UNIVERSITI PUTRA MALAYSIA

DEVELOPMENT OF AN ENZYME-AIDED PRE-TREATMENT PROCESS FOR PRODUCTION OF PUMPKIN (Cucurbita moschata. L) POWDER

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By

FOROUGH SHAVAKHI

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

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Especially dedicated to my beloved husband

Rasoul

Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfilment of the requirement for the degree of Doctor of Philosophy

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Chairman: Professor Hasanah Mohd Ghazali, PhD

Faculty : Food Science and Technology

Pumpkin powder is one of the main products of pumpkin which can be easily stored and conveniently used in processed food. Spray drying is a highly appropriate process for heat-sensitive products and has been widely used in many fruits and vegetables. Appropriate feed viscosity and solids concentration are necessary to easily pump the feed into a spray dryer and obtaining an acceptable yield. On the other hand, the main problem in the spray drying of fruits and vegetables is stickiness and a part of this problem can be solved by addition of maltodextrin which offsets spray drying performance due to increasing viscosity. The inclusion of an enzymatic maceration step in fruits and vegetables processing leads to decreasing the viscosity. Therefore, the main

objective of this research was to develop a process for production of spray-dried pumpkin powder using enzyme as a pre-drying treatment.

The aim of the first experiment of this study was the maceration of raw pumpkin flesh in order to prepare a spray drying feed with an appropriate color, aroma, viscosity, and low molecular weight sugars. Therefore, the effects of different enzymes (pectinase, cellulase and amylase) on maceration or liquefaction of pumpkin were investigated in order to select the best treatment for preparation of the base feed for spray drying. Based on the results on aroma (Principal Component Analysis of zNose data), color, viscosity and sugar analysis, the best treatment to macerate pumpkin and produce a suitable base feed for spray drying was using 2.5% v/w Pectinex® Ultra SP-L (Novozymes, Denmark).

The viscosity value of macerated pumpkin with Pectinex® Ultra SP-L, 2.5 % v/w (0.102 Pa.s) was increased after addition of maltodextrin. To get more reduction in feed viscosity, Celluclast® 1.5 L (Novozymes, Denmark) was added to Pectinex-treated pumpkin. Therefore, the next set of experiments was conducted to optimize the effects of three variables namely Celluclast concentration (0-1% v/w, x_1), maltodextrin (MD) concentration (15–30% w/w, x_2) and spray dryer inlet temperature (150-190°C, x_3) on pumpkin powder characteristics using pumpkin macerated with 2.5% v/w Pectinex® Ultra SP-L as the base feed. The results indicated that the response surface models were significantly ($p \leq 0.05$) fitted for response variables of process yield, moisture content, water activity and hygroscopicity. The overall optimum region that would result in desirable powder characteristics was predicted to be obtained by a combined air inlet

temperature of 180°C, Celluclast concentration of 0.7 % (v/w) and MD concentration of 23% w/w. Furthermore, the results showed that the glass transition temperature (T_g) of pumpkin powder increased with an increase in air inlet temperature and MD concentration. Also, the T_g of pumpkin powder was found to increase with increasing of concentration of Celluclast up to 0.5% v/w. Conversely, increasing the concentration of Celluclast above 0.5% v/w (and up to 1% v/w), decreased the T_g of the pumpkin powder. Surface morphology images of powder captured by scanning electron microscopy (SEM) confirmed the results.

In addition, the effects of the three variables mentioned above on reconstitution of pumpkin powder based on color, water solubility index (WSI) and water absorption index (WAI) were described. The results revealed that a second order polynomial regression model was fitted to all response variables studied except for *a*-value in powder and reconstituted powder, reconstituted hue-value and also ΔE and WSI with good determination coefficients (R^2>80%). Total color change (ΔE) of initial pumpkin puree and reconstituted pumpkin powder solution varied from 16.08 to 20.04. Solubility index of the pumpkin powder increased as a result of adding MD and increasing the air inlet temperature. Water absorption index of pumpkin powder in this study varied from 66.0 to 140.48, indicating that powders was able to hold water at a maximum of 1.4 times their weight. Therefore, it is not suitable as a thickening ingredient.

