ANTIOXIDANT STABILITY ASSAY OF ALPHA TOCOPHERYL ACETATE IN SOLID LIPID NANOPARTICLE SYSTEM (LIPID BASE BEESWAX AND MONOSTEARIC GLISERYL 50:50)

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ABSTRACT

Introduction : As an antioxidant, alpha tocopheryl acetate is easily degraded by light and free radical in air. Solid Lipid Nanoparticle (SLN) is a system that can provide protection of active ingredient because of its drug entrapment ability and physical protective from UV light due to its nano-sized. The aim of this study is for investigated the ability of SLN that could increase stability of antioxidant potency from alpha tocopheryl acetate. SLN is expected to have higher stability of antioxidant potency compared with simple cream.

Methods : SLN and simple cream was made SLN was made using lipid beeswax (BW) and monostearic glyceryl (GMS) as base and Tween 80 as stabilizer. SLN was produced using high shear homogenization method and simple cream was produced using hot plate magnetic stirrer. Antioxidant potency was measured by DPPH method. Sample was radiated by UV C light as free radical initiator.

Result and Discussion : Alpha tocopheryl acetate loaded in SLN system has higher stability of antioxidant potency compared with simple cream that shown with k value as constanta of antioxidant potency degradation between time. This could be due to its physical blocker of UV light and drug entrapment properties.

Conclusion : SLN was selected as antioxidant carrier due to its ability to increase antioxidant stability of alpha tocopheryl acetate.

Keywords : antioxidant, alpha tocopheryl acetate, DPPH, stability, SLN

INTRODUCTION

Solid Lipid Nanoparticle (SLN) is a dispersion system with nano-sized particles in range 40-1000 with spherical shape (Muller, 2009). SLN has widely used in cosmetic formulation because of its advantages due to its nano-sized particle such as UV protective effect and enhance emmolient (Souto, 2008; Wissing, 2002).

Alpha tocopheryl acetate is derivate from tocopherol that commonly used as an antioxidant in cosmetic due to its properties of photo protective for skin and could decrease skin's damage of free radical (Nam, 2012; Tsai, 2012). As an antioxidant, alpha tocopheryl acetate is easily degraded by light and free radical in air. SLN is one of the system that could prevent the antioxidant degradation of alpha tocopheryl acetate especially because the presence of light. It is result from SLN ability to scattering light with its nano-sized particles and produce physical UV protective effect (Golmohammadzadeh, 2012). The lipid matriks that contained in SLN also has the protective effect for active ingredient (Souto, 2008).

The aim for this study was for investigated the impact of SLN system with binary lipid beeswax and monostearic glyceryl (50:50) on antioxidant stability of alpha tocopheryl acetate compared with simple cream. Antioxidant activity is measured by DPPH method. Combination 50:50 of lipid component beeswax and monostearic glyceryl was selected due to the combination has higher physical stability and drug entrapment (Jenning, 2000; Rosita, 2013).

MATERIALS AND METHOD

Materials:

Tocopheryf acetate, DPPH (Sigma Aldrich), beeswax, monostearic glyceryf, tween 80, (PT. Bratako). Dapar component: acetic acid and sodium citrate (E. Merck), aquadest (PT Jawisesa).

Instrument:

Double beam UV Spectrophotometer UV-1800 Shimadzu, One Fourier Transform Infrared (FTIR} Spectrophotometer, pHI meter Eutech Instruments pH 700, Differential Thermal Analyzer (OTA), DelsaTMNano C Particle Analyzer, JEOLJEM-1400 Electron Microscope, Ultra Turrax IKA® T25 Digital High Shear Homogenizer, Sentrifuge Hettich Rotofix 32, Ultrasonic LC 60h Elma,, Hot plate, UV C lamp

Preparation method of SLN

SLN formed by beeswax and monostearic glyceryl (GMS) as lipid core 50: were prepared by high shear homogenization method . 10% of tween 80 was used as surfactant and 20% of propylene glycol was used as co-surfactant. Acetic buffer pH 4.2 μ=0,5 was used as aqueous phase (table 1). They were stirred at 24,000 rpm for 8 minutes, with 30 seconds intervals every two minutes, using an Ultra Turrax homogenizer T-25. The hot dispersion were cooled keep in stirring decreased speed gradually.

