CHAPTER V

CONCLUSION

In the study of antioxidant activity of Thai marigold flower extracts by two models: DPPH assay and TBARS assay compared with reference antioxidants (Trolox, quercetin and gallic acid). The Hexane (H), ethyl acetate (EA) and ethanol (Et) extracts were obtained by continuous extraction with Soxhlet apparatus. Then EA which showed the highest antioxidant activity than H and Et was selected for further isolation by vacuum column chromatography in which providing 14 fractions (F1-F14) and then these fractions were determined antioxidant capacity by both methods. The fraction 9 (F9) exhibited the highest antioxidant activity in both assays. The total phenolic content of marigold extracts were also determined. The result revealed that EA, F8 and F9 expressed the higher total phenolic content among all extracts. The total phenolic content study demonstrated the corresponding to the antioxidant activity. This is indicating that phenolic compounds might be the major contributors to the antioxidant activity of the marigold extracts.

In the part of the formulation study of NLC, The unloaded NLC dispersions consisting of 10% lipid phase and stabilized by a combination of Tween® and Span® were successfully obtained by hot high pressure homogenization. NLC dispersions with good physical stability; particle size, PDI and zeta potential, over a storage time of 90 days were obtained from three formulations with differences in type of solid lipid or liquid lipid or solid lipid: liquid lipid ratio. The incorporation of marigold

flower extracts; EA and F9 extracts which possess the high antioxidant activity into these three formulations was facilitated. The effect of surfactant on physical appearance was observed by varying the amount from 5 to 12%. The obtained EA-NLC dispersion has yellow color, good smell, smooth and homogenous texture with low viscosity while F9 NLC has the same characters with pale yellow color. After stability test, the results revealed that there is only one suitable NLC formulation for EA while there are two suitable formulations for F9 which contain 12% surfactant in formulation for both extracts. During storage at 4°C and RT, the particle size of these three ME-NLC increased. However they maintained the particle size in nanometer range, suggesting the possibility of maintaining the formulations against the aggregation when stored at these temperatures.

NLC dispersions are interested for topical application. They have to be incorporated into a semi-solid formulation such as cream, in order to have a proper consistency for cosmetic application. NLC dispersions containing antioxidant extracts, EA and F9, were incorporated into oil in water cream to obtain nano-cosmeceutical cream from marigold flower extract as anti-wrinkle cream. The ME-NLC cream exhibited no irritation after test by modified Draize model in rabbit and healthy volunteers. The ME-NLC cream were storage in H/C cycling for six cycles as stress condition and they were stable at this condition. The F9-NLC cream revealed the higher antioxidant activity than F9 cream whereas EA-NLC cream exhibited the same results. During storage, antioxidant activity of all tested creams decreased but F9-NLC creams showed significantly higher antioxidant activity than F9 cream

(p<0.05). This might say that the incorporation of F9 into NLC can increase the stability of F9 to light resulting in the higher %inhibition.

The wrinkle reducing capacity of ME-NLC creams were also evaluated in 25 volunteers by Skin Visiometer® SV 600 FW then analyzed in four parameters (volume, surface, Roughness R*a* and Rz). After 8 weeks of application, F9 and EA-NLC creams showed significantly higher wrinkle reducing capacity compared with before treatment (p<0.05) in all parameters. In addition, F9-NLC cream demonstrated considerable wrinkle reducing efficiency at the end of treatment, with significantly difference compared with F9 cream while EA-NLC cream had no significantly different wrinkle reducing efficiency compared with EA-cream (p<0.05, ANOVA). This result is corresponding to the percent inhibition result in antioxidant activity test.

There are a few studies about marigold extract as active compound in the present cosmetic market. Marigold flower extract presented as another potent antioxidant activity from natural source which can be useful in anti-aging or anti-wrinkle cosmetic products. The results from this study indicate that NLC was a promising delivery system for marigold flower extract that can be used as anti-wrinkle cosmeceuticals and this can be value added to the marigold which widely grown in Northern of Thailand.