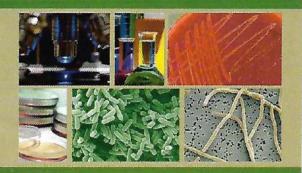
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# Program



# Seminar

# Lactic Acid Bacteria & Culture Collection

Their Role in Food, Health, and Industry and the Important of Management of Culture Collection

Organized by ISLAB, PSPG UGM, FORKOMIKRO In collaboration with PATPI Yogyakarta, UGM, IDAI, PERMI

Organized by



ndonesian Society of Lactic Acid Bactera Pangan dan Gizi



Forum Komunikasi Kurator Koleksi Mikroorganisme Indonesia

In collaboration with









16-17 Januar 2009

# Program



# Seminar

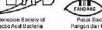
# Lactic Acid Bacteria & Culture Collection

Their Role in Food, Health, and Industry and the Important of Management of Culture Collection

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In collaboration with









# **MITRA**

- •PT YAKULT INDONESIA
- PT DIPA PUSPA LABSAINS
  - PT NESTLE INDONESIA
  - PT ELO KARSA UTAMA

Seminar Lactic Acid Bacteria and Culture Collection: Their Role in Food, Health, Industry and the Important of Management of Culture Collection. Yogyakarta January 16-17th, 2009. Faculty

of Agricultural Technology, Gadjah Mada University, Yogyakarta, Indonesia

Total 40



#### SEMINAR

"Lactic Acid Bacteria & Culture Collections: Their Role in Food, Health, Industry and the important of management of culture collection"

> 16 - 17 January 2009 Auditorium Faculty of Agricultural Technology Gadjah Mada University Yogyakarta, Indonesia

# Organized by:

ISLAB (Indonesian Society for Microbiology)
FORKOMIKRO (Communication Forum of Indonesian Culture Collection
Curators)
Center for Food and Nutrition Studies, GMU
Department of Food and Agricultural Product Technology (DFAPT),
Faculty of Agricultural Technology Gadjah Mada University

In Collaboration with:
PERMI (Indonesian Society for Microbiology)
PATPI (Indonesia Food Technologist Association) branch Yogyakarta
IDAI (Indonesian Pediatric Association) branch Yogyakarta

#### INTRODUCTION

Indonesia as a mega-diversity country has diverse microorganisms, including lactic acid bacteria. These bacteria which have varied physiological functions have been isolated and investigated associated with human life. The utilizations of lactic acid bacteria expand into many areas of food, health, and industries. Lactic acid bacteria play many roles in traditional Indonesian fermented foods such as *tape*, *kecap*, and *asinan*. Many species and strains of lactic acid bacteria have been suggested to have many beneficial effects on the health of the digestive tract of humans. Many strains of lactic acid bacteria have been applied into probiotic products. Administration of specific strains of lactobacilli and/or bifidobacteria was found to be effective in the treatment/prevention of rotavirus, antibiotic-associated, and pathogenic diarrhea. The ability of specific probiotics to enhance immune function in infant has also been reported.

Many researches have been carried related to the development of science and technology in microbiological area. To support the preservation of potential microorganisms, culture collection should be managed in a good management system. Therefore, it is necessary to disseminate these research findings and experiences as well as how to manage culture collection among researcher, pediatrician, students, industries and other stake holders.

Objectives of this seminar and workshop are: (1) To up date information related to the potency of indigenous microorganisms in food, health and industry; (2) To disseminate research in microorganisms, particularly lactic acid bacteria and other bacteria; (3) "case study" related to diarrhea and probiotic treatment; (4) To improve management of Culture Collection in Indonesia

This seminar is organized by ISLAB (Indonesian Society for Lactic Acid Bacteria), FORKOMIKRO (Communication Forum of Indonesian Culture Collection Curators), Center for Food and Nutrition Studies - , and Department of Food and Agricultural Product Technology (DFAPT), Faculty Agricultural Technology - Gadjah Mada University; in collaboration with IDAI (Indonesian Pediatric Society) branch Yogyakarta, PATPI (Indonesian Society for Food Technologist) branch Yogyakarta and PERMI (Indonesian Society for Microbiology)

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#### Preface

Many researchers in Indonesia have been isolated and characterized various kind of indigenous microbes from the nature. Many of them are recognized as industrially useful and also have significant role in food and health. The management of microbes preservation is important in order to maintain its sustainability for exploration their potential properties.

Lactic acid bacteria has historically linked with food fermentation, bacteriocins produced by several strains produced additional hurdle for spoilage and pathogenic microbes. Many species and strains of lactic acid bacteria suggested to have many beneficial effects on the health of the digestive tract of humans and animals, and had been applied for probiotics. It has been said that they have many functions as potential theurapetic properties including reduction of cholesterol, reduction in constipation, anti inflammatory and anti cancer activities.

The seminar on Lactic Acid Bacteria and Culture Collection: Their Role in Food, Health, Industry and The Important Management of Culture Collection is organized to provide opportunity for researchers, food and pharmaceutical manufacturer, pediatricians, culture collection curators, students to interact and communicate for the development of science and technology in microbiological area which have positive impact on the utilization indigenous microbes and culture collection management. In this seminar, 8 presentations will be delivered by guest speakers, while the technical session consists of 30 oral and 15 poster presentations.

On behalf of the organizing committee I would like to express my sincere thanks to Center for Food and Nutrition Studies Gadjah Mada University, Faculty Agricultural Technology Gadjah Mada University, ISLAB (Indonesian Society for Lactic Acid Bacteria), FORKOMIKRO (Communication Forum of Indonesian Culture Collection Curators) and IDAI (Indonesian Pediatric Society) branch Yogyakarta, PERMI (Indonesian Microbiological Society), PATPI (Indonesian Society for Food and Technologist), branch Yogyakarta and all sponsoring company for their support leading to the success of the whole seminar activities. I also would like to thank to all presenters and participant for their tremendous effort and time spent in the seminar.

Organizing Committee Chairman

Dian Anggraini Suroto

#### Preface Chairman of ISLAB

The Indonesian Society for Lactic Acid Bacteria (ISLAB) was established on March 12, 2003 after the establishment of Asian Federation of Society for Lactic Acid Bacteria (AFSLAB) in November 2002 in Tokyo, Japan. Currently, more than 100 Indonesian Scientists are registered as the member of ISLAB and actively conducting many researches in this area. ISLAB is a member of AFSLAB which is currently planning the 5<sup>th</sup> Asian Conference for Lactic Acid Bacteria on July 2 - 3, 2009 in Singapore.

After the 3<sup>rd</sup> Asian Conference for Lactic Acid Bacteria was held on August 25 - 26, 2005, ISLAB has no scientific meeting, nevertheless ISLAB as a member of Indonesian Society for Microbiology (PERMI) supports every annual meeting organized by PERMI. During the last PERMI meeting in Purwokerto in August 2008, the committee of PERMI, ISLAB and FORKOMIKRO had decided to arrange seminar focused on the area of lactic acid bacteria and culture collection. Therefore on January 16 - 17, 2009 we organized this seminar in collaboration with several societies, i.e., IDAI cabang Yogyakarta as well as PATPI cabang Yogyakarta and also the Center for Food and Nutrition Studies and the Faculty of Agricultural Technology, Gadjah Mada University.

During this seminar, we have international guest speakers, committee of AFSLAB, i.e. Prof. Fusao Tomita, as previous Chairman of AFSLAB, Dr. Lee Yuan-Kun, Vice President of AFSLAB, and Dr. Yoshimi Benno, as Head of RIKEN BRC-JCM, Japan. While, Chairman of AFSLAB, Prof. Tsai Ying-Chieh, who planned to attend this event, unfortunately he could not make it, and sends regard to us.

Today, the seminar is very special since the participants are not only come from universities and research institutes, but some of them come from industries. Form this seminar, we hope that we can share experience, expertise, and establish collaboration with universities, research institutes, and industries.

