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ANTIOXIDANT AND STABILITY STUDIES OF FLORAL ANTHOCYANIN IN *CLITORIA TERNATEA* EXTRACT

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ABSTRACT

Clitoria ternatea, commonly known as butterfly pea is found in great abundance in Malaysia. In this project, blue aqueous anthocyanin extract of flower petals of *Clitoria ternatea* was used to explore its potential to be used as antioxidant and as colourant in food, cosmetic and pharmaceutical industries. The antioxidant capacity of blue *Clitoria ternatea* anthocyanin extract (CTAE) and factors affecting its stability were investigated. CTAE was found to contain 171.70 ± 38.27 mg/g of total phenolics and 0.70 ± 0.09 mg/g of total anthocyanins. UV-Vis spectroscopic studies of CTAE showed the characteristics of polyacylated anthocyanins with free OH group on C-5 position. The colour of CTAE varied with changes in pH. The anthocyanin pigment gave intense red, violet, blue, blue green, green and yellow at pH ≤ 2.5, 3-4, 5-6, 7-9, 9-10 and 10-12, respectively. Such variation in colour has been attributed to structural transformation of anthocyanin in response to changes in pH. CTAE was evaluated for antioxidant capacity by using three antioxidant assay methods namely DPPH radical scavenging method, reducing power, and FTC-linoleic acid assays. It was found that blue CTAE at the optimal concentration of 1.00 mg/ml exhibited 64% radical scavenging activity and 48% reducing power relative to α-tocopherol. It showed 90% inhibition of lipid peroxidation relative to α-tocopherol at a concentration of 2.10 mg/ml. The solvent-solvent partition extract of CTAE at a concentration of 1.00 mg/ml showed 71% DPPH radical scavenging activity. Effects of temperature, light, pH and preservative at different storage period on the stability of CTAE were investigated by measuring the absorbance at the wavelength of maximum absorption (λ_{max}) in the visible region. The overall thermal stabilities of anthocyanins in blue CTAE decreased in the order of 5°C>45°C>27°C>37°C. The colour of CTAE remained blue for at least one year at 5°C. The degradation profile of CTAE did not follow first order kinetics at the temperature range of 5-45°C. However, first order kinetics were observed at temperatures 70, 100 and 160°C, with half-lives of 315, 77 and 6 minutes, respectively. Light exposure of anthocyanins at 27°C did not affect the colour stability greatly. Effect of benzoic acid on the stability was also investigated at 27°C and 37°C. It was found that the presence of benzoic acid caused a hyperchromic effect on the absorbance of anthocyanins in the visible region and lengthen the degradation process at 27°C and 37°C to about 70 and 20 days, respectively. It was observed that pH influenced the colour stability of CTAE on storage at 27°C and 37°C. Acidic solutions showed longer storage stabilities than alkaline pHs. The solutions retained 70-80% of its colour on day 60 at 27°C. Isolation and characterization of anthocyanin from CTAE have also been conducted

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CHAPTER 1

INTRODUCTION

Colour affects every moment of our lives, our daily moods and our perception of quality of material and also the foods. Red anthocyanins (E163) are the natural sources of some currently permitted natural foods colourants commonly found in fruits such as strawberries, cherries and grapes (Wissgott & Bortlik, 1996). Most of food colourant in food manufacturing used natural colourant such as strawberry in jam jar. We can see clearly the colour indicates the flavour that we will taste. However, an artificial colourant sometimes looks bright but looks inferior for the food.

Antioxidants are molecules that in lower concentration can slow or prevent the oxidation of other chemicals. Oxidation is a redox chemical reaction that transfers electrons from a substance to an oxidising agent. Oxidation reaction can involve the production of free radicals, which can form dangerous chain reactions. Antioxidants can terminate these chain reactions by removing radical intermediates and can inhibit other oxidation reactions by being oxidised themselves (Pokorny *et al*., 2001).

In recent years, one of the areas, which have attracted a great deal of attention, is antioxidants in the control of degenerative diseases in which oxidative damage has been implicated. Several plant extracts and different classes of phytochemicals have been shown to have antioxidant activity (Cao *et al*., 1996; Oomah & Mazza, 1996; Wang *et al*., 1996; Duh & Yen, 1997; Abassi *et al*., 1998; Bergman *et al*., 2001). Natural