Efficacy and Safety of Curcuminoids Loaded Solid Lipid Nanoparticles Facial Cream as an Anti-aging Agent

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Abstract

Dried turmeric powder has long been incorporated as an active ingredient in various cosmetic products. Recently, the antioxidant properties of curcuminoids have gained enormous attention as an anti-aging agent. The aims of this study were to (1) evaluate the effect of curcuminoids loaded solid lipid nanoparticles (SLN) facial cream as an anti-aging agent in healthy volunteers, and (2) test the safety of this product. This study was a prospective, half-face, randomization of face, cream base control trial. Thirty-three healthy volunteers with noticeable facial wrinkles participated in this study. The participants applied curcuminoids loaded SLN cream and cream base on the assigned side of their faces before bed for eight weeks. Skin wrinkles, hydration, melanin content, biological elasticity, and viscoelasticity were main outcomes measured. Skin irritation was indicated by transepidermal water loss index, skin pH, and physician's observations. The results indicated that, from week three onward, all measures of efficacy of the treatment side were significantly better than the control side. When compared with their baselines, all efficacy measures were significantly improved by two weeks. In addition, no sign of skin irritation was observed. In conclusion, curcuminoids loaded SLN cream was effective as an anti-aging preparation with acceptable safety.

Keywords: Curcuminoids; Solid lipid nanoparticles; SLN; Anti-aging

Introduction

Aging of the skin is a continuous process associated with increased wrinkles, deep lines and irregular pigmentation (Novoseltsev et al., 2001). An important event in the process of aging is the production of reactive radical species by oxidative phosphorylation processes and from exogenous sources (Giacomoni & Rein, 2001; Giacomoni & Rein, 2004). Free radicals are the cause of deterioration of skin's supporting structures, leading to decreased elasticity and resilience (Bolognia, 1995; Giacomoni & Rein, 2001; Wlaschek et al., 2001). As much as aging process is understood, many attempts have been made to prevent it. One approach is to slow down free-radical damage using various antioxidants.

Recently, the antioxidant properties of curcuminoids, a group of three phenolic compounds, i.e., curcumin, demethoxycurcumin, and bisdemethoxycurcumin, isolated from the rhizomes of turmeric (*Curcuma longa* Linn., Zingiberaceae), have gained enormous attention in the field of cosmetics (Miquel et al., 2002; Scott, 1999; Selvam et al., 1995). Unfortunately, curcuminoids are easily degraded by acid and alkali hydrolysis, oxidation, and photodegradation (Ansari et al., 2005; Bernabe-Pineda et al., 2004; Pfeiffer et al., 2003; Sundaryono et al., 2003; Wang et al., 1997). This makes the stability issue of the compounds a discernible problem in product formulation.

Applications of drug delivery system have been found to improve stability and enhance activity of various unstable active ingredients (Ravi-Kumar, 2000). Solid lipid nanoparticles (SLN) are among recent development of drug delivery system

that have shown to be potential carrier systems, particularly for lipophilic drugs (Lim & Kim, 2002; Shlomo & Elka, 1999; Sowbhagya, 2005).

During the last ten years, applications of SLN have emerged in topical cosmetic formulations such as insect repellents, sunscreen, and anti-aging products with well toleration (Cavalli et al., 1997; Mehnert & Mäder, 2001; Müller et al., 2002; Shlomo & Elka, 1999). SLN were found to promote stability of the active compounds by protecting them from photodegradation, hence prolonging their release from the system (Mehnert & Mäder, 2001; Müller et al., 2000). They also offered high active compound payload and odor masking effects (Müller et al., 2000; Müller et al., 2002; Shlomo & Elka, 1999). In addition, SLN possess superior occlusive effects compared to conventional emulsions and microparticles. SLN enhance penetration of active substances through stratum corneum by increasing its hydration and forming an intact film while drying (Cavalli et al., 1997; Müller et al., 2000; Müller et al., 2002).

The objectives of this study were to (1) examine the efficacy of curcuminoids loaded SLN anti-aging night cream and (2) test the product safety.

