CHAPTER V
DISCUSSIONS AND CONCLUSIONS

P-glycoprotein, an encoded product of *multidrug resistant 1* gene or *MDR1* plays a crucial function for excretion of carcinogens in human cells. The variation of *MDR1* has been found to be correlated with change of P-gp expression and activity (Milojkovic et al., 2014). Lack of P-gp physiological function may contribute to increased risk of cancer development. Therefore, this study aimed to investigate association between the variation of *MDR1* at nucleotide position 1236, 2677 and 3435 and risk of cervical cancer in Northeast of Thailand. Our results demonstrated that *MDR1* genotype at position 3435 (C3435T) alone was not associated with cervical cancer risk (Table 5). This result is agreed with those studies in breast cancer and gastric cancer (Rubis et al., 2012; Zebrowska et al., 2014). In contrast, individuals carrying TT genotype had a higher risk for acute lymphoblastic leukemia, endometrial cancer and colon cancer which suggesting that the impairment of carcinogens clearance among TT genotype (C3435T) carriers may involve in the increased risk for cervical cancer development (Kurzawski et al., 2005; Mrozikiewicz et al., 2007; Wang et al., 2013).

Gene-gene interactions was observed in this study, the significantly increased cervical cancer risk was found among women carrying TT genotype (C3435T) in combination with GA and GT genotypes (G2677T/A) at 3.50-fold compared with those of GG genotype (G2677T/A) (Table 7). Similarly, increased risk for breast cancer was found in G2677T/A-C3435T variant combination (Fajac et al., 2010). The interaction between 2 SNPs of *MDR1* lead to the use of rare codons which influences the protein folding process and results in variation of tertiary conformation (Kimchi-Sarfaty et al., 2007). Thus, the gene-gene interactions induce-protein conformation change may reduce substrate specificity and affinity of ATP-binding and consequently reduced the protection against P-gp dependent carcinogens (Brambila-Tapia, 2013; Kimchi-Sarfaty et al., 2007).

Among women who taking hormonal contraceptives, the carriers of TT genotype at 3435 SNP had a 3.30-fold elevated risk for cervical cancer as compared with those of CC genotype (Table 14). Hormonal contraceptives contain combinations of estrogen and progestin or progestin alone, which play a role in proliferation of cervical epithelium cell (Gariglio et al., 2009; Plu-Bureau et al., 2013). Several studies have been shown association between long term uses of oral contraceptive and increased risk of cervical cancer (Gierisch et al., 2013; Smith et al., 2003; Urban et al., 2012). The promoting integration of HPV-DNA into host genome is a proposed mechanism of hormone-induce progression of pre-malignant to malignant cervical lesions (Castellsague et al., 2002). Furthermore, experimental study has been shown that estradiol may stimulate transcription of E6 and E7 of HPV16 (Gariglio et al., 2009). The TT genotype of 3435 SNP could be a risk factor for cervical cancer development due to the diminished affinity of ATP-binding which lead to the decreased P-gp function (Brambila-Tapia, 2013; Kimchi-Sarfaty et al., 2007). The effect of low P-gp may cause a remaining hoarding of estrogen and/or progestin within the cell (Coles et al., 2009).

Regarding G2677T/A and C1236T SNP of *MDR1*, the relationship between *MDR1* (G2677T/A and/or C1236T) and increased cervical cancer risk was observed. The protective effect for cervical cancer was found among carriers of CT genotype (C1236T) together with TA or TT genotypes (G2677T/A) (Table 6). Surprisingly, our data differ from study of Zebrowska and coworker who found that the TT genotype of 2 SNPs, 1236 and 2677 may involve occurrence of gastric cancer (Zebrowska et al., 2014). It was suggested that the synonymous SNP at position 1236 plays an important role in the binding of substrates, the alteration of tridimensional structure that cause by interaction of 2 SNPs of *MDR1* may change P-gp function (Brambila-Tapia, 2013). Thus, polymorphism at position 1236 and 2677 may increase affinity of substrate binding together with induced ATPase activity. Finally, the potentially toxic substances and carcinogens are eliminated out of the cells (Brambila-Tapia, 2013).

Human papilloma virus or HPV is a crucial factor for the development of cervical cancer. Interestingly, HPV-infected women who carrying TT genotype (G2677T/A) had a higher trend of cervical cancer risk compared to those with CC genotype (Table 16). The mechanism of HPV-induce carcinogenesis is inactivation of

p53 and *retinoblastoma (RB)* genes which are tumor suppressor gene (de Freitas et al., 2014). According to our result, the homozygous mutant (TT) genotype may reduce affinity of ATP-binding which results in increased carcinogen accumulation within the cells (Brambila-Tapia, 2013; Fung and Gottesman, 2009). The high carcinogens accumulation in the cells due to low activity of P-gp together with infection by HPV which cause an impairment of cell cycle checkpoints leading to abnormality of cell proliferation and cancer development (Brambila-Tapia, 2013; de Freitas et al., 2014). Our findings are in agreement with studies in Chinese and Brazilian populations and suggested that genetic polymorphism interacting with HPV infection is contribute to cervical cancer susceptibility (Chagas et al., 2013; Xu et al., 2012).

In summary, our results conclude that the genetic variation of *MDR1* is associated with the risk of cervical cancer development in Northeast of Thailand. The homozygous mutant genotype of *MDR1* at position 3435 (TT genotype) may considered as a risk factor of cervical cancer, especially among women who taking contraceptives. Regarding *MDR1* at position 2677, the TT genotype could be a risk factor for cervical cancer among HPV-infected women. Moreover, the study was also found the relationship between gene-gene interaction and either risk factor or protective factor of cervical cancer. Thus, further study in *MDR1* polymorphism should be provided for a clinical use with regard to cervical cancer prevention and treatment.