## **CHAPTER 4**

## CONCLUSION

The water extract of *Hibiscus sabdariffa* Linn. (Roselle) is a strong free radical scavenger. Although it is less effective than commercial antioxidants such as ascorbic acid and  $\alpha$ -tocopherol, it shows similar activity to BHT in the DPPH radical scavenging assay. With the EC<sub>50</sub> value of  $8.45 \pm 0.35 \,\mu\text{g/ml}$ , Roselle extract can be considerate as one of the good sources for natural antioxidants. However, due to the hydrophilic nature of the extract, it exhibits remarkably low activity in inhibition of lipid peroxidation of liposome. The active components of Roselle extract are polyphenolic compounds especially anthocyanins. High linear correlations were found between the antioxidant capacity of Roselle extract and the contents of either polyphenolics or anthocyanins. These results indicate that polyphenolics or anthocyanins are major components that contribute to antioxidant capacity in Roselle extract.

Although Roselle extract shows good antioxidant capacity, it has stability problems. The color and stability of anthocyanin pigments of Roselle extract are dependent on several factors such as pH, light and temperature. In acidic solutions, Roselle extract is charmingly pink-red. However, the red hue decreases and its color gradually changes toward more bluish as pH increases, and then finally turns to brown and yellow tone in alkaline pH. These phenomena are due to the structures and concentration of anthocyanins: flavylium cation (red), quinonoidal anhydrobase (blue), carbinol pseudo-base (colorless), and chalone (colorless or light yellow) at equilibrium which vary with pH. The antioxidant capacity of Roselle extract is also

influenced by the amount of these anthocyanin structures. The activity is higher in acidic solutions than neutral and basic solutions. This activity still remains even after color fading and anthocyanin degradation has occurred.

In dry state, Roselle extract absorbed moisture from the environment very rapidly. After storage under accelerated conditions (45 °C, 75% RH) for 4 months, the color of dried Roselle extract changed progressively from pinky red to dark brown. Its degradation seemed to be related with the decreasing of red monomeric anthocyanins and the occurrence of brown polymeric compounds. However, there was no significant change of its antioxidant capacity and total phenolic contents. These results indicated that polymer derived from anthocyanin degradation increase the DPPH scavenging ability.

In the formulation development study, creams containing 5%w/w of Roselle extract can be formulated. After heating and cooling cycle tests, the limit stability of anthocyanins in the formulations was observed, but the antioxidant activities were significantly increased. Again, this may due to the activity of degradation products occurring after the heating and cooling cycle test.

Overall, Roselle extract can be considered as a good natural antioxidant. However, it is not stable in several conditions such as temperature, moisture and pH. This may limit of it use as an active component in cosmetics and pharmaceuticals. The novel pharmaceutical technology such as liposome or nanosome may require avoiding or minimize this stability problem.