I wish, in the future, ISLAB is becoming solid and stronger as society in lactic acid bacteria, and more scientists performing research in this area, and we could play role and actively participated in international scientific meeting.

Finally, I would like to express my sincere appreciation to all participants for their contributions; I wish all of you have a fruitful time during the seminar.

Chairman of ISLAB
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#### PROGRAM

3.00-09.00	Registration / Coffee break				
9.00-09.30	Opening Ceremony Welcome Speech Chairman of Organizing Committee Dean of Faculty Agricultural Technology, Gadjah Mada University Chairman of Indonesian Society of Microbiology				
9.30-10.00	Prof. Dr. Amin Subandrio / Ministry of Research and Technology -     Development of research, and technology in Indonesia				
0.00-10.30	2 Dr. M. Juffrie/UGM · Effect of problotic to the diarrhea patients				
0.30-11.00	Prof. Dr. Yuan Kun Lee / National University of Singapore -     Gastrointestinal commensals modulate immunity and energy metabolism     of the host				
1.00-11.30	PT YAKULT INDONESIA da	IN PT NESTLE INDONESIA			
1.30-13.00	A CORPORATION AND AND AND AND AND AND AND AND AND AN	LUNCH BREAK			
3.00-14.00		POSTER PRESENTATION			
4.00-17.00	Room A: Technical Session	Room B: Technical Session	Room C: Case Study		
	-Speaker: 4. Dr. I Nengah Sujaya (Udayana) - Indonesian indigenous lactic acid bacteria	-Speaker: 5, Dr. Wellyzar Sjamsuridzal (UI)- Culture collection in Indonesia	-Speaker: 6. Dr. dr. Sumadiopo SpAK (UGM) - Effect of problotic to allergic and immune suppressive patient		
	Paper presentation A1-1 to A1-5	Paper presentation B1-1 to B1-5			
	Paper presentation A2-1 to A2-5	Paper presentation B2-1 to B2-5	/Conceled.		
8.00-22.00	A SECTION AND THE PROPERTY OF THE PARTY OF T	WELCOME DINNER			
ATURDAY, 17	7 January 2009				
	Room A: Technical Session	Room B: Technical Session	Room C: Lab Work		
08.00-10.00	- Speaker: 7. Dr. Achmad Dinoto (LIPI)- Population dynamic of gastrointestinal microbes in carcinogen-treated animal	aker: Achmad Dinoto Population Intestinal Speaker: 8. Dr. Agus Wijaya (UNSRI)- Enterococci between pathogen and probiotics probiotics Speaker: 9. Prof. Dr. Sutriswati F Lactic aci foods PT ELO KAF			
	Paper presentation A3-1 to A3-5	Paper presentation B3-1 to B3-5			
10.30-11.00	PT DIPA PUSPA LABSAINS				
11.00-11.30	10. Dr. Yoshimi Benno / RIKEN-JCM, Japan - Quality control of lactic acid				
11.30-12.00	11 Prof. Dr. Yati Spenarto /UGM - Rotavirus diarhea in Indonesia				
	12. Prof. Dr. Fusao Tomita /Japan- Future trend probiotic products				
11.30-12.00	11. Prof. Dr. Yati Soenar	to /UGM - Rotavirus diarh	ea in Indonesia		

# Technical Session Room A: R 102 (Friday 14.00 - 17.00)

Time and moderator	Note	Speaker	Title
14.00 - 15.45 (Rindit	S-4	Dr. I Nengah Sujaya (Udayana)	Indonesian indigenous lactic acid
Pambayun)	A1-1	K. A. Nocianitri	Isolation and Characterization of Soy Milk Fermenting Lactic Acid Bacteria from Infant Feces
	A1-2	Nyoman Semadi Antara 2	Characterization of Lactic Acid Bacteria Isolated from Fermented Horse Milk of Bima
	A1-3	Ingrid S Surono	Scientific Evidence of Local Probiotic L. plantarum and E. faecium strains Isolated from Dadih
	A1-4	Sri Harimurti 4	Effect of Probiotic Supplementation on the Intestinal Structures of Broiler Chickens
	A1-5	Dewi Ratna Nurhayati	The Effect of Fermented Milk Metabolites in Profile Lipid on Sprague Dawley with Standard Diet
15.45 - 17.00 (I Nengah Sujaya)	A2-1	Agustin Krisna Wardani	Comparison of Lactate Reduction Strategies for Enhancement of Nisin Production by Lactococcus lactis
	A2-2	Ngatirah 7	Karakteristik Biokapsul Lactobacillus Dad 13 Dengan Berbagai Jenis dan Konsentrasi Bahan Pengkapsul
	A2-3	Rita Khairina ()	Potensi Tape Biji Teratai (Nyhmpahea sp.) Sebagai Makanan Fungsional
	A2-4	Yoyok Budi Pramono	Fermentasi Terkendali Petis Daging dengan Kultur Starter Pediococcus acidilacticii YDA3 dan Pediococcus pentosus YDA4
	A2-5	Rindit Pambayun	Sensitivity of Gram-Possitive Bacteria Toward Catechin Extracted from Gambier (Uncaria gambir Roxb)

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## Technical Session Room B: R 105 (Friday 14.00 - 17.00)

Time and moderator	Note	Speaker	Title
14.00 - 15.45 (Nanik Suhartatik)	S-5	Dr. Wellyzar Sjamsuridzal (UI)	Culture collection in Indonesia
	B1-1	Lanjar Sumarno	Growth Characteristic of Lactic Acid Bacteria Isolated from Fermented Fruit Juice of Noni (Morinda citrifolia) in Soymilk
	B1-2	Enny Karti B.S.	Peningkatan Kualitas Yoghurt Beku Susu Kacang Merah (Kajian Senyawa <i>Cryoprotectant</i> dan Suhu Pembekuan)
	B1-3	Leni Herliani Afrianti 13	Pengaruh Perbandingan Kacang Hijau dengan Air Serta Konsentrasi Starter Yoghurt Terhadap Karakteristik Yoghurt Kacang Hijau (Vigna radiata)
	B1-4	Yeyen P. Wanita	Physicochemical Characteristic o Soy and Peanut Yoghurt
	B1-5	Siti Nur Jannah	Exploiting Tofu Liquid Waste Become Healthy Food by Lactic Acid Bacteria and Determination of Its Cholesterol Levels <i>In Vitro</i>
15.45 - 17.00 (Wellyzar Sjamsuridzal)	B2-1	Rosida 16	Pengaruh Konsentrasi Starter Lactobacillus plantarum dan Lama Fermentasi Pada Kualitas dan Kerusakan Terasi
	82-2	Sri Sumarsih	Organoleptic and Chemical Quality of Rucah Fish Fermentation with Lactic Acid Bacteria as Starter
	B2-3	Nurhayati 10	Biodegradasi Limbah Cair Pabrik Tepung Tapioka Menggunakan Ragi Tape
	B2-4	B. Sulistiyanto	Effects of Filler and Propionic Acid on Microbial Performance of Processed Food Waste in the Different of Storage Time
	B2-5	Eni Suryani 20	Penggunaan Effective Microorganism (EM4) Terhadap Tanaman Cabai Merah Hibrida Varietas ietas Hot beauty di Lahan Marginal

# Technical Session Room A: R 102 (Saturday 08.00 - 10.00)

Time and moderator	Note	Speaker	Title
Saturday 08.00 - 10.00 (Lanjar Sumarno)	5-7	Dr. Achmad Dinoto (LIPI)-	Population dynamic of gastrointestinal microbes in carcinogen-treated animal
	A3-1	Ema Damayanti	Antimicrobial Activity of Lactic Acid Bacteria Isolated from Fruit Juices of Ginseng (Panax sp.)
	A3-2	Ahmad Sofyan	Response of Lactic Acid Bacteria Growth on the Media Containing Earth Worm Meals (Lumbricus rubellus) as Feed Additive
	A3-3	Tyas Utami 23	Influence of Bile on Lactobacilli Viability and Ability to Reduce Lactose in MRSL Broth
	A3-4	Basuni Hamzah	A Study of Lactic Acid Bacteria Agglutination during Manufacture of Cottage Cheese
	A3-5	Sri Luwihana	Perubahan Kimiawi pada Air Kelapa Pascabuka

