

## Microbial Analysis of Chicken “Urutan” Traditional Food with Different Smoke Processes

A.A. Made Semariyani<sup>1</sup>, I Wayan Sudiarta<sup>2</sup>, Ni Made Ayu Suardani Singapurwa<sup>3\*</sup>, Luh Suariani<sup>4</sup>, Agata Viani<sup>5</sup>, Sang Ayu Made Agung Prasetiawati Djelantik<sup>6</sup>

Fakultas Pertanian Universitas Warmadewa, Denpasar, Bali

**Corresponding Author:** Ni Made Ayu Suardani Singapurwa

[a.suardani@gmail.com](mailto:a.suardani@gmail.com)

---

### ARTICLE INFO

*Keywords:* Shelf Life, Preservation, Smoking Technique

*Received :* 12 July

*Revised :* 13 August

*Accepted:* 18 September

©2023 Semariyani, Sudiarta, Singapurwa, Suariani, Viani, Djelantik: This is an open-access article distributed under the terms of the [Creative Commons Atribusi 4.0 Internasional](https://creativecommons.org/licenses/by/4.0/).



### ABSTRACT

The chicken sequence is a processed meat product that is processed by making a complete Balinese meat, fat, and seasoning mixture and then put into a sleeve. This study aimed to investigate the impact of varying concentrations of coconut shell liquid smoke on the sequence features of chickens. The method used in this study is the descriptive analysis of microbial growth observations. The results showed that the use of coconut shell liquid smoke with a concentration of 4% could maintain chicken “urutan” for up to 20 days of storage at low temperatures (5+1 oC) with total microbial characteristics of  $2.08 \times 10^7$  CFU/g, *Escherichia coli* < 10<sup>3</sup> CFU/g and Yeast mold 0 CFU/g (meet the requirements SNI 01-3820-2015)

## **INTRODUCTION**

Food preservation techniques are used by inhibiting the growth of microorganisms that cause food spoilage. One solution that can be used is pickling with smoke (Alamsyah et al, 2021; Anshari, 2009). Smoking is the process of withdrawing water and chemically depositing it from smoke. In addition, the smoking process also combines the process of salting, heating and attaching the chemical components of the smoke (Assidiq et al, 2018). Smoking aims to preserve a food product by removing the water content in the product, so as to inhibit the growth of pathogenic organisms in order to extend shelf life. The smoking process is divided into two, namely traditional smoking and modern smoking using liquid smoke (BSN,2015; Budijanto et al, 2008)). Traditional smoking techniques require certain equipment, are complicated and require quite a long time. For the formation of distinctive taste, color and aroma in smoking products it is easier to use the liquid smoking method. The advantage of using liquid smoke is that it is easier to apply and control to produce a uniform product (Darmadji, 2009; Dewi et al, 2018).

Chemical compounds contained in smoke are formic acid, acetic, butyric, caprylic, vanillic, syringic acid, dimethoxyphenol, methyl glyoxal, furfural, methanol, ethanol, octanol, acetaldehyde, diacetyl, acetone, and 3,4-benzpyrene. The alcohols and acids originate from the decomposition of cellulose and hemicellulose at lower temperatures than lignin (Fausan and Ikhwanus, 2017; Fitriani, 2009). The formaldehyde contained in the smoke has a great preservative effect. Phenol in smoke has a function as an antioxidant that can inhibit oxidative rancidity. According to Fitriyani (2009) liquid smoke is a result of pyrolysis of plants or wood at a temperature of around 400°C. Liquid smoke is currently extensively utilised by the food industry to impart a particular aroma, texture, and flavour to various culinary products, including but not limited to meat, fish, and cheese. The preservation of food ingredients can be attributed to the acid, phenolic, and carbonyl chemicals found in liquid smoke. The advantage of using liquid smoke as a preservative is that it is able to maintain the levels of protein and fat contained in the material, when compared to other preservatives (Girard, 1992; Guillen et al., 2000).

Chicken sequence is a processed meat product that is processed by making a complete Balinese meat, fat and seasoning mixture (“Basa Genep”) (Semariyani and Sudiarta, 2018; Nggoweng et al., 2020). Then it is put into the sleeve (synthetic casing). Under normal conditions (room temperature) the average durability of the chicken order is 1-2 days. If it is more than this limit, the taste of the chicken stock will become rancid and rotten making it unfit for consumption and sometimes mushrooms will grow depending on the level of maturity when fried. Based on the description above, research on smoking techniques, smoking time, concentration of liquid smoke and soaking time will be carried out on the characteristics and shelf life of chicken “urutan”, to determine the effect of smoking technique and duration on the characteristics of chicken “urutan”.

