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# Physicochemical Characterization of Rutaecarpine-Loaded Microemulsion System

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**ABSTRACT** To develop an o/w microemulsion system containing poorly water-soluble rutaecarpine, the solubility of rutaecarpine in water, ethanol, various oils, and surfactants were investigated. Among the surfactants and oils tested, Tween 20/PEG 400 and castor oil were chosen as the surfactant system and oil phase of the microemulsion, as rutaecarpine was most soluble in them, respectively. Pseudoternary phase diagrams were constructed to obtain the concentration range of oil, surfactant, and cosurfactant for microemulsion formation, and the stability test of rutaecarpine delivered by microemulsion formation was then evaluated. Pseudoternary phase diagrams show that the areas of microemulsion appeared at those with 0–20% Smix (PEG 400/Tween80=60/40), 64–81% water, and 10–20% oil. The rutaecarpine (300 μg/g)-loaded microemulsion composed of 10.8% PEG 400, 7.2% Tween 80, 20% caster oil, and 72% water was physically and chemically stable for at least 6 months. Thus, the microemulsion system composed of castor oil, PEG 400, Tween 80, and water could be a stable dosage form for rutaecarpine.

**KEYWORDS** Microemulsion, Rutaecarpine, Stability

## INTRODUCTION

The dried, unripened fruit of *Evodia rutaecarpa* has been used as a remedy for gastrointestinal disorders (abdominal pain, dysentery), postpartum hemorrhage, headache, and amenorrhea. Rutaecarpine, a bioactive component isolated from *Evodia rutaecarpa*, is characterized by its antiplatelet activity and vasorelaxing action (Jiang et al., 2000; Sheu et al., 1996, 1998). However, it shows very low absorption, probably due to its poor water solubility. It has been attempted to improve the solubility of rutaecarpine by a cosolvent system (Wang et al., 1999), surfactants (Adeel & Luthy, 1995; Shiau et al., 1994), dissolved organic matter (Rebhun et al., 1998), inclusion complex (Archontaki et al., 2002), and solid dispersion (Verheyen et al., 2002; Watanabe et al., 2003).

Much attention has been paid to the application of microemulsion as a drug delivery system, since microemulsion systems are thermodynamically

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stable and form spontaneously by simply mixing various components (Malcolmson et al., 1998; Park et al., 1999). It was reported that the o/w microemulsion formed spontaneously by simple mixing could be employed as a drug delivery system (Corswant et al., 1998). Some of the advantages of o/w microemulsion as a drug delivery system include the improvement of drug solubilization, potential for oral and parenteral use, and production on a large industrial scale without high-energy homogenization.

In this study, to develop a microemulsion system containing poorly water-soluble rutaecarpine, the solubility of rutaecarpine in water, ethanol, various oils, and surfactants was investigated. Pseudoternary phase diagrams were then constructed to obtain the concentration range of oil, surfactant, and cosurfactant for microemulsion formation. Furthermore, the physical and chemical stability of rutaecarpine delivered by microemulsion formation was evaluated.

# MATERIALS AND METHODS

## Materials

Rutaecarpine was supplied by the Institute of Organic Synthesis, College of Pharmacy in Yeungnam University (Kyungsan, South Korea). Polyethylene glycol 400 (PEG 400) and Poloxamer 188 were purchased from Yakuri Pure Chemicals Co. (Kyoto, Japan) and BF Goodrich (Breesville, OH), respectively. Tween 80, Tween 20, Span 20, and Span 80 were purchased from Junsei Chemical Co. (Tokyo, Japan), respectively. Various oils such as sunflower seed oil, castor oil, corn oil, sesame oil, soybean oil, cotton oil, and peanut oil were obtained from Sigma Aldrich Chemical Co. (Milwaukee, WI). All other chemicals were of reagent grade and used without further purification.

## Aqueous Solubility of Rutaecarpine

Excessive eutectic drugs (100 mg) were added in 1 mL of water, ethanol, various oils, or surfactant. They were shaken at room temperature for 7 days and centrifuged at 25°C at 14,000 rpm for 10 min (Eppendorf centrifuge 5415, Germany) to remove the undissolved drugs. Aliquots of supernatant were taken, appropriately diluted with acetonitrile, and analyzed by HPLC (Jasco PU-980) equipped with an Inertsil ODS-3 $C_{18}$ column (GL science, 0.5 Mm, 15 cm × 0.46 cm i.d.) and UV detector (Jasco UV-975). The mobile phase consisted of acetonitrile, water, and orthophosporic acid (60:40:0.1, volume ratio). The eluent was monitored at 227 nm with a flow rate of 1 mL/min (Gao et al., 1998; Ko et al., 1994).

