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Effect of salam [*Syzygium polyanthum* (Wigt) Walp.] leaves extract on the microorganism population in chicken meat and shrimp and their sensory

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Abstract

Recently, there has been an increasing demand and interest in developing plant extracts as natural food sanitizer, owing to their antimicrobial properties. Hence, the aim of this study was to investigate the effect of salam (*Syzygium polyanthum* L.) leaves methanolics extract on the number of microflora on chicken meat and shrimp. Salam leaves extract at different concentrations (0.0%, 0.1%, 1.00%) and exposure times (5, and 10 min) used to treat chicken meat and shrimp by using dilution method. Result showed that the total plate count and *Staphylococcus aureus* had been detected in untreated chicken and shrimp samples with 6.66 ± 0.12, 8.66 ± 0.15 and 7.25 ± 0.21, 6.54 ± 0.21, respectively. However, there was no *Escherichia coli, Salmonella* spp. and *Vibrio cholerae* detected in both samples. The number of total plate count (TPC) and *S. aureus* in chicken meat and shrimp were starting to reduce significantly at 0.01% concentration of salam leaves extract for 5 minutes of exposure time compared to initial count. There was no significantly different between exposure times. The highest reduction in number of microorganism population was at treatment with 1.0% extract for 10 min where TPC was reduced from 6.66 ± 0.12 to 0.00 ± 0.00 $\log_{10}$ CFU/ml, and from 8.66 ± 0.15 to 4.88 ± 0.00 $\log_{10}$ CFU/ml in shrimp while *S. aureus* reduced from 7.25 ± 0.21 to 3.88 ± 0.01 and from 6.54 ± 0.21 to 4.92 ± 0.04 in chicken and shrimp, respectively. For the sensory acceptability, overall acceptability were accepted by panellists until treatment 0.10% for 5 min and 10 min of soaking time. In conclusion, salam leaves extract might be developed as natural sanitizer for rinsing raw food materials such as chicken meat and shrimp.

Introduction

Food contamination has become a serious concern with the increasing number of outbreaks of food-borne illnesses (Rane, 2011). The alleviating cases in food-borne illnesses have highlighted the importance of microbiological control in the food industry. Microbial contamination in food that leads to food-borne illnesses may be contributed by various factors including inadequate food hygiene, place of preparation, cooking or processing procedure, processing or serving utensils, raw materials, and time and temperature abuse (Annan-Prah *et al.*, 2011).

According to Scientific Status Summary (2013), types of food-borne bacterial pathogens commonly detected in raw meat and seafood are *Salmonella* sp., *Staphylococcus aureus*, and *Escherichia coli*. Besides, *Vibrio cholerae* is also commonly found in seafood. Aside from this, Abadias *et al*. (2008) reported that *Salmonella* sp. and *E. coli* are the most prominent food-borne pathogens which lead to outbreaks of illnesses through consumption of the contaminated fresh products. On the other hand, most of the *Vibrio* spp. are pathogenic for mankind and usually responsible for causing alimentary infections in countries with warm coastal waters, where fish and shellfish are consumed raw or lightly cooked. It is important to point out that these pathogens have been isolated in larger number from fish and shellfish (Slavika *et al*., 2002). Wilfred *et al.* (2014), indicated the consumption of poultry meat has been associated with the incidence of outbreaks of foodborne illness. In addition, study by Akbar & Anal (2013), showed that most of *S. aureus* species easily be found in poultry meat.

In order to resolve the problems of microbial contamination and spoilage in food, various food preservation strategies have been developed and applied to control the growth and propagation of microorganisms including chilling, freezing, water activity reduction, nutrient restriction, pasteurization

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or synthetic antimicrobials (Davidson, 1993). However, most of these procedures may result in the loss of organoleptic properties of foods and reduce consumer acceptability (Pradeep, 2011). Besides that, nowadays consumers also put a lot of concern in the use of synthetic additives due to its adverse health effect. Therefore, instead of synthetic antimicrobials, natural antimicrobials are more preferable by consumers and thus natural antimicrobials such as organic acids, essential oils, plant extracts, and bacteriocins become considerable alternatives to ensure food safety (Cleveland *et al.*, 2001; Burt, 2003). The antimicrobial activity of plant extracts are studied and found to be having phytochemical constituents such as phenolic compounds, protein-like compounds, flavonoids and tannins (Pradeep, 2011).

