EFFECT OF TAURINE SUPPLEMENT IN CONJUNCTION WITH EXERCISE ON ANTIOXIDANT ENZYMES ACTIVITIES IN ADULT AND MIDDLE-AGED RAT BRAINS

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ผลของการได้รับทอรีนเสริมร่วมกับการออกกำลังกายต่อการทำงานของเอนไซม์ต้านอนุมูลอิสระในสมองหนูหนุ่มและหนูวัยกลางคน

นางสาวจิราพร อ่อนศรี

วิทยานิพนธ์นี้เป็นส่วนหนึ่งของการศึกษาตามหลักสูตรปริญญาวิทยาศาสตรมหาบัณฑิต
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Suranaree University of Technology has approved this thesis submitted in partial fulfillment of the requirements for a Master's Degree.

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จิราพร อ่อนศรี : ผลของการได้รับทอรีนเสริมร่วมกับการออกกำลังกายต่อการทำงานของเอนไซม์ต้านอนุมูลอิสระในสมองหนูหนุ่มและหนูวัยกลางคน (EFFECT OF TAURINE SUPPLEMENT IN CONJUNCTION WITH EXERCISE ANTIOXIDANT ENZYMES ACTIVITIES IN ADULT AND MIDDLE-AGED RAT BRAINS) อาจารย์ที่ปรึกษา : ผู้ช่วยศาสตราจารย์ ดร.รุ่งฤดี ศรีสวัสดิ์, 221 หน้า.

การศึกษาในครั้งนี้ต้องการศึกษาผลของการออกกำลังกายอย่างเดียว การให้ทอรีนเสริมและการให้ทอรีนเสริมร่วมกับการออกกำลังกายต่อการทำงานของเอนไซม์ต้านอนุมูลอิสระ และ มาลอนไดออลดีไฮด์ ซึ่งสารที่บ่งชี้ถึงสภาวะเครียดออกซิเดชันในสมองหนูหนุ่มและหนูวัยกลางคน หนูหนุ่ม และหนูวัยกลางคนเพศผู้พันธุ์วิสตาร์ ถูกแบ่งออกเป็น 2 กลุ่ม (กลุ่มที่ไม่ได้ออกกำลังกายและกลุ่มที่ออกกำลังกาย) โดยทั้ง 2 กลุ่มได้รับน้ำ 1% ทวีน 80 วิตามินอี (50 IU/กิโลกรัม) หรือ ทอรีน 800 มิลลิกรัม/กิโลกรัม ทางปากเป็นประจำทุกวันในขนาด 8 มิลลิลิตรต่อกิโลกรัมเป็นเวลา 8 สัปดาห์ หนูในกลุ่มที่ออกกำลังกายถูกให้ออกกำลังกายโดยการว่ายน้ำเป็นเวลา 30 นาที 5 วันต่อสัปดาห์ เป็นเวลา 8 สัปดาห์ ส่วนหนูในกลุ่มที่ไม่ได้ออกกำลังกายถูกปล่อยไว้ในกรงโดยไม่ได้ว่ายน้ำ ทันทีหลังฝึกทั้งหมดถูกทำให้สลบโดยใช้ยาสลบ 60 มิลลิกรัม/กิโลกรัม สมองถูกเก็บ เพื่อหาระดับของ มาลอนไดออลดีไฮด์ ไฮโดรเจนเปอร์ออกไซด์ และการทำงานของเอนไซม์ต้านอนุมูลอิสระ (ประกอบด้วย ซุปเปอร์ออกไซด์ดิสมิวเทส คะตะเลส และ กลูต้าไธโอนเปอร์ออกซิเดส) การทดลองพบว่า การให้ทอรีนเสริมร่วมกับการออกกำลังกายสามารถลดระดับของมาลอนไดออลดีไฮด์ในสมองส่วนเบซอลฟอร์เบรนและฮิปโปแคมปัสของหนูวัยกลางคน เพิ่มระดับของมาลอนไดออลดีไฮด์ในสมองส่วนสไตรเอตัมของหนูหนุ่ม และลดระดับของไฮโดรเจนเปอร์ออกไซด์ ในสมองส่วนซีรีบรัลคอร์เทกซ์ ในหนูหนุ่ม และในสมองส่วนฮิปโปแคมปัสและสไตรเอตัมของหนูวัยกลางคน ผลของการให้ทอรีนเสริมร่วมกับการออกกำลังกาย มีผลในการเพิ่มระดับเอนไซม์ต้านอนุมูลอิสระ ซุปเปอร์ออกไซด์ดิสมิวเทสในสมองส่วนสไตรเอตัมของหนูหนุ่มและหนูวัยกลางคน มีการเพิ่มระดับของเอนไซม์คะตะเลสในสมองส่วนฮิปโปแคมปัสและสไตรเอตัม การเพิ่มระดับการทำงานของเอนไซม์กลูต้าไธโอนเปอร์ออกซิเดสในสมองส่วนฮิปโปแคมปัสของหนูวัยกลางคน ทอรีนทำหน้าที่เป็นสารต้านอนุมูลอิสะ และยังทำหน้าที่ป้องกันระบบประสาทในสมองส่วนกลาง นอกจากนี้การออกกำลังกายแบบแอโรบิคยังมีประโยชน์หลายด้านทางสรีรวิทยาและการทำงานของสมอง โดยการลดการทำลายออกซิเดชัน และการเพิ่มการทำงานของเอนไซม์ต้านอนุมูลอิสระ การให้ทอรีนเสริมร่วมกับการออกกำลังกายช่วยลดสภาวะเครียดออกซิเดชัน และเพิ่มการทำงานของเอนไซม์ต้านอนุมูลอิสระ ในสมองได้ดีขึ้นทั้งในหนูหนุ่มและหนูวัยกลางคน โดยจะมี

ประสิทธิภาพในสมองหนูวัยกลางคนมากกว่า ดังนั้นการให้ทอรีนเสริมร่วมกับการออกกำลังกาย น่าจะมีผลดีต่อการลดภาวะเครียดออกซิเดชันที่มีส่วนในการเกิดโรคความเสื่อมของระบบประสาท ที่สัมพันธ์กับอายุ

สาขาวิชาปรีคลินิก
ปีการศึกษา 2561

ลายมือชื่อนักศึกษา_____
ลายมือชื่ออาจารย์ที่ปรึกษา_____
ลายมือชื่ออาจารย์ที่ปรึกษาร่วม_____

JIRAPORN ONSRI : EFFECT OF TAURINE SUPPLEMENT IN CONJUNCTION WITH EXERCISE ON ANTIOXIDANT ENZYMES ACTIVITIES IN ADULT AND MIDDLE-AGED RAT BRAINS. THESIS ADVISOR : ASST. PROF. RUNGRUDEE SRISAWAT, Ph.D. 221 PP.

TAURINE, EXERCISE, ANTIOXIDANCE ENZYME

The present study investigated the effect of exercise alone, taurine supplement alone, and taurine supplement in conjunction with exercise on antioxidant enzymes activities and malondialdehyde (MDA), oxidative stress marker, in adult and middle-aged rat brains. Adult and middle-aged male Wistar rats were divided into two groups (sedentary and exercise groups), rats in both groups were daily orally administered with DDD water, 1% Tween 80, vitamin E (50 IU/kg), or taurine (800 mg/kg) for 8 weeks at a volume of 8 ml/kg. Rats in the exercise group were submitted to swimming sessions (30 minutes) at 5 days/week for 8 weeks and rat in the sedentary groups were left in cages without swimming. Immediately, after last training, rats were anesthetized by pentobarbital sodium (60 ml/kg). Brains were collected to determine level of MDA, H_2O_2, and antioxidant enzymes activities (including SOD, CAT, and GPx). The results demonstrated that taurine supplement in conjunction with exercise reduced MDA levels in cerebral cortex of adult rats, and in basal forebrain and hippocampus of middle-aged rats, increased MDA levels in striatum of adult rats, reduced H_2O_2 levels in cerebral cortex of adult rats, and hippocampus and striatum of middle-aged rats. Taurine supplement in conjunction with exercise increased levels of antioxidant enzyme SOD in striatum of adult and middle-aged rats, increased levels of CAT in hippocampus and

striatum of middle-aged rats, and increased activities of GPx in hippocampus of middle aged rats. Taurine acts as antioxidant and neuroprotective agent in central nervous system. Aerobic exercise has benefit in many physiological performance and brain function by reducing oxidative damage and enhancing antioxidant enzymes activities. Taurine supplement in conjunction with exercise is capable of reduction of oxidative stress and enhancement of antioxidant enzymes activities in adult and middle-aged rat brains, and is more effective in middle-aged rats brains. Therefore, taurine supplement in conjunction with exercise may benefit in reduction of oxidative stress which is contributed to age-related neurodegenerative diseases.

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Jiraporn Onsri

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CHAPTER I

INTRODUCTION

1.1 Introduction

Aging is the accumulation of changes which increase the risk of death. Environmental conditions, genetic factors and disease are the causes of aging. Furthermore, oxidant/antioxidant status may also cause the degenerative changes encountered in aging. Oxidative stress is proposed as a key component in the aging process (Harman, 1956). Aging is often accompanied by cognitive impairment, loss of memory and an increased susceptibility to neurodegenerative disorders (Mattson and Magnus, 2006). Aging is an inevitable biological process that is characterized by a general decline in the physiological functions. In 1956, Harman suggested that free radicals produced the process of aerobic respiration cause cumulative oxidative damage; over time that ultimately in aging and death (Harman, 1956). Age may lead to an increased damage from oxidative stress that could be elicited through increased reactive oxygen species (ROS), decreased antioxidant enzyme activities or a combination of both. Free radical mediated oxidation of proteins also increases with age (Berlett and Stadtman, 1997).

Brain is an organ most sensitive to oxidative stress and this is lead to the high metabolic rate and very high rates of iron and copper present in the organ. These interactions with the diffusible hydrogen peroxide and result in the generation of the very reactive hydroxyl radical that yields damage to proteins, lipids and DNA damage

(Halliwell and Barry, 2001). The number of oxidative modification of lipids, proteins, and DNA are generally used as markers of oxidative damage, which are increased with the neuropathology of aging, and in many cases suggested to be a causative factor in many different specific disorders (Esiri, 2007; Head, 2009; Martin, 2008).

Exercise can be accomplished only through the series of complex interactions within all of the interacting body systems. Physical exercise is a general term that refers to different types of physical exertions that may vary in its time, intensity and type. During physical exercise there is an elevated generation of ROS (Radak *et al.*, 2013) but regular exercise is known to improve the physiological performance of skeletal and cardiac muscle and associated with a decreased incidence of a wide range of diseases, including cardiovascular diseases, some cancers, osteoporosis, and diabetes II (McCarter, 2000; Radak *et al.*, 2005). Over the past few decades, it became clear that regular exercise can be beneficial for brain function, and could play an important preventive and therapeutic role in stroke affect, Alzheimer's disease, and Parkinson disease (Christophe Fleury *et al.*, 2002; Mattson and Magnus, 2006; Radak *et al.*, 2010). The complex effects of exercise appear to be very complex and could induce neurogenesis via neurotrophic factors, increase capillarization, reduce oxidative damage, and increase proteolytic degradation by proteasome in the deep brain regions such as thalamus, hypothalamus, and hippocampus (Cotman and Berchtold, 2002; Cotman and Engesser-Cesar, 2002; Johnson and Mitchell, 2003; Raffaella Molteni *et al.*, 2004). The findings of some earlier studies suggested that exercise, voluntary running, resulted in lower the level of oxidative stress in the brain in low vitamin E fed animals (Suzuki M *et al.*, 1983). Swimming training rats showed significant enhanced activities in Lipid peroxidation, and Glutathione peroxidase (GPx) in brain (Hara *et al.*,

1997). The antioxidant enzyme activities were dependent on regions of the brain, and the effects of exercise were also dependent on the brain portion. In some part of the brain such as the brainstem and corpus striatum, exercise training resulted in increases in Superoxide dismutase (SOD) and GPx activities (Rybak *et al.*, 1995).

Taurine, 2-aminoethanesulfonic acid is most abundant endogenous amino acid in the central nervous system (CNS) and plays various roles in our body: thermoregulation, stabilization of protein folding, anti-inflammatory actions, antioxidation, osmoregulation, calcium homeostasis and CNS development (Frosini *et al*., 2003; Huxtable, 1992; Kumar, 2009; S. Schaffer *et al.*, 2000). The brain synthesizes only a limited amount of taurine, but highest amount taurine synthesized occurs in the liver and derived from foods. It is probable that significant amounts of taurine will be required transport into the brain and then transport into those cellular compartments that sequester this amino acid (Pow *et al.*, 2002). The beneficial effects of taurine as an antioxidation in biological systems have been attributed to ability to stabilize membranes, to scavenge ROS, to reduce the peroxidation of unsaturated fatty acids , and attenuate production of lipid peroxidation that has MDA is an end product (A. Nandhini *et al.*, 2005). Taurine as a neuromodulator and have other roles such as neuroprotection from excitotoxic cell death and regulation of protein phosphorylation (Pow *et al.*, 2002). Pretreated with taurine rats prevented cisplatin-induced neuronal death in the cerebral and cerebellar cortices, caudo-putamen and hippocampus, as taurine exhibit improved performance and brain antioxidant status with decrease in acetylcholinesterase activity and oxidative stress indices when compared with cisplatin alone group (Owoeye *et al.*, 2018). Taurine could prevente sodium fluoride (NaF)-induced increase in oxidative stress (hydrogen peroxide and lipid peroxidation levels),

but increase acetylcholinesterase, and increase antioxidant enzymes activities and glutathione level in the hypothalamus, cerebrum and cerebellum of the rats (Adedara, Abolaji, *et al.*, 2017; Adedara, Olabiyi, *et al.*, 2017). Taurine treatments could prevent the alterations by ethanol, promote in superoxide dismutase and catalase activities that suggesting a modulatory role in enzymatic antioxidant defenses in zebrafish brain (Rosemberg *et al.*, 2010).

However, there was no evidence supporting the potential of taurine as a supplement in conjunction with exercise on antioxidation. Thus, the effects of taurine supplement in conjunction with exercise on oxidative stress and antioxidant enzyme activities in adult and middle-aged rat brains were investigated.

1.2 Research Objectives

1. To study the effect of taurine supplementation alone, exercise alone, and taurine supplementation in conjunction with exercise on antioxidant enzymes activities in adult and middle-aged rat brains.

2. To study the effect of taurine supplementation alone, exercise alone, and taurine supplementation in conjunction with exercise on malondialdehyde (MDA) oxidative stress marker in adult and middle-aged rat brains.

3. To study physiological role of taurine and exercise in prevention of natural degeneration of the brain.

1.3 Research Hypothesis

1. Supplementation of taurine in conjunction with exercise can enhance activities of glutathione peroxidase (GPx), catalase (CAT) and superoxide dismutase (SOD) as an antioxidant enzymes in adult and middle-aged rat brains.

2. Supplementation of taurine in conjunction with exercise can attenuate the level of an oxidative stress marker malondialdehyde (MDA) in adult and middle-aged rat brains.

1.4 Scope and Limitation of the Study

The effects of taurine supplementation in conjunction with exercise on the antioxidant enzyme activities and MDA level in hippocampus, cerebral cortex, basal forebrain and striatum were investigated in adult and middle-aged rats.

CHARPTER II

LITERATURE REVIEW

2.1 Taurine

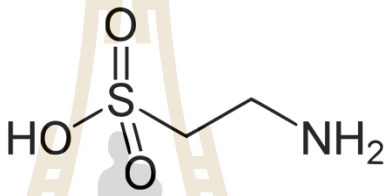

Figure 1 Chemical structure of taurine (Jang-Yen Wu and Prentice, 2010).

Taurine, 2-amino-ethanesulfonic acid (Figure 1) is one of the most abundant amino acid in mammals and presents at a high level in the mammalian brain. It also has multiple roles in physiological functions, and also importance in keeping the structural completeness of the membrane, controlling calcium binding and transport, as an osmolyte, a neuromodulator, a neurotransmitter, and a neuroprotector against L-glutamate (L-Glu) – induced neurotoxicity (Jang-Yen Wu and Prentice, 2010). Taurine may have a specific function in immature brain tissue. Taurine is an essential nutrient for cats, and possibly for mammals, so probably being essential for the growth and living of neural cells (P. Saransaari and Oja, 2000). Morphological degeneration of the ratina and tapetum lucidum was found in a dietary taurine-deficient kittens (P. Saransaari and Oja, 2000). Cardiomyopathy and the development of central retinal

degeneration was found in feline fed with taurine deficient diet (Foos and Wu, 2001). In central nervous system (CNS), Taurine play a role as a neuro-protective agent, a potent regulator for intracellular calcium homeostasis and a neurotransmitter (Jang-Yen Wu and Prentice, 2010). Taurine could protect retinal neurons from the glutamate excitotoxicity (Louzada *et al.*, 2004). Taurine is transported across the blood-brain barrier by a Na^+-dependent, carrier-mediated transport (Kang, 2000). In the perinatal period, the concentration of taurine in the brain is much higher than plasma. This ratio is expected to reverse in the adult period of life, taurine in plasma higher than the brain. This suggests that the brain has a high capacity to accumulate taurine compared to taurine synthesis capability. This is supported by the finding that the brain displays higher taurine transport capacity during neonatal, relatively GABA transport cycle is quite low. Although $NaCl^-$dependent taurine transporters are the main factor increasing taurine uptake in the brain, taurine leakage from the brain to plasma appears to be primarily mediated by $GABA_2$ transporters (Zhou *et al.*, 2012) Taurine can reduce the hepatic failure by reducing plasma activities of Aspartate aminotransferase (AST) and alanine aminotransferase (ALT), are biomarkers for liver disease. Therefore, taurine can potentially be good for liver (Louzada *et al.*, 2004).

In general, it is believed that taurine exerts neuroprotective functions because of its function in reducing concentration of intracellular free Ca^{2+} ($[Ca^{2+}]_i$) and reduced the oxidative stress (Louzada *et al.*, 2004). Neuroprotective effects of taurine was demonstrated since, taurine could protect neural damage from neuronal excitotoxic neuronal, protect the damage by ischemia and hypoxia, and reduces Ca^{2+} influx in ischemia (Saransaari and Oja, 2000). Taurine could prevent cultured neurons and inhibit the glutamate–induced release Ca^{2+} from the internal pools (cytoplasmic and

mitochondrial) regulates calcium homeostasis (Jang-Yen Wu and Prentice, 2010; P. Saransaari and Oja, 2000). Taurine possessed an important function in the alteration of intracellular calcium homeostasis. Taurine increased mitochondrial Ca^{2+} concentration of the rat cerebral cortex, and decreased cytosolic free Ca^{2+} concentration (Saransaari and Oja, 2000). Taurine's effect in a part of neuroprotective properties depends on antioxidant action. Taurine could extend cell survival, and decrease striatal damage, and protect the partial hippocampus from damage, and protect rats cerebellar granular cells from free radical damage and recovery the ethanol-induced reduction of antioxidants that protected and reduced oxidative stress in the rats (Saransaari and Oja, 2000). The taurine might have advantageous effects in Alzheimer's disease and other neurological symptoms, taurine could raise the levels of acetylcholine in the brain and enhance memory in rats disclosed to ozone low-level of taurine was found in patients with Alzheimer's disease (Saransaari and Oja, 2000). The effect of chronic administration of taurine on hippocampus neurogenesis in aging rat brain resulted in increased cell proliferation in the dentate gyrus, increased the survival of newborn neurons, and increased neurogenesis in adulthood (Figure 2). Taurine enhanced the number of immature rat neurons (Gebara *et al.*, 2015; Saransaari and Oja, 2000).

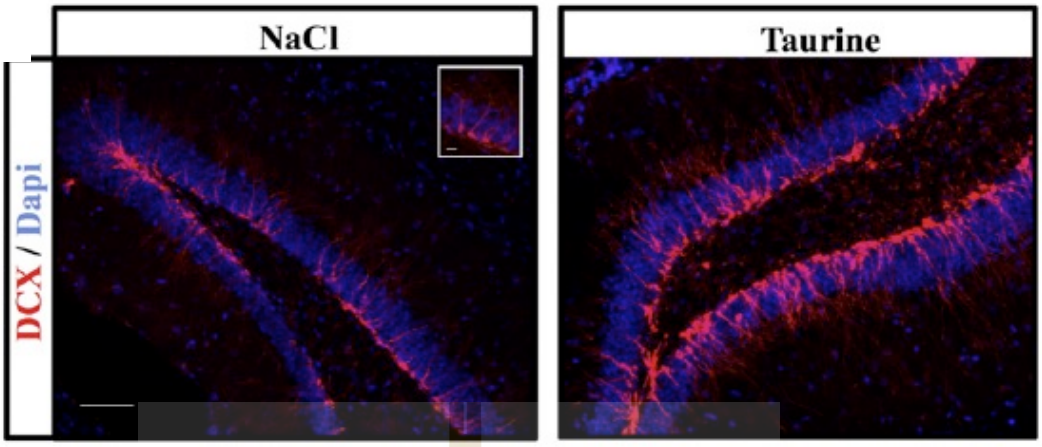

Figure 2 Taurine enhanced the number of immature neurons. Confocal maximal projection micrographs of hippocampal sections for DCX-immunostained. Inset: Higher magnification confocal microscopy of a DCX-positive cell. Dapi stained. Scale bars: 100 μm, insets 10 μm (Gebara *et al.*, 2015).

In middle-aged (13-14 months) rats taurine significantly increased the cerebellum GSH levels and significantly reduced cerebellum MDA levels compare to middle-aged control rats. The cerebellum MDA level in the young taurine group (6-7 weeks of age rat) was lower than the young control group (6-7 weeks of age rat) (Zuhal Yildirim and Kilic, 2011).

2.2 Exercise

Acute exercise was found to induce oxidative stress since a single bout of physical exercise was proved to induce generation of ROS and nitrogen species, and the related oxidative damage. On the other hand, regular exercise was known to improve the resistance against ROS induced lipid peroxidation, and to reduce the accumulation of oxidative protein and DNA damage, as an increased protein and DNA

repair (Zsolt Radak *et al.*, 2001). Untrained men received the training programs of endurance training, resistance training, and concurrent training for 8 weeks showed decreased oxidative stress and increased enzymatic and nonenzymatic antioxidant capacities in plasma (Azizbeigi *et al.*, 2014).

Long-term exercise rats revealed a lower level of ROS with a reduced amount of protein carbonyls in the hippocampus. Long-term physical exercise (treadmill running) induced an up-regulation of SOD-1 and GPx enzymes, as an antioxidant activities and protected neurons against oxidative stress in the hippocampus at the early stage of aging, and up-regulated protein synthesis, which improved cellular ability to remove damaged proteins post synthetically by free radicals in the cell (Marosi *et al.*, 2012).

Increase in CAT and GPx activities were demonstrated in the hippocampus of swimming rats (total training period of 12 weeks with five training days per week). and was irrespective of the age (Devi and Kiran, 2004).

2.3 Aging

2.3.1 Aging Theories

Many theories have been proposed to explain the process of aging, the traditional aging theories hold that aging is not an adaptation or genetically programmed aging theories. Modern biological aging theories in humans fall into two main categories: programmed and damage or error theories. The programmed theories which imply that aging follows a biological timetable, perhaps a continuation of the one that regulates child development. This regulation would be depending on changes in gene expression that effect to maintain the systems, repair, and defense responses.

The damage (error) theories emphasize environmental assaults to living organisms that induce cumulative damage at many levels, that cause of aging. (Mladen Davidovic *et al*., 2010).

- Programmed Theory

Davidovic *et al.* discuss the aging process of genetic instability in aging and dynamics. Sequential switching of certain genes will switch off and thereby cause aging, with senescence being defined as the time when age-related deficits are manifested (Mladen Davidovic *et al*., 2010).

- Endocrine Theory

Dr. van Heemst has discusses the potential mechanism underlying IIS and aging process, biological clocks act through hormones that control the speed of aging. Recent studies confirm that aging is hormonally regulated and that the evolutionarily conserved insulin/IGF-1 signaling (IIS) pathway plays a key role in the hormonal regulation of aging (Heemst, 2010).

- Immunological Theory

The immune system becoming defective over time, which leads to an increased vulnerability to infectious when infections were the leading cause of aging and mortality. It is well documented that the effectiveness of the immune system peaks at puberty and gradually declines with the advance in age. For example, as one grows older, antibodies lose their effectiveness, and fewer new diseases can be combated effectively by the immune system, this leads to cellular stress and eventually cell death (Cornelius, 1972).

- Wear and Tear Theory

Cells and tissues have vital parts that wear out from repeated use and resulting in aging. Like components of an aging car, parts of the body ultimately wear out from repeated use. First proposed scientifically by Dr. August Weismann, in 1882, Sounds like a perfectly reasonable to many people even now, because this is what happens to most common things around them (Brys *et al*., 2007).

- Rate of Living Theory

The higher rates of oxygen basal metabolism cause the shorter its lifespan. The rate-of-living theory of aging while helpful is not completely adequate in fully explains lifespan (Hulbert *et al.*, 2007).

- Cross-Linking Theory

The cross-linking theory of aging was proposed by Johan Bjorksten. The cross-linking theory, in aging, has an accumulation of cross-linked proteins cause cellular damage, slowing down bodily processes. Recent findings show that cross-linking reactions are involved in the age-related changes in the studied proteins (Bjorksten and Tenhu, 1990).

- Free Radicals Theory

This theory, which was first introduced by Dr. Gerschman in 1954, but was developed by Dr. Denham Harman (Harman, 1956; Rebeca Gerschman *et al.*, 1954), proposes that superoxide and other free radicals cause cell damage and damage to the macromolecular componants, giving rise to accumulated damage-causing cells, and eventually organs, to stop functioning. The macromolecules such as nucleic acids, lipids, sugars, and proteins are susceptible to free radical attack. Nucleic acids can get an additional base or sugar group; single-strand breaks or double strand breaks in the

backbone and cross-link of the other molecule. The body does possess some natural antioxidants enzymes, which assist to curb the harmful build-up of these free radicals, without which cell death rates would be highly increased, and subsequent life expectancies would decrease. This theory has been bolstered by experiments in which rodents fed antioxidants achieved greater mean lifespan. Igor Afanas'ev shows review that reactive oxygen species (ROS) as the most important signalling that responsible for the development of cell senescence and organismal aging and that ROS signalling might be considered as the further development of a free radical theory of aging (Afanas'ev, 2010).

Aging is a complex process involving interacts of both physiological and behavioral factors. As people grow older, the basal metabolic rate slows down, blood pressure rises, and maximum heart rate, cardiac output, and maximal oxygen consumption decrease, and overall muscle mass decrease. Functional changes in aging that occur include a decline in cognitive function, lung lead to decreased compliance, and loss of bone mass. A large sign of aging is oxidative stress and a significant volume of evidence of oxidative stress is an important pathogenic factor in Alzheimer's patients (Figure 3). In response to a stress or disuse, the age-related in physiologic reserves decreased causing a loss a homeostasis or inability to finish a work requiring near-maximal exertion (Clarke, 1977). Physiological changes involved in aging (Clarke, 1977) are as follows:

- decreased muscle mass, strength, power, endurance, speed of contraction, mitochondrial function, and oxidative enzyme activity,

- decreased maximal and submaximal aerobic fitness, cardiac output, impaired endothelial relaxation, and low heart rate variability (autonomic dysfunction),
- elevated arterial and myocardial stiffness and systolic and diastolic blood pressure,
- decreased nerve conduction velocity, impairments in proprioception and balance, slower gait velocity, and gait instability,
- decreased sensitivity insulin and glucose tolerance,
- increased of visceral fat mass, total body fat, and intramuscular lipid accumulation,
- impaired immune system,
- decreased tissue elasticity, thin collagen fibrils, collagen cross-linkage, and shortening and weakens the tendon, and
- bone mass, strength, and density reduces (Clarke, 1977).

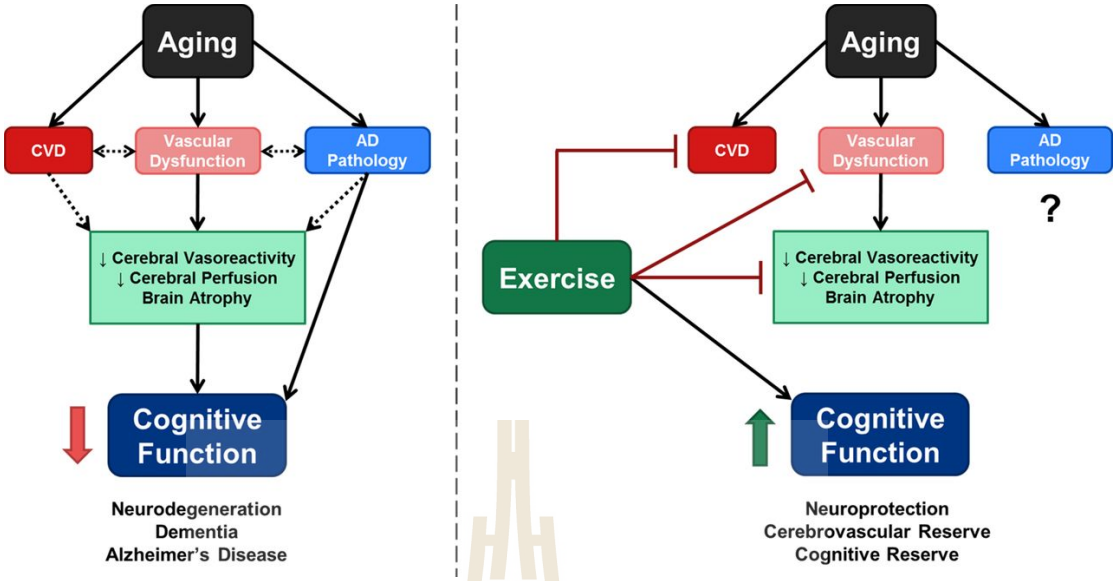

Figure 3 The potential interactions and ideas of how variables associated with aging may interact to cognitive processes affect and how exercise may inhibit this process (Barnes, 2015).

2.3.2 Neurodegenerative Disorders

The brain is exposed throughout life to oxidative stress, and certain diseases of the brain and nervous system are thought to involve free radical processes and oxidative damage, either as a primary cause or as a consequence of disease progression. Common neurodegenerative disorders are increasing as life expectancy increases. Ageing is considered to be a stochastic process combining predictable and random effects that caused to the accumulation of unrepaired cellular damage, weakening cellular repair and compensatory mechanisms. Lifestyle and the effects of the environment means that 75% that are most accounted individual variation in ageing , with genes accounting for only 25% of variability (Kirkwood, 2003). Neurodegeneration appears at different levels in normal ageing brain associated degenerative diseases. The brain is a very active organ with a fixed average energy cost per neuron (Herculano-Houzel, 2011).

Ageing is associated linked mitochondrial dysfunction, increased production of ROS, which may lead to genomic instability and DNA mutations, with telomere reduction is linked to disease evolution in some (Migliore and Coppede, 2009). Proteasome activity declines during aging that degrade damaged or ubiquitinated proteins leading to an increase in abnormal deposition of cellular brain proteins (Tai and Schuman, 2008). Aging is associated with a reduction in efficiency of chaperones and imbalance of autophagy recycling, change caused by inflammatory, complement system, microglial activation and an impaired response to brain injury, to recover from damage (Cherra and Chu, 2008). In the aging brain have a progressive iron accumulation, rendering the cell more susceptible to toxins (Zecca *et al*., 2004). Reductions tyrosine hydroxylase and dopamine in striatal, a reduction in the number of pigmented neurons in the substantia nigra and reduced dopamine receptor density, a related change with age. Although there is a loss of pigmented neurons with aging, there is evidence remaining neurons exhibit hypertrophy, as the possible compensatory mechanisms. All of these age-related changes are relevant to pathophysiology of all neurodegenerative diseases (Rudow *et al.*, 2008).

2.3.3 Alzheimer's Disease

Brain diseases contribute a major cause high morbidity and leading cause of death all over the word. Accumulation of oxidative stress and reactive oxygen species (ROS) in the pathophysiology was linked to neurodegenerative diseases as Alzheimer's disease (AD) and Parkinson disease (PD) (Figure 4) (Guerra-Araiza *et al*., 2013). Reactive oxygen species induced cellular damage and cell death (Andersen, 2004; Christophe Fleury *et al.*, 2002). Oxidative stress is defined as the imbalance between the generation of reactive oxygen/nitrogen species and ability of the cells to neutralize then by the antioxidant defence. Main source of reactive oxygen species is in the electron transport chain at the mitochondrial inner membrane where energy is produced in the form of ATP. Other sources of reactive oxygen species and reactive nitrogen species in brain are astrocytes and microglia that produce reactive species when activated and also in catalysed reaction by redox active metal ions such as copper and iron in the brain (Doorn and Petersen, 2003).

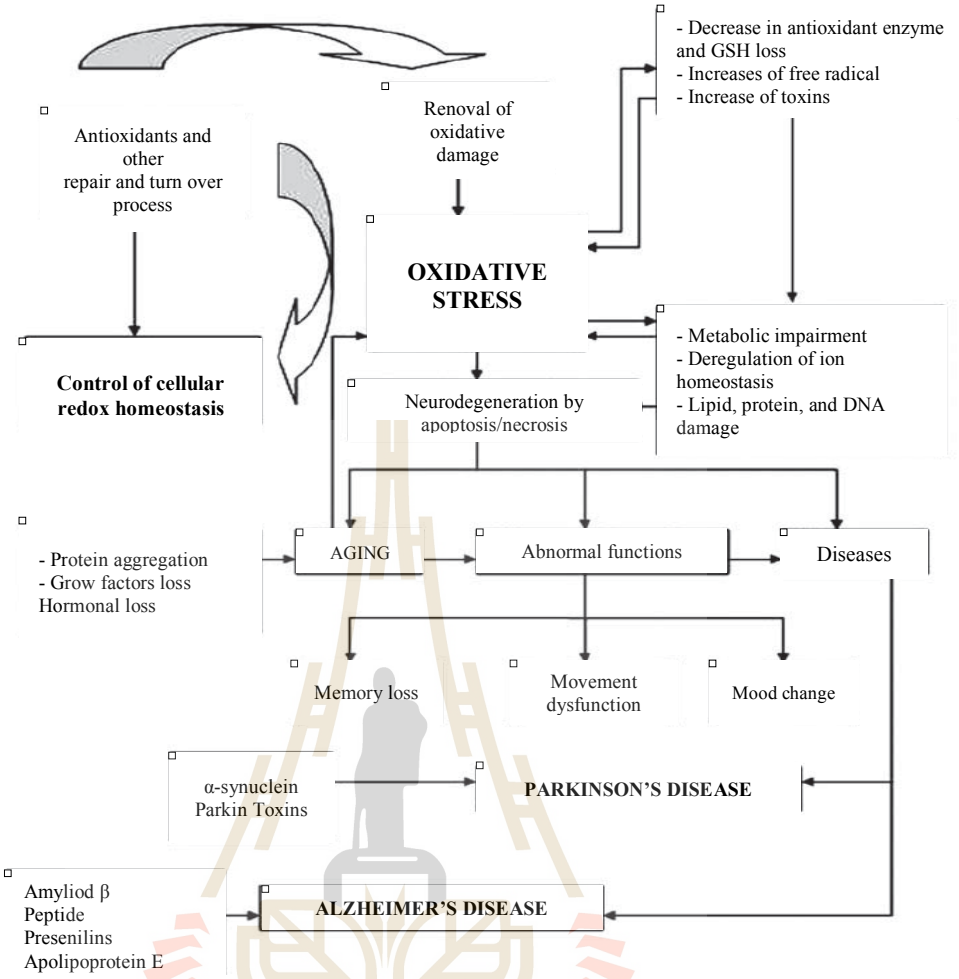

Figure 4 Oxidative stress is a common mechanism of neuronal injury in aging brain and neurodegenerative diseases and occurs in several different rate and intensity in different conditions. Oxidative stress produces neuronal damage that effects to normal functions of the central nervous systems (Guerra-Araiza *et al.*, 2013).

Oxidative stress plays a key role in the Alzheimer's disease. The brain in Alzheimer's disease has been enhanced lipid peroxidation and weakness to free radical attack to brain membrane phospholipids with high content of polyunsaturated fatty acids. Degenerative changes occurs when increased lipid peroxidation in the Alzheimer's disease (Bo Su *et al.*, 2008; William R. Markesbery, 1999). Reactive

oxygen species (ROS), such as the superoxide anion ($O_2^{\cdot -}$), hydrogen peroxide (H_2O_2), and hydroxyl radical ($OH^{\cdot}$) are produced by products of aerobic metabolism in mitochondria and can damage to DNA, lipids, and proteins, its can damage to macromolecules and can accumulate with age and may contribute to senescence and degenerative diseases associated with aging (Denham Harman, 1956).

Oxidative modifications in aging created from many ageing-related neurodegenerative diseases. Learning and memory functions in the rats are modified by free radicals and the first brain regions to go through degenerative with aged are the cerebral cortex and hippocampus. Oxidative stress and oxidative damage in the aging brain leads to increase lipid peroxidation and reduce antioxidant defences. Cells continuously generate free radical and reactive oxygen species (ROS) in a pathway of metabolic processes. Effective mechanisms to control reactive oxygen species among which is antioxidant enzymes, an elaborate antioxidant defence system consisting of endogenous enzymes such as superoxide dismutase (SOD), catalase (CAT), glutathione peroxidase (GPx), and non-enzymatic antioxidants, including vitamin A, vitamin E, and vitamin C, glutathione (GSH), ubiquinone, melatonin, and flavonoids by assist to scavenge reactive oxygen species and prevent deleterious effects in the cells. Effects of enzymatic antioxidants such as SOD, which hastens the dismutation of $O_2^{\bullet -}$ to H_2O_2, catalase and glutathione peroxidase (GPx), which convert H_2O_2 to water (Beckman and Ames, 1998; Christophe Fleury *et al.*, 2002; Devi, 2009).

In aged running mice showed significant increase in number of new neurons compared to sedentary mice (van Praag *et al.*, 2005). Many studies demonstrated that physical activity could improve cognitive performance in older rats through decline of oxidative stress. Swimming for 12 weeks effectively increased the antioxidant activity

enzyme could improve learning and memory in older rats. Exercise increased the neuronal number in the hippocampus in older rats and reduced the brain tissue loss in the humans (Devi, 2009; Stanley J. Colcombe *et al.*, 2003). Swimming in aging rat model successfully improved learning, memory tasks, and up-regulated in the antioxidant activities in the brain (Jolitha *et al.*, 2006). Exercise could increase cognition, neurogenesis, and angiogenesis (Figure 5) (Cotman *et al.*, 2007). Running enhanced learning and increased hippocampal neurogenesis in aged mice. Cognitive impairment in older may be results of decrease in dentate gyrus neurogenesis and exercise could improve cell proliferation in young and aged animal runners cell survival returned to the levels in young controls. In older (van Praag *et al.*, 2005). After 15-week training period in 15 months old, reactive oxygen species and protein carbonyls decline in hippocampus and SOD and intracellular and mitochondrial GPx level in exercise group was higher than sedentary group. Exercise was effective to elevate levels of intracellular antioxidant enzymes in aging and reduce the level of free radical in the hippocampus. Exercise may be involved in the regulation of the pathway of the antioxidant gene transcription in the hippocampus (Marosi *et al.*, 2012).

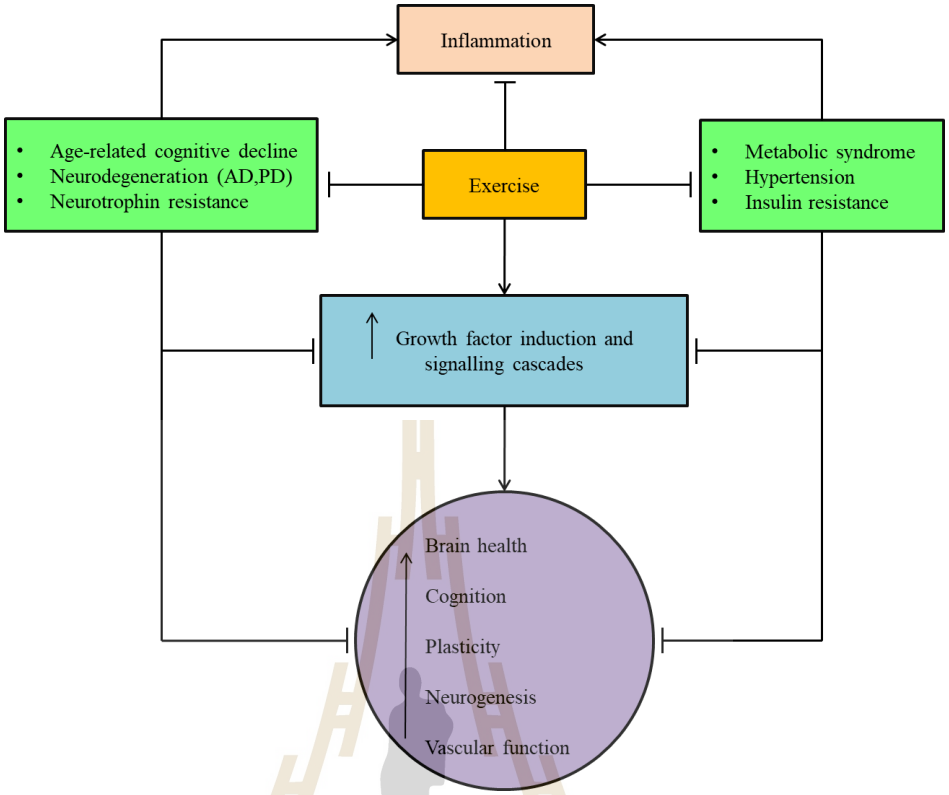

Figure 5 Exercise-induced growth factor cascades, a central mechanism mediating exercise-dependent benefits in cognitive functions, synaptic plasticity, neurogenesis and vascular function on the brain. In addition, exercise reduces peripheral risk factors for cognitive impairment such as hypertension and insulin resistance, components of the metabolic disorders that converge to increase the risk for brain dysfunction and neurodegeneration. Inflammatory, which can reduces growth factor signaling, exacerbate the metabolic disorder and accelerate cognitive decline, is reduced by exercise. Overall, exercise induces growth factor cascades and reduces peripheral risk factors for cognitive impairment, all of which converge to improve brain healthy and function, and to delay the onset of and slow down the decline in neurodegenerative disorders including Alzheimer disease (AD) and Parkinson's disease (PD) (Cotman *et al.*, 2007).

2.4 Antioxidants

Antioxidant activity is a defense mechanism of free radical-induced oxidative stress. Antioxidant activity means a reaction of inhibiting oxidation with molecule in another chemical process. The antioxidant can prevent or decrease to cell damage. The main function of antioxidant is to neutralize the production of free radicals. There are two main groups of antioxidant which are endogenous antioxidants and exogenous antioxidants. Antioxidants can be divided into two categories: enzymatic antioxidants and non-enzymatic antioxidants (Egbuna, 2017). Enzymatic antioxidant defence system includes superoxide dismutase (SOD), catalase (CAT), and glutathione peroxidase (GPx) and non-enzymatic antioxidant defence system includes alpha-tocopherol (Hardie *et al*., 1990), glutathione, carotenoids, ascorbic acid, and flavonoids (Lu *et al.*, 2014).

2.4.1 Enzymatic Antioxidants

Enzymatic antioxidants defences contain many enzymes that catalyse reactions of ROS degradation, such as superoxide dismutase (SOD), glutathione peroxidase (GPx), and catalase (CAT) (Hardie *et al.*, 1990).

- Superoxide Dismutase (SOD)

Superoxide dismutase (SOD) is the enzyme that play a vital antioxidant role in human body and presents in nearly all aerobic cells and in extracellular fluids (Johnson and Giulivi, 2005). SOD is the enzyme that catalyses the decomposition of superoxide anion into oxygen and hydrogen peroxide. SOD is a group of the metal-containing enzyme. SOD contains metal ion cofactors at the active site, depends on the isozyme

(copper, zinc, manganese or iron). SOD contains two main forms in the human cell types: copper zinc SOD (Cu-Zn-SOD) that can be found primarily in the cytosol, and manganese SOD (MnSOD) that can be found predominantly in the mitochondria. (Bannister *et al.*, 1987).

- Catalase (CAT)

Catalase (CAT) is a common enzyme present in all living organisms exposed to oxygen including bacteria, plants, and animals. CAT is found in peroxisomes, the organelle found in virtually all eukaryotic cells. CAT helps prevent damage to cell and tissue from oxidative stress can damage by reactive oxygen species. CAT is the enzyme catalyse the conversion of hydrogen peroxide to water and oxygen by using ion or manganese cofactor, the only substrate is hydrogen peroxide (Hardie *et al.*, 1990).

