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APPLICATION OF CHITOSAN FROM WASTE GURAMI FISH SCALES (*Osphronemus goramy*) AND CLOVE POWDER (*Syzygium aromaticum*) AS EDIBLE COATING ON CHICKEN MEAT

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ABSTRACT

Chicken meat is a nutritious food that can increase immunity but has perishable food properties that are easily damaged so it is necessary to add preservatives, namely chitosan as an edible coating to protect the meat from microbial contamination. Gourami (*Osphronemus goramy*) scales have the potential to be processed as chitosan. Through 3 stages, namely deproteination, demineralization, and deacetylation. The purpose of this study was to determine the effect of adding chitosan with gourami fish scales and cloves as an edible coating on the quality of raw chicken meat at room temperature for 9 h. This study used a factorial completely randomized design (3×3), with factor A: concentration of chitosan solution (0%, 1%, and 2%) and factor B: concentration of clove solution (0%, 1%, and 2%) and repeated 3 times. The results showed that the control treatment without clove and chitosan after 9 h of storage at room temperature had a significant difference in effect on the treated samples. The best treatment was found in the interaction of 2% chitosan (K2) and 2% cloves (C2) with microbial contamination of 2×10 5 CFU/g so that it still meets the standards of the National Standardization Agency (BSN) namely, chicken meat has a maximum requirement of 1×10^6 CFU/g, which can maintain the quality of meat, both in terms of pH, water content, and acceptability of organoleptic values which are still favored by panelists.

Keywords: Chitosan; poultry meat; sensory; scales fish; total bacteria

INTRODUCTION

During the current COVID-19 pandemic, one way to avoid contracting the coronavirus is to maintain the body's immunity. One of the nutritious foods that help to boost the immune system and is liked by many people is chicken meat. In addition to being cheap, one of the active components in chicken meat is an endogenous antioxidant (Al Awwaly et al., 2015) so that it helps increase the body's immune system. Enzymatic endogenous antioxidants are antioxidants produced by the human body as an antidote to exogenous free radicals and endogenous free radicals.

However, chicken meat is one of the perishable or perishable foodstuffs. This is because chicken meat is a good medium for the growth of bacteria. If the bacteria are pathogenic, the bacteria will cause various diseases and can cause the meat to rot quickly. The majority of chicken meat sales are carried out in traditional markets by placing them at room temperature and open for more than 6 h. All research samples of chicken meat from several markets in the city of Kupang showed high microbial contamination ranging from 5250000 CFU/g - 92500000 CFU/g when compared to the requirements of SNI 08-1-1-7388: 2009 which is 10^6 CFU/g (Ariesthi et al., 2011). This is because the total bacteria in chicken meat can increase up to 100 times or more when stored at room temperature for a long time.

Efforts to maintain the quality of a food ingredient are coating it with an edible coating that is biodegradable. One of the materials that can be used in the manufacture of edible coatings is chitosan. Chitosan can be obtained from the scales of gourami (Osphronemus goramy). According to Nurjanah et al. (2010) scales of gourami (Osphronemus goramy) at a weight of 260-3,315 g contain 0,4%-3,7% chitin so that it has the potential as an ingredient for making chitosan. The difference in chitosan content was obtained from the weight of the fish used. The larger the gourami whose scales are taken, the lower the chitosan content, this is because the greater the weight of the gourami proportionally the harder the texture of the scales is caused by the mineral content of hydroxyapatite. Susanti and Purwanti's research (2020) that fish scales are waste that has not been used optimally, but has the potential to make chitosan.

Tulungagung is the largest supplier of gourami in Indonesia. According to the Director-General of Fisheries and Marine Affairs (2016), the largest gourami production center in Indonesia is located in the Tulungagung district, which is 13,404.17 tons. In general, gourami fish are only used for their meat, while the scales become waste both on a large industrial scale and on a household scale (Pratama et al., 2015). The lack of processing waste carp scales causes problems in the environmental field that can extend to social problems to health.

The manufacture of chitosan is carried out in three stages namely, the deproteinized stage to remove the protein contained in fish scales, the demineralization stage to remove minerals contained in fish scales, and the deacetylation stage to remove the acetyl groups contained in chitin and form chitosan. Chitosan can also form a membrane that functions as an adsorbent at the time of the binding of organic and inorganic substances by chitosan (Susanti and Purwanti, 2020). The use of chitosan as an edible coating for effective and safe in

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preventing damage to the quality, extend shelf life and maintain nutritional value (Alhuur et al., 2020), but the chitosan has an aroma that is tart, according to Harjanti (2014) that the scent of chitosan bit pungent and can affect the aroma of the meat. This is because chitosan requires an organic acid solvent, so it is necessary to add other ingredients like a mixture for making edible coatings, namely cloves.

Cloves (Syzygium Aromaticum), are scented dried flower stalks of the Myrtaceae tree family. Clove is a spice native to Indonesia that is abundant and easy to find, especially in East Java. East Java is the highest clove producer in Indonesia (Hakim, 2015). Yusuf, et.al (2019) stated that cloves have natural antimicrobial and antioxidant activity. Tinangon et al., (2017) that the clove flower is a flavorful spice, has a warm taste, and is generally used as a flavor enhancer in meat. According to (Andries et al., 2014), eugenol is the main component contained in clove flowers reaching 70-96% so that it can inhibit bacterial growth. The eugenol content in cloves can kill bacteria including bacteria that are resistant to antibiotics (Andries et al., 2014) one of which is MRSA bacteria which are resistant to several classes of antibiotics (Azizah et al., 2017).

