



UNIVERSITI PUTRA MALAYSIA

**PHYSICO-CHEMICAL CHARACTERISTICS OF ROSELLE
(HIBISCUS SABDARIFFA L.) CALYX AND EFFECT OF
PROCESSING AND STORAGE ON THE STABILITY OF
ANTHOCYANINS IN ROSELLE JUICE**

WONG PENG KONG

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By

WONG PENG KONG

**Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia,
in Fulfilment of Requirement of the Degree of Master of Science**

November 2002



**Specially Dedicated
To
My Family**

Abstract of thesis submitted to the senate of Universiti Putra Malaysia in fulfillment of the requirement for the degree of Master Science

PHYSICO-CHEMICAL CHARACTERISTICS OF ROSELLE (*HIBISCUS SABDARIFFA* L.) CALYX AND EFFECT OF PROCESSING AND STORAGE ON THE STABILITY OF ANTHOCYANINS IN ROSELLE

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November 2002

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Faculty: Food Science and Biotechnology

The main purpose of this project was to focus on determining the physico-chemical properties of roselle calyx, to develop a processing method that would optimise the colour stability and determining the influences of different treatments on roselle juice during storage. The physico-chemical characteristics of roselle included size, weight, pH, titratable acidity, soluble solids, anthocyanin contents, colour density, polymeric colour, percent polymeric colour and browning index were studied. Succinic acid and glucose were the major organic acid and sugar found present in roselle with 0.51 g/100g and 1.29 g/100g, respectively. The most biologically effective natural antioxidant vitamins such as ascorbic acid, β -carotene and lycopene were also determined and quantified by high performance liquid chromatography (HPLC). The amounts of ascorbic acid, β -carotene and lycopene contents were 141.09 mg/100 g, 1.88 mg/100 g and 164.34 μ g/100 g, respectively. Two main anthocyanins namely delphinidin-3-sambubioside and cyanidin-3-

sambubioside were present in roselle detected by thin layer chromatography (TLC) and HPLC.

The optimum conditions for hot water extraction (HWE) of roselle juice was determined by using Response Surface Methodology (RSM). Time-temperature combinations in the range of 30-300 minutes and 30-90 °C were the independent variables and their effects on anthocyanins content, colour density and ascorbic acid were evaluated. The results implied an optimum juice extraction condition for HWE to be at 3.5 hours at 60 °C. The juice obtained from HWE method was then compared with juices obtained from hot water blending (HWB) method, cold water blending (CWB) method and screw press (SP) method for their physico-chemical characteristics and sensory evaluation. Results of the study indicated that HWE method was found to be the most effective extraction method resulting in high anthocyanins content and ascorbic acid. Sensory evaluation also showed that juice obtained from HWE method was higher in quality in terms of taste, colours and flavour. Juice produced through SP method was poor in quality and sensory result compared with other.

Four different treatments were given to the sample roselle juice: control (no additions), ascorbic acid, cysteine hydrochloride and sodium metabisulphite. The influence of storage at 4 ± 2 °C, 27 ± 2 °C and 40 ± 2 °C was investigated. Losses in anthocyanins and ascorbic acid were very small during 12 weeks storage at 4 °C as compared to 27 °C and 40 °C. Higher temperature caused anthocyanin degradation more rapidly. The addition of sodium metabisulphite in roselle juice slowed down the loss of anthocyanins and correspondingly decreased the rate of polymeric colour.

Addition of cysteine hydrochloride had only slight effect on colour stability and addition of ascorbic acid seemed to increase the degradation on anthocyanin contents.

Addition of cysteine hydrochloride in roselle juice prevents ascorbic acid loss during storage.

Abstrak tesis yang dikemukakan kepada senat Universiti Putra Malaysia
sebagai memenuhi syarat Master Sains

**SIFAT-SIFAT FIZIKO-KIMIA KALYX ROZEL (*HIBISCUS SABDARIFFA*
L.) DAN KESAN PEMROSESAN DAN PENYIMPANAN KE ATAS
KESTABILAN ANTHOSIANIN DALAM JUS ROZEL**

