Addition of andaliman to shelf life of beef nugget

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Abstract. This study aims to identify the antimicrobial properties found in andaliman and test to the shelf life of beef nuggets containing andaliman. This study method uses an exploratory method which begins with andaliman extraction and identification of antibacterial compounds. The next stage is the manufacture of beef nuggets that are treated (0; 0.5; 1; 1.5) % andaliman. The observed research parameters included: phytochemical test and contamination test (microbiology). This study uses Factorial 4x3 Completely Randomized Design (CRD) with 2 factors: factor 1: andaliman concentration and factor 2: shelf life. The result showed that Andaliman has antimicrobial properties. The 1.5% Andaliman concentration in beef nuggets (N3H0) has the best inhibitory power, because the andaliman beef nugget has the highest inhibitor when compared with other treatment.

1. Introduction

Good Manufacturing Practices (GMP) dan Good Hygienic Practice (GHP) are guidelines for handling meat to safe processing and suitable meat. If it is not handled properly, meat will be rotten because meat is known as perishable food and it has the potential hazardous foods (PHF). Type of meat are often processed are beef, chicken, lamb and pork. Preparation of meat processing are selecting materials, hygienizing equipment, cleaning location and handling of waste. Some meat product such as meatball, nugget and corned beef needs several stages before safe to eat. The stages of making the nugget are selecting any ingredients, meat grinding, mixing, patterning nugget, boiling, freezing, and finally frying the frozen nugget. Nugget still stored in the freezer before used. This is done because to prevent nugget from being contaminated with bacteria by the influence of decrease the temperature. Salmonella and Escherichia coli (E. coli) are microbes that can infects meat products. The food industries uses artificial compound to extend storability their products. This method is not good to consumer for a long term. Another methods can be used to safe meat processing. One of them is the addition of ingredients that can increase shelf life.

Andaliman (Zanthoxylum acanthopodium DC) is a type of herb that has antimicrobial properties. These antimicrobial have the potential to reduce risk of contamination in meat products. Generally, andaliman is used as spice for cooking fish. Andaliman has a unique taste and aroma. Addition of andaliman in meat product can strengthen the taste of food.

This study aims to identifying aromatic compounds that have the antimicrobial properties in andaliman and test the durability of andaliman beef nuggets from bacterial contamination. The phytochemical test such as flavonoid, alkaloid, terpenoid, steroid, tanin, and saponin.
2. Materials and Methods

2.1. Experimental design

The experiment was conducted in animal production laboratory, Agricultural Faculty and microbiology laboratory, Math and Science Faculty, Universitas Sumatera Utara, Medan, Indonesia. The study lasted for 4 months from May to September 2018. Materials consisting of beef, andaliman flour, wheat flour, tapioca flour, starch, salt, pepper, nutmeg powder, garlic, egg, bread crumb, ice, water. The tools were wrapping plastic, plastic/rubber gloves, mask, head cover, cake mold, stove, mixer, etc. The research method used was complete randomized design (RAL) with 2 factor, 4 treatments/factor and 3 replications. The treatment given as follows : [N0;N1;N2;N3] = Nugget with [0%; 0.5%; 1%; 1.5%] Andaliman and [H0;H1;H2;H3] = [0;1;2;3] days after nuggets are made.

Parameter of research were phytochemical test, quantitative analysis Total Plate Count (TPC), quantitative analysis of Salmonella and E.Coli.

3. Results and discussion

Phytochemical screening. The result of the qualitative analysis of second metabolite of andaliman extract using methanol showed positive on alkaloid, terpenoid, saponin and negative on flavanoid, steroid and tanin. The results shown in the Table 1.

| Reagent     | Flavonoid | Alkaloid | Torpedoed | Steroid | Tannin | Saponin |
|-------------|-----------|----------|-----------|---------|--------|---------|
| FeCl3 5%    | -         |          |           |         |        |         |
| NaOH 10%    | -         |          |           |         |        |         |
| H2SO4(p)    | -         |          |           |         |        |         |
| Mg-HCl      | -         |          |           |         |        |         |
| Bouchardart | -         |          |           |         |        |         |
| Dragendorff | -         |          |           |         |        |         |
| Maeyer      | +         |          |           |         |        |         |
| Salkowsky   | +         |          |           | -       |        |         |
| CeSO4 1% dalam | - |          |           |         |        |         |
| H2SO4 10%   | -         |          |           |         |        |         |
| Aquadest+Alkohol | 96%+  |     |           |         |        |         |
| HCl 2N      | +         |          |           |         |        |         |

Description : (-) = not detected by second metabolites, (+) = detected second metabolites.