Clove powder can be used as an additional ingredient in the manufacture of edible coatings, besides being able to inhibit the growth of microorganisms, it also has a fragrant aroma. Towaha (2012), stated that eugenol compounds and their derivatives which have antioxidant and antimicrobial properties can be used as raw materials for edible coatings and edible films. The use of cloves as an additional ingredient in the manufacture of edible coatings, will increase the variety of functions of cloves and minimize price fluctuations in clove flowers, such as during the current pandemic.

Coating chicken meat using an edible coating of chitosan and cloves is expected to be able to form a good edible coating and can extend the shelf life of meat. In addition, chitosan and cloves used in edible coatings can add active components that can improve the quality and delicious aroma of chicken meat. The characteristics that must be possessed by an ideal edible coating are that it is non-toxic, allergic, and digestible, its structure is stable so it can prevent mechanical damage during transportation, handling, and display, providing semi-permeability to maintain internal gas balance so as to slow down aging, preventing loss of components. which can change the organoleptic characteristics (Akhtar et al., 2015).

MATERIALS AND METHODS

This research was conducted at the Central Laboratory of Biological Sciences, Brawijaya University, Malang, East Java, for the manufacture of chitosan from gourami fish scales and the manufacture of chitosan and cloves as edible coatings, then testing pH, water content, and TPC. Wage Market chicken traders in Tulungagung Regency as panelists in organoleptic tests. The time of the study was carried out from November 2020 to February 2021.

Material

The materials used in this study were fresh chicken meat purchased from the Mergan traditional market in Malang City and the traditional market in Tulungagung Regency, chitosan powder, 2% fulltime brand acetic acid aquades, clove flower powder, Nutrient Agar media, spirtus liquid, peptone, pH buffer 4, pH buffer 7 and pH buffer 10, alcohol 70%, aluminum foil, tissue, brown paper, label paper, filter paper.

Tools

The preparation stage begins with preparing the tools that will be used in the research process, namely a styrofoam box with a capacity of 30 kg as a place to store newly purchased chicken meat, 1m × 1m filter cloth to filter the chitosan yield that has been formed, one pack plastic bag, conductor 30 liters, 20cm × 40 cm plastic

tray as a chicken meat storage container, 30 plastic bottles as a solution container, 30 small plastic cups as sample containers when treated, 1 wooden cutting board, 1 cutting knife, tweezers, 2 dropper pipettes, one pair of scissors, organoleptic assessment form as a means for organoleptic testing. The equipment used to make chitosan includes 2 glass beakers with a size of 1 L, 4 pieces of 500 ml in size and 4 pieces of 250 ml in size, analytical balance (Shimadzu-ATX224), 1 liter Erlenmeyer and 500 ml each. 2 pieces, hot plate and magnetic stirrer (Heidolph), 3 pieces measuring 100 ml glass, 2 pieces of stirring rod with a size of 30 cm. Weighing bottles (Schott Duran) as many as 27 pieces as containers for testing the water content, oven, desiccator, clamp holder, pH meter (Horiba-LAQUA) as a test indicator for pH levels. For total bacteria testing, you need tools, namely micropipettes, blue tips, 60 Petri dishes (Pyrex), mortar and pestle, test tube rack, water bath (Memerth), incubator, LAF, destruction stove, Bunsen, porcelain dish, and autoclave.

Research methods

The research method used was experimental with a factorial Completely Randomized Design (CRD) with 2 factors, namely the administration of 0%, 1%, and 2% chitosan concentrations and the administration of 0%, 1%, 2% clove powder with a total of 9 treatments and 3 replications. The control treatment in this study was without giving chitosan and cloves. The treatment is as follows:

P0 : Control treatment (without immersion)
P1 : Treatment with the addition of 0% cloves + 1% chitosan
P2 : Treatment with the addition of 0% cloves + 2% chitosan
P3 : Treatment with the addition of 1% cloves + 0% chitosan
P4 : Treatment with the addition of 1% cloves + 1% chitosan
P5 : Treatment with the addition of 1% cloves + 2% chitosan
P6 : Treatment with the addition of 2% cloves + 0% chitosan
P7 : Treatment with the addition of 2% cloves + 1% chitosan
P8 : Treatment with the addition of 2% cloves + 2% chitosan

preparation of chitosan

a. Early preparation
   The scales of gourami (Osphronemus goramy) were washed thoroughly and dried in the sun to dry. Then the scales are crushed using a flour machine and then sieved.
b. The stage of making chitosan (Susanti and Purwanti, 2020)
   1. Deproteinization
      This process was carried out at a temperature of 65-75˚C using 3% NaOH solution with a ratio of fish scales to NaOH = 1:10 (gram scales/ml 3% NaOH) and stirred for 2 h. Then filtered using filter cloth and filter paper. The precipitate obtained was washed using aquadest to pH 7 (neutral), then filtered again and dried in an oven for 15 h at 60˚C to constant weight.
   2. Demineralization
      To remove the minerals, 0.5N HCl was added. The ratio of fish scales after deproteination with HCl = 1:10 (gram powder/ml 0.5N HCl) into a glass beaker. Then soaked and stirred at a temperature of 30-40˚C for 30 min. The results obtained were filtered using filter cloth and filter paper. Then washed with distilled water until pH 7 (neutral). The obtained solids were dried again in an oven at a temperature of 60˚C. The

product of this process is called chitin.

