CHAPTER 4

Discussion and Conclusions

According to the present study, it can be summarized that 1) the estrogen deprivation aggravated the severity of metabolic parameters impairment in the obese-insulin resistant condition 2) the estrogen deprivation alone, obese-insulin resistant condition alone and combination of obese-insulin resistant followed by estrogen deprivation impaired cognitive function only hippocampal-dependent learning and memory but not hippocampal-independent learning and memory through the hippocampal dysfunction. 3) the estrogen deprivation did not aggravate these hippocampal dysfunction under an obese-insulin resistant.

According to previous studies done by others and studies done by our team used the model of ovariectomy followed by obesity to investigate the combined effects of estrogen deprivation and obese-insulin resistant (18, 149, 150, 1252, 153, 168). Those results found that obese-insulin resistant aggravated the peripheral insulin resistance, determined by increased plasma insulin, hyperglycemia, HOMA index and impaired glucose tolerance in the estrogen deprivation more than the obese-insulin resistant or ovariectomized alone (18, 149, 150, 152, 153, 168). Nowadays, the population of menopause women who suffer from obesity is seriously increased. Therefore, our present study interested in the effects of obese-insulin resistant followed by the estrogen deprivation model on peripheral disturbances, brain functions and cognitive function. It has been shown that ovariectomized rats fed with high-fat diet had higher level of peripheral insulin resistance, dyslipidemia and oxidative stress than the ovariectomized rats fed with normal diet or sham-operated rats fed with high-fat diet alone. These results demonstrated that obese-insulin resistant followed by the estrogen deprivation condition could aggravates the metabolic impairment in obese-insulin resistant condition, as determined by the aggravation of peripheral insulin resistance, dyslipidemia and oxidative stress.

It is well known that learning and memory are differentiated into explicit memory and implicit memory, which are hippocampal-dependent and hippocampal-independent memory, respectively (20, 21). Previous studies indicated that the HF consumption, the ovariectomy or the combination of both conditions could impair the spatial learning and memory (18). Nonetheless, no study had investigated the combine effect of obese-insulin resistant and estrogen deprivation on hippocampal-independent memory. Therefore, we first determined the comparative and cumulative effects of estrogen deprivation and obese-insulin resistant condition on both hippocampal-dependent and hippocampal-independent learning and memory in the same study. The results found that obese-insulin resistant, estrogen deprivation and obese-insulin resistant followed by estrogen deprivation impaired only hippocampal-dependent memory, but not impaired hippocampal-independent memory. Moreover, obese-insulin resistant, estrogen deprivation and obese-insulin resistant followed by estrogen deprivation equally lead to brain pathology in the hippocampal area, determined by the impairment of hippocampal synaptic function, increased hippocampal ROS production and hippocampal apoptosis. Additionally, data showed that obese-insulin resistant, estrogen deprivation and obese-insulin resistant followed by estrogen deprivation decreased estradiol level in the hippocampus. According to previous studies, estrogen can also be synthesized in the brain, notably in the hippocampus (169, 170). Estrogen in the hippocampus plays an important role to moderate the synaptic function which related to cognitive function (169, 171). Furthermore, preceding studies also showed that estrogen plays a role as an antioxidant effect, it can lower oxidative stress and apoptosis levels in the brain (172, 173). Thus, we contemplated that obese-insulin resistant, estrogen deprivation and obese-insulin resistant followed by estrogen deprivation resulted the hippocampal estradiol level reduction and hippocampal ROS production increment, which resulted to increase hippocampal apoptosis and led to decrease hippocampal synaptic plasticity. Finally, from these outcomes led to impair hippocampal-dependent learning and memory by the MWM test, demonstrated by increased time to reach the platform and decreased time in target quadrant. Nevertheless, obese-insulin resistant, estrogen deprivation and obese-insulin resistant followed by estrogen deprivation did not impair the hippocampal-independent memory and the brain pathology in the cortex area. We found that obese-insulin resistant, estrogen deprivation and obese-insulin resistant followed by estrogen deprivation did not

