



UNIVERSITI PUTRA MALAYSIA

**AEROBIC MESOPHILIC BACTERIA AND SENSORY QUALITIES OF
YELLOW ALKALINE NOODLES DURING STORAGE**

SITI SUWAIBAH ABD GHAFFAR

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By

SITI SUWAIBAH ABD GHAFFAR

**Thesis Submitted to the School of Graduate Studies, Universiti Putra Malaysia,
in Fulfillment of the Requirement for the Degree of Masters of Science**

June 2008



DEDICATION

To.....

My husband, Acheh, and my beloved family.

The people I LOVE the most.....



ABSTRACT

Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirements for the degree of Master of Science

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June 2008

Chairman: Associate Professor Fatimah Abu Bakar, PhD

Faculty : Food Science and Technology

A study was conducted to evaluate the aerobic mesophilic bacteria and sensory characteristics of yellow alkaline noodles (YAN) during storage. Samples were obtained from 9 commercial noodles processing plants, 3 from each category which were situated in Selangor and Kuala Lumpur. The categories of processing plants were determined according to the processing methods employed namely; automated processing method (AP samples); semi automated processing method (SM samples) and manual processing method (MP samples). The samples were collected on the day of production, placed in a polystyrene box and transported to the laboratory where they were stored at ambient temperature (28 ± 2 °C). Aerobic mesophilic counts (APC), pH and sensory evaluation of noodle samples were analysed daily for 9 days. APC in all samples increased significantly ($P < 0.05$) from around $\log 3$ CFU/g to



around log 7 CFU/g during storage. The pH of all samples decreased significantly ($P < 0.05$) from initial pH around 10 to 6 at end of storage. Sensory attributes to evaluate YAN spoilage characteristics include; off odours, firmness, elasticity and sliminess. Samples began to deteriorate and became unacceptable to panelist around day 2-3 days of storage.

Composition of aerobic mesophilic bacteria were identified using the Biolog Microstation System with its equivalent software and database. A total of 90 isolates, were isolated and the pre-dominant bacteria identified include *Staphylococcus sciuri* and *Ochrobactrum anthropi* which were common in all samples and *Alcaligenes faecalis* which was only isolated from SP and MP samples.

Another study was conducted to evaluate the growth profile of pre-dominant bacteria in YAN. Samples were treated with solutions of *S. sciuri* (SS), *A. faecalis* (AF) and *O. anthropi* (OA) at level of log 5-6 CFU/g. Samples were stored at ambient temperature (28 ± 2 °C) and analysed daily for aerobic mesophilic counts (APC), pH and sensory evaluation for 4 days. APC of SS and AF samples increased significantly ($P > 0.05$) during storage and more rapidly compared to OA samples. The pH values of SS and AF samples decreased significantly ($P > 0.05$) than OA samples during storage. Panelist associated development of off odours in SS and AF samples with off odours of samples produced from commercial processing plants. Volatiles compounds produced by *S. sciuri* and *A. faecalis* were accomplished using SPME-GCMS method.

Identification in the volatiles compounds in SS and AF samples were from alcohol groups and disulfide, dimethyl.

A different study was conducted to evaluate aerobic mesophilic bacteria and sensory characteristics of YAN treated with lactic acid and nisin. Untreated samples were regarded as control and samples treated with boric acid were analysed as comparison due to its abuse usage in YAN. Noodle samples were treated by dipping in 0.5%, 1.0% or 1.5% solution of lactic acid (v/v), nisin (w/v) or boric acid (w/v) for 30 seconds. Samples were stored at ambient temperature (28 ± 2 °C). Duplicate analyses of APC, pH, and sensory evaluation were conducted on day 0, 1, 3, 5, 7, and 10 of storage. A total of 200 isolates were isolated from noodle samples during storage and further characterized using series of biochemical tests. APC of all samples increased significantly ($P < 0.05$) from log 2.42 CFU/g for control (untreated) samples and around log 3-4 CFU/g for treated sampled to around log 10 CFU/g at end of storage. pH of all samples decreased significantly ($P < 0.05$) throughout storage period. Sensory evaluation showed that samples treated with lactic acid, nisin and boric acid were rejected by panelist due to unacceptable odour and texture. Results showed that the ability of nisin, lactic acid and boric acid to reduce microbial load in noodle samples were not significant due to the fact that the inhibitory effect were limited to certain group of bacteria only. The potential of lactic acid and nisin acting singly to inhibit growth of aerobic mesophilic bacteria in YAN were not significant however the enhancement on its potential may probably be achieved by acting in combinations or with other preservatives.