## Technical Session Room B: R 105 (Saturday 08.00 - 10.00)

Time and moderator	Note	Speaker	Title
Saturday 08.00 - 10.00 (Agustin Krisna Wardani)	5-8	8. Dr. Agus Wijaya (UNSRI)	Enterococci between pathogen and probiotics
	B3-1	Harsojo	Dekontaminasi Bakteri Salmonella Pada Beberapa Makanan Olahan yang Berasal dari Daging Ayam dengan Iradiasi Gamma
	B3-2	Murtiari Eva	Pengaruh Tingkat Pengenceran Asap Cair dan Jenis Ikan Terhadap Karakteristik Mikrobiologis Fillet Ikan Asap
	B3-3	Noviladelfia Dian Estetika	Kualitas Kecap Kacang Merah (Phaseolus vulgaris)
	B3-4	Ixa Fibriastuti	Extraction Methods Variance Roselle (Hibiscus sabdariffa Linn) on Antioxidant Activity Combucha
	B3-5	Nanik Suhartatik	Roselle (Hibiscus sabdariffa Linn) Kombucha and Its Capability as Antihypercholesterolemia

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**S-2** 

#### Effect of Probiotic to the Diarrhea Patients

M. Juffrie Faculty of Medicine, Gadjah Mada University Yogyakarta, Indonesia

#### ABSTRACT

Probiotic is live microorganism that is beneficial for human beings and animals. There are 3 genera of Lactic acid bacteria which are often known as probiotic such as Lactobacillus, Bifidobacterium and Streptococcus. Probiotics is a normal flora found in digestive system; it plays a very important role for the gastrointestinal immunity.

The important role of probiotic in association with Diarrhea has been widely known. Some specific strains, such as *Lactobacillus GG*, *L. reuteri*, *Saccharomyces baulardii*, Bifidobacteria has significantly been recognized to solve diarrhea problems within the child.

An in-vivo supplementation of milk containing the probiotic *Lactobacillus casei* DN-114 001 strain is able to reduce the clinical symptoms of diarrhea. Furthermore; probiotic is also able to lower hystopathological changes caused by rota virus.

In children, it seems that probiotics have a more specific role in diarrhea cases caused by viral infections. This may be due to the increase of IgA secretion, and the probiotics capability to stop viral spreading within the intestinal.

Episodes of acute diarrhea can be caused by viral, bacterial infections or by parasite infestation. Many studies report evidence of clinical improvement due to probiotics in diarrhea cases caused by rota virus or in cases of traveler's diarrhea, a diarrhea caused by *Clostridium difficile*.

In conclusion; probiotics holds an important role in diarrhea treatment.

# **S-4**

#### Gastrointestinal commensals modulate immunity and energy metabolism of the host

Yuan-Kun Lee
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The post embryonic development of the gastrointestinal tract is subjected to regulation by colonizing microbiota. This maturation process requires the commensal bacteria to crosstalk with host cells by way of recognizing receptors and inducing signaling pathways to activate transcriptional factors. The influence of gastrointestinal microbiota may extend beyond the gut, modifying systemic immunity and physiology. Studies using intestinal epithelial cell (IEC) lines have suggested that commensal lactic acid bacteria may "program" IECs to down-regulate pro-inflammatory responses by interfering with cytokines and Toll-like Receptors (TLRs) expression. At the same time, the expression of innate immunity genes is up-regulated in preparation of the invasion of pathogens.

RNA and Protein assay on immunology signaling pathways showed that c-Jun N-Terminal Kinase (JNK), p-38 and Peroxisome Proliferator-activated Receptor-gamma one (PPAP $\gamma$ 1) are involved in the immuno-regulatory effects of intestinal lactic acid bacteria. The signaling pathways are tightly regulated suggests that microbiota-driven regulation might be important for a number of homeostatic strategies in the gut.

PPAPy was shown to regulate metabolic target genes, including Adipose Differentiation-related Protein (ADRP) and Fasting-induced Adipose Factor (FIAF), both of these gene products modulate lipid accumulation in cells, linking lipid metabolism to control of inflammation in epithelial cells, and possibly obesity.

Given that pathogenic bacteria have various means to subvert host cell responses, it is conceivable that commensals and non-pathogenic microbiota may possess mechanisms to regain homeostatic balance. It follows that if small changes of the functional status of microbiota could regulate potent factors such as nuclear receptors, one might envisage a targeted way of improving colonic ailments.

The roles of the intestinal microbiota may open avenues for modulation of intestinal homeostasis in both health and disease conditions.

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# Indonesian Indigenous Lactic Acid Bacteria: Their Potential Application and Conservation for Sustainable Usage

1 N. Sujaya<sup>1,21</sup>, N.P. Desy Aryantini<sup>1</sup>, N.W. Nursini<sup>2</sup>, K. A. Nocianitri<sup>3</sup>, Y. Ramona<sup>4</sup>, DM. Sukrama<sup>5</sup>, K. Asano<sup>6</sup>, F. Tomita<sup>7</sup>,

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#### ABSTRACT

Lactic acid bacteria (LAB) are often associated with traditional fermented foods and a part of normal intestinal microbiota. Long history consumption of LAB derived foods revealed that the bacteria are important in human nutrition and health and considered as safe microorganisms, therefore LAB are of main target for modulating of human health through consumption of live cells (probiotic). It is thought that activity/efficacy of each probiotic is strain specific and hence observation made in one probiotic strain could not be extrapolated to other strains. This imply that validation is requires for each specific strain independently against given condition such as consumers. These provide opportunities to initiate the study on potential usage of indigenous LAB strains from Indonesian diversities.

Studies have been initiated and aimed to isolate and characterize indigenous LAB from Indonesian resources such as fermented foods and human gastro intestinal tract (GI). Several lactobacilli and bifidobacteria strains have been characterized based upon their probiotic properties. Two groups of lactobacilli isolated from traditional fermented foods and GI showed different properties, which was influenced by their sources. Intestinal origin strains showed better adaptation into bile stress and GI condition than lactobacilli isolated from fermented foods. In addition, the intestinal LAB was more adhesive than non intestinal LAB verified on in vitro studies using enterocytes of mouse. This property might be important for competitive exclusion of pathogen associated with foods, which often causing foodborne diseases in Indonesia. Some isolates showed their potency for plant based application as well as dairy product. These initial studies recommend that the Indonesian indigenous LAB is worthy to explore their industrial application and need to be conserved for sustainable usage.

### Population Dynamic of Gastrointestinal Microbes in Carcinogen-Treated Animal

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#### ABSTRACT

Eco-physiology changes in animal gastrointestinal tracts due to carcinogen treatment were observed in this study. A notorious food carcinogen, benzo[a]pyrene (BaP), was applied to mice in order to investigate the impacts of carcinogen on gastrointestinal ecosystems. Mice treated with 0.2 pmol BaP at day 1, 7, and 14 after mice birth were maintained carefully and sacrificed after seven weeks-old. The cecum performances and cecal microbial community were then observed. As the results, BaP treatment reduced the cecal content weight up to about 1.5 fold as compared to normal mice. The pH of cecal content in carcinogen-treated mice was observed to be more alkali (pH 8.3) than that of in normal mice (pH 7.2). In addition, the population of Lactobacillus spp. as detected by fluorescence in situ hybridization (FISH) was reduced four times lower by Bap. The characterization of microbial communities by terminal restriction fragment length polymorphisms (T-RFLP) is still on going to obtain more comprehensive results of population dynamic in mouse gastrointestinal tracts as the response to carcinogen. Since BaP was applied directly to mouse by intravenous injection, the changes of microbes in gastrointestinal tracts seem to be related to physiology changes of intestinal organs which is interfered by BaP. Although the mechanisms of the eco-physiology changes in mouse intestine have not yet been clarified at the moment, these results at least show the phenomenon of close interaction between macro- and microorganism in gastrointestinal tracts.