change cortical estradiol level, cortical ROS production. Accordingly, these conditions did not alter cortical apoptosis, which did not cause hippocampal-independent memory impairment by the NOR test, as determined by no change of both percent exploration time and index preference. The summaries of the obese-insulin resistant condition on hippocampal-dependent learning and memory and hippocampal-independent learning and memory, the estrogen deprivation on hippocampal-dependent learning and memory and hippocampal-independent learning and memory and the estrogen deprivation on hippocampal-dependent learning and memory and hippocampal-independent learning and memory in obese-insulin resistant condition were illustrated in Figure 4.1-4.3. The proposed mechanism of the obese-insulin resistant and estrogen deprivation on hippocampal-dependent learning and memory composed of 3 mechanisms 1) after estrogen binding with estrogen receptor lead to activate of protein kinase such as PI3K/Akt which promote insulin sensitivity (116, 174) 2) estrogen act as an antioxidant and anti-apoptosis effects, it can decrease oxidative stress and apoptosis levels in the brain (172, 173) 3) estrogen can enhance hippocampal synaptic plasticity by promoted AMPA receptor insertion (175, 176). Therefore, the condition of estrogen deprivation resulting in decreased brain insulin sensitivity, increased oxidative stress and apoptosis, impaired hippocampal synaptic plasticity and impaired hippocampal-dependent learning and memory. Previous study illustrated that the hippocampal-independent memory impairment occurred only after estrogen deprivation for 12 weeks (long term), as indicated by decreased time with novel object in NOR test (25), but not impair hippocampal-independent memory after estrogen deprivation for 6 weeks (short term). According to the present study, we used short duration of estrogen deprivation (only 7 weeks), could impaired only hippocampal-dependent learning and memory but could not impair the hippocampal-independent learning and memory, may be occurred after more long time of estrogen deprivation than 7 weeks. Moreover, we found that estrogen deprivation did not aggravate hippocampal and cortical pathology and cognitive impairment under an obese-insulin resistant condition. On the other hand, our previous studies reported that obesity can accelerate and aggravate the increase of brain oxidative stress, hippocampal synaptic dysfunction, brain mitochondrial dysfunction, brain insulin resistance and cognitive impairment (hippocampal-dependent learning and memory) in estrogen deprivation (6, 25). The duration of the estrogen deprivation in present study

(7 weeks) maybe a possible explanation for these results. Nevertheless, further studies are needed to be done to give more evidences to support this possible explanation. Properly, the longer duration of estrogen deprivation may aggravate the severity of brain pathologies and cognitive decline in the obese-insulin resistant condition. In addition, there are the difference between hippocampus and cortex including the cell types, hippocampus has more pyramidal neuron which play an important role in synaptic plasticity (177, 178). Previous studies found that ovariectomy result to decrease estrogen receptor in the hippocampus but ovariectomy did not alter estrogen receptor in cortex (24, 179). Therefore, the difference of estrogen receptor distribution in these two brain areas maybe one more possible explanation for the result in present study.

In conclusion, our present study demonstrated that estrogen deprivation aggravated the severity of peripheral insulin resistance, dyslipidemia and oxidative stress in obese-insulin resistant condition. Additionally, obese-insulin resistant, estrogen deprivation and obese-insulin resistant followed by estrogen deprivation impaired only hippocampal-dependent learning and memory through the decreased hippocampal estradiol level, increased hippocampal ROS productions, increased hippocampal apoptosis and impaired hippocampal synaptic function. Nevertheless, estrogen deprivation did not aggravate these deleterious effects under an obese-insulin resistant condition.

