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Effect of nitrite and citric acid on the bacterial count and sensory evaluation of the canned beef sausages

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ABSTRACT

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KEYWORDS:

Nitrite, Canned sausage, Citric acid The effects of nitrite and citric acid as preservatives on the quality and safety of the canned beef sausage were investigated after three months storage at room temperature (35±5°c). Two experiments were conducted in this study, the first was undertaken to determine the effect of nitrite as a preservative on total bacterial count of the canned sausages retorted at 107.2 °C (225)°F for 80 minutes, and at 115.5°C (240)°F for 40 minutes . The second one, which was based on the results of the first experiment, was conducted to determine the effects of the absence of nitrite on the canned sausage processed with meat treated by immersion in 1% citric acid before processing at (80 and 30 °C) for one minute and drained, then the product retorted at 107.2 °C for 80 minutes, The parameters included total bacterial count and sensory attributes (flavour, juiciness, tenderness and overall acceptability). The evaluation of the total bacterial count was done monthly and indicated that, there were no colonies of bacteria found in the samples evaluated even after three months of storage. The sensory evaluation, which was conducted twice; after canning, and after 3 months of storage in the second experiment, indicated that there were no significant differences among the treatments in colour, flavour, tenderness, juiciness and overall acceptability. However, it was noticed that, there was a slight decrease in flavour, colour, juiciness and the product became less tender and less acceptable by the panelists after 3 months of storage in comparison with that evaluated immediately after canning. On the other hand, Citric acid had no clear effect on the total bacterial count and sensory attributes, but caused slight decrease in the total bacterial count of the raw and cooked sausages compared to the control.

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INTRODUCTION

The Sudan has a huge amount of animal resources which has qualified it to compete in meat industry. There are approximately 102.235.000 heads of cattle, sheep, goat and camel (MARFR, 2012). In spite of all this great resource and availability of good quality red meat which has been estimated as 167.2 million tons (A.O.A.D, 2005), there is no considerable development in meat processing and preservation.

In the past Sudanese preferred fresh meat to processed meat, but now the profitability and technological advances, and the change in the life style (the working women have a little time for food preparation), will probably affect the rate and ultimate extent to which meat is processed.

Thermal processing, specifically retort processing, has been used as a common preservation technique in food industry for shelf stable low acid foods. Commercial retort processing ensures as The United States Code of Federal Regulations defines commercial sterility as "The condition achieved by application of heat, chemical sterilant (s), or other appropriate treatment that renders the equipment and containers free of viable microorganisms having public significance, as microorganisms of non-health significance, capable of reproducing in the food under normal no refrigerated conditions of storage and distribution (USFDA, 2009).. In recent times, with the development of computing technology, programs are available to determine thermal process requirements, and also online monitoring and controlling of the thermal process (Fellows, 2009).

Generally foods are processed commercially for one of the following reasons: (1) extend the shelf life of the processed food form, (2) alter the characteristics of the product, (3) separate components from the complex mixture of bio-chemicals and (4) improve the nutritional characteristics of the processed food (Lund, 1979).

Meat products that can be stored at room temperature without the risk of microbial spoilage are considered to be shelf stable products. They include canned meats such as ham, tuna and chicken, jerky, dry sausages snack sticks, summer sausage and freeze dried meat. Because these products not need cold temperatures preservation, they are popular for camping, trips, hunting and fishing expeditions and other activities where refrigeration may not be available. In addition, they are convenient product to have on hand in your cupboard (Elizabeth, 1994). So in the Sudan the farmers in agricultural areas, the army in the military regions, people in the disaster areas, and regions of the displaced people, are in need of shelf stable and acceptable product that does not require refrigeration. Meat canning is a suitable method of preservation to solve these problems that meet the availability of safe foods in those areas.

Today consumers all over the world especially in industrialized and developed countries increasingly demand minimally processed foods and food stuffs, which retain natural flavour, colour, texture, nutritional characteristics, and contain fewer additives (e.g. preservatives) especially chemical additives (Ohlsson and Bengtsson, 2002). Accordingly to encourage the consumption of processed meat we must get rid of the use of chemical additives, as nitrite has been reported to be carcinogenic above and beyond its possible role in formation of nitrosamines (Pearson and Tauber, 1989). The objectives of the study were, to investigate the use of many preservatives in canning in order to decrease the time and temperature required for commercial sterilization, to evaluate the necessity of using nitrite in canned sausages by examining its effect on microbial and palatability aspects of canned sausages, and also to develop a meat product that does not need freezing or refrigeration and accessible to people living in remote areas.