In the next stage, the effects of two anti-caking agents namely tri-calcium phosphate (TCP) and calcium silicate (CS) at 0, 0.1, 0.2 g of each/100g puree on pumpkin powder

characteristics before and after storage for 8 weeks in ambient condition were investigated. Anti-caking agents showed positive effect on the cyclone recovery in spray drying of pumpkin powder and could improve the recovery of spray-dried pumpkin powder. Powder containing anti-caking agents showed higher T_g during 8 weeks of storage indicating that these additives acted as successful anti-caking agents.

Finally, rheological characteristics of enzymaticaly-macerated pumpkin and also spray dried reconstituted powder were evaluated to determine the potential application of pumpkin powder. Results revealed that the enzymatically-macerated pumpkin exhibited a yield stress and shear stress–shear rate data was best fit to Herschel-Bulkley Model at any given temperature with $R>0.94$, while shear stress–shear rate data of reconstituted powder solutions were fitted to Power Law Model without definite yield stress. Consistency coefficients (K) of enzymatically-macerated pumpkin was decreased at 25ºC (4.44 $Pa.s^n$) to 55ºC (3.41 $Pa.s^n$) followed by an increase at 65ºC (4.78 $Pa.s^n$) and afterward. Rheologically, the reconstituted powder behaved similarly to fruit juice. Flow activation energy of consistency index and apparent viscosity were 7.37 and 7.53 kJ/mol respectively. Based on rheological properties of reconstituted powder solution, one of the potential applications of spray-dried pumpkin powder is its suitability for preparation of pumpkin juice.

The finding of this research revealed that Pectinex® Ultra SP-L in different concentrations (2.5, 3.5, 4.5 and 5.5% v/w) and incubation times (2.5, 2, 1.5 and 1 h, respectively) was able to macerate pumpkin. The optimum concentration of 2.5% v/w

was used for preparation of base feed for spray drying since it is more economical. Celluclast® 1.5 L was able to decrease the feed viscosity and increase the recovery of pumpkin powder. The best anti-caking agent was tri-calcium phosphate, based on a study on storage for 8 weeks. Results obtained showed that the pumpkin powder prepared in this study was not suitable as a thickening ingredient but can be used in the preparation of pumpkin juice.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia sebagai memenuhi keperluan untuk ijazah Doktor Falsafah

PEMBANGUNAN PROSES PRA-RAWATAN ENZIM UNTUK PENGHASILAN SERBUK LABU (*Cucurbita moschata*. L)

Oleh

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March 2011

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Serbuk labu merupakan salah satu produk utama buah labu yang mudah disimpan dan sesuai digunakan dalam penyedian makanan terproses. Pengeringan semburan (*spray-drying*) adalah proses yang paling sesuai untuk produk yang sensitif terhadap haba dan teknik ini telah banyak digunakan untuk buah-buahan dan sayur-sayuran. Kesesuaian di dalam kelikatan sampel dan kepekatan pepejal adalah diperlukan untuk memudahkan sampel makanan dipam masuk ke dalam pengering sembur (*spray dryer*), dan dengan demikian boleh memperolehi hasil yang memadai. Sebaliknya pula, masalah utama dalam pengeringan semburan buah-buahan dan sayur-sayuran adalah sifat kelekitan dan sebahagian dari masalah ini dapat diatasi dengan penambahan bahan maltodekstrin yang dapat meningkatkan prestasi penyembur kering dengan peningkatan kelikatan. Dengan

penambahan langkah maserasi enzimatik dalam pemprosesan buah-buahan dan sayur-sayuran akan menyebabkan penurunan kelikatan. Oleh itu, tujuan utama kajian ini adalah untuk membangunkan proses penghasilan semburan kering serbuk labu dengan menggunakan enzim sebagai rawatan pra-pengeringan.

