UNIVERSITI PUTRA MALAYSIA

EFFECTS OF MANUAL, CHEMICAL AND ENZYMATIC PEELING METHODS ON PHYSICOCHEMICAL AND MICROBIOLOGICAL PROPERTIES OF MALAYSIAN MANGO (Mangifera indica L. CV. 'CHOK ANAN') PUREE

NUR SADRINA BINTI MOHAMAD

FSTM 2016 11

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By

NUR SADRINA BINTI MOHAMAD

Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia, in Fulfillment of the Requirements for the Degree of Master of Science

May 2016

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirement for the Degree of Master of Science

EFFECTS OF MANUAL, CHEMICAL AND ENZYMATIC PEELING METHODS ON PHYSICOCHEMICAL AND MICROBIOLOGICAL PROPERTIES OF MALAYSIAN MANGO (*Mangifera indica* L. CV. 'CHOK ANAN') PUREE

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NUR SADRINA BINTI MOHAMAD

May 2016

Chairman : Norhayati Hussain, PhD
Faculty : Food Science and Technology

Mango is an intermediate product applied to several products as juices, nectars and purees but it requires peeling before further processing. Conventional peeling has been implemented, but it is time-consuming and requires laborious work. Therefore, this study aimed to determine the optimum conditions of selected peeling methods (chemical and enzymatic) of Malaysian 'Chok Anan' mango fruit on physicochemical properties, polyphenol oxidase (PPO), respiration rate and microbiological analysis of mango puree. Optimization of peeling conditions was carried out using the Response Surface Methodology (RSM) to study the effect of chemical (sodium hydroxide, NaOH) and enzyme (Pectinex Ultra SP-L) based on concentrations (chemical: 1.6-7.3 g/100mL; enzyme: 0.005-0.095% (v/v)), temperatures (chemical: 80-95 ℃ ; enzyme: 25-40 ℃) and soaking durations (chemical: 5-10 min; enzyme: 30-120 min) on the mango fruit. Peeling yield and the effect of the different peeling methods on moisture content (%), color changes and viscosity (Pa.s) of mango puree were investigated. The optimum concentration of 7.3 g/100mL; temperature at 95℃ and duration of 8.5 min for soaking were selected for chemical peeling. The optimized concentration and temperature of NaOH applied in this study were the most significant factors ($p<0.05$) affecting the peeling yield (88.72%), moisture content (86.19%), color (ΔdE = 62.00) and viscosity (0.2169 Pa.s) of mango puree. The optimum conditions for enzymatic peeling (pectinase solution) were at concentration of 0.009%; temperature, 25℃, and duration of soaking for 120 min. The pectinase concentration, temperature and duration of soaking had significantly ($p<0.05$) affected the quality of the Malaysian 'Chok Anan' mango puree with enhanced yellow color (ΔdE = 67.02), low viscosity (0.1739 Pa.s) and low moisture content (82.67%). Enzymatic peeling significantly ($p<0.05$) reduced the peeling time (4.46 min) of mango puree production compared to both manual (5.30 min) and chemical (6.49 min) peeling. In addition, absorption of chemical (0.84 g/100g) and enzyme (2.50 g/100g) solutions, penetration depth of

NaOH (0.45 mm), enzyme activity (0.48-0.63 g/100mL) were analysed for the peeling efficiency of chemical and enzymatic peeling methods. Based on the results, enzymatically peeled mango puree had significantly ($p<0.05$) the lowest moisture content (84.04-84.44%), lowest pH (4.54 – 4.67), acidic (TA = 0.08-0.10%), yellower in color (ΔdE = 61.72), highest total soluble solids (TSS) content (16.0-16.9 ^{o}Brix), highest vitamin C content (1.80 mg/100g) compared to manual-peeled (moisture content = 84.73-86.39%, pH = 4.51-4.88, TA = 0.09-0.14%, ΔdE = 61.00, TSS = 15.0-16.0 ^{o}Brix, vitamin C = 1.42 mg/100g) and chemical-peeled (moisture content = 84.43-88.23%, pH = 5.20–5.36, TA = 0.06-0.07%, ΔdE = 59.80, TSS = 15.0-15.7 ^{o}Brix, vitamin C = 1.42 mg/100g) mango puree. There was significantly ($p<0.05$) decreased (manual: 20.4-20.2 kPa, chemical: 20.3-20.0 kPa, enzymatic: 20.7-19.5 kPa) in oxygen composition and increased of carbon dioxide (manual: 0.64-0.80 kPa, chemical: 0.46-3.03 kPa, enzymatic: 0.64-2.60 kPa) reflected a high respiration rate occurred over storage period. Furthermore, enzymatic-peeled mango puree stored at 4±2℃ had significantly ($p<0.05$) the lowest total plate counts (6.8 log CFU/g) compared to the manual-peeled (7.8 log CFU/g) mango purees at the end of 26 days of storage. Yeast and mold counts of enzymatically peeled mango puree shows significantly ($p<0.05$) the lowest count (6.5 log CFU/g) compared to manual-peeled (7.8 log CFU/g) and chemical-peeled (8.3 log CFU/g) after 26 days of storage. Storage of mango puree at 4±2℃ shows significantly longer shelf life (26 days) compared to 20±2 ℃ (ambient temperature) for only 3 days. This study suggested that enzymatic peeling can be an alternative peeling method other than manual peeling and effective with the recyclability of the pectinase used throughout the mango processing. The development of effective peeling method may be useful to assist the puree production of mango industries.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk Ijazah Master Sains

