POTENCY OF PAPAYA SEED POWDER (Carica papaya L.) AS CHICKEN MEAT AND SHRIMP PRESERVATIVE

Nani Radiastuti1*, Gusniar Auvina Berty2, Reno Fitri3

INTRODUCTION

Chicken meat and Vaname shrimp are easily-damaged food because of its high protein contains. Meat damage is largely influenced by sanitation at the slaughterhouse, during transportation, marketing, and during the meat storage. Thus good handling is important to maintain the quality of chicken and shrimp when they arrived at consumers. The short shrimp durability is affected by the high water content of shrimp that reaching 76.14% (Suradi, 2016; Ahmad et al., 2018). The presence of microorganisms can cause damage, rottenness, and produce unpleasant odors (Winarno, 2007). One way to extend the storage time of food by providing preservatives. Providing preservatives is a method used to extend the storage time of fresh meat and other meat products (Soeparno, 2011). Food preservatives can be classified into two; first, natural preservatives obtained from natural material such as red galangal extract (Alpina purpurata) against the durability...
of chicken meat, extract of betel leaf (\textit{Piper betle}) and bay leaf (\textit{Syzygium polyanthum}) as an alternative to tofu preservatives. The second group is synthetic preservatives such as acetic acid, benzoic acid, and sorbic acid (Kharisma, 2013).

The weakness of synthetic preservatives is a side effect that appeared. Synthesis preservatives are believed to cause negative effects on health such as cancer cell growth caused by carcinogenic compounds in preservative. So, awareness of preservatives food using natural ingredients began to increase as it believed to be safe for consumption (Susanto et al., 2011).

Research on the use of natural material as a preservative in fish has been carried out by various researchers including the use of \textit{Aloe vera} as a natural material in preserving fresh milkfish (Agustini et al., 2007). Research related to papaya seed powder as a preservative has been conducted by Salim et al. (2018), he proved that the use of papaya seed powder (unknown variety) at a concentration of 4\% has antibacterial characteristic against fresh shrimp because it can reduce the pH and the total colony of fresh shrimp bacteria. The total bacterial colonies in the fresh shrimp have not met the national standard (SNI 7388:2009) in terms of the quality limit of microorganism contamination in animal-based food material (5x10^{5}\text{CFU/g}). According to Peter et al. (2014) papaya seed extract from several concentrations has the potential to inhibit the growth of \textit{Staphylococcus aureus}, \textit{Pseudomononas aeruginosa}, \textit{Escherichia coli}, and \textit{Salmonella typhi}. Papaya seed contains tannin, phenol, saponin and flavonoid compound. Tanin has the ability to inhibit bacterial growth (Taufiq et al., 2015). Therefore research applications of papaya seed powder need to be studied further, especially applications in other fresh foods with different varieties.

The research was conducted using papaya seed ‘Bangkok’ and ‘California’ varieties. These two varieties can be obtained easily and consumed by many people. This research was the first step to find out the length of storage of chicken meat and shrimp that was given papaya seed powder. Moreover, it showed the quality and organoleptic characteristic of fresh chicken meat and shrimp after giving papaya seed powder varieties ‘California’ and ‘Bangkok’ with storage duration of one and three days at 4\°C.

**MATERIALS AND METHODS**

This research used papaya seeds from different varieties, namely the Bangkok papaya variety that was obtained from a plantation in Sukabumi, and the California variety that was obtained from a plantation in Bogor. The samples were taken in June 2019.

**Papaya Seed Powder of ‘Bangkok’ and ‘California’ Varieties**

Papaya seed sample of 100 g was dried using the wind dry method for five days. Sample of papaya seeds that have been dried was then crushed by a blender to obtain papaya seed sample in the form refined powder so that it is more soluble in water. Seed powder was then filtered by siever 200 mesh (Masduqi et al., 2014).