Objektif pertama dalam kajian ini adalah untuk maserasikan isi labu mentah untuk proses pengeringan semburan dengan mendapatkan warna, rasa, kelikatan dan pengurangan berat molekul gula. Oleh itu, kesan enzim yang berbeza (pektinase, selulase, dan amilase) pada proses maserasi atau pencairan labu dikaji untuk memilih rawatan yang terbaik sebagai asas bahan untuk penyemburan kering. Berdasarkan keputusan dalam bab ini ke atas rasa aroma (Analisis komponen utama data zNose), warna, kelikatan dan analisis gula, rawatan yang terbaik untuk proses maserasi labu dan menghasilkan bahan asas untuk pengeringan semburan adalah dengan menggunakan 2.5% v/w Pectinex® Ultra SP-L (Novozymes, Denmark).

Nilai kelikatan untuk labu yang telah dimaserasikan dengan 2.5% v/w Pectinex® Ultra SP-L (0.102 Pa.s) meningkat selepas penambahan MD. Bagi mendapatkan lebih banyak pengurangan kelikatan bahan asas, Celluclast® 1.5L (Novozymes, Denmark) telah ditambahkan ke dalam labu yang telah dirawat dengan Pectinex. Oleh itu, set eksperimen yang berikutnya adalah untuk mengoptimumkan kesan daripada tiga pembolehubah, iaitu kepekatan Celluclast (0-1%, v/w, x_1), kepekatan maltodekstrin (MD) (15-30% w/w, x_2) dan suhu masuk (150-190°C, x_3) pengering sembur terhadap sifat serbuk labu dengan menggunakan labu yang dimaserasikan dengan 2.5%

Pectinex® Ultra SP-L sebagai bahan asas. Keputusan kajian menunjukkan bahawa model respon permukaan secara signifikan ($p \leq 0.05$) sesuai dengan respon pembolehubah hasil proses, kandungan air, kejelkitan, aktiviti air dan higroskopisiti. Bahagian optima keseluruhan yang menghasilkan sifat serbuk yang dikehendaki diramal akan diperoleh daripada kombinasi suhu udara masuk (180°C), kepekatan Celluclast (0.7% v/w) and kepekatan MD sebanyak 23% w/w. Tambahan lagi, hasil kajian juga menunjukkan bahawa perubahan suhu kaca (T_g) bagi serbuk labu bertambah dengan peningkatan suhu udara masuk dan kepekatan MD. Selain itu, T_g serbuk labu turut meningkat dengan peningkatan kepekatan Celluclast sehingga 0.5%. Sebaliknya, peningkatan Celluclast melebihi 0.5% (sehingga 1%) akan menurunkan kadar T_g serbuk labu. Imej morforlogi permukaan serbuk yang dirakamkan dengan menggunakan mikroskop imbasan elektron (SEM) telah memberi kepastian terhadap keputusan yang diperolehi.

Di samping itu, kesan tiga pembolehubah yang disebutkan di atas terhadap penyerapan semula serbuk labu berdasarkan warna, indeks keterlarutan air (WSI) dan indeks penyerapan air (WAI) yang dinyatakan. Keputusan kajian menunjukkan model regresi polinomial susunan kedua adalah sesuai dengan semua respon pembolehubah yang dikaji, kecuali nilai *a* dalam serbuk dan penyerapan semula serbuk, nilai rona penyerapan semula dan juga ΔE and WSI dengan penentuan pekali yang baik (R^2 lebih tinggi daripada 80%). Jumlah perubahan warna (ΔE) awal puri labu dan larutan penyerapan semula serbuk labu adalah dari 16.08 sehingga 20.04. Indeks keterlarutan serbuk labu meningkat hasil dari penambahan MD dan peningkatan suhu udara masuk.

Kadar indeks penyerapan air serbuk labu adalah dari 66.0 hingga 140.48, menunjukan serbuk labu boleh menampung air secara maksimumnya 1.4 kali ganda beratnya dan oleh itu tidak sesuai dijadikan sebagai bahan pemekat.