Ingredient	Function	Concentration
Beeswax	Lipid	10%
Monostearic glyceryl	Lipid	
Alpha tocopheryl acetate	Active ingredient	10%
Tween 80	surfactant	10%
Propilen glycol	Co-surfactamt	20%
Acetic buffer pH4.2, μ=0,5	Aqueous phase	Ad 100%

Table 1. Formula of SLN and simple cream tocopheryl acetate

Characterization of SLN

Measurement of Particle Size

Each sample was diluted with water before measurement. The particle sizes were analyzed by Delsa Nano Particle Size Analyzer at 25°C. Each measurement was performed in triplicates and the particle average diameter and polydispersity index (PI) was determined.

Observation of SLN morphology

The morphology of SLNs was observed by Transmission Electron Microscope (TEM). Either SLN blanks or SLN-PMCA were stained with phosphotungstic acid 2% w/v and placed on copper grids with former film for viewing at 120 kV (JEOL JEM-1400) and operated using software. The shape of SLN observed with Thermal Electron Microscope (TEM) and Drug entrapment of PMCA was measured by centrifugation method.

Measurement IC SO Value of Alpha Tocopheryl Acetate

IC SO value of alpha tocopheryl acetate was measured by DPPH method. The series of concentration of alpha tocopheryl acetate was prepared. 2 ml of sample in ethanol solution was centrifuged with 2 ml of DPPH solution. After 30 minutes absorbance of sample was measured by spectrophotometer in 517 nm.

Measurement of Antioxidant stability of Alpha tocopheryl acetate in SLN and simple cream

Each sample of SLN and simple cream alpha tocopheryl acetate was radiated by UV C light as free radical initiator. 1 gram of each sample was taken and diluted in ethanol and then centrifuged at 2500 rpm for 20 minutes. Supernatant was taken 2 ml and then mixed with 2 ml of DPPH. The reduction in DPPH radical was measured by spectrophotometer at 517 nm.

$$\%\ rsductium = \frac{\{A_{control} - AJam;.16\}}{A_{control}} \times 100\%$$

RESULT AND DISCUSSION

SLN-alpha tocopheryl acetate had most spherical shape (fig 1,). Range of particle size, average of particle size and their Polydispersity Index (PI) can be seen in table 2.

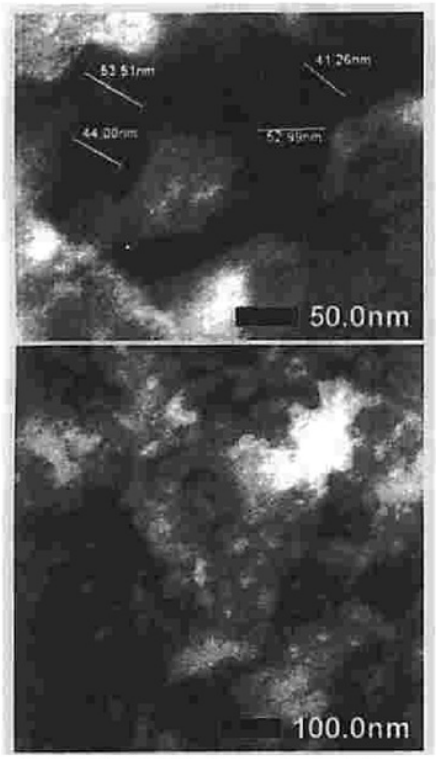

Fig 1. Morfology of SLN-alpha tocopheryl acetate measured by TEM

System	Replication	Diameter range (nm)	Poly-dispersIry Index (PO	Diameter (nm))	Mean of Particle size e SD (nm))
SLN alpha tocopheryl acetate	1	252,2 – 2544,9	0,345	900,7	1087,03 ± 256,28
	2	353,5 – 1764,3	0,386	981,1	
	3	338,4 – 6184,0	0,530	1379,3	
Simple cream alpha tocopheryl acetate	1	727,6 – 34129,2	0,766	2302,5	1957,9 1321,95
	2	414,7 – 213904,4	0,623	1664,8	
	3	417,9 – 95650,4	0,642	1906,4	

