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Antimicrobial Activity of Garlic (*Allium Sativum* L.) in The Preservation of Merguez, A Traditional Algerian Sausage

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A B S T R A C T

The objective of the present study is to determine the microbiological quality of Merguez-type sausages prepared and sold locally from meat offal and to study the antibacterial activity of fresh garlic (*Allium sativum* L.) during conservation at 4°C. Thus, fifteen representative samples of sausages are taken randomly from several butcher's shops in the commune of BBA, Algeria, then subjected to a bacteriological examination with reference to the standards established by the Algerian Ministry of Public Health. Bacteriological analysis revealed the presence of 6.88×10^5 CFU.g-^{1} of total aerobic bacteria, 5.39×10^5 CFU.g-^{1} of total coliforms, 2.23×10^5 CFU.g-^{1} of faecal coliforms, 2.43×10^3 CFU.g-^{1} of *Escherichia coli* and 1.8×10^5 CFU.g-^{1} of coagulase positive staphylococci, values higher than Algerian standards. The Addition of fresh garlic as an antibacterial preservative at concentrations of 0.06, 0.12, 0.18 and 0.24g.g-^{1} to ground beef samples and stored in the refrigerator at 4°C for 15 days. The addition of garlic to Merguez reduced significantly the presence of different bacterial groups during their refrigerated storage, compared to untreated meat by bringing it below the standards defined in the material. Thus, the use of garlic as a food additive at a concentration of 0.12 g.g-^{1} was sufficient to obtain levels under Algerian standards equal to 1.8×10^4 CFU.g-^{1} of total aerobic bacteria, 9.48×10^3 CFU.g-^{1} of Total Coliforms, 3.68×10^3 CFU.g-^{1} faecal Coliforms, 4.56×10^2 CFU.g-^{1} of *E.coli* 2.39×10^4 CFU.g-^{1} of coagulase positive Staphylococci. Through this study, we can conclude that adding garlic to Merguez reduces the aerobic bacterial load and thus increases the shelf life in a refrigerated at 4°C.

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Introduction

Microbial growth in meat and processed meat products probably hurry the lipid oxidation and other oxidative processes which cause deterioration in sensory characteristics such as flavour, and it is responsible for the nutritional value reduction (Saggiorato et al., 2012). Microbial contaminations comprise a major public health hazards and economic loss in terms of food poisoning and meat spoilage (Sallam et al., 2004).

In 2018, the Center for Disease Control in the United States estimated that 48 million people each year get sick from a foodborne illness, 128.000 are hospitalized, and 3000 dies (Center for Diseases Control and Prevention, 2018). This situation is all the more serious in emerging nations, with devastating economic consequences (Cohen et al., 2006). According to the Algerian Ministry of Health, more than 15.233 cases of food poisoning were recorded between 2016 and 2017, with 16 deaths, of which *Staphylococcus aureus* was the second leading cause (Algerian Ministry of Health, 2017).

In recent years, a huge number of researches have been performed for studying the antibacterial activity of natural products (Bal et al., 2019; Mohammed et al., 2020). Plants, particularly herbs and spices, are being given more attention. These days, there are more than one thousand plants with known antimicrobial efficacies, and over 30.000 antimicrobial outputs have been insulated from plants (Zheng et al., 2016; Sevindik et al., 2017; Mohammed et al., 2019). Spices have been utilized as food and flavouring hundreds years ago besides as remedy and food preservatives in recent times. A lot of spices; such ascinnamon, clove, curcuma, oregano, rosemary and thyme have been settled to keep food protected from pathogenic bacterial contamination (Jessica et al., 2017; Sevindik, 2018).

Garlic (*Allium sativum* L.) has been cultivated and used for culinary and medicinal purposes by many cultures for centuries. Most of its prophylactic and therapeutic effects are ascribed to specific oil- and water-soluble organosulfur compounds, which are responsible for the typical odor and flavor of garlic (Sivam, 2001). Apart from it, garlic has a wide spectrum of actions which include antibacterial, antifungal, antioxidant and beneficial effects on the cardiovascular and immune system of human (Mah et al., 2009; Gafar et al., 2012). Garlic contains many sulphur containing compounds and the most important of which is alliin (Chukwu et al., 2012).

The commodities we are interested in are specifically spicy lamb or beef-based raw sausage, a North African specialty called "Merguez". Merguez is regarded as the most popular variety of meat products widely consumed in Algeria. Due to its nutritional composition, sausage constitutes a rich medium that is very favorable to pathogen growth (Ed-Dra et al., 2018).

