Effect of tamarind (*Tamarindus indica* L.) seed polysaccharide on physical properties of itraconazole-loaded nanoemulsions

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ABSTRACT

Recent trend toward the use of plant based and natural products demands the replacement of synthetic additives with natural ones. The objective of this study was to investigate the effect of tamarind (*Tamarindus indica* L.) seed polysaccharide on physical properties of itraconazole-loaded nanoemulsions prepared by simple homogenization (20,000 rpm, 15 min). Itraconazole, a low aqueous solubility substance, was used as a model drug. Castor oil, oleic acid and caprylic capric triglyceride at the concentration of 20-50% w/w were used as an oil phase. Polysorbate 80 and sorbitan sesquioleate 83 were used as emulsifier whereas tamarind seed polysaccharide was used as co-emulsifier. The formulations containing 30% oil and 20-30% w/w polysorbate 80 provided nano-sized emulsions (100-500 nm) while the use of tamarind seed polysaccharide alone up to 3% as emulsifier could not produce the nanoemulsions. The incorporation of 1-3% tamarind seed polysaccharide in the formulations containing 20% polysorbate 80 decreased the size of emulsion droplets. For the formulations containing 3% tamarind seed polysaccharide, the droplet size of nanoemulsions decreased with the increase of concentration of polysorbate 80. The addition of itraconazole did not affect droplet size of the nanoemulsions. The zeta potentials ranged from -16.97 to -16.41 mV. The viscosity was in the range of 1,067-1,595 cPs. The nanoemulsions were stable for at least 14 days at 8°C. The results pointed that tamarind seed polysaccharide, a biodegradable, edible and harmless biopolymer, could be applied as co-emulsifier in nanoemulsions.

Keywords: Tamarind seed, *Tamarindus indica* L., Co-emulsifier, Physical stability, Itraconazole, Nanoemulsions

1. INTRODUCTION

The use of plant-based and natural products demands the replacement of synthetic additives with natural ones becomes interesting due to their safety, biodegradability, biocompatibility and non-toxicity. Tamarind (*Tamarindus indica L.*), belongs to the family Leguminosae and grows naturally in many tropical and sub-tropical regions. Tamarind is an important food resource for Thai population. Flower and leaf are consumed as vegetables while the germ obtained from the seed is used for manufacturing Tamarind gum [1]. Tamarind fruit is a pulpy mass of a light reddish-brown color, changing with age to a dark brown, containing some branching fibers and numerous reddish brown, smooth, oblong or quadrangular, compressed seeds, each enclosed in a tough membrane. Tamarind seeds contain tamarind seed polysaccharide which was reported to use as binding agent, emulsifier, suspending agent, sustained release agent, hydrogels, mucoadhesive agent, rectal drug delivery and nasal drug delivery [2-3]. Itraconazole (ITZ), a triazole antifungal agent, is poorly water-soluble drug and its absorption in gastrointestinal tract is low. The various methods that could increase the solubility of ITZ included complexation with β-cyclodextrin [4], use of mesoporous silica [5], solid dispersion [6] and nanoemulsions [7]. Nanoemulsions are often referred to emulsions with droplet sizes in the nanometric scale, generally 100-500 nm [8]. It has been found that the use of nanoemulsions for oral administration to increase the bioavailability of poorly water-soluble drugs due to an enhancement of the intestinal absorption of the drug is well documented. The previous study of nanoemulsions could contain 140 μg/mL of ITZ in caprylic/capric triglyceride as internal phase [7,9]. Therefore, the aim of this study was to investigate effect of tamarind seed polysaccharide as co-emulsifier on physical properties of itraconazole-loaded nanoemulsions prepared by simple homogenization and evaluate the changes in physical properties, such as droplet size, viscosity, zeta potential as well as physical stability, to explore their potential in pharmaceutical industry.

2. MATERIALS AND METHODS

Materials

Tamarind fruits were procured from local market (Nakhon Pathom, Thailand). Itraconazole raw material used in this study was purchased from Nosch Labs Private (India). All other chemicals used were of analytical grade and used as received. Deionized water was used as an aqueous phase in all preparations.

Extraction of tamarind seed polysaccharide

Tamarind seed kernel (TKP) powder (20 g) was added to 1000 mL of cold distilled water to prepare slurry. The slurry obtained was allowed to stand for 4 hours so most of the large particle size (TKP portion) was settled down. The precipitate was resuspended in water, heated to 80°C for 30 min and kept overnight at room temperature. A clear solution was separated and poured into ethanol with continuous stirring. The precipitate obtained was dried and stored in a desiccator.