3. Deacetylation
   To remove the acetyl group present in chitin and become chitosan. Chitin was added with the addition of 60% NaOH with a ratio of chitin to NaOH = 1:10 (gram/ml NaOH) heated at a temperature of 80-90˚C while stirring for 1.5 h using a magnetic stirrer. According to Wulandari et al. (2020) the optimum condition of the degree of deacetylation of chitosan using 60% NaOH. The higher the concentration of NaOH, the greater the value of the degree of deacetylation, so 60% of the NaOH concentration is used. This is because the degree of deacetylation shows the number of broken acetyl groups so that more and more chitosan is formed from chitin (Rumengan et al., 2018). Then filtered and washed until pH 7 (neutral), then dried chitosan in an oven at a temperature of 60˚C

c. The process of making chitosan solution.
   Chitosan was dissolved in 2% acetic acid according to the treatment. Using a ratio of 1 gram of chitosan to 100 ml of 2% acetic acid for 1% chitosan content, and 2 g of chitosan to 100 ml of 2% acetic acid for 2% chitosan content (Saraswati, 2014). The addition of making edible coating on chitosan is by dissolving the chitosan using a magnetic stirrer at a temperature of 45˚C for 3 h. Then filtered and placed in a plastic bottle. Making edible coating on chitosan is by heating and stirring the chitosan solution until it dissolves (Nurhayati et al., 2014)

Clove solution preparation

The cloves were ground to a powder and then dissolved with distilled water according to the treatment. Using a ratio of 1 gram of cloves with aquaes of up to 100 ml for a 1% concentration, and 2 g of clove powder with aquades of up to 100 ml for a 2% concentration of cloves, stirred using a magnetic stirrer for 1 hour at 45˚C. Making edible coatings using chitosan and cloves is by mixing a solution of chitosan and followed by a solution of cloves. Several types of antimicrobial ingredients that can be added to edible packaging include spices in powder form and essential oils such as cinnamon, pepper, cloves, oregano. (Winarti et al., 2012)

Application of chitosan and cloves as preservatives for chicken meat

1. Immersion

The chitosan solution was given according to the treatment and followed by the addition of the clove solution according to the treatment. In this soaking stage, the chicken breast fillet is prepared and soaked for 3 min. Soaking time of 3 min is the optimal time for soaking because it does not damage the texture, smell, and appearance. Each piece of chicken meat that has been soaked is drained using a filter container for ± 10 min then after that, it is stored for the next stage (Alhuur et al., 2020)

2. Storage

The drained chicken meat was then stored in small trays according to the treatment at room temperature (25-30˚C) for 9 h in the open state, then tested on the nine treatments. Research by Jaelani et al. (2018) stated that the storage period of 3-9 h on broiler meat that had been soaked in turmeric juice had the lowest microbial contamination compared to meat stored with more than 9 h of treatment. So that the soaked broiler chicken is stored for no more than 9 h at room temperature, because the longer the meat is stored at room temperature, the more microbial contamination.

Observation Variable

The variables observed in the experimental study of the concentration of chitosan and cloves as preservatives in chicken meat were carried out subjectively (sensory) and objectively (non-sensory). Subjective testing of samples was carried out using organoleptic tests consisting of color, odor, and texture using an assessment scoresheet on chicken meat samples, while objective testing using measurements of pH values, water content, and total bacterial colony values or Total Plate Count (TPC).

a. Organoleptic test

Organoleptic testing was carried out after 9 h of storage by 5 limited panelists. The number of standard panelists involved for one test is 3-5 people (Imbar et al., 2016) where this panel has a high sensitivity to the handling of the product being tested (Ayustaningwaro, 2014). This sensory assessment was carried out on several test parameters, namely color, smell, and texture parameters.

This is because these parameters are important indicators in assessing the freshness of chicken meat. The data analysis used was ANOVA and continued with the DMRT test. The chicken pieces were scored based on an assessment on the score sheet with a scale of 1 (one) as the lowest score and 5 (five) as the highest score by chicken traders in the Tulungagung traditional market.

b. Measurement of pH Value (AOAC, 1995)

The pH test on chicken meat soaked in chitosan and cloves according to the treatment with 9 h storage using the method (AOAC, 1995) was carried out at the Central Laboratory of Biological Sciences, Universitas Brawijaya.

The lower the pH, the higher the value of the water content so that it becomes a parameter of decay. The stability value shown by the pH meter is the value of the sample pH measurement results. The data analysis used was ANOVA and continued with the Honest Significant Difference (BNJ) test.

c. Moisture content (AOAC, 2005)

Testing the moisture content of chicken meat soaked in chitosan and cloves according to the treatment with 9 h storage using the method (AOAC, 2005) was carried out at the Central Laboratory of Biological Sciences, Universitas Brawijaya. The higher the water content in the sample indicates the more microbial activity in the sample. The method used is gravimetric by weighing the sample weight before and after the oven and only using ANOVA data analysis.

d. Calculation of the value of Total Plate Count (TPC) (Fardiaz, 1992)

TPC testing on chicken meat soaked in chitosan and cloves according to treatment with storage for 9 h using the pour plate method (Fardiaz, 1992) was carried out at the Central Laboratory of Biological Sciences, Universitas Brawijaya. This test was conducted to determine the number of bacterial colonies in the treated samples so that the rotting of the meat could be detected. Analysis of the data used was ANOVA and continued by using the Honest Significant Difference (BNJ) test.

RESULTS AND DISSCUSION

Color

The concentration treatment of chitosan solution can increase the color value through organoleptic test from an average of 2.13 to 3.22. The results of the analysis of variance showed that the interaction treatment concentrations of chitosan and clove solutions in table 1, showed no significant difference ($P > 0.05$), while the concentration treatment of chitosan solution and clove solution showed a very significant difference to the color score of chicken meat ($P < 0.01$). Research by Alhuur et al., (2020) stated that the characteristics of colorless (clear) chitosan did not affect the color of the soaked chicken meat, so it did not change the color of the chicken meat and still maintained the color of the chicken meat, while the clove concentration treatment resulted in a decrease in the color value from an average

of 3. ,13 becomes 2,3. This is in accordance with the research of Yusuf et al. (2019) that the color of the fish gills covered with clove powder is slightly light brown and the color of the mucus is slightly cloudy. This is because giving a 2% clove solution gives a more brownish color effect on chicken meat, compared to only 1% clove solution, causing the color of the soaked product to be browner than the original product.