- Glutathione Peroxidase (GPx)

Glutathione (GSH) system contains the glutathione, glutathione reductase (GR), glutathione peroxidase (GPx), and glutathione S-tranferase (GST). GSH is an important antioxidant found in all aerobic cells and it neutralizes free radical. GSH is an enzyme family of the selenium-containing enzyme that catalyses the breakdown of hydrogen peroxide (H_2O_2) and organic hydroperoxides. GSH is changed to its oxidized form, glutathione disulphide (GSSG). The primary role of the GPx activity is to protect oxidative damage of the cell. GPx reduced lipid hydro-peroxides to their corresponding alcohols, and free hydrogen peroxide to water. Especially for toxic hydroperoxide, GPx is helping to prevent harmful water. GPx has several isozymes encoded by different genes (Muller *et al.*, 2007).

2.4.2 Non-enzymatic Antioxidants

Non-enzymatic antioxidants (such as vitamin C and vitamin E) are widely used in nutritional supplement. They can be present in the animal cells and various plants. They can assist cell proliferation from damage caused by free radicals that contribute to many diseases (Hardie *et al.*, 1990).

- Vitamin E or Alpha-tocopherol

Vitamin E or alpha-tocopherol is one of major antioxidant in fruit, vegetable, and animals. Some animals are commonly known as the good source of vitamin E such as salmon (*Salmo salar* L.) (Hardie *et al.*, 1990), and rainbow trout (*Salmo gairdneri* Richardson) (Blazer and Wolke, 1984). Vitamin E can prevent oxidative stress in the body and protect against many conditions such as cancer and heart disease (I-Min Lee, 2005). In aging rats, reduction of total SOD was found in cerebral cortex, but high total SOD in the hippocampus. Vitamin E elevated SOD in swimming exercise old rats. Mn-SOD in cerebral cortex increased in exercise middle-age and old rats. Cu-Zn-SOD in hippocampus increased in adult rats supplemented with vitamin E and swimming. Age-related and region-specific increased in protein carbonyl (PrC) content with decreased sulphydryl (P-SH) was demonstrated. Vitamin E could reduce PrC and advance oxidation protein products (AOPPs) in all ages, and appreciably in the hippocampus and cerebellum (Jolitha *et al.*, 2006).

2.5 Lipid peroxidation

Free radical-mediated lipid peroxidation has been implicated in a number of human diseases. Lipid peroxidation refers to oxidative deterioration of polyunsaturated lipids. It is the process whereby free radicals "steal" electrons from the lipids in cell membranes, resulting in cell damage. Damage of cell membrane will cause increase permeability to sodium ions, rapid in flux of calcium, osmotic entrance of water into the cell leading to cell damage. This process proceeds by a free radical chain reaction mechanism. It most often affects polyunsaturated fatty acids (PUFA) (Grotto *et al.*, 2009; Gueraud *et al.*, 2010). Biomarker of lipid peroxidation includes malondialdehyde (MDA), 4-hydroxy-2-nonenal (4-HNE), and Acrolein (ACR) (Gueraud *et al.*, 2010).

2.5.1 Malondialdehyde (MDA)

The enzymatic and the free-radical peroxidation of PUFAs which contain at least three double bonds, like arachidonic acid, could cleave to the bis-aldehyde malonaldehyde (MDA, Figure 6). MDA is a major aldehyde derived from lipid peroxidation and has been described in lipid peroxidation studies and reported as a biomarker of the peroxidation of ω-3 and ω-6 fatty acids (Shibamoto, 2006). Brain has a high amount of PUFAs and high content of free ions. Peroxidation of membrane lipids can cause impairment of membrane potential, decreased fluidity, inactivation of membrane-bound receptors and enzymes, increased permeability to ions and possible eventual membrane rupture (Gutteridge, 1995). After 16 weeks of swimming exercise period could improve neuronal number in the hippocampus which was associated with decrease in lipid peroxidation and hydrogen peroxide in adult and middle-aged rats (Abhijit *et al.*, 2018).

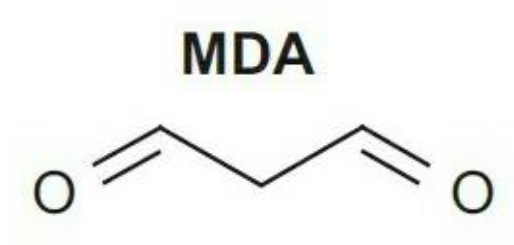

Figure 6 Structure of malondialdehyde (MDA) (Gueraud *et al.*, 2010).

2.5.2 4-hydroxy-2-nonenal (4-HNE)

4-hydroxy-2-nonenal (4-HNE) (Figure 7) is a representative compound arising from the peroxidation of ω-6 fatty acids and as a major membrane lipid peroxidation product. 4-HNE was formed under various conditions like auto-oxidation and stimulated microsomal LPO (Uchida, 2003).

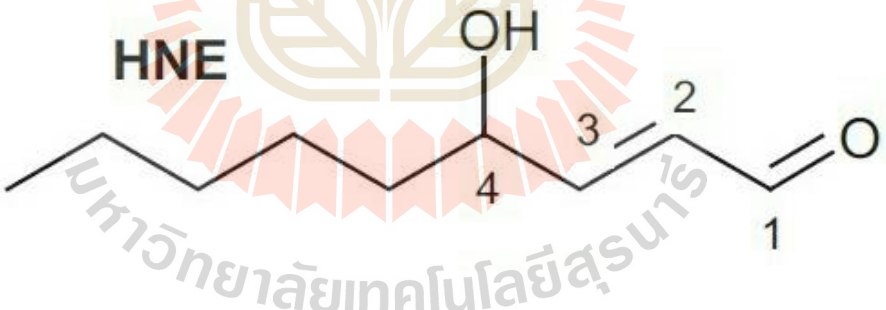

Figure 7 Structure of 4-hydroxy-2-nonenal (4-HNE) (Gueraud *et al.*, 2010).

2.5.3 Acrolein (ACR)

Acrolein (ACR) (Figure 8), a highly reactive unsaturated aldehyde and also a result of lipid peroxidation, is carcinogenic aldehyde found everywhere in the environment, by reacts with lysine residues in various proteins (Kang, 2013).

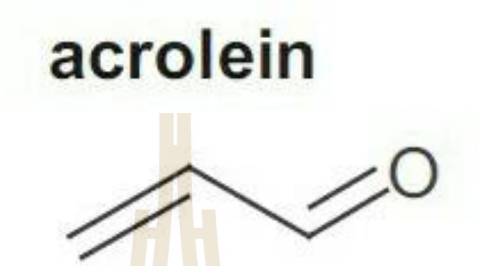

Figure 8 Structure of Acrolein (ACR) (Gueraud *et al*., 2010).

CHAPTER III

MATERIALS AND METHODS

3.1 Preparation of Taurine

Taurine was purchased from Sigma-Aldrich Chemical (St. Louis, USA). Taurine solution (100 mg/ml) was prepared by dissolving 800 mg of taurine in 8 ml of double deionized distilled (DDD) water at 60 ºC.

3.2 Preparation of Vitamin E

Vitamin E was purchased from Sigma-Aldrich Chemical (St. Louis, USA). Vitamin E was prepared by dissolving 50 IU of vitamin E in 8 ml of 1% Tween 80.

3.3 Animals

Male albino rats of Wistar strain (8 weeks old and 15 months old) were obtained from Institutional Animal Care Unit, Suranaree University of Technology (SUT), Nakhon Ratchasima Province, Thailand. Animals were housed 1-2 animals per cage and maintained at standard laboratory conditions (a temperature of 20±1 ºC and under a daily photoperiod of 12 h-light and 12 h-dark cycles) with *ad libitum* food and water. All studies were conducted with permission from the SUT Animal Care and Use

Committee. Table 1 tries to estimate the relative aged of the rats in relationship to the human (Andreollo *et al*., 2012).

Table 1 The rat's age in months and it relationship in years with human.

Rat's age in months	Human's age in years
6 months	18 years
12 months	30 years
18 months	45 years
24 months	60 years
30 months	75 years
36 months	90 years
42 months	105 years
45 months	113 years
48 months	120 years

3.4 Experimental designs

3.4.1 Effects of Taurine Supplement in Conjunction with Exercise on Antioxidant Enzymes Activities, Malondialdehyde Levels and Hydrogen Peroxide levels in Adult Rat Brains.

After one week of acclimatization, 8 weeks old male albino rats of Wistar stain (n=64) were randomly allocated to 4 sedentary groups (Se), which did not undergo physical activity, and 4 exercise groups (Ex), which were submitted to swimming exercise (Figure 9). Sedentary groups received a daily oral supplementation of 8 ml/kg of DDD water (Ad DDDW+Se, n=8), 8 ml/kg of 1% Tween 80 (Ad 1% Tween 80+Se,

n=8), 50 IU/8 ml/kg of vitamin E (Ad Vit E+Se, n=8) (Jolitha *et al*., 2006), or 800 mg/ 8 ml/kg of taurine (Ad Tau+Se, n=8) (Qiao *et al*., 2015). Exercise groups received a daily oral supplementation of 8 ml/kg of DDD water (Ad DDDW+Ex, n=8), 8 ml/kg of 1% Tween 80 (Ad 1% Tween 80+Ex, n=8), 50 IU/8 ml/kg of vitamin E (Ad Vit E+Ex, n=8) (Jolitha *et al*., 2006), or 800 mg/8 ml/kg of taurine (Ad Tau+Ex, n=8) (Qiao *et al*., 2015). Figure 9 shows experimental design of adult rats.

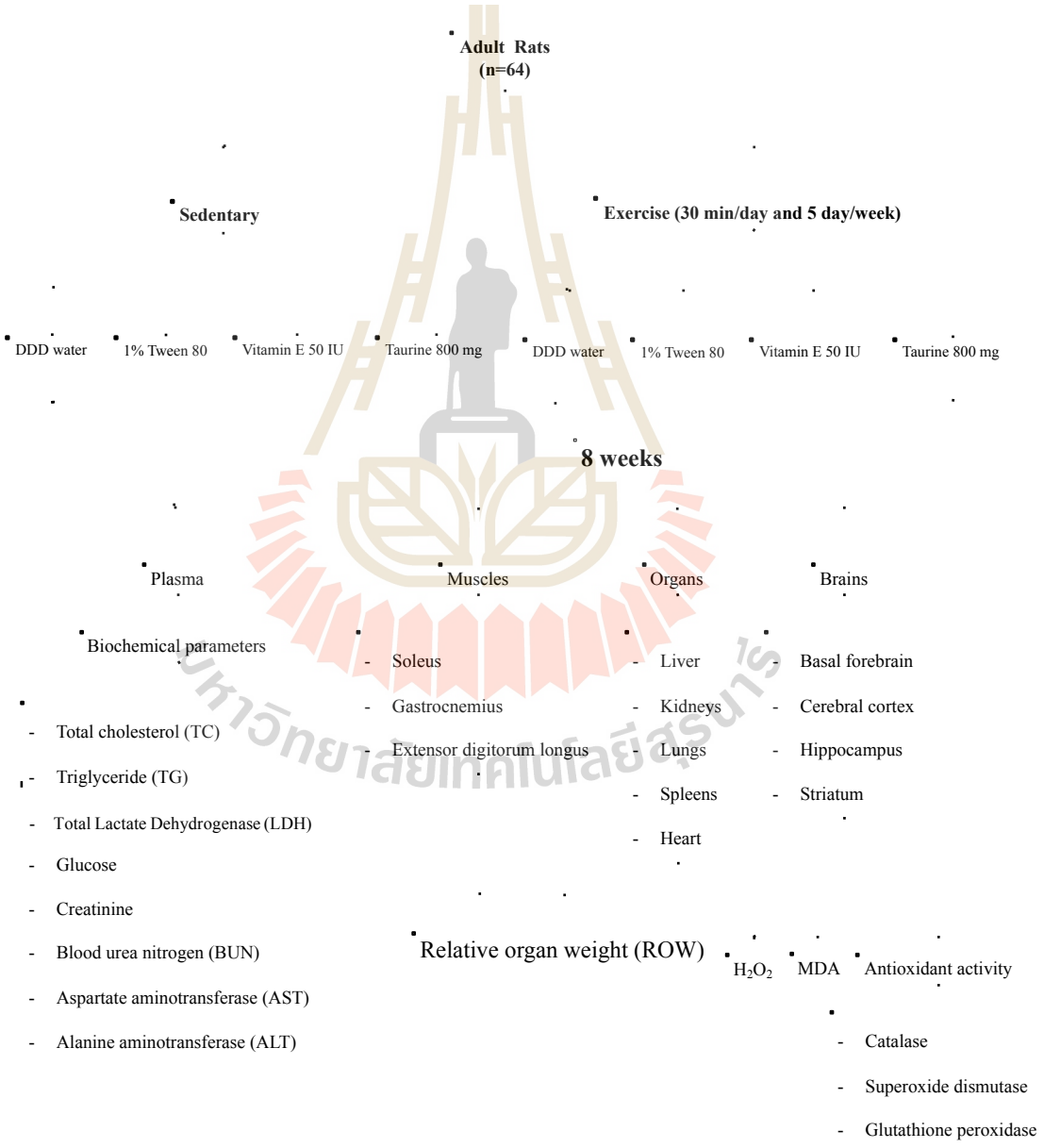

Figure 9 Schematic showing the experimental design of adult rats.

3.4.2 Effects of Taurine Supplement in Conjunction with Exercise on Antioxidant Enzymes Activities, Malondialdehyde Level and Hydrogen Peroxide Level in Middle-Aged Rat Brains.

After one week of acclimatization, male albino rats of Wistar strain (15 months old, n=64) were randomly allocated to 4 sedentary groups (Se) and 4 exercise groups (Ex) (Figure 10). Sedentary groups received a daily oral supplementation of 8 ml/kg of DDD water (Md DDDW+Se, n=8), 8 ml/kg of 1% Tween 80 (Md 1% Tween 80+Se, n=8), 50 IU/8 ml/kg of vitamin E (Md Vit E+Se, n=8) (Jolitha *et al*., 2006), or 800 mg/8 ml/kg of taurine (Md Tau+Se, n=8) (Qiao *et al.*, 2015). Exercise groups received a daily oral supplementation of 8 ml/kg of DDD water (Md DDDW+Ex, n=8), 8 ml/kg of 1% Tween 80 (Md 1% Tween 80+Ex, n=8), 50 IU/8 ml/kg of vitamin E (Md Vit E+Ex, n=8) (Jolitha *et al*., 2006), or 800 mg/8 ml/kg of taurine (Md Tau+Ex, n=8) (Qiao *et al.*, 2015). Figure 10 shows experimental design of middle-aged rats.

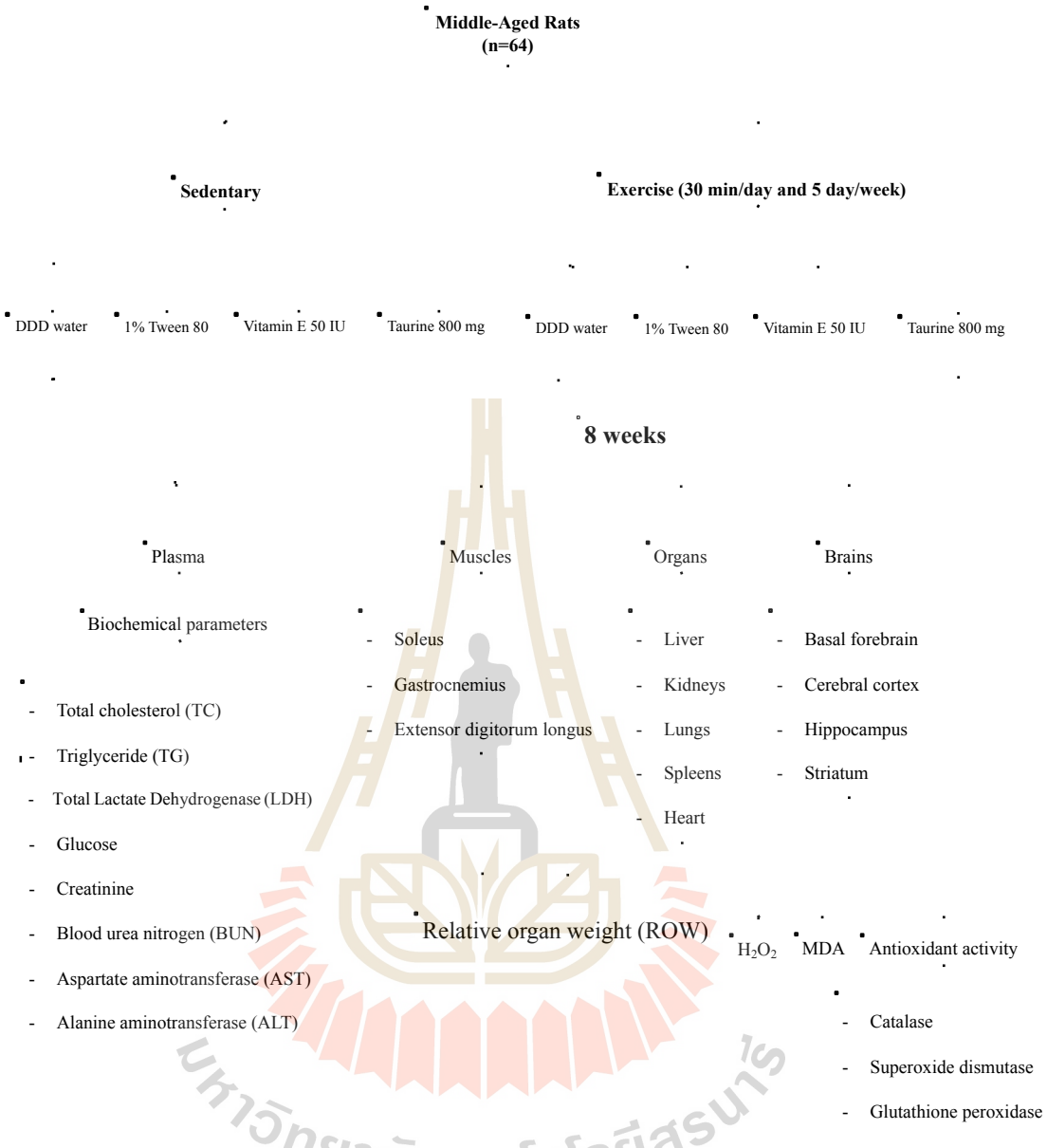

Figure 10 Schematic diagrams showing the experimental design of middle-aged rats.

3.4.3 Exercise Training Protocol

Training protocol for both adult and aging rats was followed the protocol as described earlier (Devi and Kiran, 2004) with some modifications. The experiments were performed during the day (8:00 - 17:00 hr.). Briefly, thirty minutes after oral supplementation of DDD water, 800 mg/kg of taurine, 1% Tween 80, and 50 IU/kg of

vitamin E, rats in exercise groups swam with 3% of their body weight tied load to their tails. Initially, they were swimming individually in a plastic pool (90 cm × 45 cm × 45 cm) filled with fresh water maintained at 37 ± 1 ºC, approximately 60 cm depth to make sure that rats can't support themselves by touching the bottom with their tails. Rats were made to exercise for 5 min per day with a progressive increase to 30 min per day (1^{st} = 5 min/day, 2^{nd} = 10 min/day, 3^{rd} = 15 min/day, 4^{th} = 20 min/day, and 5^{th} = 30 min/day in first week) for a total training period of 8 weeks with 5 training days per week. Rats in sedentary groups were restricted to cage activity. Rats' body weight was recorded weekly.

At the end of 8 weeks training period, all rats were fasted overnight. Immediately after last training, rats were anesthetized with pentobarbital sodium (Nembutal, Ceva Sante Animale, Libourne, France) at a dose of 60 mg/kg (i.p.).

Blood samples were collected *via* cardiac puncture into heparinized tubes and were centrifuged at 2000 × g at 4 ºC for 5 min. The plasma was obtained for analysis of biochemical parameters [total cholesterol (TC), triglyceride (TG), total lactate dehydrogenate (LDH), glucose, creatinine, blood urea nitrogen (BUN), aspartate aminotransferase (AST), and alanine aminotransferase (ALT)] using automate analyzers (Wang *et al.*, 2012). Immediately after blood collection, rats were perfused with ice-cold normal saline solution *via* cardiac puncture until clearly from blood, the organs (brain, liver, kidneys, lungs, spleen, heart, soleus muscles, extensor digitorum longus muscles (EDL), and gastrocnemius muscles) were quickly dissected and weighed individually.

The relative organ weight (ROW) of each organ was calculated using the following formula:

$$ROW = (\text{organ weight} \div \text{body weight}) \times 100$$

3.5 Brain Tissue Preparation

Immediately after collecting the organs, brains were removed, rinsed in ice-cold normal saline solution and weighed. Basal forebrain (BF), cerebral cortex (CC), hippocampus (HC), and striatum (ST) were separated and frozen on dry-ice. The brain tissue was homogenized with ice-cold 10% of 0.1 M phosphate buffer (pH 7.4) containing 0.1 mM EDTA at 1,000 rpm for 15 min at 4 ºC (Devi and Kiran, 2004).

The rat brain homogenates were used to determine activities of antioxidant enzymes (superoxide dismutase (SOD), glutathione peroxidase (GPx), and catalase (CAT)), malondialdehyde (MDA) level and hydrogen peroxide (H_2O_2) level.

3.5.1 Determination of Protein

The measurement of protein was adapted from the method of Lowry (1951). Briefly, 30 µl of samples and standards were pipetted into triplicate wells of 96-well plates and 100 µl of mixed solution A and solution B (1:3) was added. The mixture was incubated at room temperature for 60 minutes. After incubating, 150 µl of 2 N Folin-Ciocatteau reagent was added, mixed well and incubated again at room temperature for 30 minutes. After incubation, optical density (O.D.) of the mixture was read at 650 nm by Benchmark Plus Microplate Spectrophotometer.

Calculation:

$$\Delta A_{650\ nm\ Standard} = A_{650\ nm\ Standard} - A_{650\ nm\ Blank}$$

Plotted the $\Delta A_{650\ nm}$ Standard against protein concentration on the standard graph.

$$\Delta A_{650\ nm\ Sample} = A_{650\ nm\ Sample} - A_{650\ nm\ Blank}$$

Determined the mg protein from the standard curve.

3.5.2 Determination of Malondialdehyde (MDA) Level

The methods for determination of malondialdehyde (MDA) was adapted from the method of Buege and Aust (1978). Briefly, 100 µl of the sample and standard enzymes and 200 µl of TCA-TBA-HCl solution were pipetted into each tube, and mixed well. DDD water mixed with solution A was used as the control and solution A was used as blank. All determinations were performed in triplicate. All mixtures were heated on water bath 100 ºC for 15 minutes. After cooling, the mixture was centrifuged at 1,000 g for 10 minutes. The absorbance of the mixture (supernatant) was measured at 535 nm using microplate reader.

The TMP concentrations were calculated using the following equation:

$$\Delta A_{535\ nm}\ \text{Standard} = A_{535\ nm}\ \text{Standard} - A_{535\ nm}\ \text{Blank}$$

Plotted the $\Delta A_{535\ nm}$ Standard againt TMP concentration on the standard graph.

$$\Delta A_{535\ nm}\ \text{Sample} = A_{535\ nm}\ \text{Sample} - A_{535\ nm}\ \text{Blank}$$

The TMP (equivalence with MDA) level was determined as nmol/mg tissue weight from the standard curve.

3.5.3 Determination of Hydrogen Peroxide (H_2O_2) Level

The method for determination of hydrogen peroxide (H_2O_2) level was adapted from the method of Muller (1985). Briefly, 100 µl of 0.1 M phosphate buffer (pH 5.0) was pipetted into 96 well-plates and 100 µl of sample, standard H_2O_2, or 50 mM phosphate buffer (pH 7.4) containing 0.1 mM EDTA was added. Hydrogen peroxide (20 µl) was added, mixed well and then incubated at 37 ºC for 5 min. After incubation, 30 µl of 1.25 mM ABTS and 30 µl of 1 unit.ml^{-1} horseradish peroxidase were added, mixed well and incubated at 37 ºC for 10 min. The absorbance of mixture were measured at 405 nm by Benchmark Plus Microplate Spectrophotometer.

The hydrogen peroxide concentration was calculated using the following equation:

$$\Delta A_{405\ nm}\ \text{Standard} = A_{405\ nm}\ \text{Standard} - A_{405\ nm}\ \text{Blank}$$

Plotted the $\Delta A_{405\ nm}$ Standard against H_2O_2 concentration on the standard graph.

$$\Delta A_{405\ nm}\ \text{Sample} = A_{405\ nm}\ \text{Sample} - A_{405\ nm}\ \text{Blank}$$

The H_2O_2 level was determined as µM/mg protein.

3.5.4 Determination of Total Superoxide Dismutase (SOD) Level

The method for determination of superoxide dismutase level was adapted from SOD determination kit (Sigma-Aldrich). Briefly, 20 µl of sample was added into each triplicate well of 96-well plates and 200 µl of water-soluble tetrazolium (WST) working solution was added and mixed well. Enzyme working solution (20 µl) was added, mixed and incubated the mixture at 37 ºC for 20 min. After incubation, the absorbance of the mixture was measured at 450 nm using a microplate reader.

The SOD activity (inhibition rate %) was calculated using the following equation:

$$\text{SOD activity (inhibition rate \%)} = \frac{(\text{Abs control} - \text{Abs sample})}{\text{Abs control}} \text{ x } 100.$$

3.5.5 Determination of Catalase (CAT) Level

The measurement of catalase (CAT) level was adapted from the method of Goldblith and(Samuel A. Goldblith and Proctor, 1950). Briefly, 10 µl of the sample and standard enzyme were pipetted into triplicate wells of 96-well plates. Fifty microliters of 0.01 N of Hydrogen Peroxide (H_2O_2) solution was pipetted into bottom of 96-well plates. Twenty five microliters of 5 N of sulfulic acid (H_2SO_4) solution and 150 µl of Potassium permanganate ($KMnO_4$) solution were added and mixed well. The absorbance of the mixture was then measured at 490 nm using a microplate reader. The DDD water mixed with hydrogen peroxide solution, sulfulic acid solution and potassium permanganate solution was used as the control. Hydrogen peroxide solution, sulfulic acid solution and potassium permanganate solution was used as blank.

Calculation:

$\Delta A_{490\ nm}$ Standard = $A_{490\ nm}$ Standard - $A_{490\ nm}$ Blank

Plotted the $\Delta A_{490\ nm}$ Standard against catalase enzyme concentration on the standard graph.

$\Delta A_{490\ nm}$ Sample = $A_{490\ nm}$ Sample - $A_{490\ nm}$ Blank

Determined the catalase enzyme level from the standard curve.

Catalase enzyme level was expressed as unit/mg protein.

3.5.6 Determination of Glutathione Peroxidase (GPx) Activity

The method for determination of glutathione peroxidase (GPx) activity was adapted from the method of glutathione peroxidase cellular activity assay kit (Sigma-Aldrich). Briefly, glutathione peroxidase assay buffer was pipetted into each wells. The temperature of the assay buffer was kept at 25 ºC. NADPH Assay Reagent (10 µl) and 10 µl of sample and enzyme were added into 96-well plate and mixed well. The reaction was started by addition of 2 µl of the 30 mM tert-butyl hydroperoxide solution into the mixture and mixed well. The decrease in absorbance of the mixture was measured at 340 nm using a kinetic program of Benchmark Plus Microplate Spectrophotometer.

The following program was read:

Wavelength: 340 nm

Initial delay: 15 seconds

Interval: 10 seconds

Number of reading: 6

The amount of enzyme in the sample was calculated as follows:

The activity of glutathione peroxidase in the sample was calculated using the formula:

Activity per extract (mmol/min/ml = Units/ml)

$$(\Delta A_{340} \text{ x DF}) \div 6.22 \text{ x V}$$

$$\Delta A_{340} = A_{340}/\text{min}_{\text{(blank)}} - A_{340}/\text{min}_{\text{(sample)}}$$

$$6.22 = \varepsilon^{mM} \text{ for NADPH}$$

DF = dilution factor of sample before adding to reaction

V = sample volume in ml

Unit definition: 1 unit of glutathione peroxidase caused the formation of 1.0 μmol of $NADP^+$ from NADPH per minute at pH 8.0 at 25 ºC in a coupled reaction in the presence of reduced glutathione, glutathione reductase, and tret-butyl hydroperoxide.

3.6 Statistical Analysis

Data were expressed as mean ± SEM. Statistical analysis was assessed by one-way analysis of variance (ANOVA) using the SPSS software version 24, IBM®. Post hoc testing was performed for intergroup comparisons. *P-Value* less than 0.05 (p<0.05) was considered to be statistically significant.

CHAPTER IV

RESULTS

4.1 Effects of Taurine Supplement in Conjunction with Exercise on Antioxidant Enzymes Activities, Malondialdehyde Levels and Hydrogen Peroxide Levels in Adult and Middle-aged Rat Brains

4.1.1 Relative Organ Weight (ROW)

- Gastrocnemius muscle of Adult Rats

The ROWs of gastrocnemius muscle in the sedentary group of adult rats were as follows, DDDW+Se group, 1.20±0.03 g/100 g body weight; 1%Tween80+Se group, 1.15±0.02 g/100 g body weight; Vit E+Se group, 1.27±0.02 g/100 g body weight; and Tau+Se group, 1.17±0.05 g/100 g body weight. The ROWs of gastrocnemius muscle in the exercise group of adult rats were as follows: DDDW+Ex group, 1.17±0.05 g/100 g body weight; 1%Tween80+Ex group, 1.31±0.06 g/100 g body weight; Vit E+Ex group, 1.21±0.04 g/100 g body weight; and Tau+Ex group, 1.27±0.03 g/100 g body weight (Figure 11). The results showed that ROW of gastrocnemius muscle following 1% Tween 80 treatment in the exercise group was significantly higher than vitamin E supplement and 1% Tween 80 treated group in the sedentary group and DDD water treatment in the exercise group (P<0.05). In comparison between the sedentary group and the exercise group, exercise significantly increased ROW of gastrocnemius in 1% Tween 80 treatment (P<0.05).

- Gastrocnemius muscle of Middle-aged Rats

The ROWs of gastrocnemius muscle in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 1.04±0.06 g/100 g body weight; 1%Tween80+Se group, 1.06±0.03 g/100 g body weight; Vit E+Se group, 1.08±0.04 g/100 g body weight; and Tau+Se group, 1.03±0.04 g/100 g body weight. The ROWs of gastrocnemius muscle in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 1.15±0.05 g/100 g body weight; 1%Tween80+Ex group, 1.11±0.07 g/100 g body weight; Vit E+Ex group, 1.18±0.06 g/100 g body weight; and Tau+Ex group, 1.11±0.04 g/100 g body weight (Figure 11). The results showed that ROW of gastrocnemius muscle of middle-aged rats following taurine treatment in the exercise group was significantly higher than DDD water treatment in sedentary group. In comparison between the sedentary group and the exercise group, no significant difference in ROW of gastrocnemius muscle was shown in all treatment.

- The Comparison between Adult and Middle-aged Rats of ROW of Gastrocnemius Muscle

The result of the ROWs of gastrocnemius muscle when comparison between adult rats and middle-aged rats (Figure 11) found that all treatment groups in the adult rats in the sedentary group and the exercise in adult rats following 1% Tween 80 treatment and taurine treatment were significantly higher than same treatment in middle-aged rats (P<0.05).

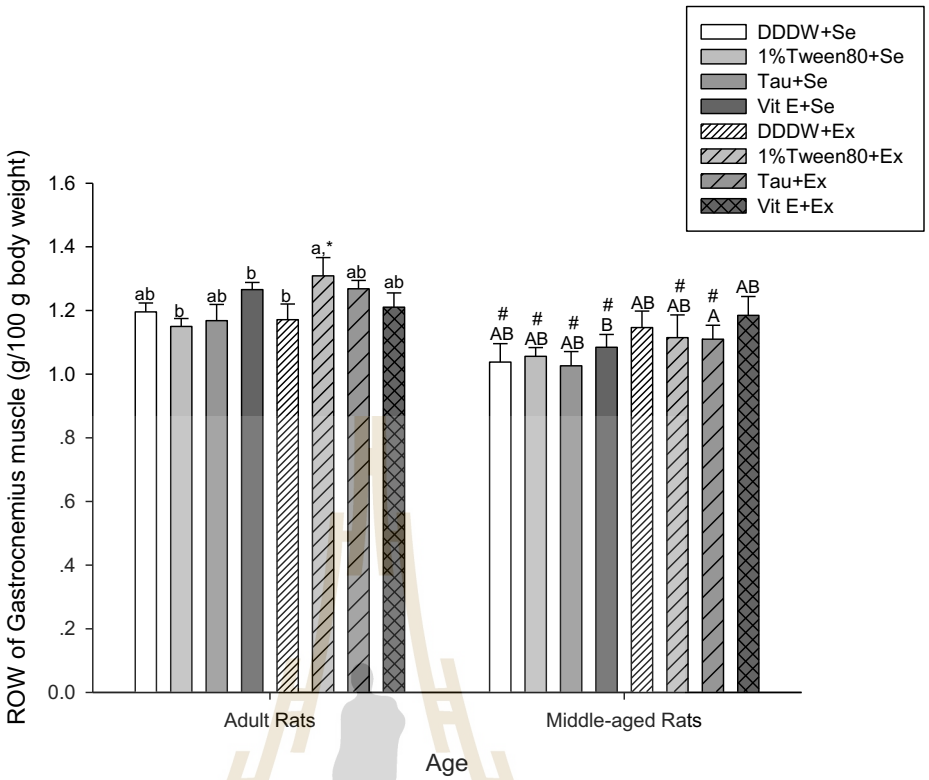

Figure 11 Effects of taurine and vitamin E supplement in conjunction with exercise on ROW of gastrocnemius muscle of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Soleus Muscle of Adult Rats

The ROWs of soleus muscle in the sedentary group of adult rats were as follows: DDDW+Se group, 0.086±0.004 g/100 g body weight; 1%Tween80+Se group, 0.085±0.003 g/100 g body weight; Vit E+Se group, 0.088±0.005 g/100 g body weight;

and Tau+Se group, 0.086±0.003 g/100 g body weight. The ROWs of soleus muscle in the exercise group of adult rats were as follows: DDDW+Ex group, 0.077±0.004 g/100 g body weight; 1%Tween80+Ex group, 0.092±0.004 g/100 g body weight; Vit E+Ex group, 0.090±0.004 g/100 g body weight; and Tau+Ex group, 0.091±0.005 g/100 g body weight (Figure 12). The results showed that there was no significant difference in ROW of soleus muscle between all groups in the sedentary group. In the exercise group, ROWs of soleus muscle following vitamin E supplement and 1% Tween 80 treatment were significantly higher than exercise alone (P<0.05). In comparison between the sedentary group and the exercise group, exercise group did not cause changes in ROW of soleus muscle in all treatments.

- Soleus Muscle of Middle-aged Rats

The ROWs of soleus muscle in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 0.088±0.010 g/100 g body weight; 1%Tween80+Se group, 0.086±0.006 g/100 g body weight; Vit E+Se group, 0.094±0.006 g/100 g body weight; and Tau+Se group, 0.074±0.005 g/100 g body weight. The ROWs of soleus muscle in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 0.093±0.005 g/100 g body weight; 1%Tween80+Ex group, 0.087±0.005 g/100 g body weight; Vit E+Ex group, 0.089±0.005 g/100 g body weight; and Tau+Ex group, 0.081±0.002 g/100 g body weight (Figure 12). The result showed that taurine supplement was significantly higher than vitamin E treatment in the sedentary group (P<0.05). In comparison between the sedentary group and the exercise group, no significant difference in ROW of soleus muscle was shown in all treatment.

- The Comparison between Adult and Middle-aged Rats of ROW of Soleus Muscle

The result of the ROWs of soleus muscle when comparison between adult rats and middle-aged rats (Figure 12) showed that there was no significant difference between all treatment groups when comparison between adult rats and middle-aged rats.

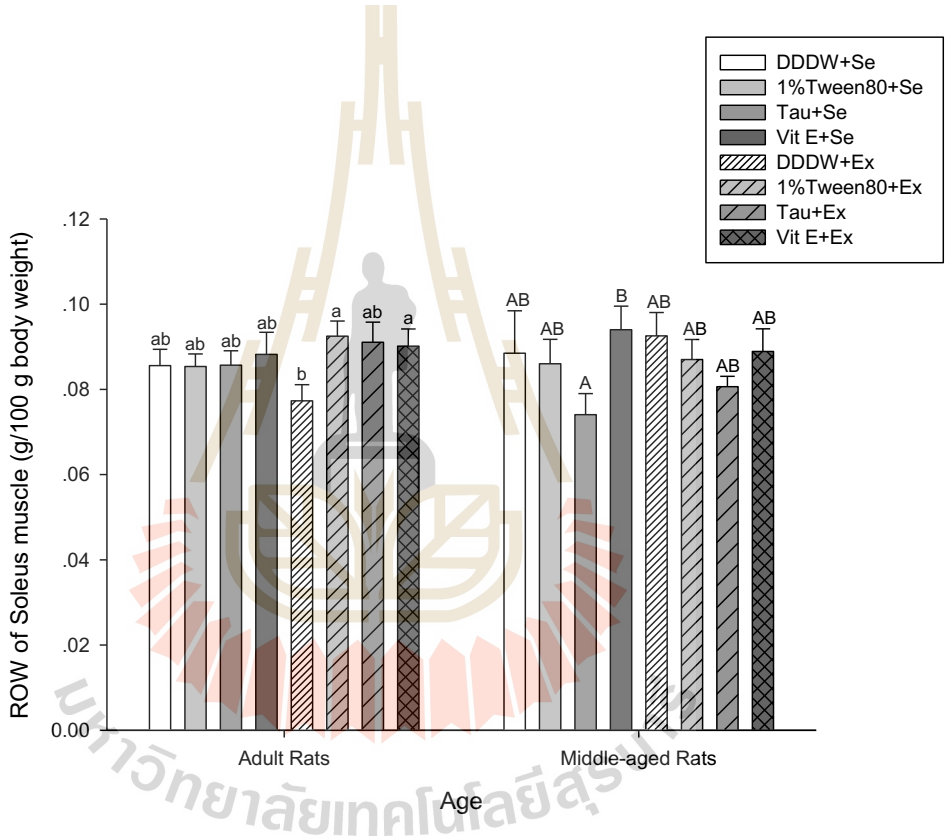

Figure 12 Effects of taurine and vitamin E supplement in conjunction with exercise on ROW of soleus muscle of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case).

- Extensor Digitorum Longus (EDL) Muscle of Adult Rats

The ROWs of EDL muscle in the sedentary group adult rats were as follows: DDDW+Se group, 0.094±0.004 g/100 g body weight; 1%Tween80+Se group, 0.090±0.003 g/100 g body weight; Vit E+Se group, 0.101±0.004 g/100 g body weight; and Tau+Se group, 0.100±0.004 g/100 g body weight. The ROW of EDL muscle in the exercise group of adult rats were as follows: DDDW+Ex group, 0.096±0.004 g/100 g body weight; 1%Tween80+Ex group, 0.113±0.004 g/100 g body weight; Vit E+Ex group, 0.104±0.005 g/100 g body weight; and Tau+Ex group, 0.102±0.008 g/100 g body weight (Figure 13). The results showed that ROWs of EDL muscle following 1% Tween 80 treatment in the exercise group was significantly higher than 1%Tween80 treatment and DDD water treatment in the sedentary group (P<0.05). In comparison between the sedentary group and the exercise group, ROW of EDL muscle in 1% Tween 80 treatment with exercise was significantly higher than its respective control (P<0.05).

- Extensor Digitorum Longus (EDL) muscle of middle-Aged Rats

The ROWs of EDL in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 0.078±0.006 g/100 g body weight; 1%Tween80+Se group, 0.095±0.007 g/100 g body weight; Vit E+Se group, 0.110±0.006 g/100 g body weight; and Tau+Se group, 0.101±0.008 g/100 g body weight. The ROWs of EDL muscle in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 0.106±0.008 g/100 g body weight; 1%Tween80+Ex group, 0.091±0.009 g/100 g body weight; Vit E+Ex group, 0.106±0.006 g/100 g body weight; and Tau+Ex group, 0.104±0.006 g/100 g body weight (Figure 13). The results showed that ROWs of EDL

muscle of the middle-aged rats following taurine supplement and vitamin E supplement were significantly higher than the DDD water treatment in the sedentary group and in the exercise group following taurine supplement, vitamin E supplement, DDD water treatment, and 1% Tween 80 treatment were significantly higher than DDD water treatment in the sedentary group ($P<0.05$). In comparison between the sedentary group and the exercise group, ROW of EDL muscle in the exercise group was significantly higher than respective control (DDD water treatment) ($P<0.05$).

- The Comparison between Adult and Middle-aged Rats of ROW of EDL Muscle

The result of the ROWs of EDL muscle when comparison between adult rats and middle-aged rats (Figure 13) showed that there was no significant difference between all treatment groups when comparison between adult rats and middle-aged rats.

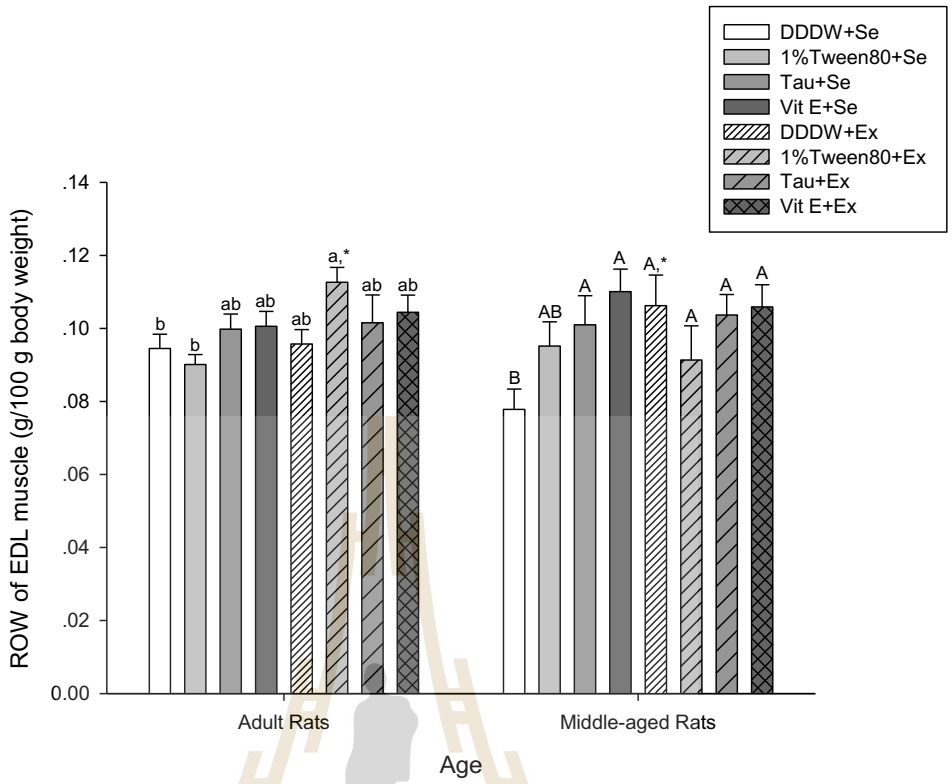

Figure 13 Effects of taurine and vitamin E supplement in conjunction with exercise on ROW of EDL muscle of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age.

- Liver of Adult Rats

The ROWs of the liver in the sedentary group of adult rats were as follows: DDDW+Se group, 2.60±0.06 g/100 g body weight; 1%Tween80+Se group, 2.99±0.16 g/100 g body weight; Vit E+Se group, 2.59±0.05 g/100 g body weight; and Tau+Se group, 2.74±0.11 g/100 g body weight. The ROWs of the liver in exercise group of adult rats were as follows: DDDW+Ex group, 2.66±0.13 g/100 g body weight;

1%Tween80+Ex group, 2.75±0.08 g/100 g body weight; Vit E+Ex group, 2.57±0.06 g/100 g body weight; and Tau+Ex group, 2.62±0.09 g/100 g body weight (Figure 14). The results showed that ROW of liver following 1% Tween 80 treatment in the sedentary group was significantly higher than taurine supplement and DDD water treatment in the sedentary groups and DDD water treatment, vitamin E treatment, and taurine supplement in the exercise groups (P<0.05). There was no significant difference in ROW of liver between all groups in the exercise group. In comparison between the sedentary group and the exercise group, exercise did not cause changes in ROW of liver in all treatments.

- Liver of Middle-aged Rats

The ROWs of liver in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 2.30±0.08 g/100 g body weight; 1%Tween80+Se group, 2.20±0.06 g/100 g body weight; Vit E+Se group, 2.28±0.09 g/100 g body weight; and Tau+Se group, 2.29±0.08 g/100 g body weight. The ROWs of liver in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 2.57±0.11 g/100 g body weight; 1%Tween80+Ex group, 2.31±0.11 g/100 g body weight; Vit E+Ex group, 2.33±0.08 g/100 g body weight; and Tau+Ex group, 2.43±0.08 g/100 g body weight (Figure 14). The results showed that ROWs of liver following DDD water treatment in the exercise group was significantly higher than taurine treatment and 1% Tween 80 treatment in the sedentary groups and 1% Tween 80 treatment in the exercise group. When compared between the sedentary group and the exercise group, in the exercise group was significantly higher ROW of liver than DDD water treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats of ROW of Liver

The result of the ROWs of liver when comparison between adult rats and middle-aged rats (Figure 14) found that all treatment groups in the adult rats in the sedentary group and the exercise in adult rats following 1% Tween 80 treatment and vitamin E treatment were significantly higher than same treatment in middle-aged rats (P<0.05).