MATERIALS AND METHODS Experiment (1)

Approximately 7 kgs of lean meat (top side) cut were taken from beef animals of similar breed and age after 24 hours from slaughter at Meat Technology Department in Kuku Animal Production Research Centre and used in sausage preparation

The meat was ground through (0.64 cm diameter) plate of an electrical meat grinder. Casings: Synthetic cellulose casings were obtained from local market.

Cans: Round sanitary acid-resistance cans (6.83cm in diameter and 10.16cm in. height) of nominal capacity 315 gm were obtained from a local processing plan

Sausage preparation

The ground meat (6 kgs) was divided into two equal batches, one batch was formulated using the ingredients in Table (1) and 155 ppm sodium nitrite while the other was

formulated using the same ingredients in Table (1) without addition of nitrite and considered as control.

Each batch was chopped separately. The chopper was started after the minced meat was introduced. Salt, nitrite (in one batch), and half of the recommended ice water were added and uniformly dispersed. Then, the binder and seasoning were added together, with the remainder of the recommended ice water. The entire mass for each batch was chopped about 5 minutes. The batter for each batch was then stuffed into cellulose casings of 22 mm in diameter and linked at length about 8 cm. The product was cooked for 15 minutes in boiling water followed by immediate cooling in ice water for 5 min. The sausages were peeled, packed in polyethylene bags and stored in refrigeration (4°C) over night. The product of each batch was divided into two treatments in one treatment the canning was done by retorting at 107°C for80 minutes, and in the other treatment the canning was done by retorting at115.5°C for 40 minutes. And considered at 0 month. The other samples were awaiting microbial test at intervals of 1, 2, 3 months of storage.

Table 1: Ingredients based on total mixed base

Ingredients	Percentage
Cold water (crushed ice)	12.2
Salt	2.4
Ground pepper	0.3
Sugar	0.3
Mustard powder	0.2
Skimmed milk powder	3.4

Canning operation:

The empty cans were spray washed with 26.5°C water in accordance with federal meat inspection regulation (Pearson and

Tauber, 1989). Each batch of the product was subdivided into two portions, one part to be canned, and retorted at 107.2° C for 80 min, and the other part to be canned and

retorted at 115.5°C for 40 minutes. The cans were filled with sausages by hand. Seven pieces(links) of sausage weighing about 160gm were put in each can. Then the remaining space was filled with decontaminated boiling water. The cans were closed by double sealer machine (Mp 502120207/s) in the Canning Unit of the Food Research Centre, Shambat.

Cans were placed into a vertical nonagitating retort. After processing cans were water cooled by immersing in a cold water tank. Temperature of water was approximately (15°c). The wet cans were dried by heat which was permitted to accelerate evaporation (Pearson and Tauber, 1989).

Product storage

The cans were labelled and stored at room temperature (Table, 2); 30gm of samples were taken randomly after 72 hours (incubation period) from canning for microbial test

Table 2: Average temperature and humidity of storage for the canned sausages

No. of experiment	Period of storage	Temperature °C	Humidity %
	First month	35.3	32.25
1	Second month	31.9	23.25
	Third month	30.5	21.3
	First month	30.1	16.2
2	Second month	30.9	14.8
	Third month	35.7	21.1

Experiment (2) Meat preparation

Meat samples were taken from beef animals of similar breed and age after 24 hours from slaughter at the Meat Technology Department in Kuku Animal Production Research Centre.

Approximately 7 kgs of lean meat (top-side) cut were trimmed to a minimum amount of fat were used in sausage preparation in each replicate (three replicates were performed for each treatment).

The lean meat was divided into two batches, one batch treated with citric acid and another batch treated with decontaminated water (free of contamination by water filtration). Each batch was divided into two parts. The first part was dipped in 1% (at 30°c) citric acid for one minute and drained for one minute. The 2nd part was dipped in 1% citric acid at (80°c) for one minute and drained for one minute. The third part was dipped in decontaminated water at 30°c for

one minute and drained for one minute. The fourth part was dipped in decontaminated water at 80°c for one minute and drained for one minute. The meat was ground through 0.25 in/plate of electrical meat grinder. The same casings and same cans as in experiment (1) were used. Four treatments were done:

- 1/ Sausages processed with meat dipped in 1% citric acid at 30°c for one minute and retorted at 107.2 °C for 80 minutes.
- 2/ Sausages processed with meat dipped in 1% citric acid at 80°c and retorted at 107.2°C for 80 minutes.
- 3/ Sausages processed with meat dipped in decontaminated water at 30°c for one minute and retorted at 107.2°C for 80 minutes.
- 4/ Sausages processed with meat dipped in decontaminated water at 80°c for one minutes and retorted at 107.2°C for 80 minutes.