Table 1. Average Organoleptic Color Value with Treatment of Chitosan and Clove Concentration

Factor B (Clove)	Factor A (Chitosan)			Average
	K0 (0%)	K1 (1%)	K2 (2%)	
C0 (0%)	2.20±0.20	3.67±0.58	3.53±0.42	3.13 ± 0.81 [b]
C1 (1%)	2.60±0.20	3.27 ± 0.23	3.13±0.23	3.00 ± 0.35 [b]
C2 (2%)	1.60±0.00	2.73±0.61	2.60±0.20	2.31 ± 0.62 [a]
Average	2.13 ± 0.50 [a]	3.22 ± 0.47 [b]	3.09 ± 0.47 [b]	

Information: [a,b] = Different superscripts in the same column showed very significant differences (P<0.01)
Values represent the mean of 3 independent replicates ± standard deviation

Duncan's follow-up test of 1% showed that there was a significant difference between chicken meat with 0% chitosan treatment (without giving chitosan) and chicken meat with 1% and 2% chitosan treatment. This significant difference indicates that the addition of chitosan can maintain the organoleptic results of chicken meat color so that the color of chicken meat after the storage is still favored by the panelists. The treatment of giving clove powder solution showed that in Duncan's further test there was a significant difference between chicken meat with 2% clove solution and 1% clove solution and without clove solution. The real difference in giving clove solution as much as 2% is because of the color of the chicken meat changes to brown. This is because the original color of the cloves itself and is also influenced by the oxidation between oxygen and fat in chicken meat so that the color is less favored by the panelists.

Smell

The results of the analysis of variance in Table 2 show that the interaction of the concentration of chitosan and cloves showed a very significant difference (P < 0.01) on the odor score of chicken meat. The interaction of treatment with the concentration of chitosan solution and clove solution can increase the odor value through organoleptic tests from an average of 1.13 to 3.87. Treatment of concentration of chitosan solution showed a very significant difference (P<0.01) to the value of the smell of chicken meat.

Chitosan solution treatment can increase the odor value from an average of 2.22 to 3.53. Chitosan edible coating treatment is able to give a good influence on the quality of the smell of chicken meat. Suptijah et al. (2008) stated that the treatment of chitosan solution had an effect on the odor of catfish fillets after 18 h of storage. This proves that the addition of chitosan solution is able to inhibit the emergence of odors that are not favored by panelists by inhibiting the release of volatile compounds that cause foul odors to come out of fish meat through the coating process on the fillets. Treatment of clove solution concentration showed a significant difference (P<0.05) to the value of the smell of chicken meat. Clove solution treatment can increase the odor value from an average of 2.80 to 3.29.

Table 2. Average Organoleptic Odor Value with Treatment of Chitosan and Clove Concentrations.

Factor B (Clove)	Factor A (Chitosan)			Average
	K0 (0%)	K1 (1%)	K2 (2%)	
C0 (0%)	1.13 ± 0.12 [a]	3.87 ± 0.23 [c]	3.40 ± 0.60 [b]	2.80 ± 1.46 [a]
C1 (1%)	2.60 ± 0.20 [b]	3.20 ± 0.40 [b]	3.40 ± 0.20 [bc]	3.07 ± 0.42 [ab]
C2 (2%)	2.93 ± 0.42 [b]	3.53 ± 0.64 [c]	3.40 ± 0.00 [b]	3.29 ± 0.32 [b]
Average	2.22 ± 0.96 [a]	3.53 ± 0.33 [b]	3.40 ± 0.00 [b]	

Information: [a,ab, b, bc, c] = Different superscripts in the same column show a very significant difference (P<0.01)
Values represent the mean of 3 independent replicates ± standard deviation

Duncan's 1% further test showed that there was a significant difference in the interaction of the concentration of the chitosan solution and the clove solution. Treatment with 0% chitosan solution concentration and 0% clove solution (without soaking) showed a significant difference in the effect of other treatments. The difference in effect between chicken meat without soaking and chicken meat with soaking showed that the presence of chitosan and cloves gave better organoleptic results of chicken meat odor, supported by Duncan's further test 1% treatment with 0% chitosan concentration (without chitosan administration) showed a difference which is significant to the treatment of 1% chitosan solution concentration (K1) and 2% chitosan solution (K2). Duncan's 5% further test also showed a significant difference between chicken meat with 0% clove concentration treatment (without giving cloves) and 2% clove solution concentration (C2), this is because clove solution can inhibit the growth of pathogens due to the presence of eugenol compounds. Oyedemi et al. (2008) stated that the eugenol content reached 70-96% so that it could inhibit the growth of bacteria. The eugenol content in cloves can kill bacteria, including bacteria that are resistant to antibiotics (Andries et al., 2014).

Texture

The results of the analysis of variance showed that the interaction of treatment with the concentration of chitosan and clove solutions in table 3, showed no significant difference (P>0.05), but the interaction of treatment with the concentration of chitosan solution and clove solution can increase the texture organoleptic score from an average of 1.87 to 3.27.