## LITERATURE REVIEW

Fumigation in pickling aims to preserve and give the product a distinctive color and smoked taste. Actually, the durability caused by smoke is very limited, so that meat can be preserved for a long time, it must be followed or preceded by other methods of preservation. Smoking also aims to remove steam from the elements of phenolic or aldehyde compounds from the type of wood attached to the product or to incorporate these elements into the product so as to produce a distinctive taste and aroma, as well as dry the product so that the expected preservative effect is obtained. The delicious taste that characterizes smoked products is obtained mainly from phenol and aldehyde compounds Dewi et al. (2018); Fausan and Ikhwanus, (2017). The phenol element melts into the fat present in the product section and controls the automatic oxidation of this fatty portion, thus preventing the reddish discoloration of the final product (Himawan, 2010).

Elements in the smoke, which are effective in preventing the growth of microorganisms are aldehydes, phenols and organic acids. States that smoke compounds can reduce the pH of the product surface thereby making the smoke product environment less favorable for most bacteria. It is also said that the formation of color during smoking is thought to be caused by the Maillard reaction in which the smoke component plays a dominant role. The anti-bacterial substance in the aldehyde element is very strong (Huseinsyah and Mosthapa, 2011). Because the compounds contained in the smoke containing these antibacterial substances do not get into the product, the anti-spoilage effect is only around the surface of the product. In cold smoking, the heat generated from the smoke does not have much effect on the fish (Megasari, 2020; Mohamed and Ahmed, 2016). So the smoking time must be long because the distance between the smoke source and the fish is quite far. Due to the long smoking, sometimes the product becomes hard like wood. In hot smoking, the distance between the product and the smoke source is usually close. Then the temperature is high enough, so the product cooks quickly. High heat can stop unwanted enzyme activity, agglomerate proteins, and evaporate some of the water in the fish's body, so that the shelf life of the product can be increased. the drying process occurs during smoking, the reduction in water content together with the preservative power of the smoke, so that smoking has the power to kill bacteria (bactericidal), whose strength depends on the amount of smoke absorbed (Muhammad, 2018; Setha, 2011).

In general, there are two smoking methods that have been used for a long time, namely hot smoking and cold smoking. But today in developed countries it has been developed using electricity (electric smoking). To save time and energy, the use of liquid smoke has also been developed, namely by dipping the product into a solution of smoke ingredients (smoke concentrate) after which it is then dried. This liquid smoke is obtained from the dry distillation of wood smoke. Experiments are still being carried out to find the type of liquid smoke that can provide satisfactory results without causing adverse consequences for consumers. In addition, excessive handling during smoking contributes to the fragility of the smoked product (Simon et al, 2005; Siskos et al., 2007) so it is

recommended to use trays so that they can be easily moved to different parts of the smoking area so that all products receive uniform smoking (Soeparno,S, 2009; Soldera et al., 2008). Furthermore, it will be explained below, there are five types of smoking processes, namely, cold smoking, warm smoking, hot smoking, liquid smoke, and electric smoking.

## **METHODOLOGY**

This research will be carried out at the Laboratory of the Faculty of Agriculture, University of Warmadewa, and at the Denpasar Veterinary Laboratory. The research was conducted from January - August 2023 for the first year. The research method was carried out in several stages. The research was conducted by sequential fumigation with coconut shell liquid smoke with a concentration of 2%, 4%, 6% and storage time for 5, 10, 15, and 20 days. Observations were made every fiutilization each treatment and the data was analyzed statistically to see if there were any real differences between treatments. Data is displayed in tabular form.

### **Observation Objectively**

Total Plate Count Test, Escherichia coli and Mold (BSN, 2015; Budijanto et al, 2008)

The MPN (Most Probable Number) method is a technique for quantifying microorganisms. It involves analyzing the growth of microorganisms in a designated liquid medium within a series of tubes. These tubes are inoculated with solid or liquid samples that are either undiluted or diluted based on the desired concentration. By observing the number of positive growths in the tube series, the MPN value per unit of sample volume or mass can be determined, providing a range of microorganism quantities. Estimator Test 1 ml of water sample is inoculated into 5 ml of tube containing LB medium, then 0.1 mL of water is inoculated into 1 tube of LB medium, and after that 0.01 ml of water sample is inoculated with 1 tube of LB medium. All tubes were incubated in an incubator at 37°C for 24 hours. Observations were recorded if gas or medium was formed in the Durham tube, which was cloudy yellow, which means it was positive. Tubes that were positive on the assay for bacteria/mold were compared to the MPN table.