Key words: Lactobacillus, gastrointestinal tracts, mouse, food carcinogen, benzo[a]pyrene

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**S-8** 

# The enterococci: between pathogen and probiotics

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#### ABSTRACT

Enterococci belong to the clostridial subdivision of the Gram-negative bacteria, together with other genera of lactic acid bacteria (Devriese and Pot, 1995). Enterococci constitute an important part of the autochthonous bacteria associated with the mammalian gastrointestinal tract. Over the last two decades enterococci, especially strains belonging to Enterococcus faecalis and Enterococcus faecium, have emerged as important nosocomial pathogens in immunocompromised patients and intensive care units (Adams, 1999). On the other side, enterococci play important role in food fermentation. Specific strains of enterococci are being used as probiotics for humans and animals. One of the best studied enterococci used as a human probiotic, especially in the treatment of diarrhea, is Enterococcus faecium SF68. Another probiotic Enterococcus is the Causido® culture that consists of two strains of Streptococcus thermophilus and one strain of Enterococcus faecium. However, the use of enterococci as probiotics remains a controversial issue. While the probiotic benefits of some strains are well established, the mergence of antibiotic resistant strains of enterococci and the increased association of enterococci with human disease has raised concern regarding their use as probiotics. Therefore, should an Enterococcus strain be considered as a human probiotic, it should be determined whether the strain is antibiotic resistant and whether it contains any virulence determinants.

#### Lactic Acid Bacteria in Foods

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Lactic acid bacteria (LAB) are defined as Gram positive, cocci or rod, non-sporing and non-respiring bacteria that produce lactic acid as the major end product during fermentation of carbohydrate. Genera that commonly occur in foods are Lactobacillus, Streptococcus, Pediococcus, and Leuconostoc, however there are several other genera exist, i.e., Enterococcus, Lactococcus, Aerococcus, Carnobacterium, Oenococcus, Tetragenococcus, Vagococcus and Weissella. The correlation of this LAB to foods is due to several reasons, such as most foods support growth of these bacteria, most food-borne LAB are capable of lowering the pH to extend no longer permitting growth of pathogens and spoilage bacteria. Lactic acid bacteria are also a normal microbiota in gastrointestinal tract. Among lactic acid bacteria found in foods, only few (Streptococcus) species are pathogen.

Lactic acid bacteria have been involved in certain food fermentation, including fruits, vegetables, milk, and meat. These fermented foods are relatively safe, usually preserved well because the souring effect of lactic acid and other organic acids that could inhibit the growth of other microorganisms, and have better organoleptic characters such as texture, taste, and flavor.

It has also recognized that LAB are capable of producing inhibitory substances other than organic acids that are antagonistic toward other microorganisms, such as bacteriocin, hydrogen peroxide, diacethyl, and other secondary metabolites.

Cells of LAB have been used as food biopreservatives in many ways such as the use of starter cultures to inhibit the growth of pathogenic and spoilage bacteria in fermentation foods and in refrigerated foods. Metabolites or products of LAB, such as hydrogen peroxide and bacteriocins have also been used as food biopreservatives, commercially.

Consumption of several species of LAB either through dairy product (called food probiotic) or as live cells has been associated with the health benefits in human. These benefits due to the ability of LAB to combat the growth of undesirable bacteria in GI tract, to control the microbiota balance in the intestine, to control lactose intolerance, the stimulate immune system & bowel movement, and to reduce serum cholesterol.

Therefore, it is important to study the occurrence of these bacteria in foods. Aim of the workshop is to study occurrence lactic acid bacteria and their role in foods, and will be focused on:

- 1. Enumeration lactic acid bacteria in foods
- 2. Isolation and identification of lactic acid bacteria
- 3. Detection of bacteriocin producer

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S-10

### Quality Control of Lactic Acid Bacteria in RIKEN BRC- Japan Collection of Microorganisms

Yoshimi BENNO, DVM, PhD Head of RIKEN BRC-JCM

The Japan Collection of Microorganisms (JCM) in the RIKEN Bio-Resource Center has contributed to bioscience and biotechnology as one of the leading Culture Collections in the world since 1981. A wide range of microbes relevant to human health and environment are collected, preserved and distributed using new technologies and by collaboration with researchers and the Culture Collections. Efforts have been also made to search the new microbes and to develop technologies for handling extremophiles and uncultured microbes, screening systems for valuable microbes and culture-independent analysis of the microbes. A total of preserved strains are 18,500 strains. Approximately 3,200 strains were annually distributed to the Universities and Research Institutes in Japanese and overseas. Type strains and reference strains of lactic acid bacteria, anaerobic bacteria, aerobic bacteria, streptomyces, archea, extremphiles, yeasts and fungi for human health and environmental research are collected to BRC-JCM. On a total of 377 Lactobacillus JCM strains, 104 species (82% of total Lactobacillus species proposed) and 275 reference strains are distributed from BRC-JCM. All the reference strains were reclassified using 16S rDNA sequence for the quality management. BRC-JCM acquired the ISO9001.2000 certification for the quality control of microbial strains on August 2007. The BRC-JCM activities include training course, the homepage renewals, publications of mail-news, the JCM newsletter and JCM catalogue. By these activities, microbial researches in Japan and overseas will be developed and be aligned our international initiatives with share and relations to the Asian Microbial Culture Collections.

#### **BRC-JCM** activities

- 1) Collection, preservation and distribution of the microbial strain(s)
- Deposition of new microorganisms and issuance of BRC-JCM certification for deposition and availability of the type strain to new taxon proposed
- Freezing and freeze-drying preservation of microbial strains for the safety purposes
- 4) Quality control of the microbial resources
- a. Confirmation of purity and characteristics of deposited strains
- b. Genetic monitoring of deposited strains
- Collection and distribution of microbial information related to deposited strains
- Transference of microbial strains is practiced according to the material transfer agreements (MTAs)

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6) The BRC-JCM management is practiced according to the Regulations of Microbial Usage in RIKEN

#### Information services

- 1. Homepage (http://www.jcm.riken.jp).
- 2. Publication of the BRC-JCM mail-news every month
- 3. Publication of the BRC-JCM Newsletter every year.
- 4. Publication of the BRC-JCM Catalogue per 3 years.

#### Specific aim

1) Collection and preservation of the microbial strain(s)

Deposit of new microorganism(s):

To contribute to the domestic, regional, and global improvements in the conservation of microbial resources in cooperation with other culture collections and institutions, BRC-JCM collects, preserves and distribute of authentic microorganisms to researchers in the fields of life sciences and biotechnology as well as of microbial taxonomy. The isolation, phenotypic characterization and phylogenetic analyses of microorganisms from natural sources are carried out to determine microbial biodiversity and to evaluate the currently recognized taxonomic systems and criteria. As of the end of November 2008 a total of 18,500 strains (bacteria, streptomyces, archaea, filamentous fungi, yeasts and 21 others) were held in BRC-JCM.

A total of 9,861 cultures were distributed to local and overseas researchers from 1st April 2005 to 31st March 2008.

BRC-JCM also provides an interface for access to its culture collection by users, as well as for accepting strains for deposit and distributing cultures (JCM homepage and on-line catalogue).

