



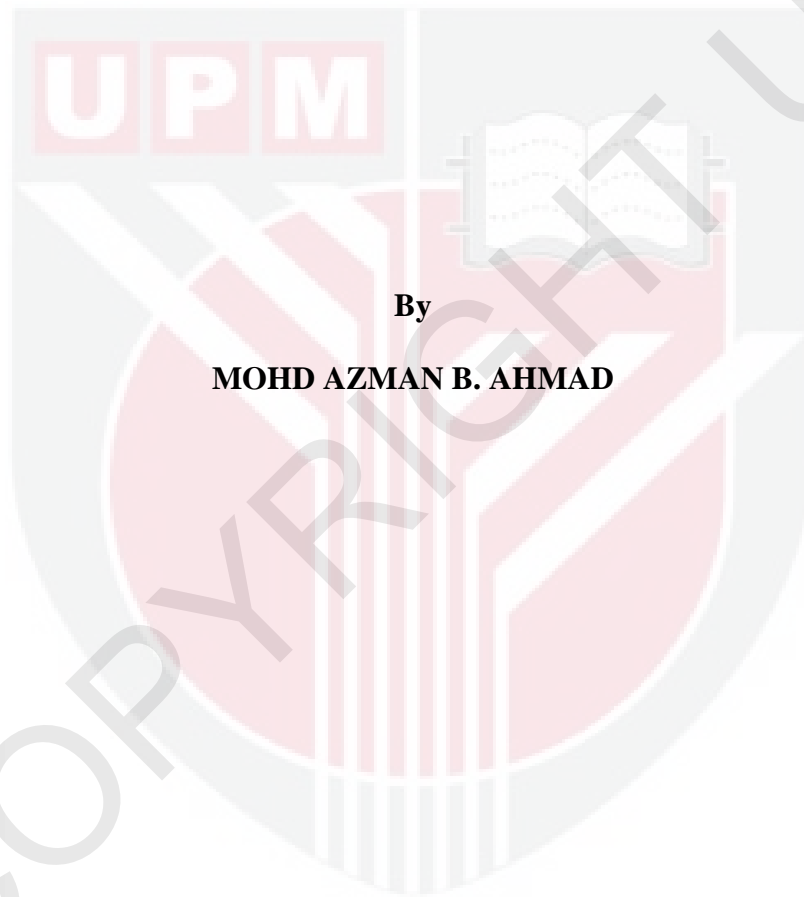
UNIVERSITI PUTRA MALAYSIA

**ENHANCEMENT OF FREEZE AND SPRAY DRYING TECHNIQUES FOR
THE PRESERVATION OF *ASPERGILLUS NIGER* SPORES**

MOHD AZMAN B. AHMAD

FBSB 2009 38

**ENHANCEMENT OF FREEZE AND SPRAY DRYING TECHNIQUES FOR THE
PRESERVATION OF *ASPERGILLUS NIGER* SPORES**



By

MOHD AZMAN B. AHMAD

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

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

ENHANCEMENT OF FREEZE AND SPRAY DRYING TECHNIQUES FOR THE PRESERVATION OF *ASPERGILLUS NIGER* SPORES

By

MOHD AZMAN B. AHMAD

SEPTEMBER 2009

Chairman: Dr. Rosfarizan Mohamad

Faculty: Biotechnology and Biomolecular Sciences

In this study, *Aspergillus niger* spores were powderised using freeze and spray drying techniques prior to the treatment of palm kernel cake (PKC) for subsequently used as an animal feed particularly for poultry. Initially, effects of several harvesting agents on the recovery of the spores from the growth solid media were investigated. Prior to freeze and spray drying processes, several cryoprotective agents (skimmed milk, maltose, glucose, lactose and leucine) were added to the prepared spore suspension in order to maintain high cell viability.

