MICROBIOLOGICAL PROFILE AND SHELF LIFE OF FRESH PASTA SUBJECTED TO SEVERAL CYCLES OF PASTEURIZATION

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Abstract – Campania, and mostly Avellino and Benevento, is the main producer of durum wheat and pasta.

The fresh pasta has a high rate of humidity, which makes it an appropriate substrate for the growth of microorganisms and it requires protection from microbial contamination and thorough microbiological controls for its commercialization.

The shelf-life of a food is the time period in which a product, in certain storage conditions, maintains a level of quality and safety "acceptable". The acceptability of the product is defined by the presence or absence of a certain number of microrganisms identified by microbiological analysis aimed basically to ensure that food meets specific microbiological criteria, established by a rigorous national and community legislation (Reg. EC 852/2004, Reg. EC 2073/2005, Reg. EC 1441/2007). Particularly, it is important to define suitable conditions of pasteurization and packaging in order to preserve the fresh pasta for a long period of commercialization and ensuring the safety of consumers.

In this study we evaluated the effects of different cycles of pasteurization on the microbiological quality and safety of the fresh pasta produced by a company of Benevento (Italy) and the effects on shelf-life of the product.

Keywords: fresh pasta, pasteurization, packaging, shelf-life, microbiological quality.

1. INTRODUCTION

Italy is the first producer and consumer of pasta in the world, this is due, in large part, to the vast areas of cultivation of durum. In Campania the provinces with the largest area devoted to the cultivation of durum wheat are Avellino and Benevento (AIDEPI 2013).

The quality and added value of the pasta , comes from the raw material. The processing of durum wheat (*Triticum durum*) into flour foresees some main steps: the cleaning of the wheat, humidification with the addition of an appropriate amount of drinking water and the grinding .

The durum wheat flour is mixed in special mixers with pure water. The process is not so simple: the determination of water and its temperature are important variables and enter in determining the final quality of the pasta.

The main form of marketing of pasta is represented by dry pasta, mainly produced at industrial level. The dry pasta is obtained by drying the finished product, this involves the removal by evaporation of a part of water, until reaching a rate of humidity less than 12.5%, a value that makes the product impervious by microbiological agents for many months.

The fresh pasta, on the contrary, suffers a milder heat treatment, the pasteurization, therefore has a high rate of humidity, at least 24% which makes it an appropriate growth substrate for bacteria and molds. The pasteurization together with the quality of the raw material is the stage that

most affects the quality of fresh pasta, in fact, many of the structural properties and cooking behavior of pre-packaged fresh pasta are definitely influenced by the intensity of the pasteurisation process.

Unlike from sterilization, the pasteurization is able to eliminate vegetative microbial cells alone (but not the spores). During the pasteurization, by operating at temperatures comprised between 87°C and 110°C for the time of a few minutes pathogenic microorganisms are potentially eliminated, such as *Salmonella*, *Listeria*, Coliforms, Mycobacteria and *Brucella*. Therefore the pasteurization in addition to extend the life of the product (shelf -life) serves to ensure the hygienic-sanitary safety of the finished product.

The fresh pasta is often packaged in a protective atmosphere MAP (Modified Atmosphere Packaging). The modified atmosphere for fresh pasta comprises generally 30% carbon dioxide and 70% nitrogen, which effectively contrasts the natural deterioration of the product, increases the shelf life, controls the humidity and prevents crushing of the product same.

Data from the FAO and the WHO and the international bibliography, show that in industrialized countries, the main food safety problems are of microbiological origin. Authoritative estimates show that in developed countries, about 30% of the population is exposed to certain diseases from microbiological contamination of food with consequent heavy health care costs.

Whereas it is not possible to keep under control in a food matrix all bacteria, fungi, viruses, protozoa, potential causes of diseases from foods; it is necessary to aim the control only of the microorganisms that compromise in an important way the state hygienic-sanitary of the product. According to the regulatory standards, it is important to the evaluation of a series of indicator organisms; divided into indicators of quality of product and indicators of product safety. The first are microorganisms and/or their metabolites (for example toxins), whose presence in food may be useful for evaluating the quality and eventually the hygienic shortage of the product, directly related to the hygienic problem of the process. The seconds, however, are potential pathogens that indicate the occurrence of a risk to the health of the consumers.