Sausages preparation

Sausages were prepared following the previously mentioned procedures in experiment one. Random samples of raw (uncooked) and cooked sausages were taken for microbial analysis.

Sausages canning

All procedures of canning were performed as in experiment one except the retorting was done for all treatments at 107°C for 80 minutes.

Product storage

The cans were labelled and stored for three months at room temperature (Table 2) waiting microbial tests every month. Each variable determination was replicated three times.

Total bacterial count (TBC)

Ten gms of the sample were homogenized (using Bench Blender) in 90 ml sterile distilled water for 1.5 minutes. Ten fold dilutions of the homogenate were prepared in 0.5% peptone water (oxoid L 37) 1 ml of suitable dilutions were mixed with 10 ml sterile plate count a gar (oxoid CM 325) and the plates were incubated at 37°c for 48 hours. Colonies were counted and the TBC was reported as log_{10} cfu/g. log_{10} cfu/g values were used for the calculation of mean log_{10} cfu/g in statistical analysis (Jarvis, 1989)

Sensory evaluation of sausages:

Samples were taken for evaluation after the TBC recorded negative results.

The sensory evaluation was conducted in the sensory evaluation facilities of the Meat Technology Department, Animal Production Research Centre. Ten (10) semi trained panellists were asked to evaluate the treatments effect Cross *et al.*, (1978) on colour, flavour, tenderness, juiciness and

over all acceptability after canning (before storage), and after 3 months of storage.

Statistical Analysis

Statistical analysis was performed on all data of the various experiments using SPSS and was subjected to analysis of variance (ANOVA). Least significant difference (LSD) was used for mean separation (Gomez and Gomez, 1984).

RESULTS AND DISCUSSION Microbiological results Experiment (1)

As shown in Table (3) microbial results indicate that there were no colonies of bacteria to be counted in all treatments. Canning at 115.5°C for 40 minutes gave the same results as canning at 107°C for 80 minutes irrespective of the addition of nitrite. These results proved that, the canned sausages were free from any bacteria and commercially sterilized so it would be safe at room temperature within 3 months of shelf life. Most canned meats that do not require refrigeration had relatively long shelf life (Dwight and Horn, 2004). Sterile canned meats should be placed in a cool, dry place since both relative humidity and temperature influence their keeping quality (Pearson and Tauber, 1989). As discussed by Aberle et al., (2001) nitrite, either as a potassium or sodium salt, is used to preserve desirable meat flavour, prevent warmed over flavour, fix a bright reddish pink colour and microbial inhibit growth, particularly outgrowth of Cl. botulinum spores. Also in a study to reduce nitrite content in hot dogs using hurdle technology without sacrificing product safety and quality, Jafari and Zahra (2007) found that, there was a decrease in total aerobic counts in hurdle treated hot dogs (with 50 ppm nitrite), compared to the control (with 120 ppm nitrite) whereas perfringens Clostridium count Clostridium botulinum detection were the

same in both hurdle treated and control samples.

Commercially sterile canned meat products generally reach an internal temperature of at least 107°C, but this temperature may be as low as 101.5°C, depending on salt and nitrite content (Pearson and Tauber, 1989).

Table 3: Effect of sausage treatment on total bacterial count (log 10 cfu/g)

Storage (Month)	225	5° F	240	S.E	
	Ni	trite	Nit		
	(-)	(+)	(-)	(+)	
0	0	0	0	0	
1	0	0	0	0	0
2	0	0	0	0	
3	0	0	0	0	

^{*} Canned sausages using 107.2 °C and 115.5 °C processing temperatures with (+) nitrite without (-) nitrite.

