UNIVERSITI PUTRA MALAYSIA

EFFECTS OF FERMENTATION AND DE-BITTERING METHODS ON PRODUCTION OF ALMOND-LIKE RAMBUTAN SEED (Nephelium lappaceum L.) SNACK

SHABNAM MEHDIZADEH

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By

SHABNAM MEHDIZADEH

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

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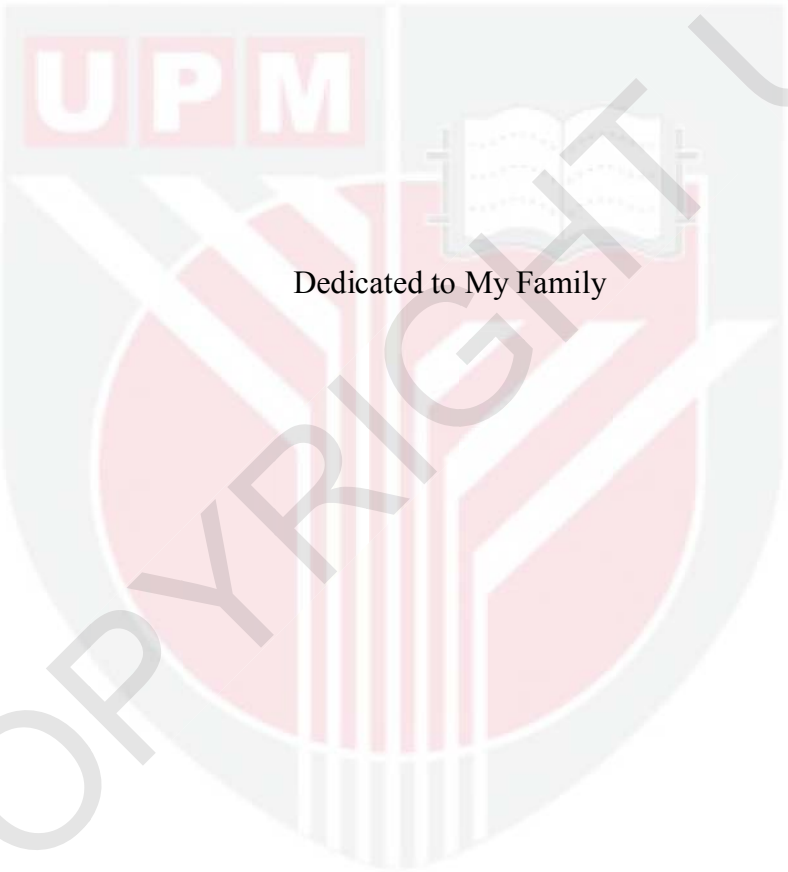
Dedicated to My Family



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

EFFECTS OF FERMENTATION AND DE-BITTERING METHODS ON PRODUCTION OF ALMOND-LIKE RAMBUTAN SEED (*Nephelium lappaceum* L.) SNACK

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December 2014

Chairman: Associate Professor Lasekan Olusegun Olaniyi, PhD

Faculty: Food Science Technology

There are so many current studies going on in the field of waste products. One of the instances is seeds of rambutan, which is discarded yearly because of its slightly bitter taste despite the fact that it contains valuable source of oleic acid, arachidic acid and vitamins. The first part of this study determined the effect of fermentation on variability of fermentation index, polyphenol constituents and amino acids of rambutan seeds (anak sekolah). Results of fermentation showed that time of fermentation had a significant decrease in pH, whereas a significant increase was evident in the fermentation index ($p<0.05$). Findings also showed that microbial activities in the pulp produce alcohols to some extent and acids such as acetic acid and lactic acid, and raises the temperature and humidity and these changes inside the seed resulted in the death of the embryo. Moreover, it was found that fermentation significantly reduced the levels of total polyphenols (59%), tannin (60%) and saponins (33%). However, fermentation appeared to have a moderate effect on geraniin, corilagin and much stronger effect on ellagic and gallic acids. During fermentation, variability in gallic acid, geraniin, corilagin and ellagic acid did not show consistent trend. In contrast, amino acids that elicited bitter taste (phenylalanine, tyrosine and leucine) showed reduction of 20%, 30% and 40% respectively after 10 days of fermentation. Furthermore, it was discovered that sucrose, which was the only sugar present in significant amount, in unfermented seed was remarkably reduced during fermentation. The second part of this study concentrated on the effect of different debittering methods on the seed of rambutan. There were 14 different methods for reducing bitterness with different times and concentrations and in different soaking medium such as soaking in water and debittering through impregnation processes by sugar and salt as well as lye treatment. Three methods were selected based on the substantial reduction of bitterness compounds. These three methods were soaking in water for 7 days, treatment of seeds with 22% sodium chloride for 8 h and treatment of seeds with 1 N NaOH for 8 h, accordingly. The three most efficient methods indicated that tannin content declined by 55.4%, 48.2% and 40.9%. Similarly, polyphenol contentswere reduced to 92.7%, 84.4% and 85.1%. Saponin, gallic acid and corilagin, which are known as