**Preservation of Chicken Meat and Shrimp**

Chicken meat and shrimp samples obtained from the sampling location were immediately packed and put in zipped plastic to maintain the cleanliness of fresh chicken meat and shrimp. The samples were taken to the laboratory to be treated by papaya seed powder varieties of ‘Bangkok and California’. The papaya seed powder in the concentrations of 0, 2, 4, 6, and 8 g was sprinkled to 100 g of fresh...
chicken meat and shrimp (without removing the skin). The fresh chicken meat and shrimp were then put in a zippered plastic and stored for three days at a cold temperature (4°C).

**Analysis of Plate Count Agar (PCA) SNI 01-2332-3-2006**

Chicken meat and shrimp samples were weighed aseptically (SNI 7388:2009) as much as 25 g. The samples were then put into a sterile plastic containing 225 mL 0.98% NaCl solution and were homogenized. The result of homogenates was then diluted using serial dilution method by dissolving 1 mL of sample solution into 9 mL of sterile NaCl, until \(10^{-6}\) dilution. The dilutions of \(10^{-4}\), \(10^{-5}\), \(10^{-6}\) were made into two series. As much as 1 mL of solution from each dilution was put into a different sterile PCA in the petri dish. Cultures were incubated in an upside-down cup position for 48 hours at 37°C. Total Plate Count (TPC) testing was carried out on the first and third days.

**Calculation Total Plate Number (ALT)**

Calculation was counted conventionally by using a colony counter. Colonies <25 were not counted whereas colony with 25 – 250 colonies were counted by using the formula (SNI 01-2332.3, 2006):

\[
N = \sum C / \left[ (1 \times n \ 1) + (0.1 \times n \ 2) \right] \times (d)
\]

\(N\) = number of colony product ( colony per g);
\(\sum C\) = number of colony in all plates counted;
\(n1\) = number of plates in the first dilution that are counted;
\(n2\) = number of plates in the second dilution that are counted;
\(d\) = the first dilution that is counted.

**pH Measurement**

\(pH\) measurements were carried out by using a pH meter. The pH meter was previously calibrated by using a buffer solution of \(pH\) 4 and buffer \(pH\) 7 (BSN, 2009).

**Organoleptic Analysis**

The organoleptic analysis was referred to BSN 3924: 2009 in accordance with the organoleptic test sheet score determined by the national standardization agency. In organoleptic testing, data were collected with the assessment sheet (score sheet). Things that were tested including color, aroma, and texture. The raw samples were presented and the panelist were asked to rate them according to their preference level with the addition of a comparative sample without treatment. There were 25 untrained panelists in each organoleptic test. The hedonic test were performed on the first day and third days with the same 25 panelists. The organoleptic test was conducted to determine the acceptance of panelists preference level for the treatment of papaya seed powder on chicken meat and shrimp with the following evaluation criteria: (1= really dislike), (2= dislike), (3= neutral), (4= likes), and (5= really like).

**Data Analysis**

The TPC, \(pH\), and organoleptic test data were analyzed using ANOVA (analysis of variance) to determine the effect of each treatment. If the effect of the treatment was significantly different with a confidence level of 95%, a further Tukey test was carried out.

**RESULTS AND DISCUSSION**

**Average Total Plate Count (TPC) Value of Chicken Meat and Shrimp**

The microbiological quality of chicken meat and shrimp can be determined by a large number of microorganisms in chicken meat and shrimp. Calculation of total bacterial colo-
nies was carried out to determine the ability of papaya seed powder to inhibit bacterial growth in chicken meat and shrimp. The maximum limit of the total number of bacterial colonies in animal-based food material as stated in SNI 7388:2009 are $1 \times 10^6$ CFU/g for chicken meat and $5 \times 10^5$ CFU/g (SNI, 2009) for shrimp. If the bacterial content in chicken meat and shrimp exceeds the specified standard, then they are unsuitable for consumption as food as their quality decrease and may cause disease. The results showed that the number of bacteria cell in chicken meat and shrimp treated with papaya seed powder varieties of ‘Bangkok’ or ‘California’ (Table 1) was decline compared to control.