Oleh

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Tujuan utama penyelidikan ini dijalankan adalah menumpu kepada penentuan sifat-sifat fizikal dan kimia kalyx rozel, mendapatkan satu kaedah pemprosesan yang berupaya mengoptimalkan kestabilan warnanya dan mengkaji pengaruh rawatan-rawatan berlaku terhadap jus rozel semasa penyimpanan. Sifat-sifat fizikal dan kimia rozel seperti saiz, berat, pH, keasidan titratan, pepejal terlarut, kandungan anthosianin, ketumpatan warna, warna polimer, peratusan warna polimer dan index kecoklatan telah dikaji. Succinik asid dan glukosa adalah asid organik and gula yang utama terdapat dalam rozel dengan kandungan 0.51 g/100g and 1.29 g/100g. Kajian juga dijalankan untuk memastikan dan menentukan jumlah vitamin antioksidan semulajadi yang mempunyai sifat biologikal yang paling efektif seperti ascorbik asid, β -carotin dan licopen dengan menggunakan Kromatografi Cecair Berpretasi Tinggi (HPLC). Kandungan ascorbik asid, β -carotin dan licopen adalah 141.09 mg/100g, 1.88 mg/100g dan 164.34 μ g/100g. Delphinidin-3-sambubioside dan

cyanidin-3-sambubioside merupakan dua anthosianin utama yang dikesan terkandung dalam rozel dengan menggunakan Kromatografi Lapisan Nipis (TLC) dan HPLC.

Keadaan optima dalam penghasilan jus rozel bagi kaedah Pengekstrakan Air Panas (HWE) telah ditentukan dengan menggunakan Methodologi “Response Surface” (RSM). Kombinasi masa-suhu dalam julat 30-300 minit dan 30-90 °C merupakan variasi tak berubah. Kesannya ke atas kandungan anthosianin, ketumpatan warna dan asid ascorbik telah ditentukan. Keputusan menunjukkan keadaan optima bagi pengekstrakan jus dengan menggunakan HWE adalah 3.5 jam pada suhu 60 °C. Kemudian, jus dihasilkan daripada kaedah HWE telah dibandingkan dengan jus daripada kaedah Pengisaran Air Panas (HWB), kaedah Pengisaran Air Sejuk (CWB) dan kaedah Tekanan Skru (SP). Sifat-sifat fizikal-kimia dan penilaian deria jus rozel yang diperolehi telah ditentukan. Keputusan menunjukkan bahawa kaedah HWE merupakan cara pengekstrakan yang paling efektif dengan kandungan anthosianin dan ascorbik asid yang paling tinggi. Penilaian deria turut menunjukkan jus daripada kaedah HWE adalah lebih berkualiti dari segi rasa, warna dan bau. Manakala jus daripada kaedah SP didapati paling rendah dari segi kualiti dan penilaian deria berbanding lain.

Empat rawatan berlainan dijalankan ke atas jus rozel iaitu kawalan, asid ascorbik, cisten hidroklorida dan natrium metabisulfit. Perubahan sampel-sampel jus rozel yang disimpan pada 4 ± 2 °C, 27 ± 2 °C dan 40 ± 2 °C diperhatikan. Kehilangan anthosianin dan asid ascorbik adalah sedikit pada suhu 4 °C berbanding dengan 27 °C dan 40 °C dalam penyimpanan selama 12 minggu. Suhu yang tinggi telah menyebabkan anthosianin dimusnahkan lebih cepat. Penambahan natrium metabisulfit

dalam jus rozel dapat melambatkan kehilangan anthosianin lalu mengurangkan kadar pembentuk warna polimer. Penambahan cisten hidroklorida hanya mendatangkan sedikit kesan ke atas kestabilan warna manakala penambahan asid askorbik meningkatkan degradasi kandungan anthosianin. Penambahan cisten hidroklorida dalam jus rozel mencegah kehilangan asid ascorbik semasa penyimpanan.

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I certify that an Examination Committee met on 21st November 2002 to conduct the final examination of Wong Peng Kong on his Master of Science thesis entitled "Physico-Chemical Characteristics of Roselle (*Hibiscus Sabdariffa* L.) Calyx and Effect of Processing and Storage on the Stability of Anthocyanins in Roselle Juice" in accordance with Universiti Pertanian Malaysia (Higher Degree) Act 1980 and Universiti Pertanian Malaysia (Higher Degree) Regulations 1981. The committee recommends that the candidate be awarded the relevant degree. Members of the Examination Committee are as follow:

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DECLARATION

I hereby declare that the thesis is based on my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously or concurrently submitted for any other degree at UPM or other institutions.



Wong Peng Kong

Date: 2/12/02

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