Table 3. Mean Value Appearance Texture with treatment Concentration of Chitosan d an Cloves

Factor B (Clove)	Factor A (Chitosan)			Average
	K0 (0%)	K1 (1%)	K2 (2%)	
C0 (0%)	1.87±0.31	3.27 ± 0.31	3.27 ± 0.12	2.80±0.81
C1 (1%)	2.73±0.46	2.80±0.00	2.93±0.31	2.82±0.10
C2 (2%)	2.20±0.35	3.20±0.72	3.13±0.70	2.84±0.56
Average	2.27 ± 0.44 [a]	3.09 ± 2.25 [b]	3.11 ± 0.17 [b]	

description: [a, b] = Different superscripts in the same column showed a very significant difference (P<0.01)
Values represent the mean of 3 independent replicates ± standard deviation

There was also no significant difference in the clove solution concentration treatment (P>0.05) on the texture score of chicken meat, but the clove solution treatment could increase the texture value from an average of 2.80 to 2.84, while

the chitosan solution concentration treatment showed a very significant difference. significantly (P<0.01) on the texture score of chicken meat. Chitosan can maintain the texture of chicken meat for 9 h of storage by increasing the texture value from an average of 2.27 to 3.11.

Duncan's follow-up test of 1% showed that there was a significant difference between chicken meat with 0% chitosan treatment (without giving chitosan) and chicken meat with 1% and 2% chitosan treatment. The significant difference between chicken meat treated with 1% and 2% chitosan and chicken meat without chitosan indicated that the addition of chitosan gave better organoleptic results of the chicken meat texture. The highest value was found in the interaction of 1% chitosan (K1) and cloves 0% (C0) and 2% chitosan (K2) and cloves 0% (C0) with an average of 3.27 this indicates that chitosan is able to maintain the quality of chicken meat texture. . Hilma et al. (2018) stated that research has proven that the effect of 2% chitosan coating can maintain the physical characteristics of green grapes for 7 d.

pH

The results of the analysis of variance in Table 4, show that the interaction between the concentration of chitosan and cloves showed no significant difference (P>0.05). The average pH of the interaction treatment concentration of chitosan solution and clove solution ranged from 5.59 to 6.24 but in the treatment interactions, the pH was still in the normal standard of chicken meat. According to Laack et al. (2000), broiler meat quality can be seen by knowing the pH and total bacteria, with a normal pH of 5.96-6.0. The concentration treatment of clove and chitosan solution also had no significant difference (P>0.05) on the pH of chicken meat. The clove solution treatment slightly increased the pH from an average of 5.90 to 5.98 because there was clove content in it, while the chitosan concentration treatment gave a decreasing pH value with an average of 6.15 to 5.82 which indicated an acidic atmosphere.

This is because chitosan uses a solvent, namely 2% acetic acid, which causes the pH of chicken meat to decrease with the addition of chitosan. The high pH value in chicken meat greatly affects the water content, the higher the pH in the meat, the higher the water content of the meat. Conversely, the lower the water content in the meat, the higher the acid content or the lower the pH. According to Sitompul et al. (2015), that the amount of free water content in meat affects the durability of meat and increases the pH value.

Table 4 . Average pH with Chitosan and Clove Concentration Treatment

Factor B (Clove)	Factor A (Chitosan)			Average
	K0 (0%)	K1 (1%)	K2 (2%)	
C0 (0%)	6.24±0.34	5.59±0.09	5.88 ± 0.33	5.90±0.33
C1 (1%)	6.04±0.19	5.99±0.19	5.83±0.06	5.95±0.11
C2 (2%)	6.16±0.27	5.88±0.02	5.90±0.04	5.98±0.16
Average	6.15 ± 0.10 [b]	5.82 ± 0.20 [a]	5.87 ± 0.03 [a]	

Information: [a,ab, b] = Different superscripts in the same column showed significant differences (P<0.05)

Values represent the mean of 3 independent replicates ± standard deviation

A further test of 1% Honest Significant Difference (BNJ) showed that there was a significant difference between chicken meat treated with 0% chitosan (without chitosan administration) and chicken meat treated with chitosan 1% (K1) and 2% (K2). The significant difference between chicken meat treated with 1% and 2% chitosan and chicken meat without chitosan treatment was due to K1 and K2

treatments lowering the pH value of the sample. In accordance with the results of research by Rahardyani (2011) which showed that beef treated with chitosan had a lower pH value than the pH value of control beef. This is because chitosan has a positive charge which is chemically very reactive to bind hydroxyl ions (OH-). This binding process will cause the amount of dissociated OH- to be less because it is bound by the positive charge of chitosan so that it becomes undissociated (Fessenden and Fessenden, 1986). This causes the treatment with chitosan to have a lower pH value compared to only giving clove solution or without the addition of cloves or chitosan.

Water Content

The results of the analysis of variance in Table 5, showed that there was no significant difference (P>0.05) in the interaction of the concentration of chitosan solution and clove solution on the water content of chicken meat after storage at room temperature for 9 h. The average water content of the interaction treatment concentration of chitosan solution and clove solution ranged from 73.24 to 75.85. There was also no significant difference in the concentration of the chitosan solution and the treatment with the clove solution concentration (P>0.05). The high water content in this study was due to the high water content when cutting chicken meat. Forests et al. (1975) stated that the water content of broiler chicken meat is 65-85% so that the water content in the research results still meets the water content standard in chicken meat. Research Sharma et al. (2017) stated that the use of clove solution also did not show a significant effect on the average water content of fresh chicken sausage (P>0.05). Similar results have also been reported by Siewe et al. (2015) that there is no significant effect on the average water content of raw beef given clove solution.

