

## Effect of Lactic Acid Bacteria and Yeasts towards Chemical, Physical and Organoleptic Qualities of Mutton Salami

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**Abstract**— Mutton has exceptional qualities such as smell, high consistency, and huge muscle fat, which makes mutton are restricted to consume. To expand mutton utilization, diversification with proper innovation is important. Salami is one of the proper mutton products that required starter such LAB that can deliver metabolites to change the texture, taste, and odor of meat. Other than LAB, yeasts additionally show up in meat processing, particularly in salami fermentation, which can build the meat taste and odor. The study aimed to identify the chemical, physical (water holding capacity, tenderness, and pH) and organoleptic (odor, color, texture, and taste) quality of mutton salami. Completely Randomized Design (CRD) was used as an experimental design with starters of *Lactobacillus Plantarum* as lactic acid bacteria (LAB) and yeasts, i.e. *Cryptococcus humicolous* and *Trichosporon beigeli*. The treatments were divided into 5 levels, i.e. R1 (0.5% LAB: Yeast 0.5%), R2 (1% LAB: Yeast 1 %), R3 (1.5% LAB: Yeast 1.5%), R4 (2% LAB: Yeast 2%), and R5 (2.5% LAB: Yeast 2.5%), with four replications. Analysis of variance was used to find out the treatment effect, and the Duncan Multiple Range Test was used to find out the difference between any treatments. The results show that the addition of *Lactobacillus Plantarum* from 0.5% to 2.5% and yeast *Cryptococcus humicolous*, *Trichosporon beigeli* from 0.5% to 2.5%, did not significantly affect chemical and physical quality, while water holding capacity and organoleptic test, i.e., color, odor, and texture showed significant effects except taste.

**Keywords**— Lactic Acid Bacteria; yeast; qualities; Mutton Salami.

### I. INTRODUCTION

Mutton chemical composition depends on the animal condition, carcass types, and animal species [1]. The chemical composition of mutton consists of water, protein, fat, ash, vitamins, and non-protein compounds [2]. The protein content of mutton between 18.70-19.58%, while the fat content is 4.24%. Mutton has special characteristics such as odor, high consistency, and large muscle fat, which makes mutton are limited to consume.

To increase mutton consumption, diversification with appropriate technology is necessary for mutton processing. One of the meat diversification products is salami. Salami production required starter such LAB that can produce metabolites to change the texture, taste, and odor of meat. During the fermentation process, LAB also produces compounds which can inhibit microbial growth. The activity of LAB could decline pH and yielding several compounds such as hydrogen peroxide, diacetyl, carbon dioxide, and a bacteriocin. Lactic acid bacteria are a group of gram-positive bacteria that not created spores, have coccus and stem forms, and can ferment carbohydrates to produce lactic acid [3].

The LAB that involves in fermenting salami is *Lactobacillus Plantarum* group, *L. sake*, *L. curvatus*, *Pedicoccus acidilactis* combined with *Pedicoccus pentosaceus* [4].

Besides LAB, yeasts also appear in meat processing, especially in salami fermentation, which can increase the meat taste and odor. Yeast is a unicellular fungi-shaped microorganism, has high endurance such as antimicrobial, high resistance to the antibiotics presences and resistant to acid, salt, and temperature [5]. The yeasts isolated from fresh red meat and fowl meat i.e. *Candida sp.*, *Rhodotorula sp.*, *Debaryomyces sp.*, *Trichosporon*, while on frozen mutton chops found *Cryptococcus laurentii*, *Candida zeylanoides*, *Trichosporon pullulans* [6]. *Debaryomyces hansenii* on Italian salami affects the quality of sausage and provides a delicious taste [7].

Smokey flavor was also important characteristics in Salami. The smoke liquid of coconut shell can be used in Salami production, and it contains various chemical components such as phenol, aldehyde, ketone, organic acid, alcohol, and esters [8]. Coconut shell pyrolysis produces smoke with 4.13% phenol, 11.3% carbonyl, and 10.2% organic acid [9]. Chemical components contained in liquid

smoke can act as antioxidants and antimicrobials. It also provides a distinctive color effect and characteristic smoke taste in Salami products [8]. The research aims to determine the effects of LAB and yeasts inoculation in Salami production towards the chemical, physical, and also the organoleptic characteristic of Salami.