Experiment (2)

Table (4) indicates that, there were no significant differences among the treatments, (P>0.05) although there was a slight decrease in total bacterial count of the cooked sausages in comparison with the uncooked sausages. Citric acid slightly decreased the total bacterial count (log₁₀ cfu/g) compared to the control. The increase in citric acid and decontaminated water temperatures were associated with slight decrease in total bacterial count except cooked sausages treated with 1% citric acid. These results agree with the findings of Bolton et al., (2001) who found that, sodium acid and citric lactate. lactic combination, did not significantly reduce E. coli O 157: H 7 numbers when applied at different stages to burger manufacturing process. Commercial trimming and washing operations are not effective means of decontaminating beef carcasses

washing approximately halves the average numbers of *E. coli*, coliform and aerobic bacteria deposited in the surface of beef carcasses (Gill *et al.*, 1996).

The results of the bacterial count (log₁₀ cfu/g), (Table 5) indicate that, there was no bacteria found in the various treatments (0). This means that, the canning process at 225°F for 80 minutes for the sausages processed from the raw meat which was dipped in citric acid at (30, 80)°C or decontaminated water at (30, 80)°C was sufficient to provide complete sterilization of the product which could be stored for 3 months.

Canned foods, whether in tin or glass jars, won't keep forever. Commercial canners work under tightly controlled conditions with careful sanitation and just the right heat and timing periods, but there are still limits to how long food quality can be preserved (University of Wisconsin, 1996)

Table 4: Means and standard error (S.E) for Total bacterial count (log_{10} cfu/g) of the various raw and cooked sausage treatments*.

Independent Variables					
	Citric a	cid (1%)	Decontamin	S.E	
	30 ° C	80 ° C	30° C	80° C	
Uncooked sausages Cooked sausages	5.82 ^a 5.70 ^a	5.79 ^a 5.73 ^a	5.85 ^a 5.81 ^a	5.84 ^a 5.79 ^a	± 0.004

^a Means bearing similar superscripts are not significantly different (P>0.05).

Table 5: Effect of storage treatment on total bacterial count (log₁₀ cfu/g) of the various canned sausage treatments*.

	Treatment *					
Storage (Month)	Citric ac	eid (1%)	Deconta Wa	S.E		
	30 ° C	80 ° C	30 ° C	80 ° C		
0	0	0	0	0		
1	0	0	0	0	0	
2	0	0	0	0		
3	0	0	0	0		

^{*} Raw meat was dipped in 1% citric acid at 30° C and at 80° C and in Decontaminated water at 30°C and at 80°C (dipping and draining were performed for 1 minute in each treatment before processing)

Sensory evaluation

From the sensory point of view Table (6) shows no significant differences among treatments (P>0.05). In the colour, flavour, tenderness and over all acceptability of the canned sausages before storage (0 month) and after 3 months of storages. The brightest colour scores were achieved by the canned sausages tested before storage (0 month) in comparison with the canned sausages after 3 months of storage. Citric acid had no effect on the colour compared to the control. Also there was a slight increase in the colour scores for the canned sausages treated with citric acid at 80°c before storage (0 month)

compared to those treated with citric acid at 30°c and increase in colour scores with a decrease in decontaminated water temperature.

As for flavour, the results show that the highest scores were given by the canned sausages before storage (0 month) followed by that canned sausages after 1, 2, 3 months respectively, that means the flavour was decreased with increasing the storage period. Citric acid gave slight decrease in flavour compared to the control. Also there was slight increase in flavour with increase of citric acid temperature and slight decrease in flavour with increase of decontaminated

^{*} Raw meat was dipped in 1% citric acid at 30° c and at 80°C and in Decontaminated water at 30° C and at 80°C (dipping and draining were Performed for 1 minute in each treatment before processing).

water temperature. The results are in agreement with the findings of Quinton et al. (1997) who reported that, meat sticks (M.S, beef, pork) or stew sticks (S.S; beef, pork, vegetables) were formulated to pH 5.2 or 4.6 within capsulated lactic or citric acid. Meat sticks were preferred, although 25% of consumer panellists rated S.S as moderately acceptable or higher on a 9-point hedonic scale. Acid type (citric or lactic) did not affect consumer panel flavour scores. Assuming equal heat treatments, high temperature short time exposures to heat are less destructive upon flavour and texture than low temperature-long-time processes, within limits (Deserosier and Desrosier, 1977). Persson and Sydow (2006) found that, "aseptic" canning and especially, HTST (High Temperature Short Time)-Sterilization had a pronounced positive effect on aroma, in the late case of samples packed in thin layers (flexible pouches).