Table 5 . Mean Water Levels by Treatment Concentration Ki t osan and Cloves (%)

Factor B (Clove)	Factor A (Chitosan)			Average
	K0 (0 %)	K1 (1%)	K2 (2 %)	
C0 (0 %)	73.85 ± 3.87	75.04±0.45	75.46 ± 0.20	74.78 ± 0.84
C1 (1 %)	75.85±0.97	75.54 ± 0.53	75.21 ± 0.34	75.53 ± 0.32
C2 (2 %)	73.24 ± 0.60	75.51 ± 0.18	73.98 ± 1.71	74.24 ± 1.15
Average	74.31 ± 1.37	75.36±0.28	74.88 ± 0.79	

Note: (ns) non-significant

The lowest water content was found in the interaction of treatment with 2% cloves (C2) and 0% chitosan (without giving chitosan solution) this is because cloves have an anti-microbial compound, namely eugenol which is able to suppress microbial growth so that it reduces the amount of water content, but for The overall water content in chicken meat is quite high and there is no significant difference, this is because the immersion of the meat with the edible coating will increase the water content of the chicken meat, besides the storage conditions and the amount of concentration also affect and cause insignificant water content. This is in accordance with Rukhana's research (2017) that the lowest water content is the control treatment compared to red chili (Capsicum annum L) which is dyed with edible coating.

Total Plate Count

The results of the analysis of variance showed that the interaction of the concentration of chitosan and cloves in Table 6 showed a very significant difference (P < 0.01) in the amount of TPC of chicken meat. The interaction of the concentration of the chitosan solution and the clove solution could reduce the number of bacterial

colonies from an average LOG value of 7.79 to 5.26. Treatment of concentration of chitosan solution showed a very significant difference (P<0.01) on the number of bacterial colonies of chicken meat. The concentration treatment of chitosan solution can reduce the number of bacterial colonies from an average LOG value of 6.88 to 5.26. This is supported by the opinion of Wittriansyah et al. (2019) that the use of chitosan is very effective in suppressing bacterial growth, the best result of the TPC test was obtained by giving 2% chitosan capable of inhibiting microbial growth from mullet by 2.7x106.

The concentration treatment of clove solution showed a very significant difference (P<0.01) to the number of bacterial colonies of chicken meat. Clove solution treatment can reduce the number of bacterial colonies from the LOG average value of 6.57 to 5.92.

Table 6 . Mean Value TPC (log CFU / g) with treatment Concentration Ki t osan and Cloves LOG

Factor B (Clove)	Factor A (Chitosan)			average**
	K0 (0%)	K1 (1%)	K2 (2%)	
C0 (0%)	7.79 ± 0.10 ^{c}	6.47 ± 0.30 ^{b}	5.44 ± 0.42 ^{ab}	6.57 ± 1.18 ^{b}
C1 (1%)	6.29 ± 0.44 ^{b}	6.20 ± 0.12 ^{b}	5.62 ± 0.15 ^{ab}	6.04 ± 0.37 ^{a}
C2 (2%)	6.54 ± 0.43 ^{b}	5.95 ± 0.28 ^{ab}	5.26 ± 0.24 ^{a}	5.92 ± 0.64 ^{a}
average**	6.88 ± 0.80 ^{c}	6.21 ± 0.26 ^{b}	5.26 ± 0.18 ^{a}	

Information: $^{a,ab, b, bc, c}$ = Different superscripts in the same column show a very significant difference (P<0.01)
Values represent the mean of 3 independent replicates ± standard deviation

A further test of 1% Honest Significant Difference (BNJ) showed that there was a significant difference in the interaction of the concentration of the chitosan solution and the clove solution. Treatment with 0% chitosan solution concentration and 0% clove solution (without soaking) showed a significant difference in the effect of other treatments. The difference in effect between chicken meat without soaking and chicken meat with soaking shows that the treatment with chitosan and cloves can suppress microbial growth in chicken meat after storage at room temperature for 9 h, supported by a further test BNJ 1% treatment with 0% chitosan concentration (without giving chitosan) showed a significant difference in the treatment of 1% chitosan solution concentration (K1) and 2% chitosan solution concentration (K2). The 1% BNJ further test also showed a significant difference between chicken meat with 0% clove concentration treatment (without giving cloves) to 1% clove solution (C1) and 2% clove solution (C2).

The interaction of treatment with 0% chitosan and 0% cloves (without soaking) was ranked the most bacteria with an average colony number of 6.3×107 CFU/g. The treatment interaction was able to suppress the growth of most bacteria so that it had the least number of bacteria, namely the treatment with the interaction of K2 and C2 with an average LOG value of 5.26. According to BSN (2009), the microbiological quality requirement (TPC) of chicken meat has a maximum requirement of 1×106 CFU/g. The research data showed that the interaction of K2 and C0 (P2) with a total number of colonies 3.6×105 CFU/g, the interaction of C1 and K2 (P5) with a total number of colonies 4.3×105 CFU/g, and the interaction of K2 and C2 (P8) with a total number of colonies 2×105 CFU/g still in accordance with the maximum standard for the total number of microbes. This proves that the administration of a solution of chitosan and cloves proved effective to suppress the growth of bacteria. Cloves produce the main component, namely eugenol which

functions as an antimicrobial (Andries et al., 2014). The results of other studies also showed that the treatment of chitosan coating on broiler chickens with concentrations of 1%, 2%, and 3% gave a significant effect ($P<0.05$) on the decrease in the total number of bacteria compared to meat without chitosan coating. The use of 3% chitosan showed the best inhibition (Alhuur et al., 2020). This is because chitosan is bacteriostatic, which means it can inhibit the growth of bacteria. The mechanism of chitosan in inhibiting bacterial growth is by damaging the structure of the bacterial cell wall. In gram-positive bacteria, the positive charge of chitosan is bound to the peptidoglycan layer which causes distortion and breakdown of the cell wall due to osmotic differences and exudation of cytoplasmic content. The mechanism of chitosan in gram-negative bacteria is by blocking the flow of nutrients in bacterial cells so that bacteria die due to a lack of nutrients (Damayanti et al., 2016).