### **Data Analysis**

The quantitative analysis of the data collected in this study was conducted using analysis of variance (ANOVA). If the data acquired demonstrates a statistically significant disparity, it is recommended to proceed with the utilisation of the Duncan's Multiple Range Test (DMRT). In 2015, statistical analysis was conducted using the SPSS programme for Windows versions 22. The acquired data was subjected to descriptive analysis and afterwards presented in tabular format.

## RESULTS AND DISCUSSION

### Total Microbes (Total Plate Count)

Table 1 displays the outcomes of the TPC examination conducted on the chicken sequence samples across all treatments. Table 1 presents the relationship between the concentration of liquid smoke and the number of microorganisms in the chicken order, indicating that an increase in liquid smoke concentration results in a decrease in microbe count. The presence of phenolic components in liquid smoke serves as an antioxidant, hence enabling it to effectively limit sausage damage. By supplying hydrogen, liquid smoke is able to effectively reduce fat auto-oxidation and subsequently minimise the detrimental effects of oxygen-induced fat oxidation. The presence of acid in liquid smoke exhibits notable efficacy in eradicating and impeding the proliferation of microorganisms in sausages. This is primarily achieved through the ability of acidic compounds to permeate the cellular membranes of microorganisms, leading to their lysis and subsequent demise. By reducing the bacterial population in food products, the detrimental impact caused by microorganisms can be effectively restrained. The acid content is a chemical property that plays a crucial role in determining the quality of the liquid smoke generated.

Table 1. Test Results for Total Plate Count, E.coli and Mold “Urutan” of Chicken in Concentration Treatment of Coconut Shell Liquid Smoke and Long Low Temperature Storage

Treatment Liquid smoke concentration and Storage time	Total Plate Count (CFU/g)	<i>E.coli</i> (CFU/g)	Mold (CFU/g)
2%, 5 days	2.4 x10 <sup>3</sup> l	1.5x10 <sup>1</sup>	0
4%, 5 days	5.9 x10 <sup>4</sup> j	7.9x10 <sup>2</sup>	0
6%, 5 days	1.9 x10 <sup>4</sup> k	9.1x10 <sup>2</sup>	0
2%, 10 days	3.7 x10 <sup>4</sup> h	2.0x10 <sup>1</sup>	0
4%, 10 days	7.3 x10 <sup>4</sup> g	2x10 <sup>1</sup>	0
6%, 10 days	1.08 x10 <sup>5</sup> c	2.2x1x100 <sup>1</sup>	0
2%, 15 days	1.2 x10 <sup>7</sup> e	1.7x10 <sup>1</sup>	0
4%, 15 days	3.1.x10 <sup>6</sup> i	9.5x10 <sup>2</sup>	0
6%, 15 days	2.7 x10 <sup>8</sup> a	5.6x10 <sup>2</sup>	0
2%, 20 days	2.1 x 10 <sup>8</sup> b	0	0
4%, 20 days	2.08x10 <sup>7</sup> d	0	0
6%, 20 days	8.2x10 <sup>6</sup> f	0	0

Note: The mean value followed by the same letter in the same column shows no significant difference at the Duncant test level of 5%

The acid compounds formed from the wood burning process are organic acid compounds which are located from the pyrolysis of wood components such as hemicellulose and cellulose at a certain temperature. Liquid smoke can be used as a preservative if it contains phenolic and acid compounds which act as antibacterial and antioxidant (Himawati, 2010). Based on SNI 01-3820-2015, the maximum total plate number in sausages is a maximum of 105 CFU/g where the number of microorganisms produced from chicken "urutan" is still within safe limits for consumption.



