ASSESSMENT OF MICROBIOLOGICAL LOAD OF RTE –MEAT PRODUCTS DURING SHELF LIFE, DECLARED IN THE LABEL

Ermelinda Nexhipi [1]

[1] The Department of Microbiology, the Institute of Food Safety and Veterinary,Tirana
E-mail: emanexhipi@yahoo.com

Abstract

The microbiological quality of RTE meat products until the shelf life declared on label, on refined establishment of Tirana's region, is the main criteria of the product's safety of consumer's health. So that, we studied 108 packed salami samples, of which 55 samples were cut in slices. The samples were stored in 5-7 ^{0}C and were analyzed for 15 days until the shelf life according to label's declaration. On the 1st production day, we took these results by the microbiological analyzes for *APC* : 25 samples resulted without an count, 47 samples with a count from 4.5x10^1 to 5.8x10^2 cfu/g, 36 samples with a count from 1.3x10^3 to 4.5x10 ^{4}cfu/g. From the last analysis made 2 days before the shelf life, 31 samples or 28.7% raised their microbiological count up to 6.0 x 10 ^{6}cfu/g, where the product with the highest count was the sliced salami, 30 samples were varied from 5.8x10^2 to 1.2 x10 ^{4}cfu/g and 47 samples resulted with a count from 1.0 x10^2 to 2.1x10 ^{3}cfu/g. The samples were analyzed for the presence of *E.Coli* and *Salmonella spp*. In no one sample it wasn't seen the salmonella presence. *E.Coli* was resulted in 10 samples or 9.2% with a primary count of 16cfu/g. At the end of the shelf life, the products resulted with a count of 70cfu/g, where the product with the highest load was the sliced salami.

***Keywords*:** *APC, E.coli, Salmonella spp during shelf life, RTE-meat products.*

Introductıon:

Meat products are one of the most consumed ready-to-eat food products around the world. The reasons that fuel the demand for such products are convenience and good acceptance by consumers. Microbiological safety of meat products ready to eat is of great importance for consumers as well as for the food industry. The fact that these meat products do not require further treatment (such as cooking) before consumption, lack of potentially pathogenic microorganisms in these products must be provided. Production of meat products generally includes traditional techniques, however, essentially the quality and safety of such products remains microbiology of these processes. During preparation and sale, these products can be contaminated by pathogenic bacteria causing illness in consumers if infective doses are reached at the time of consumption [2, 3]. Epidemiological and microbiological studies have identified cross-contamination (during preparation and sale) and subsequent bacterial growth (during storage) as the main causes of RTE contamination and illness. Slicing machines and cutting utensils are recognized as important vehicles of contamination of cooked meat products both at factory and sale points [4, 12]. Good-hygienic practices during handling arise, and also the implement of the storage conditions, as an important means to reduce cross-contamination.

Aim of study/research:

The aim of study is assessment the microbiological quality of RTE meat products until the shelf life declared on label, on refined establishment of Tirana's region.

Scientific methods:

We evaluated 108 samples, taken on the production establishments of Tirana. The samples were storage from 5-7 ^{o}C according to theirs perspective's labels. The analyzes were carried out by this time:
- The first analysis was made on the first production day.
- The second analysis was made on the 15th production day.
- The third analysis was made two days before the shelf life.

The samples were analyzed for Escherichia *coli, APC*, absence or presence of *Salmonella ssp*.

A. Microbiological analysis

Analysis of Escherichia Coli.

The "Sample preparation" was referred to ISO 6887-1 [7], where 10 gr of sample were weight and homogenized with 90 ml of BPW. We will transfer them in Petri sterile plates (1 ml of primary and serial dilutions). In every Petri sterile plate we put 15 ml of TBX media, warmed before in 44 ^{o} C – 47^{o}C. After that we mixed inoculate to the media as soon as it possible and we let it to be hardened in a cold horizontal surface. Then we turn overthrown the plates and we put them in a thermostat, at 44^{o}C for 18-24 hours. The total incubation time should not be more than 24 hours. The enumeration technique of colonies in plate in 44^{o}C by using 5-bromo-4-chloro-3-indolyl β-glucuronide and results expressed it was perform according to the standard method – ISO/TS 16649-2 [11], the horizontal method for enumeration of *Escherichia coli* β -glucuronidase-positive.

B. Aerobic Plate Count

The "Sample preparation" was referred to ISO 6887 [9], where 10 g of sample were weight and homogenized with 90 ml of BPW. We will transfer them in Petri sterile plates (1 ml of primary and serial dilutions). In every Petri sterile plate we put 15 ml of PCA media, warmed before in 44^{0} C – 47 ^{0}C. After that we mixed inoculate to the media as soon as it possible and we let it to be hardened in a cold horizontal surface. Then we turn overthrown the plates and we put them in a thermostat, at 30 ^{o} C for 72 hours. We used the regulated ISO 4833:2004 [8] the horizontal method for the enumeration of microorganisms -Colony-count technique at 30°C and expressed the result.