Pada tahap berikutnya, kesan dua ejen pencegah penggumpalan iaitu trikalsium fosfat (TCP) and kalsium silikat (CS) pada 0, 0.1, 0.2 g setiap satu/ 100g puri terhadap cirri serbuk labu sebelum dan selepas disimpan selama 8 minggu dalam keadaan ambien dikaji. Ejen pencegah penggumpalan menunjukkan kesan positif pada perolehan siklon dalam penyemburan kering serbuk labu serta dapat meningkatkan perolehan serbuk labu yang di sembur kering. Serbuk yang mengandungi ejen pencegah penggumpalan yang menunjukkan kekurangan ciri kelekitan dan lebih tinggi T_g dalam masa 8 memberi implikasi bahan tambahan ini berjaya bertindak sebagai ejen pencegah penggumpalan.

Akhirnya, sifat reologi labu yang telah dienzim dan dimaserasikan dan juga serbuk yang diserap semula dengan semburan kering dinilai untuk menentukan potensi penggunaan serbuk labu. Keputusan menunjukkan bahawa labu yang telah dienzim dan dimaserasikan secara enzimatik menghasilkan tekanan dan data ‘tegasan ricihan-kadar ricihan’ yang berpadanan dengan model Herschel-Bulkley pada semua suhu yang ditetapkan (R^2>0.94), sedangkan data ‘tegasan ricihan-kadar ricihan’’ penyerapan semula larutan serbuk berpadanan dengan hukum kuasa tanpa hasil tekanan. Konsistensi pekali (K) labu yang telah dienzim dan dimaserasikan secara enzimatik mengalami penurunan dari 25°C (4.44Pa.s^n) hingga 55°C (3.42 Pa.s^n) diikuti dengan peningkatan pada 65°C (4.78 Pa.s^n) dan seterusnya. Secara reologi, ciri penyerapan

semula serbuk hampir sama dengan jus buah. Aliran pengaktifan tenaga pada indeks konsistensi dan kelikatan yang nyata masing-masing adalah 7.37 dan 7.53 kJ/mol. Berdasarkan sifat reologi larutan penyerapan semula serbuk, salah satu potensi dalam aplikasi serbuk labu yang disembur kering adalah kesesuaiannya untuk penyediaan jus labu.

Penemuan kajian ini menunjukkan bahawa Pectinex® Ultra SP-L pada kepekatan yang berbeza (2.5, 3.5, 4.5 dan 5.5% v/w) dan masa inkubasi (2.5, 2, 1,5 dan 1 jam, masing-masing) boleh maserasikan labu. Namun, 2.5% v/w digunakan dalam penyediaan bahan asas untuk pengeringan semburan kerana lebih ekonomik. Celluclast® 1.5 L mampu menurunkan kelikatan dan meningkatkan pemulihan serbuk labu. Tri-kalsium fosfat adalah ejen pencegah penggumpalan yang terbaik untuk menyekat proses penggumpalan semasa simpanan selama 8 minggu. Serbuk labu yang disediakan dalam kajian ini aalah tidak sesuai sebagai bahan pemekat, tetapi boleh digunakan dalam penyediaan jus labu.

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I certify that a Thesis Examination Committe has met on 28 March 2011 to conduct the final examination of **FOROUGH SHAVAKHI** on her Ph.D. thesis entitled "**DEVELOPMENT OF AN ENZYME-AIDED PRE-TRETMENT PROCESS FOR PRODUCTION OF PUMPKIN (*Cucurbita moschata*. L) POWDER**" 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 candidate be awarded the Ph.D. degree. Members of the Thesis Examination Committee were as follows:

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DECLARATION

I declare that the thesis is my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously, and is not concurrently, submitted for any other degree at Universiti Putra Malaysia or at any other institution.

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FOROUGH SHAVAKHI

Data: 28 March 2011

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