CONCLUSION

The best treatment was interaction with 2% chitosan and 2% clove which was able to maintain the quality of the meat, both in terms of pH, water content, TPC value, with a total number of bacteria as much as 2×10^5 CFU/g and acceptability of organoleptic values which were still favored by panelists. This indicates the success of edible coatings in maintaining the quality and shelf life of chicken meat. Overall, the higher the concentration of chitosan and cloves, the lower the microbial contamination in chicken meat. The interaction of chitosan and cloves can suppress microbial contamination better.

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REFERENCES

Akhtar, J., Omre, P., & Azad, Z. R. A. A. (2015). Edible coating for preservation of perishable foods: A review. *Journal of Ready To Eat Food*, *2*(3), 81–88.

Alhuur, K. R. G., Juniardi, E. M., & Suradi, K. (2020). Efektivitas kitosan sebagai edible coating karkas ayam broiler. *Jurnal Teknologi Hasil Peternakan*, *1*(1), 17–24. https://doi.org/10.24198/jthp.v1i1.24093

Andries, J. R., Gunawan, P. N., & Supit, A. (2014). Uji efek anti bakteri ekstrak bunga cengkeh terhadap bakteri *Streptococcus mutans* secara in vitro. *E-GIGI*, *2*(2), 1–8. https://doi.org/10.35790/eg.2.2.2014.5763

AOAC. (1995). *Official Methods of Analysis of the Inc*. Arlington.

AOAC. (2005). *Official Methods of Analysis* (18th ed.). Association of Official Analytical Chemists inc.

Ariesthi, K. D. (2019). Analisis jumlah cemaran mikroba dan identifikasi salmonella sp dan escherichia coli pada daging ayam di beberapa tempat pemasaran wilayah Kota Kupang. *CHM-K Applied Scientific*, *2*(2), 416–423.

Awwaly, K. U., Triatmojo, S., Erwanto, Y., & Artama, W. T. (2015). Bioactive components in the meat and their functional properties : a literature study. *Jurnal Ilmu Dan Teknologi Hasil Ternak*, *10*(1), 22–34.

Ayustaningwaro, F. (2014). *Teknologi Pangan Teori Praktis dan Aplikasi*. Graha Ilmu .

Azizah, A., Suswati, I., & Agustin, S. M. (2018). Efek anti mikroba ekstrak bunga cengkeh (*Syzygium Aromaticum*) terhadap methicillin-resistant staphylococcus aureus (MRSA)secara in vitro. *Saintika Medika*, *13*(1), 31–35. https://doi.org/10.22219/sm.v13i1.5444

Badan Standardisasi Nasional. (2009). *Mutu Karkas dan Daging Ayam*. Badan Standardisasi Nasional.

Damayanti, W., Rochima, E., & Hasan, Z. (2016). Aplikasi kitosan sebagai antibakteri pada filet patin selama penyimpanan suhu rendah. *JPHPI*, *19*(3), 321–328.

Direktorat Jendral Perikanan Budidaya Kementrian Perikanan dan Kelautan. (2016). *Peta Sentra Produksi Perikanan Budidaya 2010-2015*. Buku statistik perikanan budidaya Indonesia.

Fardiaz S. (1992). *Mikrobiologi Pengolahan I*. Pusat Antar Universitas Pangan dan Gizi Institut Pertanian Bogor.

Fessenden, R., & Fessenden, S. (1986). *Kimia Organik*. Erlangga.

Forrest, J., Aberle, E., Hedrick, H., & Merkel, R. (1975). Principles of meat science. In *3rd ed. Dubuque, IA: Kendall/Hunt Publishing Co.London*.

Hakim, L. (2015). *Rempah dan Herba Kebun-Pekarangan Rumah Masyarakat*. Diandra Creative.

Harjanti, R. (2014). Kitosan dari limbah udang sebagai bahan pengawet ayam goreng. *Jurnal Rekayasa Proses*, *8*(1), 12–19. https://doi.org/10.22146/jrekpros.5018

Hilma, Fatoni, A., & Sari, D. (2018). Potensi kitosan sebagai edible coating pada buah anggur hijau (Vitis vinifera Linn). *Jurnal Penelitian Sains*, *20*(1), 25–29. https://doi.org/10.26554/jps.v20i1.497

Imbar, H. S., Harikedua, V. T., & Walalangi, R. G. (2016). Analisis organoleptik beberapa menu breakfast menggunakan pangan lokal terhadap pemenuhan kebutuhan gizi siswa sekolah dasar. *Jurnal GIZIDO*, *8*(1), 82–86. https://doi.org/10.47718/GIZI.V8I1.83

Jaelani, A. (2018). Jumlah mikroba dan sifat organoleptik daging ayam broiler yang direndam air perasan kunyit *(Curcuma Domestica VAL)* dengan lama penyimpanan yang berbeda. *Jurnal Ziraa'ah*, *43*(01), 85–95.

Nurhayati, Hanum, T., Rangga, A., & Husniati. (2014). Optimasi pelapisan kitosan untuk meningkatkan masa simpan produk buah-buahan segar potong. *Jurnal Teknologi Industri Dan Hasil Pertanian*, *19*(2), 161–178.

Nurjanah, Suwandi, R., & Yogaswari, V. (2010). Karakteristik kimia dan fisik sisik ikan gurami *(Osphronemus gouramy)*. *Jurnal Sumberdaya Perairan*, *4*(2), 7–12.