C. Detection of Salmonella spp.

Detection of Salmonella it was based in EN ISO 6579-2002- Am/2004 [6], an international accredited method, which has passed through 4 phases:
1st Phase: (Pre enrichment) in a liquid unselective media. After the preparation of the prove sample, 25 gr of the sample + 255 ml BPW, we homogenised and incubated it in at 37^{o}C for 18 hours.
2nd Phase: (Enrichment) in a liquid selective media. After we transferred 0.1 ml and incubated the culture, in a tube that has 10 ml RVS, we incubated it at 41.5^{o}C for 24 hours, plus 1 ml incubated culture in 10 ml MKTTn which was also incubated at 37^{o}C for 24 hours.
3rd Phase: The planting in a plate and isolation. It was taken with an (Anza) from RVS and was inoculated in two XLD plates, a solid selective media and two other plates from HEA solid

selective media and we incubated them at 37° C for 24 hours. Then we took with an (Anza) from the MKTTn and after we inoculate the culture to two XLD and HEA plates and incubated at 37 ° C for 24 hours.
4rth Phase: Isolation and confirmation. The plates were examined for the presence of Salmonella colonies. Suspected colonies on agar plates, were transferred to Kligler agar, and were incubated at 37°C for 24 to 48 h. Gram staining and biochemical tests such as the presence of catalase, oxidase, sulfide – indole - motility (SIM) medium (sulfur reduction test, indole production, motility) were perform. API 20 E, it was used for further biochemical identification and confirmation. Serological tests it is used to.

Data analysis:

The data of analysis was January – April 2014

Findings:

A. The evaluation of the microbiological count during the product's lifetime for E.Coli.

Sample Type	The microbiological count on the first production day			The microbiological count , 2 days before the shelf life		
	0 cfu/g	< 20cfu/g	>20cfu/g	0 cfu/g	0-20 cfu/g	20-100cfu/g
Salami	50	3		50		3
Sliced salami	48	7		48		7

Table1. Microbial growth during the shelflife of E. coli

From the analyzing of 108 samples on the first production day, we took these results: 98 samples didn't have an E.Coli presence, but 10 samples had a count to16 cfu/g. The analysis which was made 2 days before the date declared on the label showed us a raise of the microbiological count to 70cfu/g. The product with the highest count was the sliced salami. So, only 10% of the analyzed samples, had raised theirs count, during the storage process on 5-7 °C, until the shelflife day. This studies made from different authors, had shown that the microbiological loud of E.Coli for these products' category, is on a low percentage.
Wojtas & Christen [5] detected E. coli in 3/44 samples (6.8%) in a 1997 survey conducted in the ACT, while a follow-up survey the next year found E. coli in 4/65 samples (6.2%) taken from retail premises, with levels ranging from 4 to 210 cfu/g [14]. In an extensive survey of catering and retail premises in the UK, Elson, Burgess, Little, & Mitchell [13] found E. coli in 81/2894 samples (2.8%) of cooked sliced meats, with levels ranging from 20 cfu/g to greater than 107 cfu/g. Another large survey conducted in the UK on retail packaged cooked RTE meats found E. coli in 10/2981 samples (0.2%), at levels ranging from 20 to10^6 cfu/g [15]. The presence of *E. coli* in raw foods is considered an indication of direct or indirect fecal contamination. Indirect contamination can occur through sewage and polluted water. Direct fecal contamination occurs during the processing of raw foods of animal origin and because of poor personal hygiene of food handlers. In heat-processed and ready-to-eat foods its presence is a concern.

B. The microbiological count evaluation during the lifetime of the products for APC

One important parameter is the number of aerobic mesophiles. This group contains all aerobic organisms with optimal growth at 30-40 °C. These microbes are not all pathogens, but they are indicative of the overall quality of the meat.

First Day

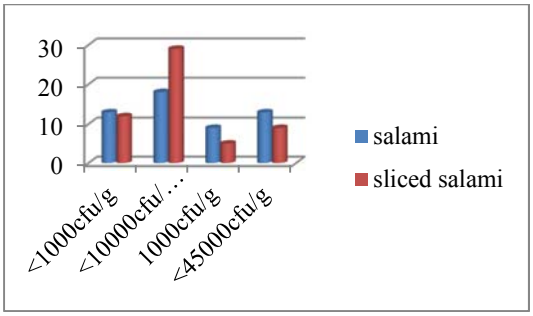

Figure1. The performance of the APC microbiological count during the shelf life

On the first analyzing day 25/108 samples resulted without an count,47 samples with a count from $4.5x10^1$ to $5.8x10^2$ cfu/g,36 samples with a count from $1.3x10^3$ to $4.5x10^4$cfu/g. So, 33.3 % of samples had an increase trend of microbiological count.

