運動對學習記憶、焦慮、憂鬱及物質濫用之影響

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計畫主持人：游一龍

計畫參與人員：鄧予涵　專任助理

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The impact of exercise on learning and memory, anxiety, depression and substance abuse

(運動對焦慮、憂鬱及物質濫用促發因子易感性之研究)

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(一) 中文摘要
焦慮、憂鬱症及物質濫用是台灣地區急速發展的兩大國民身心健康議題。焦慮症與憂鬱症主要症狀包括伴隨情緒異常而有活動力、飲食行為異常、攻擊傾向高、注意力不集中與受損的學習記憶。焦慮、憂鬱症可能有共同的病因及病理基礎。近年來的研究多指出運動合併抗憂鬱劑的使用可以回復輕、中度憂鬱症或因慢性病引發憂鬱的症狀。運動造成的長期效果及經濟特性提供了一個副作用少的非藥物治療焦慮、憂鬱的可能方式。我們在過去一年的研究以高架十字形迷津模型發現中長期及長期強迫運動效果可能改變該模型中的本能焦慮行為指標。心理運動刺激劑(例如近年來在臺灣地區氾濫使用的搖頭丸)的開始使用及戒除後復發受壓力這個因子的培基調控，長期強迫運動的效果之一是調整生物體對於壓力的易感性。我們以場地偏好物質依賴模型研究中長期及長期強迫運動是否能降低動物對心理運動刺激劑的強化效果。我們發現中長期及長期強迫運動效果皆可以改變小鼠對於搖頭丸的強化行為效果。
關鍵詞：運動、學習記憶、物質濫用、焦慮、神經傳導素

（二）Abstract
To date, affective disorders, seriously endangering both mental and physical health at gigantic cost, raise a major issue of concern in the local area. Psychomotor stimulants are, currently, substance abuse of choice in Taiwan. The symptoms of major depression and generalized anxiety disorders are frequently profiled as a cluster of motivation-related deficits in general activity, foraging and drinking, aggression, attention, learning and memory. Lately, accrued lines of clinical evidence claimed that exercise alone and/or in conjunction of anti-depressive drugs effectively alleviated the symptoms associated with a mild to moderate depression even well-controlled experiments still remain sparse. Exercise, characterized by its long-lasting effects, inexpensive cost, and less side effects, deserves a full-scale understanding. We propose to examine the impact of long-term forced exercise program on altering behavioral parameters in elevated plus maze paradigm for naïve anxiety level. Humans and animals under stress are more susceptible to the reinforcing (euphoric) effects of psychomotor stimulants. Exercise has long been known to modulate the stress susceptibility in many species. We, accordingly, hypothesize that long-term exercise may attenuate the reinforcing effects of MDMA. We propose to study the effects of long-term exercise on the euphoric/reinforcing effects of MDMA in conditioned place preference paradigm in a mouse model.

Key words: exercise, learning/memory, anxiety, substance abuse, neurotransmitter

(三) Progress report

Introduction
The growing size of senile populations and competitive working conditions are thought to induce subjective stress, anxiety and depression, with a consequent decrease in the self-evaluated quality of life, circadian rhythm disturbances, attention-disrupted poor productivity and various drug abuses. Acute stress may suppress immune function, leading to an increased incidence of infections, and chronic stress may predispose to several ailments, including digestive disturbances, cardiovascular diseases and neoplasia. Thus, control of stress becomes an important issue in all modern societies. Although stress can not be avoided, its impact may be reduced by many recommended ways. Among them, physical activity/exercise can be one of the best alternatives, since exercise was not only thought to exert many positive effects on physical and psychological health with minor side effects but to possess a money-saving character.

Physical activity/exercise is associated with lower risks of cognitive impairment, and physical inactivity may be a risk factor for the Alzheimer’s disease (Friedland et al., 2001; Laurin et al., 2001). Physical activity and habitual exercise may have beneficial effects in both young and aged subjects. The exercise program appeared to have a greater effect on physiological functioning, concentration and short-term memory of younger subjects, while both young and old subjects achieved gains in physiological functioning, and psychological well-being (Emery, 1994). High intensity aerobic exercise has positive effects on well-being in an adolescent population (Norris et al., 1992). An individualized exercise program can improve a functional balance in people aged above 75 years and such improvement was maintained at least for one month (Wolf et al., 2001).