2) Preservation of microbial strains

To preserve and maintain microorganisms safely and correctly, BRC-JCM employs two preservation methods: freezing and freeze-drying. In freezing, a harvested culture is suspended in cryoprotectant, placed in a serum tube, and kept in a deep-freezer (-80°C) or in the vapor phase of liquid nitrogen (ca - 175°C). Fundamentally, ten tubes are prepared for freezing, 6 are kept in a deep freezer, 2 are in liquid nitrogen, 1 is kept in a liquid nitrogen tank in another place to avoid risk of emergency, and the remaining 1 is used for routine check after freezing. Freeze-drying is also a well-known method to microbial preservation. BRC-JCM has devised an available freeze-drying machine with manifold (Japan Patent 59-141735). Since 15 ampoules can be placed in one manifold, 15 ampoules per batch can be prepared under the same condition for one strain. Using our manifold system, 105 ampoules (15 each for 7 strains) can be prepared as our system is equipped after acceleration test at 37°C for 2 weeks, we keep them in a cold room and use them to send out cultures for distribution in answer to requests.

Seminar Lactic Acid Bacteria and Culture Collection: Their Role in Food, Health, Industry and the Important of Management of Culture Collection. Yogyakarta January 16-17<sup>th</sup>, 2009. Faculty of Agricultural Technology, Gadjah Mada University, Yogyakarta, Indonesia

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All data concerning preservation was kept in a database and carefully compared with actual tubes or ampoules when newly stocked or used for distribution.

3) Issuance of the JCM certification for deposition and availability of the type

strain to new taxon proposed

In accordance with the policy of the International Journal of Systematic and Evolutionary Microbiology (IJSEM), authors who wish to propose a new species, a new subspecies or a new combination in IJSEM or to apply for citation of a new name in the Validation List of IJSEM are required to prove the deposition of type strains in culture collections and their availability to the public prior to the acceptance of their manuscript or the citation in a culture collection list. In response to this policy and upon a depositor's request, BRC-JCM will issue a certificate to him/her or one of the authors of a manuscript describing a new taxon after confirming the viability and purity of the deposited culture and the identity of the culture preserved in BRC-JCM.

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### Future trend of probiotic products

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Probiotics was defined as dietary supplements and live microbes containing potentially beneficial bacteria or yeasts. While according to current FAO/WHO definition, probiotics are live microbes which and when administered in adequate amounts confer a health benefit on the host. Most common microbes are Lactic acid bacteria (LAB) that have been used since our human life started about 10,000 or more years ago.

Probiotics means 'for life,' too. The first benefit proved to be beneficial to the host is to keep gastrointestinal microflora being healthy conditions, those are probed to be brought by low pH due to lactic acid fermentation, and prevention of unwanted microbes by competing with them. According to these most common microbes are Lactic acid bacteria (LAB) that have been used since our ancestors have started their settlements in defined areas together with domestic animals such as cow, goat etc.

When the concept of probiotics came into our foods, probiotics were understood as assistants to our body's naturally occurring gut flora (ecology of microbes) to re-establish themselves. Therefore when the concept of FOSHU (food of specific health use) appeared, those foods related to lactic acid fermentation, such as yoghurt, other fermented milk products.

As studies on food chemistry, food microbiology etc progress, understanding of probiotics has progressed in high speed. Now, probiotics for immune systems anti-allergies, intake of excessive alcohol intake, stress, exposure to toxic substances, lowering of cholesterol, lowering of blood pressure and so on has been developed and some of them are already on market.

The diversification of probiotics function has much to do with progresses of science in bacterial genomics and other 'omics'. In the postgenomic era, it becomes possible to study unculturable microbes, and in gut of ours and other animals' numbers of those unculturable microbes are known to be present and their functions are waiting to be uncovered. Namely, studies of microflora and their functions, cross talks among them, microbial resources contributing to human health and environment etc. are major subjects for not only probiotics but also basic microbial science in the future.

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A 1-1

# Isolation and Characterization of Soy Milk Fermenting Lactic Acid Bacteria from Infant Feces

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#### ABSTRACT

This research was aimed to isolate and characterize soy milk fermenting lactic acid bacteria (LAB) from infant feces for developing a soy milk based probiotic for infant who given soy milk as supplement of breast milk due to cow milk allergy.

The LABs were isolated from healthy infant aged one week and one month using modified TOS medium containing soy milk. Twelve out of 89 isolates of soy milk fermenting LABs were selected and characterized based upon their probiotic characters such as production of antimicrobial, resistance to bile. Two promising isolates, F16 and BY-85, were tested their capabilities to pass through gastrointestinal tract using modified gastric juice and their ability to adhere into enterocytes of rat GI.

The results showed that some isolates inhibited pathogenic bacteria through metabolites such acid and it was not due to bacteriocin production. Twelve selected isolates were resistant to bile range from 0.2-0.4 mM. Two promising isolates, F16 and BY-85 were retained their viable cells about 0.1% ( $10^8$ - $10^9$  viable cells from  $10^{11}$  initial cells) after exposed with modified gastric juice .These isolates showed good capabilities to adhere into mouse intestinal enterocytes with adherent cells number were about  $12,0\pm1,89$  and  $14,8\pm1,88$  cells/enterocyte, respectively. Identification performed by sequencing of 16S rDNA showed that the isolate F16 and BY-85 were identified as *Enterococcus avium* F16 dan *Enterococcus avium* BY-85, respectively. Further research need to be conducted to explore the possibility of these isolate to be develop as soy milk based probiotic with specific target to infant who have problem with cow milk and using soy milk as supplement of breast milk.

Keywords: soy milk, probiotic, lactic acid bacteria, infant feces, Enterococcus

A 1-1

### Isolasi dan Karakterisasi Bakteri Asam Laktat yang Dapat Memetabolisme Susu Kedelai Dari Feses Bavi

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#### **ABSTRAK**

Penelitian ini bertujuan untuk mengisolasi dan mengkarakterisasi bakteri asam laktat (BAL) asal feses bayi yang mampu memetabolisme susu kedelai guna pengembangan probiotik untuk individu bayi yang diberikan susu kedelai sebagai pengganti susu ibu karena adanya reaksi alergi terhadap susu formula.

Bakteri asam laktat diisolasi dari bayi sehat umur satu minggu dan satu bulan, dengan medium TOS yang dimodifikasi dengan menambahkan susu kedelai dan MRS. Sebanyak 89 isolat BAL diisolasi dari feses bayi sehat, 6 isolat dari feces bayi umur satu minggu dan 83 isolat dari feces bayi umur satu bulan. Skrining pertumbuhan pada medium TOS dengan susu kedelai diperoleh bahwa sebanyak 12 isolat menunjukkan pertumbuhan vang selanjutnya dikarakterisasi berdasarkan karakter probiotik. Dua isolat. F1-6 dan BY-85, menunjukkan pertumbuhan yang baik pada MRS mengandung 0,2-0,4 mM deoksi kolat, tidak mentransformasi asam kolat menjadi deoksi asam kolat, serta mempunyai ketahanan yang baik pada model kondisi saluran pencernaan dengan jumlah sel yang bertahan hidup sebanyak 108-109 dari 1011 sel awal. Kedua isolat memilki kemampuan adhesi pada sel enterosit mencit secara in vitro dengan jumlah sel yang menempel pada setiap sel enterosit mencit masing-masing 12,0 ± 1,89 dan 14,8 ± 1,88 sel. Sekuen 165 rDNA menunjukkan bahwa kedua isolat memiliki kesamaan susunan basa nukleotida dengan Enterococcus avium F1-6 dan Enterococcus avium BY-85. Kedua strain ini memerlukan penelitian lebih mendalam guna mengekplorasi potensinya sebagai probiotik BAL asal saluran pencernaan lokal Indonesia.

Kata kunci: susu kedelai, bakteri asam laktat, feces bayi, Enterococcus.

