RESEARCH ARTICLE

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Hygienic Quality of Meat Products in Qazvin Province during (2011-2014), Iran

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Background & Aims of the Study: One way to increasing shelf life of meat is the production of sausages along with additives. One of these additives contains nitrite that in an acceptance limit is not a threat for human health but if it's amount is higher than standard, it will react in the body with secondary amine and the result is the formation of carcinogenic nitrosamines. Also, the consumption of healthy and safe food is concerned for every consumer. That's why we decided to examine Nitrite, Carbohydrates, Fat, Protein, Humidity, microbial contamination and exterior features of the sausages which were produced in Qazvin province.

Materials and Methods: The study population consisted of sausages produced in Qazvin province from 2011 to 2014. A total of 74 samples of products that have been produced in the factories of meat products of Qazvin, distribution system and retail level were collected. Additive composition, microbial contamination and exterior features were examined according to standard methods of Iranian national standards No 2303.

Results: The overall result showed that all products comply with their nitrite acceptance limit. Of course, the amount of humidity (13.84%) and carbohydrates (39.62%) were infringement but the two combinations no adverse effects on human health and microorganism counts determined in overall processing were not at harmful levels for human health and microbial contamination of all meat products of our study were acceptable.

Conclusion: According to the results, we should have more control on the hygienic quality of meat products over the production, storage and supply periods in this area.

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Background

Using the meat as a food dates back to the beginning of human history. With increasing prosperity increased diversity in human tempers. That's why food manufacturers such as meat products for the presence in household consumption basket to produce meat products and various dimensions of their greed's. Over the time, the researchers came to the conclusion that processing of meat into meat products improves the product's shelf-life and microbiological safety (1).

Sausages are sustainable mix of cattle slaughter (according to Islamic law), fat and water, along with other materials in natural or synthetic coatings packed in good condition. After an appropriate heat treatment, other processes needed for human food consumption is prepared. With the advancement of science and the importance of increasing the shelf life of food, the researchers had to develop chemical

preservatives to increase shelf life of food. Among these, nitrites are preservatives which were initially due to the favorable effects on the color and flavor of meat products had been used; but later it became more important effect of clostridium. The indiscriminate use of chemical sustaining food producers has led to regulatory agencies to determine the permissible acceptance limit for chemical substances used in their food. Nitrite is a compound that exists in living systems and also, it is one of the most active intermediate species in the nitrogen cycle, where suffers a surprising metamorphosis, from a vilified substance that generates carcinogenic N-nitrosamines with amines and amides present in the stomach, to a life-saving drug that liberates a protective agent (nitric oxide or NO) during the hypoxic events (2).

When our daily excessive ingestion of nitrite ions from food and water is accumulated in the gastrointestinal tract, the nitrite is absorbed into the bloodstream and change the blood pigment hemoglobin (the oxygen-carrying part of blood) into the met-hemoglobin, which is not an oxygen carrier, thus the ability of the blood to carry oxygen throughout the body tissues is limited (3).

As a high reactive compound, nitrite can function as an oxidizing, a reducing or a nitrosylating agent, and can be converted to a variety of related compounds in meat, including nitrous acid, nitric oxide and nitrate. However, due to their bacteriostatic and bactericidal actions (4), nitrite has been recognized for centuries and is still used today as color fixer (5) and protection agent against Botulism in preservation of meat products because of its use in the first step strongly inhibitory effect on the anaerobic bacteria (Clostridium botulinum) and in the second step is a capacity to control the level of other micro-organisms (such as Listeria monocytogenes) (6).

The World Health Organization has reported that the fatal dose of nitrite ingestion is between 8.7 µM and 28.3 µM (7).

In the same time, the Legislation of the European Union (8) suggests a maximum supplement which is allowed in the first stage of the food processing, and it imposes a maximum residue of 50 mg/kg nitrites and 250 mg/kg nitrates for those meat products (expressed as $NaNO_2$), which has not been treated thermally.

Nitrites and nitrosamines effects on human health due to the formation of nitrites used in meat processing about one-fifth less than two decades ago (9). The formation of nitric acid in the human body is very complex. Nitrates and nitrites from food and water, synthesis of nitric acid, nitric acid entering the blood, saliva, conversion of nitrate to nitrite by bacteria in saliva and re-conversion to nitric acid in the blood, including factors that are involved in the formation of some negative aspects associated with particular processed meats are such as the high fat and cholesterol content and the possible cancer promoting effects related to high intakes (10,11).

The report of the World Cancer Research Fund (12) and the American Institute for Cancer Research (AICR) indicated a weak but a significant relationship between increased intake of red and processed meats and an increased risk of colorectal cancer. As a consequence, experts recommended limiting red meat and avoiding processed meat intake (13).