Salam leaves, which is synonym to *Syzygium polyanthum* L., is a deciduous tropical tree belonging to the Myrtaceae family (Kato *et al.*, 2013). This plant grows wildly on lowlands and is widely distributed in the temperate, subtropical and tropical regions in the world (Perumal *et al.,* 2012). Salam leaves has been used traditionally as medicine or therapeutic agents include effective against ulcer, hypertension, diabetes, hyperuricemia, diarrheal, gastritis, skin diseases and inflammation (Sumono and Wulan, 2008). Salam leaves had been found to have antimicrobial functions. It was found to be able to inhibit the growth of microorganisms such as *Bacillus cereus, Bacillus subtilis, E. coli, Salmonella* sp., *S. aureus* and *Pseudomonas fluorescens* (Setiawan, 2002). Other than that, salam leaves are also found to be able to cure diarrhea due to the presence of tannin and also able to treat diabetes and skin infection (Dalimartha, 2007). It was also reported to have potential antioxidant properties (Perumal *et al.,* 2012; Darusman *et al.*, 2013).

Researcher had reported the effectiveness of garlic extract to reduce microbial growth on poultry meat such as *E. coli, Salmonella typhimurium, S. aureus* and *Bacillus cereus* (Yadav *et al.*, 2002). Clove extract was found to inhibit the growth of *S. typhimurium, Aeromonas hydrophila* and toxin production by *E. coli* in chicken meat during refrigerated storage (Singh *et al.*, 2004).

However, no study has been reported on the treatment of daun salam extract against the microorganisms in chicken meat and shrimp. Therefore, the aim of this study was to determine the antimicrobial activity of salam leaves on natural microorganism population in chicken meat and shrimp as a basic screening in order to developing natural food sanitizer.

Materials and Methods

Dried salam leaves

Samples, the dried salam leaves (*Syzygium polyanthum* L.), were purchased from herbal market, Pasar Baru, Bandung, Indonesia. The samples were identified and deposited in Laboratory of Natural Products, Institute of Bioscience (IBS), Universiti Putra Malaysia (UPM), Serdang, Selangor, Malaysia. The dried salam leaves were packed in sealed plastic bags and kept at room temperature.

Chicken and shrimp

Raw chicken meat (boneless fillets) and raw shrimp (white leg type) were purchased from Tesco Hypermarket, IOI City Mall, Putrajaya, Malaysia. Both samples were purchased on the day of analysis to ensure their freshness, and to avoid contamination during storage.

Extraction of salam leaves

The salam leaves were extracted using the methods reported by Rukayadi *et al.* (2008) with slight modifications. One hundred grams of salam leaves powder were placed in 500 ml conical flask and soaked in 400 ml of absolute methanol (99.8%) (Sigma-Aldrich, Saint Louis, MO, USA). The mixture underwent sonication for a total duration of 45 min with the sonicator water bath (Delta Ultrasonic Cleaner D150H, Taiwan). Mixture was then filtered using Whatman filter paper No. 2 (Whatman International Ltd., Maidstone, England) attached to an the aspirator pump. After that, the liquid solution underwent evaporation with a rotary vacuum evaporator (Heidolph Instruments, Germany) at 50ºC and 150 rpm for 30 min to obtain methanol-free salam leaves extract. The extract was kept at 4ºC until further analysis.

