DEVELOPMENT OF ACID-HYDROLYSED AND ENZYME-HYDROLYSED WINGED BEAN AND SOYBEAN PROTEINS

By

CHOO WEE SIM

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

February 2004

Dedicated to my beloved parents

Choo Quee Bong

and

Goo Sun Yen

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

DEVELOPMENT OF ACID-HYDROLYSED AND ENZYME-HYDROLYSED WINGED BEAN AND SOYBEAN PROTEINS

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Chairman: Associate Professor Sharifah Kharidah Syed Muhammad, Ph.D.

Faculty: Food Science and Biotechnology

Hydrolysed vegetable protein (HVP) is a savoury flavouring material obtained by acid, alkaline or enzymatic hydrolysis of a proteinaceous substrate. Traditionally, hydrolysis is often carried out using hydrochloric acid. However, the presence of 3-monochloropropane-1,2-diol (3-MCPD), a chemical contaminant in acid-hydrolysed vegetable protein pose a potential health risk to its users. Enzymatically hydrolysed vegetable protein, produced using proteolytic enzymes is a newer alternative to the traditional HVP. Common source of raw material in the industry for producing HVP is soybean. Winged bean (*Psophocarpus tetragonolobus* (L.) DC.) known locally as "kacang botor" is a tropical plant that contains high protein content, and virtually duplicate soybeans in composition and nutritional value. Based on Response Surface

Methodology (RSM), the optimum conditions for producing acid-hydrolysed winged bean (aHWBP) and soybean (aHSBP) proteins were 7 hours and 5 hours of hydrolysis with hydrochloric acid at 125°C, respectively. Proteolytic hydrolysis using 2.1% Flavourzyme 500L in aqueous 29% winged bean slurry (8% protein) for 11 hours, and proteolytic hydrolysis using 2.4% Flavourzyme in aqueous 26% soybean slurry (8% protein) for 16 hours were selected as the optimum conditions for producing enzyme-hydrolysed winged bean (eHWBP) and soybean (eHSBP) proteins, respectively. The proteolysis was preceded by treatment with 2% Viscozyme L in aqueous 29% winged bean or 26% soybean slurry. Alkaline thermal treatment using sodium hydroxide at pH 8.5 for 2 hours at 100°C effectively reduced the 3-MCPD contents of aHWBP and aHSBP to undetectable levels. It did not cause major changes to other chemical and sensory properties of the HVP. Significant reduction in bitterness of eHWBP and eHSBP without changes to their chemical and sensory properties were obtained after treatment of the hydrolysates with 0.1% (w/w) β-cyclodextrin. Enzymatic hydrolysis led to the production of 3-MCPD as well but the level was very low and within the permissible level. aHWBP and aHSBP were dark brown in colour with strong savoury flavours whereas eHWBP and eHSBP were lighter in colour and had a much less pronounced savoury flavour. Both aHWBP and eHWBP have their own distinct flavours which are different from that of soybean-derived flavours. aHWBP and aHSBP can be used as both taste-donor and taste-enhancer flavouring materials whereas eHWBP and eHSBP can be used as a base note flavouring material. Winged bean seeds can indeed be a new source of raw material for producing HVP.

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

PEMBANGUNAN PROTIN KACANG BOTOR DAN KACANG SOYA TERHIDROLISIS MELALUI KAEDAH ASIDIK DAN ENZIMATIK

Oleh

CHOO WEE SIM

Februari 2004

Pengerusi: **Profesor Madya Sharifah Kharidah Syed Muhammad, Ph.D.**

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Protin sayur terhidrolisis (HVP) ialah sejenis bahan perisa savuri yang diperolehi melalui hidrolisis sumber protin secara asidik, beralkali atau enzimatik. Secara tradisional, hidrolisis biasa dilakukan dengan menggunakan asid hidroklorik. Walaubagaimanapun, pencemaran 3-monochloropropane-1,2-diol (3-MCPD) dalam protin sayur terhidrolisis secara asidik menimbulkan risiko kesihatan kepada penggunanya. Protin sayur terhidrolisis secara enzimatik yang diperolehi dengan menggunakan enzim proteolitik merupakan satu alternatif yang lebih baru daripada protin sayur terhidrolisis yand diperolehi secara tradisional. Sumber bahan mentah yang biasa digunakan dalam industri untuk memproses protein sayur terhidrolisis ialah kacang soya. "Winged bean"

(*Psophocarpus tetragonolobus* (L.) DC.) yang dikenali di tempatan sebagai kacang botor merupakan sejenis tumbuhan tropikal yang mengandungi protin yang tinggi, dan menyerupai kacang soya dari segi kandungan dan nilai nutrisi. Berdasarkan kaedah respons permukaan (RSM), keadaan optima untuk memproses protin kacang botor terhidrolisis secara asidik (aHWBP) dan protin kacang soya terhidrolisis (aHSBP) ialah masing-masing 7 jam dan 5 jam untuk jangka masa hidrolisis dengan menggunakan asid hidroklorik pada suhu 125°C. Hidrolisis proteolitik dengan menggunakan 2.1% Flavourzyme 500L dalam sluri akuas 29% kacang botor (8% protin) selama 11 jam, dan hidrolisis proteolitik dengan menggunakan 2.4% Flavourzyme 500L dalam sluri akuas 26% kacang soya (8% protin) selama 16 jam telah masing-masing dipilih sebagai keadaan optima untuk memproses protin kacang botor terhidrolisis secara enzimatik (eHWBP) dan protein kacang soya terhidrolisis secara enzimatik (eHSBP). Hidrolisis proteolitik akan didahului dengan perawatan menggunakan 2% Viscozyme L dalam sluri akuas 29% kacang botor atau sluri akuas 26% kacang soya. Perawatan haba beralkali dengan menggunakan natrium hidroksida pada pH 8.5 selama 2 jam pada suhu 100°C berjaya menurunkan kandungan 3-MCPD dalam aHWBP dan aHSBP kepada paras yang tidak dapat dikesani. Ia tidak menyebabkan perubahan besar kepada ciri-ciri kimia dan sensori HVP itu yang lain. Penurunan rasa pahit dalam eHWBP dan eHSBP tanpa menyebabkan perubahan besar kepada ciri-ciri kimia dan sensori selepas perawatan hidrolisat dengan 0.1% β-cyclodextrin (berdasarkan berat) telah diperolehi. Hidrolisis secara enzimatik juga menyebabkan pembentukan 3-MCPD tetapi pada paras yang sangat rendah dan tidak melebihi paras yang dibenarkan. Warna aHWBP dan aHSBP ialah perang tua dengan perisa savuri yang kuat manakala warna eHWBP dan eHSBP lebih

muda dan mempunyai perisa savuri yang amat kurang. Kedua-dua aHWBP dan eHWBP mempunyai perisa savuri mereka tersendiri yang berlainan daripada perisa yang berasaskan kacang soya. aHWBP dan aHSBP boleh digunakan sebagai kedua-dua bahan perisa jenis penderma rasa dan penambahan rasa manakala eHWBP dan eHSBP boleh digunakan sebagai bahan perisa jenis dasar. Biji kacang botor memang boleh menjadi sumber baru untuk memproses protin sayur terhidrolisis.

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I certify that an Examination Committee met on 16 February 2004 to conduct the final examination of Choo Wee Sim on her Master of Science thesis entitled "Development of Acid-Hydrolysed and Enzyme-Hydrolysed Winged Bean and Soybean Proteins" 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 follows:

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

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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.

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CHOO WEE SIM

Date:

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