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Validated high performance liquid chromatography for simultaneous determination of stability of madecassoside and asiaticoside in film forming polymeric dispersions

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ABSTRACT

The objective of the work was to validate the high performance liquid chromatography for simultaneous determination of stability of madecassoside and asiaticoside in *Centella asiatica* (L.) Urb., Apiaceae, extract-loaded film forming polymeric dispersions. High performance liquid chromatography method was validated in five topics: linearity and range, limit of detection and limit of quantitation, specificity, precision, and accuracy. Results showed the method had a good linearity ($R^2 > 0.9990$) in the range of 5–150 µg/ml and specific. The limit of detection and limit of quantitation of madecassoside were 81 and 245 ng/ml and asiaticoside were 21 and 64 ng/ml, respectively. The percent relative standard deviation of intraday and interday precision were less than 1 and 3%, respectively. The accuracy presented as percent recovery was 101.54–103.29% for madecassoside and 100.39–102.58% for asiaticoside. This validated high performance liquid chromatography method was used to determine the stability of the formulation containing *Centella asiatica* extract. *Centella asiatica* extract-loaded film forming polymeric dispersions used Eudragit[®] RS 30D and Eudragit[®] RL 30D as film former, glycerin as plasticizer, and absolute ethanol as solvent and penetration enhancer. Three formulations with different ratio of Eudragit[®] RS 30D and Eudragit[®] RL 30D were prepared and stored for 90 days at 4 °C, 25 °C, and 40 °C. Stability results showed that almost all of the formulations were unstable at 25 °C and 40 °C. Except, two of three formulations were stable at 4 °C. However, the formulation was further developed to improve the stability of madecassoside and asiaticoside in the formulation.

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Introduction

Centella asiatica (L.) Urb. is a well-known herbal medicinal plant in the family of Apiaceae. In Thailand, it is listed in National Essential Drug List; topical cream containing 70% ethanol extract of dried *C. asiatica* in a concentration of 7%w/w is used for wound healing purpose. Currently, *C. asiatica* is commonly used for the treatment of dermatological diseases including bacterial infection, psoriasis, scleroderma, and wound. Furthermore, antioxidant activity is reported as well (Bylka et al.,

2014). Other biological and pharmacological effects are also reported. It is used as an adaptogen, antibiotic, blood-purifier, central nervous system relaxant, detoxifier, diuretic, emmenagogue, laxative, peripheral vasodilator, and sedative (Khare, 2007). It contains various chemical compounds including pentacyclic triterpenoids called centelloids such as madecassic acid, asiatic acid, and its glycosides; madecassoside and asiaticoside is a main chemical compound that exhibits wound healing property. The chemical structures of madecassoside (1) and asiaticoside (2). *Centella asiatica* also contains other compounds e.g., asiaticoside C, asiaticoside D, asiaticoside E, asiaticoside F, centellasaponin B, centellasaponin C, isothankunic acid and oleanane-type saponins (e.g. terminolic acid and centellasaponin D) (Bylka et al., 2014).

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