Preparation of salam leaves extract for treatment

In this preparation, 3 g salam leaves extract was diluted with 30 ml 10% dimethyl sulfoxide (DMSO) (Sigma-Aldrich, Saint Louis, MO, USA) with the ratio (1:10) to make up a 10% salam leaves extract solution. The 10% salam leaves extract solution was then serially diluted in the sterile de-ionized water (DIW) (B Braun Medical Industries, Penang, Malaysia) to make up extract solutions of three different concentrations (1%, 0.1% and 0.01%) which were used on the raw chicken meat and shrimp. concentration of salam leaves extract solution were prepared and ready to be used for the application on raw chicken meat and shrimp. This methodology was referred to Yusoff *et al*. (2015)

with slight modification.

Media for enumeration of microorganism by spread plate method

Media were chosen based on Microbiology Manual (12th ed, Merck, Darmstadt, Germany) which included Plate count agar (Merck, Darmstadt, Germany), Tryptic soy agar (Merck, Darmstadt, Germany), Mac Conkey agar (Hardy Diagnostics, Santa Mania, USA), X.L.D. medium (OXOID Ltd., Basingstoke, Hampshire, England) and T.C.B.S. Cholera medium (OXOID Ltd., Basingstoke, Hampshire, England).

Treatment of chicken and shrimp samples with salam leaves extract

Ten grams of raw chicken meat and shrimp samples were immersed in 25 ml of tap water, DIW and different concentrations (1.0%, 0.1% and 0.01%) of salam leaves extract solutions at room temperature for 5 and 10 min exposure time. A sample without soaking with any solution served as the positive control, for both chicken meat and shrimp.

Enumeration of microorganisms by spread plate method

All treated samples were aseptically removed using a flame-sterilized spatula and drained on absorbent paper for drying. The treated samples were placed into the stomacher bag (Baglight, BagSystem, Interscience, France) with 90 ml of 0.1% peptone water. The mixture was homogenized using a stomacher machine (BagMixer 400-P, Interscience, France) for 2 min at 250 rpm. The enumeration of microorganisms was conducted by spread plate methods. The colony forming unit per gram (CFU/g) of samples were calculated after 24 h of incubation at 37ºC.

Sensory evaluation

The sensory evaluation acceptability test was performed as according to Brasil *et al.* (2012), with slight modification. A preliminary test was conducted to determine whether the treated food would be acceptable to consumers. The sensory evaluation was carried out for the visual sensory test of each treatment of the raw chicken meat and shrimp samples. All samples were treated with tap water, DIW and different concentrations (1.0%, 0.1% and 0.01%) of salam leaves extract solutions for 5 and 10 min exposure time. A group of 30 random panellists were invited to perform the evaluation on the treatment samples. The evaluation was conducted based on the 9-point hedonic scale for acceptance in terms of colour (observed with eyes), odour (smell with nose), texture (touch with finger) and the overall acceptability on each treatment samples. The rating for each analysis of samples were given in a range of scale from extremely like (scale of 9) to extremely dislike (scale of 1). Scores ≥5 were "acceptable".

Statistical analysis

Data were analysed by using MINITAB 16 for the analysis of variance (ANOVA), one-way, unstacked, whereby Tukey's test was employed to determine the significance difference ($p<0.05$) between different treatments. Results were expressed as means ± standard deviation (SD) of duplicate analyses, unless otherwise stated.

Results

Yields of salam leaves extraction

A 100 g of dried weight of salam leaves extracted with methanol solvent yielded 3.06 g extract (3.06%).

Effect of salam leaves extract on number of microorganisms in treated chicken meat and shrimp

Chicken meat and shrimp samples were treated with different extract concentrations (1.0%, 0.1%, 0.01%, tap water and DIW) and different soaking times (5 min and 10 min). TPC and selective agar media were used to enumerate the number of natural microflora and food-borne pathogens in chicken meat and shrimp samples after treatment. The results were interpreted in $\log_{10}$ CFU/ml and summarized in Table 1 and Table 2, respectively.

Sensory evaluation of raw chicken meat and shrimp treated with salam leaves extract

Results for sensory evaluation are presented in Table 3 and Table 4, respectively.