Table 1. The average TPC value of chicken meat and shrimp treated with papaya seeds powder varieties ‘Bangkok’ and ‘California’

<table>
<thead>
<tr>
<th>Treatment chicken meat (g)</th>
<th>Storage time (days)</th>
<th>TPC (CFU/g)</th>
<th>Standard Deviation</th>
<th>TPC (CFU/g)</th>
<th>Standard Deviation</th>
<th>TPC (CFU/g)</th>
<th>Standard Deviation</th>
<th>TPC (CFU/g)</th>
<th>Standard Deviation</th>
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<tr>
<td>Control</td>
<td>$1 \times 10^6$</td>
<td>$4.2 \times 10^7$</td>
<td>0.46</td>
<td>$1.72 \times 10^6$</td>
<td>0.5</td>
<td>$1.87 \times 10^7$</td>
<td>0.5</td>
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<td>2</td>
<td>$5.3 \times 10^5$</td>
<td>$8.2 \times 10^6$</td>
<td>0.35</td>
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<td>4</td>
<td>$1.2 \times 10^6$</td>
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<td>$1 \times 10^6^*$</td>
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<td>$1 \times 10^6^*$</td>
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Statistical analysis result showed significant difference in papaya seed powder treatment in different concentration and storage time to the number of bacterial cells in chicken meat compared to control ($p<0.05$). This shows that papaya seed powder has the ability to inhibit bacterial growth.

The result showed that giving papaya seed powder as much as 8 g has a very significant effect on the number of chicken meat bacterial cells, compared to the control. This is because papaya seed powder contains several antibacterial compounds such as flavonoids that are able to inhibit bacterial growth. The flavonoids contains phenol called carabolic acid which has an acid characteristic. The structure of cell walls and cytoplasm of bacterial consist of proteins and fats. The presence of antibacterial compounds such as flavonoids can denature proteins and damage cell membranes. Papaya seed powder ‘Bangkok’ variety is more effective than ‘California’ variety,
because by the concentration of 2 g. It can re-
duce bacterial concentration \( (8.2 \times 10^6) \) under control (Table 1).

Treatment of papaya seed powder to shrimp at different concentrations and storage
time was significantly affected the number of bacterial cells compared to controls \((p<0.05)\),
which means that papaya seed powder has the ability to inhibit the growth of different
bacteria in each concentration. Storage time also affected the number of bacterial cells in
shrimp significantly compared to control. The treatment of papaya seed powder as much as 8
g affected significantly the bacterial cell number compared to controls. This is because pa-
paya seed powder has content that can inhibit the growth of microorganisms. That is in ac-
cordance with the opinion of Cahyadi (2008), the antimicrobial character of formaldehyde
is the result of its ability to inactivate proteins into hydrocolloids. Treatment of papaya seed
powder ‘Bangkok’ or ‘California’ variety as much as 2, 4, 6, and 8 g on first day and on
third day resulted in the decreasing number of bacterial cells. The higher the administration
of papaya seed powder to the shrimp, the lower the number of bacteria obtained.

Salim et al. (2018) reported that soaking of papaya seed extract as much as 2, 4,
and 6\% for two hours can increase storabil-
ity of shrimp for 12 days storage time. Ac-
cording to Peter et al. (2014) papaya seed extract with concentrations of 2.5, 5, 7.5, and
10\% have the potential to inhibit the growth of \( Staphylococcus aureus, Pseudomonas
aeruginosa, Escherichia coli, Salmonella
\)typhi.

TPC values increase during storage
because bacterial growth begins to increase
since the shrimp die. The shrimp temperature
rises, causing bacteria to attack immediately.
After tissue damage occurs in the body of the
shrimp, there will be changes in the composi-
tion of the meat that resulting in decay (Salim
et al., 2018). According to Suparno (2012),
decay in shrimp is caused by the presence of
blood vessels and dirt in the shrimp’s head.
Shrimp undergoes autolysis of fats and pro-
teins very fast, and black spots arise on the
treatment carried out in high temperatures.
According to Suradi (2005), microbial activi-
ty during storage produces alkali compounds.
The strong alkali caused by the decomposi-
tion of chemical compounds especially pro-
teins, so that the decomposition process will
continue to increase.

