



UNIVERSITI PUTRA MALAYSIA

EFFECTS OF LIQUID METABOLITES FROM *LACTOBACILLUS PLANTARUM* ON GASTROINTESTINAL HEALTH, CHOLESTEROL LEVEL AND GROWTH PERFORMANCE OF BROILER CHICKENS

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Thesis Submitted to the school of Graduate Studies, Universiti Putra Malaysia, in
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DEDICATION

This write up is dedicated to my beloved son who grows up with my absence for 4 years and is always all my love.



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

EFFECTS OF LIQUID METABOLITES FROM *LACTOBACILLUS PLANTARUM* ON GASTROINTESTINAL HEALTH, CHOLESTEROL LEVEL AND GROWTH PERFORMANCE OF BROILER CHICKENS

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The use of antibiotics as growth promotants has become unacceptable. Liquid metabolites seem to have a bright future as antibiotic substitutes because of their natural antibacterial properties. Three experiments were conducted to study the effect of feeding liquid metabolite (LM) mixtures produced from 4 strains of *Lactobacillus plantarum* on growth performance in broiler chickens.

Different liquid metabolites produced by RG14, RG11, RS5, and RI11 of locally isolated *L. plantarum* were studied against the indicator strain *P. acidilactici* in the first experiment. The pH, bacteriocin activity, inhibitory zone diameter and viable plate count

of liquid metabolites from four *L. plantarum* strains RG11, RG14, RI11 and RS5 and their mixtures were determined. The studies showed that metabolites produced by the strains of *L. plantarum* have different abilities to inhibit the indicator; especially metabolite mixtures exhibited stronger bacteriocin activity and clear zone diameter compared to single RS5, RI11, RG14 and RG11 strains. Mixtures of these metabolites further enhance the inhibitory effects against the indicator *P. acidilactici*. These metabolites have synergistic effects as they could be applied as biopreservatives in food industry and feed additives for animal.

In the second experiment, liquid metabolite mixtures produced by four strains of *L. plantarum* were used in feed as antibiotics replacement in broiler chickens. A total of 294 male Cobb broiler chickens were divided into 7 treatments with different diets: 5 levels of liquid metabolites (LM) (0.5%, 1.0%, 1.5%, 2.0%, and 2.5%) and 2 controls (negative control and positive control). Improved growth performance, reduced *Enterobacteriaceae* and increased lactic acid bacteria counts and increased small intestine villi height were found in the treatments supplemented different levels of metabolite mixtures. The chicken supplemented with 1.0% LM had the best results among the treatment groups, but not significantly different ($P < 0.05$) with 0.5% LM treatment.

The third experiment was conducted using drinking water to study growth performance, apparent digestibility, meat and blood plasma total cholesterol in 6 weeks broiler chicken. Compared with metabolites in feed, administering metabolites in drinking water is easy to handle and not sensitive to environment temperature especially in the

farm conditions. A total of 432 male Cobb broiler chickens were divided into 6 groups based on metabolites level in drinking water: four levels of liquid metabolites mixtures (0.25%, 0.50%, 0.75% and 1.00%) and 2 controls (negative control and positive control). Improved growth performance, increased feed apparent digestibility and lowered meat and blood plasma total cholesterol concentrations were found in the treatments supplemented different levels of metabolite mixture. Furthermore, similar or better effect in enhancing chicken growth performance of metabolites compared to antibiotic growth promotant was observed in this study. The chickens supplemented with LM in drinking water had no significantly different with each other. The chickens supplemented with 0.25% LM had the best results among the treatment groups. LM combinations of 4 strains RS5, RI11, RG11, and RG14 at level of 0.25% can be used in drinking water as replacement of antibiotic in poultry production.

In conclusion, mixture of four strains RG11, RG14, RI11 and RS5 from *Lactobacillus plantarum* has synergistic effects against the indicator strain *P. acidilactici*; feeding this metabolite mixture in diets or drinking water has beneficial effects on growth performance, faecal microflora, small intestine villi height, apparent digestibility, meat and blood plasma cholesterol in broiler chickens. It implies liquid metabolites produced by RG14, RG11, RS5, and RI11 of locally isolated *L. plantarum* are potential alternatives to antibiotics that could be used as growth promoter in broiler chickens.

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

KESAN CECAIR METABOLIT DARI *LACTOBACILLUS PLANTARUM*
TERHADAP KESIHATAN USUS, KOLESTEROL PERINGKAT DAN PRESTASI
TUMBESARAN AYAM PEDAGING

Oleh

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Penggunaan antibiotic sebagai penggalak tumbesaran tidak dapat diterima. Cecair metabolik mempunyai masa depanyay cerah sebagai pengganti antibiotik disebabkan oleh ciri-ciri anti-bakteria semulajadi. Tiga eksperimen telah dijalankan untuk mengkaji kesan pemberian makanan campuran metabolik cecair (LM) dari 4 strain *Lactobacillus plantarum* terhadap prestasi pertumbuhan ayam pedaging.