Many clinical studies reported that exercise may reduce age-related lean body mass loss and risk for several chronic diseases. Moderate muscle strength training demonstrated positive effects on clinical parameters in chronic heart failure patients (Radzewitz et al., 2002). Although inconclusive results have been reported for most treatment modalities, exercise appeared to decrease the myriad of physical symptoms associated with Fibromyalgia, a rheumatological disorder of unknown origin (Meyer and Lemley, 2000; Gowans et al., 2001). Graded exercise therapy have shown promising results in reversing the symptoms of chronic fatigue syndrome, characterized by severe disabling fatigue and a variety of musculoskeletal, cognitive and sleep disorders lasting at least six months (Youssefi and Linkowski, 2002). To date, the general public is, perhaps, aware of the physical benefits of exercise, but less cognizant of the psychological merits of regular exercise. Psychological states such as mild-to-moderate anxiety, depression, and chronic diseases-associated dysthymia (a mild depression) have been found reduced by exercise and/or physical activity. Likewise, exercise has been claimed to elevate mood, increase intellectual functioning, and improve self-concept. Although people with depression tend to be less physically active than non-depressed individuals, increased aerobic exercise or strength training has been shown to reduce their depressive symptoms (Paluska and Schwenk, 2000). Anxiety symptoms and panic disorder also improve with regular exercise (Paluska and Schwenk,
An exercise training program may be considered an alternative to antidepressants for treatment of depression in senile persons. Although antidepressants may facilitate a more rapid initial therapeutic response than exercise, exercise was equally effective in reducing depression among patients with major depressive disorder after 16-wk of antidepressant treatment (Blumenthal et al., 1999). An appropriate application of exercise program was effective in relieving depression or anxiety in the long-term maintenance hemodialysis patients with common complaints of depression and anxiety (Suh et al., 2002). Mild to moderate aerobic exercise may be of therapeutic value to breast cancer survivors with respect to depressive and anxiety symptoms (Segar et al., 1998). Healthy subjects showed increased physiological and psychological indices of relaxation after underwater exercise (Oda et al., 1999). Overall results revealed that exercise-induced increases in aerobic fitness have beneficial short-term and long-term effects on psychological outcomes (DiLorenzo et al., 1999). Exercise was associated with decreases in total mood disturbance, as well as increases in vigor in physically active postpartum women (Koltyn and Schultes, 1997).

However, claims for the psychological benefits of physical exercise appear to precede solid evidence. The emotional effects of acute exercise still remain controversial. Some claimed that acute exercise, a dynamic version of Takwondo, induced positive mood state changes and long-term exercise, extensive Takwondo skill, did not necessarily elicit beneficial changes in affect (Toskovic, 2001), while others emphasized the paucity of effects following acute exercise. Long-term exercise exerts the antidepressant and anxiolytic effects and mitigates the harmful consequences of stress mostly limited in subclinical studies. Studies attempt to link exercise habits to protection from harmful effects of stress on mental health, but causality remains unclear. Moreover, conflicting data on physical activity modalities hinder any general conclusion. For example, a buffering effect for leisure physical activity was suggested against physical symptoms and anxiety associated with minor stress, while no moderating effect for aerobic fitness was found in this regard (Carmack et al., 1999). Physical activity of long duration amongst men conferred protection against common mood and anxiety states, while no such protection for women (Bhui and Fletcher, 2000). Furthermore, exercise, as performed strenuously, could be associated with increased production of reactive oxygen species, consuming endogenous antioxidants and eventually damaging biological molecules and cellular components. Likewise, excessive physical activity is thought to result in overtraining and generate psychological symptoms that mimic depression. Thus, well controlled studies are needed to elucidate the mental benefits of exercise in differing populations and to address the biological mechanisms underlying the benefits of exercise on mental health.

As for animal studies, sparse paradigms have been attempted to examine the modulating effects of exercise/physical activity on reversing behavioral, neurochemical, or neuroendocrine parameters associated with depression and/or anxiety. Dishman (1997) reported that voluntary (chronic activity wheel running) and forced exercise (treadmill exercise training) exerted anxiolytic and antidepressant effects via reversing behavioral, neurochemical versus neuroendocrine parameters, respectively. The voluntary wheel running increases neurogenesis and long-term potentiation in the dentate gyrus, and enhances spatial learning performance (Fordyce et al., 1993; van Praag et al., 1999a; van Praag et al., 1999b), while stress caused atrophy of dendrites in the CA3 region and suppresses the neurogenesis of dentate gyrus neurons (McEwen, 1999). However, the results of most these studies compromise with the methodological flaws, such as the loosely-controlled intensity of exercise and genetic backgrounds, dissimilar exercise protocols as well as the incompatible sampling timing. In an attempt to avoid these weaknesses, we employed consistently the same paradigms (CPP and EPM), forced exercise protocols, sampling timing in mouse models with similar genetic backgrounds.

**Specific Aims**
1. We attempted to differentiate the effects of medium- and long-term forced exercise on locomotor activity and naïve anxiety levels.
2. We decided to examine the modulating effects of of medium- and long-term forced exercise on the reinforcing/ euphoric effects of MDMA.