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A 1-2

## Characterization of Lactic Acid Bacteria Isolated from Fermented Horse Milk of Bima

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#### ABSTRACT

The aim of the research was to characterize of the six species of lactic acid bacteria (LAB) (Lactobacillus acidophilus, Lb. salivarius, Lb. brevis, Lb. delbrueckii, Lb. plantarum and Lactococcus lactis) isolated from the fermented horse milk of Bima. The observation was done to cover safety aspect, ability to survive on low pH condition, bile salt tolerance, adherence and colonization, anti bacterial activity and effect toward cholesterol level reduction. The six LAB species were not pathogen, not toxic and not invasive and were able to grow well on media which have bile salt at 0.750%. Lb. acidophilus and Lb. brevis have ability to survive on media pH 2.5 for 3 hours and were able to adherence and colonization on gut mucosa epithel. Neutralized supernatant of Lb. acidophilus and Lb. brevis showed anti bacterial activity toward pathogen bacteria (Escherichia coli ATCC8739 and Salmonella enteritidis ATCC 13076). Lb. acidophilus and Lb. brevis have ability to reduce cholesterol level of blood serum of rabbits on hypercholesterolemia condition significantly at 53.74% and 51.70% respectively. Based on the characteristics, two out of the six species. Lb. acidophilus and Lb. brevis, can be used as probiotic.

Key words: lactic acid bacteria, characterization, probiotic, horse milk

A 1-3

# Scientific Evidence of Local Probiotic L. plantarum and E. faecium strains Isolated from Dadih

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#### **ABSTRACT**

Indonesian traditional fermented buffalo milk originated from West Sumatra, dadih has been believed to have a beneficial impact on human health related to the properties of the lactic acid bacteria (LAB) involved in its fermentation process. Adhesion to the intestinal mucosa would allow colonization, although transient, of the human intestinal tract and has been related to the ability to modulate the immune system, especially during its development. In addition, adhesion to and colonization of the mucosal surfaces are possible protective mechanisms against pathogens through competition for binding sites and nutrients or immune modulation. Hence, adhesion is one of the main selection criteria for new probiotic strains. The indigenous dadih LAB strains were Lactobacillus plantarum IS-10506 and IS-20506 and Enterococcus faecium IS-27526, IS-23427, and IS-16183, isolated from dadih fermented milk and were identified by 16S rRNA gene sequencing as L. plantarum (GenBank accession no. DQ860148) and DC860149) and E. faecium (GenBank accession no. EF068251, EF068250, and EF068249). All strains were kept in the University of Turku culture collection. The adhesion of tested LAB strains was strain-dependent and varied from 1.4 to 9.8%. The most adhesive Lactobacillus plantarum strain was IS-10506, with 9.8% adhesion. The competition assay between dadih LAB isolates and pathogens showed that a 2-h pre-incubation with L. plantarum at 37°C significantly reduced pathogen adhesion to mucus. All tested LAB strains displaced and inhibited pathogen adhesion, but the results were strain-specific and dependent on time and pathogen strains. In general, L. plantarum IS-10506 showed the best ability against pathogen adhesion.

Keywords: local probiotics, dadih, L. plantarum, E. faecium, adhesion, competitiveness against pathogens

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A 1-4

# Effect of Probiotic Supplementation on the Intestinal Structures of Broiler Chickens

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#### ABSTRACT

The aim of the study was to investigate the effect of probiotic supplementation on the intestinal structures of broiler chickens. *Cornsoybean rations* antibiotic free were formulated to meet the nutrient requirement of broiler chicken as basal diet for the control treatment. The other treatment groups were fed basal and supplemented with three concentrations of probiotic 10<sup>7</sup>; 10<sup>8</sup> and 10<sup>9</sup> cell/ml/bird per day via oral respectivelly. The probiotic preparation consisted of *Lactobacillus murinus*, *Streptococcus thermophillus*, and *Pediococcus acidilactici*. Probiotic treatments significantly (P<0.05) increased duodenum, jejunum, and ileum villus height, villus width and crypt depth. Suplementation of probiotic improved the intestinal structures of the broiler chickens.

Keywords: probiotic, intestinal structures, broiler chicken

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# Pengaruh Suplementasi Probiotik Terhadap Struktur Usus Ayam Broiler

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#### ABSTRAK

Penelitian ini bertujuan untuk mengetahui pengaruh suplementasi probiotik terhadap struktur usus halus ayam broiler. Ransum ayam berbasis jagung, bungkil kedelai yang bebas dari antibiotik disusun sesuai dengan standar kebutuhan ayam broiler, digunakan sebagai pakan dasar pada perlakuan kontrol. Untuk tiga kelompok perlakuan lainnya juga digunakan pakan dasar tersebut dengan suplementasi probiotik pada konsentrasi 10<sup>7</sup>, 10<sup>8</sup> 10<sup>9</sup> sel bakteri/ml yang diberikan melalui tetes mulut 1 ml per ekor setiap hari. Probiotik yang digunakan dalam penelitian ini terdiri dari Lactobacillus murinus, Streptococcus thermophillus, dan Pediococcus acidilactici. Hasil penelitian menunjukkan bahwa probiotik secara signifikan (P<0,05) meningkatkan tinggi villi, lebar villi, dan kedalaman crypta pada usus halus di segmen duodenum, jejunum, dan ileum. Suplementasi probiotik terbukti memperbaiki struktur usus halus ayam broiler.

Kata kunci : probiotik, struktur usus, ayam broiler

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# A 1-5

## The Effect of Fermented Milk Metabolites in Profile Lipid on Sprague Dawley with Standard Diet

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#### ABSTRACT

Probiotic products such as fermented milk has been widely distributed in the market and proved to have beneficial health effect. Previous research indicated that milk products supplemented with indigenous *Lactobacillus* Dad 13 cells have problems such as reduction of cell viability and organoleptic stability and requirement of low temperature during storage. Therefore it is necessary to study the effect of fermented milk metabolites on cholesterol reducing capability to improve its stability.

The objectives of research were to study the effect of pasteurized and sterilized fermented milk with Lactobacillus Dad 13 on cholesterol lowering capability in vivo. In vivo study was carried out by using 35 male rats 2 months old. The rats were divided into ten groups and fed with standard diet AIN-93 at libitum. Once a week rats were fed with standard diet and to have hypercholesterol diet for a week. At the end of the first week, lipid profile was measured. When experiments started, rats were assigned to standard fed groups, was treated with diet intervention for 4 weeks. During diet intervention period, the rats were given water (placebo), skim milk 10%, sterilized fermented milk (SFM), pasteurized fermented milk (PFM), and commercial fermented milk (CFM). At the end of the treatments, was carried any analysis of lipid profiles.

The *in vitro* results showed that SFM and PFM still have cholesterol binding capability and bile acid deconjugation activity. However their capability reduced by heating, i.e. SFM 10.75 µg/ml, and the ability deconjugation of bile acid is 35.59 µmol/ml. By *in vivo*, pasteurized fermented milk (PFM) and sterilized fermented milk (SFM) reduced total cholesterol level by (38.5 - 43.5%), low density lipoprotein (LDL) by 7.08% - 8.50%, high density lipoprotein (HDL) by 50.04% - 56.00%, and triglyceride (TG) by 31.06% - 34.75%.

Keywords: fermented milk metabolites, LAB, cholesterol, *Lactobacillus* sp Dad 13

## Comparison of Lactate Reduction Strategies for Enhancement of Nisin Production by Lactococcus lactis

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#### ABSTRACT

Chemical food preservatives are generally used to combat microorganism which are the major food-related diseases and spoilage in the production of food. However due to the potential danger of these substances and demand for purer and safer food, i.e. food with less chemical additives, there is a growing interest to replace chemical preservatives by natural preservatives that are easily degradable and harm neither the individual nor the environment. Nisin, a small peptide produced by Lactococcus lactis that exhibits an antimicrobial, should be the promising alternative to meet the consumers' demand. However, there is a problem in nisin production. This is due to the high level of lactate concentration produced by L. lactis lead to the decreasing of pH and inhibition of cell growth. Since nisin production in L. lactis is growth-associated, the inhibition of cell growth will lead to the decreasing of nisin production, resulting low yield of nisin. To overcome this problem, strategy for lactate reduction is applied to avoid the inhibitory effect of lactate on cell growth and nisin production.