In the freeze drying process, cell viability was reduced from 1.2×10^{12} CFU/g to 2.7×10^{10} CFU/g when using 10% w/v of glucose as the cryoprotective agent and achieved about 1.65 of log reduction. Meanwhile, highest cell viability (1.8×10^{11} CFU/g) was exhibited using 10% w/v of maltose compared to 10% w/v of skimmed milk (2.7×10^{10} CFU/g), which examined immediately after freeze drying process. However, 10% w/v skimmed milk showed the best cryoprotective agent for long storage life. The viable cells were survived up to 61 weeks of storage using 10% w/v skimmed milk, while it was only 49 weeks for both 10% w/v glucose and 10% w/v maltose.

In the spray drying process, the cell viability reduced from 6.0×10^{12} CFU/g to 6.1×10^{10} CFU/g when using skimmed milk as the protective agent. The combination of 10% w/v lactose and 10% w/w leucine as the protective agents prior to spray drying process showed drastically decreased of cell viability from 5.0×10^{12} CFU/g to 4.3×10^{10} CFU/g. However, increased of leucine percentage (12% w/w leucine) in the combination of the cryoprotective agents slightly improved the cell viability from 4.3×10^{10} CFU/g to 6.0×10^{10} CFU/g.

Comparison between freeze drying and spray drying process in terms of drying methods, cell viability immediately after drying process, survival rate, and long term storage product was made. As a result, the freeze drying process using combination of selected cryoprotective agents was showed to be the preferred method for long storage life. Generally, freeze drying process exhibited highest cell viability, high survival rate of spores and long term storage stability of product. Meanwhile, spray drying process showed shorter storage life of viable cells, which only last up to week 29 as compared to week 61 for freeze drying process and there were no viable cells detected at the end of each storage life.

Higher reducing sugars were obtained from both spray dried (141.10 mg/g PKC) and freeze dried (86.83 mg/g PKC) *A. niger* spores compared to fresh *A. niger* spores that produce 58.57 mg/g PKC. While for mannanase production for spray dried spores was 341.75 U/g of PKC, freeze dried was 330.25 U/g of PKC and fresh *A. niger* 346.75 U/g of PKC. Powderised spores have shown promising results to reduced NDF, ADF, and CF in PKC and increase CP.

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

**PENINGKATAN TEKNIK PENYEJUK KERING DAN PENYEMBUR KERING
UNTUK PENYIMPANAN SPORA *ASPERGILLUS NIGER***

Oleh

MOHD AZMAN B. AHMAD

SEPTEMBER 2009

Pengerusi: Dr. Rosfarizan Mohamad

Fakulti: Bioteknologi dan Sains Biomolekul

Dalam kajian ini, spora *Aspergillus niger* dijadikan serbuk menggunakan teknik penyejuk kering dan penyembur kering sebelum dicampurkan kepada kek isirong kelapa sawit (PKC) untuk digunakan seterusnya sebagai makanan haiwan untuk industri ternakan. Pada mulanya, kesan beberapa agen penuaian dalam mendapatkan semula spora daripada media pepejal pertumbuhan dikaji terlebih dahulu. Sebelum proses penyejuk kering dan penyembur kering, beberapa agen perlindungan (susu skim, maltosa, glukosa, laktosa dan leucine) ditambah untuk menyediakan ampaiian spora untuk mengekalkan kadar sel hidup yang tinggi.

Dalam proses penyejuk kering, kadar sel hidup berkurangan dari 1.2×10^{12} CFU/g kepada 2.7×10^{10} CFU/g menggunakan 10% w/v glukosa sebagai agen perlindungan dan mencapai 1.65 log pengurangan. Sementara itu, kadar sel hidup yang tinggi (1.8×10^{11} CFU/g) diperolehi seurus selepas proses penyejuk kering menggunakan maltosa berbanding dengan susu skim (2.7×10^{10} CFU/g). Walaubagaimanapun, susu skim menjadi agen perlindungan yang terbaik untuk penyimpanan jangka panjang. Sel hidup

dapat hidup sehingga 61 minggu dengan menggunakan susu skim berbanding dengan glukosa dan maltosa hanya bertahan selama 49 minggu.