**Materials and methods**
**Animal models:** Since one report documented that there were sexual differences in exercise-generated benefits, only male C57BL/6 mice (3 months old) were used as the animal model in the experiments. Mice were group-housed (5 per cage) with free access to mouse chow and tap water in a humidity- and temperature-controlled colony room and laboratory maintained on a 12 h light/dark cycle unless mentioned otherwise. All experimental procedures and use of animals have been approved by the local committee at National Cheng Kung University College of Medicine (see attached).

**Exercise protocols:** Under the forced exercise protocol, following one-week familiarization, mice in the medium- and long-term exercise groups run on a treadmill at the speed of 12 m/min for 60 min/day, 5 days/wk, 1 and 12 weeks in total, respectively. In contrast, the sedentary groups experienced one-week familiarization, then, were placed on the yoked treadmill for 10 min each day except any exercise training.

**Elevated plus maze:** A custom-made dark plastic maze with two anxiety-provoking open arms and two closed arms with high walls specially for mice were used. The plus maze was elevated 75 cm above the ground. Mice were allowed to freely explore these arms on this maze. Voluntary passage onto the open arms is associated with elevated plasma corticosterone concentrations, increased freezing and production of fecal boli. Normal exploration is in favor of the closed arms. Percentage of total open arm entry and time spent were recorded to examine the anxiolytic effects of exercise.

**Conditioned Place Preference:** Mouse Place Preference Instrument ENV-3013 and Software (Georgia, Vermont, Med Associate, USA) were used. Mice were first translocated from their home cages to the instrument for a 10-min adaptation to assure the unbiased design. Mice, then, were injected, intraperitoneally, with MDMA (Ecstasy at 1.67 mg/kg) and refrained in one compartment chamber for 30 minutes and back to their home cages. Vehicle were given 8 hours later in the same day and animals were restricted in the other compartment chamber for 30 minutes. The regimen were repeated for totally 3 times. On day 4, mice were translocated into the neutral chamber and started immediately (program-controlled) a 15-min test session with free access to all compartment chambers. The total time spent and locomotor activity in every chamber (methamphetamine-associated, vehicle-associated, neutral) were automatically recorded.

**Statistical Analysis:** Data were expressed as mean ± SEM. The results were analyzed by unpaired Student’s t test, ANOVA, or nonparametric analysis whenever applicable. The P values less than 0.05 were considered statistically significant.

**Results and discussion**

1. Medium-term forced exercise—
   We found that 1-week forced exercise did not alter the post-exercise indices in the locomotor activity or in the elevated plus maze. In contrast, we observed conflicting results in the MDMA-induced CPP (Figures 1 and 2).

2. Long-term forced exercise—
   We found no difference in the indices of locomotor activity or the elevated plus maze. However, we found that long-term forced exercise appeared to reliably attenuate the reinforcing/euphoric effects of MDMA in CPP paradigm (Figure 3). Moreover, we examined the CYP1A1 in their liver samples and observed no differences in exercise and control groups. Thus, we suspect the modulating effects of exercise on the reinforcing/euphoric effects of MDMA is via the central impact of exercise.

(四) References:


Carmack CL, Boudreaux E, Amaral-Melendez M, Brantley PJ, de Moor C. Aerobic fitness and leisure physical activity as moderators of the stress-illness relation. *Ann Behav Med* 21(3):251-7,
1999.


(五) Self evaluation

We are comfortable with the current results with regard to the compatibility of experimental execution and the proposed studies and the degree of progress. Most importantly, we found that long-term exercise seemed to mitigate the reinforcing/euphoric effects of MDMA (Ecstasy). We expect to examine the exercise effects on some other abused drug of choice to reinforce the rationale that long-term exercise may reduce the naïve anxiety levels and mitigate the attractiveness of psychomotor stimulants.
Figure 1 The effects of medium-term forced exercise on locomotor activity, elevated plus maze and MDMA-induced conditioned place preference (1)
Figure 2 The effects of medium-term forced exercise on locomotor activity, elevated plus maze and MDMA-induced conditioned place preference (2)
Long Term Exercise (1-wk adaptation, 12-wks exercise)

CPP

Mean
Exer 3  -59.2063
Con 3  258.7938
SEM
Exer 3  32.23062
Con 3  31.11383

Mean
Exer 3  28.55
Con 3  108.4375
SEM
Exer 3  39.19432
Con 3  56.41603

EPM (pre-exercise)

Mean
Exer 3  36.5
Con 3  29.88
SEM
Exer 3  3.703
Con 3  3.811

Mean
Exer 3  12.37
Con 3  10.75
SEM
Exer 3  3.703
Con 3  3.811

EPM (post-exercise)

Mean
Exer 3  13
Con 3  10.38
SEM
Exer 3  4.61
Con 3  2.97

Mean
Exer 3  32.63
Con 3  32.13
SEM
Exer 3  4.062
Con 3  7.465
Figure 3 The effects of long-term forced exercise on locomotor activity, elevated plus maze and MDMA-induced conditioned place preference