## Construction of Phase Diagrams

Pseudoternary phase diagrams were constructed to obtain the components and their concentration ranges, which can result in the existence of large areas of microemulsion without the drug Gao et al., 1998. The surfactant-cosurfactant mixtures (Smix) were prepared by mixing surfactant (Tween 80) and cosurfactant (PEG 400) in a fixed weight ratio of 60:40. Aliquots of each surfactant-cosurfactant mixture (Smix) were then mixed with oil and finally with aqueous phase. Mixtures were shaken and kept at 25°C to obtain equilibrium. The physical states were represented on a pseudo-ternary phase diagram with one axis representing water, one representing oil, and the third representing $S_{mix}$ (Gao et al., 1998; Ko et al., 1994).

## Preparation and Stability of Rutaecarpine-Loaded Microemulsion

### *Preparation of Rutaecarpine-Loaded Microemulsion*

Based on the phase diagram experiments, the oily phases with soluble rutaecarpine were mixed in with various ratios of Smix and the water, and passed through a high-pressure homogenizer (Emulsiflex®-B3 Avestin Inc., Ottawa, Canada).

### *Physical Stability*

The particle size of oil droplets in rutaecarpine (300 μg/mL)-loaded microemulsion was measured at 25±1°C by photon correlation spectroscopy. A light scattering spectrophotometer (LPA-3100; Otsuka Electronics, Osaka, Japan) equipped with a data processing unit (LPA-3000; Otsuka Electronics) was used for characterizing particle size in the 3–5000 nm range using the dynamic light scattering method.

For measuring particle size by photon correlation spectroscopy, microemulsions were diluted with aqueous phase, which is the continuous phase of microemulsions. The continuous phase of microemulsions did not exhibit light scattering and the particle size of diluted microemulsion (up to 20-fold dilution) was not significantly changed (Constantinides & Yiv, 1995; Park & Kim, 1999).

### *Chemical Stability*

The chemical stability of rutaecarpine-loaded microemulsion was evaluated by the contents of rutaecarpine in the microemulsion over 6 months at 25°C.

## RESULTS AND DISCUSSION

Rutaecarpine showed very low absorption, probably due to its poor water solubility. It has been attempted to improve the solubility of rutaecarpine by a cosolvent system (Wang et al., 1999), surfactants (Adeel & Luthy, 1995; Shiau et al., 1994), dissolved organic matter (Rebhun et al., 1998), inclusion complex (Archontaki et al., 2002), and solid dispersion (Verheyen et al., 2002; Watanabe et al., 2003). However, the inclusion complex and solid dispersion methods could not greatly improve the solubility of rutaecarpine (Adeel & Luthy, 1995; Archontaki et al., 2002; Rebhun et al., 1998; Shiau et al., 1994; Verheyen et al., 2002; Wang et al., 1999; Watanabe et al., 2003). Thus, in this study, an alternative method, the microemulsion system, has been attempted to improve the solubility of rutaecarpine.