Several studies related to the use of bio-
active compounds as antibacterial have been
carried out. soaking papaya seed extracts on
the storability of shrimp as much as 2, 4 and
6\% for two hours has a storage time of 12 days
(Salim et al., 2018). Papaya seeds have anti-
bacterial effects that useful for curing chronic
skin diseases, such as ekimia. Papaya seeds
also have antibacterial activity against
\( Trichomonas vaginalis \). Papaya seed powder
contains fatty acid bioactive compounds that
have the ability to inhibit bacterial growth.

According to research of Torar et al.
(2017), papaya seeds has antibacterial activi-
ty because it contains alkaloids (Carpaine)
which can inhibit bacterial growth. Carpaine is
a lactonate ringed alkaloid with seven groups
of methylene chains so it is effective to inhibit
the activity of several microorganisms. Car-
paine can digest microorganisms proteins and
turn them into derivatives compound called
peptones. Papaya seeds also contain another
compound such as flavonoids. Flavonoids are
polyphenol compounds that are widespread in
nature, based on chemical structure flavonoid
compounds consist of flavonol, flavon, flavo-
noid catechin, anthocyanidins, and chalcons
(Taufiq et al., 2015).

Papaya seeds powder contains tannin
which tends to have the highest bioactive
compounds content value among the phenol compound, saponins, and flavonoids. Tanin generally has a bland taste and more acidic, this causes the papaya seeds to have a bitter taste. Tannins have the ability to inhibit bacterial growth (Salim et al., 2018). Tannins have the character of wrinkles membranes cell. The action mechanism of tannin as an antibacterial is to inhibit the reverse transcriptase enzyme so that bacterial cells cannot be formed.

**pH Value of Chicken Meat and Shrimp**

Microbiological quality of chicken meat and shrimp is one of the parameters in looking at the quality and damage level to chicken meat and shrimp. Microbiological quality of chicken meat and shrimp can be selected from changes in physical and chemical quality including pH value and organoleptic properties (color, aroma, and texture). The pH value is one of the important parameters that affect the growth of bacteria and other microorganisms.

The initial pH value in both chicken meat samples treated by papaya powder ‘Bangkok’ and ‘California’ varieties showed a neutral pH (7) and remained neutral (6.6 – 7.2) after three days of storage (Table 2). The pH measured in this study was classified as neutral pH. According to Suradi (2008) reported that chickens before slaughter have a pH value of 6.51 and then declined to 5.96 and 5.82 at the time of 10 hours and 12 hours after slaughter at room temperatures. The pH value obtained in this study until the 12th hour was still in accordance with the initial pH value of chicken meat. Data shows that the higher pH level of papaya seed powder ‘Bangkok’ and ‘California’ varieties in chicken meat causes the pH value increase (Table 2). This is because papaya seed powder has a pH value of 7 (neutral), so papaya seed powder affected the chicken meat pH value.

### Table 2. The pH value of chicken meat and shrimp treated with papaya seed powder varieties ‘Bangkok’ and ‘California’

<table>
<thead>
<tr>
<th>Treatment chicken meat (gr)</th>
<th>Storage time (days)</th>
<th>pH</th>
<th>Standard Deviation</th>
<th>pH</th>
<th>Standard Deviation</th>
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<td>1 (day)</td>
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<tr>
<td>Control</td>
<td>7</td>
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<thead>
<tr>
<th>Treatment shrimp (gr)</th>
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<tr>
<td>Control</td>
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<td>8.5</td>
<td>0.42</td>
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The initial pH value of both shrimp samples treated with papaya seed powder ‘Bangkok’ and ‘California’ varieties showed a neutral pH (7). After three days of storage the pH value increased to the alkali (8.2 – 8.9) (Table 2). The pH value continues to increase during storage due to the degradation of proteins that will produce simpler nitrogen compounds such as the free amino acids and nitrogen alkali acid. The addition of nitrogen alkali will increase pH. According to Suradi (2005), the activity of micro-organisms during storage results in the decomposition of chemical compounds. Especially proteins that produce alkali compounds and strong alkali, so that the decomposition process will be followed by an addition in pH.