Discussion

Potential contamination in chicken meat and shrimp by the foodborne pathogens can be caused through the unhygienic practices in handling, cooking or post cooking storage of products. During washing process of raw foods material is also one of the causes for the foodborne pathogens to growth with rapidly. However, those numbers of microorganisms in raw foods can be reduced by using the antimicrobial compounds in the sanitizers. Therefore, sanitizers which possess antimicrobial property become important and common in washing raw food materials (Nychas, 1995).

In this experiment, salam leaves extract was

Table 1.Number of microorganism presence in untreated chicken meat (initial bacterial load) and treated chicken meat samples (log10 CFU/ml)

Sample	Chicken meat							
Bacterial species	Total plate count		*E. coli*		*S. aureus*		*Salmonella* sp.	
Initial bacterial Load (log_{10} CFU/ml)	6.66 ± 0.12 ^{a}		0.00 ± 0.00^{a}		7.25 ± 0.21 ^{a}		0.00 ± 0.00 ^{a}	
[1]ST/ Treatment	5 min	10 min	5 min	10 min	5 min	10 min	5 min	10 min
Tap Water	6.61 ± 0.03^{aA}	6.55 ± 0.01^{aA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}	7.13 ± 0.03^{aA}	7.15 ± 0.00^{aA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{Aa}
[2]DIW	6.36 ± 0.01^{aA}	6.36 ± 0.01^{aA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}	7.10 ± 0.01^{aA}	7.11 ± 0.01^{aA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}
0.01%	5.00 ± 0.03^{bA}	4.84 ± 0.04^{bA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}	6.55 ± 0.05^{bA}	6.54 ± 0.00^{bA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}
0.1%	3.35 ± 0.11^{cA}	3.01 ± 0.01^{cA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}	5.61 ± 0.01^{cA}	5.44 ± 0.01^{cA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}
1.0%	0.00 ± 0.00^{dA}	0.00 ± 0.00 ^{dA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}	4.11 ± 0.01^{dA}	3.88 ± 0.01^{dA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}

Mean values ± standard deviation with different lowercase letter in the same column have significant difference (P<0.05).
[1] ST = soaking time
[2] DIW = deionized water
[3]TFTC = too few to count (colonies < 30)

obtained through extraction. Extraction was done by using absolute methanol due to its capability to produce high yield of extract containing high chemical compounds from the plant sample (Caunii *et al*., 2012). Table 1 shows the number of microorganisms detected in chicken meat before and after soaking treatment. The number of microorganisms in total plate count (TPC) detected was log_{10} CFU/ml 6.66 ± 0.12, *S. aureus* was log_{10} CFU/ml 7.25 ± 0.21 and none were detected for *E. coli* and *Salmonella* sp. respectively. In this finding, *S. aureus* was the highest number of species found in the samples, and this correlated with findings from Akbar and Anal (2013) who also found *S. aureus* to be predominant in poultry meat. The number of total plate count (TPC) and *S. aureus* in chicken meat were starting to reduce significantly at 0.01% concentration of salam leaves extract for 5 minutes of exposure time compared to initial count. However, reduction of the bacterial count was not significantly different between the soaking times.

Table 1 also shows the highest reduction of bacterial population (TPC) occurred in 1.0% concentration from initial bacterial load log_{10} CFU/ml 6.66 ± 0.12 to 'Too Few To Count' (TFTC) and the population was decreased with the decreasing concentration of extract solutions. This finding trend was correlated with Boziaris *et al*., (2010) who indicated that plant extract delayed the growth of microorganisms in a concentration-dependent manner where the higher the concentration, the higher the growth in inhibition and reduction. It showed that the reduction number of microorganisms in chicken meat was strongly correlated with the concentration of salam leaves extract.