In the present work, we will carry out a microbiological diagnosis on the sanitary quality of sausages, widely manufactured and marketed by local butchers. We will try to identify the different microorganisms responsible for food poisoning and finally, to study the impact of food additives such as garlic in the observation and limitation of the growth of different bacterial groups in sausages Merguez type during cold storage**.**

Material and Methods

Sampling

During 2 months from April 4th to May 30th 2021, 15 fresh sausage samples were collected from five different butcheries in Bordj Bou Arreridj, Algeria. The samples were taken within four hours after its exposure for selling. The samples (100 g) were conditioned in sterile bags, maintained in a refrigerator containing frozen cooling blocks. The main microbiological groups of the sausages were analysed immediately upon their arrival.

For study antimicrobial effect of fresh white local Garlic (*A. sativum*), 15 samples of Merguez (100g) prepared in Butcher with Incorporation of Garlic according to this distribution: A lot of 3 samples prepared without Garlic, as a control. Four lots each of which consists of 3 samples prepared with increasing concentrations of Garlic, ranging from 0.06 to 0.24 g/g. Samples were kept in a refrigerator (4 °C) for 15 days. The same microbiological groups of the sausages were tested over the storage period for the evaluation of antimicrobial impacts of the fresh garlic.

Microbiological Analysis

At the laboratory, each sample initially separated into 5 units then was cut out separately in small pieces in sterile Petri dish, a Stomacher sachet is tarred and 25 g of each unit were weighed exactly there. Then, 225 mL of a solution of tryptone salt (TSE) (Pasteur Institute, Algeria) were introduced into the sachet. The unit was crushed during 2 to 3 min in Stomacher. The supernatant obtained after crushing was recovered in a sterile bottle (it is the stock solution (SM) of concentration 10^{-1}. The latter was left at rest during 45 min, to allow the reactivation of the shocked or stressed microorganisms.

Serial ten-fold dilutions were prepared using TSE (Pasteur Institute, Algeria) starting from the stock solution in accordance with the standards ISO 6887-1 (Standard ISO 6887-1, 1999).

The bacteria investigated and counted were the total viable aerobic bacteria, total coliforms group, faecal coliforms group, thermotolerant, *E. coli* and coagulase positive staphylococci.

The total viable aerobic bacteria evaluation were performed according to (ISO 4833, 2003), which aerobic plate count (APC) (enumerated on plate count agar (PCA) and incubated for 24–48 h at 30 °C).

Total coliforms group were determined using the Most Probable Number (MPN) technique. MacConkey and Brilliant Green Bile Lactose Broth (BGB) were used for presumptive and confirmed tests for coliforms respectively. For faecal coliform presence, positive tubes from Brilliant Green Bile Lactose Broth (BGB) medium were subcultured into *E. coli* broth medium and then incubated at 44.5 °C for 48 hours. The Most Probable Number (MPN) for both coliforms and faecal coliforms was recorded using the MPN table. The indol production by *E. coli* was carried out by the Mac Kenzie test (ISO 7251, 2005).

The coagulase positive staphylococci were cultivated in Giolitti-Contoni broth supplemented with potassium tellurite and Chapman medium were used for presumptive and confirmed tests for staphylococci respectively. The bacterial number is evaluated after an incubation of 48 h at 37 °C, their identity was confirmed by Gram stain, and the enzymatic based research of catalase and coagulase (Bourgeois et Leveau, 1991).

All microbial counts were expressed as decimal logarithm of colony forming units per gram ($Log_{10}CFU.g^{-1}$). The results were compared with the criteria required by the Algerian inter-ministerial decree of July 2nd 2017, related to the quality of foodstuffs. The maximum concentrations accepted for the counted bacteria are: Total Aerobic bacteria 5.10^6 $CFU.g^{-1}$, Total coliforms and faecal coliforms 10^4 $CFU.g^{-1}$, *E.coli* 5.10^2 $CFU.g^{-1}$, coagulase positive staphylococci 10^3 $CFU.g^{-1}$ (Interministerial decree, 2017).

Statistical Analysis

The results were expressed in terms of mean and standard deviation (SD) of mean. The means were compared by one way ANOVA followed by Duncan's multiple range Test at 5% level of significance (Duncan, 1995). A probability value of $P<0.05$ was described as significant using SPSS 25 statistical software (SPSS 25, 2020).

Results

The total mesopholic aerobic flora always tells us about the hygienic quality. On the PCA medium, we obtained a collection of different colonies of sizes This germs form small round colonies of varying diameter between 1 and 10 mm, with a smooth, opaque and whitish appearance (Figure 1, A).

Total and faecal coliforms were appeared in small colonies inferior to 5mm, coloured purplish pink and lenticular in shape (Figure 1, B, C and D). The two groups of coliforms were fermented lactose with the production of gas on BGB.