Figure 1. "Urutan" Traditional Food of Chickens

According to Soldera et al. (2008) recommend that the number of bacteria in good fresh meat is  $< 10^4$  CFU/g and the number of critical bacteria is between  $> 10^4 - 10^5$  CFU. The results of research by Assidiq et al. (2018) stated that the use of coconut shell liquid smoke with a concentration of 0.25% - 6.0% was able to inhibit the growth of the fungi *Collectichum gloeosporoides* and *Fusarium oxysporum* colonies. In testing liquid smoke with a concentration of 0; 0.2; 0.6; 0.8; and 1% for four bacterial cultures on TSB media with an initial population of 108 CFU/g, after 24 hours of incubation showed that the growth of *Staphylococcus aerus* and *Pseudomonas fluorescens* was inhibited at a concentration of 0.6%. At the same time, *Escherichia coli* and *Bacillus subtilis* began to inhibit at a concentration of 0.8% (Siskos et al., 2007). Darmadji, 2009 states that liquid smoke with a concentration of 0.6% causes a decrease in the pH of TSB (Trypticase Soy Broth) to 6.36. The TSB pH of 6.36 caused a normal decrease in four bacterial cultures where the minimum growth pH for *E.coli*, *S. aureus*, *P. fluorescens*, and *B. subtilis* was 4.4: 4.0, respectively; 5,6 ; 4,9.

The addition of 0.6% liquid smoke has inhibited the growth of *S. aureus* and *P. fluorescens*, this indicates that the inhibition of the growth of these two bacteria is not caused by a decrease in the pH of their living environment but by the activity of antimicrobial compounds in coconut shell liquid smoke. Likewise the growth of *E. coli* and *B. subtilis* at the addition of 0.8% smoke concentration. The decrease in the number of *E. coli* and *B. subtilis* at the addition of 1% liquid smoke concentration was caused by the activity of antimicrobial compounds in liquid smoke (Siskos et al, 2007). The compounds present in smoke are substances that have bacteriostatic and bactericidal properties. Phenolic chemicals and acetic acid are examples of antibacterial agents. According to Darmadji (2009), the growth of *Pseudomonas fluorescens* bacteria, *Bacillus subtilis*, *Escherichia coli*, and *Staphylococcus aureus* was inhibited by smoke at a dilution of 10 times. The identification of *E. coli* bacteria within the chicken sample is likely attributed to the introduction of contaminants through the utilisation of tools during the manufacturing procedure. According to Darmadji (2009), the presence of Coliform in food preparation facilities can be attributed to its introduction via raw materials, subsequently leading to its contamination of cooked food by various means such as contact with hands, surfaces of utensils, cooking surfaces, and other equipment.

A negative correlation was seen between the concentration of coconut shell liquid smoke and the population of *E. coli*, indicating a drop in the latter as the former increased. After a storage period of 20 days, no presence of *E. coli* bacteria, which could potentially contaminate the sample, was detected. The observed phenomenon can likely be attributed to the presence of phenolic chemicals and acetic acid in coconut shell liquid smoke, which have been shown to inhibit the proliferation of *E. coli* bacteria under refrigerated conditions.

## **CONCLUSIONS AND RECOMMENDATIONS**

The use of coconut shell liquid smoke with a concentration of 4% can maintain the sequence characteristics of chickens up to 20 days of storage with a total of  $2.08 \times 10^7$  CFU/g microbes, *E. coli* <103 CFU/g and Yeast mold 0 CFU/g

## **FURTHER STUDY**

Further research was conducted to observe the characteristics and proximate values of the cured chicken “urutan” by smoking.

## **ACKNOWLEDGMENT**

Thank you to the Chancellor, Dean, Head of the Research Institute, Head of the Food Science and Technology Study Program, Head of the Laboratory, and to Laboratory Assistant at Warmadewa University, so that this research can run smoothly

## **REFERENCES**

- Alamsyah, F.H., Irfan, dan D. Yunita. 2021. Penggunaan Asap Cair Sebagai Pengawet Alami Bakso Ikan Tuna Sirip Kuning (*Thunnus albacares*). Banda Aceh. 12 (1) : 106-112.
- Anshari, D. 2009. Impregnasi Asap Cair Tempurung Kelapa, Poliester Tak Jenuh Yukalac 157 BQTN-EX dan Toluena Diisiosianat Terhadap Kayu Kelapa Sawit. Tesis. Medan: Sekolah Pasca Sarjana Universitas Sumatra Utara.
- Assidiq, F., T. D. Rosahdi, dan B. V. E. Viera. 2018. Pemanfaatan Asap Cair Tempurung Kelapa Dalam Pengawetan Daging Sapi. Bandung. Volume 5 (1): 34-41
- Badan Standardisasi Nasional. 2008. SNI 2897:2008 tentang Metode Pengujian Cemarkan Mikroba dalam Daging, Telur dan Susu, serta Hasil Olahannya. Departemen Pertanian, Jakarta.
- Badan Standardisasi Nasional. 2015. SNI 01-3820-2015: Syarat Mutu Sosis Daging .Departemen Pertanian. Jakarta
- Budijanto, S., R. Hasbullah, S. Prabawati, Setyadjit, Sukarno, dan I. Zuraida. 2008. Identifikasi dan Uji Keamanan Asap Cair Tempurung Kelapa untuk Produk Pangan. Jurnal Pascapanen, 5(1): 32-40
- Darmadji P, 2009. Teknologi Asap Cair dan Aplikasinya pada Pangandan Hasil Pertanian. Pidato Ilmiah Pengukuhan Guru Besar. Universitas Gajah Mada. Yogyakarta
- Dewi, J., A. Gani, dan M. Nazar. 2018. Analisis Kualitas Asap Cair Tempurung Kelapa dan Ampas Tebu Sebagai Bahan Pengawet Alami Pada Tahu. Banda Aceh. Jurnal IPA dan Pembelajaran IPA. 2 (2): 106-112
- Fausan F dan M. Ikhwanus. 2017. Pemurnian Asap Cair Tempurung Kelapa melalui Destilasi dan Filtrasi Menggunakan Zeolit dan Arang Aktif. Prosiding Seminar Nasional Sains dan Teknologi