Dalam proses penyembur kering, kadar sel hidup berkurangan dari 6.0×10^{12} CFU/g kepada 6.1×10^{10} CFU/g apabila menggunakan susu skim sebagai agen perlindungan. Kombinasi 10% w/v laktosa dan 10% w/w leucine sebagai agen perlindungan sebelum proses penyembur kering menunjukkan penurunan drastik pada kadar sel hidup dari 5.0×10^{12} CFU/g kepada 4.3×10^{10} CFU/g. Walaubagaimanapun, peningkatan peratusan leucine (12% w/w leucine) dalam kombinasi agen perlindungan meningkatkan kadar sel hidup dari 4.3×10^{10} CFU/g kepada 6.0×10^{10} CFU/g.

Perbandingan di antara proses penyejuk kering dan proses penyembur kering dalam pelbagai aspek seperti, kaedah pengeringan, kadar daya hidup sel selepas proses pengeringan, kadar keupayaan untuk hidup, dan penyimpanan produk jangka panjang dilakukan. Dapat disimpulkan, proses penyejuk kering menggunakan kombinasi agen perlindungan yang terpilih menunjukkan kaedah yang lebih sesuai untuk penyimpanan jangka panjang. Secara umumnya, proses penyejuk kering memberikan kadar sel hidup yang paling tinggi, kadar keupayaan hidup spora yang tinggi, dan kadar kestabilan penyimpanan produk yang lama. Sementara itu, proses penyembur kering menunjukkan kadar sel hidup yang pendek, di mana hanya bertahan sehingga minggu ke 29 berbanding 61 minggu bagi proses penyejuk kering dan tiada sel hidup dikesan pada akhir jangka hayat penyimpanan.

Hasil guna penurun yang tinggi diperolehi dari kedua-dua spora penyembur kering (141.10 mg/g PKC) dan penyejuk kering (86.83 mg/g PKC) daripada spora *A. niger*

yang segar menghasilkan 58.57 mg/g PKC. Sementara itu, penghasilan enzim mannanase daripada spora penyembur kering ialah 341.75 U/g PKC, penyejuk kering 330.25 U/g PKC dan spora segar 346.75 /g PKC. Serbuk spora menunjukkan keputusan yang memberangsangkan untuk mengurangkan NDF, ADF, CF dan meningkatkan CP.



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I certify that an Examination Committee met on **4 September 2009** to conduct the final examination of **Mohd Azman B. Ahmad** on his **Master** thesis entitled “**Enhancement of Freeze and Spray Drying Techniques for the Preservation of *Aspergillus Niger* Spores** 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.

Members of the Theses Examination Committee were as follows:

Associate Professor Dr. Suraini Abdul Aziz, PhD.

Faculty Biotechnology and Biomolecular Sciences
Universiti Putra Malaysia
(Chairman)

Associate Professor Dr. Ling Tau Chuan, PhD.

Faculty Engineering
Universiti Putra Malaysia
(Internal Examiner)

Dr. Nor Aini Abdul Rahman, PhD.

Faculty Biotechnology and Biomolecular Sciences
Universiti Putra Malaysia
(Internal Examiner)

Associate Professor Dr. Aidil Abdul Hamid, PhD.

Faculty Science and Technology
Universiti Kebangsaan Malaysia
(External Examiner)

BUJANG KIM HUAT, PhD.

Professor and Deputy Dean
School of Graduate Studies
Universiti Putra Malaysia

Date:

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

Main Supervisor, Ph.D.

Associates Professor Dr. Rosfarizan Mohamad, PhD

Faculty Biotechnology and Biomolecular Sciences

Universiti Putra Malaysia

(Chairman)

Co-Supervisor, PhD.

Professor Dr. Arbakariya Ariff, PhD

Faculty of Biotechnology and Biomolecular Sciences

Universiti Putra Malaysia

(Member)

HASANAH MOHD GHAZALI, PhD.

Professor and Dean

School of Graduate Studies

Universiti Putra Malaysia

Date:

DECLARATION

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

MOHD AZMAN B. AHMAD

Date:

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