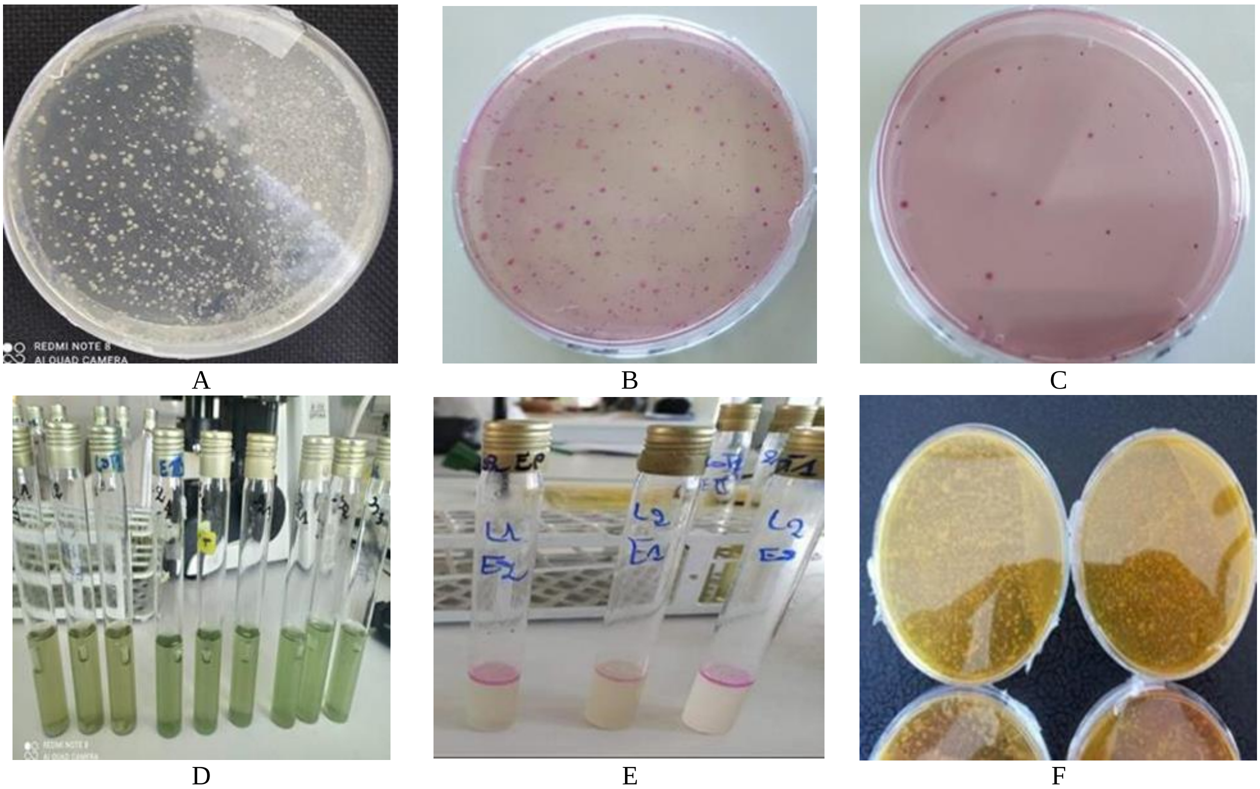

Figure 1. Microbiological groups of the sausages analysed; A) Total Aerobic bacteria; B) Total coliforms; C) Faecal coliforms; D) confirmed tests for coliforms; E) Indole production; F) Staphylococci.

Escherichia coli are a common Gram-negative microorganism which may be present in food and water samples. The presence of *E. coli* cells may be indicative of faecal contamination, although their ingestion does not necessarily have adverse effects on health. Nevertheless, certain strains are responsible for diarrhoeal disease and can also lead to more serious forms of illness. EC Broth is a selective medium and confirmatory test for *Escherichia coli* from food and environmental samples. It is also suitable for the enumeration of presumptive *E. coli* using the most probable number technique. Indole production in peptone water is major signs of presence of *E. coli* (Figure 1, E).

The coagulase positive staphylococci consider as were cultivated in Giolitti-Contoni broth supplemented with potassium tellurite and Mannitol salt medium were used for presumptive and confirmed tests for staphylococci respectively. The bacterial number is evaluated after an incubation of 48 h at 37°C; their identity was confirmed by Gram stain, and the enzymatic based research of catalase and coagulase.