A decrease in pH due to long storage will cause the water holding capacity to decrease. This is in accordance with the opinion of Soeparno (2009) which stated that the difference in the holding capacity of water is also partly due to the rate and magnitude of the decrease in pH. This is reinforced by the statement of Bahar (2003) which states that the holding capacity of water is influenced by the rate and magnitude of the pH value. The lower the pH, the lower the holding capacity of water to meat. According to Salim et al. (2018), the treatment of papaya seed powder shows a slower pH change because papaya seed powder binds to the binding capacity of water.

**Chicken and Shrimp Organoleptic Value**

Organoleptic testing of chicken meat and shrimp greatly determines its quality. The fresh meat products are chewy and if we pressed with a finger the scar will return soon. Meat that undergoes a decay process will become physically damaged, lose its texture, and watery. According to Torar et al. (2017) for shrimp, the condition of the body segment or leg segment usually determine the freshness level of a segmented fishery product. The fresh condition fishery product such as a kind of shrimp is characterized by the complete limbs that are still attached and the strength of the attached limbs.

From The organoleptic test, it is obtained that organoleptic value experienced decreases after longer storage time. The results of analysis of variance showed that treatment of papaya seed powder Bangkok and California varieties to chicken meat with a storage time of three days was significantly different (p˂0.05) on color, aroma and texture compared to control. Duncan’s further test result showed that the treatment of papaya seed powder at a concentration of 2 g was produced the color, aroma, and texture of chicken meat that was very favorably compared to the concentration of 4, 6, and 8 g.

The color, aroma, and texture of chicken meat treated by papaya seed powder as much as 4, 6, and 8 g were less favorable with scoring of less than 3. It means that the panelists did not like the color in the treatment of papaya seed powder. This is because the result obtained from organoleptic testing based on the level of preference of the panelists which have paler color compare to fresh chicken meat which has a fresh aroma like papaya seed powder.

Organoleptic test obtained showed that low organoleptic values resulted from experience a decrease in color, aroma, and texture caused by a longer storage time. However, the texture has a good level of preference with a scoring of 3. It means panelists like the texture of chicken meat that has a hard texture compared to meat control chicken (Figure 1, 2, 3 & 4).
Figure 1. Organoleptic of papaya seed powder variety of ‘Bangkok’ in chicken meat for one day of storage

Figure 2. Organoleptic of papaya seed powder variety of ‘Bangkok’ in chicken meat for three day of storage

Figure 3. Organoleptic of papaya seed powder variety of ‘California’ on chicken meat for one day of storage
Unlike the treatment of papaya seed powder which has a stable decrease in value during storage, a decrease in organoleptic appearance shows that there is decay during storage. Based on the research by Rajesh (2008) stated that changes in the texture of food material during storage can be caused by changes in water content which affects the hardness of food material (getting softer).

The main components affecting meat hardness are connective tissue, meat fiber, and fat (Aberle et al., 2001). According to Fellows (1992), the texture of the meat is also affected by the type, moisture content, and the number of structural carbohydrates and protein. Texture changes are caused by fat loss, emulsion damage, hydrolysis from carbohydrate polymers, coagulation and protein hydrolysis, as well as the temperature level of the environment and the meat itself. Sitindoan (2007) found that the meat age also influences the hardness level. Old meat has more rough muscle fiber compare to young meat as older meat binds more water which can affect the texture of the meat.