the prime causes of bitterness, were completely removed by these different treatments. However, geraniin was reduced by 20% during debittering methods. Tyrosine, leucine and phenylalanine were the three amino acids, which contain bitter taste in rambutan seeds were also reduced by the different debittering methods. The third part of this study focused on the effect of roasting on the texture, color and flavor of debittered seeds of rambutan and almond. Debittered rambutan seeds and almonds were roasted at 125 °C for 15 minutes, the volatile extract of rambutan seeds produced 13 odors and roasted almond produced 122 odors-active compounds by GC-MS. Findings showed that the highest peaks were associated to acids and ester groups. The results also showed that roasted debittered by 22% NaCl for 8 h and 7 days of soaking treatment were more similar to roasted almond than the other two treatments. In general, therefore, not only do the current findings add to a growing body of literature on the different debittering methods of rambutan seed, but also some efficient approaches were identified for debittering rambutan seed in which this may contribute significantly to produce a new almond-like snack.

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

KAJIAN KESAN PENAPAIAN DAN KAEDAH NYAH PAHIT TERHADAP PENGHASILAN SNEK BIJI RAMBUTAN (*Nephelium lappaceum* L.) KE SERUPA ALMOND

Oleh

SHABNAM MEHDIZADEH

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Pengerusi: Professor. Madya Lasekan Olusegun Olaniyi, PhD

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Pada masa ini, beberapa kajian telah dijalankan ke atas bahan buangan. Salah satu contoh adalah biji rambutan yang dibuang setiap tahun kerana ia rasanya pahit walaupun ia mengandungi sumber yang berharga seperti asid oleik asid, arakidik dan vitamin. Bahagian pertama kajian ini telah menentukan kesan penapaian pada kebolehubahan indeks penapaian, juzuk polifenol dan asid amino rambutan. Keputusan penapaian menunjukkan bahawa masa penapaian mempunyai penurunan ketara dalam pH, manakala peningkatan yang ketara jelas didapati pada indeks penapaian ($p<0.05$). Dapatan kajian juga menunjukkan bahawa aktiviti mikrob pada pulpa menghasilkan sedikit sebanyak alkohol dan asid seperti asid asetik dan asid laktik, dan meningkatkan suhu dan kelembapan serta perubahan beginimengakibatkan kematian embrio pada benih. Selain itu, didapati bahawa penapaian mengurangkan aras jumlah polifenol (59%), tanin (60%) dan saponin (33%). Walau bagaimanapun, penapaian mempunyai kesan sederhana pada geraniin, korilagin dan kesan lebih kukuh pada elagik dan asid galik. Semasa penapaian, kepelbagaian asid galik, geraniin, korilagin dan asid elagik tidak menunjukkan tren yang konsisten. Sebaliknya, asid amino yang memberi rasa pahit (fenylalanina, tirosina dan leusina) menunjukkan pengurangan masing –masing 20%; 30% dan 40% selepas 10 hari penapaian. Tambahan pula, didapati bahawa sukrosa, yang merupakan satu-satunya sumber gula yang hadir dengan jumlah yang besar, di dalam biji yang tidak beralkohol telah berkurangan dengan ketara akibat penapaian. Bahagian kedua kajian ini tertumpu kepada kesan kaedah penyahpahitan berbeza pada rambutan. Terdapat 14 kaedah yang berbeza untuk mengurangkan kepahitan dalam pada masa yang berbeza dan melalui konsentrasi yang berbeza biji seperti rendaman di dalam air selama 3 hari dan 7 hari dan penyahpahitan melalui proses gula dan garam serta rawatan ‘lye’. Tiga kaedah telah dipilih berdasarkan pengurangan sebatian kepahitan yang berpotensi. Tiga kaedah ini adalah dengan merendamnya di dalam air selama 7 hari, rawatan biji rambutan dengan 22% natrium klorida selama 8 jam dan rawatan biji dengan 1 N NaOH selama 8 jam. Dengan menggunakan tiga kaedah yang berkesan, hasil telah menunjukkan bahawa kandungan tanin masing-masing berkurangan menjadi 55.4%, 48.2% dan 40.9%.