Coagulase positive staphylococci (*S. aureus*) are considered pathogenic bacteria and their presence in food indicates poor handling conditions during preparation as well as the poor hygienic quality of the materials used. Coagulase positive staphylococci are formed a small round colonies with a diameter of less than 1mm, with a yellow appearance surrounded by a yellow halo on the medium (Figure 1, F).

Microbiological quality of Merguez

One of the main elements limiting the shelf life of food products is the development of microbial groups such as pathogens and spoilage microorganisms during the preservation. Meat products through their favorable growth conditions for a broad spectrum of microorganisms are often prone to be degraded by microbial activities.

Counts of mesophilic aerobic microorganisms are commonly used to indicate the sanitary quality of food (Franco and Landgraf, 2005) and to detect the number of aerobic or facultative mesophilic bacteria, which are present both in vegetative form and also as spores in food.

The microbiological analysis (table 1) shows a variable and very highly significant presence at 1% level intra and inters bacteria groups in the different Merguez samples analysed. It can be seen that in terms of the means of the total aerobic mesophilic counts 5.84 $Log_{10}CFU.g^{-1}$; majority of samples (66.66%) showed a value less than or equal to the criterion fixed by Algerian standards. The results also showed that sample number 5 had the least amount of the total aerobic bacteria 5.54 $Log_{10}CFU.g^{-1}$ (Figure 2).

Out of the 15 samples, 100% samples present a load in total coliforms, thermotolerant coliforms and coagulase positive staphylococci higher than the fixed standards; theirs averages concentrations were 5.73; 5.34 and 5.25 $Log_{10}CFU.g^{-1}$, respectively, while sample 5 carried the lowest amount of both total coliforms and *S. aureus* where the following values are recorded 4.41 and 4.75 $Log_{10}CFU.g^{-1}$, respectively (Figure 2).

Regarding thermotolerant coliforms, sample 10 was least pollution at a rate equal to 4.75 $Log_{10}CFU.g^{-1}$. In the same way, 93.33% of the samples show a load in *E. coli* higher than the standards, the average number is 3.83 $Log_{10}CFU.g^{-1}$. For samples less contaminated with *E. coli*, we found sample number 6 with 2.60 $Log_{10}CFU.g^{-1}$ (Figure 2).

Table 1. Results of one way ANOVA Analysis of microflora counted in Merguez sausages

Source	Sum of squares type III	ddl	Medium square	F	Signification
Total Aerobic Bacteria	0,576	15	0,038	3871,675	0,000
Total Coliforms	15,937	15	1,062	255,032	0,000
Faecal Coliforms	14,696	15	0,980	243,924	0,000
E. coli	3,449	15	0,230	26,186	0,000
S. aureus	21,549	15	1,437	46,598	0,000

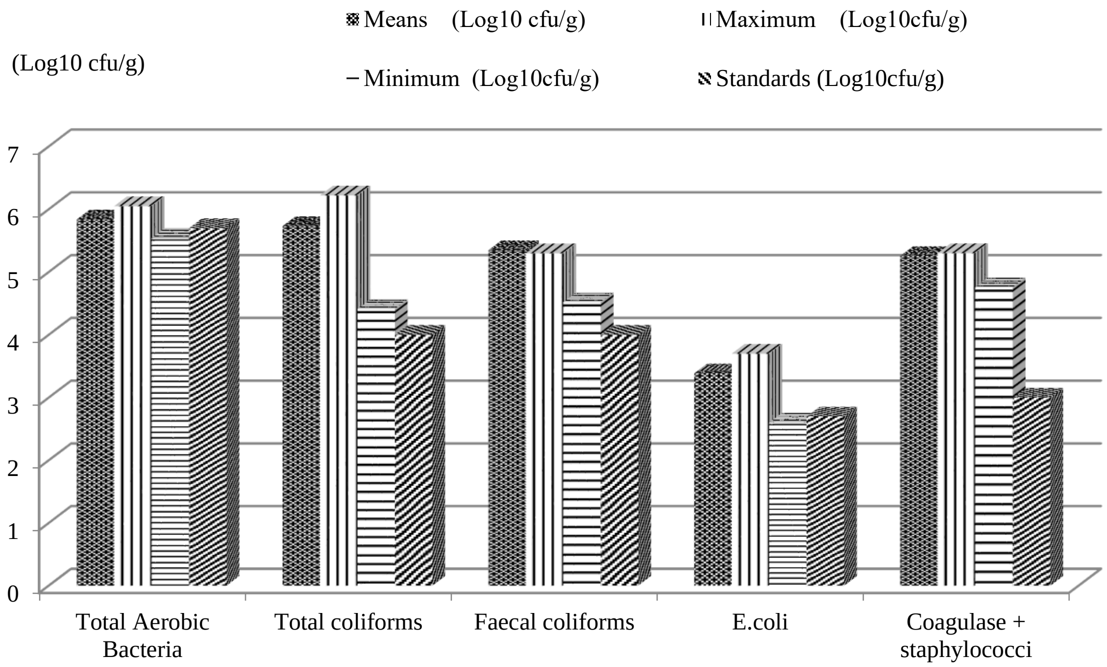

Figure 2. The average values of the microflora counted in Merguez sausages in Log10UFC/g compared to the required standards