Ciri-ciri antibakteria metabolik cecair yang dihasilkan oleh RG14, RG11, RS5, dan RI11 dari pemencilan *L. plantarum* tempatan dan dibandingkan dengan strain *P. acidilactici* telah dikaji dalam eksperimen pertama. pH, aktiviti bakteriosin, diameter zon jelas dan

kiraan ‘viable plate’ dari 4 strain *L. plantarum* iaitu RG11, RG14, RI11, dan RS5 serta kombinasi mereka telah dikenalpasti. Kajian ini menunjukkan bahawa metabolik yang dihasilkan oleh strain *L. plantarum* mempunyai keupayaan-keupayaan yang berbeza dalam perencutan patogen; terutamanya gabungan-gabungan metabolit yang memperkenalkan aktiviti bakteriosin dan diameter pusat zon jelas yang lebih kukuh berbanding dengan strain tunggal RS5, RG14, RI11, dan RG11. Metabolit-metabolit ini mempunyai kesan sinergistik yang boleh diaplikasikan sebagai biopreservasi dalam industri makanan serta bahan aditif makanan haiwan.

Dalam eksperimen kedua, gabungan-gabungan metabolit cecair yang dihasilkan oleh 4 strain *L. plantarum* telah digunakan dalam makanan sebagai pengganti antibiotik dalam ayam pedaging. Sejumlah 294 ayam pedaging Cobb jantan telah dibahagikan kepada 7 rawatan dengan diet yang berbeza: 5 kepekatan metabolik cecair (LM) (0.5%, 1.0%, 1.5%, 2.0%, dan 2.5%) dan 2 kawalan (kawalan negatif dan kawalan positif). Peningkatan prestasi pertumbuhan, pengurangan *Enterobacteriaceae* dan penambahan LAB serta penambahan ketinggian vilus usus kecil telah ditemui dalam rawatan yang dibekalkan tahap gabungan metabolit yang berlainan. Ayam-ayam dibekalkan dengan 1.0% LM merupakan antara kumpulan-kumpulan rawatan terbaik, tetapi perbezaanya tidak ketara ($P>0.05$) dibandingkan dengan 0.5% rawatan LM.

Eksperimen ketiga dijalankan dalam air minuman untuk mengkaji prestasi pertumbuhan ayam pedaging, kebolehcernaan ketara, daging dan jumlah kolesterol dalam daging dan plasma keatas ayam pedaging berumu 6 minggu. Berbanding dengan metabolik dalam makanan, penambahan metabolik dalam air minuman lebih mudah dikendalikan dan

tidak sensitif pada suhu persekitaran terutama sekali dalam keadaan ladang. Sejumlah 432 ayam pedaging Cobb jantan telah dibahagikan kepada 6 kumpulan berdasarkan kepada kepekatan metabolik dalam air minuman: 4 tahap gabungan metabolik cecair (0.25%, 0.50%, 0.75% dan 1.00%) dan 2 kawalan (kawalan negatif dan kawalan positif). Prestasi pertumbuhan yang lebih baik, penambahan kebolehcernaan ketara makanan dan perendahan kolesterol daging dan plasma kolesterol dalam ayam pedaging telah ditemui dalam rawatan yang dibekalkan dengan kepekatan gabungan metabolit yang berlainan. Tambahan pula, kesan setara atau lebih baik dalam mempertingkatkan prestasi pertumbuhan ayam yang debekalkan metabolik mempunyai berbanding dengan penggalak tumbesaran antibiotik dalam kajian ini. Ayam-ayam dibekalkan dengan LM dalam air minuman tidak mempunyai perbezaan yang nyata antara satu sama lain ($P<0.05$). Ayam-ayam dibekalkan dengan 0.25% LM menunjukkan keputusan terbaik berbanding dengan kumpulan-kumpulan rawatan. Gabungan-gabungan LM 4 strain RS5, RI11, RG11, dan RG14 pada tahap 0.25% boleh digunakan dalam air minuman sebagai pengganti antibiotik dalam pengeluaran ternakan ayam.

Kesimpulannya, kombinasi 4 strain RG11, RG14, RI11 dan RS5 dari *L. plantarum* mempunyai kesan sinergistik berlawan terhadap petunjuk *P. acidilactici*; penambahan kombinasi metabolit ini dalam diet serta air minuman mempunyai kesan manfaat dalam prestasi pertumbuhan, mikroflora dalam tahi, usus kecil, ketinggian villi, daya pencernaan ketara, kolesterol daging dan plasma dalam ayam pedaging. Ini menunjukkan cecair metabolit yang dihasilkan oleh RG14, RG11, RI11 dan RS5 dari pemencilan *L. Plantarum* mempunyai potensi sebagai alternatif untuk antibiotik dan boleh digunakan sebagai penggalak tumbesaran ayam pedaging.

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I certify that a Thesis Examination Committee has met on July 27, 2011 to conduct the final examination of Cheng Xiang Feng on his thesis entitled "Effects of liquid metabolites from Lactobacillus Plantarum on gastrointestinal health, Cholestrol Level and growth performance of broiler chickens" 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 degree of Science.

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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 concurrently, submitted for any other degree at Universiti Putra Malaysia or other institutions.

CHENG XIANG FENG

Date: 27 July 2011

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