ABSTRAK

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

BAKTERIA AEROBIK MESOFILIK DAN PENILAIAN DERIA MEE KUNING BERALKALI SEMASA PENSTORAN

Oleh

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Satu kajian mengenai bakteria aerobik mesofilik dan penilaian deria mee kuning beralkali (YAN) telah dijalankan. Sampel-sampel diperolehi daripada 9 kilang mee yang beroperasi secara komersil dan berlokasi di sekitar Selangor dan Kuala Lumpur. Pengilang dibahagikan mengikut kaedah pemprosesan mee iaitu sampel-sampel daripada kilang automatik (sampel AP), kilang separa automatik (sampel SP) dan kilang manual (sampel MP). Sampel diperolehi daripada 3 kilang bagi setiap kategori pada hari pengeluaran, diletakkan dalam kotak polystyrene dan disimpan pada suhu persekitaran (28 ± 2 °C). Kiraan bakteria aerobik mesofilik (APC), nilai pH dan penilaian deria dianalisa setiap hari dalam tempoh 9 hari. Kiraan awal APC untuk semua sampel adalah lebih kurang $\log 3$ CFU/g dan meningkat secara signifikan

($P < 0.05$) kepada lebih kurang $\log 7$ CFU/g pada hari terakhir penyimpanan. Ciri-ciri sampel yang dinilai dalam penilaian deria terdiri daripada bau, kekerasan, keanjalan dan penghasilan lendir. Sampel AP, SP and MP mulai tidak diterima oleh ahli panel selepas 2-3 hari tempoh penyimpanan.

Identifikasi bakteria aerobik mesofilik ditentukan menggunakan sistem BIOLOG Microsystem Biolog Microstation. Sejumlah 90 isolat, 30 daripada setiap sampel AP, SP dan MP telah dikenalpasti dan dipencilkan. Jenis bakteria yang mendominasi dalam YAN telah dikenalpasti iaitu *Staphylococcus sciuri* and *Ochrobactrum anthropi* yang didapati hadir di dalam setiap sampel manakala *Alcaligenes faecalis* hanya terdapat di dalam sampel SP dan MP.

Satu kajian lain telah dijalankan untuk menilai pertumbuhan bakteria aerobik mesofilik mendominasi YAN. Sampel dirawat dengan larutan *S. sciuri* (SS), *A. faecalis* (AF) dan *O. anthropi* (OA) pada kadar $\log 5-6$ CFU/g. APC, nilai pH dan penilaian deria dianalisa setiap hari sepanjang tempoh simpanan selama 4 hari yang disimpan pada suhu ambien (28 ± 2 °C). APC sampel AF dan SS meningkat secara signifikan ($P > 0.05$) pada kadar lebih mendadak berbanding sampel OA. Nilai pH sampel SS dan AF menurun secara signifikan ($P > 0.05$) berbanding sampel OA semasa tempoh penyimpanan. Ciri bau dan kekerasan dalam penilaian deria menunjukkan tahap kerosakan sampel SS adalah yang paling ketara. Ahli panel mengaitkan ciri bau yang terhasil dalam sampel SS dan AF dengan ciri bau dari sampel komersil. Pencirian kompaun mudah terpeluwap oleh *S. sciuri* dan *A. faecalis* telah dijalankan