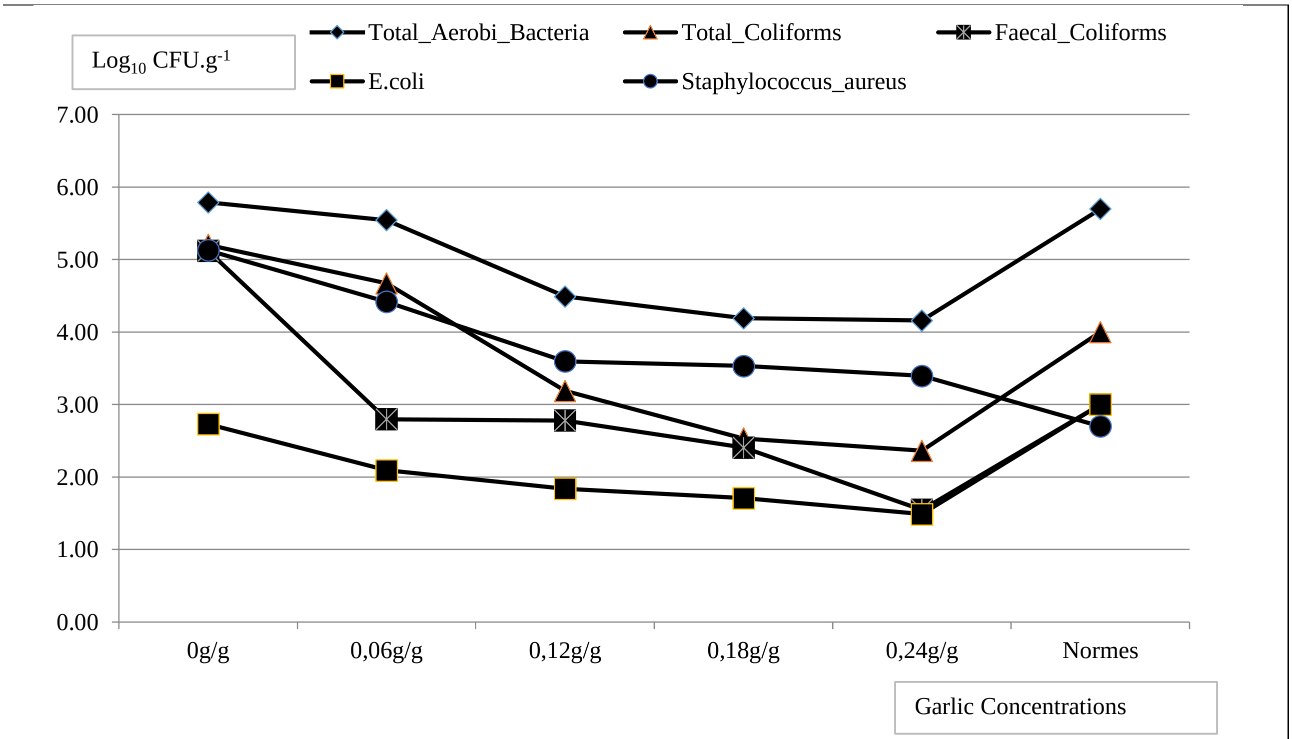

Figure 3. Effect of five concentrations of fresh garlic (FG) on the number of colonies of different bacterial groups after 15 days conservation of sausages stored at 4 °C

Antimicrobial Activity of Garlic Incorporated in Sausage

During the final period of storage (15 days) the mesophilic aerobic microorganism count rose and the treatments with added fresh Garlic showed significant difference compared with the control (5.79 $Log_{10}CFU.g^{-1}$); Therefore, it was possible to see the influence of the addition of Garlic in the sausages because the lowest counts for mesophilic aerobic bacteria occurred in the sausages with added fresh white Garlic. These values were 4.48; 4.19 and 4.15 $Log_{10}CFU.g^{-1}$ on day 15 for the treatments with 0.12; 0.18 and 0.24 g/g of added Garlic, respectively (Figure 3).