Also, the microbial contamination which is another factor affecting the quality of meat products is much higher standard threatens for consumer health. In Kamkar and Associates study, the average chemical agents, moisture, fat, protein and ash in Iran were compared with standard acceptance limit. The results of this study showed that a standard for the production of meat products cannot be fully complies in Iran (14).

Also, in the study of Babaei et al. (2011) the average nitrate in sausage was 81.14 and 115.1 mg/kg, respectively, which have said in this case the acceptance limit is nitrite 60 ppm (15).

Therefore, it should be a careful monitoring and continuous, integral part of maintaining health and microbial quality of meat products. But given the above people still have an interest in consumption of meat products which can be the source of this interest is the increase in population and new conditions of urban societies that physical access to fresh food such as fruit and vegetables lowered and people are forced for economic prosperity for their families, most of the day to spend outside the home and at work; So, to have the fast food. This food because of their business should be prepared attractive for the customer, the high-fat, spicy and salty. Its diversity purchase price for all people of the wealthiest to the most underprivileged class of society to provide comfort. In the study of Yaghoubi Far et al. (2009) mentioned per capita consumption of sausages for every Iranian citizen is about 4 kg (16).

Aims of the study:

The aim of this study was to assess the hygienic quality of meat products in Qazvin province of Iran during (2011-2014).

Materials & Methods

Sampling: This research is a descriptive cross sectional study and the study population consisted of sausage and kielbasa produced in Qazvin province of Iran from 2011 to 2014. A total of 74 samples of meat products that have been produced in the factories of meat products of Qazvin, distribution and retail level were collected.

We evaluated the additive composition, microbial contamination and the exterior features according to the methods of Iranian National standard in 2303.

Sampling in accordance with the principles of IR-ISO NO 690 sampling took place. In accordance with the specified form in this sampler instruction, sampling, transport and delivery to the laboratory conditions reflected examples and to complete the testing of the samples to measure the quality of results from quality and safety of each product and the comparison of them with the Iranian national standard the number 2303 were done. Tests used for various meat products assessment are showed in the following tables. Tests used for bacterial and fungal assessment of sausage and kielbasa, acceptable limit and reference methods are shown in the following tables.

The purpose of the microbial status and health evaluated in this study were organoleptic characteristics such as texture, the appearance, packaging, odor, color, miscellaneous and markings that make up the general features and moisture, nitrite and carbohydrates that make up the chemical properties and Staphylococcus aurous, Escherichia coli, Salmonella, total count, Clostridium perfringens, coliforms and molds and yeasts that form the biological characteristics.

Tests used for bacterial, fungal and compounds forming assessment of Sausage and kielbasa, acceptable limit and reference methods are shown in the following tables 1 and 2.

Total counts of microorganisms (Standard No. 5272): From 1/10 diluted sample 1 mL was poured into a sterile plate and then 15 cc of PCA medium (with a temperature of 44°C) was added and stirred. After hardening of medium, plates were incubated upside down at 30°C for 24-48 hours. After the incubation period, colonies were counted. In the absence of colony growth, result was reported in less than dilution was cultured (17).

Coliform counted (Standard No. 9263): From 1/10 diluted sample 1 mL was poured into in a sterile plate and then 15 cc of VRBA medium (With a temperature of 44°C) was added and stirred. After hardening of medium, plates were incubated upside down at 37°C for 24-48 hours. After incubation period, colonies were counted. In the absence of colony growth result was reported in less than dilution was cultured (18).

Escherichia coli Count: For counting of this bacteria EMBA was used (Standard No. 2946) (19).

Staphylococcus aureus were counted (Standard No. 6806-1 and 3):

Standard No. 6806-1 is used if the limit upper than 100: 1 ml of prepared dilution distributed on Baired-Parker agar medium containing egg yolk and incubated at 37 ° C for 1-2 days. Black colonies with a colorless zoon are Staphylococcus aureus. To confirm the colonies, coagulase test is used. Staphylococcus aureus colonies are coagulase positive (20).

Clostridium perferingens were counted (Standard No. 2197): From 1/10 diluted sample 1 mL was poured into in a sterile plate and then 15 cc of Sulfite agar medium (With a temperature of 44°C) was added and stirred. After hardening of medium, plates were incubated upside down at 37°C for 24-48 hours. Colonies were counted after incubation period. In the absence of colony growth result was reported in less than dilution was cultured. Black colonies are due to reduction of sulfite by Clostridium (21).

Mold and Yeast were counted (Standard No. 10899 and 3): On sterile plates containing DRBC, 0.1 mL of desired dilution distributed. Molds colonies were counted after 3-5 days of incubation at 25 °C [10].