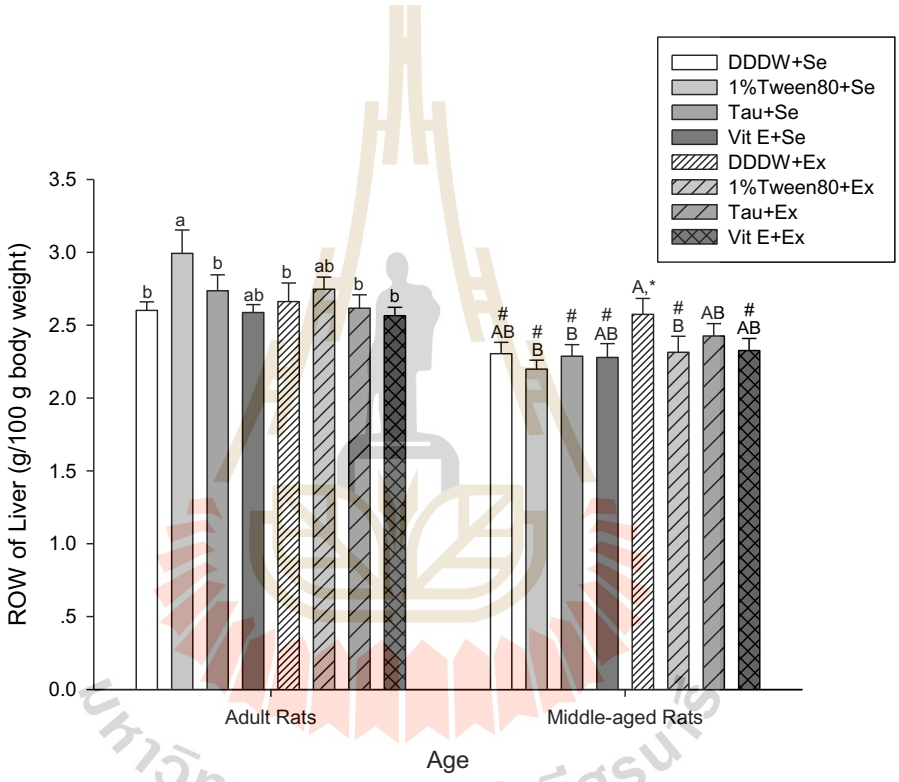

Figure 14 Effects of taurine and vitamin E supplement in conjunction with exercise on ROW of liver of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Kidney of Adult Rats

The ROWs of the kidney in the sedentary group of adult rats were as follows: DDDW+Se group, 0.50±0.03 g/100 g body weight; 1%Tween80+Se group, 0.51±0.03 g/100 g body weight; Vit E+Se group, 0.52±0.03 g/100 g body weight; and Tau+Se group, 0.51±0.03 g/100 g body weight (Figure 15). The results showed that there was no significant difference in ROW of kidney between all groups in both the sedentary group and the exercise group. In comparison between the sedentary group and the exercise group, exercise did not cause changes in ROW of kidney in all treatments.

- Kidney of middle-aged Rats

The ROWs of kidney in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 0.49±0.03 g/100 g body weight; 1%Tween80+Se group, 0.49±0.02 g/100 g body weight; Vit E+Se group, 0.52±0.04 g/100 g body weight; and Tau+Se group, 0.51±0.03 g/100 g body weight. The ROWs of kidney in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 0.51±0.02 g/100 g body weight; 1%Tween80+Ex group, 0.47±0.03 g/100 g body weight; Vit E+Ex group, 0.55±0.03 g/100 g body weight; and Tau+Ex group, 0.55±0.03 g/100 g body weight (Figure 15). The results showed that there was no significant different in ROWs of liver between in all treatment of the sedentary group and the exercise group. When compared between the sedentary group and the exercise group, there was no significant difference in all treatment groups.

- The Comparison between Adult and Middle-aged Rats of ROW of Kidney

The result of the ROWs of kidney when comparison between adult rats and middle-aged rats (Figure 15) showed that there was no significant difference between all treatment groups when comparison between adult rats and middle-aged rats.

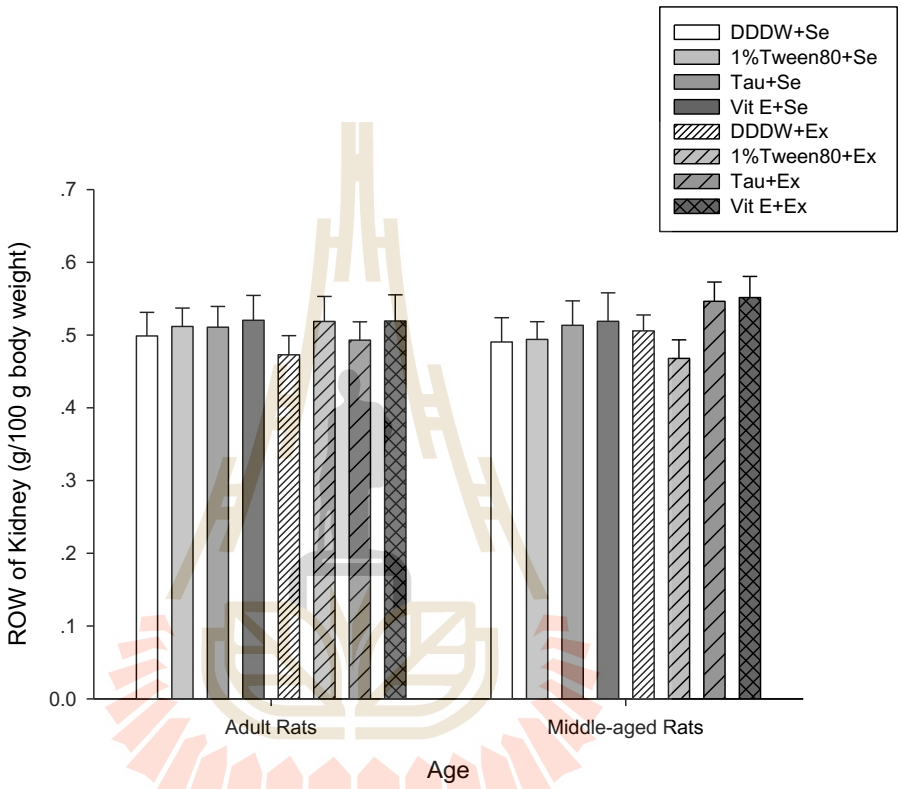

Figure 15 Effects of taurine and vitamin E supplement in conjunction with exercise on ROW of kidney of adult and middle-aged rats. Values are expressed as mean ± SEM.

- Lung of Adult Rats

The ROWs of the lung in the sedentary group of adult rats were as follows: DDDW+Se group, 0.56±0.06 g/100 g body weight; 1%Tween80+Se group, 0.50±0.05 g/100 g body weight; Vit E+Se group, 0.49±0.05 g/100 g body weight; and Tau+Se group, 0.48±0.06 g/100 g body weight. The ROWs of the lung in exercise group of adult rats were as follows: DDDW+Ex group, 0.44±0.02 g/100 g body weight;

1%Tween80+Ex group, 0.51±0.05 g/100 g body weight; Vit E+Ex group, 0.49±0.05 g/100 g body weight; and Tau+Ex group, 0.44±0.04 g/100 g body weight (Figure 16). There was no significant difference in ROW of lung between all groups in both the sedentary group and the exercise group. In comparison between the sedentary group and the exercise group, exercise did not cause changes in ROW of lung in all treatments.

- Lung of Middle-aged Rats

The ROWs of lung in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 0.48±0.04 g/100 g body weight; 1%Tween80+Se group, 0.47±0.05 g/100 g body weight; Vit E+Se group, 0.43±0.05 g/100 g body weight; and Tau+Se group, 0.47±0.04 g/100 g body weight. The ROWs of lung in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 0.48±0.08 g/100 g body weight; 1%Tween80+Ex group, 0.43±0.05 g/100 g body weight; Vit E+Ex group, 0.48±0.04 g/100 g body weight; and Tau+Ex group, 0.41±0.04 g/100 g body weight (Figure 16). The results showed that there was no significant difference in ROW of lung between in all treatment groups of both the sedentary group and the exercise group. When compared between the sedentary group and the exercise group, there was no significant difference in all treatment groups.

- The Comparison between Adult and Middle-aged Rats of ROW of Lung

The result of the ROWs of lung when comparison between adult rats and middle-aged rats (Figure 16) showed that there was no significant difference between all treatment groups when comparison between adult rats and middle-aged rats.

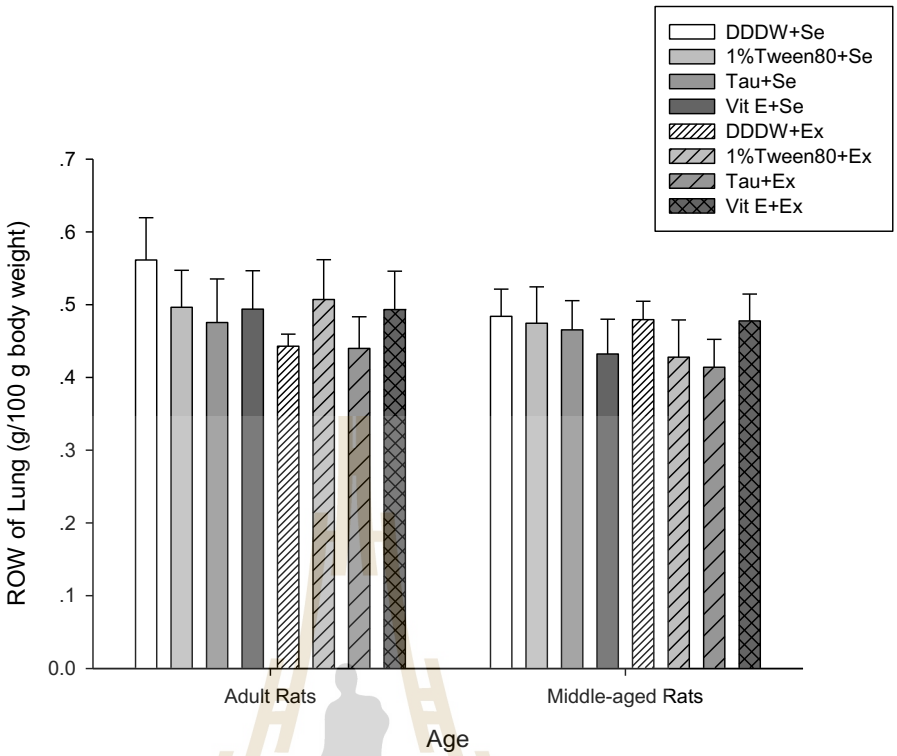

Figure 16 Effects of taurine and vitamin E supplement in conjunction with exercise on ROW of lung of adult and middle-aged rats. Values are expressed as mean ± SEM.

- Spleen of Adult Rats

The ROWs of the spleen in the sedentary group of adult rats were as follows: DDDW+Se group, 0.198±0.007 g/100 g body weight; 1%Tween80+Se group, 0.204±0.007 g/100 g body weight; Vit E+Se group, 0.202±0.009 g/100 g body weight; and Tau+Se group, 0.211±0.008 g/100 g body weight. The ROWs of the spleen in the exercise group of adult rats were as follows: DDDW+Ex group, 0.183±0.010 g/100 g body weight; 1%Tween80+Ex group, 0.205±0.007 g/100 g body weight; Vit E+Ex group, 0.200±0.009 g/100 g body weight; and Tau+Ex group, 0.187±0.009 g/100 g body weight (Figure 17). The results showed that ROWs of spleen following vitamin E treatment in the sedentary group was significantly higher than vitamin E treatment and DDD water treatment in the exercise group (P<0.05). In comparison between the

sedentary group and the exercise group, exercise significantly decreased ROW of spleen in taurine treatment (P<0.05).

- Spleen of Middle-aged Rats

The ROWs of spleen in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 0.179±0.009 g/100 g body weight; 1%Tween80+Se group, 0.206±0.011 g/100 g body weight; Vit E+Se group, 0.196±0.007 g/100 g body weight; and Tau+Se group, 0.204±0.006 g/100 g body weight. The ROWs of spleen in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 0.195±0.015 g/100 g body weight; 1%Tween80+Ex group, 0.178±0.012 g/100 g body weight; Vit E+Ex group, 0.196±0.012 g/100 g body weight; and Tau+Ex group, 0.184±0.011 g/100 g body weight (Figure 17). The results showed that ROWs of spleen following taurine supplement, vitamin E treatment and 1% Tween 80 treatment in the sedentary groups and DDD water treatment in the exercise group were significantly higher than 1% Tween 80 treatment in the exercise group (P<0.05). When compared between the sedentary group and the exercise group, there was no significant difference in all treatment groups.

- The Comparison between Adult and Middle-aged Rats of ROW of Spleen

The result of the ROWs of spleen when comparison between adult rats and middle-aged rats (Figure 17) found that there was no significant difference in all treatment groups in the sedentary group and in the exercise in adult rats following 1% Tween 80 treatment was significantly higher than same treatment in middle-aged rats (P<0.05).

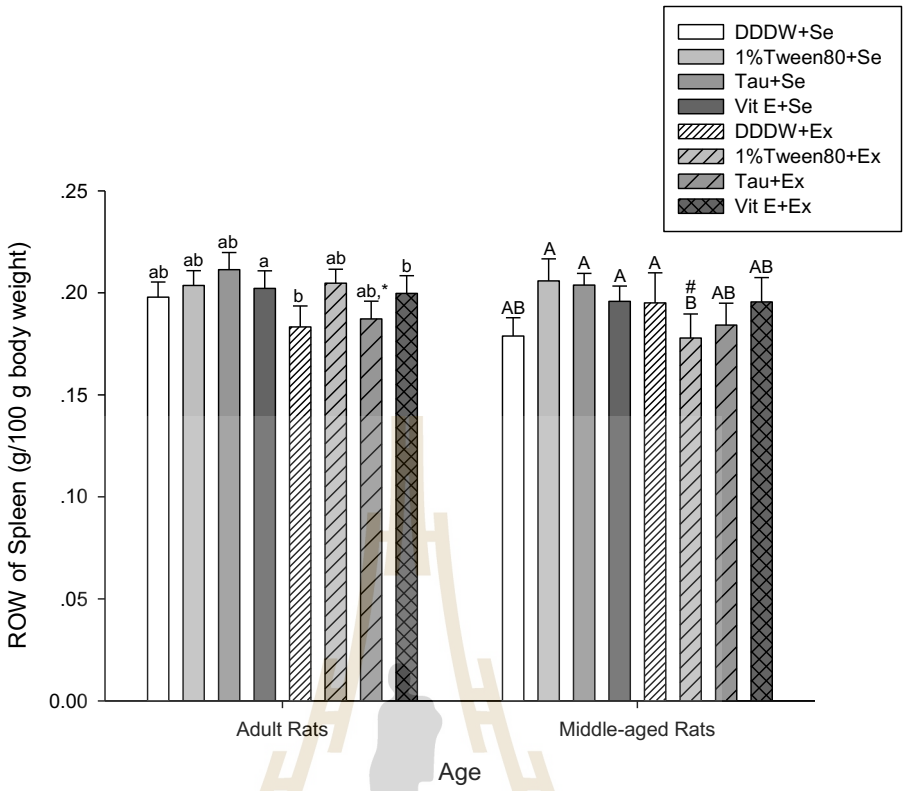

Figure 17 Effects of taurine and vitamin E supplement in conjunction with exercise on ROW of spleen of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Heart of Adult Rats

The ROWs of the heart in the sedentary group of adult rats were as follows: DDDW+Se group, 0.32±0.02 g/100 g body weight; 1%Tween80+Se group, 0.30±0.01 g/100 g body weight; Vit E+Se group, 0.32±0.01 g/100 g body weight; and Tau+Se group, 0.32±0.02 g/100 g body weight. The ROWs of the heart in exercise group of

adult rats were as follows: DDDW+Ex group, 0.31±0.01 g/100 g body weight; 1%Tween80+Ex group, 0.34±0.01 g/100 g body weight; Vit E+Ex group, 0.33±0.01 g/100 g body weight; and Tau+Ex group, 0.32±0.01 g/100 g body weight (Figure 18). The result of ROW of heart in the adult rats following 1% Tween 80 treatment and taurine supplement in the exercise groups were significant higher than 1% Tween 80 treatment in the sedentary group (P<0.05). In comparison between the sedentary group and exercise group, exercise significantly increased ROW of heart in 1% Tween 80 treatment (P<0.05).

- Heart of Middle-aged Rats

The ROWs of heart in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 0.25±0.01 g/100 g body weight; 1%Tween80+Se group, 0.27±0.02 g/100 g body weight; Vit E+Se group, 0.26±0.01 g/100 g body weight; and Tau+Se group, 0.27±0.01 g/100 g body weight. The ROWs of heart in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 0.32±0.01 g/100 g body weight; 1%Tween80+Ex group, 0.33±0.02 g/100 g body weight; Vit E+Ex group, 0.29±0.02 g/100 g body weight; and Tau+Ex group, 0.31±0.01 g/100 g body weight (Figure 18). The results showed that ROW of heart following DDD water treatment and 1% Tween 80 treatment in the exercise group were significantly higher than all treatment group of the sedentary groups (P<0.05). Vitamin E treatment in the exercise group was significantly higher than taurine treatment and DDD water treatment in the sedentary group (P<0.05). In comparison between the sedentary group and exercise group, ROWs of heart following vitamin E supplement and DDD water treatment in the exercise groups were significantly higher than their respective control (P<0.05).

- The Comparison between Adult and Middle-aged Rats of ROW of Spleen

The result of the ROWs of gastrocnemius muscle when comparison between adult rats and middle-aged rats (Figure 18) found that the adult rats in the sedentary group following DDD water treatment and taurine treatment and the exercise in adult rats following vitamin E treatment were significantly higher than same treatment in middle-aged rat (P<0.05).

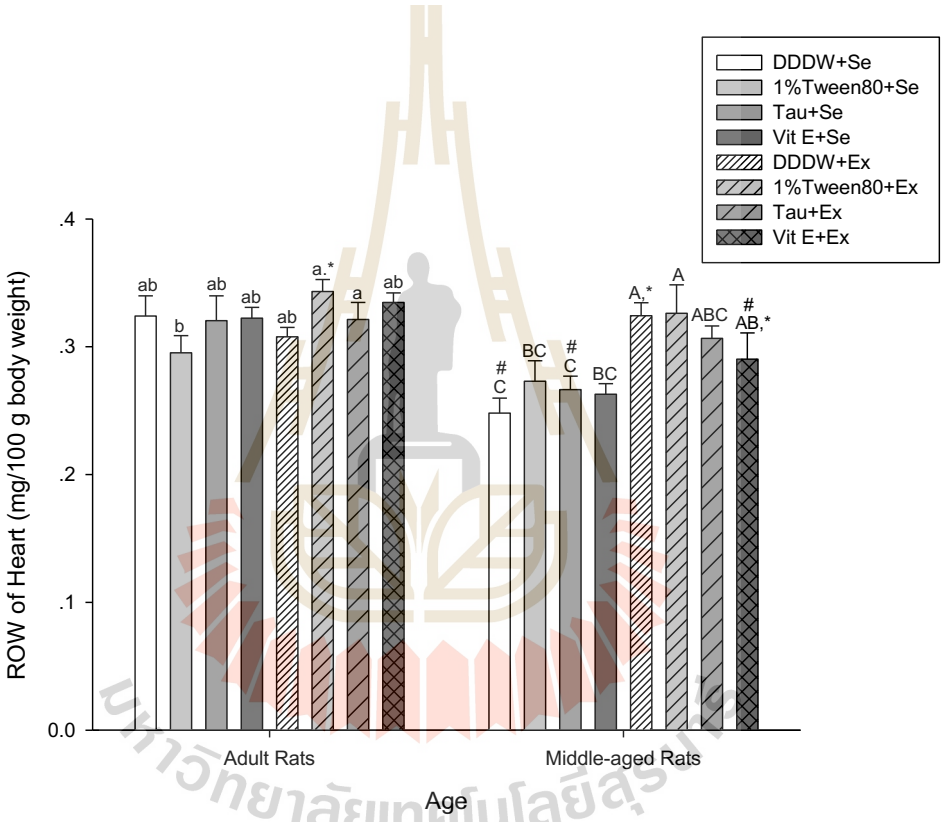

Figure 18 Effects of taurine and vitamin E supplement in conjunction with exercise on ROW of heart of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

4.1.2 Blood Biochemical Analysis

- Blood Sugar Levels of Adult Rats

The blood sugar levels in the sedentary group of adult rats were as follows: DDDW+Se group, 124.67±5.99 mg/dL; 1%Tween80+Se group, 132.00±1.59 mg/dL; Vit E+Se group, 129.00±4.15 mg/dL; and Tau+Se group, 126.33±3.72 mg/dL. The blood sugar levels in exercise group of adult rats were as follows: DDDW+Ex group, 132.00±7.86 mg/dL; 1%Tween80+Ex group, 110.25±2.40 mg/dL; Vit E+Ex group, 133.33±4.01 mg/dL; and Tau+Ex group, 116.20±2.16 mg/dL (Figure 19). The results showed that in the adult rats following vitamin E treatment and DDD water treatment in the exercise groups and 1% Tween 80 treatment in the sedentary group were significantly higher than taurine treatment and 1% Tween 80 treatment in the exercise groups, and vitamin E treatment, taurine treatment, and DDD water treatment in the sedentary groups were significantly higher than 1% Tween 80 treatment in the exercise group (P<0.05). In comparison between the sedentary group and the exercise group, exercise significantly decreased blood sugar level in 1% Tween 80 treatment and taurine supplement (P<0.05).

- Blood sugar levels of Middle-aged Rats

The blood sugar levels in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 159.33±9.47 mg/dL; 1%Tween80+Se group, 155.50±2.24 mg/dL; Vit E+Se group, 142.29±5.93 mg/dL; and Tau+Se group, 145.50±8.68 mg/dL. The blood sugar level in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 137.60±11.49 mg/dL; 1%Tween80+Ex group, 143.60±14.42

mg/dL; Vit E+Ex group, 132.40±7.89 mg/dL; and Tau+Ex group, 119.57±4.59 mg/dL (Figure 19). The results showed that in the sedentary group following DDD water treatment was significantly higher than vitamin E treatment and taurine treatment in the exercise groups, and 1% Tween 80 treatment and taurine supplement in the sedentary groups and 1% Tween 80 treatment in the exercise group were significantly higher than taurine supplement in the exercise group (P<0.05). In comparison between sedentary group and exercise group, exercise significantly lower blood sugar level in taurine supplement (P<0.05).

- The Comparison between Adult and Middle-aged Rats of blood sugar levels

The result of the blood parameters of blood sugar levels when comparison between adult rats and middle-aged rats (Figure 19) found that the middle-aged rats in the sedentary group following DDD water treatment, 1% Tween 80 treatment and taurine treatment were significantly higher than same treatment in adult rats (P<0.05).

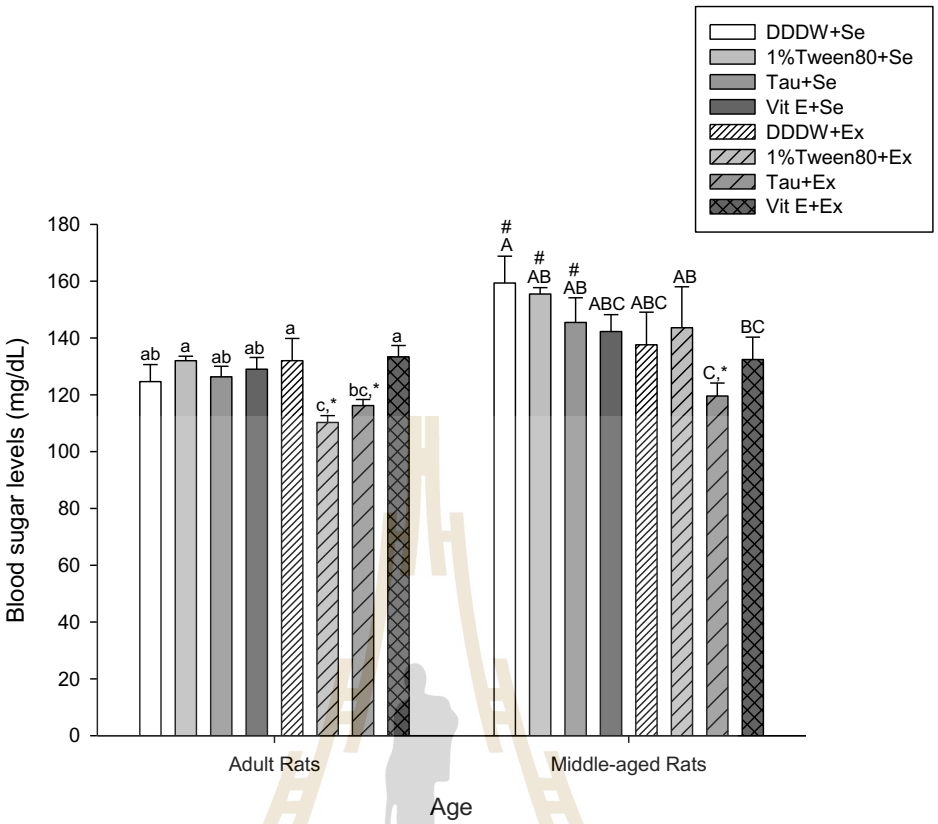

Figure 19 Effects of taurine and vitamin E supplement in conjunction with exercise on blood sugar levels of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Lipid Profile Levels

- Total Cholesterol Levels of Adult Rats

The plasma total cholesterol levels in the sedentary group of adult rats were as follows: DDDW+Se group, 57.57±2.47 mg/dL; 1%Tween80+Se group, 60.89±2.92 mg/dL; Vit E+Se group, 64.20±4.56 mg/dL; and Tau+Se group, 63.75±2.07 mg/dL.

The plasma total cholesterol levels in the exercise group of adult rats were as follows: DDDW+Ex group, 59.88±0.98 mg/dL; 1%Tween80+Ex group, 56.63±2.20 mg/dL; Vit E+Ex group, 60.88±2.41 mg/dL; and Tau+Ex group, 63.22±3.18 mg/dL (Figure 20). The results showed that there was no significant difference in plasma total cholesterol levels between all treatments of adult rats in both the sedentary group and the exercise group. The results showed that there was no significant difference in plasma total cholesterol levels all treatments of adult rats in both the sedentary group and the exercise group. In comparison between the sedentary group and the exercise group, no significant difference in plasma total cholesterol levels were found in all treatments.

- Total cholesterol levels of Middle-aged Rats

The plasma total cholesterol levels in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 103.75±5.99 mg/dL; 1%Tween80+Se group, 93.44±3.30 mg/dL; Vit E+Se group, 88.56±4.42 mg/dL; and Tau+Se group, 119.14±3.77 mg/dL. The plasma total cholesterol levels in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 99.63±7.80 mg/dL; 1%Tween80+Ex group, 98.14±7.29 mg/dL; Vit E+Ex group, 84.50±6.29 mg/dL; and Tau+Ex group, 103.88±7.38 mg/dL (Figure 20). The results showed that cholesterol levels of middle-aged rats following taurine supplement in the sedentary group was significantly higher than vitamin E supplement, 1% Tween 80 treatment and DDD water treatment in the exercise groups and 1% Tween 80 treatment and vitamin E treatment in the sedentary groups (P<0.05), and DDD water treatment in the sedentary group and taurine treatment in the exercise group were significantly higher than vitamin E supplement in the exercise group (P<0.05). There was no significant difference

between all treatment groups of exercise group. When compared between the sedentary group and the exercise group, there was no significant difference in all treatment groups.

- The Comparison between Adult and Middle-aged Rats of Total Cholesterol Levels

The result of the blood parameters of total cholesterol levels when comparison between adult rats and middle-aged rats (Figure 20) found that the middle-aged rats all treatment group of both the sedentary groups and the exercise groups were significantly higher than same treatment in adult rats ($P<0.05$).

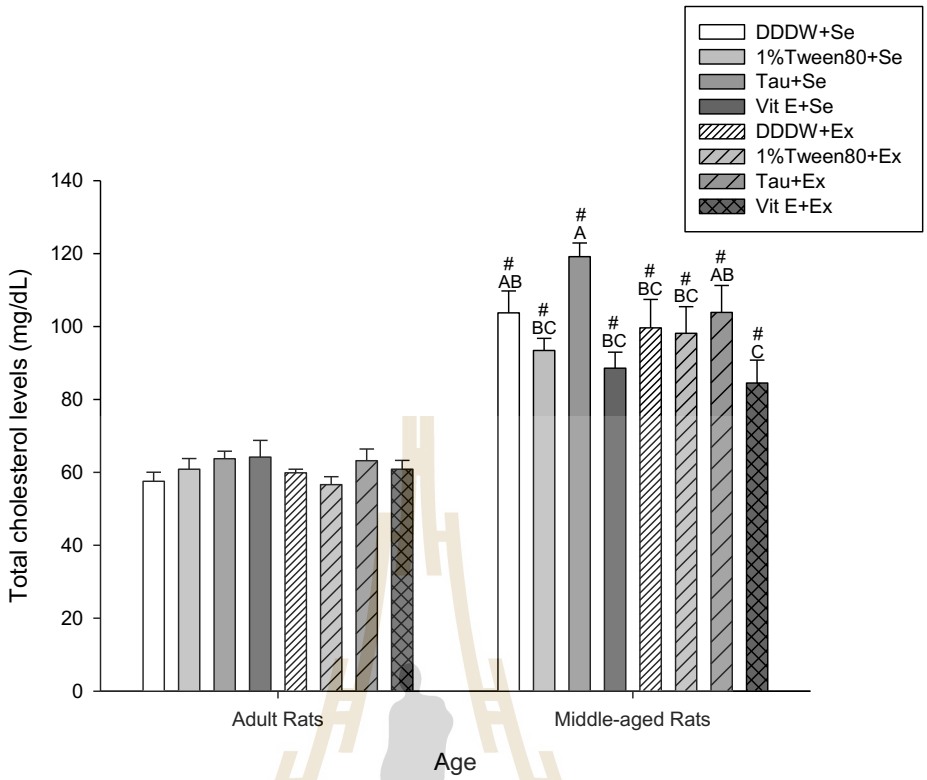

Figure 20 Effects of taurine and vitamin E supplement in conjunction with exercise on total cholesterol levels of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in middle-aged group (upper case). Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity. In not have symbol showed no significant difference between all comparison.

- Triglyceride Levels of Adult Rats

The plasma triglyceride levels in the sedentary group of adult rats were as follows: DDDW+Se group, 83.67±7.39 mg/dL; 1%Tween80+Se group, 94.60±10.06 mg/dL; Vit E+Se group, 74.60±6.66 mg/dL; and Tau+Se group, 78.00±9.29 mg/dL. The plasma triglyceride levels in the exercise group of adult rats were as follows:

DDDW+Ex group, 91.00±5.63 mg/dL; 1%Tween80+Ex group, 91.22±8.68 mg/dL; Vit E+Ex group, 87.80±7.95 mg/dL; and Tau+Ex group, 71.60±3.54 mg/dL (Figure 21). The results showed that there was no significant difference in plasma triglyceride levels in all treatments of adult rats in both the sedentary group and the exercise group.

In comparison between the sedentary group and the exercise group, no significant difference in plasma triglyceride levels were found in all treatments.

- Triglyceride levels of Middle-aged Rats

The plasma triglyceride level in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 135.33±10.89 mg/dL; 1%Tween80+Se group, 106.89±9.51 mg/dL; Vit E+Se group, 99.89±12.38 mg/dL; and Tau+Se group, 98.33±8.90 mg/dL. The plasma triglyceride level in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 141.43±11.89 mg/dL; 1%Tween80+Ex group, 130.29±10.22 mg/dL; Vit E+Ex group, 100.89±14.71 mg/dL; and Tau+Ex group, 91.00±6.70 mg/dL (Figure 21). The results showed that plasma triglyceride levels in the middle-aged rats following DDD water treatment in the exercise group was significantly higher than both taurine supplement and vitamin E supplement in both sedentary group and exercise groups, DDD water treatment in the sedentary group was significantly higher than both taurine supplement and vitamin E supplement in both sedentary group and exercise groups, and 1% Tween 80 treatment in the exercise group was significantly higher than vitamin E treatment in the exercise group ($P<0.05$). When compared between the sedentary group and the exercise group, there was no significant difference in all treatment groups.

- The Comparison between Adult and Middle-aged Rats of Triglyceride Levels

The result of the blood parameters of triglyceride levels when comparison between adult rats and middle-aged rats (Figure 21) found that the middle-aged rats in the sedentary group following DDD water treatment and the exercise in middle-aged rats following DDD water treatment, 1% Tween 80 treatment and taurine treatment were significantly higher than same treatment in adult rats ($P<0.05$).

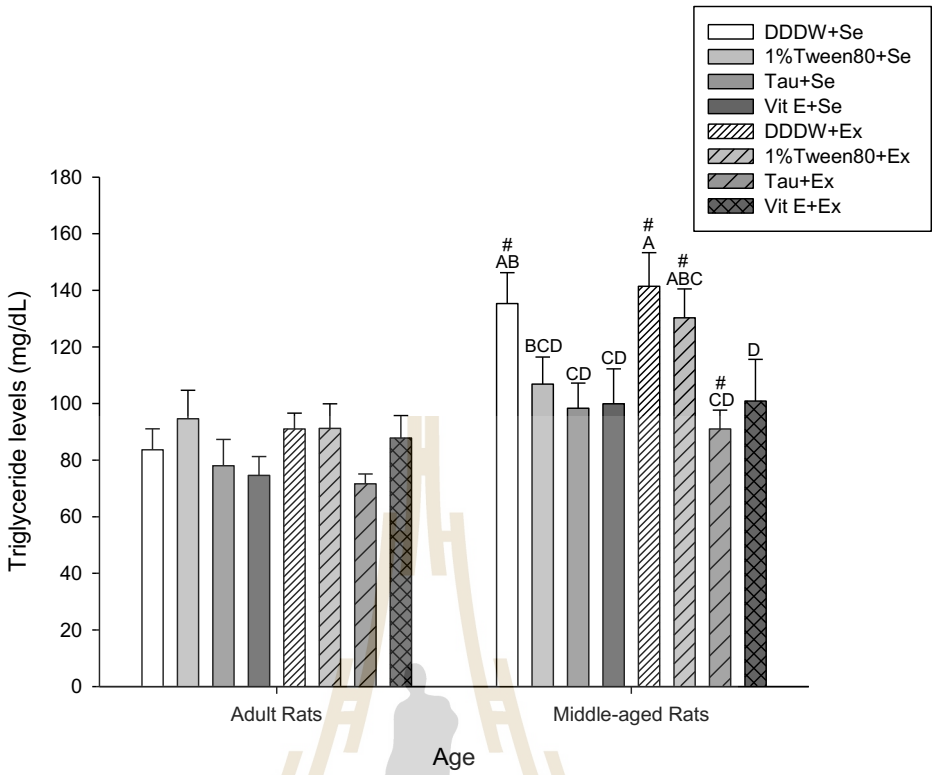

Figure 21 Effects of taurine and vitamin E supplement in conjunction with exercise on triglyceride level of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in middle-aged group (upper case). Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity. In not have symbol showed no significant difference between all comparison.

- Total Lactate Dehydrogenase (LDH) Levels of Adult Rats

The plasma LDH levels in the sedentary group of adult rats were as follows: DDDW+Se group, 4,092.43±404.14 U/L; 1%Tween80+Se group, 3,566.60±290.18 U/L; Vit E+Se group, 3,222.30±317.21 U/L; and Tau+Se group, 3,176.00±342.29 U/L. The plasma triglyceride levels in the exercise group of adult rats were as follows:

DDDW+Ex group, 4,354.00±311.10 U/L; 1%Tween80+Ex group, 3,979.40±450.54 U/L; Vit E+Ex group, 3,077.00±357.82 U/L; and Tau+Ex group, 3,317.60±297.16 U/L (Figure 22). The results showed that plasma LDH levels following DDD water treatment in the exercise group was significantly higher than taurine supplement and vitamin E supplement in the sedentary groups and taurine supplement in the exercise group (P<0.05). In comparison between the sedentary group and the exercise group, no significant difference in plasma LDH levels were found in all treatments.

- Total Lactate Dehydrogenase (LDH) Levels of Middle-aged Rats

The plasma total LDH levels in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 1,058.60±202.01 U/L; 1%Tween80+Se group, 3,093.14±525.26 U/L; Vit E+Se group, 2,500.00±629.32 U/L; and Tau+Se group, 1,441.50±277.30 U/L. The plasma total LDH levels in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 3,112.00±626.65 U/L; 1%Tween80+Ex group, 4,688.40±592.41 U/L; Vit E+Ex group, 2,554.13±752.02 U/L; and Tau+Ex group, 1,403.57±439.23 U/L (Figure 22). The results showed that plasma total LDH levels following 1% Tween 80 treatment in the exercise group was significantly higher than taurine supplement, vitamin E supplement, and DDD water treatment in the sedentary groups and taurine supplement and vitamin E treatment in the exercise groups (P<0.05) and DDD water treatment in the exercise group and 1% Tween 80 treatment in the sedentary group were significantly higher than DDD water treatment in the sedentary group (P<0.05). In comparison between the sedentary group and the exercise group, exercise significant increased the plasma total LDH in DDD water treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats of Total Lactate Dehydrogenase (LDH) Levels

The result of the blood parameters of total LDH levels when comparison between adult rats and middle-aged rats (Figure 22) found that the adult rats in the sedentary group following DDD water treatment and taurine treatment and in the exercise in adult rats following taurine treatment were significantly higher than same treatment in middle-aged rats ($P<0.05$).

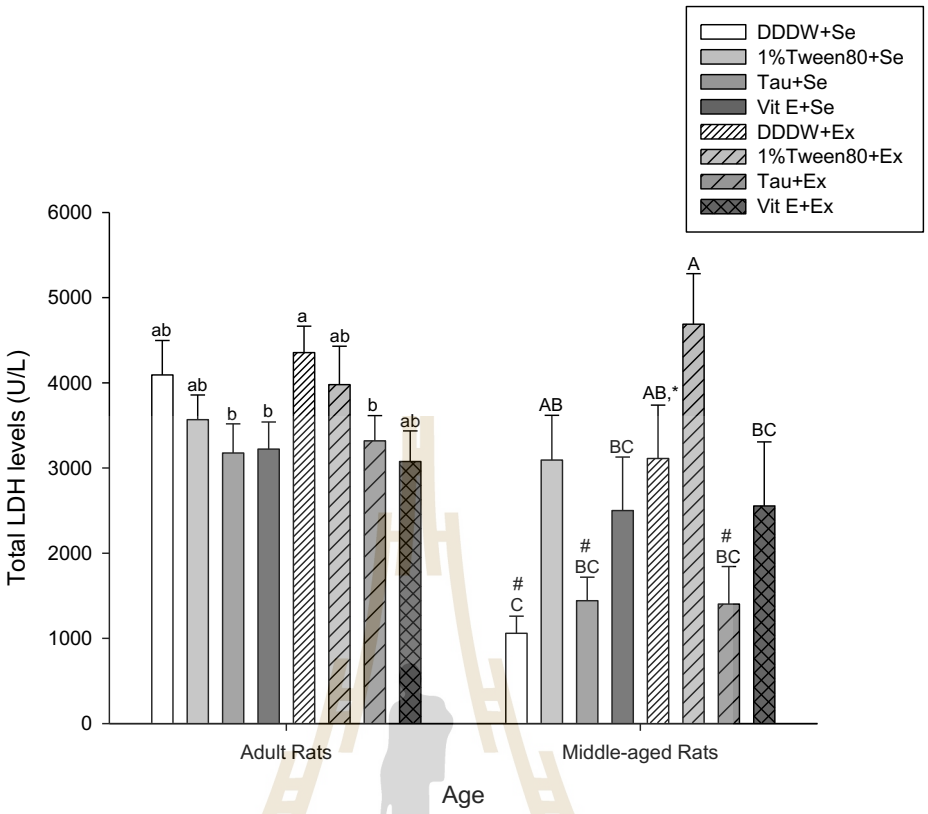

Figure 22 Effects of taurine and vitamin E supplement in conjunction with exercise on total LDH levels of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Liver Function Test

- Aspartate Transaminase (AST) Levels of Adult Rats

The plasma AST levels in the sedentary group of adult rats were as follows: DDDW+Se group, 126.60±19.50 U/L; 1%Tween80+Se group, 89.40±3.12 U/L; Vit E+Se group, 91.00±4.60 U/L; and Tau+Se group, 95.80±3.48 U/L. The plasma AST levels in the exercise group of adult rats were as follows: DDDW+Ex group, 114.78±8.61 U/L; 1%Tween80+Ex group, 93.90±1.45 U/L; Vit E+Ex group, 105.56±6.11 U/L; and Tau+Ex group, 101.70±5.45 U/L (Figure 23). The results showed that DDD water treatment in the sedentary group was significantly higher than both the sedentary and the exercise groups following taurine supplement, vitamin E treatment, and 1% Tween 80 treatment groups, and DDD water treatment in the exercise group was significantly higher than 1% Tween 80 treatment in the exercise group and taurine supplement, vitamin E treatment, and 1% Tween 80 treatment in the sedentary groups ($P<0.05$). In comparison between the sedentary group and the exercise group, no significant difference in plasma AST levels were found in all treatments.

- Aspartate Transaminase (AST) Levels of Middle-aged Rats

The plasma AST levels in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 83.67±7.39 U/L; 1%Tween80+Se group, 94.60±10.06 U/L; Vit E+Se group, 74.60±6.66 U/L; and Tau+Se group, 78.00±9.29 U/L. The plasma AST levels in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 91.00±5.63 U/L; 1%Tween80+Ex group, 91.22±8.68 U/L; Vit E+Ex group, 87.80±7.95 U/L; and Tau+Ex group, 71.60±3.54 U/L (Figure 23). The results in the

sedentary group showed that plasma AST levels following 1% Tween 80 treatment was significantly higher than DDD water treatment (P<0.05). There was no significant difference in plasma AST levels of middle-aged in all treatments of exercise group. When compared between the sedentary group and the exercise group, there was no significant difference in all treatment groups.

- The Comparison between Adult and Middle-aged Rats of AST Levels

The result of the blood parameters of AST levels when comparison between adult rats and middle-aged rats (Figure 23) found that the middle-aged rats in the sedentary group following 1% Tween 80 treatment and vitamin E treatment and in the exercise in middle-aged rats following DDD water treatment and 1% Tween 80 treatment were significantly higher than same treatment in adult rats (P<0.05).

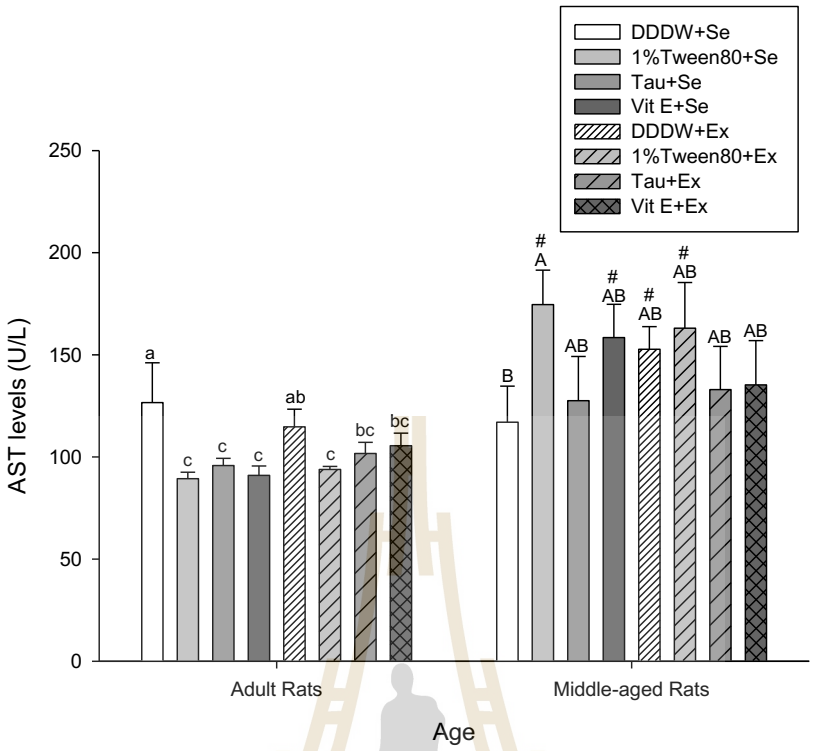

Figure 23 Effects of taurine and vitamin E supplement in conjunction with exercise on AST levels of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case).. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Alanine Transaminase (ALT) Levels of Adult Rats

The plasma ALT levels in the sedentary group of adult rats were as follows: DDDW+Se group, 39.75±2.43 U/L; 1%Tween80+Se group, 36.00±1.71 U/L; Vit E+Se group, 33.20±1.92 U/L; and Tau+Se group, 34.20±1.19 U/L. The plasma ALT levels in the exercise group of adult rats were as follows: DDDW+Ex group, 33.63±2.12 U/L; 1%Tween80+Ex group, 35.80±1.22 U/L; Vit E+Ex group, 37.30±2.17 U/L; and

Tau+Ex group, 33.90±2.19 U/L (Figure 24). The results showed that plasma ALT levels following DDD water treatment in the sedentary group was significantly higher than vitamin E supplement in the sedentary group and taurine supplement and DDD water treatment in the exercise groups (P<0.05). In comparison between the sedentary group and the exercise group, no significant difference in plasma ALT levels were found in all treatments.