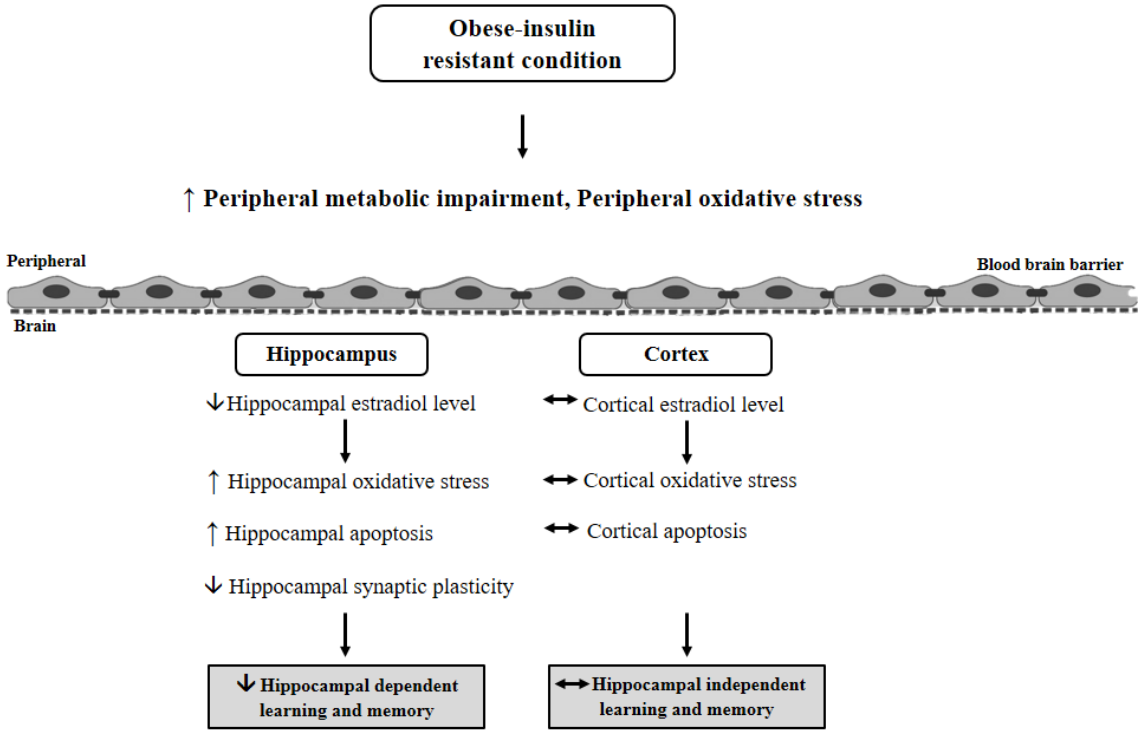

Figure 4.1 Summary of the obese-insulin resistant condition on hippocampal-dependent learning and memory and hippocampal-independent learning and memory; ↑ = increased of the parameters; ↓ = decreased of the parameters; ↓ = lead to; ↔ = no alteration in the parameter