Materials and Methods

The product

Curcuminoids extract was purchased from Thai-China Flavours and Fragrances Industry Co., Ltd. (Bangkok, Thailand). The extract, produced on September 2, 2004, contained 92% curcuminoids (5.25% bisdemethoxycurcumin, 18.75% emethoxycurcumin, and 68.04% curcumin). The cream preparation was prepared by a microemulsion technique at moderate temperature (Gasco, 1997). The details of formulation as well as its physical and chemical stability of the product were confirmed (Tiyaboonchai et al., 2007). The tested product was curcuminoids loaded SLN cream. Control was cream base with the same composition as the tested product except the active ingredient.

Location

This study was conducted at Cosmetics and Natural Products Research Center, Naresuan University, Phitsanulok, Thailand, between August and November, 2005.

The participants

Participants were enrolled in the study on the following criteria: healthy persons aged 30-55; had visible facial wrinkles as determined by a dermatologist; had no history of smoking, alcohol or drug use; and no history of cosmetic product allergy. Participants were excluded if they: had history of any kind of allergic reaction; had history of eczema or psoriasis within six months prior to enrollment; used steroids, antibiotics, NSAIDs, and antihistamines within three days prior to enrollment; applied any topical agents on the face within three weeks prior to enrollment; had major operation within one year prior to enrollment; were pregnant or lactating. In addition, the investigators might discontinue a participant if he/she showed any sign of allergy to the tested products, concomitantly used any moisturizing products during the trial, and requested for discarding from the study.

Study design

The study was open-labeled, match paired design with randomization of treatment to the side of face. The duration of the study was eight weeks. The protocol was approved by Naresuan University Institutional Review Board.

On the first day, the participants reported at the research laboratory for baseline measurements. $\%SE_w$, SCH, melanin value, U_r/U_f, U_v/U_e, TEWL, and skin pH were recorded for both sides of the face. After that, the test product (curcuminoids loaded SLN cream) and control product (cream base) were given to the participants. The participants were instructed to apply approximately 2 mg of test and control products on the assigned side of the face once daily at night. Each volunteer received a diary for recording adverse events and monitoring their compliance. The follow up visits were scheduled every week after beginning of the trial (weeks 1-8). Efficacy as well as safety measures were conducted on every visit.

Outcome measurements

Efficacy of curcuminoids loaded SLN cream was determined by skin wrinkles ($\%SE_w$, Visiometer® SV 600 USB), skin hydration (stratum corneum hydration, SCH, Corneometer® CM 825), melanin content (Mexameter® MX 18), biological elasticity (U_r/U_f, Cutometer® 580 MPA), and viscoelasticity (U_v/U_e, Cutometer® 580 MPA).

Skin irritation, a measurement of product safety, was assessed objectively by transepidermal water loss (TEWL, Tewameter® TM 210), and skin pH (Skin-pH-Meter® PH 905). In addition, a subjective evaluation of product safety was conducted by a dermatologist on a predetermined 0-3 scale, i.e., 0 = no sign of irritation, 1 = mild irritation, 2 = moderate irritation, and 3 = strong irritation, and a self-report diary.

Statistical analysis

Paired t-test was utilized to determine the difference of mean outcomes between tested and control sides, and between the mean outcomes of the test side at weeks 1-8 with baseline. The level of statistical significance was set at 0.05.

Results

Participants' characteristics

Thirty-three persons were eligible for enrollment. The vast majority (30, 91.9%) were females, with an average age of 39.7±6.6 years.

Baseline characteristics

Baseline characteristics were measured on both sides of the face before the beginning of the trial. The characteristics were found to be comparable between sides, except biological elasticity that the treatment side was significantly higher than the control side (Table 1).

Table 1. Baseline characteristics of the participants

Characteristics	Control side (mean±SD)	Treatment side (mean±SD)
Skin hydration: Capacitance (mg/cm^2)	65±10.8	66±9.8
Skin pH value	5.3±0.54	5.3±0.53
Skin whitening: Melanin content	319±91.0	315±81.0
Skin wrinkles: %SE$_w$	58±12.0	60±12.0
Skin firmness: Distensibility	0.73±0.140	0.75±0.150
Gross elasticity*	0.08±0.040	0.13±0.040
Viscoelasticity	1.74±0.620	1.80±0.480
Skin irritation: TEWL (g/hm^2)	18±4.3	18±5.1

*p<0.001 (control vs. treatment side)

Product efficacy

The efficacy of curcuminoids cream was evaluated by comparing the difference between treatment and control sides at each follow up week, and between the test sides at each follow up week with its baseline. The data are shown in Table 2.

