



Isolation and identification of lactic acid bacteria from raw meat

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ABSTRACT

The study was carried out to isolate Lactic Acid Bacteria (LAB) from cow feces meat as well as screening bioactive compound produced from isolated LAB via well-in agar method. The identification of LAB through biochemical test consists of sugar test and milk curdles. Isolated LAB was fermented at 37°C for 24 hours. One type of LAB was successfully isolated, namely *Lactococcus sp.*

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Keywords

Lactic Acid Bacteria (LAB),
Antimicrobial compound.

Introduction

Lactic acid bacteria (LAB) are potential to produce lactic acid as a result of carbohydrate fermentation. These microbes are broadly used for the productions of fermented food products such as yogurt (*Streptococcus spp.* and *Lactobacillus spp.*), cheeses (*Lactococcus spp.*) and sauerkraut (*Leuconostoc spp.*) (Daeschel, 1989). These organisms are heterotrophic and generally have complex nutritional requirements because they lack many biosynthetic capabilities (Khalisanni 2011). Thus, LAB are abundant only in communities where these requirements are provided. They often associate with animal oral cavities and intestines (e.g. *Enterococcus faecalis*), plant leaves (*Lactobacillus*, *Leuconostoc*) as well as decaying plant or animal matter such as rotting vegetables, fecal matter and compost (Khalisanni et al. 2011).

Materials

Samples

Processed meat obtained from local stall located at Bandar Baru Seri Iskandar, Perak. The meat was used to isolate Lactic Acid Bacteria.

Test Microorganisms

The test microorganisms were obtained from department of Microbiology, MARA University of Technology, Shah Alam, Selangor. 6 types of test microorganisms were used namely *Escherichia coli*, *Bacillus subtilis*, *Staphylococcus aureus*, *Streptococcus pyogenes*, *Klebsiella sp.* and *Clostridium sp.*

Methods

Meat

0.5 ml of crushed meat suspension were poured onto MRS agar plates and incubated anaerobically at 37°C for 48 hours with modified pH at 5.5. The growth microorganisms were streaked on MRS agar. Gram stained was conducted to identify the types of LAB from the isolation.

Biochemical Tests

Two types of biochemical test were conducted:

1. Sugar Tests

2. Milk Curdle Test

Fermentation

Pure culture of the LAB species from stock culture was inoculated into 150 ml MRS broth and shake at 150 rpm at 37°C for 24 hours anaerobically.

Antimicrobial Test

The fermented broth was centrifuged at 5000 rpm. The supernatant was dispensed into wells onto Nutrient Agar plates which was lawn with test microorganisms. Sterilized penicillin was used as positive control and sterilized Nutrient Broth was used as negative control. The plates were incubated at 37°C for 24 hours. Antimicrobial activities were indicated by the presence of inhibition zone around the wells.

Discussion

Based on the results, there was a genera of LAB successfully isolated from the sample which is *Lactococcus sp.* The LAB fermented sugars via Homolactic Pathway because from the biochemical test result, it shown that sugar test contained Andrades indicator changed color without gas production detected. As refers to Table 3, the reduction of pH indicates the production of lactic acid. The presence of antimicrobial compound from the LAB was proved by the appearance of inhibition zone against all test microorganisms.

Conclusion

One genera of Lactic Acid Bacteria (LAB) was successfully isolated from meat samples and able to produce antimicrobial compound against some gram positive and gram negative bacteria.

Reference

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Table 1: Characteristics of isolated LAB from meat

Source	Macroscopic observation	Microscopic observation	Lactic acid bacteria genera
Meat	White, circular, small-sized	Gram positive, cocci	<i>Lactococcus sp.</i>

Table 2: Biochemical test observation

Biochemical Tests		<i>Lactococcus sp.</i>
a)	Sugar test	
i)	Fructose	+ve
ii)	Galactose	
iii)	Glucose	+ve
		+ve
b)	Milk curdle	+ve

Table 3: Antimicrobial test observation

Samples	Meat
Isolated LAB	<i>Lactococcus sp.</i>
pH of MRS broth after fermentation	4.93
Test Microorganisms	Inhibition zone:
a) <i>Bacillus subtilis</i>	+ve
b) <i>Clostridium sp.</i>	
c) <i>Staphylococcus aureus</i>	+ve
d) <i>Streptococcus pyogenes</i>	
e) <i>Klebsiella sp.</i>	+ve
f) <i>E. coli</i>	+ve
	+ve
	+ve
	+ve