



UNIVERSITI PUTRA MALAYSIA

**EVALUATION OF SOLID STATE FERMENTATION BY ASPERGILLUS  
NIGER TO IMPROVE THE NUTRITIVE VALUE OF PALM KERNEL  
CAKE FOR BROILERS**

**KHIN HNIN SWE.**

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**By**

**KHIN HNIN SWE**

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the requirement for the degree of Doctor of Philosophy

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**KHIN HNIN SWE**

**December 2004**

**Chairman : Professor Abd. Razak Bin Alimon, Ph.D.**

**Faculty : Agriculture**

A study was conducted to examine the effects of solid-state fermentation (SSF) by *Aspergillus niger* on the nutrient contents of palm kernel cake (PKC). The effects of different types and levels of alcohols were investigated on the enzyme activity and nutrient contents of fermented PKC+ rice bran (RB). The results showed that the neutral detergent fibre (NDF) and acid detergent fibre (ADF) contents of fermented substrate (FS) (PKC+RB) were decreased by 47.3% and 22.6% respectively, while the crude protein (CP) was increased by 28.7% after 8 days of fermentation and the activity of mannanase was 109.7 U/g at 6 days of fermentation. There were no significant effects ( $P>0.05$ ) of ethanol and methanol on the nutrient contents and citric acid production in the fermented substrate.

An experiment was conducted to improve the fermentation process of *A. niger* on PKC. Rice bran was added to PKC in varying proportions. Three different proportions of rice bran (10%, 20% and 30% of total substrate) were used and the fermentation was carried out for 8 days. The results showed that the nutrient contents of FS were not significantly ( $P>0.05$ ) affected by the RB added. Nevertheless, among the different substrate groups used, 70% PKC+ 30% RB showed the lowest concentration of ochratoxin (43.3ppb). The ochratoxin level of 100% PKC substrate group was 49.8 ppb. Therefore, based on the ochratoxin level, the substrate group consisting of 70% PKC + 30% RB was used as the substrate for SSF. The fermented substrate was used as an ingredient in the broiler ration.

Four experimental diets were used in the feeding trial. They were: 1) control diet (without PKC), 2) diet containing 20% PKC, 3) diet containing 20% FS and 4) diet containing 25% FS. A total of 60 birds, 15 birds per treatment, 3 weeks old Arbor Acre were used. The birds were placed in individual cages. The growth trial was continued up to 42 days. The results showed poor ( $P<0.05$ ) growth performances of birds fed diets containing FS and the best performance was recorded in birds fed control diet. There were no significant ( $P>0.05$ ) differences in the performance of birds fed diet containing 20% PKC and those fed control diet. Cumulative FCR of birds fed control diet was 1.85 whereas that of birds fed dietary treatment 4 was 2.42.

The study on histopathological examination of visceral organs of chickens fed diet containing FS showed some lesions of ochratoxicosis where the immune organ (bursa

of Fabricius) was adversely affected. The lesions in liver were less than those of the kidney.

An attempt was carried out to delay sporulation of fungal culture. In this study, 14% ammonium sulphate was observed as the best concentration to add in culture during SSF of substrate (PKC+RB) by *A. niger* to delay sporulation. The digestibility of fibre was improved by adding ammonium sulphate in the culture.

Although the nutritive value of fermented PKC was enhanced by SSF with *A. niger*, the feeding trial carried out in broilers showed no beneficial effect of using the fermented substrate as a ration component in poultry feed. Nevertheless, the information obtained in this study would be beneficial in the understanding of the biochemical changes that occur in PKC during SSF with *A. niger*.

Abstrak yang dikemukakan kepada Senat Universiti Putra Malaysia untuk memenuhi keperluan bagi ijazah Doktor Falsafah.

**PENILAIAN FERMENTASI DALAM BENTUK PEPEJAL HAMPAS KELAPA  
OLEH *ASPERGILLUS NIGER* UNTUK MENINGKATKAN NILAI NUTRITIF  
HAMPAS KELAPA SAWIT UNTUK AYAM PEDAGING**

**Oleh**

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Satu kajian telah dijalankan untuk menilai kesan fermentasi dalam bentuk pepejal oleh *Aspergillus niger* ke atas kandungan nutrien dari hampas kelapa sawit.. Dalan kajian pertama, kesan dari jenis dan tahap kandungan alkohol yang berbeza telah dikaji ke atas aktiviti enzim dan kandungan nutrien dari hampas padi + PKC yang telah difermentasikan. Keputusan telah menunjukkan kandungan NDF dan ADF dari substrat yang difermentasi (FS) (PKC+RB) adalah menurun sebanyak 47.3% dan 22.6%, sementara protein kasar meningkat 28.7% selepas 8 hari fermentasi dan aktiviti mannanase adalah 109.7 U/g pada fermentasi selama 6 hari. Tiada perbezaan signifikan ( $P>0.05$ ) kesan dari ethanol dan methanol pada kandungan nutrien dan penghasilan asid sitrik dalam substrat telah difermentasi.