There was a decrease in tenderness with the increase of storage period. Citric acid seemed to reduce the tenderness compared to the control except the sample tested after 3 months treated with 1% citric acid at 80°c which appeared tendered than the control. There was no effect of increase in citric acid or decontaminated water temperatures on tenderness. Gover and Hostetler (1960) reported that, the rate of heat penetration in meat differs with cooking medium and seems to affect tenderness. On the other hand, they stated that it was quite evident that the time-temperature combination had an effect on the tenderness of the meat, probably causing both physical chemical changes in the connective tissue and in muscle fibres.

With regard to juiciness although the results show that there were no significant differences among the treatments (P>0.05).

the canned sausages showed a decrease in juiciness with the increase of storage period. Citric acid slightly decreased the juiciness compared to the control, also increasing temperature of citric acid lead to slight decrease in juiciness. As we mentioned before, there was a decrease in moisture content with increase of storage period and this is attributed to the decrease in juiciness. These results are in agreement with the report of Aberle et al., (2001) who reported that, the degree of shrinkage on cooking is correlated with loss of juiciness. The principal source of juiciness in meat, as detected by consumer, is the intramuscular lipids and the water content.

As for overall acceptably, the results indicate that the panellists could not pick any significant difference among the treatments (P>0.05). However, the acceptably decreased with the increase of storage period and this could be due to the decrease in flavour, juiciness, colour scores and tenderness. The results agree with the findings of Leblanc and Leblanc (2007) who found in their study of the effect of retort process time on the physical and sensory quality of canned lobster meat, that Quantitative Descriptive Analysis (QDA) panel for after taste, inner-grey colour and showed acceptance development of grey colour and after taste with longer processing time that is congruent with decreased acceptability. Ruiz et al., (1998) in their study of canning process and storage on lipid composition and palatability of canned sardines in olive oil for five years, found that the palatability of the sardines was significantly higher after 6 months of maturation than immediately canning and this quality maintained, for at least 5 years of storage.

Table 6: Means and standard errors (S.E) for sensory attributes of the various canned sausage treatments as assessed by panellists*.

	Treatment **								
Independent variables	Citric Acid 1%				Decontaminated water				
	30° c		80° c		30° c		80° c		S.E
	Storag	e (month)	Storage (month)		Storage (month)		Storage (month)		-
	0	3	0	3	0	3	0	3	
Colour	5.20 a	4.50 ^a	5.40 ^a	4.40 ^a	5.53 ^a	4.33 ^a	5.27 ^a	4.27 ^a	±0.06
Flavour	4.87 ^b	4.63 ^b	5.23 ^b	4.70 ^b	5.23 ^b	5.06 ^b	5.03 ^b	4.87 ^b	± 0.05
Tenderness	5.43°	4.53°	5.53°	4.47°	5.47 ^c	4.57 °	5.50°	4.27 ^c	± 0.05
Juiciness	5.27 ^d	4.40^{d}	5.22 ^d	4.33^{d}	5.67 ^d	4.90^{d}	5.23 ^d	4.50 ^d	$\pm~0.04$
Over all acceptably	5.13 ^e	4.47 ^e	5.30 ^e	4.60 ^e	5.40 ^e	4.53 ^e	5.47 ^e	4.77 ^e	± 0.07

^{abcd} Means in the same row bearing similar superscripts are not Significantly different (p > 0.05).

CONCLUSION

In the conclusion, the canned beef sausages will be of good quality and safe if processed without added nitrite and retorting at 107.2°C for 80 minutes.

Also it had better palatability characteristics at least for 3 months of storage at room temperature. So, we could get rid of nitrite which was caused some problems in the meat industry. The effect of 1% citric acid was not clear, however, it caused reducing in total bacterial count of the raw and cooked sausages.

Recommendations

Meat canning should be encouraged in Sudan and further researches should be applied to cover sufficient aspect and parameter such as:

Effect of canning on nutritive value of beef, Influence of different preservatives other than nitrite and citric acid on food poisoning bacteria.

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^{* 1-7} point scale was used, where 1= extremely dislike 7= extremely like.

^{*} Raw meat was dipped in 1% citric acid at 30° C and at 80°C and in Decontaminated water at 30°C and at 80°C (dipping and draining were performed for 1 minute in each treatment before processing).

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