## II. MATERIALS AND METHOD

Five kg meat obtained from the thigh of 3-year-old local male mutton were used as a sample for making salami. Seasoning of Salami consists of garlic, ginger, pepper, nutmeg, sugar, salt, corn-starch, skim milk and fat. The starter used *Cryptococcus humicola* and *Trichosporon pullulans* as yeasts and *Lactobacillus Plantarum* as LAB. Mutton salami processed using meat and fat ratio of 80:20 (g/g) which mixed and frozen for 24h [10]. After that, the mixture was mixed using food processor within the seasoning, like garlic, ginger, pepper, nutmeg, sugar, salt, corn-starch, and skim milk. The starter of *Lactobacillus Plantarum*, *Cryptococcus humicola* and *Trichosporon beigeli* were inoculated with various treatment dose of R1 (0.5% LAB: Yeast 0.5%), R2 (1% LAB: Yeast 1%), R3 (1.5% LAB: Yeast 1.5%), R4 (2% LAB: Yeast 2%), and R5 (2.5% LAB: Yeast 2.5%). The mixture was inserted into a 50 mm diameter casing and tied up to a vertical distance of 8 cm, then hung on a rack and held for 24h at room temperature [11].

Furthermore, the fermentation occurred at room temperature for 6d and interspersed with curing for 1h every

two days. The curing temperature was maintained between 27-30°C with dry coconut shell used as fuel. To control the temperature, ice was added to the curing room if the heat exceeds over than 30°C.