Table 2. Range of particle size, average of particle size and their Polydispersity Index (PI)

The existence of the particle size of SLN alpha tocopheryl acetate that are outside the range because the weakness of the manufacturing method of SLN with high shear homogenizer / high speed homogenizer which can result in the size of the microparticles in the SLN (Lason, 2011).

From table 3 it shown the IC 50 value of alpha tocopheryl acetate. The mean value of IC 50 alpha tocopheryl acetate is 2,56% ± 0,0850.

Replication	IC 50 Value (%)	Mean of IC 50 value± SD (%)
1	2,66	
2	2,53	2,56±0,0850
3	2,50	

Table 3. IC 50 value of alpha tocopheryl acetate

The antioxidant stability of SLN and simple cream alpha tocopheryl acetate was shown in table 4 and fig.2.

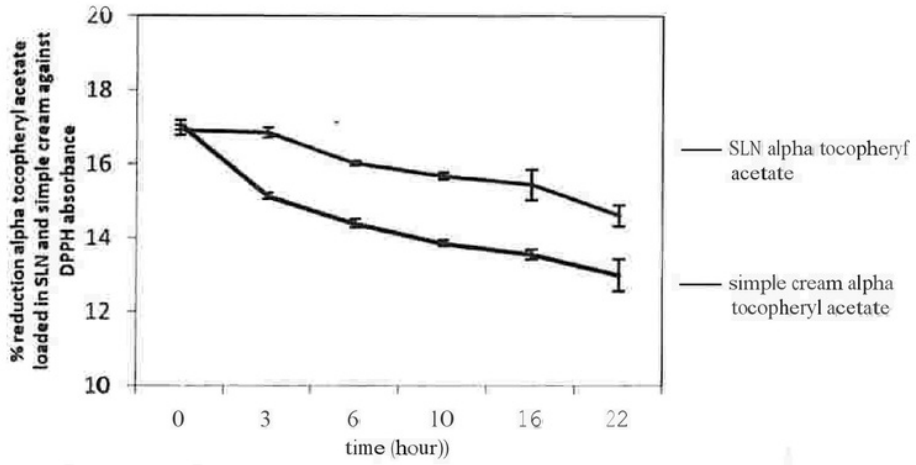

Fig. 2. Graphic% reduction alpha tocopheryl acetate loaded In SLN and simple cream against DPPH absorbance between time

System	Replication	constants of antioxidant potency degradation between time (/hour)	Mean of constanta of antioxidant potency degradation between time±SD
SLN alpha tocopheryl acetate	1	0,0913	0,1030±0,0175
	2	0,0946	
	3	0,1232	
Simple cream kouvensional alpha tocopheryl acetate	1	0,1714	0,1558±0,0169
	2	0,1379	
	3	0,1580	

Table 4. Constanta value of antioxidant potency degradation between time

From graphic in fig.2 it can be determined that SLN alpha tocopheryl acetate shown less sloping than simple cream alpha tocopheryl acetate. It means that decrease of antioxidant stability of alpha tocopheryl acetate loaded in SLN over the time is smaller than loaded in simple cream. This result was confirmed with constanta value of antioxidant potency degradation between time among SLN and simple cream loaded alpha tocopheryl acetate. SLN alpha tocopheryl acetate has smaller value of constanta of antioxidant potency degradation between time than simple cream alpha tocopheryl acetate. This phenomenon may occur as a result of UV blocker by SLN and drug entrapment effect of SLN that could protecting active ingredient.

CONCLUSION

SLN was selected as antioxidant carrier due to its ability to increase antioxidant stability of alpha tocopheryl acetate.

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