PERBEZAAN KESAN PROSES PENGUPASAN SECARA MANUAL, KIMIA DAN ENZIM KE ATAS SIFAT- SIFAT FIZIKOKIMIA DAN MIKROBIOLOGI PURI BUAH MANGGA (*Mangifera indica* L. CV. ' CHOK ANAN') MALAYSIA

Oleh

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Mangga merupakan produk perantaraan yang digunakan dalam beberapa produk seperti jus, nektar dan puri tetapi ia memerlukan pengupasan sebelum proses selanjutnya dilakukan. Pengupasan secara manual telah lama dipraktikkan, tetapi ia memakan masa dan memerlukan lebih tenaga pekerja. Oleh itu, kajian ini bertujuan untuk menentukan proses pengupasan yang optimum (kaedah kimia dan enzim) ke atas buah mangga 'Chok Anan' Malaysia ke atas sifat-sifat fizikokimia, polifenol oksida (PPO), kadar respirasi dan analisis mikrobiologi puri mangga. Pengoptimuman proses pengupasan telah dijalankan menggunakan 'Response Surface Methodology' (RSM) untuk mengkaji kesan bahan kimia (natrium hidroksida, NaOH) dan enzim (Pectinex Ultra SP-L) ke atas kepekatan (1.6-7.3 g/100ml; 0.005-0.095%), suhu (80-95 ℃; 25-40 ℃) dan tempoh merendam (5-10 min; 30-120 min) buah mangga. Kesan proses pengupasan yang berbeza pada hasil (%), kandungan kelembapan (%), perubahan warna dan kelikatan (Pa.s) mangga puri telah dikaji. Kepekatan optimum 7.3 g/100ml; pada suhu 95℃ dan tempoh rendaman selama 8.5 min telah dipilih untuk mengupas kulit mangga menggunakan bahan kimia (NaOH). Kepekatan dan suhu NaOH yang digunakan dalam kajian ini merupakan faktor yang paling memberikan kesan signifikan ($p<0.05$) ke atas hasil (88.72%), kandungan kelembapan (86.19%), warna ($\Delta dE = 62.00$) dan kelikatan (0.2169 Pa.s) puri mangga. Walau bagaimanapun, pengoptimuman bagi proses pengupasan menggunakan enzim pektinas adalah dengan menggunakan kepekatan 0.009%; suhu, 25℃, dan tempoh merendam selama 120 min. Kepekatan, suhu, dan tempoh rendaman pektinas memberikan kesan signifikan ($p<0.05$) ke atas kualiti puri mangga 'Chok Anan' Malaysia dengan warna yang lebih kuning ($\Delta dE = 67.02$), kelikatan rendah (0.1739 Pa.s) dan mengandungi kelembapan yang rendah (82.67%). Proses pengupasan menggunakan enzim juga memberikan kesan signifikan ($p<0.05$) dengan mengurangkan masa pemprosesan puri mangga selama 4.46 min berbanding secara manual (5.30 min) dan penggunaan bahan kimia (6.49 min). Di samping itu, penyerapan larutan kimia (0.84 g/100g) dan enzim (2.50 g/100g), penetrasi larutan