In this research, some strategies for lactate reduction are considered via metabolic engineering, hemin addition and via biological means using co-culture with yeast. Metabolic engineering was performed by introduction of alanine dehydrogenase gene (alaD) from B. sphaericus is considered to create a new pathway for alanine synthesis. By this strategy, it is expected that carbon flux from lactate will flow to alanine resulting low production of lactate. Metabolic distribution change is also expected by hemin addition during cultivation. By addition of hemin, L. lactis can undergo respiration due to the hemin activate the cytochrome leads to the high consumption of NADH. As a consequence, the available NADH for LDH reaction decreases, resulting the lactate reduction. Co-culture with yeast is also considered as strategy for lactate reduction. In this strategy, microbial interaction is manipulated to maintain lactate concentration at low level during the process. In this system, maltose is used by L. lactis as carbon source to produce lactate, then lactate is assimilated by yeast. By this mechanism, lactate production is possible to be controlled through out the process.

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A 2-2

# Karakteristik Biokapsul *Lactobacillus* Dad 13 Dengan Berbagai Jenis dan Konsentrasi Bahan Pengkapsul

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#### **ABSTRAK**

Telah dilakukan penelitian mengenai penggunaan berbagai jenis bahan pengkapsul dan variasi konsentrasi bahan dalam pembuatan biokapsul Lactobacillus Dad 13. Penelitian ini bertujuan untuk menyeleksi bahan pengkapsul yang dapat melindungi sel dari pengaruh lingkungan (pH rendah dan garam empedu) sehingga viabilitasnya dapat dipertahankan. Jenis bahan pengkapsul yang digunakan meliputi alginate, kitosan, karagenan dan pectin, sedangkan konsentrasi yang digunakan 3 % dan 3,5%. Penelitian dilakukan dengan menggunakan rancangan blok lengkap teracak factorial dengan jenis bahan pengkapsul sebagai Faktor I dan konsentrasi bahan pengkapsul sebagai Faktor II.

Hasil penelitian menunjukkan bahwa jenis bahan pengkapsul berpengaruh terhadap ketahanan sel pada garam empedu namun tidak berpengaruh pada berat, diameter, jumlah sel dalam kapsul maupun ketahanan pH rendah. Sedangkan konsentrasi bahan pengkapsul tidak berpengaruh pada senua parameter. Berdasarkan jumlah sel, ketahanan pH dan ketahanan terhadap garam empedu, maka didapatkan dua bahan pengkapsul yang terbaik yang mampu mempertahankan viabilitas sel vaitu kitosan dan karagenan.

Kata kunci: biokapsul, Lactobacillus Dad 13, bahan pengkapsul

A 2-3

### Potensi Tape Biji Teratai (Nyhmpahea sp.) Sebagai Makanan Fungsional

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#### ABSTRAK

Penelitian ini bertujuan untuk mengembangkan pemanfaatan biji teratai sebagai bahan dasar pembuatan tape dan mempelajari fungsionaliti tape biji teratai sebagai agensia prebiotik dan atau probiotik. Potensi tape biji teratai sebagai agensia probiotik dilakukan dengan suplementasi tape dengan Lactobacillus acidhophillus SNP-2 dan diamati pada relawan yang mengonsumsi tape biji teratai selama 1 bulan. Hasilnya menunjukkan Total Bakteri Asam Laktat, Lactobacillus dan Enterobacter pada feses relawan yang mengonsumsi tape biji teratai selama pengamatan tidak menunjukkan pengaruh yang nyata. Tidak terdapat perbedaan nilai mikroflora feses relawan sebelum dan sesudah mengonsumsi tape biji teratai yang disuplementasi biomassa. Rata-rata kisaran BAL adalah Log 8,73 CFU/g - Log 10,12 CFU/g, Lactobacillus Log 7.37 CFU/g - Log 10.38 CFU/g dan Enterobacter Log 9.19 CFU/g - Log 10,49 CFU/g. Nilai pH feses relawan selama pengamatan berbeda nyata dengan kisaran berada pada nilai 6.11 - 6.68.

Kata kunci: teratai, tape biji teratai, tape probiotik, Lactobacillus acidophillus,

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A 2-4

# Fermentasi Terkendali Petis Daging dengan Kultur Starter Pediococcus acidilacticii YDA3 dan Pediococcus pentosus YDA4

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#### **ABSTRAK**

Pada fermentasi petis daging yang bersifat spreadable sering terkontaminasi bakteri patogen, pembusuk dan penghasil histamin yang membahayakan kesehatan bagi yang mengkonsumsinya untuk itu perlu tindakan pengendalian. Pada fermentasi petis daging dengan kadar garam 20% (b/b) yang mampu bertahan adalah bakteri halofil. Salah satu bakteri asam laktat yang tahan garam adalah Pediococcus. Untuk itu dalam penelitian ini digunakan Pediococcus acidilacticii YDA3 dan Pediococcus pentosus YDA4, yang secara alami berasal dari mikrobiota fermentasi petis daging. Penelitian ini bertujuan mengungkap peran kedua bakteri asam laktat tersebut sebagai kultur starter pada fermentasi tersebut.

Penelitian ini merupakan penelitian eksplorasi awal ("exploration experimental method") dengan 4 ulangan dan 2 satuan percobaan. Isolat bakteri asam laktat yang digunakan merupakan hasil isolasi penelitian sebelumnya dari fermentasi petis daging tradisional, yaitu Pediococcus acidilacticii YDA3 dan Pediococcus pentosus YDA4. Fermentasi dilakukan secara terkendali dengan menggunakan kedua bakteri asam laktat tersebut sebagai kultur starter.

Hasil penelitian menunjukan bahwa kedua kultur starter mampu menekan kelompok coliform dan bakteri penghasil histamin 1 log cycle; protein terlarut meningkat hingga 10,3%, mampu menekan histamin hingga 5,8 mg/100g; mampu menekan TVN hingga 42,8 mg/100g; mampu menekan TMA hingga 23,2 mg/100g.

Kata kunci:

# Sensitivity of Gram-Possitive Bacteria Toward Catechin Extracted from Gambier (Uncaria gambir Roxb)

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#### ABSTRACT

Research about sensitivity of gram-possitive bacteria toward catechin extracted from gambir product has been conducted. The results showed that inhibition of catechin on the Gram positive bacteria due to the leakage of cell wall, indicated by increasing protein and nucleic acid in the media. The culture of Streptococcus mutans, Staphylococcus aureus, and Bacillus subtilis treated with 0, 60, 120, 180, and 240 ppm of catechin extracts showed that the higher catechin concentration added the higher protein and nucleic acid leakages. Leakage levels of 5. mutans, S. aureus, and B. subtilis were 3.51 and 21.50 %, 3.36 and 18.91 %, and 3.15 and 7.58 %, respectivelly. Furthermore, addition of 8 % (w/v) of the extracts to the media showed that both of 5. mutans and S. aureus decline during one hour, while B. subtillis was still resistant to the catechin. It could be concluded that S. mutans is the most sensitive bacteria to the catechin followed by S. aureus and B. subtilis. These results indicated that binding of catechin to the peptide units of peptidoglycans caused cell wall leakage which could induce the leakage of membrane. Thus, protein and nucleic acid leakage could be used as an indicator of sensitivity of gram-positif bacteria toward catechin extracted from gambier.

Keywords: gram-possitive bacteria, catechin, gambier

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# A 3-1

# Antimicrobial Activity of Lactic Acid Bacteria Isolated from Fruit Juices of Ginseng (Panax sp.)

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#### ABSTRACT

Ginseng roots (Panax sp.) have long been used for medical purposes. Beside of the root, Ginseng had red fruit ripen and its extracts also have health effect. Ginseng fruit juice (GFJ) which kept would be spontaneous fermented. The previous study showed that GFJ had antibacterial activities to Escherichia coli and Staphylococcus aureus, have low pH and contained total lactic acid bacteria (LAB) was 2.7 x 10<sup>5</sup> cfu/ml. The objective of this study was to isolate Lactobacillus strain potential for antimicrobial agent, Isolation was conducted using MRSA medium, followed by incubation at 37° C for 48 hours. Identification covered Gram staining, morphological, catalase and gas production tests. Antimicrobial activity was conducted using diffusion agar method with E. coli, S. aureus and C. albicans as microbial test. The result indicated that Lactobacillus sp. was found in GJF. This strain belongs to hetero-fermentative group. The antimicrobial activity of GFJ showed against S. aureus, E. coli and C. albicans with clearing zone diameters were 11.07, 10.5 and 2.83 mm respectively.