Selain itu, kandungan polifenol masing-masing dikurangkan sebanyak 92.7%, 84.4% dan 85.1%. Saponin, asid galik dan korilagin yang diketahui merupakan punca utama kepahitan telah dibuang sepenuhnya oleh rawatan-rawatan yang berbeza. Walau bagaimanapun geraniin dikurangkan sebanyak 20% dengan kaedah penyahpahitan. Tirosina, leusina dan fenilalanina merupakan tiga asid amino, yang mengandungi rasa pahit dalam biji rambutan, juga dikurangkan dengan kaedah penyahpahitan berbeza dengan nilai-nilai yang berbeza. Bahagian ketiga kajian ini memberi tumpuan kepada kesan memanggang terhadap tekstur, warna dan rasa biji rambutan dan badam yang dinyahpahit. Biji rambutan yang dinyahpahit dan badam telah dipanggang pada 125°C selama 15 minit, ekstrak volatile biji rambutan menghasilkan 13 bau dan badam panggang menghasilkan 122 sebatian bau-aktif oleh GC-MS. Dapatan kajian menunjukkan bahawa puncak tertinggi adalah berkaitan dengan asid dan kumpulan ester. Keputusan juga menunjukkan bahawa hasil panggang yang dinyahpahit oleh NaCl 22% selama 8 jam dan 7 hari rawatan rendaman lebih serupa dengan badam panggang berbanding dengan dua rawatan yang lain. Secara umum, oleh kerana itu, kajian ini bukan sahaja menghasilkan penemuan yang menambahkan bahan literatur kepada badan literatur yang semakin berkembang tentang kaedah penyahpahitan yang berbeza pada biji rambutan, tetapi juga kepada beberapa pendekatan yang cekap yang telah dikenalpasti untuk penyahpahitan biji rambutan di mana ini boleh menyumbang dengan ketara untuk menghasilkan makanan ringan ala badam yang baharu.

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This thesis was submitted to the Senate of Universiti Putra Malaysia and has been accepted as fulfillment of the requirement for the degree of Master of Science. The members of the Supervisory Committee are as follows:

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LIST OF ABBREVIATIONS

ANOVA	Analysis of variance
C	Corilagin
°C	Celsius
EA	Ellagic acid
FI	Fermentation Index
G	Geraniin
GA	Gallic Acid
GC-MS	Gas Chromatography- Mass Spectrometry
g	Gram
hr	Hour
h°	hue angels
HCl	Hydrochloric acid
HPLC	High Performance Liquid Chromatography
KCl	Potasium chloride
Kg	Kilogram
mg	Milligram
µg	Microgram
µl	Microliter
Min	Minute
N	Normality
NaCl	Sodium Chloride
NaoH	Sodium Hydroxide
Na_2CO_3	Sodium Carbonate
R^2	Coefficient of determination
SG	Solid gain

TPC	Total Phenolic Content
tR	Retention time
WL	Water loss
WR	Weight reduction
w/w	Weight per weight
w/v	Weight/volume
3D	Three-dimensional
%	Percentage

CHAPTER 1

INTRODUCTION

Rambutan (*Nephelium lappaceum* L.) is one of the famous fruit that typically can be found in Southeast Asia belongs to the family of Sapindaceae (Marisa, 2006). Rambutan comes from the Malay word 'rambut' meaning hair. In Malaysia the rambutan bear its fruit two times in a year, the first, being in June and a second one in December. The rambutan is well adapted to humid climate and tropical area. It is widely cultivated in Malaysia with an annual production rate of (80, 000 tons/year), Indonesia (148, 000 tons/year), Thailand (430, 000 tons/year), and the Philippines (20, 000 tons/year).A warm climate is ideal for its growth and high yield. Rambutan tree is about 10-20 feet (3.05-6.1 m) high. The color of the skin can be red, yellow or green and the shape is round to oval, size is about 2" to 3" inch. The surface of fruit is made of spiny peel while inside of the fruit is a white and juicy acid pulp that is rich in vitamin C (Dadshani, 2002). The pulp is sweet, succulent and adheres to the seed (Nakasone; Paull, 1998; Wall, 2006). Among many appealing and desirable attributes of fruits from the tropics and subtropics is their characteristic flavor, which is the most remarkable to consumers. While rambutan fruit is still little known in many parts of the world, it is an important commercial crop in Asia (Morton 1987; Ong, Acree & Lavin, 1998).