Oyedemi, S. O., Okoh, A. I., Mabinya, L. V., Pirochenva, G., & Afolayan, A. J. (2009). The proposed mechanism of bactericidal action of eugenol, A-terpineol and Y-terpinene against Listeria monocytogenes, Streptococcus pyogenes, Proteus vulgaris and escherichia coli. *African Journal of Biotechnology*, *8*(7), 1280–1286. https://doi.org/10.4314/AJB.V8I7.60106

Pratama, S. M., Barqly, G. J. K., Widyastuti, R., Wardani, R. N., Sielma, D. F., & Munawir, A. (2016). Pengaruh lama perendaman terhadap absorpsi tetrasiklin pada adsorben limbah sisik ikan gurami *(Osphronemus Gouramy)*. *Majalah Kedokteran Gigi Indonesia*, *20*(2), 161–166. https://doi.org/10.22146/majkedgiind.9231

Rukhana, I. (2017). *Pengaruh Lama Pencelupan dan Penambahan Bahan Pengawet Alami dalam Pembuatan Edible Coating Berbahan Dasar Pati Kulit Singkong Terhadap Kualitas Pasca Panen Cabai Merah (Capsicum annum L)*. Universitas Islam Negri Maulana Malik Ibrahim.

Rumengan, I. F., Suptijah, P., Salindeho, N., Wullur, S., & Luntungan, A. (2018). *Nanokitosan dari Sisik Ikan: Aplikasinya Sebagai Pengemas Produk Perikanan*. Lembaga Penelitian dan Pengabdian Kepada Masyarakat Universitas Sam Ratulangi.

Saraswati, D. (2014). Aktivitas bubuk bunga cengkeh *(Eugenia aromatic)* terhadap kepekaan bakteri escherichia coli. *Jurnal Entropi*, *9*(1), 721–840.

Sharma, H., Mendiratta, S. K., Agrawal, R.

K., Gurunathan, K., Kumar, S., & Singh, T. P. (2017). Use of various essential oils as bio preservatives and their effect on the quality of vacuum packaged fresh chicken sausages under frozen conditions. *LWT - Food Science and Technology*, *81*(1), 118–127. https://doi.org/10.1016/j.lwt.2017.03.048

Siewe, F. B., Mbougueng, P. D., Tatsadjieu, L. N., Noumo, T. N., & Mbofung, C. M. F. (2015). The potential application of essential oils as natural preservatives of beef patties. *Food and Nutrition Sciences*, *06*(03), 374–385. https://doi.org/10.4236/fns.2015.63038

Sitompul, M., Siswosubroto, E., Rumondor, D., Tamasoleng, M., & Sakul, S. (2015). Penilaian kadar air, ph dan koloni bakteri pada produk daging babi merah di Kota Manado. *ZOOTEC*, *35*(1), 117–130. https://doi.org/10.35792/zot.35.1.2015.7108

Suptijah, P., & Gushagia, Y. (2008). Study of Inhibitory effects of chitosan on quality deterioration of catfish *(Pangasius hypopthalmus)* fillet at room temperature storage. *Buletin Teknologi Hasil Perikanan*, *11*(2), 89–101. https://doi.org/10.17844/JPHPI.V11I2.907

Susanti, N., & Purwanti, A. (2020). Pembuatan kitosan dari limbah sisik ikan (variabel konsentrasi larutan NaOH dan waktu ekstraksi). *Jurnal Inovasi Proses*, *5*(1), 40–45.

Tinangon, R., Djalal, R., Lilik, E. R., & Purwadi. (2017). Senyawa bioaktif cengkeh (Zyzygium Aromaticum) dapat menghambat pertumbuhan mikroba pada daging burger. *Seminar Nasional Peternakan 3 Universitas Hasanuddin*, 297–299.

Towaha, J. (2012). Manfaat eugenol cengkeh dalam berbagai industri di Indonesia. *Perspektif*, *11*(2), 79–90.

Van Laack, R. L. J., Liu, C.-H., Smith, M. O., & Loveday, H. D. (2000). Characteristics of pale, soft, exudative broiler breast meat. *Poultry Science*, *79*(7), 1057–1061. https://doi.org/10.1093/ps/79.7.1057

Winarti, C., Miskiyah, & Widaningrum. (2013). Teknologi produksi dan aplikasi pengemas Edible antimikroba berbasis pati. *Jurnal Penelitian Dan Pengembangan Pertanian*, *31*(3), 85–93. https://doi.org/10.21082/jp3.v31n3.2012.p%p

Wittriansyah, K., Soedihono, S., & Satriawan, D. (2019). Aplikasi kitosan emerita sp. sebagai bahan pengawet alternatif pada ikan belanak (Mugil cephalus). *Jurnal Ilmiah Perikanan Dan Kelautan*, *11*(1), 34–42. https://doi.org/10.20473/jipk.v11i1.12458

Wulandari, W. T., Nurzaman., Pratita, A. T. K., & Idacahyati, K. (2020). Pengaruh variasi konsentrasi NaOH terhadap nilai derajat deasetilasi kitosan dari limbah cangkang kerang hijau *(Perna viridis L). KOVALEN: Jurnal Riset Kimia*, *6*(3), 171–176. https://doi.org/10.22487/kovalen.2020.v6.i3.15277

Yusuf, F., Husain, R., & Naiu, A. S. (2020). Mutu organoleptik ikan selar *(Caranx Leptolepis)* segar yang dilumuri bubuk cengkeh *(Syzygium Aromaticum)* selama penyimpanan. *Jambura Fish Processing Journal*, *1*(2), 1–11. https://doi.org/10.37905/jfpj.v1i2.5423