The highest reduction of *S. aureus* population also occurred in treatment 1.0% extract from initial load of log_{10} CFU/ml 7.25 ± 0.21 to 4.11 ± 0.01, and the reduction in population decreased with the decreasing concentration of extract solutions. On the contrary, chicken meat samples soaked in tap water and DIW gave only slight reduction in bacterial population for both 5 min and 10 min soaking. For TPC, the reduction only from log_{10} CFU/ml 6.66 ± 0.12 to 6.61 ± 0.03 and 6.55 ± 0.0 whereas the population of *S. aureus* reduced from log_{10} CFU/ml 7.25 ± 0.21 to 7.13 ± 0.03 and 7.15 ± 0.00 respectively.

Table 2 shows the number of microorganisms detected in raw shrimp before and after soaking treatment. The number of natural microorganisms in total plate count (TPC) detected was log_{10} CFU/ml 8.66 ± 0.15, whereas for *S. aureus* was log_{10} CFU/ml 6.54 ± 0.21 and *E. coli* and *V. cholerae* not detected. Results also showed that the total surviving bacterial population (TPC) on shrimp was reduced after treated with different concentrations of salam leaves extracts (1.0%, 0.1% and 0.01%), tap water and DIW. Basically, the reduction trend in shrimp samples after treatment was the same as the trend in chicken meat samples. The number of total plate count (TPC) and *S. aureus* in shrimp were starting to reduce significantly at 0.01% concentration of salam leaves extract for 5 min of exposure time compared to initial count. The highest reduction for both total bacterial population and *S. aureus* occurred in 1.0% concentration at 10 min soaking from initial bacterial load of log_{10} CFU/ml 8.66 ± 0.15 to 4.88 ± 0.00 and log_{10} CFU/ml 6.54 ± 0.21 to 4.92 ± 0.04, respectively.

On the other hand, there was no bacterial count from untreated chicken and shrimp samples for

Table 2. Number of microorganism presence in untreated shrimp (initial bacterial load) and treated shrimp samples (log_{10} CFU/ml).

Sample	Shrimp							
Growth media (bacterial species)	TPC (all species)		*E. coli*		*S. aureus*		*Vibrio cholerae*	
Initial bacterial Load (log_{10}CFU/ml)	8.66 ± 0.15 [a]		0.00 ± 0.00 [a]		6.54 ± 0.21 [a]		0.00 ± 0.00 [a]	
[1]ST/ Treatment	5 min	10 min	5 min	10 min	5 min	10 min	5 min	10 min
Tap water	8.63 ± 0.01^{aA}	8.61 ± 0.01^{aA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}	6.53 ± 0.01^{aA}	6.49 ± 0.01^{aA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}
[2]DIW	8.55 ± 0.02^{bA}	8.51 ± 0.00^{bA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}	6.51 ± 0.01^{aA}	6.47 ± 0.01^{aA}	0.00 ± 0.00^{aA}	0.00 ± 0.00 ^{aA}
0.01%	8.13 ± 0.01^{cA}	7.96 ± 0.03^{cA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}	6.02 ± 0.00^{bA}	5.99 ± 0.00^{bA}	0.00 ± 0.00^{a}	0.00 ± 0.00^{aA}
0.1%	6.52 ± 0.01^{dA}	6.24 ± 0.00^{dA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}	5.75 ± 0.02^{cA}	5.61 ± 0.01^{cA}	0.00 ± 0.00^{a}	0.00 ± 0.00^{aA}
1.0%	5.14 ± 0.01^{eA}	4.88 ± 0.00^{eA}	0.00 ± 0.00^{aA}	0.00 ± 0.00^{aA}	5.02 ± 0.00^{dA}	4.92 ± 0.04^{dB}	0.00 ± 0.00^{a}	0.00 ± 0.00^{aA}

Mean values ± standard deviation with different lowercase letter in the same column have significant difference (P<0.05).
[1] ST = soaking time
[2] DIW = deionized water

Table 3. Result for sensory evaluation on the application of *salam* leaves extracts on raw chicken at different concentrations at different soaking times