During this period visual changes in Total coliforms were observed in greater quantity in the standard 5.21 $Log_{10}CFU.g^{-1}$ than in the other treatments. The action of Garlic can be seen in reducing the number of total coliforms compared to the standard treatment. This reduction was very clear in relation to the treatment with the addition of 0.24 g/g of sausage compared to the standard treatment during the period of storage, where the mean equal to 2.36 $Log_{10}CFU.g^{-1}$(Figure 3).

The average count of faecal coliforms there was significant difference between the treatments. The use of Garlic was more effective in controlling these microorganisms; the values were less than the criterion fixed by Algerian standards (3 $Log_{10}CFU.g^{-1}$). These values were 2.8; 2.78; 2.42 and 1.55 $Log_{10}CFU.g^{-1}$ on day 15 for the treatments with 0.06; 0.12; 0.18 and 0.24 g/g of added Garlic, respectively (Figure 3).

The main pathogenic microorganisms which potentially could have been in the products developed in the present study were *S. aureus* and *E. coli*. The latter can enter slaughterhouses through live animals and also humans working in the premises (Birzele et al., 2005). During the final period of storage (15 days), it was possible to see the influence of the addition of Garlic in the sausages because the lowest counts for *E. coli* occurred in the sausages with added fresh white Garlic. The values were less than the criterion fixed by Algerian standards (2.69 $Log_{10}CFU.g^{-1}$). These values were 2.12; 1.88; 1.73 and 1.49 $Log_{10}CFU.g^{-1}$ on day 15 for the treatments with 0.06; 0.12; 0.18 and 0.24 g/g of added Garlic, respectively (Figure 3).

The count for coagulase-positive *Staphylococcus* was showed difference between the treatments during the storage period. All the treatments in the present study remained higher than the legally limit (3 $Log_{10}CFU.g^{-1}$) during storage. These values were 5.13; 3.46; 3.60; 3.53 and 3.41 $Log_{10}CFU.g^{-1}$ on day 15 for the treatments with 0; 0.06; 0.12; 0.18 and 0.24 g/g of added Garlic, respectively (Figure 4).

To illustrate the relationship between the number of colonies of bacterial groups and the concentration of Garlic added to Merguez sausages, we have calculated the coefficient of determination R^2 which measures the linear intensity between the amounts of bacterial groups and concentration of Garlic. Thus, R^2 recorded after garlic treatment were equal to 0.93, 0.86, 0.97, 0.96 and 0.95 for Total Aerobic Bacteria, Faecal Coliforms, Total Coliforms, *E. coli* and *S. aureus*, respectively. Which means that 93, 86, 97, 96 and 95 % of variations in the number of these Bacterial groups can be explained by the influence of garlic concentration in Merguez sausages (Figure 4).

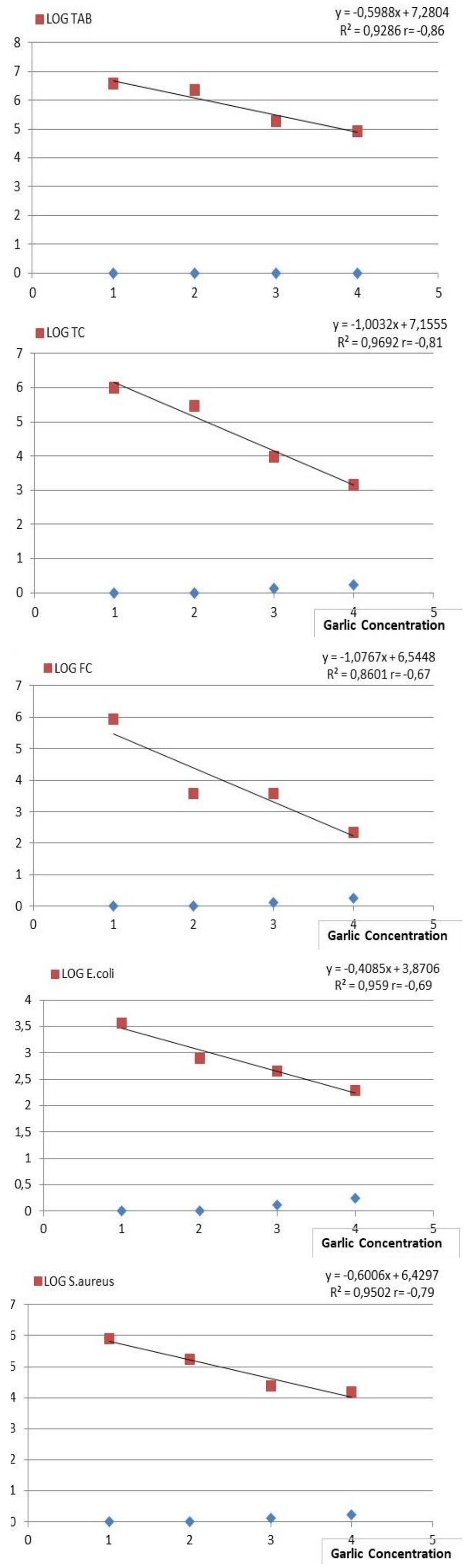

Figure 4. Relationship between the concentration of Garlic (x) and the count of bacterial groups (y) in Merguez sausages