- Alanine Transaminase (ALT) Levels of Middle-Aged Rats

The plasma ALT levels in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 49.22±5.08 U/L; 1%Tween80+Se group, 53.22±5.03 U/L; Vit E+Se group, 68.10±13.68 U/L; and Tau+Se group, 55.11±6.60 U/L. The plasma ALT levels in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 64.44±3.01 U/L; 1%Tween80+Ex group, 52.71±7.70 U/L; Vit E+Ex group, 48.88±4.60 U/L; and Tau+Ex group, 71.11±7.56 U/L (Figure 24). The results showed that taurine supplement in the exercise group was significantly higher than all treatment groups of both the sedentary and exercise groups (P<0.05). In comparison between the sedentary group and the exercise group, exercise significantly increased plasma ALT level in DDD water treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats of ALT Levels

The result of the blood parameters of ALT levels when comparison between adult rats and middle-aged rats (Figure 24) found that the middle-aged rats in the sedentary group following 1% Tween 80 treatment, taurine treatment and vitamin E treatment and in the exercise in middle-aged rats following DDD water treatment,

taurine treatment and vitamin E treatment were significantly higher than same treatment in adult rats (P<0.05).

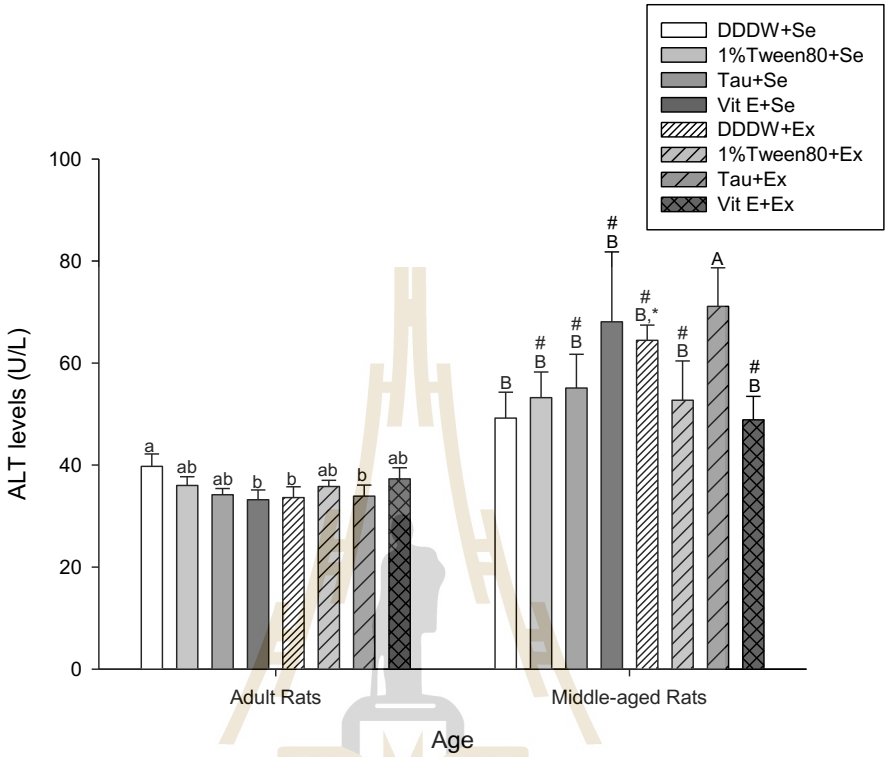

Figure 24 Effects of taurine and vitamin E supplement in conjunction with exercise on AST levels of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Kidney Function Test

- Blood Urea Nitrogen (BUN) Levels of Adult Rats

The plasma BUN levels in the sedentary group of adult rats were as follows: DDDW+Se group, 20.33±0.8 mg/dL; 1%Tween80+Se group, 18.22±0.8 mg/dL; Vit E+Se group, 21.30±1.0 mg/dL; and Tau+Se group, 19.90±1.3 mg/dL. The plasma BUN levels in the exercise group of adult rats were as follows: DDDW+Ex group, 21.00±1.1 mg/dL; 1%Tween80+Ex group, 21.50±0.9 mg/dL; Vit E+Ex group, 21.50±1.1 mg/dL; and Tau+Ex group, 21.20±1.3 mg/dL (Figure 25). The results showed that vitamin E treatment and 1% Tween 80 treatment in the exercise group were significantly higher than 1% Tween 80 treatment in the sedentary group. In comparison between the sedentary group and the exercise group, exercise significantly increased plasma BUN levels in 1% Tween 80 treatment ($P<0.05$).

- Blood Urea Nitrogen (BUN) Levels of Middle-aged Rats

The plasma BUN levels in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 17.78±0.9 mg/dL; 1%Tween80+Se group, 19.11±0.8 mg/dL; Vit E+Se group, 17.20±0.8 mg/dL; and Tau+Se group, 15.67±0.7 mg/dL. The plasma BUN levels in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 21.78±1.0 mg/dL; 1%Tween80+Ex group, 20.25±1.7 mg/dL; Vit E+Ex group, 19.56±1.0 mg/dL; and Tau+Ex group, 16.67±0.8 mg/dL (Figure 25). The results showed that plasma BUN levels following DDD water treatment was significantly higher than DDD water treatment, vitamin E treatment, and taurine supplement in the sedentary group and taurine supplement in the exercise group ($P<0.05$), 1% Tween 80 treatment in the exercise group was significantly higher than

vitamin E treatment and taurine treatment in the sedentary groups and taurine treatment in the exercise group (P<0.05), vitamin E treatment in the exercise group was significantly higher than both the sedentary and the exercise group following taurine treatment (P<0.05), and 1% Tween 80 treatment in the sedentary group was significantly higher than taurine supplement in the sedentary group (P<0.05). In comparison between the sedentary group and the exercise group, exercise significantly increased plasma AST levels in DDD water treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats of BUN Levels

The result of the blood parameters of BUN levels when comparison between adult rats and middle-aged rats (Figure 25) found that the adult rats in the sedentary group following DDD water treatment, taurine treatment and vitamin E treatment and in the exercise in middle-aged rats following taurine treatment were significantly higher than same treatment in middle-aged rats (P<0.05).

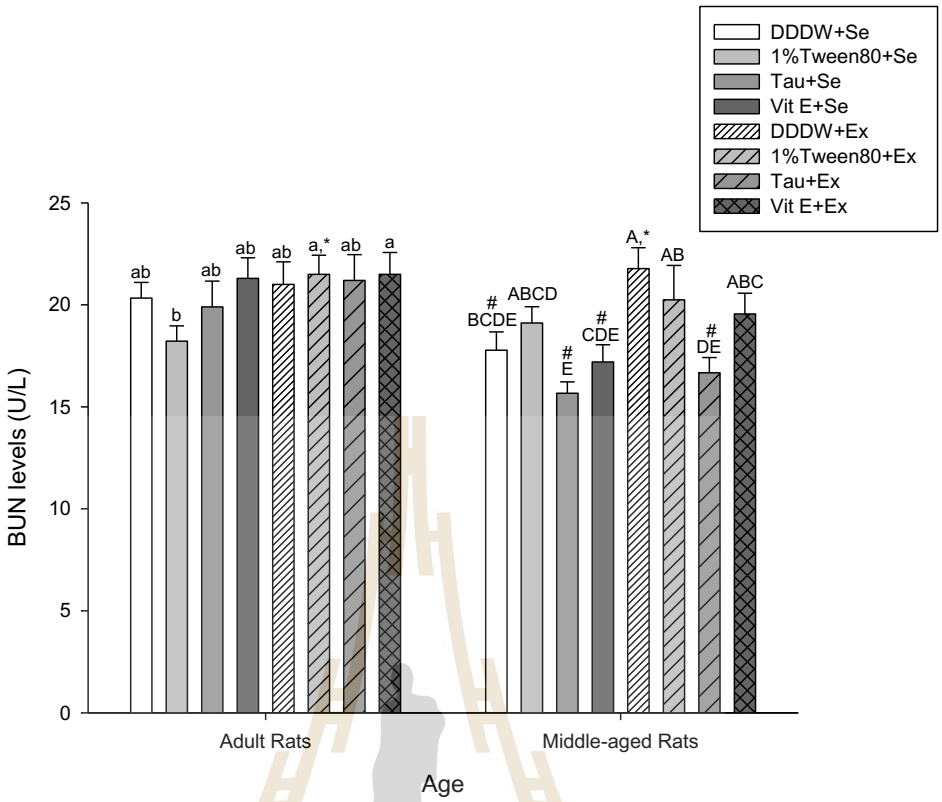

Figure 25 Effects of taurine and vitamin E supplement in conjunction with exercise on BUN levels of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Creatinine Levels of Adult Rats

The plasma creatinine levels in the sedentary group of adult rats were as follows: DDDW+Se group, 0.633±0.03 mg/dL; 1%Tween80+Se group, 0.608±0.03 mg/dL; Vit E+Se group, 0.681±0.03 mg/dL; and Tau+Se group, 0.882±0.03 mg/dL. The plasma creatinine levels in the exercise group of adult rats were as follows:

DDDW+Ex group, 0.594±0.02 mg/dL; 1%Tween80+Ex group, 0.663±0.02 mg/dL; Vit E+Ex group, 0.655±0.04 mg/dL; and Tau+Ex group, 0.748±0.03 mg/dL (Figure 26). The results showed that the plasma creatinine levels following taurine supplement in the sedentary group was significantly higher than all treatments in both the sedentary group and the exercise group (P<0.05), taurine supplement in the exercise group was significantly higher than both the sedentary and exercise group following 1% Tween 80 treatment and DDD water treatment and vitamin E treatment in the exercise groups (P<0.05). In comparison between the sedentary group and the exercise group, exercise significantly decreased the plasma creatinine level in taurine supplement (P<0.05).

- Creatinine Levels of Middle-aged Rats

The plasma creatinine levels in the sedentary group of middle-aged rats were as follows: DDDW+Se group, 0.647±0.03 mg/dL; 1%Tween80+Se group, 0.653±0.03 mg/dL; Vit E+Se group, 0.630±0.02 mg/dL; and Tau+Se group, 0.713±0.03 mg/dL. The plasma creatinine levels in the exercise group of middle-aged rats were as follows: DDDW+Ex group, 0.720±0.05 mg/dL; 1%Tween80+Ex group, 0.620±0.03 mg/dL; Vit E+Ex group, 0.642±0.02 mg/dL; and Tau+Ex group, 0.754±0.03 mg/dL (Table 8). The results showed that creatinine levels following taurine supplement in the exercise group was significantly higher than both the sedentary and exercise groups following 1% Tween 80 treatment and vitamin E treatment and DDD water treatment in the sedentary groups (P<0.05), DDD water treatment in the exercise group and taurine treatment in the sedentary group were significantly higher than 1% Tween 80 treatment in the exercise group (P<0.05). In comparison between the sedentary group and the exercise group, there was no significant difference between all treatment groups.

- The Comparison between Adult and Middle-aged Rats of Creatinine Levels

The result of the blood parameters of creatinine levels when comparison between adult rats and middle-aged rats (Figure 66) found that the adult rats in the sedentary group following taurine treatment and in the exercise in middle-aged rats following DDD water treatment were significantly higher than same treatment (P<0.05).

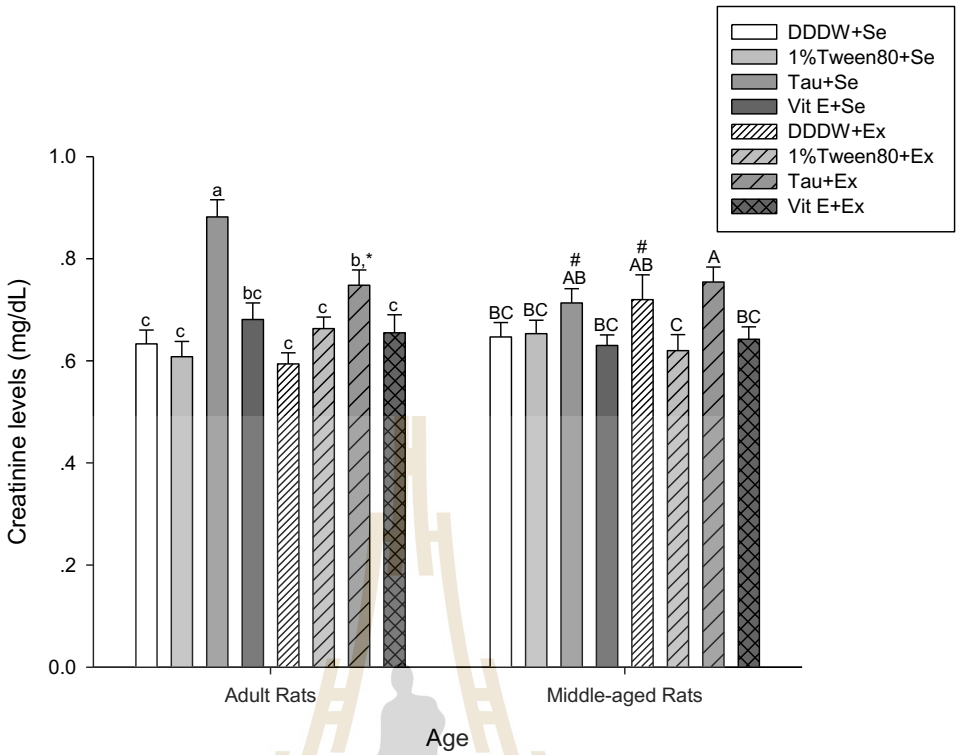

Figure 26 Effects of taurine and vitamin E supplement in conjunction with exercise on creatinine levels of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

4.1.3 Brains Biochemical Analysis

4.1.3.1 Malondialdehyde (MDA) Levels

- Basal Forebrain of Adult Rats

The MDA levels in basal forebrain of adult rats in the sedentary groups were as follows: DDDW+Se group, 0.0038±0.0003 nmol/mg protein; 1%Tween80+Se group, 0.0042±0.0004 nmol/mg protein; Vit E+Se group, 0.0048±0.0007 nmol/mg protein; and Tau+Se group, 0.0056±0.0005 nmol/mg protein. The MDA levels in basal forebrain of adult rats in the exercise group were follows: DDDW+Ex group, 0.0041±0.0008 nmol/mg protein; 1%Tween80+Ex group, 0.0035±0.0003 nmol/mg protein; Vit E+Ex group, 0.0062±0.0011 nmol/mg protein; and Tau+Ex group, 0.0051±0.0003 nmol/mg protein (Figure 27). MDA level in basal forebrain following vitamin E treatment in the exercise group was significantly higher than both the sedentary and the exercise groups of 1% Tween 80 treatment and DDD water treatment (P<0.05), taurine treatment in the sedentary group was significantly higher than DDD water treatment in the sedentary group and 1% Tween 80 treatment in the exercise group (P<0.05). When compared between sedentary group and exercise group, there was no significant difference between all treatment groups.

- Basal Forebrain of Middle-aged Rats

The MDA levels in basal forebrain of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.0056±0.0009 nmol/mg tissue; 1%Tween80+Se group, 0.0055±0.0009 nmol/mg tissue; Vit E+Se group, 0.0062±0.0004 nmol/mg tissue; and Tau+Se group, 0.0078±0.0008 nmol/mg tissue. The MDA levels in basal forebrain of middle-aged rats in the execise groups were as follows: DDDW+Ex group,

0.0038±0.0003 nmol/mg tissue; 1%Tween80+Ex group, 0.0042±0.0004 nmol/mg tissue; Vit E+Ex group, 0.0048±0.0007 nmol/mg tissue; and Tau+Ex group, 0.0056±0.0005 nmol/mg tissue (Figure 27). The results in the sedentary group showed that MDA level in basal forebrain following taurine treatment was significantly higher than both the sedentary and exercise groups following DDD water treatment and 1% Tween 80 treatment and taurine treatment and vitamin E treatment in the exercise group (P<0.05), vitamin E treatment in the sedentary group was significantly higher than 1% Tween 80 treatment and DDD water treatment in the exercise group(P<0.05), 1% Tween 80 treatment and DDD water treatment in the sedentary group and taurine treatment in the exercise group were significantly higher than DDD water in the exercise group (P<0.05). In the exercise group, MDA levels in basal forebrain following taurine treatment was significantly higher than DDD water treatment (P<0.05). When compared between the sedentary group and the exercise group, exercise significantly reduced MDA levels in basal forebrain following vitamin E treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats in Basal Forebrain of Malondialdehyde (MDA) Levels

The result of MDA levels of basal forebrain when comparison between adult rats and middle-aged rats (Figure 27) found that the middle-aged rats in the sedentary group following taurine treatment was significantly higher than same treatment (P<0.05). In the exercise group, there was no significant difference in all treatment groups.

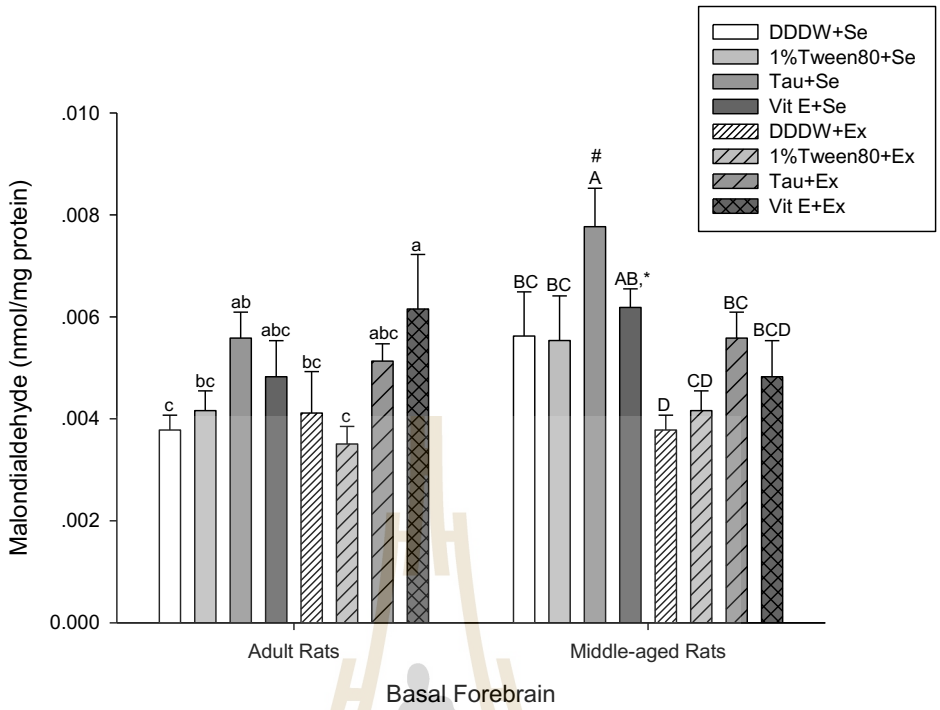

Figure 27 Effects of taurine and vitamin E supplement in conjunction with exercise on MDA levels in basal forebrain of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Cerebral Cortex of Adult Rats

The MDA levels in cerebral cortex of adult rats in the sedentary group were as follows: DDDW+Se group, 0.0048±0.0005 nmol/mg tissue; 1%Tween80+Se group, 0.0052±0.0004 nmol/mg tissue; Vit E+Se group, 0.0051±0.0003 nmol/mg tissue; and Tau+Se group, 0.0060±0.0004 nmol/mg tissue. The MDA levels in cerebral cortex of

adult rats in the exercise group were follows: DDDW+Ex group, 0.0044±0.0003 nmol/mg tissue; 1%Tween80+Ex group, 0.0053±0.0003 nmol/mg tissue; Vit E+Ex group, 0.0053±0.0005 nmol/mg tissue; and Tau+Ex group, 0.0043±0.0005 nmol/mg tissue (Figure 28). The results showed that taurine supplement in the sedentary group was significantly higher than taurine treatment and DDD water treatment in the exercise groups. In comparison between sedentary and exercise group, exercise significantly decreased MDA levels in cerebral cortex of adult rats treated with taurine supplement (P<0.05).

- Cerebral Cortex of Middle-aged Rats

The MDA levels in cerebral cortex of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.010±0.0019 nmol/mg tissue; 1%Tween80+Se group, 0.012±0.0014 nmol/mg tissue; Vit E+Se group, 0.0009±0.0007 nmol/mg tissue; and Tau+Se group, 0.0010±0.0009 nmol/mg tissue. The MDA levels in cerebral cortex of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.010±0.0016 nmol/mg tissue; 1%Tween80+Ex group, 0.009±0.0007 nmol/mg tissue; Vit E+Ex group, 0.007±0.0008 nmol/mg tissue; and Tau+Ex group, 0.009±0.0004 nmol/mg tissue (Figure 28). The results showed that MDA levels in cerebral cortex following 1% Tween 80 treatment in the sedentary group was significantly higher than vitamin E treatment in the exercise group (P<0.05). When compared between sedentary group and exercise group, exercise significantly lower MDA levels in cerebral cortex following 1% Tween 80 treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats in Cerebral Cortex of Malondialdehyde (MDA) Levels

The result of MDA levels of cerebral cortex when comparison between adult rats and middle-aged rats (Figure 28) found that the middle-aged rats in all treatment group in the sedentary group and in the exercise group following DDD water treatment, 1% Tween 80 treatment and taurine treatment were significantly higher than same treatment in adult rats ($P<0.05$).

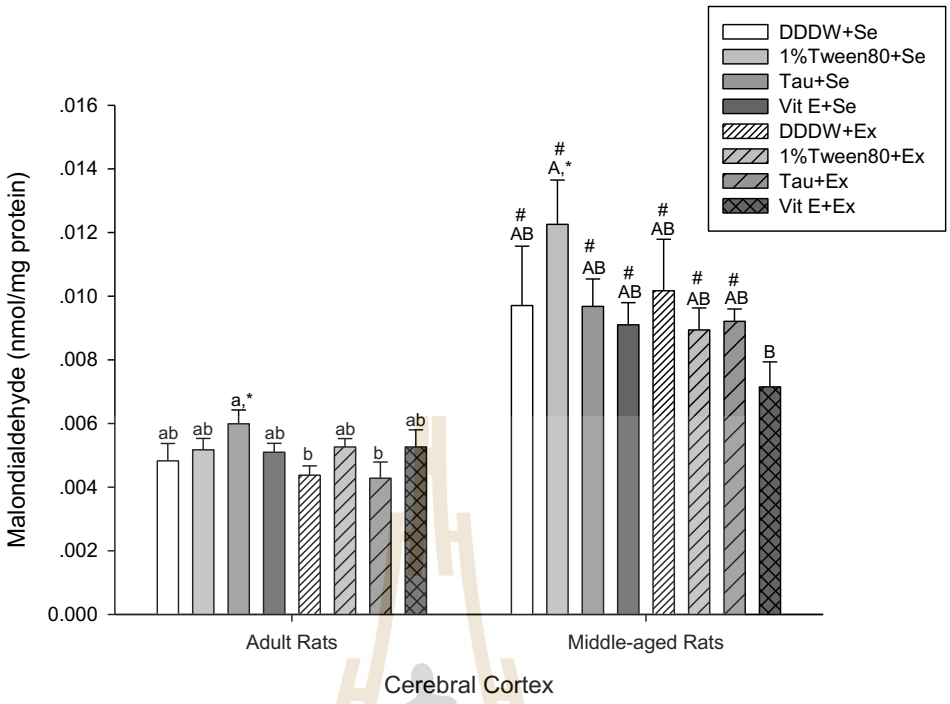

Figure 28 Effects of taurine and vitamin E supplement in conjunction with exercise on MDA levels in cerebral cortex of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Hippocampus of Adult Rats

The MDA levels in hippocampus of adult rats in the sedentary group were as follows: DDDW+Se group, 0.0038±0.0002 nmol/mg tissue; 1%Tween80+Se group, 0.0034±0.0003 nmol/mg tissue; Vit E+Se group, 0.0045±0.0006 nmol/mg tissue; and Tau+Se group, 0.0041±0.0003 nmol/mg tissue. The MDA levels in cerebral cortex of

adult rats in the exercise group were follows: DDDW+Ex group, 0.0036±0.0002 nmol/mg tissue; 1%Tween80+Ex group, 0.0032±0.0003 nmol/mg tissue; Vit E+Ex group, 0.0035±0.0004 nmol/mg tissue; and Tau+Ex group, 0.0035±0.0004 nmol/mg tissue (Figure 29). The result showed that vitamin E treatment in the sedentary group was significantly higher than 1% Tween 80 treatment in the exercise group (P<0.05). When compared between the sedentary group and exercise group, there was no significant difference between all treatment groups.

- Hippocampus of Middle-aged Rats

The MDA levels in hippocampus of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.0042±0.0004 nmol/mg tissue; 1%Tween80+Se group, 0.0043±0.0005 nmol/mg tissue; Vit E+Se group, 0.0035±0.0002 nmol/mg tissue; and Tau+Se group, 0.0045±0.0005 nmol/mg tissue. The MDA levels in hippocampus of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.0035±0.0002 nmol/mg tissue; 1%Tween80+Ex group, 0.0037±0.0003 nmol/mg tissue; Vit E+Ex group, 0.0043±0.0004 nmol/mg tissue; and Tau+Ex group, 0.0031±0.0002 nmol/mg tissue (Figure 29). The results showed that the MDA levels in hippocampus following taurine treatment, 1% Tween 80 treatment, and DDD water treatment in the sedentary group and vitamin E treatment in the exercise group were significantly higher than taurine treatment in the exercise group (P<0.05). When compared between sedentary group and exercise group, exercise significantly lower MDA levels in hippocampus following taurine treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats in Hippocampus of Malondialdehyde (MDA) Levels

The result of MDA levels of hippocampus when comparison between adult rats and middle-aged rats (Figure 29) found that there was no significant difference in all treatment group both the sedentary and exercise groups.

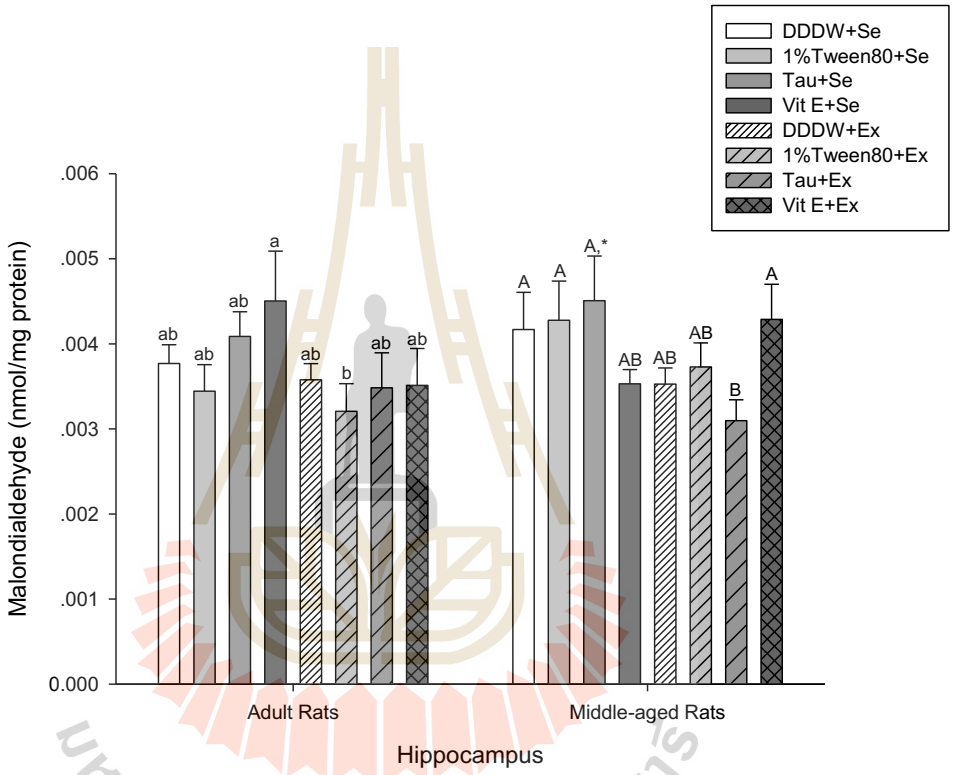

Figure 29 Effects of taurine and vitamin E supplement in conjunction with exercise on MDA levels in hippocampus of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age.

- Striatum of Adult Rats

The MDA levels in striatum of adult rats in the sedentary group were as follows: DDDW+Se group, 0.0032±0.0005 nmol/mg tissue; 1%Tween80+Se group, 0.0034±0.0003 nmol/mg tissue; Vit E+Se group, 0.0046±0.0003 nmol/mg tissue; and Tau+Se group, 0.0042±0.0003 nmol/mg tissue. . The MDA levels in cerebral cortex of adult rats in exercise group were follows: DDDW+Ex group, 0.0036±0.0006 nmol/mg tissue; 1%Tween80+Ex group, 0.0039±0.0003 nmol/mg tissue; Vit E+Ex group, 0.0034±0.0005 nmol/mg tissue; and Tau+Ex group, 0.0053±0.0003 nmol/mg tissue (Figure 14). The MDA levels in striatum of adult rats taurine treatment in the exercise group was significantly higher than both the sedentary and the exercise groups following 1% Tween 80 treatment and DDD water treatment group and vitamin E treatment in the exercise group (P<0.05), vitamin E treatment in the sedentary group was significantly higher than vitamin E treatment in the exercise group and 1% Tween 80 treatment and DDD water treatment in the sedentary groups (P<0.05). When compared between sedentary group and exercise group, exercise significantly decreased the MDA levels in striatum in vitamin E treatment (P<0.05), and significantly increased the MDA levels in striatum in taurine supplement (P<0.05).

- Striatum of Middle-aged Rats

The MDA levels in striatum of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.0061±0.0006 nmol/mg tissue; 1%Tween80+Se group, 0.0074±0.0009 nmol/mg tissue; Vit E+Se group, 0.0080±0.0007 nmol/mg tissue; and Tau+Se group, 0.0078±0.0005 nmol/mg tissue. The MDA levels in hippocampus of middle-aged rats in the exercise groups were as follows: DDDW+Ex group,

0.0062±0.0005 nmol/mg tissue; 1%Tween80+Ex group, 0.0079±0.0005 nmol/mg tissue; Vit E+Ex group, 0.0065±0.0005 nmol/mg tissue; and Tau+Ex group, 0.0080±0.0008 nmol/mg tissue (Figure 30). The results showed that there was no significant difference in MDA levels in striatum rats between all treatment groups in both the sedentary and exercise groups. When compared between sedentary group and exercise group, there was no significant difference in the MDA levels in striatum between all treatment groups.

- The Comparison between Adult and Middle-aged Rats in Striatum of Malondialdehyde (MDA) Levels

The result of MDA levels of striatum when comparison between adult rats and middle-aged rats (Figure 30) found that the middle-aged rats in all treatment group in the sedentary group and in the exercise group following DDD water treatment, 1% Tween 80 treatment and vitamin E treatment were significantly higher than same treatment in adult rats ($P<0.05$).

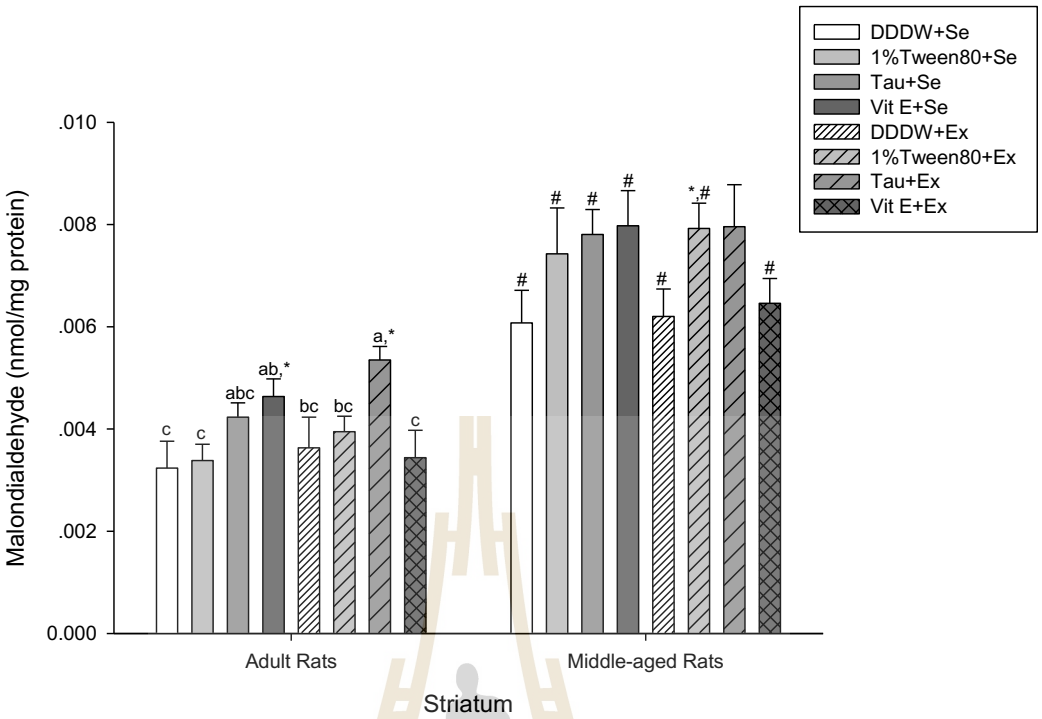

Figure 30 Effects of taurine and vitamin E supplement in conjunction with exercise on MDA levels in striatum of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

4.1.3.2 Hydrogen Peroxide (H_2O_2) Levels

- Basal Forebrain of Adult Rats

The H_2O_2 levels in basal forebrain of adult rats in the sedentary group were as follows: DDDW+Se group, 5.55±0.57 µM/mg protein; 1%Tween80+Se group, 6.75±0.64 µM/mg protein; Vit E+Se group, 7.71±1.12 µM/mg protein; and Tau+Se group, 9.41±0.74 µM/mg protein. The H_2O_2 levels in basal forebrain of adult rats in the exercise group were as follows: DDDW+Ex group, 6.34±0.42 µM/mg protein; 1%Tween80+Ex group, 8.53±1.15 µM/mg protein; Vit E+Ex group, 11.68±1.34 µM/mg protein; and Tau+Ex group, 9.45±0.090µM/mg protein (Figure 31). The H_2O_2 levels in basal forebrain in adult rats following vitamin E treatment in the exercise group was significantly higher than both the sedentary and exercise groups following 1% Tween 80 treatment and DDD water treatment, and vitamin E treatment in the sedentary group (P<0.05), both the sedentary and the exercise group following taurine treatment were significantly higher than both the sedentary and the exercise groups of DDD water treatment and 1% Tween 80 treatment in the sedentary group (P<0.05), and 1% Tween 80 treatment in the exercise group was significantly higher than DDD water in the sedentary group (P<0.05). There was no significant difference between all treatments in exercise group. In comparison between the sedentary group and the exercise group, there was no significant difference between all treatment groups.

- Basal Forebrain of Middle-aged Rats

The H_2O_2 levels in basal forebrain of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 7.31±1.16 µM/mg protein; 1%Tween80+Se group, 12.85±1.79 µM/mg protein; Vit E+Se group, 9.09±2.16 µM/mg protein; and Tau+Se

group, 10.97±0.74 µM/mg protein. The H_2O_2 levels in basal forebrain of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 10.93±1.97 µM/mg protein; 1%Tween80+Ex group, 7.81±0.89 µM/mg protein; Vit E+Ex group, 8.50±1.04 µM/mg protein; and Tau+Ex group, 12.64±1.46 µM/mg protein (Figure 31). The results showed that H_2O_2 levels in middle-aged basal forebrain following 1% Tween 80 treatment in the sedentary group and taurine supplement in the exercise group were significantly higher than DDD water treatment in the sedentary group (P<0.05). In comparison between sedentary and exercise group, exercise significantly lower of the H_2O_2 levels in basal forebrain following 1% Tween 80 treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats in Basal Forebrain of Hydrogen Peroxide (H_2O_2) Levels

The result of H_2O_2 levels of basal forebrain when comparison between adult rats and middle-aged rats (Figure 31) found that the middle-aged rats in the sedentary group following 1% Tween 80 treatment and in the exercise in middle-aged rats following DDD water treatment were significantly higher than same treatment in the adult rats (P<0.05).

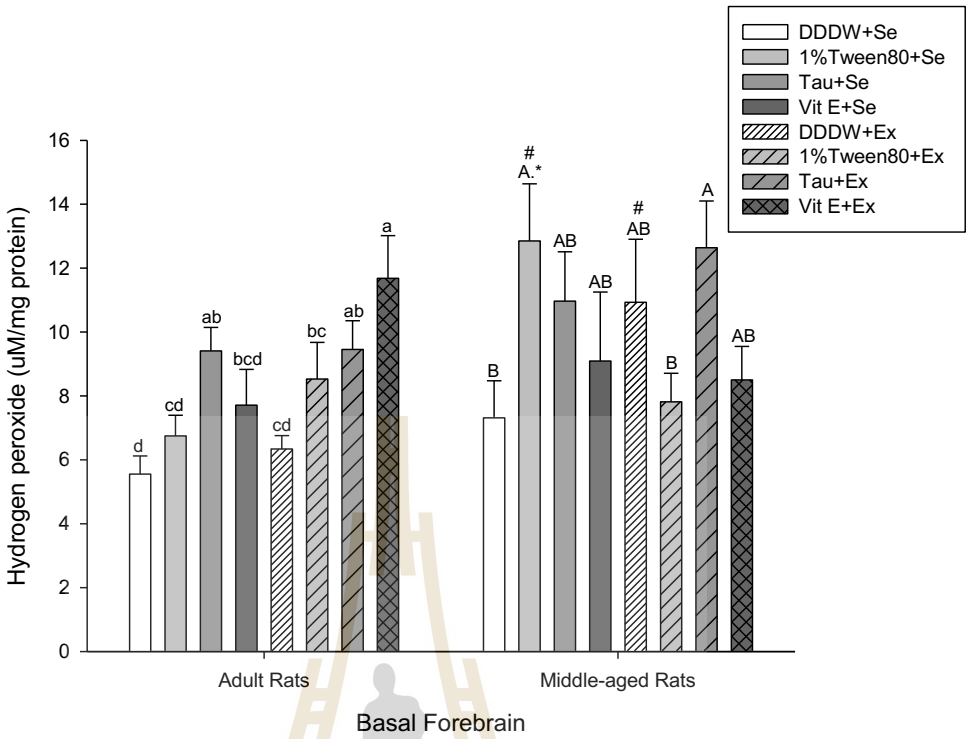

Figure 31 Effects of taurine and vitamin E supplement in conjunction with exercise on H_2O_2 levels in basal forebrain of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Cerebral Cortex of Adult Rats

The H_2O_2 levels in cerebral cortex of adult rats in the sedentary group were as follows: DDDW+Se group, 3.83±0.73 µM/mg protein; 1%Tween80+Se group, 4.23±0.63 µM/mg protein; Vit E+Se group, 4.63±0.61 µM/mg protein; and Tau+Se

group, 5.68±0.77 μM/mg protein. The H_2O_2 levels in cerebral cortex of adult rats in the exercise group were as follows: DDDW+Ex group, 4.85±0.76 μM/mg protein; 1%Tween80+Ex group, 5.17±0.52 μM/mg protein; Vit E+Ex group, 5.29±0.43 μM/mg protein; and Tau+Ex group, 3.70±0.48 μM/mg protein (Figure 32). The results showed that there was no significant difference in H_2O_2 levels in cerebral cortex between all treatment groups in both sedentary and exercise groups. When compared between the sedentary group and exercise group, the H_2O_2 levels in cerebral cortex following taurine supplement in conjunction with exercise was significantly lower than taurine supplement in the sedentary group ($P<0.05$).

- Cerebral Cortex of Middle-aged Rats

The H_2O_2 levels in cerebral cortex of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 6.11±0.44 μM/mg protein; 1%Tween80+Se group, 8.80±0.61 μM/mg protein; Vit E+Se group, 9.49±0.81 μM/mg protein; and Tau+Se group, 9.09±1.00 μM/mg protein. The H_2O_2 levels in cerebral cortex of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 9.34±1.39 μM/mg protein; 1%Tween80+Ex group, 10.23±1.60 μM/mg protein; Vit E+Ex group, 9.13±1.13 μM/mg protein; and Tau+Ex group, 7.44±0.87 μM/mg protein (Figure 32). The H_2O_2 levels in cerebral cortex of middle-aged rats in 1% Tween 80 treatment and DDD water treatment in the exercise group and vitamin E treatment in the sedentary group were significantly higher than DDD water treatment in the sedentary group ($P<0.05$). When compared between the sedentary group and the exercise group, exercise significantly increased the H_2O_2 levels in cerebral cortex following DDD water treatment ($P<0.05$).

- The Comparison between Adult and Middle-aged Rats in Cerebral Cortex of Hydrogen Peroxide (H_2O_2) Levels

The result of H_2O_2 levels of cerebral cortex when comparison between adult rats and middle-aged rats (Figure 32) found that in the middle-aged rats in all treatment groups both the sedentary and exercise groups were significantly higher than same treatment in the adult rats (P<0.05).

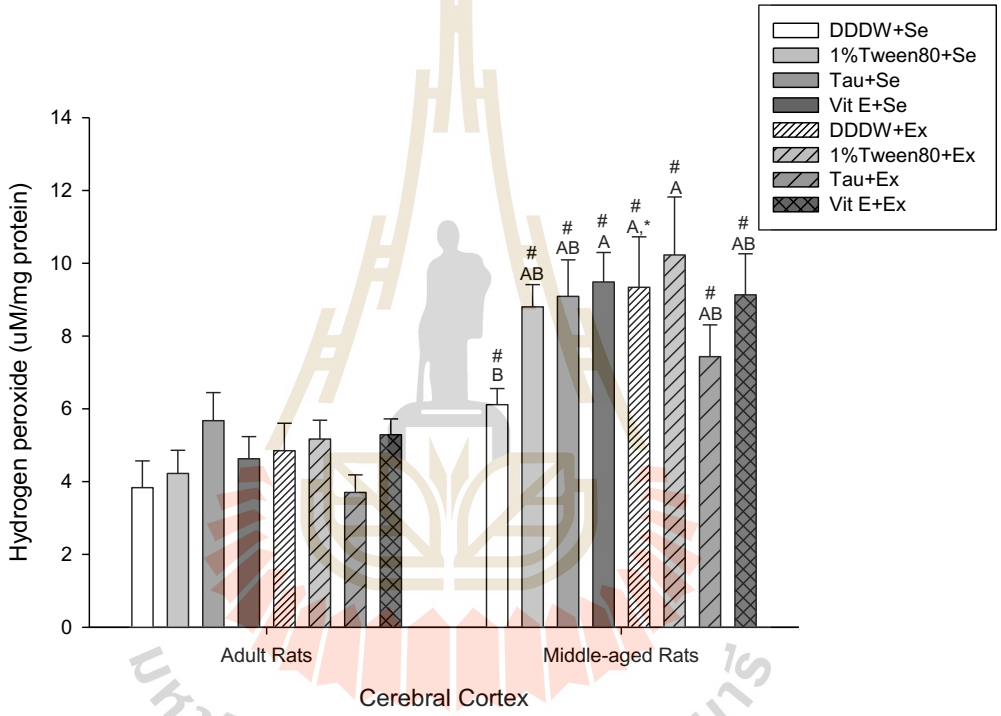

Figure 32 Effects of taurine and vitamin E supplement in conjunction with exercise on H_2O_2 levels in cerebral cortex of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity. In not have symbol showed no significant difference between all comparison.

- Hippocampus of Adult Rats

The H_2O_2 levels in hippocampus of adult rats in the sedentary group were as follows: DDDW+Se group, 3.71±0.28 µM/mg protein; 1%Tween80+Se group, 3.87±0.44 µM/mg protein; Vit E+Se group, 4.25±0.54 µM/mg protein; and Tau+Se group, 4.03±0.36 µM/mg protein. The H_2O_2 levels in hippocampus of adult rats in the exercise group were as follows: DDDW+Ex group, 3.26±0.31 µM/mg protein; 1%Tween80+Ex group, 3.55±0.20 µM/mg protein; Vit E+Ex group, 3.53±0.22 µM/mg protein; and Tau+Ex group, 4.39±0.74 µM/mg protein (Figure 33). The results showed that there was no significant difference in H_2O_2 levels in hippocampus between all treatment groups of both the sedentary group and the exercise groups. When compared between the sedentary group and exercise group, there was no significant difference in H_2O_2 levels in hippocampus between all treatment groups.

