of task (METs) of 1.5 or less or activities involving energy expenditure at the level of 1.0-1.5 METs. Such the sedentary lifestyle is often associated with decreased performance, risks of cardiovascular disease and overweight or obese, and finally becomes a major cause of death[2]. It places a large financial burden on the government each year.

Stress is an emotion or feeling caused by three factors such as mental state, changes in life and physical illness. It stimulates sympathetic nervous system resulting in catecholamine neurotransmitter and salivary alpha amylase (sAA) releases from saliva glands[3] leading to various diseases such as depression, cancer, immune deficiency and cardiovascular disease[4].

Physical activity is defined as any bodily movement proceeded by skeletal muscles that requires energy expenditure[5] such as playing, working, doing the housework and gardening[6]. Aerobic exercise by brisk walking, which is easy to do and do not require equipments, has been shown to enhance aerobic capacity[7]. Moreover, it also decreases blood glucose, increases high density lipoprotein cholesterol (HDL-C), increases respiratory muscle strength[8] and peak oxygen consumption[9]. Although continuous exercise helps enhance health and prevent cardiovascular diseases, diabetes, obesity, cancer, anxiety and depression[10], previous studies have shown

that exercise may affect physical and mental stress and subsequently physical health. Acute and heavy exercise may increase sAA response. Chatterton et al. studied among skydiving athletes reporting significantly increased cortisol and sAA concentration after competition[11]. Besides, a study on marathon runners has demonstrated that the sAA level significantly increased after 20 km of running but decreased 1 hr after the run[12]. In triathlete and youth taekwondo athlete, sAA concentration was increased after competition[13,14]. Moreover, it was also found that a secretion of stress hormones is associated with a change in the immune system following acute or heavy exercise[15, 16].

Purpose of the study

We explored sAA and white blood cell responses to brisk marching exercise of 12 consecutive weeks in sedentary Thais.

Methodology

This study recruited 50 healthy sedentary Thai aged between 20 to 55 years old with a body mass index (BMI) between 18.5 to 24.9 kg/m^2. They were non-regular alcohol drinkers and non-smokers. Each participant was fully informed the purpose of the study and the possible risks and provided written an informed consent.

Procedures

All participants works at Srinagarind Hospital, and Khon Kaen University, Khon Kaen, Thailand. A run-in period consisting of walking 40 to 60 min/day for 2 weeks was performed by all participants. They were randomly assigned to be control or exercise groups. Salivary and blood samples were collected at week 0 and week 12 in the control group. In the exercise group, a session of 30 min (5-min warm up, 20-min exercise by brisk marching of 75 to 80% HRmax, 5-min cool down), 3 to 5 sessions/week for 12 weeks were performed. There were 4 periods of data collection. Salivary and blood samples were collected prior to the exercise program (week 0) and post-exercise program (week 12), whereas only salivary samples were collected at week 4 and 8.

Equipments

Sedentary participants were included according to self-assessment questionnaire and physical activity questionnaire by a physician. A Polar for heart rate measurement, a weighing scale, a blood pressure monitor, a measuring tape and a flat chip sensor for sAA measurement were used.

Statistical analyses

Results are expressed as means ± SD. A paired t-test was used to compare pre- and post-exercise program for the immune function. sAA parameters between pre- and post-exercise program were analysed by analysis of variance (ANOVA). An analysis of covariance (ANCOVA) was used for comparisons of sAA between control and exercise groups. Significance was accepted as $p<0.05$. Data were analysed with SPSS software version 16.

Research place

This study was done at Srinagarind Hospital, the Faculty of Medicine, Khon Kaen University.

Results

Data are summarized in Table 1, 2 & 3 and Figure 1. Two groups were similar with regard to age, body weight, height pre-exercise. However, results showed that body weight, body mass index (BMI), waist circumference (WC) and hip circumference (HC) were significantly decreased ($p< 0.05$) following 12 weeks of brisk walking (Table 1).