Soaking Time	5 min					10 min				
Attributes/ Treatment	TW	DIW	0.01%	0.1%	1%	TW	DIW	0.01%	0.1%	1%
Colour	6.37 ± 1.73^{ab}	5.57 ± 2.13^{ab}	5.93 ± 1.72^{ab}	5.86 ± 1.55^{ab}	2.63 ± 1.79^{c}	5.20 ± 1.83^{b}	5.47 ± 1.85^{ab}	6.73 ± 1.41^{a}	5.27 ± 1.78^{ab}	2.83 ± 2.07^{c}
Odour	5.23 ± 1.91^{ab}	3.77 ± 2.05^{b}	5.17 ± 2.04^{ab}	4.77 ± 1.91^{ab}	3.67 ± 2.38^{b}	4.90 ± 1.95^{ab}	4.70 ± 1.82^{ab}	5.63 ± 1.85^{a}	4.87 ± 2.06^{ab}	3.63 ± 2.31^{b}
Texture	5.93 ± 1.87^{a}	5.10 ± 2.0^{a}	5.63 ± 1.67^{a}	5.37 ± 1.50^{a}	3.53 ± 2.15^{bc}	5.0 ± 1.98^{ab}	5.67 ± 1.94^{a}	6.47 ± 1.31^{a}	5.67 ± 1.54^{a}	3.43 ± 2.24^{c}
Overall Acceptance	5.80 ± 1.67^{ab}	4.53 ± 2.05^{bc}	5.67 ± 1.56^{ab}	5.20 ± 1.47^{ab}	3.37 ± 2.24^{c}	5.13 ± 1.80^{ab}	5.23 ± 1.78^{ab}	6.10 ± 1.63^{a}	5.23 ± 1.79^{ab}	3.30 ± 2.10^{c}

Mean values ± standard deviation with different lowercase letter in the same column have significant difference (P<0.05).

E. coli, Salmonella sp. and *V. cholerae*, indicating that the samples from hypermarket were fresh and clean at the time the experiment was performed. *Enterobacteriaceae* is a hygiene indicator of food (Zeitoun *et. al.*, 1994). Enterobacteriaceae family is Gram-negative bacteria that include many of more familiar pathogens such as *Salmonella, E. coli, Klebsiella, Shigella* and *Proteus*. Hence, the initial population of Enterobacteriaceae can be an indicator of adequate hygiene conditions of food.

Finding showed that the effect of salam leaves extract was positive which induced a reduction in microbial population. Besides that, studies from Setiawan (2002) also proved that salam leaves extract possesses antimicrobial activities to inhibit the growth of microorganisms such as *E. coli, V. cholerae* and *Salmonella* sp. Roughly, mechanism of antimicrobial action from plant extract can be explained as the membrane disruption pathway by phenolics and metal chelation by flavonoids in inhibiting the growth of microorganisms. According to Lau (2015), salam leaves contain different active compounds where each of them functions differently for inhibition of pathogens. Among the compounds identified which have antibacterial activity include hexadecanoic acid and phytol (Preethi *et al.*, 2010). Besides that, Jananie *et al.* (2011) also stated that phytol (0.66%) possess antimicrobial activity.

According to Irawan *et al.* (2012), salam leaves extract contained carbohydrate, tannins alkaloid, steroid, triterpenoid, and flavonoid and saponins. Flavonoids have been reported to possess antibacterial, antioxidant, anti-inflammatory, antiallergic, antimutagenic, and vasodilatory activity. Tannin is believed to be responsible for the antibacterial activity of salam leaves. The mechanisms of inhibition of the bacteria growth involved precipitation forming and denaturing of the bacteria protein (Nur Amalina, 2014).

Since salam leaves has been proved to have antimicrobial functions, as also demonstrated in this study, sensory evaluation was also carried out in order to determine the effect of salam leaves extract application in the food system on the sensory properties, even at low concentrations. The variations in colour, odour and texture of the food samples after

Table 4. Result for sensory evaluation on the application of salam leaves extracts on raw shrimp at different concentrations at different soaking times.