Table 1 show that andaliman extract has 3 active component such as alkaloid, terpenoid and saponin. Alkaloid is active component in most types of plants. Alkaloids can be attracted to ethanol solvents because alkaloids are polar. The positive reaction that occurred in the alkaloid test was formed orange deposits on the dragendorff reagent and yellow precipitate in Maeyer reagent, because the ligan turnover reaction. Alkaloids that have nitrogen atoms that have free electron pairs can replace the iodo ion in the reagents. Terpenoid is known as the main compound in plants that are essential for composing essential oils. Andaliman extract has contain much essensial oils. Saponins are generally in the form of glycosides so that they are generally polar and are surface active compounds that can cause foam if shake in the water. The foam in the test occurs because saponins have polar groups and non-polar groups which will form micelles. A micelle is formed causing the polar group to face out and the nonpolar group facing inward and this is what looks like foam. Saponins can be antibacterial because their surface active substances are similar to detergents, as a result saponins will reduce the surface tension of bacterial cell wall and damage membrane. This damage to cell membranes greatly
disrupts the bacteria. Saponins diffuse through the outer membrane and susceptible cell walls then bind to the cytoplasmic membrane and reduces the stability of the cell membrane. Microbiology quality of foods are determined by the number and type of microorganism. So, it will be determine the shelf life of the product from contamination and food safety. The total plate count test results on beef nuggets can be seen as follows (Table 2).

**Table 2.** Total Plate Count test on Andaliman Beef Nugget

| Concentration (%) | Days |
|-------------------|------|
|                   | 0    | 1    | 2    | 3    |
| 0                 | 166 x 10^5 | 2,877.33 x 10^5 | 3,074.67 x 10^5 | 3,762.00 x 10^5 |
| 0.5               | 67.67 x 10^5 | 2,682.67 x 10^5 | 2,690.67 x 10^5 | 3,512.00 x 10^5 |
| 1                 | 34.67 x 10^5 | 2,302.67 x 10^5 | 2,524.00 x 10^5 | 3,086.67 x 10^5 |
| 1.5               | 12.67 x 10^5 | 1,894.67 x 10^5 | 2,176.00 x 10^5 | 2,556.00 x 10^5 |

Table 2 shows that the difference in the andaliman concentration influence number of microbes. This difference is seen in the 0th day observations. The higher andaliman concentration was reduce the total number of bacteria on the media plate count agar (PCA). However, the observations after 0 until 3 days have the maximum of microbial contamination. The number of bacteria can be affected by conditions of materials, processing, packaging, storage, distribution and temperature. Microbes have maximum growth at high temperatures, especially at the optimum temperature. High temperature can cause enzyme denaturation. Optimum temperature can increase microbial activity. The oxygen concentration of food and the environment was affected food microorganisms. Non-airtight packaging was determined the microbial growth found in beef nuggets, especially aerobic microbes. Although, some microorganisms are heterotrophic. These bacteria need nutrients for their lives and growth. While the content of beef protein nuggets is very high so that it becomes a good substrate for growth.

The shelf life of beef nuggets on day 0 with 0% andaliman flour (Control) was a positive result in the E. coli test as many as 6 colonies with the characteristics of a metallic green colony on eosin methylene blue (EMB) medium while Salmonella sp. 5 colonies with black colony characteristics on salmonella shigella agar (SSA) media (Table 3). While giving andaliman flour with different concentrations (N1H0, N2H0, N3H0) there were no contamination of E. coli and Salmonella sp. The addition of antibacterial compound concentrations is thought to increase the penetration of antibacterial compounds into the inside of microbial cells which will damage the cell's metabolic system and can cause cell death.

**Table 3.** Contamination of E. coli and Salmonella sp at 0 day

| Concentration (%) | E. coli | Salmonella sp. |
|-------------------|---------|----------------|
| N0H0              | 6       | 5              |
| N1H0              | -       | -              |
| N2H0              | -       | -              |
| N3H0              | -       | -              |

Alkaloids as antibacterials are by interfering with the constituent components of peptidoglycan in bacterial cells, so that the cell wall layer is not formed intact and causes cell death. While saponin is a form of glycosides which have aglycones in the form of steroids and triterpenes. The mechanism of action of saponin as an antibacterial that can cause leakage of proteins and enzymes in the cell. Saponins can be antibacterial because their surface active substances are similar to detergents, consequently saponins will reduce the surface tension of bacterial cell walls and damage membrane permeability. This damage to cell membranes greatly disrupts the survival of bacteria. Saponins diffuse through the outer membrane and susceptible cell walls then bind to the cytoplasmic membrane so that it disrupts and reduces the stability of the cell membrane.
4. Conclusions
Andaliman has antimicrobial properties. Andaliman has potential to inhibit microbial growth because it active component content consisting of terpenoids, alkaloids and saponins. 1.5% Andaliman concentration in beef nuggets (N3H0) has the best inhibitory power, because the andaliman beef nugget has the highest inhibitor when compared with other treatment.

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Acknowledgments
The research was funded by the Research Central of Universitas Sumatera Utara, Medan for Internal Grand Research (Talenta 2018 Number: 2590/UN5.1.R/PPM/2018 at 16th March 2018).