While regression coefficient (r) calculated after garlic treatment were equal to -0.86, -0.81, -0.67, -0.69 and -0.79 for Total Aerobic Bacteria, Faecal Coliforms, Total Coliforms, *E. coli* and *S. aureus*, respectively. This shows that when the concentration of garlic increases, the number of bacterial colonies decreases for all groups, demonstrating a strong correlation between the two variables.

Discussion

Quality assessment of Merguez sausage

The Merguez sausage quality was evaluated and interpreted with microbiological criteria for foodstuffs according to the Algerian Food Codex for assessing the microbiological in the artisanal sausage. Merguez quality is attributed as "unsatisfactory quality" with contamination levels exceeding the norms.

Total aerobic Mesophilic-Flora (TAMF)

It provides information on the overall degree of meat contamination. The results (5.84 $Log_{10}CFU.g^{-1}$) are superior to those obtained previously in Algeria equal to 3.70 $LogCFU.g^{-1}$ by Hahemi et al. (2019) and in Morocco by El Allaoui et al. (2012), with 5.54 $Log_{10}CFU.g^{-1}$. The results are similar to that found by Dalla Santa et al. (2012) in Brazil, with 6 Log_{10} $CFU.g^{-1}$. The differences can be explained by the degree of hygiene monitoring at different stages of the meat production and preparation.

Total Coliforms (TC)

The determination of *Enterobacteriaceae* is an essential element in quality assessment of slaughter hygiene, slaughter-houses and consumption meats (Zweife et al., 2008). The average charge in TC is of 5.73 $LogCFU.g^{-1}$, the results are similar to that found by El Allaoui et al (2012), 5.74 $Log_{10}CFU.g^{-1}$, and superior to that found by Ed-Dra et al. (2018) in Meknes (Morocco) 5.05 $LogCFU.g^{-1}$. The high rate of TC in the beef sausage can be due to inadequate cleaning and disinfection, contaminant materials (ex: packaging), bad conditions of storage, source of untreated water, and deficiency in disinfection treatment.

Faecal Coliforms (FC)

Faecal coliforms live in the human and animal intestines; their presence would reflect bad conditions during the slaughter operation. They survive difficulty for a long time outside of the intestine so; their presence indicates a recent faecal contamination (Joffin and Joffin 1999). The average of the enumerations of these faecal originated bacteria is 5.34 $Log_{10}CFU.g^{-1}$, these results is very higher than reported by Hamiroune et al. (2017a), in the region of M'Sila (Algeria) and Cohen and Karib (2006) in Casablanca which present averages of 2.0 and 3.7 $Log_{10}CFU.g^{-1}$, respectively.

This average of high contamination is probably due to the improper handling during slaughtering operation. According to Mescle and Zucca (1998), the contamination occurs during the chopping and the manufacturing. These operations lead to a homogenization of the flora of the various ingredients and to a modification of the structure of the products. That makes it possible the contamination of surface to be introduced into the mass. Meat contamination is unavoidable during slaughtering operations, especially during evisceration by the workers hands, equipment, the water used or by the eventual rupture of the gastric reservoir.

Escherichia coli

Cutter (2000) has been found that bacterial contamination, such as *E. coli* O157:H7 and *Listeria monocytogenes*, impacted meat safety. Based on these observations, it appears that the oxidations of OxyMb and lipid as well as microbial contamination are serious concerns for meat producers and consumers. Results obtained show that 93.33% of the sausages studied are highly contaminated with *E. Coli* than the standards with a load of $6.10x10^2$ (2.7 $Log10CFU.g\text{-}^1$) which are 3 and 3.69 LogCFU lower than those obtained by Dalla Santa et al. (2012) and Ed-Dra et al. (2018), respectively.