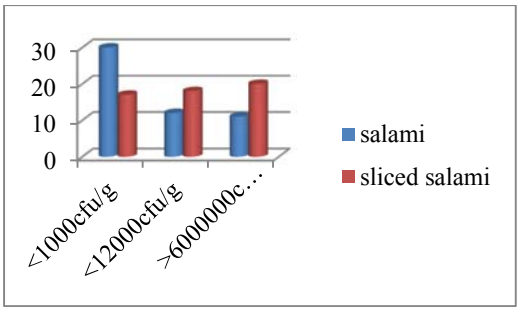

Figure2. Two days before the shelflife

From the last analysis made 2 days before the shelf life, 31 samples or 28.7% raised their microbiological count up to 6.0 x 10^6cfu/g, where the product with the highest count was the sliced salami, 30 samples were varied from $5.8x10^2$ to 1.2 x10^4cfu/g and 47 samples resulted with a count from 1.0 x10^2 to 2.1x10^3cfu/g. Their presence in large numbers in food indicates inadequate processing/or recontamination due to cross contamination by raw materials, dirty equipment or poor hygienic handling [1].

The "indicator" organisms are so called because their presence in large numbers in food signifies one of three contamination possibilities: disease bacteria or filth; spoilage or low quality; or preparation under insanitary conditions. a high APC may indicate that a food has been grossly mishandled or that it contains a poor quality ingredient.

C. The analyzing of Salmonella spp presence and absence

Sample Type	Salmonella spp	
	Presence	Absence
Salami	0	53
Sliced salami	0	55

On 108 analyzed samples, no one resulted with a presence of Salmonella spp.

Conclusions:

Hygienic quality of raw material has an important effect on final microbial load of salami. Heat application is also the main stage for the elimination of non-desired microorganisms during the production of salami. In order to prolong the shelf life and to improve the microbiological quality of salami, lower initial microbial levels of meat and other ingredients, effective heat treatment during heat application, careful handling of salami and maintenance of appropriate chill temperature during storage are necessary.

Recommendations:

The use of HACCP based control programs improves the quality and safety of the sausage during processing stages because hygienic status of the processing environment and equipment plays an essential role in the microbial stability and safety of the final products.

References:

[1] A. I. Ikeme. Fermented *sausage –dry and semi -dry*. In Meat Science and technology. 1st edition. The African fep publishers Limited Nigeria. (1990): 210

[2] B. Norrung & S. Buncic. Microbial *safety of meat in the European Union.* Meat Science, (2008). 78, 14−24

[3] EFSA (European Food Safety Authority) (2007). *The Community summary report of trends and sources of Zoonoses, Zoonotic agents, antimicrobial resistances and Foodborne outbreaks in the European Union in 2006*. EFSA Journal ,130, 3

[4] F. Pérez-Rodríguez, A. Valero, E. C. D. Todd, E. Carrasco, R. M. Carcía-Gimeno & G. Zurera. *Modeling transfer of Escherichia coli O 157: H 7 and Staphylococcus aureus during slicing of a cooked meat product.* Meat Science, (2007). 76 , 692−699

[5] F. Wojtas & S. Christen (1997). *Preservative levels and microbiological quality of manufactured meat*. Retrieved 21 April 2008, from http://health.act.gov.au/c/health?a=da&did=10017576&pid=1053857549

[6] International Standard EN ISO 6579: *Microbiology of food and animal feeding stuffs.* In: Horizontal method for the detection of *Salmonella* spp.: 2002, 2004.

[7] International Standard ISO 6887-1: *Microbiology of food and animal feeding stuffs-Preparation of test samples, initial suspension and decimal dilutions for microbiological examination.* Part 1: In: General rules for the preparation of test samples, initial suspension and decimal dilutions 1999

[8] International Standard ISO 4833: *Microbiology of food and animal feeding stuffs. Horizontal method for counting microorganisms.* In: Counting technique colonies in the plate at 30º: 2003.

[9] International Standard ISO 6887: *Microbiology of food and animal feeding stuffs.* In: Preparation of test samples, initial suspension and decimal dilutions for microbiological examination: 2003.

[10] International Standard ISO 7218: *Microbiology of food and animal feeding stuffs.* In: General requirements for the performance of laboratory tests: 1996.

[11] International Standard ISO/TS 16649-2: *Microbiology of food and animal feeding stuffs. Horizontal method for counting Escherichia coli β -glucuronidase-*

positive. In: Counting technique colonies in the plate at 44˚C using 5-bromo-4-chloro-3-indolyl β-glucuronide 2003.

[12] K. L. Vorst, E. C. D. Todd & E.T. Ryser. *Transfer of Listeria monocytogenes during slicing of turkey, bologna, and salami with simulated kitchen knives.* Journal of Food Protection, (2006). 69, 619−626

[13] R. Elson, F. Burgess, C. L. Little, & R. T. Mitchell *Microbiological examination of Ready-to-eat cold sliced meats and pâté from catering and retail premises in the UK.* Journal of Applied Microbiology (2004). 96, 499-509.

[14] S. Christen & G. Millard (1998). *Microbiological and chemical quality of cured and salted meats.* Retrieved 21 April 2008, from hhtp://health.act.gov.au/c/health?a=da&did=10017576&pid=1053858104

[15] S. K. Sagoo, C. L., Little, G. Allen, K. Williamson, K. A. Grant (2007). *Microbiological safety of retail vacuum-packed and modified-atmosphere-packed cooked meats and end of shelf life***.** Journal of Food protection 70 (4), 943-951