- Hippocampus of Middle-aged Rats

The H_2O_2 levels in hippocampus of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 4.24±0.42 µM/mg protein; 1%Tween80+Se group, 5.81±0.53 µM/mg protein; Vit E+Se group, 4.27±0.55 µM/mg protein; and Tau+Se group, 5.91±0.60 µM/mg protein. The H_2O_2 levels in hippocampus of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 5.38±0.31 µM/mg protein; 1%Tween80+Ex group, 5.12±0.41 µM/mg protein; Vit E+Ex group, 5.58±0.79 µM/mg protein; and Tau+Ex group, 4.09±0.45 µM/mg protein (Figure 33). The results showed that the H_2O_2 levels in hippocampus, following taurine treatment and 1% Tween 80 treatment in the sedentary group were significantly higher than vitamin E treatment and DDD water treatment in the sedentary group and taurine supplement in the exercise

group (P<0.05). In comparison between sedentary group and exercise group, in hippocampus following taurine treatment, 1% Tween 80 treatment in the sedentary group and DDD water treatment in the exercise group (P<0.05).

- The Comparison between Adult and Middle-aged Rats in Hippocampus of Hydrogen Peroxide (H_2O_2) Levels

The result of H_2O_2 levels of basal forebrain when comparison between adult rats and middle-aged rats (Figure 33) found that the middle-aged rats in the sedentary group following 1% Tween 80 treatment and taurine treatment and in the exercise in middle-aged rats following DDD water treatment, 1%Tween 80 treatment and vitamin E treatment were significantly higher than same treatment in the adult rats (P<0.05).

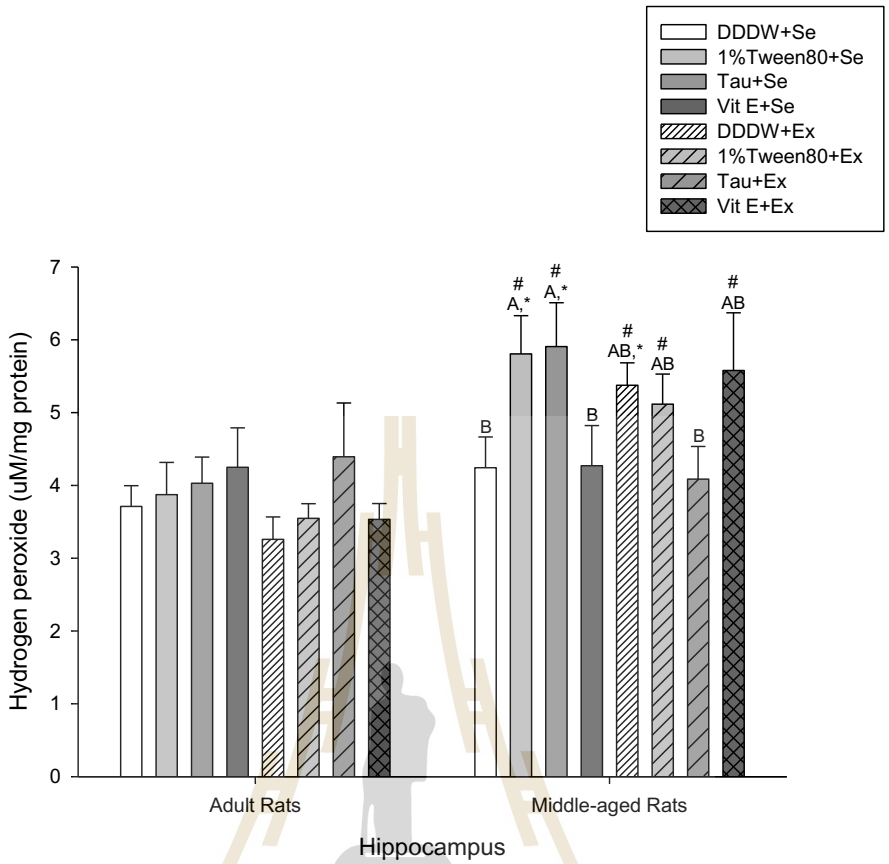

Figure 33 Effects of taurine and vitamin E supplement in conjunction with exercise on H_2O_2 levels in hippocampus of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity. In not have symbol showed no significant difference between all comparison.

- Striatum of Adult Rats

The H_2O_2 levels in striatum of adult rats in the sedentary group were as follows: DDDW+Se group, 3.60±0.33 μM/mg protein; 1%Tween80+Se group, 5.49±0.35 μM/mg protein; Vit E+Se group, 4.69±0.47 μM/mg protein; and Tau+Se group, 4.80±0.43 μM/mg protein. The H_2O_2 levels in striatum of adult rats in the exercise group were as follows: DDDW+Ex group, 3.96±0.42 μM/mg protein; 1%Tween80+Ex group, 5.79±0.62 μM/mg protein; Vit E+Ex group, 5.53±0.64 μM/mg protein; and Tau+Ex group, 3.79±0.34 μM/mg protein (Figure 34). The results showed that both the sedentary and the exercise group following 1% Tween 80 treatment and vitamin E treatment in the exercise group were significantly higher than both sedentary and exercise group of DDD water treatment, and taurine treatment in the exercise group (P<0.05). When compared between the sedentary group and the exercise group, there was no significant difference in H_2O_2 levels in hippocampus between all treatment groups.

- Striatum of Middle-aged Rats

The H_2O_2 levels in striatum of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 7.78±1.30 μM/mg protein; 1%Tween80+Se group, 5.89±1.15 μM/mg protein; Vit E+Se group, 7.78±0.64 μM/mg protein; and Tau+Se group, 8.13±0.86 μM/mg protein. The H_2O_2 levels in striatum of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 10.70±0.97 μM/mg protein; 1%Tween80+Ex group, 6.44±0.88 μM/mg protein; Vit E+Ex group, 6.09±0.81 μM/mg protein; and Tau+Ex group, 4.61±0.60 μM/mg protein (Figure 34). The results showed that the H_2O_2 levels in striatum following DDD water treatment was significantly

higher than all treatment group of both the sedentary and exercise groups (P<0.05), taurine supplement, vitamin E supplement, and DDD water treatment in the sedentary group were significantly higher than taurine supplement in the exercise group (P<0.05). In comparison between sedentary group and exercise group, exercise significantly lower the H_2O_2 levels in striatum following taurine treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats in Striatum of Hydrogen Peroxide (H_2O_2) Levels

The result of H_2O_2 levels of striatum when comparison between adult rats and middle-aged rats (Figure 34) found that the middle-aged rats in the sedentary group following DDD water treatment, vitamin E treatment and taurine treatment and in the exercise in middle-aged rats following DDD water treatment were significantly higher than same treatment in the adult rats (P<0.05).

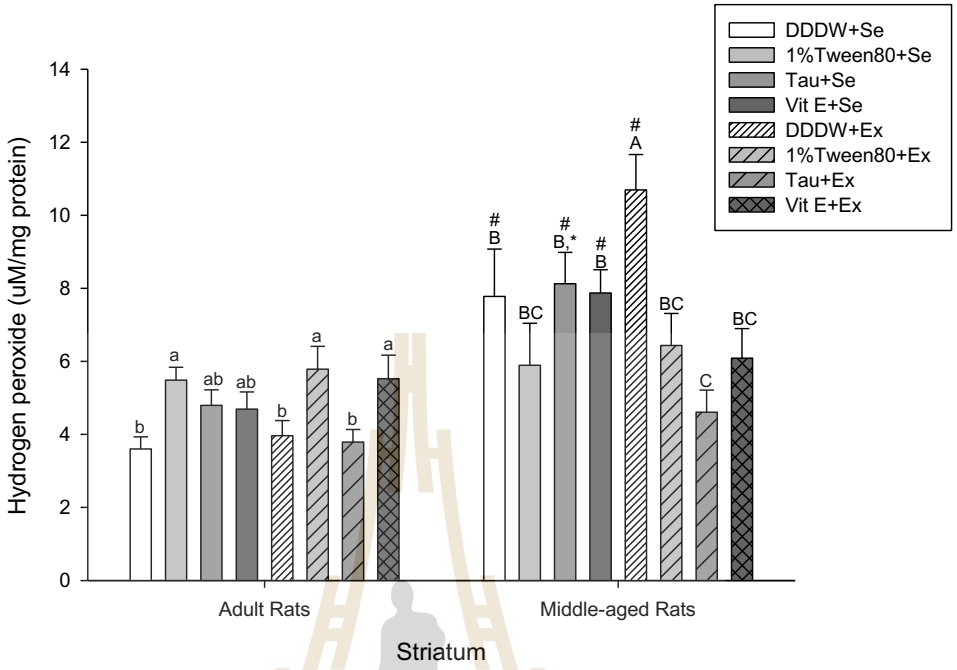

Figure 34 Effects of taurine and vitamin E supplement in conjunction with exercise on H_2O_2 levels in striatum of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

4.1.3.3 Superoxide Dismutase (SOD) Levels

- Basal Forebrain of Adult Rats

The SOD levels in basal forebrain of adult rats in the sedentary group were as follows: DDDW+Se group, 0.252±0.04 mg/mg protein; 1%Tween80+Se group, 0.263±0.04 mg/mg protein; Vit E+Se group, 0.276±0.06 mg/mg protein; and Tau+Se group, 0.344±0.06 mg/mg protein. The SOD levels in basal forebrain of adult rats in the exercise group were as follows: DDDW+Ex group, 0.255±0.05 mg/mg protein; 1%Tween80+Ex group, 0.374±0.08 mg/mg protein; Vit E+Ex group, 0.317±0.05 mg/mg protein; and Tau+Ex group, 0.436±0.05 mg/mg protein (Figure 35). The results showed that the SOD levels in basal forebrain following taurine supplement the exercise group was significantly higher than vitamin E treatment, 1% Tween 80 treatment, and DDD water treatment in the sedentary group ($P<0.05$), and DDD water treatment in the exercise group ($P<0.05$). When compared between the sedentary group and the exercise group, there was no significant difference in H_2O_2 levels in hippocampus between all treatment groups.

- Basal Forebrain of Middle-aged Rats

The SOD levels in basal forebrain of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.146±0.01 mg/mg protein; 1%Tween80+Se group, 0.153±0.02 mg/mg protein; Vit E+Se group, 0.176±0.03 mg/mg protein; and Tau+Se group, 0.218±0.02 mg/mg protein. The SOD levels in basal forebrain of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.177±0.04 mg/mg protein; 1%Tween80+Ex group, 0.162±0.01 mg/mg protein; Vit E+Ex group, 0.268±0.06 mg/mg protein; and Tau+Ex group, 0.262±0.04 mg/mg protein (Figure 35).

The SOD levels in basal forebrain of middle-aged rats following taurine supplement and vitamin E treatment in the exercise group were significantly higher than both the sedentary and the exercise group following 1% Tween 80 treatment, DDD water treatment and vitamin E treatment in the sedentary group ($P<0.05$). When compared between the sedentary group and exercise group, there was no significant difference in SOD levels in basal forebrain of middle-aged rats in all treatment groups.

- The Comparison between Adult and Middle-aged Rats in Basal Forebrain of Superoxide Dismutase (SOD) Levels

The result of SOD levels of basal forebrain when comparison between adult rats and middle-aged rats (Figure 35) found that the adult rats in the sedentary group following DDD water treatment and 1% Tween 80 treatment and in the exercise in middle-aged rats following 1% Tween 80 treatment and taurine treatment were significantly higher than same treatment in the middle-aged rats ($P<0.05$).

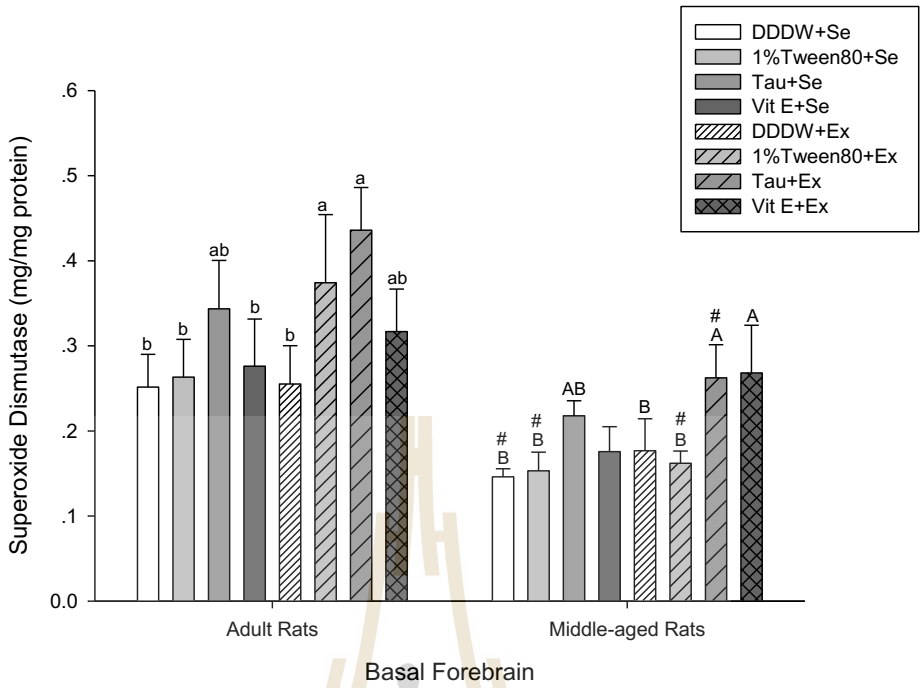

Figure 35 Effects of taurine and vitamin E supplement in conjunction with exercise on SOD levels in basal forebrain of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Cerebral Cortex of Adult Rats

The SOD levels in cerebral cortex of adult rats in the sedentary group were as follows: DDDW+Se group, 0.074±0.004 mg/mg protein; 1%Tween80+Se group, 0.083±0.005 mg/mg protein; Vit E+Se group, 0.105±0.005 mg/mg protein; and Tau+Se group, 0.091±0.007 mg/mg protein. The SOD levels in cerebral cortex of adult rats in the exercise group were as follows: DDDW+Ex group, 0.078±0.005 mg/mg protein; 1%Tween80+Ex group, 0.099±0.004 mg/mg protein; Vit E+Ex group, 0.114±0.010

mg/mg protein; and Tau+Ex group, 0.089±0.010 mg/mg protein (Figure 36). The results showed that the SOD levels in cerebral cortex showed that there was no significant difference between all treatment groups both the sedentary group and exercise group. When compared between the sedentary group and the exercise group, the SOD levels in cerebral cortex of adult rats in 1% Tween 80 treated group in the exercise group was significantly higher than its respective control (P<0.05).

- Cerebral Cortex of Middle-aged Rats

The SOD levels in cerebral cortex of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.128±0.02 mg/mg protein; 1%Tween80+Se group, 0.122±0.01 mg/mg protein; Vit E+Se group, 0.138±0.02 mg/mg protein; and Tau+Se group, 0.170±0.02 mg/mg protein. The SOD levels in cerebral cortex of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.134±0.01 mg/mg protein; 1%Tween80+Ex group, 0.128±0.01 mg/mg protein; Vit E+Ex group, 0.139±0.02 mg/mg protein; and Tau+Ex group, 0.198±0.02 mg/mg protein (Figure 36). The SOD levels in cerebral cortex of middle-aged rats following taurine supplement and vitamin E treatment in the exercise group were significantly higher than both the sedentary and the exercise group following 1% Tween 80 treatment, DDD water treatment and vitamin E treatment in the sedentary group (P<0.05). When compared between the sedentary group and exercise group, there was no significant difference in SOD levels in cerebral cortex of middle-aged rats in all treatment groups.

- The Comparison between Adult and Middle-aged Rats in Cerebral Cortex of Superoxide Dismutase (SOD) Levels

The result of SOD levels of cerebral cortex when comparison between adult rats and middle-aged rats (Figure 36) found that the adult rats in both the sedentary and exercise groups following DDD water treatment, 1% Tween 80 treatment, and taurine supplement were significantly higher than same treatment in the middle-aged rats ($P<0.05$).

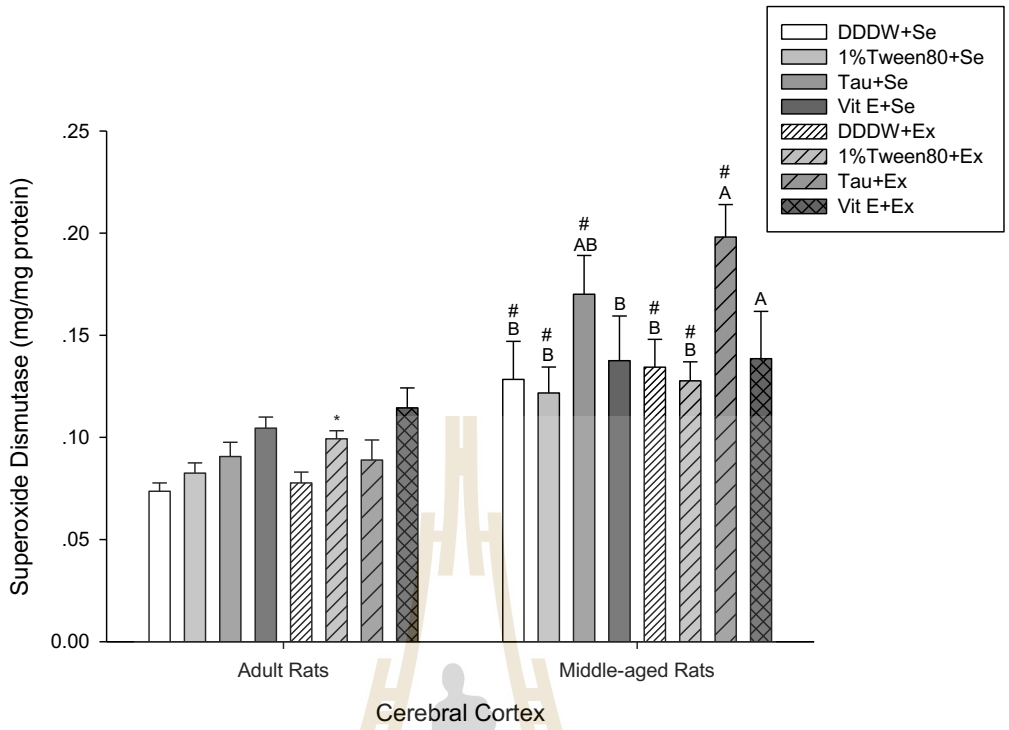

Figure 36 Effects of taurine and vitamin E supplement in conjunction with exercise on SOD levels in cerebral cortex of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Hippocampus of Adult Rats

The SOD levels in hippocampus of adult rats in the sedentary group were as follows: DDDW+Se group, 0.138±0.01 mg/mg protein; 1%Tween80+Se group, 0.129±0.01 mg/mg protein; Vit E+Se group, 0.137±0.02 mg/mg protein; and Tau+Se group, 0.149±0.03 mg/mg protein. The SOD levels in hippocampus of adult rars in the

exercise group were as follows: DDDW+Ex group, 0.146±0.01 mg/mg protein; 1%Tween80+Ex group, 0.147±0.02 mg/mg protein; Vit E+Ex group, 0.147±0.04 mg/mg protein; and Tau+Ex group, 0.186±0.03 mg/mg protein (Figure 37). There was no significant difference in the levels of SOD in hippocampus of adult rats between all groups in both sedentary and exercise groups. When compared between the sedentary group and exercise group, there was no significant difference between all treatment groups.

- Hippocampus of Middle-aged Rats

The SOD levels in hippocampus of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.141±0.01 mg/mg protein; 1% Tween 80 +Se group, 0.149±0.02 mg/mg protein; Vit E+Se group, 0.128±0.01 mg/mg protein; and Tau+Se group, 0.126±0.01 mg/mg protein. The SOD levels in hippocampus of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.152±0.01 mg/mg protein; 1%Tween80+Ex group, 0.155±0.01 mg/mg protein; Vit E+Ex group, 0.140±0.02 mg/mg protein; and Tau+Ex group, 0.148±0.02 mg/mg protein (Figure 37). The results showed that the SOD levels in hippocampus of middle-aged rats were not significantly different between all treatment groups in both the sedentary group and the exercise group. When compared between the sedentary group and exercise group, there was no significant difference in SOD levels in hippocampus of middle-aged rats in all treatment groups.

- The Comparison between Adult and Middle-aged Rats in Hippocampus of Superoxide Dismutase (SOD) Levels

The result of SOD levels of hippocampus when comparison between adult rats and middle-aged rats (Figure 37) found that there was no significant difference in all treatment group both the sedentary and exercise groups.

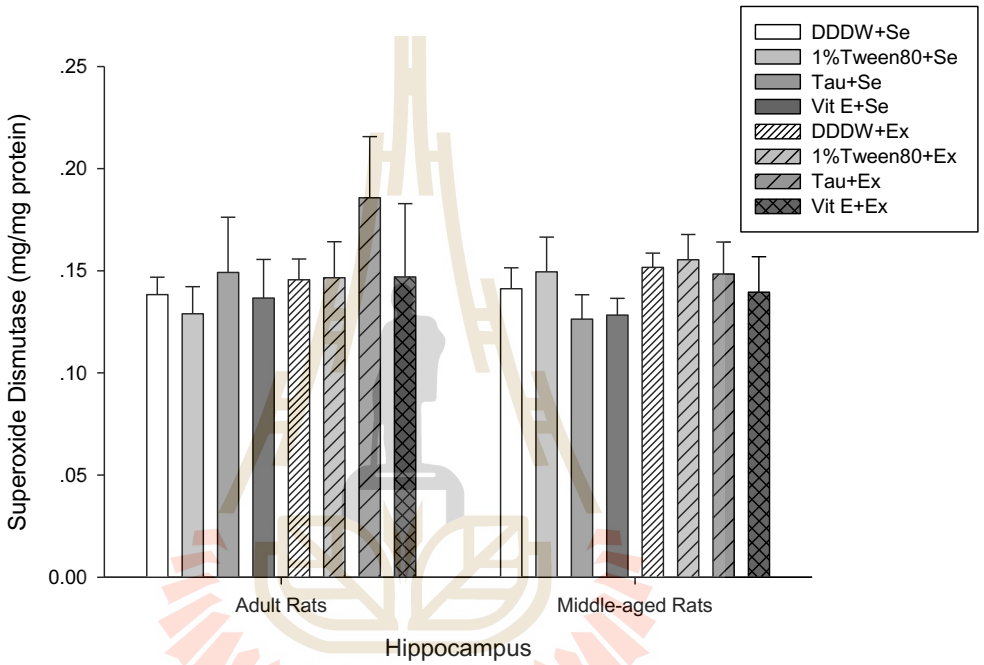

Figure 37 Effects of taurine and vitamin E supplement in conjunction with exercise on SOD levels in hippocampus of adult and middle-aged rats. Values are expressed as mean ± SEM.

- Striatum of Adult Rats

The SOD levels in striatum of adult rats in the sedentary group were as follows: DDDW+Se group, 0.206±0.02 mg/mg protein; 1%Tween80+Se group, 0.296±0.04 mg/mg protein; Vit E+Se group, 0.340±0.02 mg/mg protein; and Tau+Se group, 0.286±0.02 mg/mg protein. The SOD levels in striatum of adult rats in the exercise

group were as follows: DDDW+Ex group, 0.216±0.03 mg/mg protein; 1%Tween80+Ex group, 0.285±0.02 mg/mg protein; Vit E+Ex group, 0.326±0.04 mg/mg protein; and Tau+Ex group, 0.371±0.03 mg/mg protein (Figure 38). The SOD levels in striatum of adult rats following taurine supplement in the exercise group was significantly higher than taurine treatment and DDD water treatment in the sedentary group and 1% Tween 80 treatment and DDD water treatment in the exercise group (P<0.05), both the sedentary and the exercise group following vitamin E treatment were significantly higher than both the sedentary and the exercise group of DDD water treatment (P<0.05). The SOD levels in striatum of adult rats in taurine supplement group was significantly higher than exercise alone (P<0.05). When compared between the sedentary group and the exercise group, the SOD levels in striatum of adult rats in taurine supplement in the exercise group was significantly higher than exercise alone (P<0.05).

- Striatum of Middle-aged Rats

The SOD levels in striatum of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.130±0.03 mg/mg protein; 1%Tween80+Se group, 0.109±0.01 mg/mg protein; Vit E+Se group, 0.126±0.01 mg/mg protein; and Tau+Se group, 0.160±0.02 mg/mg protein. The SOD levels in striatum of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.136±0.02 mg/mg protein; 1%Tween80+Ex group, 0.144±0.01 mg/mg protein; Vit E+Ex group, 0.143±0.04 mg/mg protein; and Tau+Ex group, 0.233±0.03 mg/mg protein (Figure 38). The results showed that SOD levels in striatum of middle-aged rats following taurine supplement in the exercise group was significantly higher than all treatment groups in both the

sedentary and the exercise groups (P<0.05). In comparison between sedentary group and exercise group, exercise significantly increased in the SOD levels in striatum of middle-aged rats following 1% Tween 80 treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats in Striatum of Superoxide Dismutase (SOD) Levels

The result of SOD levels of striatum when comparison between adult rats and middle-aged rats (Figure 38) found that the adult rats in all treatment both the sedentary groups and the exercise groups were significantly higher than same treatment in the middle-aged rats (P<0.05).

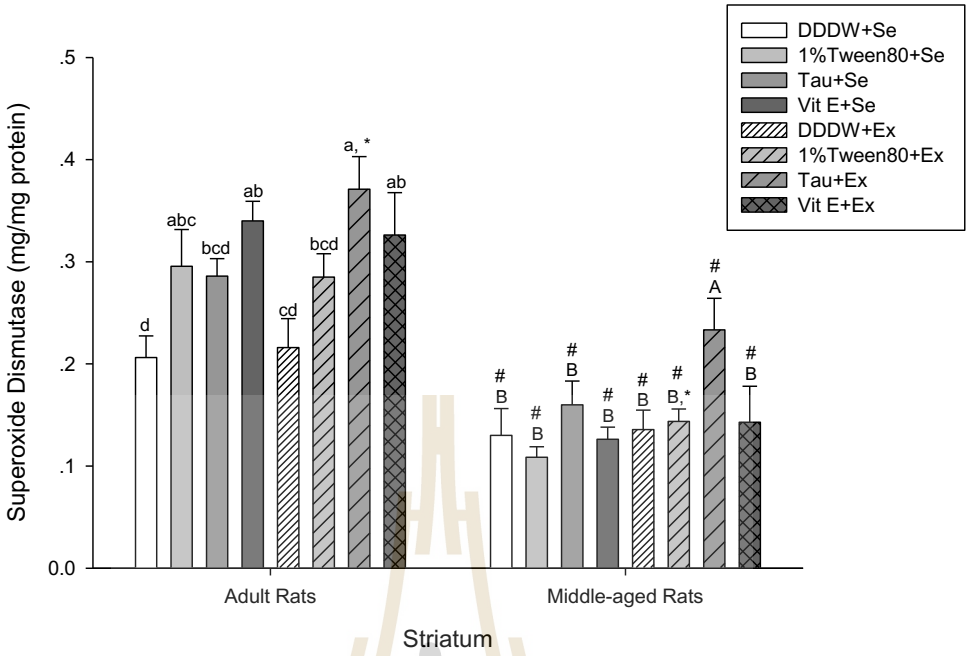

Figure 38 Effects of taurine and vitamin E supplement in conjunction with exercise on SOD levels in striatum of adult rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

4.1.3.4 The Antioxidant Activity of Catalase (CAT) Levels

- Basal Forebrain of Adult Rats

The CAT levels in basal forebrain of adult rats in the sedentary group were as follows: DDDW+Se group, 0.060±0.02 unit/mg protein; 1%Tween80+Se group, 0.070±0.01 unit/mg protein; Vit E+Se group, 0.059±0.01 unit/mg protein; and Tau+Se group, 0.094±0.02 unit/mg protein. The CAT levels in basal forebrain of adult rats in

the exercise group were as follows: DDDW+Ex group, 0.066±0.02 unit/mg protein; 1%Tween80+Ex group, 0.057±0.01 unit/mg protein; Vit E+Ex group, 0.122±0.03 unit/mg protein; and Tau+Ex group, 0.104±0.02 unit/mg protein (Figure 39). The results showed that the CAT levels in basal forebrain in adult rats following vitamin E supplement in the exercise group was significantly higher than 1% Tween 80 treatment, vitamin E treatment, and DDD water treatment in the sedentary group, and DDD water treatment and 1% Tween 80 treatment in the exercise group (P<0.05). In comparison between the sedentary group and the exercise group, there was no significant difference between all treatment groups.

- Basal Forebrain of Middle-aged Rats

The CAT levels in basal forebrain of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.029±0.005 unit/mg protein; 1%Tween80+Se group, 0.026±0.004 unit/mg protein; Vit E+Se group, 0.024±0.004 unit/mg protein; and Tau+Se group, 0.038±0.009 unit/mg protein. The CAT levels in basal forebrain of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.047±0.016 unit/mg protein; 1%Tween80+Ex group, 0.055±0.010 unit/mg protein; Vit E+Ex group, 0.059±0.015 unit/mg protein; and Tau+Ex group, 0.080±0.016 unit/mg protein (Figure 39). The results showed that in basal forebrain of middle-aged rats following taurine supplement in the exercise group was significantly higher than DDD water treatment, 1% Tween 80 treatment, and vitamin E treatment in the sedentary group (P<0.05). When compared between the sedentary group and exercise group, there was no significant difference in CAT levels in basal forebrain of in all treatment groups middle-aged rats.

- The Comparison between Adult and Middle-aged Rats in Basal Forebrain of Catalase (CAT) Levels

The result of CAT levels of basal forebrain when comparison between adult rats and middle-aged rats (Figure 39) found that there was no significant difference in all treatment groups in the sedentary group. In the exercise groups of the adult rats following taurine supplement was significantly higher than same treatment in the middle-aged rats ($P<0.05$).

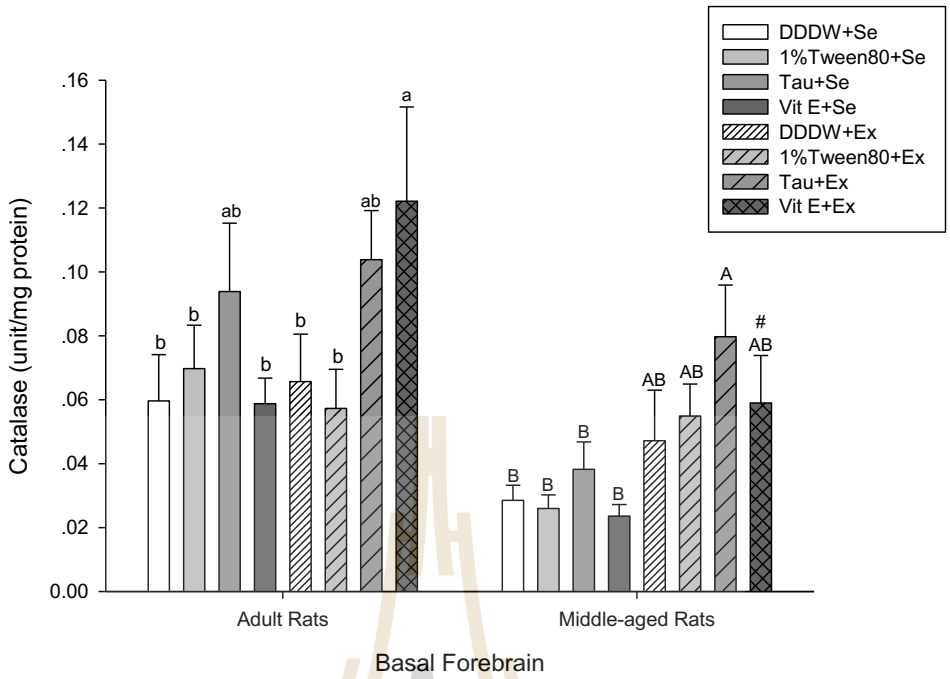

Figure 39 Effects of taurine and vitamin E supplement in conjunction with exercise on CAT levels in basal forebrain of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Cerebral Cortex of Adult Rats

The CAT levels in cerebral cortex of adult rats in the sedentary group were as follows: DDDW+Se group, 0.054±0.01 unit/mg protein; 1%Tween80+Se group, 0.066±0.02 unit/mg protein; Vit E+Se group, 0.079±0.01 unit/mg protein; and Tau+Se group, 0.085±0.01 unit/mg protein. The CAT levels in cerebral cortex of adult rats in

the exercise group were as follows: DDDW+Ex group, 0.057±0.01 unit/mg protein; 1%Tween80+Ex group, 0.079±0.01 unit/mg protein; Vit E+Ex group, 0.107±0.01 unit/mg protein; and Tau+Ex group, 0.113±0.01 unit/mg protein (Figure 40). The results showed that the CAT levels in cerebral cortex following taurine supplement and vitamin E supplement in the exercise group were significantly higher than exercise alone and DDD water treatment and 1% Tween 80 treatment in the exercise group (P<0.05). When compared between the sedentary group and the exercise group, there was no significant difference in the CAT levels in cerebral cortex between all treatment groups.

- Cerebral Cortex of Middle-aged Rats

The CAT levels in cerebral cortex of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.043±0.01 unit/mg protein; 1%Tween80+Se group, 0.093±0.02 unit/mg protein; Vit E+Se group, 0.049±0.01 unit/mg protein; and Tau+Se group, 0.055±0.01 unit/mg protein. The CAT levels in cerebral cortex of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.086±0.01 unit/mg protein; 1%Tween80+Ex group, 0.085±0.03 unit/mg protein; Vit E+Ex group, 0.056±0.01 unit/mg protein; and Tau+Ex group, 0.071±0.01 unit/mg protein (Figure 40). The result of CAT level in cerebral cortex in middle-aged rats following 1% Tween 80 treatment in the sedentary group was significantly higher than vitamin E treatment and DDD water treatment in the sedentary group (P<0.05), 1% Tween 80 treatment and DDD water treatment in the exercise group were significantly higher than DDD water treatment in the sedentary group (P<0.05). When compared

between the sedentary group and exercise group, there was no significant difference in CAT levels of cerebral cortex in middle-aged rats in all treatment groups.

- The Comparison between Adult and Middle-aged Rats in Cerebral cortex of Catalase (CAT) Levels

The result of CAT levels of basal forebrain when comparison between adult rats and middle-aged rats (Figure 40) found that there was no significant difference in all treatment groups in the sedentary group. In the exercise groups of the adult rats following taurine supplement was significantly higher than same treatment in the middle-aged rats ($P<0.05$).

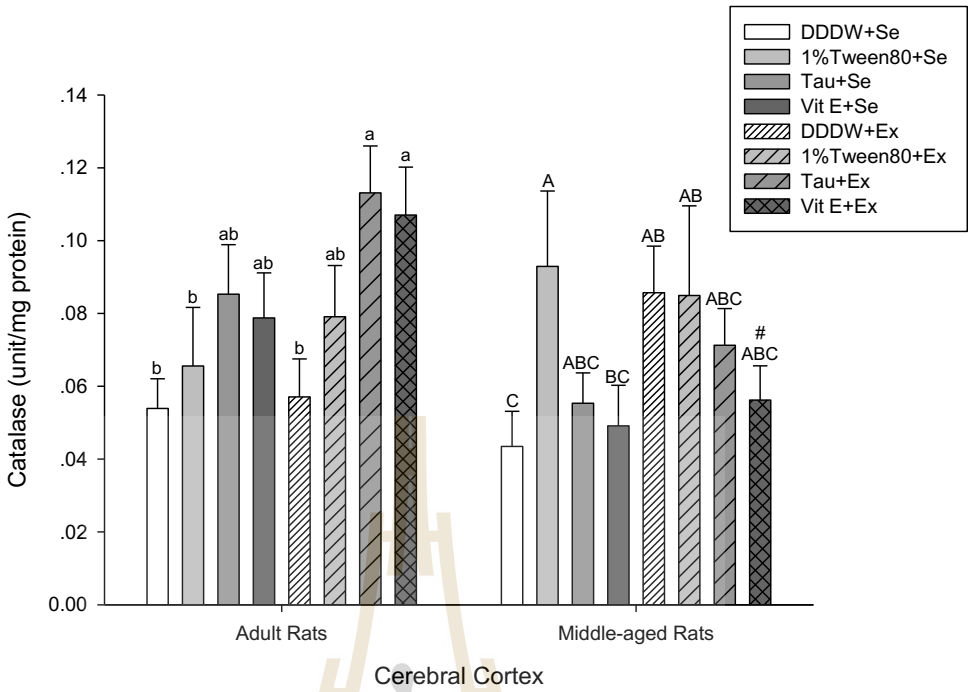

Figure 40 Effects of taurine and vitamin E supplement in conjunction with exercise on CAT levels in cerebral cortex of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Hippocampus of Adult Rats

The CAT levels in hippocampus of adult rats in the sedentary group were as follows: DDDW+Se group, 0.021±0.004 unit/mg protein; 1%Tween80+Se group, 0.037±0.008 unit/mg protein; Vit E+Se group, 0.056±0.009 unit/mg protein; and Tau+Se group, 0.048±0.008 unit/mg protein. The CAT levels in hippocampus of adult rats in the exercise group were as follows: DDDW+Ex group, 0.037±0.008 unit/mg protein; 1%Tween80+Ex group, 0.047±0.009 unit/mg protein; Vit E+Ex group,

0.057±0.009 unit/mg protein; and Tau+Ex group, 0.051±0.010 unit/mg protein (Figure 41). The results showed that the CAT levels in hippocampus following taurine supplement and vitamin E supplement in the sedentary group and vitamin E treatment, taurine treatment, and 1% Tween 80 treatment in the exercise group were significantly higher than DDD water in sedentary group (P<0.05). When compared between the sedentary group and the exercise group, there was no significant difference in the CAT levels in hippocampus between all treatment groups.

- Hippocampus of Middle-aged Rats

The CAT levels in hippocampus of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.010±0.003 unit/mg protein; 1%Tween80+Se group, 0.016±0.007 unit/mg protein; Vit E+Se group, 0.025±0.006 unit/mg protein; and Tau+Se group, 0.013±0.004 unit/mg protein. The CAT levels in hippocampus of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.013±0.004 unit/mg protein; 1%Tween80+Ex group, 0.013±0.004 unit/mg protein; Vit E+Ex group, 0.035±0.008 unit/mg protein; and Tau+Ex group, 0.031±0.006 unit/mg protein (Figure 41). The CAT level in hippocampus of middle-aged rats following vitamin E supplement and taurine supplement in the exercise group were significantly higher than DDD water treatment, 1% Tween 80 treatment, and taurine treatment in the sedentary group (P<0.05), and 1% Tween 80 treatment and DDD water treatment in the exercise group (P<0.05). In comparison between the sedentary group and exercise group, exercise significantly increased the CAT levels in taurine supplement (P<0.05).

- The Comparison between Adult and Middle-aged Rats in Hippocampus of Catalase (CAT) Levels

The result of CAT levels of hippocampus when comparison between adult rats and middle-aged rats (Figure 41) found that the adult rats in the sedentary group following DDD water treatment, vitamin E treatment and taurine treatment and in the exercise in middle-aged rats following DDD water treatment and 1% Tween 80 treatment were significantly higher than same treatment in the middle-aged rats (P<0.05).

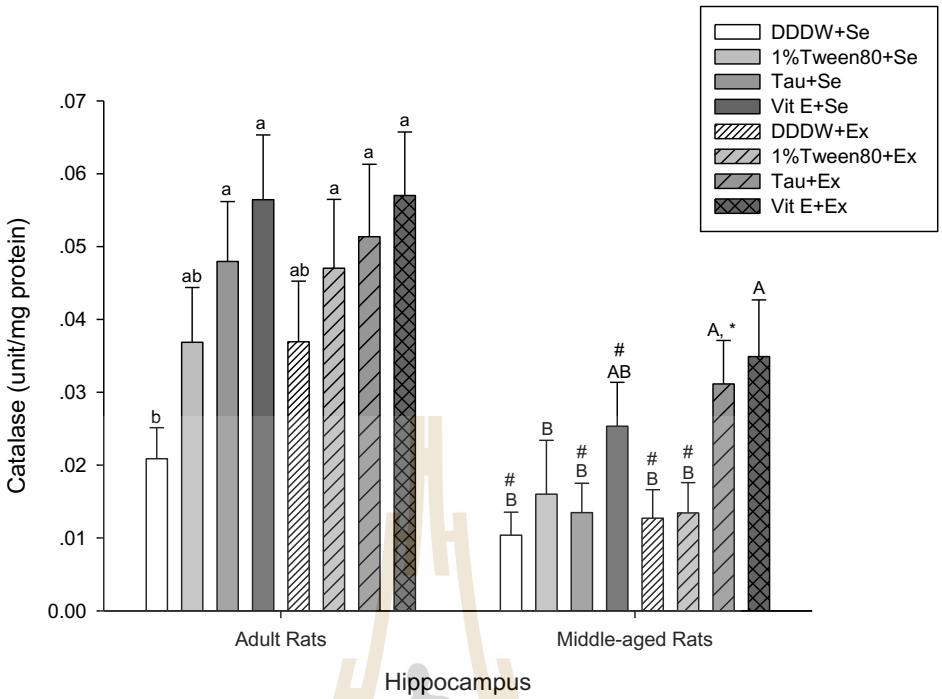

Figure 41 Effects of taurine and vitamin E supplement in conjunction with exercise on CAT levels in hippocampus of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Striatum of Adult Rats

The CAT levels in striatum of adult rats in the sedentary group were as follows: DDDW+Se group, 0.049±0.01 unit/mg protein; 1%Tween80+Se group, 0.088±0.02 unit/mg protein; Vit E+Se group, 0.093±0.01 unit/mg protein; and Tau+Se group, 0.098±0.02 unit/mg protein. The CAT levels in striatum of adult rats in the exercise

group were as follows: DDDW+Ex group, 0.038±0.01 unit/mg protein; 1%Tween80+Ex group, 0.077±0.02 unit/mg protein; Vit E+Ex group, 0.114±0.02 unit/mg protein; and Tau+Ex group, 0.110±0.02 unit/mg protein (Figure 42). In the CAT level is striatum following taurine supplement and vitamin E treatment in the exercise groups and taurine supplement in the sedentary group were significantly higher than both the sedentary and exercise groups following DDD water treatment (P<0.05), vitamin E treatment and 1% Tween 80 treatment in the sedentary group were significantly higher than DDD water treatment in the exercise group (P<0.05). When compared between the sedentary group and the exercise group, there was no significant difference in the CAT levels in striatum between all treatment groups.

- Striatum of Middle-aged Rats

The CAT levels in striatum of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.024±0.01 unit/mg protein; 1%Tween80+Se group, 0.031±0.01 unit/mg protein; Vit E+Se group, 0.055±0.02 unit/mg protein; and Tau+Se group, 0.038±0.01 unit/mg protein. The CAT levels in striatum of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.052±0.01 unit/mg protein; 1%Tween80+Ex group, 0.028±0.01 unit/mg protein; Vit E+Ex group, 0.058±0.01 unit/mg protein; and Tau+Ex group, 0.101±0.03 unit/mg protein (Figure 42). The results showed that in the CAT levels in striatum of middle-aged rats taurine treatment in the exercise group was significantly higher than all treatment groups of both the sedentary and exercise groups (P<0.05). In comparison between the sedentary group and the exercise group, exercise significantly increased the CAT levels in striatum following taurine treatments and DDD water treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats in Striatum of Catalase (CAT) Levels

The result of CAT levels of striatum when comparison between adult rats and middle-aged rats (Figure 42) found that the adult rats in the sedentary group following 1% Tween 80 treatment, vitamin E treatment and taurine treatment and in the exercise in adult rats following 1% Tween 80 treatment and vitamin E treatment were significantly higher than same treatment in the middle-aged rats ($P<0.05$).