Preparation of nanoemulsions

Castor oil, oleic acid and caprylic capric triglyceride at the concentration of 20-50% w/w were used as an oil phase. Polysorbate 80 and sorbitan sesquioleate 83 were used as emulsifier while tamarind seed polysaccharide was used as co-emulsifier. Itraconazole, a low aqueous solubility substance, was used as a model drug. The nanoemulsions were prepared by using homogenizer (Ultra-Turrax® T50 Basic, IKA, Germany) at a speed of 20,000 rpm for 15 min in an ice-bath to avoid over-heating.

Determination of physical properties

Droplet size measurement

The prepared nanoemulsions were dispersed or diluted in deionized water with gentle stirring. The droplet size of nanoemulsions was investigated by static light scattering method (Laser scattering particle size distribution analyzer LA-950, Horiba, Japan). The median particle size was measured under continuous stirring. The measurements were repeated at least three batches of nanoemulsions.

Zeta potential measurement

The zeta potential analyzer (ZetaPlus, Brookhaven, USA) was used to measure zeta potential of the prepared nanoemulsions. Nanoemulsions were dispersed in deionized water at the ratio of 1:50 (v/v) and the electric field applied was 1 V. The measurement were performed three times and reported as the average and standard deviation.

Stability of nanoemulsions

All nanoemulsions, kept in the glass vials, were separated into two groups; the first group was stored in a refrigerator at 8°C while the other group was subjected to temperature cycling test by keeping at 4°C for 24 h and at 40°C for 24 h. The stability of nanoemulsions was examined by calculation of percent creaming [7, 9]. A greater

value of the percent creaming is an indication of a more stable emulsion. The size of the nanoemulsion droplets after stability test was also measured by a static light scattering method as described above.

3. RESULTS AND DISCUSSION

The effect of emulsifier types in the concentration of 20% w/w on droplet size of emulsions in various oils (30% w/w) is shown in Figure 1A. Castor oil, oleic acid and caprylic/capric triglyceride were used in this study. In all types of oil, polysorbate 80 alone provided nano-sized emulsions (100-500 nm) while the use of sorbitan monooleate 83 alone and the combination of polysorbate 80 and sorbitan monooleate 83 in the ratio of 1:1 could not produce the nanoemulsions. In addition, tamarind seed polysaccharide (TSP) was investigated for emulsifying activity. It was found that TSP alone up to 3% w/w could not produce nano-sized emulsions by the simple homogenizer (data not shown). The effect of oil concentration was investigated when polysorbate 80 was used as emulsifier. It was found that the increase in oil concentration tended to increase the size of the emulsion (Figure 1B). Due to high solubility of itraconazole and stability of nanoemulsion, 30% w/w castor oil was selected to prepare itraconazole-loaded nanoemulsions. The effect of co-emulsifying system between polysorbate 80 and TSP on droplet size of nanoemulsions is summarized in Figure 2. When 3% w/w TSP was incorporated in the formulation as co-emulsifier, the droplet size of nanoemulsions decreased with the increased concentration of polysorbate 80 (Figure 2A). The effect of TSP concentration as co-emulsifier is summarized in Figure 2B. When polysorbate 80 used was 20% w/w, the increasing of TSP concentration up to 3% w/w resulted in a decrease in droplet size of nanoemulsions (Figure 2B). Using excess biopolymer, however, may lead to the flocculation of emulsion droplets via depletion and bridging mechanism [10]. Therefore, it is suggested that an appropriate amount of polymer should be considered in nanoemulsion formulations.