Coagulase-positive Staphylococci

Their detection and enumeration allow assessing the risk of food for consumers, given that they are the major species which can eventually produce a proteinic enterotoxin responsible of food poisoning (Joffin and Joffin, 1999). Coagulase positive staphylococci are considered to be pathogenic bacteria and their presence in food is due to poor handling conditions during preparation as well as poor hygienic quality of the material used in the food chain (Salihu et al., 2010). Our results (5.25 $LogCFU.g^{-1}$) are higher than those of Scagna et al. (2000) who reported an average level of contamination of 1.1 $LogCFU.g^{-1}$. Besides, they remain clearly superior to those obtained by El Allaoui et al. (2012), Ed-Dra et al. (2018), Hamiroune et al. (2017a) and Hamiroune et al. (2017b) with an average of 4.3; 3.42; 2.2 and $Log_{10}CFU.g^{-1}$, respectively.

These results reveal a lack of respect of the good practices of hygiene and manufacture as well as temperature of storage. Moreover, the medical status of staff plays a fundamental role in the contamination of the foodstuffs by the coagulase positive staphylococci (International Biological Standards Commission Relating To Food, 1974).

Antibacterial Activity of Garlic

Garlic, as an anti-bacterial agent, is effective against many more gram negative and gram positive bacteria like *Helicobacter pylori, E. coli, Lactobacillus casei* and that this effect is sourced from allicin inside it (Celli et al., 1996; Lemar et al., 2005). The antibacterial effect garlic apparently results from interaction of sulphur compounds, like allicin, with sulphur (thiol) groups of microbial enzymes (such as trypsin and other proteases), leading to an inhibition of microbial growth. Many bacterial strains, both gram-positive and gram-negative, can be inhibited with garlic, and some strains were inhibited much more strongly by allicin or garlic extract compared to antibiotics (Bakri and Douglas, 2005).

Adding fresh garlic as an antibacterial preservative at concentrations of 0.06, 0.12, 0.18 and 0.24 g.g-1 to samples of ground beef and stored in the refrigerator at 4 °C for 15 days has Significantly reduces the presence of different bacterial groups during their refrigerated storage, compared to untreated meat by bringing it below the standards defined in the material. Confirming the antibacterial effect of garlic, which corroborates with the results found in other research (Karuppiah and Rajaram, 2012). Garlic is also rich in anionic components such as nitrates, chlorides and sulfates and other water soluble components found in plants and these components may have antimicrobial properties (Shobana et al., 2009). Allicin has been found to be the active ingredient in garlic and it works as an antimicrobial agent by inhibiting DNA and protein synthesis moderately and inhibiting RNA synthesis completely as a primary target (Rahiman et al., 2011). Previous authors have described the antibacterial activity of garlic extract against microorganisms. Bulbs belonging to the Allium genus had the most antibacterial activity against *Streptococci* (Ohara et al., 2008; Alsaid et al., 2010; Silva and Fernandes, 2010; Daka, 2011); *Escherichia coli*, *Salmonella typhi*, *Shigella flexineri*, *Proteus mirabilis* (Shobana et al., 2009); and *Vibrio parahaemolyticus*, *Escherichia coli* and *Staphylococcus aureus* (Vuddhakul et al., 2007). Few studies have shown some bacteria to be resistant towards garlic extract and these include *E. coli* and *Staphylococcus aureus* (Esimone et al., 2010).

Thus, the addition of garlic as a food additive at a concentration of 0.12 gg-^{1} was sufficient to obtain levels below Algerian standards (1.8 10^4 CFU.g-^{1} of total aerobic bacteria, 9.48 10^3 CFU.g-^{1} of total coliforms, 3.68 10^3 CFU.g-^{1} Faecal coliforms, 4.56 10^2 CFU.g-^{1} of *E. coli* 2.39 10^4 UFC.g-^{1} of coagulase staphylococci positive). Therefore and through this study, we can conclude that adding garlic to Merguez reduces the aerobic bacterial load and thus increases the shelf life in a refrigerator at 4 °C. These results were in agreement with those previously reported (; El-Khateib et al., 1987; Kalkan et al., 2017; Mahros et al., 2021). Likewise, Sallam et al. (2004) reported that adding FG (30g/kg) or garlic powder (9 g/kg) to chicken sausages significantly reduced their CPAs and subsequently extended their shelf life to 21 days. In ground beef with added garlic, the reduction in APCs, compared to control samples, can be attributed to organosulfur compounds and allicin from their precursor in garlic (Kyung, 2012).

Conclusion

This study concluded that addition of garlic to Merguez sausages reduced their aerobic bacteria and increased their shelf life in refrigerated storage (at 4 °C for 15 days). The addition of 0.12g.g-1 to Merguez sausages reduced the bacterial loads below the required norms, remains to be verified the acceptability of such concentration and such flavour by consumers. It is therefore interesting to incorporate garlic as a preservative in meat products such as sausages, as a natural herb, to reduce bacterial contamination and extend shelf life instead of chemical preservatives.

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