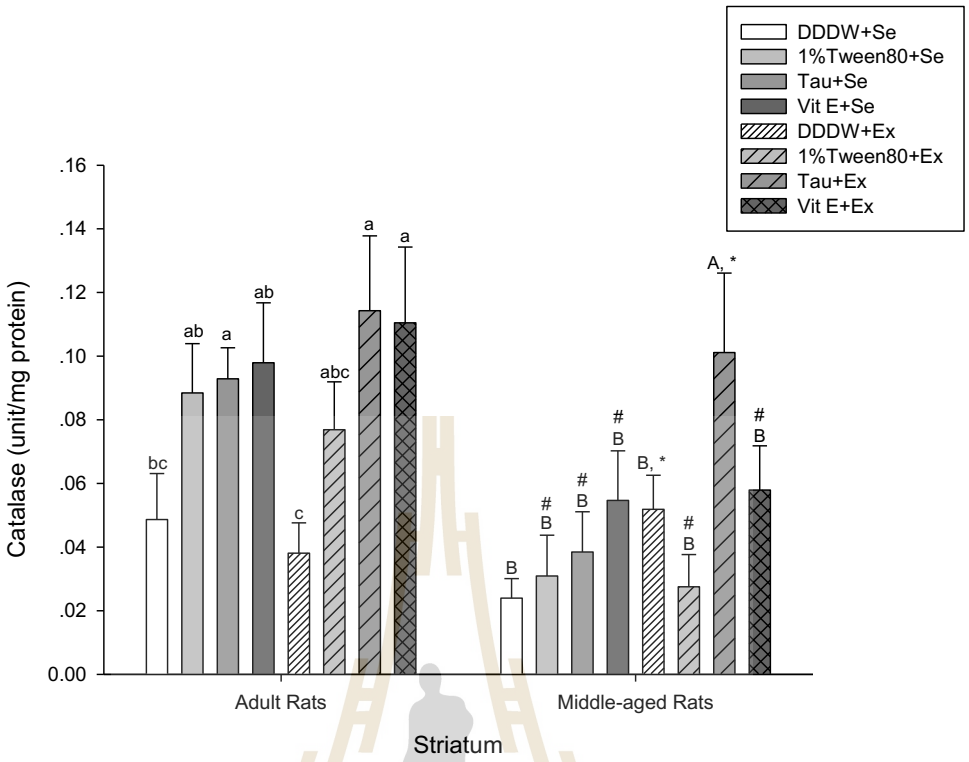

Figure 42 Effects of taurine and vitamin E supplement in conjunction with exercise on CAT levels in striatum of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

4.1.3.5 The Antioxidant Activity of Glutathione Peroxidase (GPx)

- Basal Forebrain of Adult Rats

The GPX enzyme activity levels in basal forebrain of adult rats in the sedentary group were as follows: DDDW+Se group, 0.0043±0.0004 mmol/min/ml/mg protein; 1%Tween80+Se group, 0.0081±0.0010 mmol/min/ml/mg protein; Vit E+Se group, 0.0071±0.0011 mmol/min/ml/mg protein; and Tau+Se group, 0.0093±0.0012 mmol/min/ml/mg protein. The GPx enzyme activity levels in basal forebrain of adult rats in the exercise group were as follows: DDDW+Ex group, 0.0059±0.0006 mmol/min/ml/mg protein; 1%Tween80+Ex group, 0.0064±0.0012 mmol/min/ml/mg protein; Vit E+Ex group, 0.0092±0.0024 mmol/min/ml/mg protein; and Tau+Ex group, 0.0073±0.0005 mmol/min/ml/mg protein (Figure 43). The GPx enzyme activity levels in basal forebrain of adult rats following taurine supplement alone and vitamin E supplement in the erercise group were significantly higher than DDD water treatment in the sedentary and the exercise group (P<0.05), 1% Tween 80 treatment in the sedentary group was signifivcantly higher than control group (P<0.05). In comparison between the sedentary ground and experiment group, GPx enzyme activity levels following DDD water treatment in exercise group was significantly higher than DDD water treatment in sedentary group.

- Basal Forebrain of Middle-aged Rats

The GPx enzyme activity levels in basal forebrain of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.0046±0.001 mmol/min/ml/mg protein; 1%Tween80+Se group, 0.0070±0.001 mmol/min/ml/mg protein; Vit E+Se group, 0.0078±0.0004 mmol/min/ml/mg protein; and Tau+Se group, 0.0100±0.001

mmol/min/ml/mg protein. The GPx enzyme activity levels in basal forebrain of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.0053±0.001 mmol/min/ml/mg protein; 1%Tween80+Ex group, 0.0087±0.001 mmol/min/ml/mg protein; Vit E+Ex group, 0.0107±0.002 mmol/min/ml/mg protein; and Tau+Ex group, 0.0105±0.002 mmol/min/ml/mg protein (Figure 43). The results showed that the GPx enzyme activity levels in basal forebrain of middle-aged rats following vitamin E supplement in the exercise group was significantly higher than the DDD water treatment and 1% Tween 80 treatment in the sedentary groups and exercise alone (P<0.05), taurine supplement in both the sedentary and exercise group were significantly higher than control group and exercise alone group (P<0.05), 1% Tween 80 treatment in the exercise group was significantly higher than control group (P<0.05). In comparison between the sedentary group and the exercise group, exercise significantly increased the GPx enzyme activity levels in basal forebrain of middle-aged rats in vitamin E treatment and 1% Tween 80 treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats in Basal Forebrain of Glutathione Peroxidase (GPx) Levels

The result of GPx levels of basal forebrain when comparison between adult rats and middle-aged rats (Figure 43) found that there was no significant difference in all treatment group both the sedentary and exercise groups.

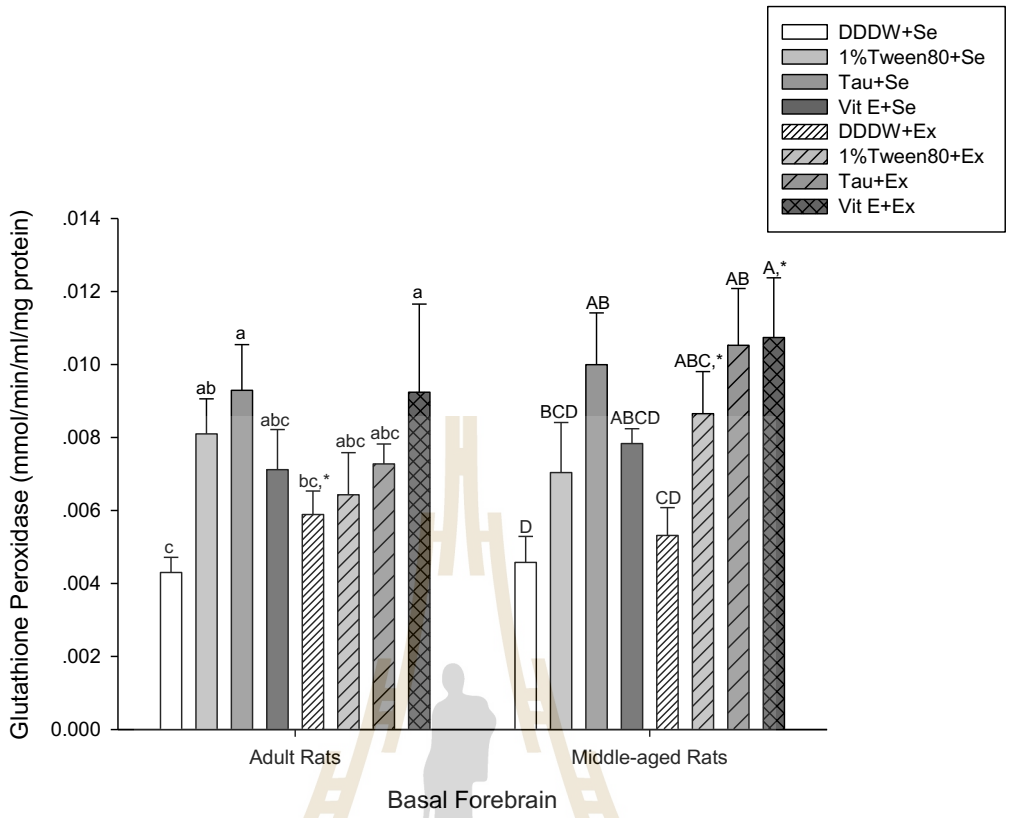

Figure 43 Effects of taurine and vitamin E supplement in conjunction with exercise on GPx enzyme activity levels in basal forebrain of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age.

- Cerebral Cortex of Adult Rats

The GPx enzyme activity levels in cerebral cortex of adult rats in the sedentary group were as follows: DDDW+Se group, 0.0067±0.001 mmol/min/ml/mg protein; 1%Tween80+Se group, 0.0074±0.001 mmol/min/ml/mg protein; Vit E+Se group, 0.0104±0.001 mmol/min/ml/mg protein; and Tau+Se group, 0.0098±0.002

mmol/min/ml/mg protein. The GPx enzyme activity levels in cerebral cortex of adult rats in the exercise group were as follows: DDDW+Ex group, 0.0076±0.001 mmol/min/ml/mg protein; 1%Tween80+Ex group, 0.0110±0.001 mmol/min/ml/mg protein; Vit E+Ex group, 0.0129±0.002 mmol/min/ml/mg protein; and Tau+Ex group, 0.0108±0.001 mmol/min/ml/mg protein (Figure 44). The GPx enzyme activity levels in cerebral cortex following vitamin E supplement in the exercise group was significantly higher than the DDD water treatment group in both the sedentary and exercise group and 1% Tween 80 treatment in the sedentary group (P<0.05), 1% Tween 80 treatment in the exercise group was significantly higher than DDD water treatment and 1% Tween 80 treatment in the sedentary group (P<0.05), taurine supplement in the exercise group and vitamin E supplement in the sedentary group were significantly higher than DDD water treatment in the sedentary group (P<0.05). When compared between the sedentary group and the exercise group, the GPx enzyme activity levels in cerebral cortex following 1% Tween 80 treatment in exercise group was significantly higher than 1% Tween 80 treatment in sedentary group (P<0.05).

- Cerebral Cortex of Middle-aged Rats

The GPx enzyme activity levels in cerebral cortex of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.0052±0.001 mmol/min/ml/mg protein; 1%Tween80+Se group, 0.0060±0.001 mmol/min/ml/mg protein; Vit E+Se group, 0.0069±0.001 mmol/min/ml/mg protein; and Tau+Se group, 0.0075±0.002 mmol/min/ml/mg protein. The GPx enzyme activity levels in cerebral cortex of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.0068±0.002 mmol/min/ml/mg protein; 1%Tween80+Ex group, 0.0063±0.002 mmol/min/ml/mg

protein; Vit E+Ex group, 0.0089±0.003 mmol/min/ml/mg protein; and Tau+Ex group, 0.0100±0.002 mmol/min/ml/mg protein (Figure 44). The results showed that there was no significant difference in the GPx enzyme activity levels in cerebral cortex of middle-aged rats in all treatment groups of both the sedentary group and the exercise group. When compared between the sedentary group and exercise group, there was no significant difference in the GPx enzyme activity levels in cerebral cortex of middle-aged rats in all treatment groups.

- The Comparison between Adult and Middle-aged Rats in Cerebral Cortex of Glutathione Peroxidase (GPx) Levels

The result of GPx levels of cerebral cortex when comparison between adult rats and middle-aged rats (Figure 44) found that the adult rats in the sedentary group following vitamin E treatment and in the exercise in adult rats following 1% Tween 80 treatment were significantly higher than same treatment in the middle-aged rats ($P<0.05$).

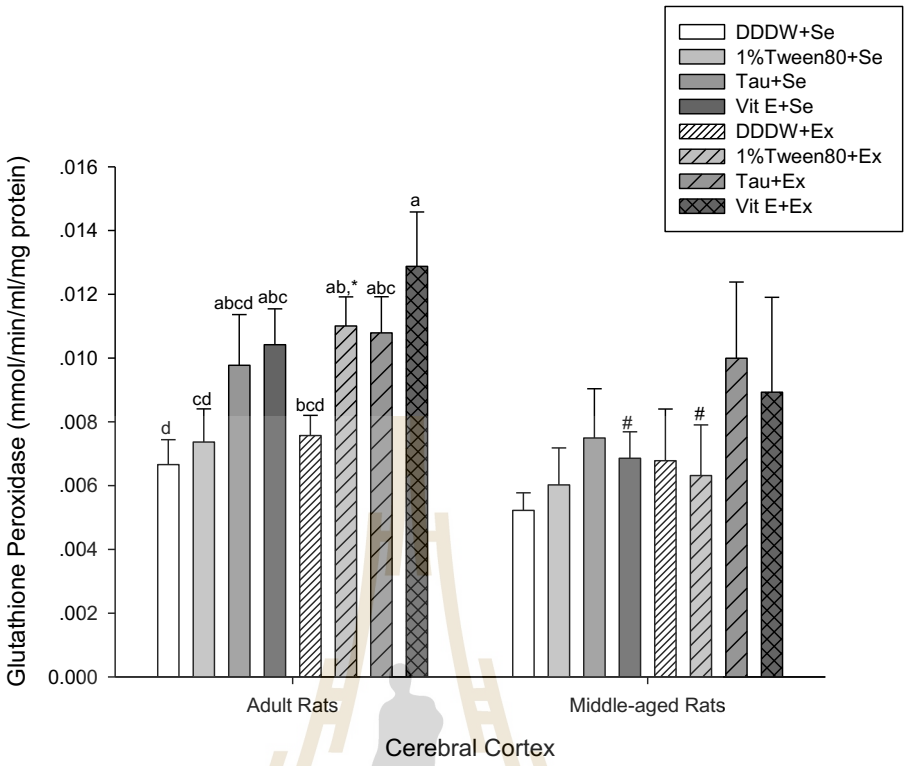

Figure 44 Effects of taurine and vitamin E supplement in conjunction with exercise on GPx enzyme activity levels in cerebral cortex of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Hippocampus of Adult Rats

The GPx enzyme activity levels in hippocampus of adult rats in the sedentary group were as follows: DDDW+Se group, 0.0046±0.001 mmol/min/ml/mg protein; 1%Tween80+Se group, 0.0065±0.001 mmol/min/ml/mg protein; Vit E+Se group, 0.0071±0.001 mmol/min/ml/mg protein; and Tau+Se group, 0.0059±0.001

mmol/min/ml/mg protein. The GPx enzyme activity levels in hippocampus in the exercise group were as follows: DDDW+Ex group, 0.0060±0.001 mmol/min/ml/mg protein; 1%Tween80+Ex group, 0.0051±0.001 mmol/min/ml/mg protein; Vit E+Ex group, 0.0079±0.001 mmol/min/ml/mg protein; and Tau+Ex group, 0.0070±0.001 mmol/min/ml/mg protein (Figure 45). The GPx enzyme activity levels in hippocampus of adult rats following vitamin E supplement in the exercise group was significantly higher than DDD water treatment in the sedentary group and 1% Tween 80 treatment in the exercise group ($P<0.05$). In comparison between sedentary group and exercise group, the GPx enzyme activity levels in hippocampus in all treatment groups were not significantly different.

- Hippocampus of Middle-aged Rats

The GPx enzyme activity levels in hippocampus of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.0046±0.001 mmol/min/ml/mg protein; 1%Tween80+Se group, 0.0069±0.001 mmol/min/ml/mg protein; Vit E+Se group, 0.0060±0.001 mmol/min/ml/mg protein; and Tau+Se group, 0.0064±0.001 mmol/min/ml/mg protein. The GPx enzyme activity levels in hippocampus of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.0057±0.001 mmol/min/ml/mg protein; 1%Tween80+Ex group, 0.0075±0.001 mmol/min/ml/mg protein; Vit E+Ex group, 0.0073±0.001 mmol/min/ml/mg protein; and Tau+Ex group, 0.0100±0.001 mmol/min/ml/mg protein (Figure 45). The GPx enzyme activity levels in hippocampus of middle-aged rats following taurine supplement in the exercise group was significantly higher than all treatment groups of both the sedentary and the exercise groups ($P<0.05$), vitamin E treatment and 1% Tween 80 treatment in the exercise

groups were significantly higher than control group (P<0.05). In comparison between the sedentary group and the exercise group, exercise significantly increased the GPx enzyme activity levels in hippocampus of middle-aged rats in taurine treatment (P<0.05).

- The Comparison between Adult and Middle-aged Rats in Hippocampus of Glutathione Peroxidase (GPx) Levels

The result of GPx levels of cerebral cortex when comparison between adult rats and middle-aged rats (Figure 45) found that there was no significant difference in all treatment groups in the sedentary groups. In the exercise groups in middle-aged rats following taurine supplement was significantly higher than same treatment in the adult rats (P<0.05).

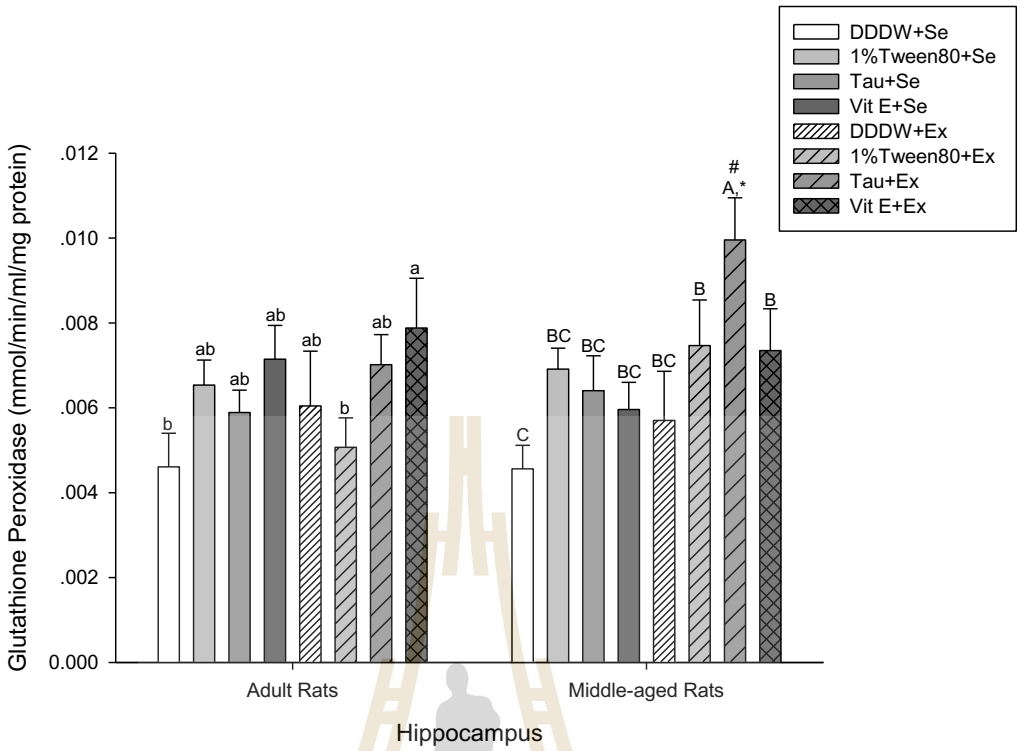

Figure 45 Effects of taurine and vitamin E supplement in conjunction with exercise GPx enzyme activity levels in hippocampus of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case) or middle-aged group (upper case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

- Striatum of Adult Rats

The GPx enzyme activity levels in striatum of adult rats in the sedentary group were as follows: DDDW+Se group, 0.0126±0.002 mmol/min/ml/mg protein; 1%Tween80+Se group, 0.0154±0.002 mmol/min/ml/mg protein; Vit E+Se group,

0.0212±0.003 mmol/min/ml/mg protein; and Tau+Se group, 0.0185±0.003 mmol/min/ml/mg protein. The GPx enzyme activity levels in striatum in the exercise group were as follows: DDDW+Ex group, 0.0090±0.001 mmol/min/ml/mg protein; 1%Tween80+Ex group, 0.0190±0.002 mmol/min/ml/mg protein; Vit E+Ex group, 0.0250±0.004 mmol/min/ml/mg protein; and Tau+Ex group, 0.0214±0.003 mmol/min/ml/mg protein (Figure 46). The results showed that the GPx enzyme activity levels in the striatum following vitamin E supplement in the exercise group was significantly higher than the DDD water treatment and 1% Tween 80 treatment in the sedentary and exercise alone (P<0.05), taurine treatment in the exercise group and vitamin E treatment in the sedentary group were significantly higher than control and exercise alone groups (P<0.05), taurine treatment in the sedentary group and 1% Tween 80 treatment in the exercise group were significant higher than exercise alone (P<0.05). In comparison between sedentary group and exercise group, the GPx enzyme activity levels in striatum in all treatment groups were not significantly different.

- Striatum of Middle-aged Rats

The GPx enzyme activity levels in striatum of middle-aged rats in the sedentary groups were as follows: DDDW+Se group, 0.0055±0.001 mmol/min/ml/mg protein; 1%Tween80+Se group, 0.0074±0.001 mmol/min/ml/mg protein; Vit E+Se group, 0.0061±0.001 mmol/min/ml/mg protein; and Tau+Se group, 0.0072±0.002 mmol/min/ml/mg protein. The GPx enzyme activity levels in striatum of middle-aged rats in the exercise groups were as follows: DDDW+Ex group, 0.0062±0.001 mmol/min/ml/mg protein; 1%Tween80+Ex group, 0.0068±0.001 mmol/min/ml/mg protein; Vit E+Ex group, 0.0093±0.001 mmol/min/ml/mg protein; and Tau+Ex group,

0.0090±0.001 mmol/min/ml/mg protein (Figure 46). The results showed that the GPx enzyme activity levels in striatum of middle-aged rat was not significantly different between all treatment groups of both the sedentary group and the exercise. When compared between the sedentary group and exercise group, exercise significantly increased the GPx enzyme activity levels in striatum of middle-aged rats in vitamin E treatment ($P<0.05$).

- The Comparison between Adult and Middle-aged Rats in Striatum of Glutathione Peroxidase (GPx) Levels

The result of GPx levels of striatum when comparison between adult rats and middle-aged rats (Figure 46) found that the adult rats in all treatment both the sedentary groups and the exercise groups were significantly higher than same treatment in the middle-aged rats ($P<0.05$).

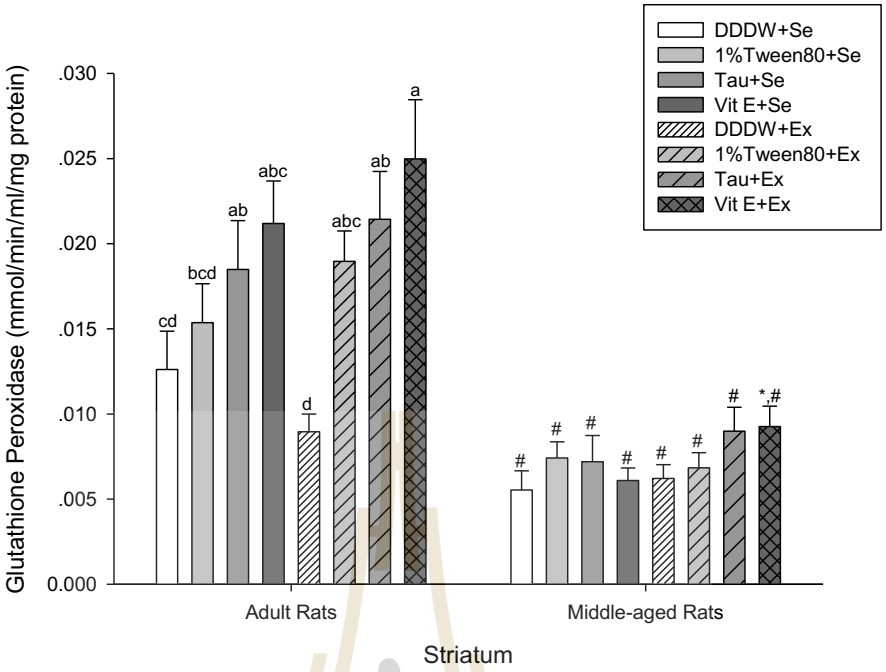

Figure 46 Effects of taurine and vitamin E supplement in conjunction with exercise on GPx enzyme activity levels in striatum of adult and middle-aged rats. Values are expressed as mean ± SEM. Different letters indicate statistically significant differences (P<0.05) among different treatments in adult groups (lower case). Asterisk indicates significant difference (P<0.05) between exercise and sedentary groups received same treatment and same age. Hash sign indicates significant difference (P<0.05) between adult and exercise groups received same treatment and same activity.

4.2 Summary of Findings

The present finding on the effect of taurine supplement in conjunction with exercise on antioxidant enzymes activity, malondialdehyde levels, and hydrogen peroxide levels in adult and middle-rats are summarized in Table 2 and Table 3, respectively.

The present finding on the effect of Vitamin E supplement in conjunction with exercise on antioxidant enzymes activity, malondialdehyde levels, and hydrogen peroxide levels in adult and middle-rats are summarized in Table 4 and Table 5, respectively.

The summary of the effects of Vitamin E supplement on the level of malondialdehyde (MDA), hydrogen peroxide (H_2O_2) in middle-aged rat brains when compared between middle-aged and adult rat brains is shown in Table 6.

The summary of the effects of Vitamin E supplement on the level of superoxide dismutase (SOD), catalase (CAT), and activity of glutathione peroxidase (GPx) when compared between middle-aged and adult rat brains is shown in Table 7.

The present findings on the effect of taurine supplement in conjunction with exercise on relative organ weights of skeletal muscles, liver, kidney, heart, spleen and lung in adult and middle-rats are summarized in Table 8.

The present findings on the effect of Vitamin E supplement in conjunction with exercise on relative organ weights of skeletal muscles, liver, kidney, heart, spleen and lung in adult and middle-rats are summarized in Table 9.

The present findings on the effect of taurine supplement in conjunction with exercise on blood parameters in adult and middle-rats are summarized in Table 10.

The present findings on the effect of Vitamin E supplement in conjunction with exercise on blood parameters in adult and middle-rats are summarized in Table 11.

The summary of the effects of Vitamin E supplement in conjunction with exercise on relative organ weights of skeletal muscles, liver, kidney, heart, spleen and lung in adult and middle-age rats is shown in table 12.

The summary of the effects of Vitamin E supplement in conjunction with exercise on blood parameters in adult and middle-aged rats is shown in table 13.

Table 2 Summary of the effects of taurine supplement on the level of malondialdehyde (MDA), hydrogen peroxide (H_2O_2), superoxide dismutase (SOD), catalase (CAT), and the activity of glutathione peroxidase (GPx) in adult rat brains.

		MDA	H_2O_2	SOD	CAT	GPx
BF	Tau+Se VS DDD water+Se	↑	↑	↔	↔	↑
	Tau+Ex VS DDD water+Ex	↔	↑	↑	↔	↔
	Tau+Ex VS Tau+Se	↔	↔	↔	↔	↔
CC	Tau+Se VS DDD water+Se	↔	↔	↑	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↔	↔	↑	↔
	Tau+Ex VS Tau+Se	↓	↓	↔	↔	↔
HC	Tau+Se VS DDD water+Se	↔	↔	↔	↑	↔
	Tau+Ex VS DDD water+Ex	↔	↑	↑	↔	↔
	Tau+Ex VS Tau+Se	↔	↔	↑	↔	↔
ST	Tau+Se VS DDD water+Se	↔	↔	↔	↑	↑
	Tau+Ex VS DDD water+Ex	↑	↔	↑	↑	↑
	Tau+Ex VS Tau+Se	↔	↔	↑	↔	↔

↔ no change,

↑ significantly increased,

↓ significantly reduced.

(BF = basal forebrain, CC = cerebral cortex, HC = hippocampus, ST = striatum, Se = sedentary, and Ex = exercise, Tau = Taurine, DDD = double deionized distilled).

Table 3 Summary of the effects of taurine supplement on the level of malondialdehyde (MDA), hydrogen peroxide (H_2O_2), superoxide dismutase (SOD), catalase (CAT), and the activity of glutathione peroxidase (GPx) in middle-aged rat brains.

		MDA	H_2O_2	SOD	CAT	GPx
BF	Tau+Se VS DDD water+Se	↑	↔	↔	↔	↑
	Tau+Ex VS DDD water+Ex	↑	↔	↑	↔	↑
	Tau+Ex VS Tau+Se	↓	↔	↔	↑	↔
CC	Tau+Se VS DDD water+Se	↔	↔	↔	↔	↑
	Tau+Ex VS DDD water+Ex	↔	↔	↑	↔	↑
	Tau+Ex VS Tau+Se	↔	↔	↔	↔	↑
HC	Tau+Se VS DDD water+Se	↔	↑	↔	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↔	↔	↑	↑
	Tau+Ex VS Tau+Se	↔	↓	↔	↑	↑
ST	Tau+Se VS DDD water+Se	↑	↔	↔	↔	↔
	Tau+Ex VS DDD water+Ex	↑	↓	↑	↑	↔
	Tau+Ex VS Tau+Se	↔	↓	↑	↑	↔

↔ no change,

↑ significantly increased,

↓ significantly reduced.

(BF = basal forebrain, CC = cerebral cortex, HC = hippocampus, ST = striatum, Se = sedentary, and Ex = exercise, Tau = Taurine, DDD = double deionized distilled).

Table 4 Summary of the effects of Vitamin E supplement on the level of malondialdehyde (MDA), hydrogen peroxide (H_2O_2), superoxide dismutase (SOD), catalase (CAT), and the activity of glutathione peroxidase (GPx) in adult rat brains.

		MDA	H_2O_2	SOD	CAT	GPx
BF	Vit E+Se VS 1%Tween80+Se	↔	↔	↔	↔	↔
	Vit E+Ex VS 1%Tween80+Ex	↑	↑	↔	↑	↔
	Vit E+Ex VS Vit E+Se	↔	↔	↔	↔	↔
	Vit E+Se VS 1%Tween80+Se	↔	↔	↑	↔	↔
CC	Vit E+Ex VS 1%Tween80+Ex	↔	↔	↑	↔	↔
	Vit E+Ex VS Vit E+Se	↔	↔	↔	↔	↔
	Vit E+Se VS 1%Tween80+Se	↔	↔	↔	↔	↔
HC	Vit E+Ex VS 1%Tween80+Ex	↔	↔	↔	↔	↑
	Vit E+Ex VS Vit E+Se	↔	↓	↔	↔	↔
	Vit E+Se VS 1%Tween80+Se	↑	↔	↔	↔	↔
ST	Vit E+Ex VS 1%Tween80+Ex	↔	↔	↔	↔	↔
	Vit E+Ex VS Vit E+Se	↓	↔	↔	↔	↔

↔ no change,

↑ significantly increased,

↓ significantly reduced.

(BF = basal forebrain, CC = cerebral cortex, HC = hippocampus, ST = striatum, Se = sedentary, and Ex = exercise, Tau = Taurine, DDD = double deionized distilled).

Table 5 Summary of the effects of Vitamin E supplement on the level of malondialdehyde (MDA), hydrogen peroxide (H_2O_2), superoxide dismutase (SOD), catalase (CAT), and activity of glutathione peroxidase (GPx) in middle-aged rat brains.

		MDA	H_2O_2	SOD	CAT	GPx
BF	Vit E+Se VS 1%Tween80+Se	↔	↔	↔	↔	↔
	Vit E+Ex VS 1%Tween80+Ex	↔	↔	↑	↔	↔
	Vit E+Ex VS Vit E+Se	↔	↔	↑	↔	↑
	Vit E+Se VS 1%Tween80+Se	↔	↔	↔	↓	↔
CC	Vit E+Ex VS 1%Tween80+Ex	↔	↔	↑	↔	↑
	Vit E+Ex VS Vit E+Se	↔	↔	↑	↔	↑
	Vit E+Se VS 1%Tween80+Se	↔	↓	↓	↔	↔
HC	Vit E+Ex VS 1%Tween80+Ex	↔	↔	↔	↑	↔
	Vit E+Ex VS Vit E+Se	↔	↔	↔	↔	↔
	Vit E+Se VS 1%Tween80+Se	↔	↔	↔	↔	↔
ST	Vit E+Ex VS 1%Tween80+Ex	↓	↔	↔	↔	↑
	Vit E+Ex VS Vit E+Se	↓	↔	↔	↔	↑

↔ no change,

↑ significantly increased,

↓ significantly reduced.

(BF = basal forebrain, CC = cerebral cortex, HC = hippocampus, ST = striatum, Se = sedentary, and Ex = exercise, Tau = Taurine, DDD = double deionized distilled).

Table 6 Summary of the effects of Vitamin E supplement on the levels of malondialdehyde (MDA), hydrogen peroxide (H_2O_2) in middle-aged rat brains when compared between middle-aged and adult rat brains.

		DDD+Se	1% Tween80+Se	Tau+Se	Vit E+Se	DDD+Ex	1% Tween80+Ex	Tau+Ex	Vit E+Ex
MDA	BF	↔	↔	↑	↔	↔	↔	↔	↔
	CC	↑	↑	↑	↑	↑	↑	↑	↔
	HC	↔	↔	↔	↔	↔	↔	↔	↔
	ST	↑	↑	↑	↑	↑	↑	↔	↑
H_2O_2	BF	↔	↑	↔	↔	↑	↔	↔	↔
	CC	↑	↑	↑	↑	↑	↑	↑	↑
	HC	↔	↑	↑	↔	↑	↑	↔	↑
	ST	↑	↔	↑	↑	↑	↔	↔	↔

↔ no change, ↑ significantly increased, ↓ significantly reduced, (BF = basal forebrain, CC = cerebral cortex, HC = hippocampus, ST = striatum, Se = sedentary, and Ex = exercise, Tau = Taurine, DDD = double deionized distilled).

Table 7 Summary of the effects of Vitamin E supplement on the levels of superoxide dismutase (SOD), catalase (CAT), and the activity of glutathione peroxidase (GPx) when compared between middle-aged and adult rat brains.

		DDD+Se	1% Tween80+Se	Tau+Se	Vit E+Se	DDD+Ex	1% Tween80+Ex	Tau+Ex	Vit E+Ex
SOD	BF	↓	↓	↔	↔	↔	↓	↓	↔
	CC	↑	↑	↑	↔	↑	↑	↑	↔
	HC	↔	↔	↔	↔	↔	↔	↔	↔
	ST	↓	↓	↓	↓	↓	↓	↓	↓
CAT	BF	↔	↔	↔	↔	↔	↔	↔	↓
	CC	↔	↔	↔	↔	↔	↔	↔	↓
	HC	↓	↔	↓	↓	↓	↓	↔	↔
	ST	↔	↓	↓	↓	↔	↓	↔	↓
GPx	BF	↔	↔	↔	↔	↔	↔	↔	↔
	CC	↔	↔	↔	↓	↔	↓	↔	↔
	HC	↔	↔	↔	↔	↔	↔	↑	↔
	ST	↓	↓	↓	↓	↓	↓	↓	↓

↔ no change, ↑ significantly increased, ↓ significantly reduced, (BF = basal forebrain, CC = cerebral cortex, HC = hippocampus, ST = striatum, Se = sedentary, and Ex = exercise, Tau = Taurine, DDD = double deionized distilled).

Table 8 Summary of the effects of taurine supplement in conjunction with exercise on relative organ weights of skeletal muscles, liver, kidney, heart, spleen and lung in adult and middle-aged rats.

Organ		Adult rats	Middle-Aged rats
Gastrocnemius muscle	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↔
	Tau+Ex VS Tau+Se	↔	↔
Soleus muscle	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↔
	Tau+Ex VS Tau+Se	↔	↔
EDL muscle	Tau+Se VS DDD water+Se	↔	↑
	Tau+Ex VS DDD water+Ex	↔	↔
	Tau+Ex VS Tau+Se	↔	↔
Liver	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↔
	Tau+Ex VS Tau+Se	↔	↔
Kidney	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↔
	Tau+Ex VS Tau+Se	↔	↔

Table 8 Summary of the effects of taurine supplement in conjunction with exercise on relative organ weights of skeletal muscles, liver, kidney, heart, spleen and lung in adult and middle-aged rats (Cont.).

Organ		Adult rats	Middle-Aged rats
Heart	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↔
	Tau+Ex VS Tau+Se	↔	↔
Spleen	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↔
	Tau+Ex VS Tau+Se	↔	↔
Lung	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↓
	Tau+Ex VS Tau+Se	↔	↓

↔ no change,

↑ significantly increased,

↓ significantly reduced.

(BF = basal forebrain, CC = cerebral cortex, HC = hippocampus, ST = striatum, Se = sedentary, and Ex = exercise, Tau = Taurine, DDD = double deionized distilled).

Table 9 Summary of the effects of Vitamin E supplement in conjunction with exercise on relative organ weights of skeletal muscles, liver, kidney, heart, spleen and lung in adult and middle-age rats.

Organ		Adult rats	Middle-Aged rats
Gastrocnemius muscle	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↔
	VitE+Ex VS VitE+Se	↔	↔
Soleus muscle	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↔
	VitE+Ex VS VitE+Se	↔	↔
EDL muscle	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↔
	VitE+Ex VS VitE+Se	↔	↔
Liver	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↔
	VitE+Ex VS VitE+Se	↔	↔
Kidney	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↑
	VitE+Ex VS VitE+Se	↔	↔

Table 9 Summary of the effects of Vitamin E supplement in conjunction with exercise on relative organ weights of skeletal muscles, liver, kidney, heart, spleen and lung in adult and middle-aged rats (Cont.).

Organ		Adult rats	Middle-Aged rats
Heart	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↔
	VitE+Ex VS VitE+Se	↔	↔
Spleen	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↔
	VitE+Ex VS VitE+Se	↔	↔
Lung	VitE+Se VS 1% Tween 80+Se	↔	↓
	VitE+Ex VS 1% Tween 80+Ex	↔	↑
	VitE+Ex VS VitE+Se	↔	↑

↔ no change,

↑ significantly increased,

↓ significantly reduced.

(BF = basal forebrain, CC = cerebral cortex, HC = hippocampus, ST = striatum, Se = sedentary, and Ex = exercise, Tau = Taurine, DDD = double deionized distilled, EDL muscle= extensor digitorum longus muscle).

Table 10 Summary of the effects of taurine supplement in conjunction with exercise on blood parameters in adult and middle-aged rats.

parameters		Adult rats	Middle-Aged rats
Blood Sugar	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↓	↔
	Tau+Ex VS Tau+Se	↔	↓
Total cholesterol	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↔
	Tau+Ex VS Tau+Se	↔	↔
Triglyceride	Tau+Se VS DDD water+Se	↔	↓
	Tau+Ex VS DDD water+Ex	↓	↓
	Tau+Ex VS Tau+Se	↔	↔
Total LDH	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↓	↔
	Tau+Ex VS Tau+Se	↔	↔
AST	Tau+Se VS DDD water+Se	↓	↔
	Tau+Ex VS DDD water+Ex	↔	↔
	Tau+Ex VS Tau+Se	↔	↔

↔ no change, ↑ significantly increased, ↓ significantly reduced.

(LDH = lactate hydrogenase, AST = Aspartate aminotransferase, ALT = alanine aminotransferase, BUN= blood urea nitrogen).

Table 10 Summary of the effects of taurine supplement in conjunction with exercise on blood parameters in adult and middle-aged rats (Cont.)

parameters		Adult rats	Middle-Aged rats
ALT	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↑
	Tau+Ex VS Tau+Se	↔	↑
BUN	Tau+Se VS DDD water+Se	↔	↔
	Tau+Ex VS DDD water+Ex	↔	↓
	Tau+Ex VS Tau+Se	↔	↔
Creatinine	Tau+Se VS DDD water+Se	↑	↔
	Tau+Ex VS DDD water+Ex	↑	↔
	Tau+Ex VS Tau+Se	↑	↔

↔ no change, ↑ significantly increased, ↓ significantly reduced.

(LDH = lactate hydrogenase, AST = Aspartate aminotransferase, ALT = alanine aminotransferase, BUN= blood urea nitrogen).

Table 11 Summary of the effects of Vitamin E supplement in conjunction with exercise on blood parameters in adult and middle-aged rats.

Organ		Adult rats	Middle-Aged rats
Blood Sugar	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↑	↔
	VitE+Ex VS VitE+Se	↔	↔
Total cholesterol	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↔
	VitE+Ex VS VitE+Se	↔	↔
Triglyceride	VitE+Se VS 1% Tween 80+Se	↓	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↓
	VitE+Ex VS VitE+Se	↔	↔
Total LDH	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↓
	VitE+Ex VS VitE+Se	↔	↔
AST	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↔
	VitE+Ex VS VitE+Se	↔	↔

↔ no change, ↑ significantly increased, ↓ significantly reduced.

(LDH = lactate hydrogenase, AST = Aspartate aminotransferase, ALT = alanine aminotransferase, BUN= blood urea nitrogen).

Table 11 Summary of the effects of Vitamin E supplement in conjunction with exercise on blood parameter in adult and middle-rats (Cont.)

Organ		Adult rats	Middle-Aged rats
ALT	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↔
	VitE+Ex VS VitE+Se	↔	↔
BUN	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↔
	VitE+Ex VS VitE+Se	↔	↔
Creatinine	VitE+Se VS 1% Tween 80+Se	↔	↔
	VitE+Ex VS 1% Tween 80+Ex	↔	↔
	VitE+Ex VS VitE+Se	↔	↔

↔ no change, ↑ significantly increased, ↓ significantly reduced.

(LDH = lactate hydrogenase, AST = Aspartate aminotransferase, ALT = alanine aminotransferase, BUN= blood urea nitrogen).

Table 12 Summary of the effects of Vitamin E supplement in conjunction with exercise on relative organ weights of skeletal muscles, liver, kidney, heart, spleen and lung in adult and middle-age rats.

	DDD+Se	1% Tween80+Se	Tau+Se	Vit E+Se	DDD+Ex	1% Tween80+Ex	Tau+Ex	Vit E+Ex
Gas	↓	↓	↓	↓	↓	↓	↓	↔
Sol	↔	↔	↔	↔	↔	↔	↔	↔
EDL	↔	↔	↔	↔	↔	↔	↔	↔
Liver	↓	↓	↓	↓	↔	↓	↔	↓
Kidney	↔	↔	↔	↔	↔	↔	↔	↔
Heart	↓	↔	↓	↔	↔	↔	↔	↓
Spleen	↔	↔	↔	↔	↔	↓	↔	↔
Lung	↔	↔	↔	↔	↔	↔	↔	↔

↔ no change, ↑ significantly increased, ↓ significantly reduced.

(Gas = gastrocnemius, Sol = Soleus, EDL = extensor digitorum longus).

Table 13 Summary of the effects of Vitamin E supplement in conjunction with exercise on blood parameters in adult and middle-aged rats.

	DDD+Se	1% Tween80+Se	Tau+Se	Vit E+Se	DDD+Ex	1% Tween80+Ex	Tau+Ex	Vit E+Ex
Blood Sugar	↑	↑	↑	↔	↔	↔	↔	↔
TC	↑	↑	↑	↑	↑	↑	↑	↑
TG	↑	↔	↔	↔	↑	↑	↑	↔
LDH	↓	↔	↓	↔	↔	↔	↓	↔
AST	↔	↑	↔	↑	↑	↑	↔	↔
ALT	↔	↑	↑	↑	↑	↑	↔	↑
BUN	↓	↔	↓	↓	↔	↔	↓	↔
Creatinine	↔	↔	↓	↔	↑	↔	↔	↔

↔ no change, ↑ significantly increased, ↓ significantly reduced.

(TC = total cholesterol, TG = triglyceride, LDH = lactate hydrogenase, AST = Aspartate aminotransferase, ALT = alanine aminotransferase, BUN= blood urea nitrogen).

CHAPTER V

DISCUSSION

5.1 Effects of Taurine Supplement in Conjunction with Exercise on Antioxidant Enzymes Activities, Malondialdehyde Levels and Hydrogen peroxide levels in Adult Rat Brains.

5.1.1 Effects of Taurine Supplement in Conjunction with Exercise on Relative Organ Weight of Adult and Middle-Aged Rats

In adult rats, regular swimming exercise for 8 weeks didn't caused change in muscle weight and all organ weight which is inconsistent with endurance training by running for 10 weeks that significantly increased ROWs of soleus and EDL muscles in adult rats (Maxwell *et al*., 1992; Shinoda *et al*., 2002). Increases in soleus muscle weight were found in 12 months old rats after wheel running over a period for 6 months and in 18 months old rats after 12 weeks swimming training (Huang *et al*., 2016; Schreckenberg *et al*., 2017). In middle-aged rats, regular swimming exercise for 8 weeks increased in EDL muscle. Aerobic exercise could increase muscle fiber in old man (Harber *et al*., 2012). Endurance training by running for 10 weeks increased soleus and EDL muscle weight in adult *ad libitum* fed rats (Maxwell *et al.*, 1992). The present study found that taurine supplement in the sedentary rats didn't caused change muscle weight and all organs weight in adult rats. In the middle-aged rats, taurine supplement alone significantly increased EDL muscle weight. Taurine supplement alone reduced

gastrocnemius muscle weight when compared to sedentary group (Manabe *et al*., 2003). Taurine supplement with running exercise could enhance muscle weight (EDL, soleus, and gastrocnemius muscle) (Manabe *et al*., 2003; Yatabe *et al*., 2003).

The present study found that swimming exercise for 8 weeks significantly increased heart weight in the middle-aged rats. Previous study showed that regular exercise training induced a myocardial hypertrophy and increased heart weight in adult rats (Blr *et al.*, 1970; Broderick *et al.*, 2011; Sakamoto and Grunewald, 1986). Exercise can reduce liver weight (Broderick *et al*., 2011) and lung weight (Shinoda *et al.*, 2002). Forced swimming stress in adult rats for 1 week caused increases in kidney and liver weights (Nayanatara *et al*., 2005). Taurine supplement in this study didn't not cause change ROWs in both adult and middle-aged rats. Previous study demonstrated that liver weight of adult rats was not changed after taurine supplement for 28 days (Mahalakshm et al., 2003). Administration of taurine (0.01% and 0.05%) didn't cause changes in liver weight and spleen weight in adult rats (Wang *et al.*, 2009). Healthful effects of taurine on hepatic lipids and oxidative stress were demonstrated in ethanol treated rats after taurine treatment for 4 weeks (Pushpakiran *et al*., 2004a). Exercise alone didn't not cause change in ROWs of both adult and middle-aged rats in the present study. Previous study showed that exercise alone could increase the heart weight (Sakamoto and Grunewald, 1986). Exercise training induced a myocardial hypertrophy and heart weight in middle-aged rat (Fiebig *et al*., 1996; Mazzeo and Horvath, 1986). Effects of taurine supplement in conjunction with exercise on ROWs of adult rats significantly decreased spleen weight, in middle-aged taurine supplement in conjunction with exercise didn't not cause change all organs weight same in previous study (Mahalakshm *et al*., 2003; Manabe *et al*., 2003). Taurine supplement may

decrease spleen weight resulting of exercise. Spleen weight was decreased as result of an expulsion of storage blood during exercise (Gollnick *et al*., 1967) and it was related in mass of lymphoid centers in the exercise animals.