From desired dilution of sample, 1 mL was poured on sterile plate containing DRBC and after that 15 mL of Y coca medium (with a temperature of 44°C) was added and stirred. After hardening of medium plates were incubated upside down at 25 ° C for 3-5 days (22).

Salmonella Isolation (Standard No. 1810): This experiment was done in three days. First day: 5 g of sample is added in 45 ml and mixed well (0.1 dilution). Second day: 1 mL of diluted sample is added into tubes containing 10 ml tetrationate broth. Third day: 0.1 ml of prepared solution of second day are cultured on SS agar medium and incubated at 37 °C for 24-48 hours (23).

Table 1) Tests, acceptable limits and reference methods for bacterial and fungal assessment of Sausage and kielbasa

No	Reference method	Acceptable limit	test
1	Iranian national standard no. 5272 (17)	Max 10^5	Total count (CFU/g)
2	Iranian national standard no. 9263 (18)	Max 10	Coliforms (CFU/g)
3	Iranian national standard no. 2946 (19)	Negative	*Escherichia coli* (CFU/g)
4	Iranian national standard no. 6806-1 and 3 (20)	<10	Coagulase Positive *Staphylococcusaureus*(CFU/g)
5	Iranian national standard no. 2197 (21)	Max 50	*Clostridium perferingens*(CFU/g)
6	Iranian national standard no. 10899 (22)	Max 10^2	Mold and yeast (CFU/g)
7	Iranian national standard no. 1810 (23)	Negative	*Salmonella* per 25 g

Determination of nitrite (Standard No. 16721) (24).
Determination of Fat (Standard No. 742) (25).
Determination of Protein (Standard No. 924) (26).
Determination of Humidity (Standard No. 745) (27).

Table 2) Tests, acceptable limits and reference methods for Nitrite, Carbohydrates, Fat, Protein and Humidity in assessment of Sausage and kielbasa

No	Reference method	Acceptable limit	test
1	Iranian national standard no. 16721 (24)	<80	Nitrite
2	Iranian national standard no. 742 (25)	<23	Fat
3	Iranian national standard no. 924 (26)	11.5-9.5	Protein
4	Iranian national standard no. 745 (27)	<60	Humidity

Results

The results of the Chemical composition, microbiological contamination and compare general features were presented in tables 3, 4 and 5.

Most of the violations on the formulation related to moisture (86.15%) and carbohydrates (60.37%) are the main reasons for the increase of this type of crime, is the violation of economic fraud because they add water and carbohydrates to make the cheaper product; then they added the meat to the formulation. Excessive moisture can reduce the shelf life of the product due to increased water activity is also due to wetting in order to disassemble the product surface texture and shape of the product to create air bubbles under the cover of the package and conditions for the growth of aerobic germs is provided as mold. That is why the greatest change in the exterior features (4.16%) and the apparent color (4.16%).

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Table3) Compare Nitrite, Carbohydrates, Fat, Protein and Humidity in samples of meat products (n=74).

Product Name		Nitrite		Carbohydrates		Fat		Protein		Humidity	
		Standard	Non-standard	Standard	Non-standard	Standard	Non-standard	Standard	Non-standard	Standard	Non-standard
Sausage	Number	50	0	32	18	50	0	50	0	43	7
	Percent	100	0	56.25	43.75	100	0	100	0	83.72	16.27
kielbasa	Number	24	0	21	3	24	0	24	0	22	2
	Percent	100	0	85.71	14.28	100	0	100	0	90.90	9.09
Total	Number	74	0	53	21	74	0	74	0	65	9
	Percent	100	0	60.37	39.62	100	0	100	0	86.51	13.84

Table 4) Microbiological contamination of meat products samples (n=74).

Product Name		*Staphylococcus aureus*		*E. coli*		*Salmonella*		*Clostridium perfringens*		Coliforms		Total Count		Mold and Yeast	
		Standard	Non-standard	Negative	Positive	Negative	Positive	Standard	Non-standard	Standard	Non-standard	Standard	Non-standard	Standard	Non-standard
Sausage	Number	50	0	50	0	50	0	50	0	50	0	50	0	50	0
	Percent	100	0	100	0	100	0	100	0	100	0	100	0	100	0
kielbasa	Number	24	0	24	0	24	0	24	0	24	0	24	0	24	0
	Percent	100	0	100	0	100	0	100	0	100	0	100	0	100	0
Total	Number	74	0	74	0	74	0	74	0	74	0	74	0	74	0
	Percent	100	0	100	0	100	0	100	0	100	0	100	0	100	0

Table 5) Compare General characteristics in meat products samples (n=74).