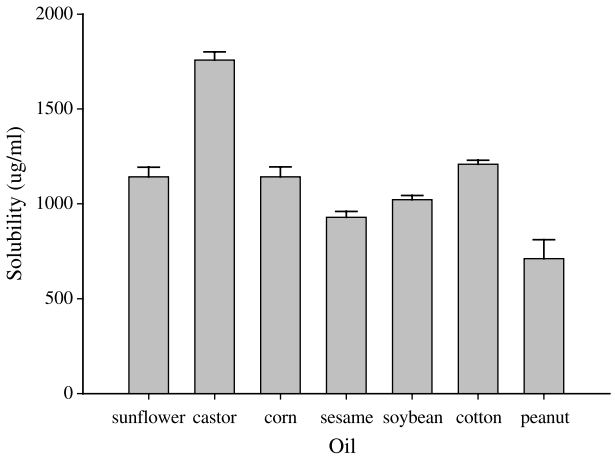


**FIGURE 1** **Solubility of Rutaecarpine in Various Oils at 25°C.**

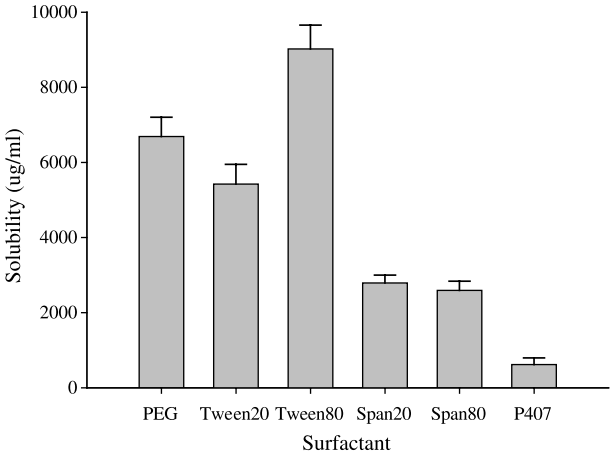


**FIGURE 2** **Solubility of Rutaecarpine in Various Surfactants at 25°C.**

To investigate the solubility of a poorly water-soluble rutaecarpine in various mediums, excess rutaecarpine was added in water; ethanol; various oils such as sunflower seed oil, castor oil, corn oil, sesame oil, soybean oil, cotton oil, and peanut oil; and various surfactants such as PEG 400, Tween 80, Tween 20, Span 20, Span 80, and 5% poloxamer 188, respectively, and then the solubility of rutaecarpine was evaluated.

The solubility of rutaecarpine was about $0.05 \pm 0.02$ and $1358 \pm 15$ μg/mL in water and ethanol, indicating that rutaecarpine was poorly soluble in water, which was insufficient to provide the desired amount in a solution for pharmaceutical dosage form with enhanced bioavailability (Park & Kim, 1999). As shown in Fig. 1, among the oils tested, drug was most soluble in castor oil ($1758 \pm 8$ μg/mL). On the other hand, the solubility of rutaecarpine in Tween series and PEG 400 was higher than the solubility in the Span series and poloxamer solution (Fig. 2). For the development of a rutaecarpine-loaded microemulsion, castor oil, Tween 80, and PEG 400 were selected as an oil phase, surfactant, and cosurfactant, respectively, because these materials with high solubility of rutaecarpine could be used in the pharmaceutical dosage form with enhanced bioavailability (Gao et al., 1998; Khan et al., 1999).

Phase studies were carried out to investigate the effect of surfactant to cosurfactant ratio on the extent of stable o/w microemulsion region (Gao et al., 1998; Park & Kim, 1999). The microemulsions in the present study were formed spontaneously at ambient temperature when their components were brought into contact with each other (Tarr & Yalkowsky, 1989).

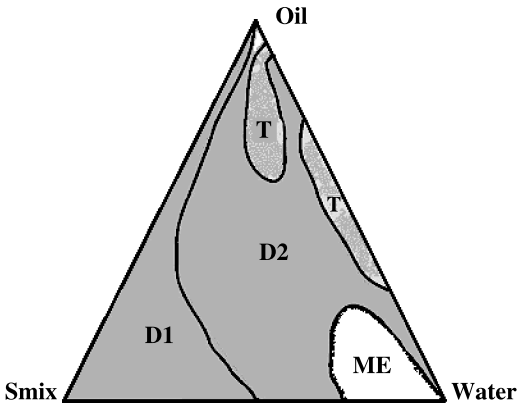


**FIGURE 3** **Pseudoternary Phase Diagrams of Microemulsion Composed of Oil (Castor Oil), Surfactant System (PEG 400-Tween 80) and Water. ME, Microemulsion Region; D1, Transparent Double-Layer; D2, Opaque Double-Layer; T, Triple-Layer.**

The areas of microemulsion appeared at those with 0–20% Smix (PEG 400/Tween80=60/40), 64–81% water, and 10–20% oil (Fig. 3). To select the optimal formulation, four randomly chosen samples within the areas of microemulsion (Smix/water/oil=18/72/10%, 16/64/20%, 9/81/10%, 8/72/20%) were kept at 25°C, and their visual appearances were investigated for 4 weeks. The three samples (Smix/water/oil=18/72/10%, 16/64/20%, and 9/81/10%) showed the oil droplet on the upper surface of preparations. However, the sample (Smix/water/oil=8/72/20%) did not show phase separation for 4 weeks. Thus, Smix/water/oil (8/72/20%) was selected as a formula for rutaecarpine-loaded microemulsion system.