5.1.2 Effects of Taurine Supplement in Conjunction with Exercise on Blood Parameters of adult and Middle-Aged Rats

Regular swimming exercise for 8 weeks didn't caused change in blood sugar levels in both adult and middle-aged rats which is inconsistent with the previous study in adult rats underwent running for 10 weeks showing a reduction in blood glucose (Broderick *et al*., 2011). Treatment of taurine benefits many kinds of pathologies. Many studies demonstrated the effectiveness of taurine supplementation against both insulin dependent, non-insulin dependent diabetes mellitus and insulin resistance in rats (Foda *et al*., 2016; Kaplan *et al*., 2004). Taurine is involved in glucose homeostasis, but the specific molecular mechanisms are unknown (Franconi *et al.*, 2004). Taurine physically help hypoglycemic effects by enhancing insulin action, as well as by promote the interaction of insulin with its receptor (Lampson *et al.*, 1983). Taurine increased glycogen synthesis, glycolysis and glucose uptake in the liver and heart of adult rats (Higo *et al.*, 2008). These effects were shown to be dependent of insulin concentration. In addition, taurine has been shown to be benefit in insulin sensitivity in type 2 diabetes when compared to N-acetylcystein in the study of men (Xiao *et al*., 2008). The present study found that taurine supplement in conjunction with exercise significantly reduced blood sugar level in adult rats, may be due to combined effects of exercise and taurine. From previous report showed taurine alone has benefit to increased glycogen synthesis,

glycolysis and glucose uptake in the liver and heart and exercise has effect to generate energy from glucose.

The present study found that swimming exercise for 8 weeks didn't not cause change lipid profile in both adult and middle-aged rats. Running exercise could enhance total cholesterol level in adult rats (Shinoda *et al*., 2002), running for 10 weeks could reduce triglyceride level in adult rats (Broderick *et al*., 2011). The results were contrary to previous studies. Taurine supplement could reduce total cholesterol, and triglyceride in diabetic rat (Foda *et al.*, 2016) but taurine supplement and taurine supplement in conjunction with exercise in this study was not change in lipid profile in both adult and middle-aged-rats. In aging cause decline in many organisms and mechanism, once mechanism that decline with aged is metabolic rate and hormone (Heemst, 2010). In many blood parameters in middle-aged rats in this study found that has higher than adult rats.

Exercise alone significantly reduced total LDH levels in adult rats and increased total LDH levels in middle-aged rats. Taurine supplement alone didn't not cause change in both adult and middle-aged rats, Taurine supplement in conjunction with exercise significant reduced in adult rat. Taurine supplement could reduce total LDH in diabetic rat (Foda *et al.*, 2016). Administration of taurine (500, 1000 mg/kg) significantly reduced total LDH level in adult rats (Heidari *et al.*, 2016). Taurine supplement in conjunction with exercise may benefit to reduced cell damage from aged.

All of rats in sedentary and swimming group with or without taurine supplementation in adult rats had no effect on serum AST and ALT levels. In human, reduction of serum AST and ALT levels was shown after running exercise 135 min/week for 8 weeks (Shamsoddini *et al*., 2015). Endurance training and resistance

training exercise could reduce AST and ALT levels in human (Shamsoddini *et al*., 2015). Taurine at 400 mg/kg body weight administered by gavage could reduce AST level and ALT level (Ezekiel *et al*., 2015). Taurine at 200 and 1000 mg/kg body weight could reduce serum AST and ALT levels in diabetic rats (Foda *et al*., 2016; Heidari *et al*., 2016). It is remarkable that the hepatoprotective property of taurine is due to its ability to decrease ROS, enhance mitochondrial function and modify cytoplasmic and mitochondrial Ca^{2+} homeostasis in biological systems (Asha and Devadasan, 2013; Heidari *et al*., 2016). Previous study found that taurine supplement with iron could prevent iron-overloaded in liver mice (Zhang *et al*., 2014). Taurine could prevent liver damage from alcohol after received 30% ethanol and 2% taurine in drinking water for 28 day in rats (Pushpakiran *et al*., 2004a). Heidari and co-workers (2016) demonstrated the hepatoprotective effect of taurine as taurine could reduce AST and ALT levels in adult rats.

In sedentary and regular swimming rats, taurine supplement alone and taurine supplement in conjunction with exercise had no effect on BUN levels. Exercise alone significantly increased BUN levels. Taurine supplement significantly increased creatinine levels in both sedentary and exercise adult rats. Exercise alone had no effect on creatinine levels, but taurine supplement in conjunction with exercise significantly reduced creatinine levels in adult rats. Exercise induces changes in the renal haemodynamics and in electrolyte and protein excretion. During exercise cause reducing renal blood flow. The reduction of renal blood flow is related to the intensity of exercise, strenuous exercise may cause renal blood flow fall to 25% of the resting value (Alyea *et al*., 1958). The reduction of renal blood flow during exercise produces a concomitant effect on the glomerular filtration rate, though the latter decreases

relatively less than the former during exertion (Bellinghieri *et al.*, 2008). An antidiuretic hormone effect is observed during strenuous exercise. Changes in urine flow are dependent on the plasma antidiuretic hormone levels which are increased by strenuous exercise (Kozlowski *et al.*, 1967). The kidney plays a importance role in regulating body stores of taurine. It possesses the electrogenic Na^{+}- $C1^{-}$ coupled co-transporter in the proximal tubule brush border membrane (Zelikovic *et al.*, 1989). Taurine help many functions including bile acid conjugation, modulation of neurotransmission, stabilization of the retinal membrane and osmoregulation (Gregory, 1994). Renal impairment is common during exercise (Bellinghieri *et al*., 2009). Creatinine is known to be an important biochemical parameter indicating kidney function disorders, and serum creatinine concentrations might be influenced by skeletal muscle damage. Creatinine is a breakdown product from the muscles and depends on muscle mass (Vinge *et al*., 1999). Taurine has protective effect on renal from antitumor drug cisplatin by increases in the serum albumin levels, kidney GSH contents, kidney GPx activity, and reduced BUN and creatinine levels (Saad and Al-Rikabi, 2002). In middle-aged sedentary and regular swimming exercise rats, taurine alone and regular swimming exercise alone had no effect on blood glucose levels. Taurine supplement in conjunction with exercise significantly reduced blood glucose levels in middle-aged rats. Low-intensity of exercise could reduce blood-glucose in diabetic rats (Gimenes *et al*., 2015; Kim *et al*., 2014; Oliveira *et al*., 2019). Taurine supplement caused reduction of blood glucose in fructose-fed rats (Anitha Nandhini *et al*., 2005). Taurine could improve glucose metabolism and lowers insulin resistance in OLETF rats supplemented with 2% taurine for 12 weeks (Kim *et al*., 2012). Taurine can improve insulin sensitivity

which increases blood flow to muscle and glucose uptake by muscle and may improve energy production from glucose during exercise (Nakaya *et al*., 2000).

In middle-aged sedentary rats, taurine supplement alone significantly reduced plasma TG levels. In middle-aged regular swimming exercise rats, taurine supplement alone had no effect on plasma TC levels, but significantly reduced plasma TG levels. Taurine supplement in conjunction with exercise had no effect on both plasma TC and TG levels in middle-aged rats. Previous study showed that running for 8 weeks could increase cholesterol and triglyceride level in aging rat (Hajighasem *et al*., 2018). On the other hand, swimming exercise for 12 weeks could reduce triglyceride and low-density lipoprotein cholesterol, and increased high-density lipoprotein cholesterol in middle-aged women (Kang, 2013). Aerobic exercise could reduce total cholesterol and triglyceride levels in chronic hemodialysis patients (Afshar et al., 2010). Previous study showed that taurine supplement could increase cholesterol level in sedentary rats (Manabe *et al*., 2003). Taurine supplement with exercise could reduce serum cholesterol and triglyceride levels compared to taurine supplement alone (Manabe *et al*., 2003). Taurine has effect in conjugation of bile acids that effect on the solubility of cholesterol, increasing its excretion, and administration of taurine could reduce plasma cholesterol levels and triglyceride levels (Mizushima *et al.*, 1996).

Total LDH were not changed in taurine supplement alone in both sedentary and exercise middle-aged-rats. Exercise alone significantly increased total LDH levels in middle-aged rats. Taurine supplement in conjunction with exercise had no effect on total LDH levels in middle-aged rats. Previous study demonstrated that exercise training could enhance LDH level in middle-aged rats (Fiebig *et al*., 1996). Taurine treatments could prevent cell damaged from exercise and aged.

the tubules in middle-aged rats. Taurine supplement can prevent renal toxicity by cyclophosphamide or gentamicin through reduction of renal toxicity biomarkers such as, serum creatinine and BUN in male rats (Alhumaidha *et al*., 2016; Erdem *et al*., 2000). Taurine involves in several physiologic functions of the kidney, including renal blood flow, glomerular filtration and its rate, osmoregulation, ion reabsorption and secretion, and composition of urine (Chesney *et al.*, 2012). The present findings suggested that taurine supplement in conjunction with exercise has benefit to kidney in middle-aged rats by acting at glomerular to increase glomerular filtration and increase reabsorption in the tubules.

5.1.3 Effects of Taurine Supplement in Conjunction with Exercise on MDA and Hydrogen Peroxide levels of Adult and Middle-Aged Rat Brains.

Regular swimming exercise for 8 weeks didn't cause change in MDA levels and H_2O_2 levels in all studies area of adult rat brains. Stress swimming exercise for 1 week could increase lipid peroxidation in adult rats (Nayanatara *et al*., 2005), daily stress exercise may impair the antioxidant defenses in the body by changing the balance between oxidant and antioxidant factors, the increased concentration of glucocorticoids hormones exacerbate reactive oxygen species (ROS) generation in the body (McIntosh and Sapolsky, 1996). Swimming for 8 weeks could reduce MDA level in whole brain, cerebral cortex, cerebellum, and hippocampus in adult rats (Devi and Kiran, 2004; Nonato *et al.*, 2016; Ravikiran *et al*., 2016). Stress running for 5 minutes could increase MDA and protein carbonyl in rat serum (Trofin *et al*., 2014). In the adult rats, endurance training exercise did not cause change in MDA and H_2O_2 levels through antioxidant enzyme defense system to scavenging ROS during exercise.

In sedentary adult rats, taurine supplement caused increases in MDA levels in basal forebrain and in H_2O_2 levels in basal forebrain. In exercise adult rats, taurine supplement caused no change in MDA levels in all studied brain areas, but coursed the increase in H_2O_2 levels in basal forebrain. Taurine supplementation can protect brain injury, apoptosis, and oxidative stress from traumatic brain injury, reduced MDA levels and attenuated ROS levels in rat brain by enhancing antioxidant enzyme activity (Niu *et al*., 2018). Administration of taurine can reduce MDA levels in the brain and protect brain injury in closed-head injury in adult rats (Sun *et al.*, 2015).

In adult rats, taurine supplement in conjunction with exercise significantly decreased levels of MDA and H_2O_2 in cerebral cortex and significantly increased MDA levels in striatum, compared to taurine supplement in sedentary group. In previous study, MDA levels in cerebellum of adult rat brains was lower than control group when received single dose of taurine at 200 mg/kg/day for 7 days (Zuhal Yildirim and Kilic, 2011). Taurine supplement could reduce MDA levels and increase GPx enzyme activity levels in liver, heart and stomach of (Anand P. *et al*., 2011; Yildirim Z. *et al*., 2007). Taurine supplement at high-dose (800 mg/kg/day) for 4 weeks could decline MDA content in brain of rats treated with aluminum (281.40 mg/kg/day in first week) (Qiao *et al.*, 2015). Taurine has protective effect in the brain by reduced MDA level and increased antioxidant enzyme activity in rat brains (Niu *et al*., 2018). Taurine supplementation has been shown to substantially reduce infarct volume, brain swelling, cell death, and neurological deficits in a stroke-induced rat model. Several researchers have associated mitochondrial dysfunction with increased ROS and superoxide production, glutathione oxidation, and reduced antioxidant enzymes (Sun *et al.*, 2012; Wang *et al*., 2016). Taurine increases antioxidant activity by reducing superoxide

production, which leads to improved mitochondrial function (Schaffer *et al*., 2012). Taurine supplementation can reduce brain injury by reduced ROS and MDA. On the other hand, taurine supplementation can increase antioxidant enzyme activity such as CAT, SOD, and GPx in the brain. Reduction of the levels of MDA and H_2O_2, markers of peroxidation and ROS, in cerebral cortex following taurine supplement with exercise found in the present study suggesting that taurine supplement in conjunction with exercise could attenuate ROS and prevent damage from free radical in some part of the brain involving learning and memory and may protect neurodegenerative disease.

Regular swimming exercise for 8 weeks caused decreases in MDA levels in basal forebrain and increases in H_2O_2 levels in cerebral cortex and striatum in middle-aged rats brains. Swimming exercise for 8 weeks training could reduce MDA level in middle-aged hippocampus (Ravikiran *et al*., 2016). Long term exercise caused adaptive to scavenging ROS in the tissue, in this study found that swimming exercise for 8 weeks can reduced MDA levels in basal forebrain as the site of learning and increases in H_2O_2 levels in cerebral cortex and striatum in middle-aged rats may cause from aged because aging cause from accumulation of the free radical and imbalance of free radical and antioxidant enzyme in cerebral cortex and striatum.

In sedentary middle-aged rats, taurine supplement causes increases in MDA levels in basal forebrain and in H_2O_2 levels in hippocampus and striatum. In exercise middle-aged rats, taurine supplement caused decreased in MDA levels in basal forebrain and decreased in H_2O_2 levels in hippocampus and striatum. Taurine supplement for 24 hours in B16F10 Melanoma Cells was shown cause reduction of ROS in the cell (Yu and Kim, 2009). Taurine supplement in single dose at 200 mg/kg/day for 7 days could reduce MDA level in middle-aged cerebellum (Zuhal

Yildirim and Kilic, 2011). Taurine has protective effect to against oxidative stress caused by ethanol, taurine treatment for 28 days could lower the TBARS in the brain of the rat received ethanol (Pushpakiran *et al*., 2004b).

In sedentary middle-aged rats, taurine supplement causes increases in MDA levels in basal forebrain and in H_2O_2 levels in hippocampus and striatum. Taurine supplement in conjunction with exercise caused decreased in MDA levels in basal forebrain and decreased in H_2O_2 levels in hippocampus and striatum. Taurine possesses antioxidant properties in peroxidatively damaged tissues. Increased MDA level, were found in taurine-deficient rats (Harada *et al*., 1990). Regular exercise could reduce MDA level in middle-aged rat brain (Radak *et al*., 2006). Taurine could decrease MDA level produced by pyrazinamide, suggesting its efficacy in scavenging the ROS and modulating the oxidative stress caused by pyrazinamide (Taziki *et al*., 2018). Taurine supplement in conjunction with exercise could reduce ROS and MDA levels in some part of the brain that may be due to duration and intensity of exercise and oxidative stress related with aged. Taurine supplement in conjunction with exercise in this study showed that reduction of ROS levels in some part of the brain involving in learning and memory in the part of short term and long term memories. Exercise training with taurine supplement may cause adaptive changes in the antioxidant defenses system by reduced free radical in the striatum in the middle-aged rats.

5.1.4 Effects of Taurine Supplement in Conjunction with Exercise on Antioxidant Activities of CAT, SOD, and GPx in Adult and Middle-Aged Rat Brains.

Regular swimming exercise for 8 weeks didn't not cause change all antioxidant enzyme in all brain regions in the present study. Previous study found that swimming exercise for 8 weeks could increase SOD levels in whole brain (Nonato *et al*., 2016). Swimming exercise could enhance all antioxidant enzyme activities in hippocampus and cerebral cortex in both male and female rats (Devi and Kiran, 2004; Ozkaya *et al*., 2002; Stone *et al*., 2014). Swimming training 30 minuets/day with a load of 3 % of their body weight tied to their tails (30 day for 5 day/week) could increase antioxidant enzyme activities in cerebral cortex, hippocampus, and cerebellum regions of rat brains (Ravikiran *et al*., 2016). Stress running exercise for 5 minutes could reduce antioxidant enzyme activities in rat serum (Trofin *et al*., 2014). Exercise training in diabetic rats could increase antioxidant enzyme activities of GPx and CAT but not Cu, Zn-SOD in the brain (Ozkaya *et al*., 2002).

In adult sedentary rats, taurine supplement significantly increased CAT levels in hippocampus and striatum, and GPx enzyme activity in basal forebrain. In adult exercise rats, taurine supplement significantly increased SOD levels in basal forebrain and striatum, CAT levels in cerebral cortex and striatum, and GPx enzyme activity levels in striatum. Previous study demonstrated that increased GPx levels in adult rat brains were found after receiving 30% ethanol and 2% taurine in drinking water for 28 days (Pushpakiran *et al*., 2004a). It is possibly that taurine can suppress brain damage resulting from alcohol by extend GPx enzyme activity in the brain since GPx enzyme activity has protective effect to damaging from oxidative stress (V.D. Almeida *et al*.,

1994). Taurine supplementation has protective effect on traumatic brain injury by increasing CAT, SOD, and GPx levels in the brain (Niu *et al.*, 2018; Sun *et al*., 2015). Taurine supplement in conjunction with exercise in adult rats caused significantly increased SOD levels in striatum, compared to sedentary rats. The present results indicated that taurine supplement in conjunction with exercise has beneficial effect in increased antioxidant enzyme activity in some parts of the brain at the site of short term memory in adult rats.

Regular swimming exercise for 8 weeks caused increase in CAT levels in cerebral cortex in middle-aged brains. Previous study showed that swimming exercise could enhance enzymatic antioxidant enzyme in middle-aged and aging rat cerebral cortex and hippocampus (Devi and Kiran, 2004). Exhaustive exercise could enhance antioxidant enzyme activity in heart, liver, and skeletal muscle in aging rats (Ji, 1993). Exercise training could enhance SOD activity in brainstem and corpus striatum, but lower SOD activity in hippocampus, which may be due to different brain regions contained different activities of antioxidant enzymes (Rybak *et al.*, 1995). Swimming training 30 minutes/day 5 day/week for 30 days could increase antioxidant enzyme activity in cerebral cortex, hippocampus, and cerebellum regions of the rat brains (Ozkaya *et al*., 2002; Ravikiran *et al.*, 2016).

In middle-aged sedentary rats, taurine supplement significantly increased in GPx enzyme activity levels in basal forebrain. In middle-aged exercise rats, taurine supplement significantly increased SOD levels in basal forebrain, cerebral cortex and striatum, CAT levels in hippocampus, and striatum, and GPx enzyme activity in basal forebrain and hippocampus. Taurine treatment for 24 hour could enhance antioxidant enzyme level in B16F10 melanoma cell (Yu and Kim, 2009). Taurine supplement for

7 days could increase glutathione and SOD levels in cerebellum of middle-aged rats (Zuhal Yildirim and Kilic, 2011). Protective effect of taurine treatment against brain damaged from ethanol by increasing CAT, SOD and GPx enzymatic antioxidant activity in rat whole brains were demonstrated (Pushpakiran *et al*., 2004b).

SOD scavenging a large amount of ROS to H_2O_2 and O_2 result in the decrement of ROS activity, and then CAT and GPx stimulate metabolized peroxide, including H_2O_2, and protect the cellular membranes from lipid peroxidation. Taurine can act as antioxidation, previous reported that taurine can improve glutathione levels by promoting cysteine into the biosynthetic pathway of glutathione (Zuhal Yildirim and Kilic, 2011). Regular exercise can attenuate damaged of ROS through an elevated generation of antioxidant enzyme activities and exercise have benefits to increase the growth factors that protects neurons, assists neuronal plasticity and induces learning and memory (Marosi *et al*., 2012; Rybak *et al*., 1995). Perhaps a taurine supplement in conjunction with exercise may have induced adaptive changes in the antioxidant defenses in order to compensate for greater free radical generation in the both adult and middle-aged rat brain. From the previous has to talk about aging theory, that have many theory that describe about aging, few of theories are “Free Radicals Theory” and “Wear and Tear Theory”, free radicals theory that reactive oxygen species (ROS) as the most important signaling that responsible for the development of cell senescence and organismal aging and free radicals cause cell damage and damage to the macromolecular components, giving rise to accumulated damage-causing cells, and eventually organs, to stop functioning (Afanas’ev, 2010). Wear and Tear Theory is cells and tissues have vital parts that wear out from repeated use and resulting in aging (Brys *et al*., 2007). In aging cause from imbalance between the antioxidant defenses and

among of the free radical, brain is an organ most sensitive to oxidative stress and free radical. The large sign of aging is oxidative stress and a significant volume of evidence of oxidative stress is an important pathogenic factor involving neurodegenerative disorder such as Alzheimer's disease (AD) and Parkinson disease (PD) (Guerra-Araiza *et al*., 2013). In present study, study in four brain regions including basal forebrain, cerebral cortex, hippocampus, and striatum that involving learning and memory. From 2 theories in previous review, I found that in middle-aged rat brains have reduced the antioxidant enzymes than adult rat brains conform to previous theory. Taurine act as antioxidant and exercise has benefit to attenuate damaged of ROS, the present study suggest that effect of taurine supplement in conjunction with exercise have effect to enhanced antioxidant enzyme in some part of the brain involve learning and memory both short term and long term memory in the middle-aged rat brain. Taurine supplement in conjunction with exercise significantly increased CAT levels and GPx enzyme activity levels in hippocampus and striatum, suggestion that taurine supplement in conjunction with exercise in single dose at 800 mg/kg/day for 8 weeks could attenuate ROS in some part of middle-aged rat brain, involving learning and memory. Taurine supplement alone in middle-aged rats showed effect to increase in SOD in the cerebral cortex.

5.2 Effects of Vitamin E Supplement in Conjunction with Exercise on Antioxidant Enzymes Activities, Malondialdehyde Levels and Hydrogen Peroxide in Adult and Middle-Aged Rat Brains

5.2.1 Effects of Vitamin E Supplement in Conjunction with Exercise on Relative Organ Weight of Adult and Middle-Aged Rats

Vitamin E supplement alone significantly increased soleus muscle in middle-aged rats vitamin E supplement in conjunction with exercise was no change in muscle weight. Vitamin E supplement alone on ROWs in adult and middle-aged rats, there was no change in all organ weight in present study. Vitamin E supplement in the exercise group was no change in all organ weight in present study. Vitamin E supplement in conjunction with exercise significantly reduce in ROW of heart in the middle-aged rats. Vitamin E or alpha-tocopherol is one of major antioxidant in fruit, vegetable, and animals (Hardie *et al*., 1990). Vitamin E can prevent oxidative stress in the body and protect against many conditions such as cancer and heart disease (I-Min Lee, 2005). Wear and Tear Theory, cells and tissues have vital parts that wear out from repeated use and resulting in aging. In parts of the body ultimately wear out from repeated use (Brys *et al.*, 2007) it's become from ROW of heart in present study decreased than adult rats. Exercise training induced a myocardial hypertrophy and heart weight in middle-aged rat (Fiebig *et al*., 1996; Mazzeo and Horvath, 1986) the ROW of heart in the exercise group in present study significantly increased than vitamin E supplement alone may cause from effect of combination between vitamin E and exercise in middle-aged rats.

5.2.2 Effects of Vitamin E Supplement in Conjunction with Exercise on Blood Parameters of Adult and Middle-Aged Rats

Vitamin E supplement alone on blood parameters in adult and middle-aged rats was no change in all blood parameters. Vitamin E supplement in the exercise group significantly increase blood sugar levels in adult rats. In the middle-aged rats in the exercise group significantly reduce triglyceride levels and total LDH levels, exercise increases the generation of oxygen free radicals and lipid peroxidation (Radak *et al*., 2013) benefit of vitamin E is scavenge reactive oxygen species and prevent deleterious effects and repair in the cells (Devi, 2009). Total LDH levels is the maker of cell damage, reduced total LDH levels in vitamin E supplement in the exercise group may cause from adaptive from exercise or vitamin E supplement or the effect of combination of exercise and vitamin E.

5.2.3 Effects of Vitamin E Supplement in Conjunction with Exercise on MDA and Hydrogen Peroxide Levels of Adult and Middle-Aged Rat Brains.

In adult sedentary rats, vitamin E supplement significantly increased MDA levels in striatum. In adult exercise rats, vitamin E supplement significantly increased both MDA and H_2O_2 levels in basal forebrain.

In middle-aged rats, vitamin E supplement significantly decreased MDA levels in hippocampus. In middle-aged exercise rats, vitamin E supplement was not cause change MDA and H_2O_2 levels in all brain regions of this study.

The increase in lipid peroxidation with age in many tissues has been reported to occur concomitantly with increased DNA and protein oxidation (Migliore and Coppede, 2009). During physical exercise there is an elevated generation of ROS in

term of high intensity of exercise (Radak *et al.*, 2013). Swimming training rats showed significant enhanced activities in lipid peroxidation, and glutathione peroxidase (GPx) in brain (Hara *et al.*, 1997). Lipid peroxidation is frequently used as an index of tissue oxidative stress which results from free radical damage to membrane components of the cell and can determined which levels of MDA and H_2O_2 (Grotto *et al.*, 2009; Gueraud *et al.*, 2010). The result in this study suggest that vitamin E supplement in adult rats both sedentary and exercise has effect on basal forebrain and striatum are the part of learning and short term memory, but in hippocampus of middle-aged rats vitamin E supplement can reduced H_2O_2 levels, at the part of long term memory.

5.2.4 Effects of Vitamin E Supplement in Conjunction with Exercise on Antioxidant Activities of CAT, SOD, and GPx in Adult and Middle-aged Rat Brains.

In adult sedentary rats, vitamin E supplement there was no significant difference in all study of brain regions. In adult exercise rats, vitamin E supplement significantly increased CAT levels in basal forebrain, and GPx enzyme activity levels in hippocampus, but SOD levels there was no significant difference in all study of brain regions.

In middle-aged rats, vitamin E supplement there was no significant difference in all study of brain regions. In middle-aged exercise rats, vitamin E supplement significantly increased SOD levels in basal forebrain and cerebral cortex, CAT levels in hippocampus, and GPx enzyme activity levels in basal forebrain.

Vitamin E is one of non-enzymatic antioxidants has effect to scavenge reactive oxygen species and prevent deleterious effects in the cells that contribute to many

diseases (Hardie *et al.*, 1990). Vitamin E can prevent oxidative stress in the body and protect against many conditions of disease (I-Min Lee, 2005). Vitamin E elevated SOD in swimming exercise old rats. Mn-SOD in cerebral cortex increased in exercise middle-age and old rats. Cu-Zn-SOD in hippocampus increased in adult rats supplemented with vitamin E and swimming. Age-related and region-specific increased in protein carbonyl (PrC) content with decreased sulphydryl (P-SH) was demonstrated. Vitamin E could reduce PrC and advance oxidation protein products (AOPPs) in all ages, and appreciably in the hippocampus and cerebellum (Jolitha *et al.*, 2006). The brain is the most sensitive to oxidative damage. Age-related diseases of the brain such as Alzheimer's disease, that found to increased oxidative stress that generates free radicals and the antioxidant defense is the main mechanism to reduced oxidative stress that damage to the cells and tissues (Hardie *et al.*, 1990). In present study found that vitamin E supplement in the exercise group significantly increased antioxidant enzyme in some part of the brain both adult and middle-aged rats in this study that involving learning and memory in long term memory. The result suggests that swimming training for 8 weeks in the vitamin E supplement in rats can enhance antioxidant enzyme in both age may benefit to scavenging oxidative stress in the brain and can protect tissue and brain damage, that can reduced generation of neurodegenerative diseases such as Alzheimer's disease in aging.

5.3 Conclusion

The present study demonstrated the beneficial effect of taurine supplement in conjunction with exercise on the important parts of the brain involving learning and memory (basal forebrain, cerebral cortex, hippocampus and striatum) by causing

reduction of the end product of lipid peroxidation (such as MDA) and oxidative stress induced by H_2O_2, and enhancement of antioxidant enzyme activities (SOD, CAT, and GPx) in these brain areas in adult and middle-aged rats. Taurine supplement in conjunction with exercise can prevent oxidative damage in adult rat brains and may benefit in reduction of oxidative stress which is contributed to age-related neurodegenerative diseases.

The possible mechanism of taurine supplement in conjunction with exercise on antioxidant enzyme activities by scavenging reactive oxygen species in both adult and middle-aged rat brains can be explained in Figure 51.

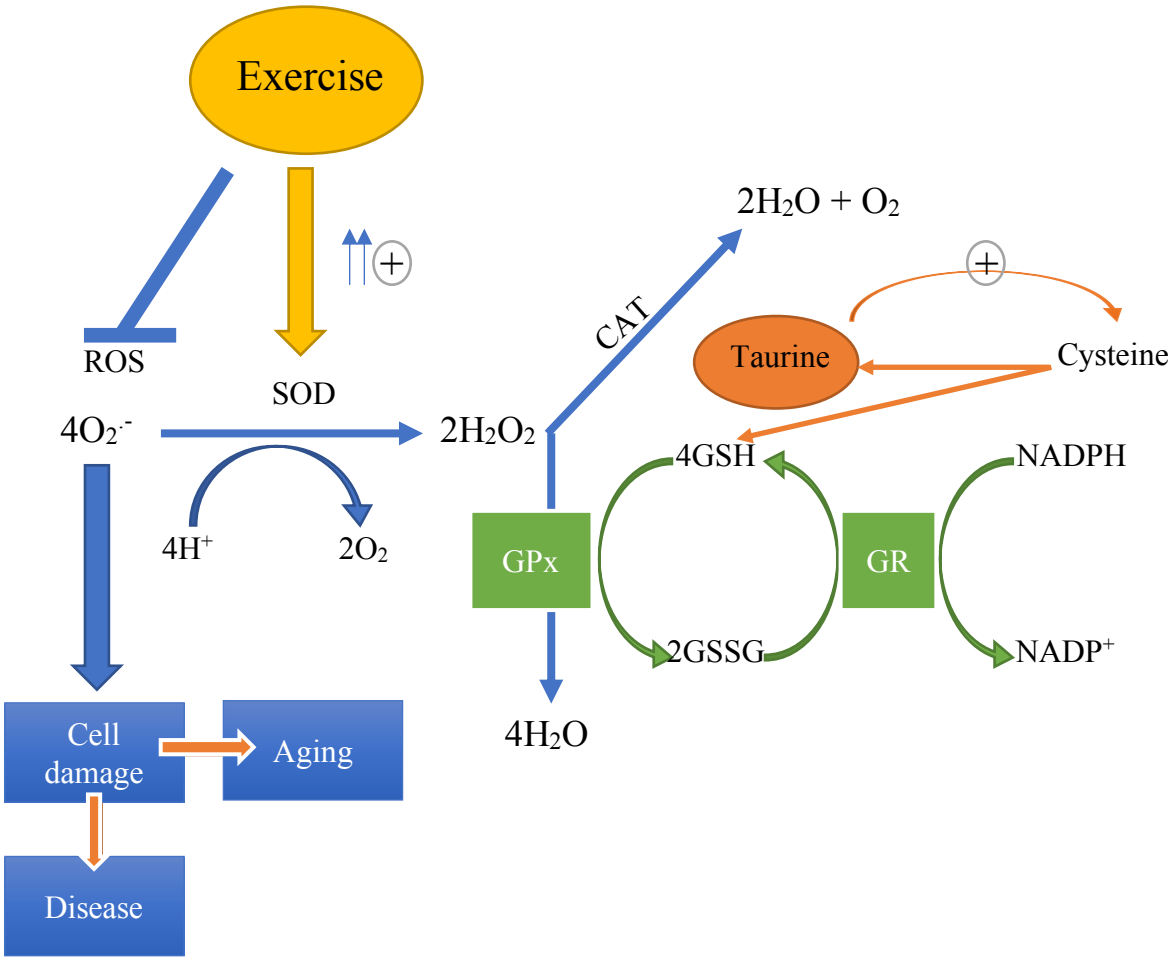

Figure 47 The possible mechanism of taurine supplement in conjunction with exercise on antioxidant enzyme activity by scavenging reactive oxygen species in both adult and middle-aged rat brains.

(ROS = reactive oxygen species, $O_2^{\cdot-}$ = Superoxide anion, SOD = superoxide dismutase, H^+ = ,O_2 = oxygen, CAT = catalase, GPx = glutathione peroxidase, H_2O_2 = hydrogen peroxide, GSH = glutathione, GR = glutathione reductase, GSSG = oxidized glutathione, NADPH = Dihydronicotinamide-adenine dinucleotide phosphate, $NADP^+$ = nicotinamide adenine dinucleotide phosphate).

REFERENCES

REFERENCES

Nandhini, A., Thirunavukkarasu, V., Ravichandran, M., and Anuradha, C. (2005). Effect of taurine on biomarkers of oxidative stress in tissues of fructose-fed insulin-resistant rats. **Singapore Medical Journal**. 42(2): 82.

Abhijit S., Tripathi S. J., Bhagya, V., Shankaranarayana Rao, B. S., Subramanyam, M. V., and Asha Devi, S. (2018). Antioxidant action of grape seed polyphenols and aerobic exercise in improving neuronal number in the hippocampus is associated with decrease in lipid peroxidation and hydrogen peroxide in adult and middle-aged rats. **Experimental Gerontology Journal**. 101: 101-112.

Adedara, I. A., Abolaji, A. O., Idris, U. F., Olabiyi, B. F., Onibiyo, E. M., Ojuade, T. D., and Farombi, E. O. (2017). Neuroprotective influence of taurine on fluoride-induced biochemical and behavioral deficits in rats. **Chemico-Biological Interactions**. 261: 1-10.

Adedara, I. A., Olabiyi, B. F., Ojuade, T. D., Idris, U. F., Onibiyo, E. M., and Farombi, E. O. (2017). Taurine reverses sodium fluoride-mediated increase in inflammation, caspase-3 activity, and oxidative damage along the brain–pituitary–gonadal axis in male rats. **Canadian Journal of Physiology and Pharmacology**. 95(9): 1019-1029.

Afanas'ev, I. (2010). Signaling and Damaging Functions of Free Radicals in Aging—Free Radical Theory, Hormesis, and TOR. **Aging and Disease**. 1(2): 75-88.

Afshar, R., Shegarfy, L., Shavandi, N., and Sanavi, S. (2010). Effects of aerobic exercise and resistance training on lipid profiles and infl ammation status in patients on maintenance hemodialysis. **Indian Journal of Nephrology**. 20(4): 185-189.

Alhumaidha, K. A., Saleh, D. O., Fattah, M. A. A. E., El-Eraky, W. I., and Moawad, H. (2016). Cardiorenal protective effect of taurine against cyclophosphamide-induced toxicity in albino rats. **Canadian Journal of Physiology and Pharmacology**. 94(2): 131-139.

Almeida V.D., Monteiro, M. G., Oliveira, M. G. M., Pomarico, A. C., Bueno, O. F. A., and Silva-Fernandes, M. E. d. (1994). Long-lasting effects of chronic ethanol administration on the activity of antioxidant enzymes. **Journal of Biochemical Toxicology**. 9(3): 141-143.

Alyea, E. P., Jr., H. H. P., and Durham, N. C. (1958). Renal response to exercise-urinary findings. **Journal of the American Medical Association**. 167(7): 807-813.

Andersen, J. K. (2004). Oxidative stress in neurodegeneration: cause or consequence? **Journal of Natural Medicines**. 10 18-25.

Anand, P., Rajakumer, D., Mathew Jeraud, Jhon, A., William Felix, and Balasubramanian, T. (2011). Effects of taurine on glutathione peroxidase, glutathione reductase and reduced glutathione levels in rats. **Pakistan Journal of Biological Sciences**. : 1-7.

Andreollo, N., Santos, E., Araújo, M., and Lopes, L. (2012). Rat's age versus human's age: what is the relationship? **Arquivos Brasileiros de Cirurgia Digestiva**. 25(1): 49-51.

Anitha Nandhini, A., Thirunavukkarasu, V., and Anuradha, C. (2005). Taurine modifies insulin signaling enzymes in the fructose-fed insulin resistant rats. **Diabetes & Metabolism**. 31(4): 337-344.

Asha, K. K., and Devadasan, K. (2013). Protective effect of taurine on the mitochondria of albino rats induced with fulminant hepatic failure. **Biomedicine & Preventive Nutrition**. 3(3): 279-283.

Azizbeigi, K., Stannard, S. R., Atashak, S., and Mosalman Haghighi, M. (2014). Antioxidant enzymes and oxidative stress adaptation to exercise training: Comparison of endurance, resistance, and concurrent training in untrained males. **Journal of Exercise Science & Fitness**. 12(1): 1-6.

Bannister, J. V., Bannister, W. H., and Rotilio, G. (1987). Aspects of the Structure, Function, and Applications of Superoxide Dismutas. **Critical Reviews in Biochemistry**. 22(2): 111-180.

Barnes, J. N. (2015). Exercise, cognitive function, and aging. **Advances in Physiology Education**. 39(2): 55-62.

Beckman, K. B., and Ames, B. N. (1998). The Free Radical Theory of Aging Matures. **The American Physiological Society**. 78(2): 547-582.

Bellinghieri, G., Savica, V., and Santoro, D. (2008). Renal Alterations During Exercise. **Journal of Renal Nutrition**. 18(1): 158-164.

Bellinghieri, G., Savica, V., and Santoro, D. (2009). Renal Alterations During Exercise. **Journal of Renal Nutrition**. 18(1): 158-164.

Berlett, B. S., and Stadtman, E. R. (1997). Protein Oxidation in Aging, Disease, and Oxidative Stress. **The Journal of Biological Chemistry**. 272(33): 20313-20316.

Bijeh, N., and Farahati, S. (2013). The Effect of Six Months of Aerobic training on Renal Function Markers in Untrained Middle-Aged Women. **International Journal of Sport Studies**. 3(2): 218-224.

Bjorksten, J., and Tenhu, H. (1990). The crossinking theory of aging - added evidence. **Experimental Gerontology**. 25: 91-95.

Blazer, V. S., and Wolke, R. E. (1984). The effects of α-tocopherol on the immune response and non-specific resistance factors of rainbow trout (Salmo gairdneri Richardson). **Aquaculture**. 37(1): 1-9.

Blr, C. M., Pasyk, S., and Leon, A. S. (1970). Interaction of Age and Exercise on Organ and Cellular Development. **The American Journal of Pathology**. 58(2): 185-199.

Bo Su, Xinglong Wang, Akihiko Nunomura, Paula I. Moreira, Hyoung-gon Lee, George Perry, Mark A. Smith, and Zhu, X. (2008). Oxidative Stress Signaling in Alzheimer's Disease. **NIH Public Access Author Manuscript**. 5(6): 525-532.

Broderick, T. L., Midaoui, A. E., Chiasson, J. L., Wang, D., Jankowski, M., and Gutkowska, J. (2011). The effects of exercise training on γ-butyrobetaine hydroxylase and novel organic cation transporter-2 gene expression in the rat. **Applied Physiology, Nutrition, and Metabolism**. 36: 781-789.

Brys, K., Vanfleteren, J. R., and Braeckman, B. P. (2007). Testing the rate-of-living/oxidative damage theory of aging in the nematode model Caenorhabditis elegans. **Experimental Gerontology**. 42(9): 845-851.

Cherra, S. J., 3rd, and Chu, C. T. (2008). Autophagy in neuroprotection and neurodegeneration: A question of balance. **Future Neurol**. 3(3): 309-323.

In middle-aged sedentary and regular swimming exercise rats, taurine alone and exercise alone had no effect on AST and ALT levels. Taurine supplement in conjunction with exercise had no effect on AST levels, but significantly increased ALT levels in middle-aged rats. Previous study found that swimming for 12 weeks and running exercise for 8 weeks could reduce AST and ALT levels in aging rat (Hajighasem *et al*., 2018; Huang *et al*., 2013). Taurine reduced the hepatotoxicity of pyrazinamide in rats treatment with 1 ml/kg of 500 mg/kg pyrazinamide for 4 weeks (Taziki *et al.*, 2018). Taurine supplement with iron could prevent iron-overloaded in liver mice (Zhang *et al*., 2014).

Taurine supplement alone had no effect on BUN and creatinine levels in sedentary middle-aged rats. Taurine supplement alone significantly reduced BUN levels, but had no effect on creatinine levels in exercise middle-aged rats. Exercise alone significantly increased BUN levels, but not creatinine levels, in middle-aged rats. Taurine supplement in conjunction with exercise had no effect on BUN and creatinine levels in middle-rats. Previous study found that, aerobic exercise could reduce creatinine in aging patients (Afshar *et al*., 2010). Aerobic training in middle-aged women showed no change in BUN and creatinine levels after training for 6 months (Bijeh and Farahati, 2013). In human, short term effect of stetanous exercise in marathon runners and other endurance athletes for 4 hours could elevate BUN and creatinine as a result of dehydration and decreased renal perfusion (Foran *et al*., 2003). Urea is excreted by the kidneys. Urea is filtered by the glomerular capillaries, and enter to the renal tubule. Some part of urea is passively reabsorbed by diffusion, but the remainder is excreted in the urine. High BUN levels following exercise may be due to decreased glomerular filtration and excretion in the urine or diminished reabsorption in

Chesney, R. W., Patters, A. B., and Han, X. (2012). Taurine and the kidneys. **Taurine in Health and Disease**. : 75-99.

Christophe Fleury, Bernard Mignotte, and Vayssière, J.-L. (2002). Mitochondrial reactive oxygen species in cell death signaling. **Biochimie**. 84: 131-141.

Clarke, H. H., Ed. (1977). Exercise and aging. **Physical Fitness Research Digest**. 7(2).

Cornelius, E. A. (1972). Increased Incidence of Lymphomas in Thymectomized Mice - Evidence for an Immunological Theory of Aging **Specialia**. : 459.

Cotman, C. W., and Berchtold, N. C. (2002). Exercise: a behavioral intervention to enhance brain health and plasticity. **Trends In Neurosciences Journal**. 25: 295-301.

Cotman, C. W., Berchtold, N. C., and Christie, L. A. (2007). Exercise builds brain health: key roles of growth factor cascades and inflammation. **Trends In Neurosciences Journal**. 30(9): 464-472.

Cotman, C. W., and Engesser-Cesar, C. (2002). Exercise Enhances and Protects Brain Function. **Exercise and Sport Sciences**. 30(2): 75-79.

Denham Harman, M. D., Ph.D. (1956). Aging: A Theory based on free radical and radiation chemistry. **Forest Park Foundation to the Journal of Gerontology**. : 298-300.

Devi, S. A. (2009). Aging brain: prevention of oxidative stress by vitamin E and exercise. **The Scientific World Journal**. 9: 366-372.

Devi, S. A., and Kiran, T. R. (2004). Regional responses in antioxidant system to exercise training and dietary Vitamin E in aging rat brain. **Neurobiology of Aging**. 25(4): 501-508.

Doorn, J. A., and Petersen, D. R. (2003). Covalent adduction of nucleophilic amino acids by 4-hydroxynonenal and 4-oxononenal. **Chemico-Biological Interactions**. 143-144: 93-100.

Egbuna, C. (2017). Oxidative Stress and Nutrition. **Tropical Journal of Applied Natural Sciences**. 2(1): 110-116.

Erdem, A., Gundogan, N. U., Usubutun, A., Kilinc, K., Erdem, S. R., Kara, A., and Bozkurt, A. (2000). The protective effect of taurine against gentamicin-induced acute tubular necrosis in rats. **Nephrology Dialysis Transplantation**. 15(8): 1175-1182.

Esiri, M. M. (2007). Ageing and the brain. **The Journal of Pathology**. 211(2): 181-187.

Ezekiel, I., Ayo, J. O., Tanko, Y., and Magaji, R. A. (2015). Effect of Taurine on Acute Restraint Stress Induced Biochemical Alterations in Wistar Rats. **International Journal of Scientific & Engineering Research**. 6(5): 958-966.

Fiebig, R., Gore, M. T., Chandwaney, R., Leeuwenburgh, C., and Ji, L. L. (1996). Alteration of myocardial antioxidant enzyme activity and glutathione content with aging and exercise training. **Age**. 19(3): 83-89.

Foda, D. S., Farrag, E. K., Metwally, N. S., Maghraby, A. S., Farrag, A. R. H., and Rawi, S. M. (2016). Protective and therapeutic impact of taurine on some biochemical, immunological and histological parameters in diabetic rats. **Journal of Applied Pharmaceutical Science**. 6(10): 45-54.

