CHAPTER 5

CONCLUSION

This research project is focused on the development of lycopene loaded NLC for cosmeceutical applications. Lycopene identification was performed by UV/Vis spectroscopy and HPLC. Three absorption peaks observed from UV/Vis were recorded at 445, 475, and 503 nm of which the maximum absorption peak is 475 nm. The HPLC fingerprint of lycopene showed the typical peak at a retention time of about 11.4 min. Lycopene is soluble in solid lipids, liquid lipids and non-polar solvents. It is partially soluble in acetone, acetonitrile, alcohol and insoluble in water, PBS pH 7.4, Transcutol^{TM} P, glycerin and polar solvents. The melting point of lycopene was observed at 171.03°C and its crystalline characteristic was confirmed by WAXS. Lycopene is the potent antioxidant substance with TEAC value of 2646.2 ± 3.8 μM whereas the IC_{50} value is 0.078 ± 0.001 mg/mL. Due to containing conjugated double bonds, it is rapidly degraded when exposed to light and oxygen and subsequently bleaches its color.

Orange wax was selected as the main solid lipid ingredient according to its benefit properties and particularly the smaller particle size could be obtained. In preparation of nano delivery systems, the most suitable production conditions was found to be 3 homogenization cycles and 500 bar homogenization pressure unless otherwise was required. Formulation using rice bran oil yielded the smaller particle size. In comparison with NE, the NLC is found to be the most potential carrier systems for lycopene. The NLC showed higher entrapment efficiency. After 30 days

of storage at room temperature the NLC showed sufficient physical stability. Both NLC and NE formulations showed free radical scavenging activities after tested with ABTS and DPPH assays, but at different levels. The highest antioxidant capacities were found from NLC load, NLC free, NE load, and NE free, respectively. The antioxidant capacity of the formulations was according to the amount of lycopene loaded. Electrostatic stabilizer gave better long term stability. The stability data showed that Eumulgin^{TM} SG surfactant could provide a better long term stability of formulation over 1 year.

In NLC development, incorporation of liquid lipid 20% into solid lipid leads to the separation of solid lipid out of the particle. This data suggested the maximum loading of oil up to 10%. Lycopene-loaded NLC formulations were successfully developed using skin-friendly materials. The observed particle diameter by LD (0.99) of SLN was 405 nm whereas that of NLC without cholesterol was 350 nm. The NLC with cholesterol had a diameter by LD (0.99) of 287 nm. Rice bran oil and cholesterol facilitated the reduction of particle size. Both NLC systems showed high zeta potential and physical stability. Nevertheless, the chemical stability profile of lycopene was unfavorable when cholesterol was incorporated in the NLC. To prolong the chemical stability profile of lycopene, the NLC should be stored below 25°C and free from cholesterol.

High crystalline characteristics as anisotropic molecular organization crystal of orange wax and lycopene can be clearly investigated by PLM, DSC, and WAXS. Low crystallinity of lipid nanocarriers like SLN or NLC containing small amount of lycopene and solid orange wax is hardly detected by those techniques. Electron diffraction mode of TEM shows potential detection of the crystalline characteristics of

small concentration of crystalline substances of both SLN and NLC. It could be concluded that TEM is a potent method to detect the low level crystallinity of the solid nanocarriers. In this study, NE presents in an amorphous form. The internal structure of obtained SLN, and NLC organized as polycrystalline structure. Cryo-TEM images revealed lycopene loaded NLC with a spherical shape.

Increasing amount of lycopene, the particle size was only slightly increased, whereas the surface properties were not affected. The entrapment efficiency is higher than 99% in all tested concentrations. It was found that NLC showed a biphasic release pattern provided a fast release initially for skin saturation followed by a slow and prolonged release profile to maintain the skin concentration of lycopene. The high occlusion factor could be obtained by increasing amount of lycopene incorporation. NLC formulations showed free radical scavenging activities after tested with ABTS and DPPH assays, but at different levels. The highest antioxidant capacities were found from 0.050% lycopene loading, followed by 0.025% and 0.005%, respectively. The antioxidant capacity of the formulations was according to the amount of lycopene loaded.

The promising NLC dispersion was incorporated into seven base cream formulations. Addition of NLC to the base cream increases drastically the occlusive effect. The good chemical stability of lycopene was obtained by entrapping in the cream. It can be concluded from the *in vitro* ABTS and DPPH assays that the antioxidant capacities of base creams could be ultimately adjusted in a controlled way by incorporating with the lycopene-loaded NLC.

It can be concluded that the lycopene-loaded NLC was potentially developed. The chemical stability of lycopene entrapped in the NLC was drastically enhanced.

This study emphasized the success on the utilization of NLC to retard the degradation profile of lycopene. Lycopene-loaded NLC and its cosmeceutical formulations provided efficacy occlusive and efficacious anti-oxidative properties.