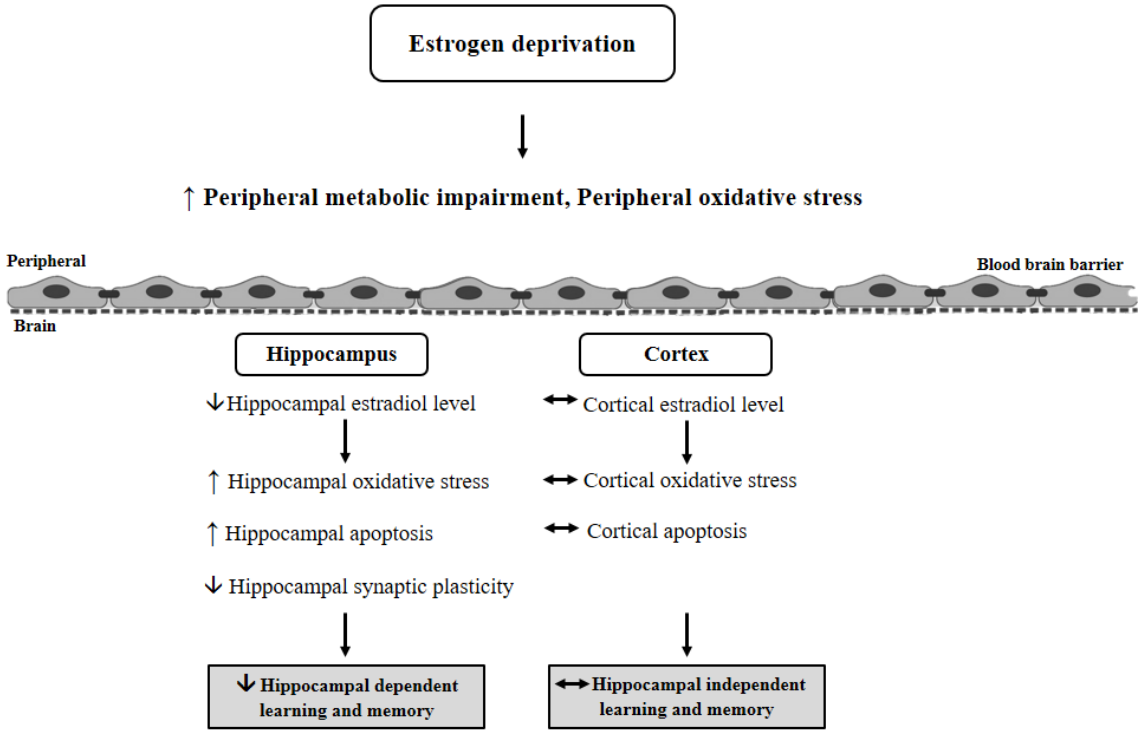

Figure 4.2 Summary of the estrogen deprivation on hippocampal-dependent learning and memory and hippocampal-independent learning and memory; ↑ = increased of the parameters; ↓ = decreased of the parameters; ↓ = lead to; ↔ = no alteration in the parameter

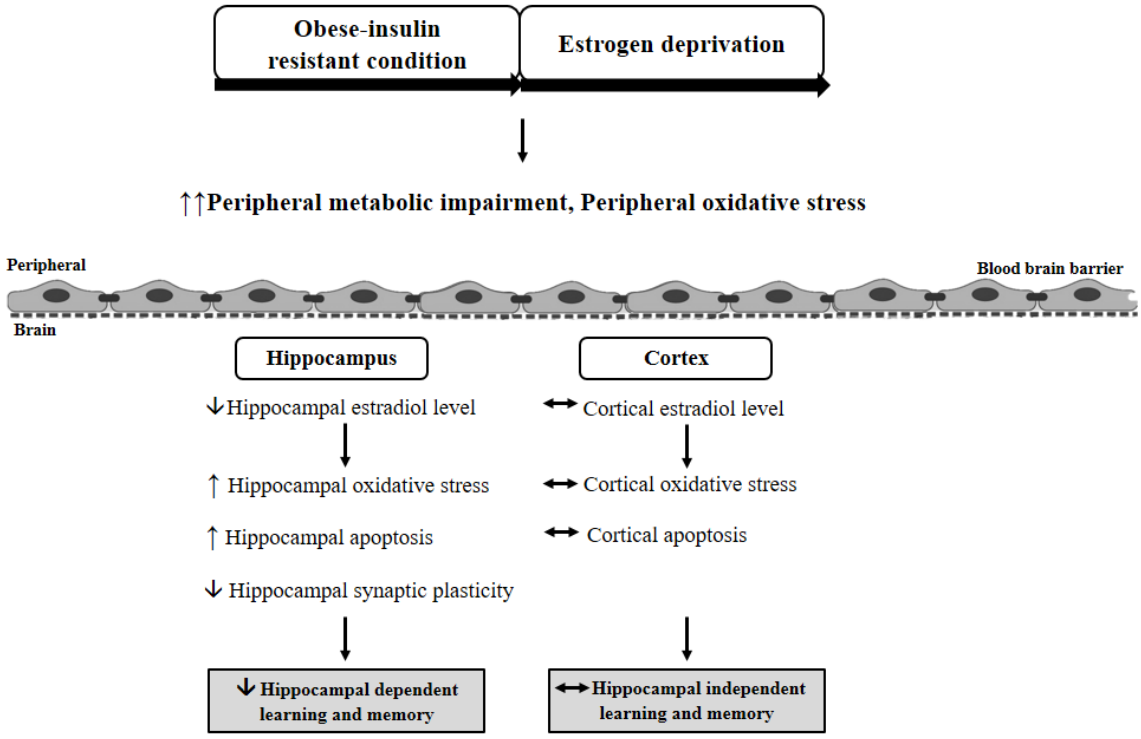

Figure 4.3 Summary of the obese-insulin resistant condition and estrogen deprivation on hippocampal-dependent learning and memory and hippocampal-independent learning and memory in obese-insulin resistant condition; ↑↑ = increased of the parameters from the combination effects of obese-insulin resistant followed by estrogen deprivation; ↓ = decreased of the parameters; | = lead to; ↔ = no alteration in the paramet