Skin wrinkles: %SE$_w$ began to show significant difference between sides of application from the first follow up. In addition, wrinkles on treatment side showed significant improvement from baseline at the first week.

Skin hydration: SCH value began to show significant difference between sides of application and between after and before use from week one.

Melanin content: The skin melanin content began to show significant difference between sides of application from week 1. In addition, the treatment side showed significant melanin reduction from baseline from week 1 onward.

Gross elasticity: The result from Table 2 indicated that gross elasticity significantly improved for the treatment side compared with the control side from week 2 onward. The same effect was observed comparing to the baseline.

Skin viscoelasticity: From week one, skin viscoelasticity of the test side began to decrease significantly compared to baseline value. The difference between treatment and control sides were observed from week three onward.

Product safety

Table 3 showed that all TEWL values were lower than 40 g/(hm^2), indicating that curcuminoids cream was considered safe for the volunteers. In addition, the results showed the sign of reduced TEWL values on the treatment side over time. The values were significantly lower than those of the control side from week 4 onward.

Another measure of skin irritation is the pH value. The result in Table 3 indicated no significant change in skin pH of the volunteers, both between sides of application and comparison to the baseline. In addition, the average skin pH was found to be optimal, i.e., approximately 5.5, throughout the study.

Skin irritation was also determined by a dermatologist and volunteers' self-report. The result of both clinician's evaluation and self report showed no sign of skin irritation throughout the study (data not shown).

Table 2. Efficacy parameters of curcuminoids loaded SLN cream

	Mean±SD											
	Skin hydration		Melanin content		Skin wrinkles		Firmness		Gross elasticity		Viscoelasticity	
	Treatment	Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment	Control
Week 1	71.58±8.09 *,**	64.45±9.98	279±78.0 *,**	311±88.0	56±8.0 *,**	62±11.0	0.73±0.09	0.72±0.03	0.13±0.05	0.13±0.09	0.52±0.68 **	0.47±0.62
Week 2	71.88±6.12 *,**	60.94±7.34	268±74.0 *,**	303±84.0	53±6.0 *,**	64±9.0	0.55±0.07 *,**	0.84±0.04	0.20±0.12 *,**	0.12±0.09	0.16±0.02 **	0.16±0.02
Week 3	63.76±8.63 *,**	63.76±8.63	262±65.0 *,**	303±73.0	52±9.0 *,**	61±10.0	0.51±0.09 *,**	0.84±0.04	0.30±0.07 *,**	0.17±0.09	0.14±0.02 *,**	0.16±0.01
Week 4	76.39±6.28 *,**	64.97±7.01	262±77.0 *,**	309±80.0	48±5.0 *,**	62±11.0	0.54±0.11 *,**	0.82±0.05	0.20±0.12 *,**	0.10±0.04	0.13±0.02 *,**	0.14±0.02
Week 5	61.39±8.61 *,**	76.06±7.73	266±75.0 *,**	318±79.0	50±8.0 *,**	67±11.0	0.50±0.07 *,**	0.75±0.05	0.17±0.06 *,**	0.11±0.05	0.12±0.02 *,**	0.14±0.03
Week 6	76.67±6.77 *,**	58.94±7.34	272±78.0 *,**	314±77.0	44±5.0 *,**	65±8.0	0.49±0.07 *,**	0.74±0.04	0.25±0.05 *,**	0.19±0.05	0.20±0.08 *,**	0.24±0.08
Week 7	78.70±7.03 *,**	78.70±7.03	258±72.0 *,**	259±72.0	43±4.0 *,**	67±7.0	0.33±0.14 *,**	0.57±0.12	0.33±0.14 *,**	0.33±0.14	0.17±0.12 *,**	0.22±0.11
Week 8	85.09±4.31 *,**	61.45±6.72	251±75.0 *,**	318±84.0	42 2.0 *,**	68±10.0	0.25±0.09 *,**	0.53±0.13	0.55±0.12 *,**	0.42±0.11	0.14±0.07 *,**	0.24±0.15

*p<0.05 by paired t-test, compared with control side on the same week.
**p<0.05 by paired t-test, compared with baseline of the same side.