Soaking Time	5 min					10 min				
Attributes/ Treatment	TW	DIW	0.01%	0.1%	1%	TW	DIW	0.01%	0.1%	1%
Colour	6.33 ± 1.56^{a}	5.80 ± 1.73^{a}	6.63 ± 1.54^{a}	6.43 ± 1.83^{a}	4.17 ± 2.28^{b}	6.20 ± 1.65^{a}	6.43 ± 1.94^{a}	6.40 ± 1.77^{a}	5.63 ± 1.43^{a}	3.73 ± 2.07^{b}
Odour	6.27 ± 1.57^{a}	5.50 ± 1.83^{ab}	5.93 ± 1.66^{a}	5.97 ± 1.61^{a}	4.23 ± 2.47^{bc}	5.30 ± 1.66^{abc}	5.97 ± 1.71^{a}	5.97 ± 1.77^{a}	5.37 ± 1.65^{abc}	3.87 ± 2.27^{c}
Texture	6.17 ± 1.82^{ab}	5.30 ± 1.60^{abc}	6.23 ± 1.70^{a}	6.30 ± 1.24^{a}	4.73 ± 2.23^{bc}	5.80 ± 1.71^{ab}	6.33 ± 1.79^{a}	6.17 ± 1.84^{ab}	5.37 ± 1.77^{abc}	4.10 ± 2.14^{c}
Overall Acceptance	6.30 ± 1.64^{a}	5.60 ± 1.63^{ab}	6.17 ± 1.34^{a}	6.23 ± 1.55^{a}	4.33 ± 2.44^{bc}	5.67 ± 1.63^{ab}	5.67 ± 1.63^{ab}	6.10 ± 1.63^{a}	5.43 ± 1.65^{ab}	3.90 ± 2.25^{c}

Mean values ± standard deviation with different lowercase letter in the same column have significant difference ($P<0.05$).

treatment do account for variations in food quality and consumer acceptance rather than caused by spoilage bacteria (Solomon *et al*., 2014). Sensory evaluation results were interpreted and presented in Table 3 for chicken meat and Table 4 for shrimp. Sensory evaluation was conducted in visual testing basis with 30 random panellists. Four attributes including colour, texture, odour and overall appearance were evaluated in a 9-point hedonic scale.

Based on the data in Table 3 and Table 4, overall acceptability were accepted by panellists until treatment 0.10% for 5 min and 0.1% for 10 min of soaking time. This meaning the treatment by tap water, DIW, 0.01% and 0.1% extract at 5 and 10 min were not significantly different and this types of solutions does not affected colour, odour, texture and overall acceptability of the samples. In this test, tap water was act as a control. The highest overall acceptability of treated chicken meat at 5 min and 10 min treatment was by soaking in 0.01% extract.

Based on Table 4, for raw shrimp samples, the highest preference of treated sample by panellists was 0.1% extract at 5 min soaking which scored at 6.23 ± 1.55 and 0.01% at 10 min with 6.10 ± 1.63. Many studies had reported the antimicrobial activity of plant extract as natural food sanitizer without concern on sensory acceptability which might affect consumer preference in future. Study conducted by Bingol *et al.* (2011) showed the use of lemon juice extract caused slight reduction of *S.* Enteritidis and *E. coli* in raw meatball, and interestingly results gave no significant differences for the sensory attributes; flavor intensity, acidic flavor, juiciness and overall acceptability.

Conclusion

The effect of salam leaves extract on the microorganism population in chicken meat and shrimp was positive. The reduction of the number of microorganisms by salam leaves extract was dependent on extract concentration. The higher the concentration of extracts resulted the higher the microbial reduction. In chicken meat and shrimp, the numbers of microorganisms were starting to reduce significantly at 0.01% concentration of daun salam extract for 5 minutes of exposure time compared to initial count. There was no significantly different between exposure times. For the sensory acceptability, overall acceptability was accepted by panellists until treatment 0.10% for 5 min and 10 min of soaking time. Overall, these results suggested that salam leaves extract exhibited antimicrobial activity and might be further developed as a natural food sanitizer.

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