NaOH (0.45 mm) dan aktiviti enzim (0.48-0.63 g/100ml) turut dikaji untuk menilai kecekapan proses pengupasan menggunakan bahan kimia dan enzim. Berdasarkan kajian, terdapat perbezaan yang signifikan ($p<0.05$) pada puri mangga yang dikupas menggunakan enzim di mana ia mempunyai kandungan kelembapan (84.04-84.44%) dan pH (4.54-4.67) yang rendah, berasid (TA = 0.08-0.10%), lebih kekuningan (ΔdE = 61.72), tinggi kandungan pepejal terlarut (TSS = 16.0-16.9 ^{o}Brix) dan kandungan vitamin C (1.80 mg/100g) yang tinggi berbanding puri mangga yang diproses secara manual (kandungan kelembapan = 84.73-86.39%, pH = 4.51-488, TA = 0.09-0.14%, ΔdE = 61.00, TSS = 15.0-16.0 ^{o}Brix, vitamin C = 1.42 mg/100g) dan puri mangga yang diproses menggunakan bahan kimia (kandungan kelembapan = 84.43-88.23%, pH = 5.20-5.36, TA = 0.06-0.07%, ΔdE = 59.80, TSS = 15.0-15.7 ^{o}Brix, vitamin C = 1.42 mg/100g). Penurunan kandungan oksigen (manual: 20.4-20.2 kPa, bahan kimia: 20.3-20.0 kPa, enzim: 20.7-19.5 kPa) dan kenaikan kandungan karbon dioksida (manual: 0.64-0.80 kPa, bahan kimia: 0.46-3.03 kPa, enzim: 0.64-2.60 kPa) yang signifikan ($p<0.05$) dapat dilihat dengan perubahan kadar respirasi yang telah berlaku sepanjang tempoh penyimpanan. Tambahan pula, puri mangga yang dikupas menggunakan enzim yang disimpan pada 4±2℃ mempunyai jumlah kiraan plat yang signifikan ($p<0.05$) rendah (6.8 log CFU/g) jika dibandingkan dengan puri mangga yang dikupas secara manual (7.8 log CFU/g) pada akhir penyimpanan selama 26 hari. Jumlah yis dan kulat yang signifikan ($p<0.05$) untuk puri mangga yang dikupas menggunakan enzim dapat dilihat dengan jumlah kiraan yang paling rendah (6.5 log CFU/g) berbanding puri mangga yang dikupas secara manual (7.8 log CFU/g) dan penggunaan bahan kimia (8.3 log CFU/g) selepas penyimpanan selama 26 hari. Terdapat perbezaan yang signifikan ($p<0.05$) untuk penyimpanan puri mangga pada suhu 4±2℃ dengan jangka hayat selama 26 hari berbanding pada suhu bilik 20±2℃ yang hanya bertahan selama 3 hari. Kajian ini mencadangkan bahawa pengupasan menggunakan enzim sesuai dijadikan salah satu kaedah alternatif selain kaedah manual, murah dan berkesan melalui penggunaan semula larutan pektinas sewaktu pemprosesan mangga. Penambahbaikan proses pengupasan yang lebih berkesan mungkin berguna dan lebih membantu pihak industri dalam penghasilan puri buah mangga tempatan.

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I certify that a Thesis Examination Committee has met on 20 May 2016 to conduct the final examination of Nur Sadrina binti Mohamad on her thesis entitled "Effects of Manual, Chemical and Enzymatic Peeling Methods on Physicochemical and Microbiological Properties of Malaysian Mango (*Mangifera indica* L. Cv. 'Chok Anan') Puree" in accordance with the Universities and University Colleges Act 1971 and the Constitution of the Universiti Putra Malaysia [P.U.(A) 106] 15 March 1998. The Committee recommends that the student be awarded the Master of Science.

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CHAPTER 1

INTRODUCTION

Mango (*Mangifera indica L.*) is one of the important tropical fruits produced and marketed globally especially in Asia (Berardini et al., 2005). Many countries in Southeast Asia such as Philippines, Indonesia, Thailand, Burma, and Malaysia are involved in the cultivation of mango (Nor Hazlina et al., 2008). Mangoes are nutritionally containing a good source of vitamins A, C and fiber (Tharanathan et al., 2006). It is famous because of its attractive color, exotic flavor and fragrance, delicious taste, nutritious and provides about 64-86 calories energy to human's diet (Pott et al., 2003; Tharanathan et al., 2006; Rathore et al., 2007).

About 90% of tropical fruits are consumed in the producing countries themselves, while 10% are traded internationally. Fresh mangoes dominate the export market and it is also processed into several products such as puree, concentrate, juice, nectar, mango blends and dried slices (Madamba et al., 2002). There are a lot of fruits purees available worldwide such as strawberry, orange, apple, kiwi, pineapple, and mango. Nevertheless, a large number of mango products produce commercially are based on ripe mango fruits which have high sugar content (15 -20%) and low acid content (0.2- 0.5%) (Tharanathan et al., 2006).