Keywords: antimicrobial activity, ginseng fruit juice, Lactobacillus

## Aktivitas Antimikrobia Bakteri Asam Laktat yang Diisolasi dari Sari Buah Ginseng (Panax sp.)

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#### **ABSTRAK**

Akar ginseng (Panax sp.) lama digunakan untuk tujuan pengobatan. Disamping dimanfaatkan akarnya, Ginseng juga memiliki buah masak berwarna merah dan ekstraknya juga memiliki efek kesehatan. Sari buah Ginseng yang disimpan akan mengalami fermentasi secara alami. Penelitian sebelumnya menunjukkan bahwa sari buah Ginseng memiliki aktivitas antibakteri terhadap Escherichia coli dan Staphylococcus aureus, memiliki pH rendah dan mengandung bakteri asam laktat total sebesar 2,7 x 10<sup>5</sup> cfu/ml. Tujuan penelitian ini adalah adalah untuk mengisolasi strain Lactobacillus yang berpotensi sebagi agensia antimikroba. Isolasi dilakukan dengan menggunakan media seletif MRSA, dilanjutkan dengan inkubasi pada suhu 37° C selama 48 jam. Identifikasi meliputi pewarnaan gram, morfologi, uji katalase dan uji produksi gas. Aktivitas antimikroba dilakukan dengan menggunakan metode difusi agar dengan E. coli, S. aureus dan C. albicans sebagai mikroba uji. Hasil penelitian menunjukkan bahwa Lactobacillus sp. ditemukan dalam sari buah Ginseng. Strain ini termasuk kelompok heterofermentatif. Sari buah ginseng menunjukkan aktivitas antimikroba terhadap S. aureus, E. coli dan C. albicans dengan diameter zona hambat berturut-turut sebesar 11,07, 10,5 dan 2,83 mm.

Kata kunci: aktivitas antimikroba, sari buah ginseng, Lactobacillus

A 3-2

# Response of Lactic Acid Bacteria Growth on the Media Containing Earth Worm Meals (Lumbricus rubellus) as Feed Additive

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#### ABSTRACT

The previous study reported that usage of earth worm meals (Lumbricus rubellus) as feed additive had capable to reduce E. coli and S. aureus growth, however its activity to inhibit lactic acid bacteria (LAB) is still unknown. This experiment was to evaluate lactic acid bacteria growth on the de Man Rogosa Sharpe (MRS) medium containing earth worm meals as feed additive. LAB was isolated from feed silage which made from Kolonjono grass (Pannicum coloratum) and rice bran added by water up to ± 60%. The treatments were addition of earth worm meals (TCT) onto medium at 0, 0.5, 1.0, 1.5 and 2.0%. Culture was incubated during 1 x 24 hours (37°C). Variable identified was total colony of LAB which was counted using spread plate method. Percentage of LAB growth was calculated by deviation of colony divided initial colony (log cfu). Results showed that initial colony around 2.8 -  $6.6 \times 10^{-4}$  cfu / ml, and the highest total colony was obtained from treatment 0.5% TCT. Total colony of LAB observed at 24 hours had increase up to 2 - 40 x 10 8 cfu / ml. The highest increasing colony was obtained from treatment 0.5% TCT with the percentage of growth 99.2% and followed by treatment 0, 2.0, 1.0, and 1.5% with the growth 95.7, 81.6, 77.9 and 77.8% respectively. Addition of TCT more than 0.5% tended to decrease LAB growth. Isolate of LAB was obtained had good stability on the medium contain TCT and potentially used symbiotic as feed additive.

Keywords: lactic acid bacteria, L. rubellus, silage, feed additive

Respon Pertumbuhan Bakteri Asam Laktat dalam Media Mengandung Tepung Cacing Tanah (Lumbricus rubellus) sebagai Aditif Pakan

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#### ABSTRACT

Penggunaan tepung cacing tanah (Lumbricus rubellus) sebagai aditif pakan telah terbukti menghambat beberapa bakteri seperti E. coli dan S. aureus tetapi belum diketahui daya hambatnya terhadap bakteri asam laktat. Penelitian ini bertujuan untuk mengetahui respon pertumbuhan bakteri asam laktat (BAL) dalam media de Man Rogosa Sharpe (MRS) yang diberi tambahan tepung cacing tanah sebagai aditif pakan. BAL diisolasi dari silase yang dibuat dari campuran rumput kolonjono (Pannicum coloratum) dan dedak padi dengan kadar air + 60%. Perlakuan yang diberikan adalah taraf penambahan tepung cacing tanah (TCT) yaitu 0, 0,5, 1, 1,5 dan 2% dalam media tumbuh BAL. Inkubasi dilakukan selama 1 x 24 jam dalam inkubator (37 °C). Peubah yang diamati adalah jumlah koloni BAL yang dihitung dengan metode spread plate. Perhitungan pertumbuhan dikalkulasikan dengan perbandingan selisih jumlah koloni (log cfu) akhir terhadap koloni awal. Total koloni awal berkisar 2.8 - 6.6 x 10 4 cfu / ml dengan jumlah koloni tertinggi pada perlakuan penambahan 0,5% TCT. Jumlah koloni pada pengamatan ke-24 jam terjadi peningkatan dengan jumlah koloni mencapai 2 - 40 x 10 8 cfu / ml. Peningkatan total koloni tertinggi dicapai pada perlakuan 0.5% TCT dengan tingkat pertumbuhan mencapai 99.2% diikuti dengan perlakuan 0, 2, 1, dan 1,5% dengan tingkat pertumbuhan berturut-turut 95,7, 81,6, 77,9 dan 77,8%. Penambahan TCT melebihi 0,5% memiliki kecenderungan menurunkan pertumbuhan BAL. Isolat BAL yang didapatkan dari silase pakan memiliki ketahanan terhadap penambahan TCT sehingga berpotensi dijadikan sebagai simbiotik dalam aditif pakan.

Kata kunci: bakteri asam laktat, L. rubellus, silase, aditif pakan

A 3-3

# Influence of Bile on Lactobacilli Viability and Ability to Reduce Lactose in MRSL Broth

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#### ABSTRACT

Lactobacillus bugaricus (yogurt starter), Lactobacillus acidophilus N2 (isolated from fecal material), and Lactobacillus sp Dad 13 (isolated from dadih, a West Sumatera traditional fermented buffalo milk) were investigated for properties relevant to the production of fermented milk products for consumption by lactose-intolerant humans. In vitro assessment of acid tolerance of lactobacilli was carried out in MRSL at pH values of 2.0, 2.5, 3.0, and 3.5. The three lactobacilli were tested their bile sensitivity in MRSL broth containing 5% lactose, supplemented with various concentration oxgall, and evaluated their ability to reduce lactose content.

There was relatively no loss of viability for all strains at pH 3 and 3.5, while at pH 2.0 they showed reduced viability for 2-3 log cycles after three hours incubation. All three lactobacilli can grow in MRSL containing 0.5% oxgall, although their growths were more slowly than in the absence of oxgall. Their viability tends to decrease with increase in oxgall concentration. The ability of the three lactobacilli to reduce lactose content in MRSL was higher in the presence of oxgall and tend to increase with the increase in oxgall concentration. The presence of 0.5 to 5% oxgall did not enhance of lysis of cells. It could be that the permeability of lactobacilli cells increased in the presence of bile, permitting more lactose substrate to enter the cells, thus increasing the ability to reduce lactose in the media.

Keywords: lactobacilli, acid tolerance, bile, lactose

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