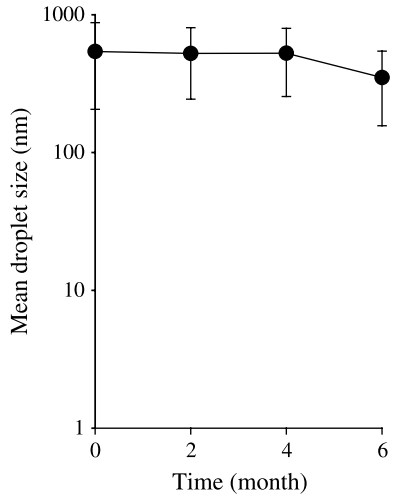


**FIGURE 5** **Mean Particle Size of Rutaecarpine-Loaded Microemulsion at 25°C. The Rutaecarpine (300 μg/g)-Loaded Microemulsion was Composed of 10.8% PEG 400, 7.2% Tween 80, 20% Caster Oil, and 72% Water. Each Value Represents the Mean±S.E. (n=3).**

Figures 4 and 5 show the particle size distribution and mean droplet size distribution of rutaecarpine-loaded microemulsion observed for 6 months at 25°C, respectively. The initial mean droplet size of rutaecarpine-loaded microemulsion was about 500 nm, and the droplet sizes did not change significantly. Furthermore, the chemical stability of rutaecarpine-loaded microemulsion was evaluated by the drug content in microemulsion over 6 months at 25°C (Gao et al., 1998). The drug content of more than 97% of initial amount in the microemulsion did not change significantly, indicating

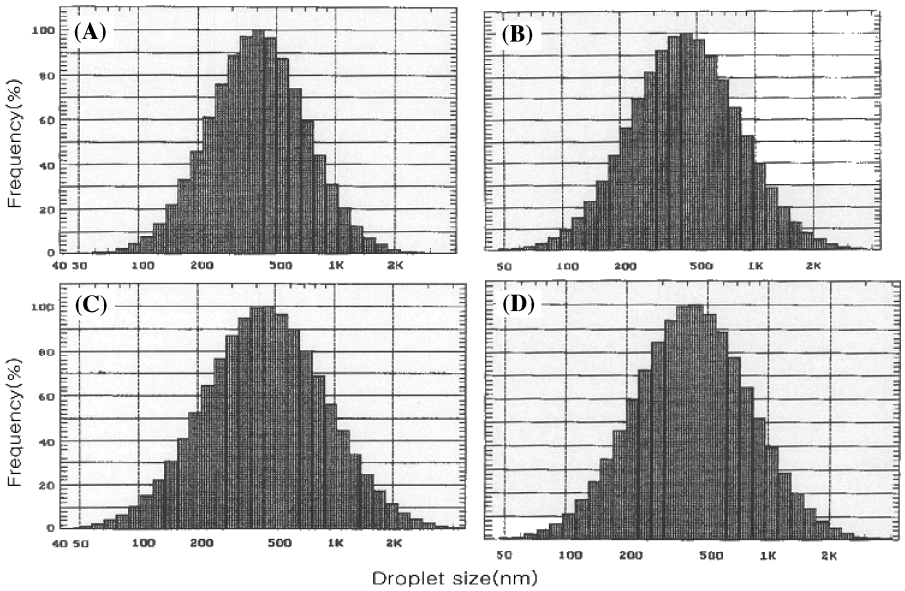


**FIGURE 4** **Particle Size Distributions of Rutaecarpine-Loaded Microemulsion at 25°C. The Rutaecarpine (300 μg/g)-Loaded Microemulsion was Composed of 10.8% PEG 400, 7.2% Tween 80, 20% Caster Oil, and 72% Water. A, 0 Month; B, 2 Months; C, 4 Months; D, 6 Months.**

that rutaecarpine-loaded microemulsion could be chemically stable for at least 6 months. The results suggest that the rutaecarpine-loaded microemulsion was very stable, being a proper carrier for rutaecarpine (Park & Kim, 1999; Tarr & Yalkowsky, 1989).

## CONCLUSION

It is concluded the rutaecarpine-loaded microemulsion composed of castor oil, PEG 400, Tween 80, and water could be a stable parenteral dosage form for rutaecarpine. Further study on the parenteral and oral pharmacokinetics of rutaecarpine-loaded microemulsion will be performed.

## ACKNOWLEDGMENT

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