Compared to the pre-exercise level (week 0), the sAA level was significantly increased at week 4 ($P=0.003$) and week 8 ($p = 0.019$) but not at week 12 ($p = 1.0$). Nonetheless, no significant change in sAA between week 0 and week 12 in the control group ($p = 0.72$) (Table 2 & Figure 1).

In regard to the white blood cells, there were no significant differences in the amount of white blood cells between week 0 and week 12 in either group of participants (Table 3).

Table 1 Clinical characteristics of participants.

	Control group (n=25)		Exercise group (n=25)	
	Pre-	**Post-**	**Pre-**	**Post-**
Age (years)	32.2 ± 9.1		33.9 ± 9.6	
Weight (kg)	52.1 ± 5.3	52.2 ± 5.5 [a]	54.7 ± 6.2	54.0 ± 6.7* [a]
Height (cm)	158.3 ± 6.3	158.3 ± 6.3 [a]	161.0 ± 6.9	161.0 ± 6.9 [a]
BMI (kg/m^2)	20.8 ± 1.5	20.8 ± 1.7 [a]	21.1 ± 1.8	20.0 ± 4.6* [a]
SBP (mm Hg)	101.8 ± 9.8	104.6 ± 8.6 [a]	104.2 ± 10.2	101.3 ± 6.5 [a]
DBP (mm Hg)	69.7 ± 7.3	67.8 ± 7.9 [a]	68.7 ± 9.2	68.4 ± 6.4 [a]
MAP (mm Hg)	80.4 ± 6.4	79.9 ± 7.4 [a]	80.2 ± 1.8	79.1 ± 1.2 [a]
HR (/min)	71.3 ± 8.3	71.2 ± 9.1 [a]	69.5 ± 8.1	68.5 ± 7.8 [a]
WC (cm)	71.4 ± 5.1	72.2 ± 5.5 [a]	74.2 ± 5.9	71.9 ± 5.2* [b]
HC (cm)	90.8 ± 3.8	91.4 ± 3.9 [a]	93.0 ± 5.7	91.8 ± 5.5* [b]
WHR	0.79 ± 0.6	0.79 ± 0.06 [a]	0.80 ± 0.06	0.78 ± 0.05* [b]

Data are presented as mean ± SD. BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; MAP, mean arterial pressure; HR, heart rate; WC' waist circumference; HC, hip circumference; WHR, waist to hip ratio. Data were tested by a paired t-test[a] or a sign rank test[b].

* $p < 0.05$, significantly different than pre-exercise.

Table 2 Changes in salivary alpha amylase concentrations in control and exercise groups.

Week	sAA concentration (kIU/l)	
	Control group (N=21)	**Exercise group (N=25)**
0	105.95 ± 81.75	125.90 ± 13.31
4	-	196.46 ± 19.19 (p=0.003)
8	-	177.69 ± 17.45 (p=0.019)
12	112.11 ± 55.82 (p=0.72)	127.87 ± 16.89 (p=1.00)

Data are presented as mean ± SD tested by ANCOVA.