Mangoes for export are often processed into puree to extend shelf life and facilitate transportation. Fresh mangoes are subjected to chilling injury during storage, and increasing storage temperature leads to rapid decay in fruit quality (Mohammed et al., 2002; Nair et al., 2009). In addition, mango fruits have short production season and storage life limited is to 2–3 weeks at 10°C−15°C. Thus, in extending the shelf life of the mangoes, peeling is indeed a necessary and important processing step. Peeling process is required in removing the unwanted skin before undergo further processes into other final products such as puree. Conventionally, peeling process was done manually using peeler or machine (mechanical peeling), steam peeling and freezer peeling (Toker et al., 2003; Pretel et al., 2008; Pagán et al., 2010). Unfortunately, it is time-consuming and requires laborious work (Pretel et al., 2008; Pagan et al., 2010).

Despite providing different advantages, most of the conventional methods of peeling often cause high peeling losses and damage the flesh, thus affecting the quality of the fruit (Toker et al., 2003). As discussed by Sivakumar et al. (2010), the quality of mangoes depends largely on the external quality (bruises, latex or sap injury, decay, uniform weight, color and shape) and internal quality parameters such as consistent and intense flesh color, free from damage, adequate acidity and total soluble solid (Brix°) that depends on cultivar and type of consumer preferences. In addition, quality and authenticity are of particular importance with respect to consumer expectation (Fügel et al., 2005). Fruit purees and fruit preparations command premium prices and, therefore, represent favored targets for adulterations such as by

blending high- priced fruits with cheaper fruits. In addition to the admixture of adulterants, the specified fruit contents may not be met. For this reason, numerous attempts at finding suitable methods for authenticity control and determination of the fruit content in fruit based products have been made. The major analytical problem is due to the complexity of the products and to the substantial variance of the fruit specific components (Fügel et al., 2005).

The greatest hurdle to the commercial marketing of mango is the limited shelf life, which is due to excessive tissue softening and browning (Soliva-Fortuny et al., 2003). Polyphenol oxidase (PPO) was demonstrated to be the color-related enzymes and played a key role in color degradation due to enzymatic browning (MacDonald et al., 2000). Discoloration represents a major restraint because color is considered as the main quality determining an attribute of the fruit itself. Apart from extrinsic factors such as heat, light and oxygen, polyphenol oxidases (PPO) have been reported to considerably contribute to this phenomenon (color) (Lopez-Serrano et al., 2002; Chisari et al., 2007). Qualitative attributes generally change with time, as part of the normal metabolism of the product (Tijskens et al., 1996). In addition, storage temperature is also a part of extrinsic factors mention above. According to Majidi et al., 2011, low temperature is the most important factor in maintaining quality and extending the shelf-life of fruits and vegetables after harvest.

Various methods have been utilized for peeling, including the use of chemical and enzymatic treatment methods (Fellows, 2000; Toker et al., 2003; Das et al., 2006). Practically, each peeling treatments have their own benefits and limitations. Chemical peeling is applied by immersing (tomato fruit) in the hot sodium hydroxide or caustic soda for a certain time (Das et al., 2006). According to Lavelli et al. (2009), chemical peeling leads to a decrease in the antioxidant content of peach-based products, and it affects the color and stability during storage (23℃ and 37℃).

As an alternative, enzymatic peeling using pectinase, cellulase or hemicellulase has been suggested (Rouhana et al., 1994; Pretel et al., 1997, 1998, 2005, 2007; Toker et al., 2003; Pagán et al., 2005; Srikaeo et al., 2011). The principle of enzymatic peeling is based on the digestion, through an enzyme preparation, of pectic substances existing in the cell wall of the plant (Bruemmer et al., 1978; Berry et al., 1988; Pretel et al., 2008). Pectin, cellulose and hemicellulose are the polysaccharides responsible for the adherence of the peel to the fruit (Whitaker et al., 1984; Toker et al., 2003). Therefore, treating the fruit with the corresponding glycohydrolases provides ease of peeling the fruit (Pretel, et al., 1997; Pagan et al., 2010). Enzymatic peeling has been studied in citrus fruits, some stone fruits and vegetables (Prakash et al., 2001; Pretel et al., 2005; Toker et al, 2003; Kaur et al., 2009; Suutarineen et al., 2003). Application of the enzymatic peeling for other local fruits has not been widely introduced and studied with the exception of citrus fruit such as mandarin (Pretel et al., 1998; Liu et al., 2000; Liu et al., 2004), pomelo (Soffer et al., 1994; Aziz et al., 1999) and limau kasturi (Hazniza et al., 2001).

Hence, the objectives of this study were (i) to optimize the peeling methods (chemical and enzymatic) for mango, (ii) to determine the mango peeling efficiency and (iii) to determine the effects of different storage conditions on physicochemical and microbial properties of mango puree. High quality local mango puree may expand the market availability and application of local seasonal mango. The optimized peeling method helped to prolong the shelf life of mango puree as natural as possible and further distributed in larger quantity for industrial usage.

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