- Fitryani, F.2009. Pengaruh Asap Cair Selama Penyimpanan Dingin Terhadap Kualitas Kimia dan Mikrobiologis Sosis Sapi.
- Girard, J.P. 1992. Technology of Meat and Meat Production. Ellis Horwood. New York
- Guillen, M.D., P. Sopelana and M.A. Partearroyo. 2000. Polycyclic aromatic hydrocarbons in liquid smoke flavorings obtained from different types of wood, effect of storage in polyethylene flasks on their concentrations. J Agric Food Chem. 48:5083-6087.
- Himawati, E. 2010. Pengaruh Penambahan Asap Cair Tempurung Kelapa Destilasi dan Redestilasi Terhadap Sifat Kimia, Mikrobiologi dan Sensoris Ikan Pindang Layang (*Decapterus spp*) Selama Penyimpanan. Skripsi. Solo: Universitas Sebelas Maret.
- Husseinsyah, S. and Mosthapa, M. 2011. The Effect of Filter Content on Properties of Coconut Shell Filled Polyester Composites. Malaysian Polymer Journal. 6(1): 87-97
- Megasari, R. 2020. Analisis Kandungan Kimia Asap Cair Dari Tempurung Kelapa dan Sabut Kelapa Dengan Metode Destilasi. Journal of Agritech Science. Gorontalo. 4 (2): 61-68
- Mohamed, A.A. and S.H. Ahmed. 2016. Effect of Drying on proximate and microbial analysis of dried *Clarias sp.* and *Tilapia sp.* Collected from Gedaref market, Gedaref state, Sudan. International Journal of Current Microbial and Applied Science, 5(6): 79-84
- Muhammad, F. 2018. Kajian Kadar Air Tempurung Kelapa Terhadap Kualitas Keasaman Produksi Asap Cair. Jurnal Penelitian Mahasiswa Teknik Sipil dan Teknik Kimia. Malang: Universitas Tribhuwana Tunggaladewi. Halaman 202- 207.
- Nggoweng, S., Singapurwa, N.M.A.S., Rudianta, N. 2020. "Base Genep" Instant Spices Quality for Sale In East Denpasar. Sustainable Environment Agricultural Science (SEAS). 4(2):142-150
- Semariyani, A.A..M dan I Wayan Sudiarta. 2018. Keamanan Pangan Urutan Ayam Ditinjau dari Cemaran Mikroba. Warmadewa. Denpasar
- Setha, B. 2011. Pengaruh Penggunaan Asap Cair terhadap Kualitas Fillet Ikan Cakalang Asap. Jurnal Ilmu Pengetahuan dan Teknologi. (1):28-37
- Simon R, Calle B, palmer S, Meler D, Anklam E.2005. Composition and Analysis of Liquid Smoke Flavouring Primary Product . J. Food Science 28:87;871-882

Siskos, I., A Zotos S, Melidou R, Tsikritzi. 2007. The effect of liquid smoking of fillets of trout (*Salmo gairdnerii*) on sensory, microbiological and chemical changes during chilled storage. *Food Chem* 101:458-464.

Soeparno,S. 2009. Ilmu dan Teknologi Daging. Universitas Gadjah Mada Press.

Soldera, S. Sebastianutto N, Bortolomeazzi. R,. 2008. Composition of phenolic compounds and antioxidant activity of commercial aqueous smoke