Kajian telah dijalankan untuk menambahbaik proses fermentasi oleh *A. niger* ke atas PKC. Hampas padi telah ditambah kepada PKC dalam kadar yang berbeza. Tiga kadar hampas padi (10%, 20% dan 30% dari jumlah substrat) telah digunakan dan fermentasi telah dilakukan selama 8 hari. Keputusan telah menunjukkan bahawa kandungan nutrien dari FS adalah tambahan hampas padi tiada perbezaan yang signifikan. Walaupun begitu, antara kumpulan substrat yang digunakan, 70% PKC + 30% RB menunjukkan kepekatan oktratoksin (43.3 ppb) yang terendah. Oktratoksin pada kumpulan tahap substrat PKC 100% adalah 49.8 ppb. Oleh itu, berdasarkan kepada tahap oktratoksin kumpulan substrat yang mengandungi 70% PKC + 30% RB telah digunakan sebagai substrat untuk SSF. Substrat yang diperlakukan telah digunakan sebagai bahan kandungan dalam ransum ayam daging.

Empat ujian diet telah digunakan dalam pengujian pemakanan. Mereka adalah: 1) diet kawalan (tanpa PKC), 2) diet mengandungi 20% PKC, 3) diet mengandungi 20% FS dan 4) diet mengandungi 25% FS. Sejumlah dari 60 ekor ayam Arbor Acre, 15 ekor setiap kumpulan, dan berumur 3 minggu digunakan. Ujian tumbesaran dilakukan dalam sangkar berasingan sehingga berumur 42 hari. Keputusan menunjukkan tumbesaran adalah rendah untuk ayam yang diberi diet FS dan terbaik adalah kumpulan ayam yang diberi diet kawalan. Tiada perbezaan yang signifikan ( $P>0.05$ ) dalam tumbesaran ayam yang diberi makan diet mengandungi 20% PKC dan yang diberi makanan diet kawalan. FCR terkumpul dari ayam yang diberi makan diet kawalan. FCR terkumpul dari ayam yang diberi makan diet kawalan adalah 1.85 sementara ayam yang diberi diet 4 adalah 2.42.

Kajian ke atas histopatologi terhadap organ visceral ayam yang diberi diet mengandungi FS menunjukkan kerosakan oktratoksikosis di mana bursa Fabricius menunjukkan kesan sebaliknya. Kerosakan pada hati adalah kurang berbanding dengan buah pinggang

Kajian telah dijalankan untuk menangguhkan sporulisasi kulat. Dalam kajian ini, 14% ammonium sulfat dicatatkan sebagai kepekatan terbaik untuk ditambahkan dalam kulur sewaktu SSF dari substrat (PKC +RB) oleh *A. niger* untuk penangguhan sporulisasi. Pencernaan tenaga dan gentian adalah meningkat dengan penambahan ammonium sulfat pada kulur.

Walaupun nilai nutritif dan PKC difermentasi adalah ditingkatkan oleh SSF dengan *A. niger*, percubaan pemakanan dalam ayam daging menunjukkan tiada kesan baik dari penggunaan bahan difermentasi sebagai bahan ration makanan ayam. Walaupun begitu, maklumat yang telah diperolehi dari kajian ini akan memanfaat dalam kefahaman perubahan yang berlaku dalam PKC semasa SSF dengan *A. niger*.

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## **LIST OF ABBREVIATIONS**

AABA	- L- $\alpha$ -Amino-n-butyric acid
ADF	- Acid Detergent Fibre
AME	Apparent metabolizable energy
Ca	- Calcium
CFU	- Colony Forming Unit
C:N	- Carbon Nitrogen Ratio
CP	- Crude Protein
C/P ratio	- Calorie Protein ratio
DM	- Dry Matter
EE	- Ether Extract
FCR	- Food Conversion Ratio
FS	- Fermented substrate (PKC+RB)
GE	- Gross Energy
HPLC	- High Performance Liquid Chromatography
ISR	Internal Standard Ratio
LD <sub>50</sub>	- Lethal Dose at 50%
MBTH	- 3-methyl-2-benzothiazolone hydrochloride
NDF	- Neutral Detergent Fibre
NRC	- National Research Council
NS	Not Significantly Different
NSPs	- Non Starch polysaccharides

<b>OA</b>	Ochratoxin A
<b>P</b>	- Phosphorous
<b>PDA</b>	- Potato Dextrose Agar
<b>PKC</b>	- Palm Kernel Cake
<b>PKM</b>	- Palm Kernel Meal
<b>POME</b>	- Palm Oil Mill Effluent
<b>POS</b>	Palm Oil Sludge
<b>PPF</b>	- Palm Press Fibre
<b>RB</b>	Rice Bran
<b>SD</b>	Standard Deviation of t-test
<b>SDW</b>	Sample Dry Weight
<b>SPSS</b>	- Statistical Package for Social Scientist
<b>SSF</b>	- Solid-State Fermentation
<b>TME</b>	- True Metabolizable Energy