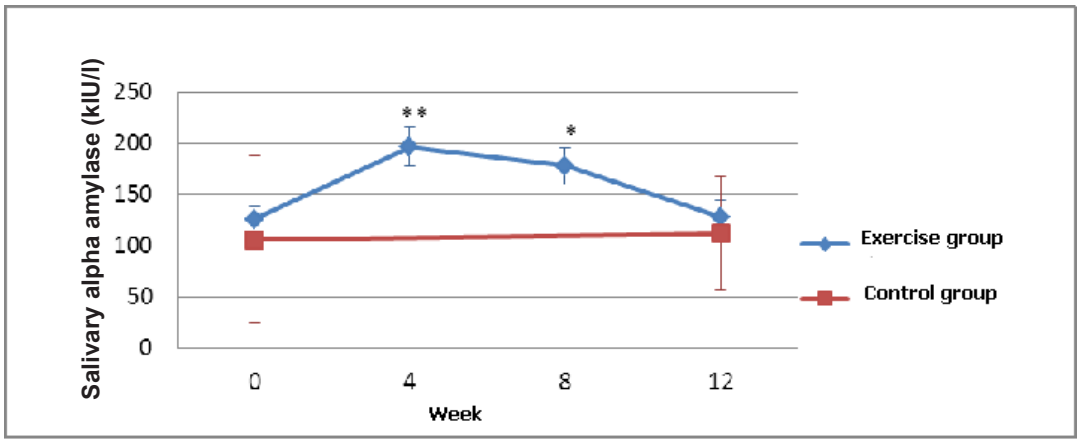

Figure 1 Salivary alpha amylase (sAA) levels in control and exercise groups at week 0, 4, 8 and 12. *p < 0.05; ** P < 0.005, significantly different than pre-exercise.

Table 3 White blood cell count before and after exercise in control and exercise groups

White blood cell	Control group (N=25)		p-value	Exercise group (N=25)		p-value
	Week 0	Week 12		Week 0	Week 12	
WBC (10^3/uL)	5.82 ± 1.65	6.05 ± 1.62	0.24[b]	6.48 ± 2.40	6.14 ± 1.78	0.25[a]
NE (%)	54.40 ± 5.87	54.82 ± 9.84	0.85[a]	57.58 ± 10.12	55.92 ± 8.42	0.69[b]
LY (%)	33.92 ± 5.68	32.75 ± 7.41	0.54[a]	31.94 ± 8.90	33.74 ± 7.64	0.31[a]
MO (%)	7.49 ± 2.05	7.40 ± 2.09	0.42[a]	6.62 ± 1.93	6.36 ± 1.35	0.10[b]
EO (%)	3.68 ± 2.75	4.36 ± 3.98	0.54[b]	3.36 ± 2.66	3.41 ± 1.97	0.25[b]
BA (%)	0.49 ± 0.18	0.64 ± 0.67	0.63[b]	0.48 ± 0.18	0.53 ± 0.17	0.65[b]

Data are presented as mean ± SD tested by paired t-test[a] and sign-rank test[b].

Discussion

In the present study, we examined the effect of exercise on stress and the immune system. We found that an aerobic exercise program derived from brisk marching for 12 weeks is beneficial for reducing stress in healthy subjects. Brisk walking training resulted in increased concentrations of sAA at week 4 and week 8 but decreased at week 12. Increases in sAA levels at weeks 4 and 6 of exercise may be due to stress-activated sympathetic nervous system which, in turn, stimulates the salivary glands to secrete the alpha amylase enzyme. In addition, the levels of sAA may increase for several reasons such as stress, anxiety, expectations, and competitive sports[14] or the tissue injured from playing sports[12]. However, the stress at week 4 and 8 of exercise was subsequently reduced at week 12. Consistent with a previous study, continuous exercise at 70 to 85% of HRmax intensity demonstrated that the sAA levels did not change[17]. Thus, exercise over a long period can reduce a response to arousal and stress, which consequently enhances performance of the body.

The characteristics such as body weight, fat loss after aerobic exercise by brisk marching for 12 weeks is consistent with the study by Roubenoff et al. in patients with

human immunodeficiency virus[18]. This also confirms that the fat content decreased. The body uses oxygen in the metabolism of food using fat as the primary energy source. Consequently, waist circumference and hip circumference as the source of fat were decreased.

Moreover, the immune function assessed by the WBC after 12 weeks of exercise did not demonstrate any change. As we did not study activities of WBC, therefore, this study did not confirm improved immune function following chronic exercise. Nevertheless, it is likely that continuous moderate exercise reduces inflammation induced by CD14+ and CD15+ monocyte[15], natural killer cell (NK cell)[19] and NK cytotoxicity, and improving efficiency of what? also reduces the responsiveness of the norepinephrine[20].

Conclusion

The present study suggests that continuous and regular exercise by brisk walking has no effects on physical stress and on increasing the amount of white blood cells providing that it lasts at least 12 weeks. Therefore, regular exercise should be encouraged in sedentary people. In addition, it is also applicable in athletes during recovery after heavy training.

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