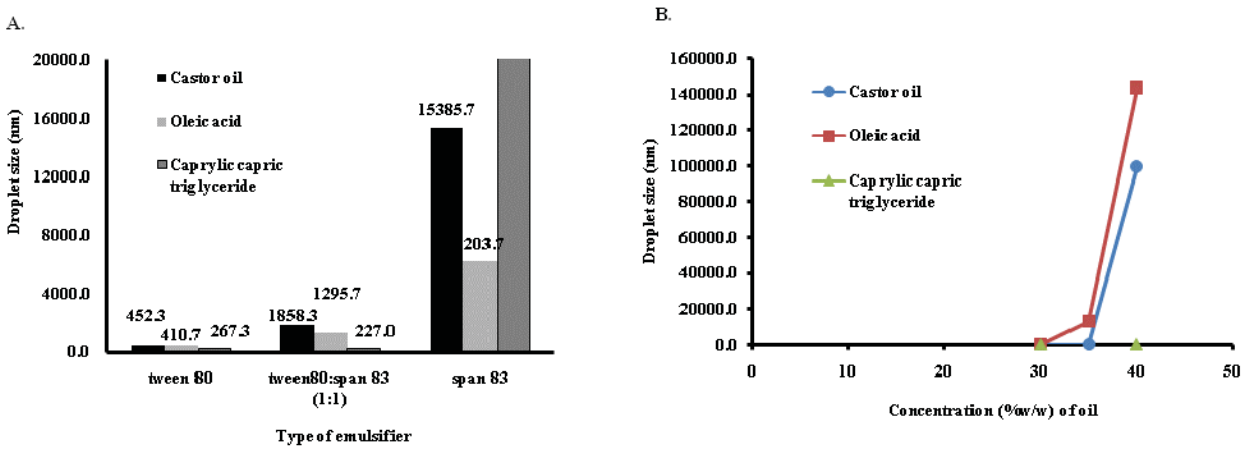

Figure 1. Effect of (A) emulsifier types and (B) oil concentration on droplet size of emulsions. The various oils used were 30% w/w (A) and the total emulsifier used was 20% w/w (B).

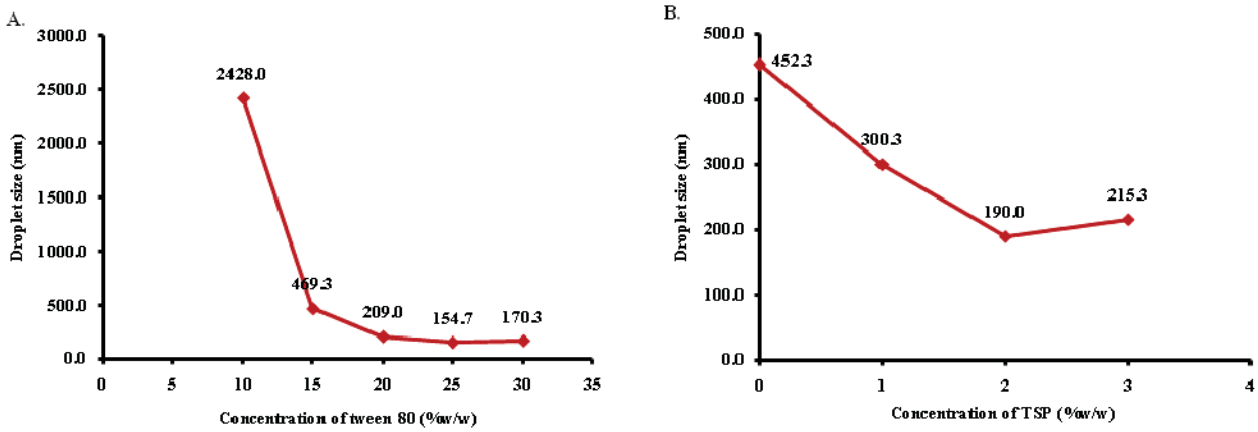

Figure 2. Effect of co-emulsifying system between polysorbate 80 and TSP on droplet size of nanoemulsions. (A) polysorbate 80 in various concentrations and 3% w/w TSP and (B) TSP in various concentrations and 20% w/w polysorbate 80. Castor oil was used in the concentration of 30% w/w.

The addition of itraconazole slightly decreased droplet size of the nanoemulsions. The zeta potentials ranged from -15.39 to -29.38 mV. The viscosity was in the range of 248.5-4,909 cPs. The percent creaming was 100%. The prepared nanoemulsions were subjected to stability test and found that the nanoemulsions were stable for at least 14 days at 8°C. After the temperature cycling test by storage at 4°C for 24 h and at 40°C for 24 h was performed, it was found that the formulation containing 30% w/w castor oil, 20% w/w polysorbate 80 and 1-3% w/w TSP was stable for 3 cycles.

4. CONCLUSIONS

Type and concentration of oils and emulsifiers affected stability of nanoemulsions prepared by simple homogenization. The decrease in the concentration of emulsifier and an increase in the oil concentration affected the stability of nanoemulsions. For the co-emulsifying system between polysorbate 80 and TSP, the increase in TSP concentration up to 3% w/w tended to decrease the droplet size of nanoemulsions. The total concentration of emulsifier also affected the size of the nanoemulsions. The formulation containing castor oil of 30% w/w, polysorbate 80 of 20% w/w and TSP of 1-3% w/w provided stable nanoemulsions. The results pointed that tamarind seed polysaccharide, a biodegradable, edible and harmless biopolymer, could be applied as co-emulsifier in nanoemulsions.

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