Fig. 1 Mutton Salami

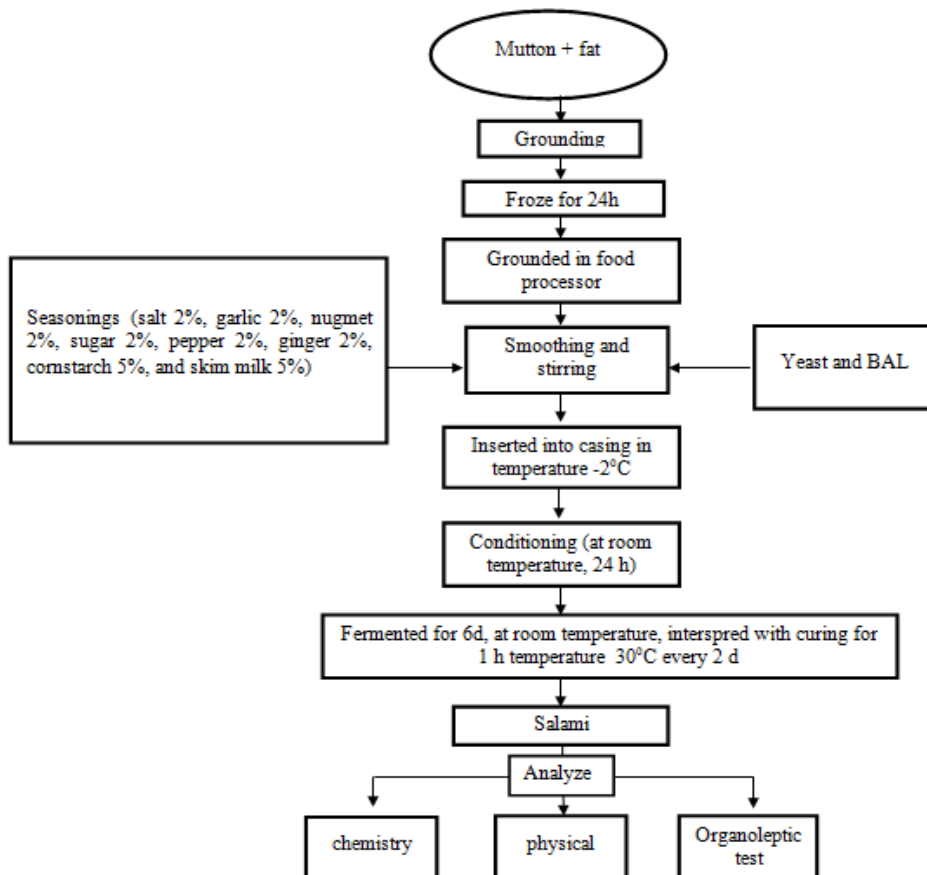


Fig. 2 Research Flow Chart

The parameter consists of chemical quality, i.e., moisture content, protein, and fat, physical quality, i.e., WHC, tenderness, and pH, and organoleptic test performed by hedonic test or preferences on the five treatments [12]–[15]. A panelist group of 50 consumers conducts the organoleptic test. The panelists played a role in the color, odor, texture and taste tests with the hedonic scale, i.e. Like Extremely (1) Like Very Much (2) Like Moderately (3) Like Slightly (4) Neither Like nor Dislike (5) Dislike Slightly (6) Dislike Moderately (7) Dislike Very Much (8) Dislike Extremely (9). The experimental design used a Completely Randomized Design (CRD) with five treatments and four replications to obtain 20 units [16]. Chemical and physical qualities were analyzed using verbal test. The statistical analysis will be continued with Duncan test, after the ANOVA test resulting in a significant difference between treatments, while the effect of treatment on the organoleptic test was analyzed using Kruskal-Wallis test [16], [17].

### III. RESULTS AND DISCUSSION

Lactic acid bacteria of *Lactobacillus plantarum* and yeasts of *Cryptococcus humicolous* and *Trichosporon beigelii* showed good interaction to help each other in the growth process. The increasing levels of yeast and LAB have no significant effect on the fat content, due to the fat degradation by lipase enzymes in salami and producing a group of free fatty acids that give the characteristic such as taste, odor and the structure which depends on the type of acid. Degradation of phospholipids may change the structure of lipids in meal or food, although lipid metabolism by yeasts includes lipase lipid production outside the cells [18].

The fat resulted in lower than the research result of Piotrowicz and Mellado [19] that is 20 percent this matter caused by raw material used differently even though the same processing method. It is called a fumigation process rancidity that will not occur even with there a fumigation process. Microorganism will die like *Escherichia collie*, *Salmonella enterica*, *Staphylococcus aureus*, *Listeria monocytogenes*, *Clostridium botulinum*, and *Toxoplasma gondii*.

TABLE I  
PERCENTAGE OF FAT, PROTEIN AND WATER CONTENT ON MUTTON SALAMI WITH STARTER LACTIC ACID BACTERIA AND YEAST

Treatment	Means		
	Fat (%)	Protein (%)	Water Content (%)
T <sub>1</sub>	9.45 a	15.63 a	62.98 a
T <sub>2</sub>	10.28 a	15.75 a	62.43 a
T <sub>3</sub>	11.49 a	15.22 a	61.30 a
T <sub>4</sub>	11.40 a	15.30 a	61.84 a
T <sub>5</sub>	10.76 a	15.32 a	61.31 a

No difference in protein levels in any treatment caused by yeast has an ability of the cellular *proteolytic* enzyme to hydrolyze peptide bonds in salami protein. This is in line with Setyaningsih, *et al.* [20] which states that the ability to break the protein molecules in food is limited to a few species microorganisms which can produce extracellular *proteolytic* enzymes.

The protein requirement for yeast comes from amino acids, non-protein nitrogen or peptide, while the energy source of yeast and lactic acid bacteria are carbohydrates. Even though the mutton salami contains relatively few carbohydrates, yeast and LAB can use proteins as energy sources. Because the nitrogen requirement for microorganisms can be derived from amino acids, nitrogen non-proteins or peptides and proteins [20].

The protein content generated is not different for every treatment, this matter caused by meet used is not occurring surimi process first, like finding by Yoruk and Guner [21] that the making of sausage or salami if occur surimi process its meat that is leaching process first then addition anti denaturant will increase protein level from sausage or salami which resulted.

The protein content of mutton obtained about 15.22 until 15.75%, that protein contains almost same with the research of Piotrowicz and Mellado [19] use pomegranate peel 1% until 3% generate protein 15.85, where pomegranate peel contains material which can inhibit protein denaturation so that protein can be maintained. Besides that also function as preservative material same like yeast and can increase the quality of salami which generated.