The edible portion of rambutan (*Nephelium lappaceumlin* L.) is sweet and juicy; the light brown oval seed is generally remains as a waste material annually in large amount in Southeast Asia because it is slightly bitter (Kamel & Kakuda, 1992). The bitterness of the seeds of rambutan has been linked to traces of alkaloid, tannin, saponin and phenolic content like ellagic acid corillagin, geraniin and gallic acid (Kondo, Tsuda, Muto, Ueda, 2002; Singh, Murthy, Jayaprakasha, 2002). Another study has also shown that some Amino acids (AA) could have contributed to the bitter taste (Kano & Goto, 2003). As far as our literature survey could establish, there are little or no studies on the identification of phenolic compounds in the seed of *Nephelium.* Although a few polyphenolic components of longan seeds (same family with rambutan) have been characterized (Zheng et al., 2009). Although roasted rambutan seed are considered edible in some Asian countries, for more safety the seed must be evaluated before used (Gray, 2011). Furthermore, seeds are highly rich in vitamins such as vitamin C. They are also a valuable source of zinc, iron, Mg and Mn. Some experiments have revealed that seed of rambutan contains almost great quantity of fat with values between 14 g/100 g and 41 g/100 g (Solís- Fuentes, Camey-Ortíz, Hernández-Medel, Pérez-Mendoza & Durán-de-Bazúa, 2010) that is highly valued for manufacturing of food and production of soap and candle. The seed oil provides enough minerals necessary for human requirements (Standard from Institute of Medicine 2001). Main fatty acids in seed of rambutan (*Nephelium lappaceum* L.*)* are oleic acid (40.3%) and arachidic acid (34.5%). The processing of rambutan seeds for the reduction of bitterness (Supplied by MARDI) involves two major stages namely fermentation and application of debittering methods on ramburan seeds. Fermentation is used as the first stage in the bitterness reduction.

Fermentation, cause several chemical reactions and a deep adjustment of the biochemical feature of seeds (Timbie, Sechrist & Keeney, 1978; Gill, Macleod & Moreau, 1984). Certainly, sugars in the mucilage are changed into alcohol by micro-organism, during fermentation, heat is generated and many organic substances are produced such as ethanol, lactic, acetic acids and other organic acids leading to the death of the seed embryo (Kostinek, Ban-Koffi, Ottah-Atikpo, Teniola, Schillinger, Holzapfel & Franz, 2008). The mass then becomes runny and drops off from the beans. Enzymatic reactions occur, leading to the creation of flavor and changes in the color of cotyledons. These biochemical variations inside the beans cause the decrease of astringency and bitterness and the production of flavor precursors (Lagunes-Gálvez et al., 2007). Fermentation is one of the most widely used methods for food preparation before humans recognized the benefits. Firstly, fermentation was utilized to preserve food, modify the flavor, color as well as producing foods that are more digestible, and it could reduce undesirable and toxic constituents in foods, which is another significant aspect of the fermentation process (Jinap, Dimick & Hollander, 1995; Afoakwa, Quao, Takrama, Budu & Saalia, 2012). Fermentation is an earliest method for improving the shelf life, nutritional and organoleptic quality of foods (Doblado, Frias, Muñoz & Vidal-Valverde, 2003). Lately, fermentation as a kind of bioprocess system has been applied for the creation and removal of bioactive constituents in foodstuff, and in chemical and pharmaceutical productions (Martins, Mussatto, Martínez-Avila, Montañez-Saenz, Aguilar & Teixeira, 2011). Apart from fermentation, other debittering methods that have been used are soaking in water, blanching with sodium hydroxide and impregnation processes by sugar and salt. Roasting is an important stage in coffee bean and nuts processing (Ozdemir & Devres, 2000; Pittia, Rosa & Lerici, 2001; Saklar, Katnas & Ungan, 2001). Roasting is performed in order to promote flavor, color and desirable textural changes that eventually promote the complete accessibility of nuts. It is becoming increasingly difficult to ignore the enormous capacities of agricultural-food biomass waste discarded yearly all around the world. Moreover, disposal of agricultural-food waste can have a serious ecological impact on the environment. In recent years, there have been increased interests in numerous investigations on waste reuse (Singh, Chidambara Murthy & Jayaprakasha, 2002). Nowadays, large amounts of fruit most especially rambutan seeds (at least ~8000 metric tons of seed in Malaysia and ~ 60000 tons in other countries) are discarded annually at processing plants. Kamel and Kakuda, (1992) reported that to be economically viable, however, both oil and meal from these fruit seeds must be used.

Almond (*Prunus dulcis*) is one of the most reputable nuts with a high nutritional value, which have been used as a snack all over the world. In this study almond was used as control for roasting since the texture, volatile compounds and some nutritional values are similar to rambutan seed.

The main objectives of this study are:

1) To examine the effect of fermentation on fermentation index, poly phenol and amino acids profile of rambutan
2) To evaluate the effect of different debittering methods on the reduction of bitterness in rambutan seeds and
3) To determine the effect of roasting conditions on the aroma profile of debitterd seeds.

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LIST OF PUBLICATIONS

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