Table 3. Skin irritation parameters of curcuminoids loaded SLN cream

	Mean±SD			
	Transepidermal water loss (TEWL)		Skin pH	
	Treatment	Control	Treatment	Control
Week 1	18±5.0	18±4.0	5.5±0.39	5.5±0.36
Week 2	18±5.0	17±4.0	5.5±0.25	5.4±0.29
Week 3	17±5.0	17±4.0	5.5±0.22	5.5±0.25
Week 4	15±3.0*	16±3.0	5.4±0.25	5.4±0.28
Week 5	16±4.0*	17±4.0	5.4±0.19	5.4±0.23
Week 6	15±3.0*	16±3.0	5.4±0.21	5.4±0.21
Week 7	15±3.0*	16±4.0	5.4±0.20	5.3±0.20
Week 8	15±4.0*	16±4.0	5.3±0.20	5.4±0.16

*$p<0.05$ by paired t-test, compared with baseline of the same side.

Discussion and Conclusions

There has been no clinical trial study to examine the effect of curcuminoids as a topical anti-aging (Thornfeldt, 2005). Most studies focused on its systemic effect, and used only curcumin as the test product. This might be because of the stability problem in topical preparation. This study utilized SLN to overcome stability obstacle, hence the first to be able to examine curcuminoids efficacy as a topical anti-aging agent. In this study, the prominent effect of curcuminoids loaded SLN cream on the outcome measures were hypothesized to be contributed to active ingredient, i.e., curcuminoids, and its delivery carrier, SLN. The antioxidant property of curcuminoids improved skin wrinkles and exhibited skin whitening effect, while the enhancement of skin hydration, elasticity, and viscoelasticity were the result of the occlusive characteristic of SLN.

The effect of our test product on skin hydration and viscoelasticity began to emerge from week three onward compared to cream base. This finding was consistent with that of Wissing and Müller (2003) who also confirmed the higher effectiveness of cream containing SLN on skin hydrating and viscoelasticity compared to conventional cream. In addition to increased stability, the occlusion effect of SLN delivery system facilitated curcuminoids penetration by enhancing hydration of the stratum corneum (Jenning & Schäfer-Korting, 2000; Zur Muhlen et al., 1998). Therefore, the antioxidant effect of curcuminoids was evident after the first week of application.

In our study, the application of cream containing curcuminoids SLN resulted in significantly lower melanin content on the first week and showed a trend toward significantly lower melanin values afterward. This effect might be due to its skin lightening effect that was mentioned by Phan and colleagues (2001). However, another explanation to this finding might be the property of curcuminoids themselves. That is, stratum corneum of the skin acted as hydrolipid films, and was the main barrier for the highly lipophilic curcuminoids. A lipophilic characteristic of curcuminoids gave rise to possible fitting on the skin surface. Furthermore, curcuminoids showed a band of UV absorbance between 400-800 nm (Bosari et al., 2002). This would probably interfere with the Mexameter® readings, and resulting in the reduction of melanin index.

In this study, the efficacy of cream containing curcuminoids SLN mainly focused on skin wrinkles, hydration, color, elasticity and viscoelasticity. All parameters were measured with well-established techniques. Since various factors such as temperature, humidity, and skin temperature may interfere with equipment readings, all measurements

in this study were performed in a room with controlled environment at 22±5 ^{o}C and 50±5% relative humidity (Curdy et al., 2004; Levin & Maibach, 2005). Furthermore, cosmetic products such as creams and powders are known to alter skin moisture and pH value. Therefore, volunteers were instructed not to apply any cosmetic product to the appointment.

From our exit questionnaires, 93.93% of volunteers were more satisfied with cream containing curcuminoids SLN than cream base. They reported that they "felt" the test product could improve wrinkles, hydration, softness and firmness of their skin after two weeks (data not shown).

In conclusion, this study exhibited that curcuminoids loaded SLN cream was statistically significant in improving skin wrinkles, hydration, melanin content, biological elasticity, and viscoelasticity, compared with the cream base and the baseline from week 3 onward. The product showed no sign of skin irritation, both objectively and subjectively, throughout the study.

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