The variance analysis result showed that the addition of lactic acid bacteria *Lactobacillus plantarum* and yeasts *Cryptococcus humicolous* and *Trichosporon beigelii* did not significantly influence moisture content of salami, with the range from 61.31% to 62.98%. It was due to the high ability and strengthens of yeast and LAB in protein meat to hold water. However, the levels of yeast and LAB varies. During fermentation, salami moisture content had an opportunity to spread evenly and quickly throughout the sausage, although the percentage varies. This is appropriate with Sriphecanart, and Skolpap [15] statement that fermentation allows water can spread evenly around part of sausages. Furthermore, Anal [22] had reported on the addition of 2% concentration of crude yeast extract biopreservatives without adding LAB, resulting in the moisture content of 65%. Mughtadi *et al.* [23] said that the moisture content of smoked chicken sausage with a curing temperature of 50°C for 20 minutes, followed by fogging 50°C for 90 minutes resulted in 56.53% moisture content. Roostita [24] reported that pork sausage decreased water content regularly with fat and nonfat without yeast and LAB as a starter treatment.

According to Wambui *et al.* [25], a processed product like sausage, salami, hamburger, and meatball if implemented ISO Food Security System will make products which consumed by consumer become more safety from the health field.

Yeast grows earlier than LAB because yeast prefers to grow in acidic conditions, i.e., at pH 4- 4.5 with optimum temperature 25-30°C and maximum temperature 35-47°C. Most of the yeasts grow well in the circumstances with sufficient water supply. Yeast produces biochemical like enzymes, antimicrobial compounds (organic acids such as *hexanoic*, *octanoic*, and *decanoate*), and proteins.

Looked from the research the water content of the result same, the highest LAB and yeast are not influenced water content of sheep's salami which obtained, the water content is one of the indicators of quality from salami like declared by the AA. Water content is one of the factors determining

the quality of processed meat like a burger, salami, etc. Water content highly influenced by storage, the longer storage decreases the water content.

TABLE II  
TENDERNESS, pH AND WATER HOLDING CAPACITY OF MUTTON SALAMI WITH STARTER LACTIC ACID BACTERIA AND YEAST

Treatment	Means		
	Tenderness (mm/min)	pH	WHC (%)
R <sub>1</sub>	0.11 a	4.46 a	25.33 a
R <sub>2</sub>	0.11 a	4.58 a	25.34 a
R <sub>3</sub>	0.11 a	4.49 a	36.00 ab
R <sub>4</sub>	0.14 a	4.57 a	36.46 ab
R <sub>5</sub>	0.12 a	4.54 a	39.46 b

The addition of yeast and LAB in spite of different concentrations can break the bundles of the same rough muscle fibers, causing the mutton salami to be obtained to have an unequal tenderness.

Tenderness depends on water content which storage in the product. This matter suitable with Holck *et al.* [26] statement which declares if the water content has a relation with tenderness and have a positive correlation. Where if the food has a high water content so that that food production will have a high tenderness level.

The final pH value of the meat or in dairy products will determine other characteristics such as water holding capacity, the growth of microorganisms, protein and enzyme denaturation, tenderness, and emulsification.

The pH value obtained 4.46-4.58 appropriate with the research of Roselino *et al.* [27] declare if pH obtained at salami or fermented sausage under 4.7 is the best quality of pH seen from a health side because a pathogenic microorganism will die. According to Sriphocant and Skolpap [15] at making salami use, *Enterococcus faecium* CRL 183 and *Lactobacillus acidophilus* CRK 1014 obtained pH about 4.47 almost same.

The lower pH obtained on each treatment, it is because of yeast and LAB as a starter in salami processing is in acidic condition. Yeast and bacteria growth with controlled in mutton salami makes the microbes can produce lactic acid, which is decreased pH in mutton salami and contribute to establishing taste, texture, color, and preservation [28].

Table 2. showed that 0.5% to 2.5% of yeast and LAB levels were not significantly different in tenderness and pH, excepting in water holding capacity. Starter of Yeast and LAB significantly affected ( $P < 0.05$ ) the water holding capacity in mutton salami, because the yeast has a strong cooperative with LAB. The higher levels of yeast and LAB will increase water-binding protein, so the water blocked and the content still high [29]. Salami with high WHC correlated with upper tenderness. Yeast can change the meat texture and then affect the WHC. The higher the added yeast level, the higher the water holding capacity [30].

Treatment LAB and yeast with a high concentration have a high water binding capacity. According to Wambui *et al.* [31] that water binding capacity has a correlation with texture or tenderness, where sausage has a high water binding capacity will have a high juice flavor too so that will affect the quality of that sausage.