menggunakan kaedah SPME-GCMS. Kompaun yang dikenalpasti adalah dari kumpulan alkohol dan disulfide, dimetil. Satu kajian berbeza dijalankan untuk menilai perubahan dalam pertumbuhan bakteria aerobik mesofilik dalam YAN yang telah dirawat dengan asid laktik dan nisin. Sampel yang tidak dirawat adalah sampel kawalan dan sampel yang dirawat dengan asid borik dijadikan perbandingan atas sebab ia selalu disalahgunakan dan di dalam YAN. Sampel dirawat dengan merendam sampel mee dalam larutan yang mengandungi asid laktik (v/v), nisin (w/v) atau asid borik acid (w/v) pada kadar 0.5%, 1.0% atau 1.5% selama 30 saat. Sampel disimpan pada suhu persekitaran (28 ± 2 °C). Kiraan bakteria aerobik mesofilik (APC), pH dan penilain deria dianalisa pada 0, 1, 3, 5, 7 dan 10 hari penyimpanan. Sejumlah 200 isolat telah dipencilkan dari sampel mi selama tempoh penyimpanan dan seterusnya diperincikan menggunakan ujian-ujian biokimia. Kiraan awal APC bagi sampel yang tidak dirawat ialah log 2.42 CFU/g manakala bagi sampel yang dirawat ialah sekitar log 3-4 CFU/g yang meningkat secara signifikan ($P < 0.05$) kepada sekitar log 10 CFU/g pada hari terakhir penyimpanan. Nilai pH bagi semua sampel menurun secara signifikan selama tempoh penyimpanan. Penilaian deria menunjukkan bahawa ciri bau dan tekstur adalah penyebab utama kerosakan dalam sampel yang dirawat dengan asid laktik, nisin dan asid borik. Data-data daripada hasil ujian menunjukkan potensi asid laktik dan nisin yang berfungsi secara individu adalah tidak signifikan untuk mengehadkan pertumbuhan bakteria aerobik mesofilik tetapi terdapat kemungkinan untuk mempertingkatkan potensi ini apabila asid laktik dan nisin berfungsi secara kombinasi atau bersama pengawet yang lain.

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I certify that a Thesis Examination Committee has met on 23 June 2008 to conduct the final examination of Siti Suwaibah binti Abd Ghaffar on her thesis entitled “**Aerobic Mesophilic Bacteria and Sensory Qualities of Yellow Alkaline Noodles During Storage**” 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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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.

SITI SUWAIBAH ABD GHAFAR

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LIST OF ABBREVIATIONS

AP	automated processing plants
APC	aerobic mesophilic counts
AF	<i>Alcaligenes faecalis</i>
MP	manual processing plants
OA	<i>Ochrobactrum anthropi</i>
SP	semi-automated processing plants
SS	<i>Staphylococcus sciuri</i>
YAN	yellow alkaline noodles
CFU	colony forming unit



CHAPTER 1

INTRODUCTION

Noodles and paste product have been in existence since ancient times. The origin of paste products is somewhat obscure (Matsuo, 1978). The cultivation of wheat and its utilization was in existence in Mesopotamia about 6000 to 7000 B.C. The Egyptian used wheat by about 2000 B.C. The art of making bread and paste products was believed to spread from the Middle East to the East, China to the West through Greece and Italy. Noodle products were presumably brought to Malaysia by the Chinese immigrants.

Noodles have evolved into diverse forms and preparations and become an essential part of local cuisine since its introduction from China to other Asian countries (Hou, 2001). There are many varieties of noodles produced from wheat flour which include yellow alkaline noodles (YAN), dry white salted noodles (mee sua), raw noodles (wanton noodles), instant noodles and udon noodles. Other type of noodles which are common in Asia but produce from various sources of cereal flours are bean thread noodles, vermicelli noodles, starch noodles and soba noodles (Coyle, 1982).