Product Name		Appearance Check		packing		Odor		Apparent color		taste		foreign material		Marking	
		Suitable	Unsuitable	Suitable	Unsuitable	Suitable	Unsuitable	Suitable	Unsuitable	Suitable	Unsuitable	Negative	Positive	Complete	Incomplete
Sausage	Number	50	0	50	0	50	0	50	0	50	0	50	0	50	0
	Percent	100	0	100	0	100	0	100	0	100	0	100	0	100	0
kielbasa	Number	21	0	24	0	24	0	21	3	24	0	24	0	24	0
	Percent	85.71	14.28	100	0	100	0	85.71	14.28	100	0	100	0	100	0
Total	Number	72	3	74	0	74	0	71	3	74	0	74	0	74	0
	Percent	95.56	4.16	100	0	100	0	95.53	4.16	100	0	100	0	100	0

Discussion

The survey results of Health Status sausage and kielbasa produced in the province of Qazvin showed that these meat products have ideal conditions in accordance with the Iranian national standards, 100% of the meat products had an acceptable bacterial and fungal quality and microbial numeration, but most studies have been inconsistent with the results of our survey. In the research of Soltan Dallal et al. (2008) on 1047 food sample, their results showed that 100 samples (9.60%) were contaminated with S. aureus (29). In the study of Bradeeba et al. (2013) on surface microbial contamination of meat supplied in the Chidambaram, the results of this study indicated that gram-negative and gram- positive bacteria such as Pseudomonas aeruginosa 38 (50%), Proteus mirabilus 30(40%), Bacillus cereus 25(33%), Salmonella sp. 24(32%), Klebsiella pneumonia 21(28%) and E.coli 18(24%) predominantly constituted the total viable count, whereas frequently observed bacteria included Aeromonas sp. 16 (21%), S. aureus 8(10%) and Acinobacter 7 (9.30%) (30). In other study on 1634 samples of meat and dairy products to assess S. aureus showed the contamination in 12.80% of all samples (31). Another study showed 4 of 360 samples of kababs and cooked burgers contaminated with S. aureus in south area of Tehran (32). In the study of Kheyri et al. (2014), 95% of the meat products had an acceptable microbial contamination (33). In the study of Assaye et al. (2014), Salmonella were isolated in two of 90 samples of dried sausage (2.20%) (34). In the study of Antwi-Agyei et al. (2014) on meat and

fish products showed pathogens like S. aureus, mould, Pseudomonas spp. Salmonella and Bacillus spp. were also isolated from both the meat and fish products (35). Also, the study of Syne (2013) showed S. aureus the most common pathogen detected in pre-cooked products. This pathogen was also found in unacceptable levels in 4 (16.70%) of 24 post-cooked samples. Fifty percent (10 of 20) of precooked mixtures of bacon and bologna were contaminated with Listeria spp. including four with L. monocytogenes. Pre-cooked mixtures of franks and bologna also contained E.coli (35 and 0.72 log10 CFU/g, respectively) while 5 (12.50%) of 40 pre-cooked mixtures of chicken franks had Salmonella spp. (36) while the contamination of all meat products of our study was acceptable.

The chemical composition of the formulation include nitrite, fat and protein in all samples as standard and matched with studies Babaei et al. (2011), Golkari et al. (2012), NasehiNia et al. (2008) and Kamkar et al. (2004) but in other survey did not matching with our study such as Sadeghi et al. (2015) (37) who were showed nitrite and nitrate, 76% and 70% respectively, over of the allowed limit and also in study of Fekri et al. (2013) (38-41).

Abdulwahab Al-Amin et al. (2015) in the study of a variety of textures and flavor of meat examined in Sudan was consistent with our results (28).

The absence of microorganisms in the product of symbolizes health conditions in the workplace, equipment and raw materialsare measured. In the study of Sepidarkish et al. (2014), none of sausages had Staphylococcus aureus infections; that the results were matched, but 13.3% of the kielbasa that were evaluated which were infected, so that the results were not matched (42).

Qazvin province is one of the centers of livestock and poultry production in Iran. Therefore, it is necessary to prevent the spread of microbial contaminants such as Salmonella, E.coli, coliforms, Staphylococcus, Clostridium and mold and yeast in order to providing consumer health. As regards to obtained results of this study in comparison with previous studies, 100% of consumable meat products and without contamination shows proper sanitation and good hygienic quality of the production process of meat products in the farm, slaughtering, cold storage and sanitary production process is in Qazvin province of Iran.

Conclusion

According to the results of this study (microbial contamination, the exterior features and an additive composition of meat products samples), we should have more control on the hygienic quality of meat products over the production, storage and supply periods in this area.

Footnotes

Conflict of Interest:

The authors declared no conflict of interest.

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