

## Article

# Optimisation of Stingless Bee Honey Nanoemulsions Using Response Surface Methodology

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**Abstract:** Nanoemulsions (NEs) have been used in a wide range of products, such as those produced by the food, cosmetics, and pharmaceutical industries, due to their stability and long shelf life. In the present study, stingless bee honey (SBH) NEs were formulated using SBH, oleic acid, tween 80, glycerol, and double-distilled water. SBH NEs were prepared using a high-pressure homogeniser and were characterised by observing their stability and droplet size. Fourier Transform-Infrared (FTIR) analysis was used to observe the functional groups of the SBH NEs after being subjected to high-pressure homogenisation. Transmission Electron Microscopy (TEM) images were then used to confirm the particle size of the SBH NEs and to investigate their morphology. The effects of the independent variables (percentage of oleic acid, storage time, and storage temperature) on the response variables (particle size and polydispersity index) were investigated using the response surface methodology, along with a three-level factorial design. The results showed that the models developed via the response surface methodology were reliable, with a coefficient of determination ( $R^2$ ) of more than 0.90. The experimental validation indicated an error of less than 10% in the actual results compared to the predicted results. The FTIR analysis showed that SBH NEs have the same functional group as SBH. Observation through TEM indicated that the SBH NEs had a similar particle size, which was between 10 and 100 nm. Thus, this study shows that SBH NEs can be developed using a high-pressure homogeniser, which indicates a new direction for SBH by-products.

**Keywords:** stingless bee honey; nanoemulsions; high-pressure homogeniser; response surface methodology; oleic acid



**Citation:** Rozman, A.S.; Hashim, N.; Maringgal, B.; Abdan, K. Optimisation of Stingless Bee Honey Nanoemulsions Using Response Surface Methodology. *Foods* **2021**, *10*, 2133. <https://doi.org/10.3390/foods10092133>

Academic Editors: Federico Marini and Alessandra Biancolillo

Received: 5 July 2021

Accepted: 4 August 2021

Published: 9 September 2021

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## 1. Introduction

Stingless bees are highly eusocial insects and can be found in tropical and subtropical regions. They belong to the Hymenoptera order, under the *Apidae* family and *Meliponini* sub-family. Similar to *Apis* bees, the stingless bees also produce and store honey in their hive [1]. Studies have shown that stingless bee honey (SBH) contains antimicrobial activity against bacteria, yeast, and fungal strains [2]. Further, SBH is considered to be a premium product with a market value that may reach up to \$100/kg, which is approximately twice the price of the honey of the *Apis* bee, which ranges from \$20 to \$40/kg [3]. In Malaysia, the SBH industry is expanding due to its potential and benefits. According to the Malaysian Department of Agriculture [4], the total farming area of stingless bees was reported to be about 1606.6 ha in 2017 and this figure was expected to increase yearly.

The composition of SBH consists of a mixture of carbohydrates, glucose, fructose, amino acids, organic acids, minerals, aromatic substances, vitamins, pigments, beeswax, and pollen, all of which affects its colour, smell, and taste. In addition, water is one of the main compositional elements of SBH and is considered a highly important feature that can determine the viscosity, specific weight, maturity, flavour, and crystallisation properties [5].