Foos, T. M., and Wu, J.-Y. (2001). The Role of Taurine in the Central Nervous System and the Modulation of Intracellular Calcium Homeostasis. **Neurochemical Research**. 27: 21-26.

Foran, S. E., Lewandrowski, K. B., and Kratz, A. (2003). Effects of exercise on laboratory test results. **Laboratory Medicine**. 34(10): 736-742.

Franconi, F., Leo, M. A. S. D., Bennardini, F., and Ghirlanda, G. (2004). Is Taurine Beneficial in Reducing Risk Factors for Diabetes Mellitus? **Neurochemical Research**. 29(1): 143-150.

Frosini, M., Sesti, C., Saponara, S., Ricci, L., Valoti, M., Palmi, M., Machetti, F., and Sgaragli, G. (2003). A specific taurine recognition site in the rabbit brain is responsible for taurine effects on thermoregulation. **British Journal of Pharmacology**. 139(3): 487-494.

Gebara, E., Udry, F., Sultan, S., and Toni, N. (2015). Taurine increases hippocampal neurogenesis in aging mice. **Stem Cell Research**. 14(3): 369-379.

Gimenes, C., Gimenes, R., Rosa, C. M., Xavier, N. P., Campos, D. H. S., Fernandes, A. A. H., Cezar, M. D. M., Guirado, G. N., Cicogna, A. C., Takamoto, A. H. R., Okoshi, M. P., and Okoshi, K. (2015). Low Intensity Physical Exercise Attenuates Cardiac Remodeling and Myocardial Oxidative Stress and Dysfunction in Diabetic Rats. **Journal of Diabetes Research**. : 1-10.

Gollnick, P. D., Struck, P. J., and Bogy, T. P. (1967). Lactic dehydrogenase activities of rat heart and skeletal muscle after exercise and training. **Journal of Applied Physiology**. 22(4): 623-627.

Gregory, R. L. (1994). The Biological Role and Clinical Implications of IgA. **Laboratory Medicine**. 25(11): 724-728.

Grotto, D., Maria, L. S., Valentini, J., Paniz, C., Schmitt, G., Garcia, S. C., Pomblum, V. J., Rocha, J. B. T., and Farina, M. (2009). Importance of the lipid peroxidation biomarkers and methodological aspects for malondialdehyde quantification. **Química Nova**. 32(1): 169-174.

Gueraud, F., Atalay, M., Bresgen, N., Cipak, A., Eckl, P. M., Huc, L., Jouanin, I., Siems, W., and Uchida, K. (2010). Chemistry and biochemistry of lipid peroxidation products. **Free Radical Research Journal**. 44(10): 1098-1124.

Guerra-Araiza, C., Alvarez-Mejia, A. L., Sanchez-Torres, S., Farfan-Garcia, E., Mondragon-Lozano, R., Pinto-Almazan, R., and Salgado-Ceballos, H. (2013). Effect of natural exogenous antioxidants on aging and on neurodegenerative diseases. **Free Radical Research Journal**. 47(6-7): 451-462.

Gutteridge, J. M. C. (1995). Lipid peroxidation and antioxidants as biomarkers of tissue damage. **Clinical Chemistry Journals**. 14(12): 1819-1828

Hajighasem, A., Farzanegi, P., Mazaheri, Z., Naghizadeh, M., and Salehi, G. (2018). Effects of resveratrol, exercises and their combination on Farnesoid X receptor, Liver X receptor and Sirtuin 1 gene expression and apoptosis in the liver of elderly rats with nonalcoholic fatty liver. **Peer Journals**. : 1-11.

Halliwell, and Barry. (2001). Role of Free Radicals in the Neurodegenerative Diseases. **Drugs & Aging**. 18(9): 685-716.

Hara, M., Iigo, M., Ohtani-Kaneko, R., Nakamura, N., Suzuki, T., Reiter, R. J., and Hirata, K. (1997). Administration of Melatonin and Related Indoles Prevents Exercise-Induced Cellular Oxidative Changes in Rats. **Neurosignals**. 6(2): 90-100.

Harada, H., Cusack, B. J., Olson, R. D., Stroo, W., Azuma, J., Hamaguchi, T., and Schaffer, S. W. (1990). Taurine deficiency and doxorubicin: interaction with the cardiac sarcolemmal calcium pump. **Biochemical Pharmacology**. 39(4): 745 - 751.

Harber, M. P., Konopka, A. R., Undem, M. K., Hinkley, J. M., Minchev, K., Kaminsky, L. A., Trappe, T. A., and Trappe, S. (2012). Aerobic exercise training induces

skeletal muscle hypertrophy and age dependent adaptations in myofiber function in young and older men **Journal of Applied Physiology**. 113(9): 1495 -1504.

Hardie, L. J., Fletcher, T. C., and Secombes, C. J. (1990). The effect of vitamin E on the immune response of the Atlantic salmon (Salmo salar L.). **Aquaculture**. 87(1): 1-13.

Harman, D. (1956). Aging: Theory based on free radical and radiation chemistry. **Journal of Gerontology**. 11: 298-300.

Head, E. (2009). Oxidative damage and cognitive dysfunction: antioxidant treatments to promote healthy brain aging. **Neurochemical Research Journal**. 34(4): 670-678.

Heemst, D. v. (2010). Insulin, IGF-1 and longevity. **Aging and Disease**. 1(2): 147-157.

Heidari, R., Jamshidzadeha, A., Niknahada, H., Mardani, E., Ommati, M. M., Azarpira, N., Khodaei, F., Zarei, A., Ayarzadehb, M., Mousavi, S., Abdoli, N., Yeganehf, B. S., Saeedi, A., and Najibi, A. (2016). Effect of taurine on chronic and acute liver injury: Focus on blood and brain ammonia. **Toxicology Reports**. 3: 870-879.

Herculano-Houzel, S. (2011). Scaling of brain metabolism with a fixed energy budget per neuron: implications for neuronal activity, plasticity and evolution. **PLoS One Journal**. 6(3): 17514.

Higo, S., Miyata, S., Jiang, Q. Y., Kitazawa, R., Kitazawa, S., and Kasuga, M. (2008). Taurine administration after appearance of proteinuria retards progression of diabetic nephropathy in rats. **Kobe Journal of Medical Sciences**. 54(1): 35-45.

Huang, C.-C., Chiang, W.-D., Huang, W.-C., Huang, C.-Y., Hsu, M.-C., and Lin, W.-T. (2013). Hepatoprotective Effects of Swimming Exercise against d-

Galactose-Induced Senescence Rat Model. **Evidence-Based Complementary and Alternative Medicine**. 2013: 1-9.

Huang, C.-C., Wang, T., Tung, Y.-T., and Lin, W.-T. (2016). Effect of Exercise Training on Skeletal Muscle SIRT1 and PGC-1α Expression Levels in Rats of Different Age. **International Journal of Medical Sciences**. 13(4): 260-270.

Hulbert, A. J., Pamplona, R., Buffenstein, R., and Buttemer, W. A. (2007). Life and death: metabolic rate, membrane composition, and life span of animals. **Physiological Reviews**. 87(4): 1175-1213.

Huxtable, R. J. (1992). Physiological action of taurine. **The American Physiological Society**. 72(1): 101-163.

I-Min Lee, N. R. C., J. Michael Gaziano, David Gordon, Paul M Ridker, JoAnn E. Manson, Charles H. Hennekens, and Julie E. Buring. (2005). Vitamin E in the Primary Prevention of Cardiovascular Disease and Cancer. **American Medical Association**. 294(1).

Jang-Yen Wu, and Prentice, H. (2010). Role of taurine in the central nervous system. **Journal of Biomedical Science**. 17: 1-6.

Ji, L. L. (1993). Antioxidant enzyme response to exercise and aging. **Medicine & Science in Sports & Exercise**. 25(2): 225-231.

Johnson, F., and Giulivi, C. (2005). Superoxide dismutases and their impact upon human health. **Molecular Aspects of Medicine**. 26(4-5): 340-352.

Johnson, R. A., and Mitchell, G. S. (2003). Exercise-induced changes in hippocampal brain-derived neurotrophic factor and neurotrophin-3: effects of rat strain. **Brain Research**. 983(1): 108-114.

Jolitha, A. B., Subramanyam, M. V., and Asha Devi, S. (2006). Modification by vitamin E and exercise of oxidative stress in regions of aging rat brain: studies on

superoxide dismutase isoenzymes and protein oxidation status. **Experimental Gerontology Journal**. 41(8): 753-763.

Kang, J. H. (2013). Acrolein, A Reactive Product of Lipid Peroxidation, Induces Oxidative Modification of Cytochrome c. **Bulletin of the Korean Chemical Society**. 34(11): 3295-3300.

Kang, Y.-S. (2000). Taurine transport mechanism through the blood-brain barrier in spontaneously hypertensive rats **Advances in Experimental Medicine and Biology**. 483: 321-324.

Kaplan, B., Karabay, G., Zag˘yapan, R. D., zer, C. O., Sayan, H., and Duyar, I. (2004). Effects of taurine in glucose and taurine administration. **Amino Acids**. 27(3-4): 327-333.

Kim, J.-S., Lee, Y.-H., Kim, J.-C., Ko, Y.-H. K., Yoon, C.-S., and Yi, H.-K. (2014). Effect of Exercise Training of Different Intensities on Anti-Inflammatory Reaction in Streptozotocin-Induced Diabetic Rats. **Biology of Sport**. 33(1): 73-79.

Kim, K. S., Oh, D. H., Kim, J. Y., Lee, B. G., You, J. S., Chang, K. J., Chung, H., Yoo, M. C., Yang, H.-I., Kang, J.-H., Hwang, Y. C., Ahn, K. J., Chung, H.-Y., and Jeong, I.-K. (2012). Taurine ameliorates hyperglycemia and dyslipidemia by reducing insulin resistance and leptin level in Otsuka Long-Evans Tokushima fatty (OLETF) rats with long-term diabetes. **Experimental and Molecular Medicine**. 44(11): 665-673.

Kirkwood, T. B. L. (2003). The most pressing problem of our age. **British Medical Journal**. 326: 1297-1299.

Kozlowski, S., Szczepanska, E., and Zielinski, A. (1967). The hypothalamo-hypophyseal antidiuretic system in physical exercises. **Archives Internationales de Physiologie et de Biochimie**. 75(2): 218-228.

Kumar, R. (2009). Role of naturally occurring osmolytes in protein folding and stability. **Archives of Biochemistry and Biophysics**. 491(1): 1-6.

Lampson, W. G., Kramer, J. H., and Schaffer, S. W. (1983). Potentiation of the actions of insulin by taurine. **Canadian Journal of Physiology and Pharmacology**. 61(5): 457-463.

Louzada, P. R., Paula Lima, A. C., Mendonca-Silva, D. L., Noel, F., De Mello, F. G., and Ferreira, S. T. (2004). Taurine prevents the neurotoxicity of beta-amyloid and glutamate receptor agonists: activation of GABA receptors and possible implications for Alzheimer's disease and other neurological disorders. **Faseb Journal**. 18(3): 511-518.

Lu, Y., Khoo, T. J., and Wiart, C. (2014). Antioxidant Activity Determination of Citronellal and Crude Extracts of Cymbopogon citratus by 3 Different Methods. **Journal of Pharmacy and Pharmacology**. 05(04): 395-400.

Mahalakshm, K., Pushpakiran, G., and Anuradha, C. V. (2003). Taurine prevents acrylonitrile-induced oxidative stress in rat brain. **Polish Journal of Pharmacology**. 55: 1037-1043.

Manabe, S., Kuroda, I., Okada, K., Morishima, M., Okamoto, M., Harada, N., Takahashi, A., Sakai, K., and Nakaya, Y. (2003). Decreased Blood Levels of Lactic Acid and Urinary Excretion of 3-Methylhistidine after Exercise by Chronic Taurine Treatment in Rats. **Journal of Nutritional Science and Vitaminology**. 49: 375-380.

Marosi, K., Bori, Z., Hart, N., Sarga, L., Koltai, E., Radak, Z., and Nyakas, C. (2012). Long-term exercise treatment reduces oxidative stress in the hippocampus of aging rats. **Neuroscience**. 226: 21-28.

Martin, L. J. (2008). DNA damage and repair: relevance to mechanisms of neurodegeneration. **Journal of Neuropathology and Experimental Neurology**. 67(5): 377-387.

Mattson, M. P., and Magnus, T. (2006). Ageing and neuronal vulnerability. **Journal of Neuroscience**. 7: 278.

Maxwell, L. C., Enwemeka, C. S., and Fernandesd, G. (1992). Effects of exercise and food restriction on rat skeletal muscles. **Tissue and Cell**. 4(24): 491-498.

Mazzeo, R. S., and Horvath, S. M. (1986). Effects of training on weight, food intake, and body composition in aging rats. **The American Journal of Clinical Nutrition**. 44: 732-738.

McCarter, R. J. M. (2000). Caloric restriction, exercise and aging. **Handbook of Oxidants and Antioxidants in Exercise**. : 797-829.

McIntosh, L. J., and Sapolsky, R. M. (1996). Glucocorticoids Increase the Accumulation of Reactive Oxygen Species and Enhance Adriamycin-Induced Toxicity in Neuronal Culture. **Experimenttal Neurology**. 141(0154): 201-206.

Migliore, L., and Coppede, F. (2009). Environmental-induced oxidative stress in neurodegenerative disorders and aging. **Mutation Research**. 674(1-2): 73-84.

Mizushima, S., Nara, Y., Sawamura, M., and Yamori, Y. (1996). Effects of Oral Taurine Supplementation on Lipids and Sympathetic Nerve Tone. **Taurine 2**. : 615-622.

Mladen Davidovic, Goran Sevo, Petar Svorcan, Dragoslav P. Milosevic, Nebojsa Despotovic, and Erceg, P. (2010). Old age as a privilege of the "selfish ones". **Aging and Disease**. 1(2): 139-146.

Muller, F. L., Lustgarten, M. S., Jang, Y., Richardson, A., and Van Remmen, H. (2007). Trends in oxidative aging theories. **Free Radical Biology and Medicine**. 43(4): 477-503.

Nakaya, Y., Minami, A., Harada, N., Sakamoto, S., Niwa, Y., and Ohnaka, M. (2000). Taurine improves insulin sensitivity in the Otsuka Long-Evans Tokushima Fatty rat, a model of spontaneous type 2 diabetes. **The American Journal of Clinical Nutrition**. 71(1): 54-58.

Nayanatara, A. K., Nagaraja, H. S., and Anupama, B. K. (2005). The effect of repeated swimming stress on organ weights and lipid peroxidation in rats. **Thai Journal of Physiological Sciences**. 18(1): 3-9

Niu, X., Zheng, S., Liu, H., and Li, S. (2018). Protective effects of taurine against inflammation, apoptosis, and oxidative stress in brain injury. **Molecular Medicine Reports**. 18: 4516-4522.

Nonato, L. F., Rocha-Vieira, E., Tossige-Gomes, R., Soares, A. A., Soares, B. A., Freitas, D. A., Oliveira, M. X., Mendonc, V. A., Lacerda, A. C., Massensini, A. R., and Leite, H. R. (2016). Swimming training attenuates oxidative damage and increases enzymatic but not non-enzymatic antioxidant defenses in the rat brain. **Brazilian Journal of Medical and Biological Research**. 49(10): 5310-5315.

Oliveira, A. R. d., Silva, F. v. S. d., Bortolin, R. H., Marques, D. f. E. d. S., Ramos, G. V., Marqueti, R. C., Silva, N. B. d., Medeiros, K. C. d. P., Corrêa, M. r. A., Paulo, J., Lima, M. S., Rezende, A. A. d., Ackermann, P. W., Abreu, B. J., and

Vieira, W. H. r. d. B. (2019). Effect of photobiomodulation and exercise on early remodeling of the Achilles tendon in streptozotocin-induced diabetic rats. **Plos One**. 14(2): 1-16.

Owoeye, O., Adedara, I. A., and Farombi, E. O. (2018). Pretreatment with taurine prevented brain injury and exploratory behaviour associated with administration of anticancer drug cisplatin in rats. **Biomedicine & Pharmacotherapy**. 102: 375-384.

Ozkaya, Y. G., Agar, A., Yargicoglu, P., Hacioglu, G., Bilmen-Sarikcioglu, S., Ozen, I., and Aliciguzel, Y. (2002). The effect of exercise on brain antioxidant status of diabetic rats. **Diabetes Metab (Paris)**. 28: 377-384.

Pow, D. V., Sullivan, R., Reye, P., and Hermanussen, S. (2002). Localization of taurine transporters, taurine, and 3H taurine accumulation in the rat retina, pituitary, and brain. **Glia**. 37(2): 153-168.

Pushpakiran, G., Mahalakshmi, K., and Anuradha, C. V. (2004a). Protective effects of taurine on glutathione and glutathione-dependent enzymes in ethanol-fed rats. **Pharmazie**. 59(11): 869-872.

Pushpakiran, G., Mahalakshmi, K., and Anuradha, C. V. (2004b). Taurine restores ethanol-induced depletion of antioxidants and attenuates oxidative stress in rat tissues. **Amino Acids**. 27(1): 91-96.

Qiao, M., Liu, P., Ren, X., Feng, T., and Zhang, Z. (2015). Potential protection of taurine on antioxidant system and ATPase in brain and blood of rats exposed to aluminum. **Biotechnology Letters Journal**. 37(8): 1579-1584.

Radak, Z., Chung, H. Y., and Goto, S. (2005). Exercise and hormesis: oxidative stress-related adaptation for successful aging. **Biogerontology**. 6(1): 71-75.

Radak, Z., Hart, N., Sarga, L., Koltai, E., Atalay, M., Ohno, H., and Boldogh, I. (2010). Exercise plays a preventive role against Alzheimer's disease. **Journal of Alzheimers Disease**. 20(3): 777-783.

Radak, Z., Marton, O., Nagy, E., Koltai, E., and Goto, S. (2013). The complex role of physical exercise and reactive oxygen species on brain. **Journal of Sport and Health Science**. 2(2): 87-93.

Radak, Z., Toldy, A., Szabo, Z., Siamilis, S., Nyakas, C., Silye, G., Jakus, J., and Goto, S. (2006). The effects of training and detraining on memory, neurotrophins and oxidative stress markers in rat brain. **Neurochemistry International**. 49(4): 387-392.

Raffaella Molteni, Jun-Qi Zheng, Z. Y., Fernando Go´mez-Pinilla, and Twiss, J. L. (2004). Voluntary exercise increases axonal regeneration from sensory neurons. **Proceedings of the National Academy of Sciences**. 101: 8473-8478.

Ravikiran, T., Sowbhagya, R., Anupama, S. K., Anand, S., and Bhagyalakshmi, D. (2016). Age-related changes in the brain antioxidant status: modulation by dietary supplementation of Decalepis hamiltonii and physical exercise. **Molecular and Cellular Biochemistry**. 419(1-2): 103-113.

Rebeca Gerschman, Daniel L. Gilbert, Sylvanus W. Nye, Peter Dwyer, and .Fenn, W. (1954). Oxygen Poisoning and X-irradiation: A Mechanism in Common. **Science Journal**. 119: 623-626.

Rosemberg, D. B., da Rocha, R. F., Rico, E. P., Zanotto-Filho, A., Dias, R. D., Bogo, M. R., Bonan, C. D., Moreira, J. C. F., Klamt, F., and Souza, D. O. (2010). Taurine prevents enhancement of acetylcholinesterase activity induced by acute ethanol exposure and decreases the level of markers of oxidative stress in zebrafish brain. **Neuroscience**. 171(3): 683-692.

Rudow, G., O'Brien, R., Savonenko, A. V., Resnick, S. M., Zonderman, A. B., Pletnikova, O., Marsh, L., Dawson, T. M., Crain, B. J., West, M. J., and Troncoso, J. C. (2008). Morphometry of the human substantia nigra in ageing and Parkinson's disease. **Acta Neuropathologica Journal**. 115(4): 461-470.

Rybak, L. P., Somani, S. M., and Ravi, R. (1995). Effect of exercise training on antioxidant system in brain regions of rat. **Pharmacology Biochemistry and Behavior**. 50(4): 635-639.

Schaffer, S., Takahashi, K., and Azuma, J. (2000). Role of osmoregulation in the actions of taurine. **Amino Acids**. 19: 527-546.

Saad, S. Y., and Al-Rikabi, A. C. (2002). Protection Effects of Taurine Supplementation against Cisplatin-Induced Nephrotoxicity in Rats **Chemotherapy**. 48(1): 42-48.

Sakamoto, K., and Grunewald, K. K. (1986). Beneficial effects of exercise on growth of rats during intermittent fasting. **Natrition and Physical Activity**. 390-395.

Saransaari, P. and Oja, S. S. (2000). Taurine and neural cell damage. **Journal of Amino Acids**. 19: 509-526.

Samuel A. Goldblith, and Proctor, B. E. (1950). Photometric determination of catalase activity. **Biological Chemistry**. (187): 705-709.

Schaffer, S. W., Jong, C. J., Ito, T., and Azuma, J. (2012). Effect of taurine on ischemia-reperfusion injury. **Amino Acids**. 46(1): 21-30.

Schreckenberg, R., Horn, A.-M., Rebelo, R. M. d. C., Simsekyilmaz, S., Niemann, B., Li, L., Rohrbacn, S., and Schlüter, K.-D. (2017). Effects of 6-months' Exercise on Cardiac Function, Structure and Metabolism in Female Hypertensive Rats–The Decisive Role of Lysyl Oxidase and Collagen III. **Frontiers in Physiology**. 8: 1-13.

Shamsoddini, A., Sobhani, V., Chehreh, M. E. G., Alavian, S. M., and Zaree, A. (2015). Effect of Aerobic and Resistance Exercise Training on Liver Enzymes and Hepatic Fat in Iranian Men With Nonalcoholic Fatty Liver Disease. **Hepatitis Monthly**. 15(10): 1-7.

Shibamoto, T. (2006). Analytical methods for trace levels of reactive carbonyl compounds formed in lipid peroxidation systems. **Journal of Pharmaceutical and Biomedical Analysis**. 41(1): 12-25.

Shinoda, M., Latour, M., and Lavoie, J.-M. (2002). Effects of physical training on body composition and organ weights in ovariectomized and hyperestrogenic rats. **International Journal of Obesity**. 26(3): 335-343.

Sigma-Aldrich. Glutathione Peroxidase Cellular Activity Assay Kit. **Sigma-Aldrich chemical**.

Sigma-Aldrich. SOD determination kit. **Sigma-Aldrich chemical**.

Stanley J. Colcombe, Kirk I. Erickson, Naftali Raz, Andrew G. Webb, Neal J. Cohen, Edward McAuley, and Kramer, A. F. (2003). Aerobic Fitness Reduces Brain Tissue Loss in Aging Humans. **Journal of Gerontology**. 58(2): 176-180.

Stone, V., Kudo, K. Y., Marcelino, T. B., August, P. M., and Matté, C. (2014). Swimming exercise enhances the hippocampal antioxidant status of female Wistar rats. **Redox Report**. 20(3): 133-138.

Sun, M., Zhao, Y., Gu, Y., and Xu, C. (2012). Anti-inflammatory mechanism of taurine against ischemic stroke is related to down-regulation of PARP and NF-jB. **Amino Acids**. 42: 1735-1747.

Sun, M., Zhao, Y., Gu, Y., and Zhang, Y. (2015). Protective Effects of Taurine Against Closed Head Injury in Rats. **Jorunal of Neurotrauma**. 32(1): 66-74.

Suzuki M, Katamine S, and S., T. (1983). Exercise-induced enhancement of lipid peroxide metabolism in tissues and their transference into the brain in rat. **Journal of Nutritional Science and Vitaminology**. 29: 141-151.

Tai, H. C., and Schuman, E. M. (2008). Ubiquitin, the proteasome and protein degradation in neuronal function and dysfunction. **Nature Reviews Neuroscience Journal**. 9(11): 826-838.

Taziki, S., Khori, V., Jahanshahi, M., Seifi, A., Babakordi, F. B., and Nikmahzar, E. N. (2018). Protective role of taurine against hepatotoxicity induced by pyrazinamide in rats. **National Journal of Physiology, Pharmacy and Pharmacology**. 8(6): 824-828.

Trofin, F.-P., Ciobica, A., Cojocaru, D., Chirazi, M., Honceriu, C., Trofin, L., Serban, D., Timofte, D., Cojocaru, S. I., and Anton, E. (2014). Increased oxidative stress status in rat serum after five minutes treadmill exercise. **Central European Journal of Medicine**. 9(5): 722-728.

Uchida, K. (2003). 4-Hydroxy-2-nonenal: a product and mediator of oxidative stress. **Progress in Lipid Research**. 42(4): 318-343.

Van Praag, H., Shubert, T., Zhao, C., and Gage, F. H. (2005). Exercise enhances learning and hippocampal neurogenesis in aged mice. **Journal of Neuroscience**. 25(38): 8680-8685.

Vinge, E., Lindergård, B., Nilsson–Ehle, P., and Grubb, A. (1999). Relationships among serum cystatin C, serum creatinine, lean tissue mass and glomerular filtration rate in healthy adults. **Scandinavian Journal of Clinical and Laboratory Investigation**. 59(8): 587-592.

Wang, F. R., Dong, X. F., Tong, J. M., Zhang, X. M., Zhang, Q., and Wu, Y. Y. (2009). Effects of dietary taurine supplementation on growth performance and immune

status in growing Japanese quail (Coturnix coturnix japonica) **Poultry Science**. 88: 1394-1398.

Wang, Q., Fan, W., Cai, Y., Wu, Q., Mo, L., Huang, Z., and Huang, H. (2016). Protective effects of taurine in traumatic brain injury via mitochondria and cerebral blood flow. **Amino Acids**. 48(9): 2169-2177.

Wang, S. Y., Huang, W. C., Liu, C. C., Wang, M. F., Ho, C. S., Huang, W. P., Hou, C. C., Chuang, H. L., and Huang, C. C. (2012). Pumpkin (Cucurbita moschata) fruit extract improves physical fatigue and exercise performance in mice. **Molecules Journal**. 17(10): 11864-11876.

William, R., and Markesbery, M. (1999). The role of oxidative stress in Alzheimer disease. **Archives Of Neurology Journal**. 56.

Xiao, C., Giacca, A., and Lewis, G. F. (2008). Oral taurine but not N-acetylcysteine ameliorates NEFA-induced impairment in insulin sensitivity and beta cell-function in obese and overweight, non-diabetic men. **Diabetologia**. 51(1): 139-146.

Yatabe, Y., Miyakawa, S., Miyazaki, T., Matsuzaki, Y., and Ochiai, N. (2003). Effects of taurine administration in rat skeletal muscles on exercise. **Journal of Orthopaedic Science**. 8(3): 415-419.

Yu, J., and Kim, A. K. (2009). Effect of Taurine on Antioxidant Enzyme System in B16F10 Melanoma Cells. **Taurine 7**: 491- 499.

Yildirim, Z., Kilic, N., Ozer, C., Babul, A., Take, G.,and Erdogan, D. (2007). Effects of taurine in cellular responses to oxidative stress in young and middle-aged rat liver. **Annals of the New York Academy of Sciences**. 1100: 553-561.

Zecca, L., Youdim, M. B., Riederer, P., Connor, J. R., and Crichton, R. R. (2004). Iron, brain ageing and neurodegenerative disorders. **Nature Reviews Neuroscience Journal**. 5(11): 863-873.

Zelikovic, I., Stejskal-Lorenz, E., Lohstroh, P., Budreau, A., and Chesney, R. W. (1989). Anion dependence of taurine transport by rat renal brush-border membrane vesicles. **American Journal of Physiology-Renal Physiology**. 256(4): 646-655.

Zhang, Z., Liu, D., Yi, B., Liao, Z., Tang, L., Yin, D., and He, M. (2014). Taurine supplementation reduces oxidative stress and protects yhe liver in an iron-overload murine model. **Molecular Medicine Reports**. 10: 2255-2262.

Zhou, Y., Holmseth, S., Guo, C., Hassel, B., Hofner, G., Huitfeldt, H. S., Wanner, K. T., and Danbolt, N. C. (2012). Deletion of the gamma-aminobutyric acid transporter 2 (GAT2 and SLC6A13) gene in mice leads to changes in liver and brain taurine contents. **Journal of Biological Chemistry**. 287(42): 35733-35746.

Zsolt Radak, Maria Sasvari, Csaba Nyakas b Hideki Ohno, Takao Kaneko, Shoichi Tahara, and Goto, S. (2001). Single bout of exercise eliminates the immobilization-induced oxidative stress in rat brain. **Neurochemistry International**. 39: 33-38.

Zuhal Yildirim, and Kilic, N. (2011). Effects of Taurine and Age on Cerebellum Antioxidant Status and Oxidative Stress. **International Journal of Gerontology**. 5: 166-170.

APPENDICES

APPENDIX A

DETERMINATION OF PROTEIN

Reagents:

1. Solution A

- Copper sulphate	($CuSO_4.5H_2O$)	0.15 g.
- Sodium tartate	($Na_2C_4H_4O_6.2H_2O$)	0.30 g.
- Sodium azide		0.05 g.

Dissolve all chemicals in DDD water and make up to a final volume of 250 ml and store in refrigerator.

2. Solution B

- Sodium hydroxide (NaOH)	2.0 g.
- Sodium carbonate (Na_2CO_3)	8.0 g.
- Sodium dodecyl sulfate	1.0 g.

Dissolve these chemicals in DDD water, adjust to the final volume of 250 ml, and store at room temperature.

3. Solution C

Dilute the 2 N Folin-Ciocatteau reagent with DDD water (1:20). Store in an amber bottle at room temperature.

4. Standard protein solution

Dissolve bovine serum albumin (BSA) with DDD water to make up 0, 15.625, 31.25, 62.5, 125, 250, 500, and 1000 μg/ml BSA solution.

Procedures

1. Pipette 30 μl of samples and standards into triplicate wells of 96-well plates.

2. Freshly mix solution A and solution B (1:3), and add 100 μl of this mixed solution to each well of 96-well plates and incubate at room temperature for 60 minutes.

3. Add 150 μl of solution C into triplicate wells of 96-well plates.

Pipette the solution into each tube as follow:

	Sample (μl)	Standard (μl)	Blank (μl)
Standard	-	30	-
Sample (brain homogenates)	30	-	-
Solution A+B	100	100	100
Solution B	150	150	150
50 mM PB, pH 7.4	-	-	30

50 mM PB, pH 7.4 = 50 mM phosphate buffer (pH 7.4) containing 0.1 mM EDTA

4. Mix and incubate all mixtures at room temperature for 30 minutes.

5. Read optical density (O.D.) at 650 nm by Benchmark Plus Microplate Spectrophotometer.

Calculation

$\Delta A_{650\ nm}$ Standard = $A_{650\ nm}$ Standard - $A_{650\ nm}$ Blank

Plot the $\Delta A_{650\ nm}$ Standard against protein concentration on the standard graph.

$\Delta A_{650\ nm}$ Sample = $A_{650\ nm}$ Sample - $A_{650\ nm}$ Blank

Determine the mg protein from the standard curve.

APPENDIX B

DETERMINATION OF MALONDIALDEHYDE LEVEL

Reagents:

1. Solution A: TCA-TBA-HCl solution

- 0.375% (w/v) Thiobabituric acid ($C_2H_4N_2O_2S$, TBA) 0.375 g.

- 15% (w/v) Trichloroacetic acid (Cl_3CCOOH, TCA) 15 g.

- 0.25 N Hydrocholic acid (HCl) 2.475 ml.

Dissolve all chemicals in DDD water (100 ml) and make up to final volume of 100 ml.

2. Solution B: 1,1,3,3-tetramethoxy propane (TMP)

Dissolve 10 µl of 1,1,3,3-tetramethoxy propane (TMP) in DDD water and make up to final volume of 1000 µl and store at room temperature. TMP was diluted with DDD water to make serial concentrations of 0 nmol/ml, 0.46 nmol/ml, 0.93 nmol/ml, 1.85 nmol/ml, 3.71 nmol/ml, 7.41 nmol/ml, 14.82 nmol/ml, 29.65 nmol/ml, and 59.30 nmol/ml, respectively.

Procedure

1. Pipette 100 µl of the sample and standard enzyme into tube.

2. Pipette 200 µl of solution A into each tube, and mix well.

3. DDD water mixed with solution A is use as the control and solution A is use blank. All determinations were performed in triplicate.

Pipette the solution into each well as follow:

	Sample (µl)	Standard (µl)	Blank (µl)
50 mM PB, pH 7.4	-	-	100
Standard TMP	-	100	-
Sample (brain homogenates)	100	-	-
Solution A	200	200	200

50 mM PB, pH 7.4 = 50 mM phosphate buffer (pH 7.4) containing 0.1 mM EDTA

4. The mixture was heated on water bath 100 ºC for 15 minutes.

5. After cooling, the mixture was centrifuged at 1,000 g for 10 minutes.

6. The absorbance of the mixture (supernatant) was measured at 535 nm using microplate reader.

Calculation

The TMP concentrations was calculated using the following equation:

$$\Delta A_{535\ nm}\,\text{Standard} = A_{535\ nm}\,\text{Standard} - A_{535\ nm}\,\text{Blank}$$

Plot the $\Delta A_{535\ nm}$ Standard againt TMP concentration on the standard graph.

$$\Delta A_{535\ nm}\,\text{Sample} = A_{535\ nm}\,\text{Sample} - A_{535\ nm}\,\text{Blank}$$

The TMP (equivalence with MDA) level is determined as nmol/mg tissue weight from the standard curve.

APPENDIX C

DETERMINATION OF HYDROGEN PEROXIDE LEVEL

Chemicals:

- Disodium phosphate (Na_2HPO_4, Sigma-Aldrich; St. Louis, USA)

- Sodium dihydrogen phosphate monohydrate ($NaH_2PO_4.H_2O$, Sigma-Aldrich; St. Louis, USA)

- Hydrogen peroxide (H_2O_2, Sigma-Aldrich; St. Louis, USA)

- 2,2'-azino-bis-3-ethylbenzthiazoline-6-sulphonic acid (ABTS, Sigma-Aldrich; St. Louis, USA)

- Horseradish peroxidase type II, Sigma-Aldrich; St. Louis, USA

- Sodium hydroxide anhydrous pellets (NaOH, Carlo Erba Reagents, France)

- Hydrochloric acid (HCl, Carlo Erba Reagents, France)

Reagents:

1. 1 M HCl

Stock solution: 37% HCl

Preparation for 1 M HCl: mixed 9.85 ml of 37% HCl with double deionized distilled water (DDD water) and adjusted to the final volume at 100 ml.

2. 1 M NaOH

Preparation: dissolved 3.997 g of NaOH in DDD water and then adjusted to the final volume at 100 ml.

3. 1.25 mM ABTS

Dissolved 0.3 g of ABTS in 100 ml of DDD water and stored at room temperature.

4. 1 M phosphate buffer (pH 5.0)

Dissolved 10.56 g of di-sodium hydrogen phosphate anhydrous (Na_2HPO_4, BDH) and 3.968 g of sodium dihydrogen orthophophate 1-hydrate ($NaH_2PO_4.H_2O$, BDH) in 80 ml of DDD water and this solution was stirred on hot plate magnetic stirrer (VELP Scientifica, Europe) at 60 ºC for 1 hr. After 1 hr, solution left to be cool down to room temperature and then adjusted to final volume at 100 ml with DDD water in volumetric flask and adjusted to pH 5.0 with 1 M HCl or 1 M NaOH.

5. 0.1 M phosphate buffer solution, pH 5.0 (500 ml)

Mixed 50 ml of 1 M phosphate buffer solution (pH 5.0) with DDD water and adjusted to the final volume at 500 ml. And then the solution was adjusted to pH 5.0 with 1 M HCl or 1 M NaOH.

6. 10 mM H_2O_2 solution from 30% H_2O_2 solution

Diluted 78 µl of 30% H_2O_2 solution in DDD water and adjusted to final volume at 100 ml.

7. Standard hydrogen peroxide .

Dissolved 10 mM H_2O_2 solution from 30% H_2O_2 solution with DDD water to make up 0, 0.448, 0.976, 1.953, 3.90, 7.8125, 15.625, 31.25, 62.5, 125, 250, 500, and 1000 µM/ml H_2O_2 solution.

8. 1 unit.ml^{-1} horseradish peroxidase

Dissolved 0.1 mg of horseradish peroxidase in 25 ml of DDD water and stored at refrigerator

Procedure:

1. Pipetted 100 µl of 0.1 M phosphate buffer (pH 5.0) into 96 well-plates

2. Pipetted 100 µl of sample, standard H_2O_2, or 50 mM phosphate buffer (pH 7.4) containing 0.1 mM EDTA

3. Pipetted 20 µl of hydrogen peroxide and mixed well and then incubated at 37 ºC for 5 min.

4. After incubated, pipetted 30 µl of 1.25 mM ABTS

5. And add 30 µl of 1 unit.ml^{-1} horseradish peroxidase, mix all solution and incubated at 37 ºC for 10 min.

Pipette the solution into each well as follow:

	Blank (µl)	Sample (µl)	Standard (µl)
0.1 M phosphate buffer, pH 5.0	100	100	100
Sample (brain homogenates)	-	100	-
50 mM PB, pH 7.4	100	-	-
standard H_2O_2	-	-	100
10 mM H_2O_2	20	20	20
1.25 mM ABTS	30	30	30
1 unit.ml^{-1} horseradish peroxidase	30	30	30

50 mM PB, pH 7.4 = 50 mM phosphate buffer (pH 7.4) containing 0.1 mM EDTA

6. The absorbance of mixture were measure at 405 nm by Benchmark Plus Microplate Spectrophotometer.

Calculation

The hydrogen peroxide concentration were calculate using the following equation:

$$\Delta A_{405\ nm}\,\text{Standard} = A_{405\ nm}\,\text{Standard} - A_{405\ nm}\,\text{Blank}$$

Plotted the $\Delta A_{405\ nm}$ Standard against H_2O_2 concentration on the standard graph.

$$\Delta A_{405\ nm}\,\text{Sample} = A_{405\ nm}\,\text{Sample} - A_{405\ nm}\,\text{Blank}$$

The H_2O_2 level was determine as µM/mg protein.

APPENDIX D

DETERMINATION OF SUPEROXIDE DISMUTASE ENZYME ACTIVITY LEVEL

Reagents:

1. Water-solution tetrazolium salt (WST) working solution

Dilute 1 ml of WST with 19 ml of buffer solution.

2. Enzyme working solution

Centrifuge the Enzyme Solution tube for 5 sec. Pipette 15 µl of enzyme solution with 2.5 ml of dilution buffer.

Procedure

1. Add 20 µl of sample into each triplicate well of 96-well plates.

2. Add 200 µl of WST working solution into each triplicate wells, and mix.

3. Add 20 µl of enzyme working solution into each triplicate wells, mix and incubate the mixture at 37 ºC for 20 min.

Pipette the solution into each well as follow:

	Sample (µl)	Blank (µl)
Sample (brain homogenates)	20	-
50 mM PB, pH 7.4	-	20
WST working solution	200	200
Enzyme working solution	20	20

4. After incubate, the absorbance of the mixture was measured at 450 nm by using a microplate reader.

5. The SOD activity (inhibition rate %) will be calculated using the following equation:

$$\text{SOD activity (inhibition rate \%)} = \frac{(\text{Abs control} - \text{Abs sample})}{\text{Abs control}} \text{ x } 100.$$

APPENDIX E

DETERMINATION OF CATALASE ENZYME ACTIVITY LEVEL

Reagents:

1. 0.01 N of Hydrogen Peroxide (H_2O_2) solution

Dissolve 56.88 µl of hydrogen peroxide 30% (w/w) in 99.943 ml of 50 mM potassium phosphate buffer, pH 7 at 25 ºC.

2. 0.005 N Potassium permanganate ($KMnO_4$) solution

Dissolve 0.016 g of potassium permanganate in 100 ml of DDD water at room temperature.

3. 5 N of sulfulic acid solution

Dissolve 3.507 ml of sulfulic acid in DDD water and make up to final volume at 25 ml and then store at room temperature.

4. 50 mM Potassium phosphate buffer solution, pH 7 at 25 ºC

Dissolve 6.80 g of potassium phosphate in DDD water and store at 25ºC temperature.

5. Standard catalase enzyme solution. .

Immediately before use, serial concentrations of standard catalase enzyme solution were prepared in 50 mM phosphate buffer (pH 7.4) containing 0.1 mM EDTA solution (0, 0.39, 0.78, 1.56, 3.12, 6.25, 12.5, and 25 unit/ml.)

Procedures

1. Pipette 10 µl of the sample and standard enzyme into triplicate well of 96-well plates.

2. Pipette 50 µl of 0.01 N of Hydrogen Peroxide (H_2O_2) solutions into bottom of 96-well plates.

3. Add 25 µl of 5 N of sulfulic acid (H_2SO_4) solution into 96 well plates.

4. Pipette 150 µl Potassium permanganate ($KMnO_4$) solutions into 96-well plate and mix all solution.

Pipette the solution into each well as follow:

	Sample (µl)	Standard (µl)	Blank (µl)
50 mM PB, pH 7.4	-	-	10
Standard CAT enzyme	-	10	-
Sample (brain homogenates)	10	-	-
Hydrogen peroxide (H_2O_2)	50	50	50
Sulfulic acid (H_2SO_4)	25	25	25
Potassium permanganate ($KMnO_4$)	150	150	150

50 mM PB, pH 7.4 = 50 mM phosphate buffer (pH 7.4) containing 0.1 mM EDTA

5. The absorbance is then measured at 490 nm using a microplate reader.

6. The DDD water mix with hydrogen peroxide solution, sulfulic acid solution and potassium permanganate solution was used as the control.

7. Hydrogen peroxide solution, sulfulic acid solution and potassium permanganate solution was used as blank.

Calculation:

$\Delta A_{490\ nm}$ Standard = $A_{490\ nm}$ Standard - $A_{490\ nm}$ Blank

Plot the $\Delta A_{490\ nm}$ Standard against catalase enzyme concentration on the standard graph. .

$\Delta A_{490\ nm}$ Sample = $A_{490\ nm}$ Sample - $A_{490\ nm}$ Blank .

Determine the catalase enzyme activity from the standard curve. Catalase enzyme activities was express as unit/mg protein.

APPENDIX F

DETERMINATION OF GLUTATHIONE PEROXIDASE ENZYME ACTIVITY LEVEL

Reagents:

1. Glutathione peroxidase assay buffer .

Bring an appropriate aliquot of to room temperature. For long term stability of the solution after opening, handle the solution in an aseptic manner.

2. NADPH Assay Reagent .

Reconstitute 1 vial of in 1.25 ml of DDD water. Store the solution at 2-8 ºC.

3. 30 mM *tert*-Butyl Hydroperoxide Solution .

Dilute 21.5 µl of Luperox tert-Butyl Hydroperoxide (TBH) 70X with DDD water to make a total volume of 5 ml.

4. Glutathione Peroxidase Standard .

Dissolve a 100 unit vial of glutathione peroxidase in 1 ml of Glutathione Peroxidase Assay Buffer. Dilute to 0.25 unit/ml with Glutathione Peroxidase Assay Buffer supplement with 1 mg/ml IgG and 1 mM DTT.

5. Sample preparation .

Dilute brain homogenates in Glutathione Peroxidase Assay Buffer.

Procedure

1. Pipette Glutathione Peroxidase Assay Buffer by the volume as shown in the Table below. Keep the temperature of the assay buffer at 25 ºC.

2. Add 10 µl of NADPH Assay Reagent and 10 µl of sample and enzyme to 96-well plate and mix.

3. Start the reaction by addition of 2 µl of the 30 mM tert-Butyl Hydroperoxide Solution. And mix all solution.

Pipette the solution as follow:

	Blank	Sample
GPx Assay Buffer (µl)	188	178
NADPH Assay Reagent (µl)	10	10
Sample (µl) (brain homogenates)	-	10
30 mM t-Bu-OOH (µl)	2	2

4. Follow the decrease in absorbance at 340 nm using a kinetic program of Benchmark Plus Micriplate Spectrophotometer. The following program is recommended:

Wavelength: 340 nm

Initial delay: 15 seconds

Interval: 10 seconds

Number of reading: 6

5. Calculate the amount of enzyme in the sample.

The activity of glutathione peroxidase in the sample can be calculated using the formula:

Activity per extract (mmol/min/ml = Units/ml)

$$(\Delta A_{340} \text{ x DF}) \div 6.22 \text{ x V}$$

$\Delta A_{340} = A_{340}/\text{min}_{\text{(blank)}} - A_{340}/\text{min}_{\text{(sample)}}$

$6.22 = \varepsilon^{mM}$ for NADPH

DF = dilution factor of sample before adding to reaction

V = sample volume in ml

Unit definition: 1 unit of glutathione peroxidase will cause the formation of 1.0 μmol of $NADP^+$ from NADPH per minute at pH 8.0 at 25ºC in a coupled reaction in the presence of reduced glutathione, glutathione reductase, and tret-butyl hydroperoxire.

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