According to Table 3, there is no different color on treatments, because the brown color on the mutton salami surface is the result of a browning or Maillard reaction. The browning mechanism involves the reaction of free amino acid groups to proteins with carbonyl groups of sugar and carbohydrate because carbonyl is a major compound produced during curing [32]. Hydroxyacetaldehyde (glycoaldehyde) causing brown color form. Other components of glycoaldehyde such as glyoxal and pyruvic aldehyde, allow for the brown coloring in fumigation [33]. The mutton salami color also influenced by pH and myoglobin, meat pigment concentration factor. Myoglobin is easily damaged sarcoplasmic proteins in an acidic atmosphere [34]. Yeast and LAB added a sour taste in salami, thus the same color of salami in the treatments is caused by damaged the sarcoplasmic protein.

TABLE III  
ORGANOLEPTIC RESULT TEST OF MUTTON SALAMI WITH STARTER YEAST AND LACTIC ACID BACTERIA

Treatment	Means			
	Color	Odor	Texture	Taste
P <sub>1</sub>	3.13 a	3.40 a	3.46 a	3.06 a
P <sub>2</sub>	3.46 a	3.63 a	3.43 a	3.23 a
P <sub>3</sub>	3.33 a	3.43 a	3.26 a	3.46 a
P <sub>4</sub>	3.16 a	3.67 a	3.50 a	3.73 ab
P <sub>5</sub>	3.23 a	3.73 a	3.43 a	4.00 b

The fermented sausage color includes dark, this matter caused by H<sub>2</sub>O<sub>2</sub> which generated by a microorganism from fermented aerobic. The ability of H<sub>2</sub>O<sub>2</sub> to oxidation cause alteration of enzyme system in the microbe cell so that used as an antimicrobial [34]. The food product color has a very important role in determining the quality and has sale value and attraction from consumer so that consumer can give judgment on that food material, and that color depends on meat which used.

Salami odor produced by smoke, which comes from phenol. Phenol is the main compound in a distinctive smoke odor forming. According to Mendoca *et al.* [35], a nice taste arising from the presence of acids and phenols from the smoke. There is a different taste found during the curing and only the basic ingredients of the meat. Taste depends on the reaction between smoke and protein components in salami [36]. Salami has a characteristic odor from chemical reactions during fermentation [37]. Salami odor depends on the seasoning and the curing process. Suryaningsih [32] argues that the meat processing products odor is influenced by varying factors, i.e. species, nation, time and conditions of meat storage after cutting and also the length and temperature of cooking.

Lactic Acid Bacteria and yeast as a starter in processing fermented sausage give an effect on a quality sensory especially smell. A compounds volatiles 6 rings that are 2-metil-3-tetrahydrofuran, 2-pentil-puran, tiofen-2-carbocsaldehyd, 5-metiltoifuraldehyd, 2-5 dimethyl and 2, 4, 5-trimethyl-oxazoline [15], [31].

Texture relates to the balance of adding water, fat, and protein. If the product contains a low fat so that will generate the dry and hard product [38]. Yeast in a dough of salami fermented will increase gas so that cause the dough to

expand. If fermentation process run with good, that will generate the perfect processed product like volume, texture, and flavor [39].

Meat texture is related to the softness of collagen at 60°C and will lower at the higher temperatures for a long time. Tenderness will be achieved without hardening myofibril protein at the temperature of 57-60°C. Tenderness in mutton salami involves of several aspects, includes easy penetration of teeth, soft chewed and the amount of residue after chewing [15].

Mutton salami tastes showed a significant difference between treatments. The higher yeast and LAB level the lower the delights because it gives a sour taste into salami. Tastes assessment depends on panelist taste sensitivities, so it was given a random taste values in mutton salami. In Mendoca *et al.* [35], curing will provide a distinctive flavor, which produced by acid and phenol, formaldehyde and furaldehyde, and other compounds, such as seasoning, appearing nice flavor in curing products.

#### IV. CONCLUSIONS

The addition of starter *Lactobacillus plantarum* from 0.5% to 2.5% and yeasts *Cryptococcus humicolus* and *Trichosporon beigeli* from 0.5% to 2.5% did not significantly affect chemical quality, physical quality except water holding capacity and organoleptic test of color, odor, texture except taste of salami.

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