Variance analysis results revealed that a provision of papaya seed powder variety Bangkok on shrimp with storage of three days was significantly preferred (p<0.05) in terms of color, aroma, and texture compared to shrimp control. Duncan’s further test result showed that treatment of provision papaya seed powder at concentration of 2, 4, 6, and 8 g, was significantly favored in terms of color, aroma, and texture of shrimp compared to control. The color as well as aroma and texture of shrimp treated by papaya seed powder Bangkok variety with a concentration of 2 g received an assessment of 3, which means that panelists liked the color, aroma and texture of shrimp even after three days of storage. This is because shrimp did not have pale color, fishy aroma, but has a hard texture. The results were similar with organoleptic shrimp with the treatment of papaya seed powder California variety.

Results of analysis of variance showed that giving papaya seed powder California variety on the shrimp with three days storage time was significantly preferred (p<0.05) in terms of color, aroma, and texture compared to control. Duncan’s further test results showed that the treatment of giving papaya seed powder at concentrations of 2, 4, 6, and 8 g were significantly preferred compare to control. The color, as well as aroma and texture of shrimp by treatment of papaya seed powder in a concentration of 2 g were more favorable than a concentration 4, 6, and 8 g. The higher concentration of papaya seed powder results in the harder the texture and the paler the color. The addition of papaya seed powder to shrimp can reduce the pH value of shrimp thus causing color changes (Figure 5, 6, 7 & 8).
Figure 5. Organoleptic of papaya seed powder variety of ‘Bangkok’ in shrimp for one day of storage

Figure 6. Organoleptic papaya seed powder variety of ‘Bangkok’ in shrimp for three days of storage

Figure 7. Organoleptic papaya seed powder variety of ‘California’ in shrimp for one day of storage
According to Soeparno (2009), the changes in meat color is affected by oxygen exposure, pH, and water content. A decrease in organoleptic appearance showed that there was decay during storage. Based on the research of Karnila et al. (2006). The pre-rigor stage of giant prawns (Macrobrachium rosenbergii) lasted for 2.5 hours after it died. It is marked by soft and supple textured meat with a very fresh odor. The organoleptic value of giant prawns at this stage was 8.9 for the rigor mortis stage of giant prawns (M. rosenbergii) occurring for 6.5 hours starting with muscle lengthening and the body beginning to curve with fresh specific odors. Shrimp organoleptic value at this stage was 7.1 to 8.6, while the post-rigor stage prawn (M. rosenbergii) took place at the 12th hour after it dies which characterized by softening of meat, black fluid discharge from its body, and the emergence of reddish spots on the head, legs, and antenna. At this stage, the organoleptic value of shrimp freshness obtained was 5 (Karnila et al., 2006).

Shrimp texture on third day in the control was soft and less elastic. The organoleptic test results of shrimp texture during cold storage will continue to experience a decrease both in preservatives as well as non-preservatives. This is because the preservative levels decrease along with storage time at cold temperature. Although the level of elasticity is reduced, the preservative shrimp texture is better because the preservative causes shrimp meat texture more cohesive.

Chicken meat texture is perishable media and suitable for microorganism growth because of its high water content and nutrients, especially protein. Some pathogen microorganisms that commonly contaminate shrimp are Salmonella sp., E. coli, and Staphylococcus sp. They are the main type of microorganisms that can cause the decay in fresh fish.

Papaya seed powder of Bangkok variety contains chemical compounds such as alkaloids group, flavonoids terpenoids, and saturated fatty acids. Papaya seed powder California variety contains chemical compounds such as carboxylic acid group, alkaloids, unsaturated fatty acids, flavonoids terpenoids, steroids and saturated fatty acids, which have antibacterial properties. Extract of papaya seed powder of Bangkok variety is obtained four groups of compounds while from ethanol extract of papaya seed powder of California variety it was obtained seven class of compounds. Papaya seeds contain a class of com-
pounds that act as antibacterial agents such as alkaloids, terpenoids, flavonoids, steroids, and fatty acids. The antibacterial activity of saturated fatty acids found in plants generally the most effective activity compared to unsaturated fatty acids.

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