Amongst the noodle family, YAN are one of the most popular noodles in South East Asia. History of YAN in Malaysia probably began when it is brought by the Chinese immigrants' long time ago. From then on, they become a popular food among all races



in Malaysia (Jiman, 1983) especially among the Chinese and Malays. Noodles based on wheat are prepared mainly from three basic ingredients; flour, water and salt (Fu, 2007). YAN are made from flour, “kan sui” (a mixture of sodium and potassium carbonate and bicarbonate), salt and water and is usually parboiled. It is characterized by the presence of alkaline salts, giving the noodles their characteristics yellowness, texture, alkaline flavour, and high pH.

The high demands of YAN cause a rapid growth of noodles manufacturers in Malaysia. The noodles industry forms one of the major cottage food manufacturing industries in Malaysia. It is stated that there are not less than 200 YAN manufacturers all over in Malaysia (Jiman, 1983). Almost all of the factories produce noodles only for local consumption and virtually no export were done. Most of the YAN manufacturers are small family business operating on small scale basis catering local or neighborhood clients (Hatcher, 2001) and some operate from the backyard. The production of these products mostly involved batch processing operations (Karim, 1989), but in modern automated processing plants, manufacturers will have to invest in the purchase of automatic machine for the production of YAN. The importance of noodle products in Malaysia can best be summarized by the fact that amount of flour used for production of noodles in Malaysia accounts to about 40% of the total flour consumed in South East Asia (Anon, 1998). This shows the huge production of noodles products and in particular, YAN in Malaysia and it is expected that the figures have increased immensely although no proper documentation has been made so far. On the basis that there are high demands of YAN, unfortunately no specific studies

have been done to explore and enhance its market potential due to problems involving noodles shelf life. This product is highly susceptible to spoilage and had short shelf lives which are common problems amongst noodle manufacturers in Malaysia. The rapid deterioration of YAN during storage may result in high level of wastage and causes great losses to noodle manufacturers. The microbiological status of YAN has never been evaluated although it is one of the major foods consumed in Malaysia. Literature provides little information on microbiological quality of noodles and in particular, YAN. Therefore, shelf life is one of the most important criteria that need to be studied and look upon thoroughly to formulate YAN which are more shelf life stable.

The current research is an initial study to investigate and understand spoilage of YAN in term of microbiological and sensory qualities. Nonetheless, this study focuses on aerobic mesophilic bacteria because YAN are normally stored and displayed in grocery stores at ambient temperature ($28 \pm 2^{\circ}\text{C}$) or even if displayed in much sophisticated supermarket or hypermarkets in chill temperature ($5-10^{\circ}\text{C}$), temperature abuse during transportation or storage are likely to occur. Therefore this study aims to evaluate the microbial profile and spoilage characteristics of YAN associated with aerobic mesophilic bacteria. Studies on these profiles hopefully will be helpful in developing a more shelf stable YAN in the future.

Knowledge of predominant bacteria isolated from YAN in this study can create path to researchers whom are much interested in evaluating the spoilage potentials and

contributions of the composition of bacteria which probably lead to establishment of spoilage indicators for YAN.

At the moment, the solution to produce noodles which is shelf stable and cost effective has yet to be discovered and due to this, irresponsible noodle manufacturers used prohibited chemical substances such as boric acid which expose consumers to hazardous health risks. Consumer demand on “natural” preservatives following the abuse usage of boric acid in YAN drives this research to evaluate the potential of using nisin and lactic acid as preservatives in YAN. Information on the changes of growth profile and composition of aerobic mesophilic bacteria in YAN treated with lactic acid and nisin will be valuable in development of suitable preservation methods to prolong its shelf life in the future. The objectives of this research are:

1. To study aerobic mesophilic bacteria and sensory qualities of YAN produced from commercial processing plants during storage.
2. To isolate and identify composition of aerobic mesophilic bacteria isolated from YAN produced from commercial processing plants during storage and further investigate the spoilage potentials of the predominant bacteria.
3. To study aerobic mesophilic bacteria and sensory qualities of YAN treated with lactic acid and nisin during storage.