Inserted in a larger project of biodiversity research and production of fresh pasta in Alto Tammaro (Benevento, Italy), the purpose of the study was to:

1) monitor and improve the microbiological safety of the fresh pasta of locally made, starting from the microbiological evaluation of the raw material (the durum wheat flour) used in the production;

2) evaluate the effects of different treatments of pasteurization on the microbiological quality of the pasta and the shelf life of the product.

2. PHASES AND ANALYTICAL METHODS

Microbiological studies were conducted by conventional methods of microbiology for the isolation, the counts and the identification of microorganisms.

With these surveys it was possible to evaluate the microbiological quality of the raw material used and the level of hygienic-sanitary attention of the entire production process.

The analysis were conducted on different groups of sample units, subjected to different cycles of pasteurization (100°C x 80''; 102°C x 97''; 102°C for 100'') at regular intervals of 1, 7, 21, and 28 days from the packaging in MAP.

The fresh pasta samples (25g) were homogenized in 250 mL 0.1% peptone solution by a paddle peristaltic homogenizer Stomacher 400 Circulator (Seward LTD, UK) and an aliquot of appropriate serial dilutions was transferred to suitable agar media, in duplicate. Plates were then incubated in suitable conditions for 1-5 days and viable count was performed. Results were expressed as colony forming units per gram of sample (CFU/g). Appropriate biochemical tests were performed in the identification phase of the microrganisms.

Due to the high values of water activity (0.92 <Aw <0.97), or of that provided for the microbial cells, the fresh pasta is considered a highly perishable food. Water activity (Aw) is just one of the factors that influence the perishability of fresh pasta and other food products. The development of such contamination depends on a large number of factors such as the richness in nutrients (proteins, carbohydrates, fats, vitamins and mineral salts) which makes them an excellent growth medium for microorganisms, the salt content, the pH , the presence or absence of oxygen, the storage

temperatures, the time between preparation and consumption of the food. There is not always a close correlation between the number of microorganisms present in a food and the degree of alteration of the same. Typically only a portion of the total population participates to the alterations. This microflora is referred to as: Specific Spoilage Organisms (*Specific Spoilage Organisms- SSO).*

For this reason the Health Ministry has set also for fresh pasta, standard microbiological indicators of appropriate processes, and of safety (Table 1). So the fresh pasta to be able to be marketed must contain: $<10^5$ CFU/g Total Mesophilic Aerobic Bacteria, $<10^2$ Enterobacteriaceae, $<10^2$ Staphylococcus, $<10^2$ Clostridia, $<10^2$ Listeria, $<10^2$ Molds, $<10^3$ Yeast, no Salmonella in 25 g of product.

Instead the Table 2 shows the microbiological parameters and quantitative analytical methods used in this study.

Parameter	Limit	Method
Total Mesophilic Aerobic Bacteria	<105 CFU/g	ISO 4833
Enterocacteriaceae	$<10^2$ CFU/g	ISO 21528-2
Molds	$<10^2$ CFU/g	ISO 21527-1
Yeasts	$<10^3$ CFU/g	ISO 21527-2
Escherichia coli	$<10^2$ CFU/g	ISO 16649-2
Staphylococcus aureus	$<10^2$ CFU/g	ISO 6888-1
Salmonella spp	Absent in 25g	ISO 6579
Clostridium perfrigens	$<10^2$ CFU/g	ISO7937
Listeria monocytogenes	$<10^2$ CFU/g	ISO 11290

Table 1. Microbiological limits for fresh pasta and durum wheat flour.

Parameter	Method	Media and incubation conditions
Total Mesophilic Aerobic Bacteria	ISO 4833	Brain Heart Infusion Agar 37°C x 24h - 48h
Enterocacteriaceae	ISO 21528-2	MacConkey Agar 37°C x 24h - 48h
Molds	ISO 21527-1	Sabouraud Agar 25°C x 1-5 days
Yeasts	ISO 21527-1	Sabouraud Agar 25°C x 1-5 days
Escherichia coli	ISO16649-2	Tryptone Bile X-glucuronide 37°C x 24h - 48h
Staphylococcus aureus	ISO6888-1	Baird Parker Agar 37°C x 24h - 48h
Salmonella spp	ISO6579	Xilosio, Lisina ,Deossicolato 37°C x 48h
Clostridium perfrigens	ISO7937	Tryptose Sulfite Cycloserina 37°C x 24h - 48h
Listeria monocytogenes	EN7ISO11290-2	Agar Listeria Ottaviani & Agosti 37°C x 24h - 48h

Table 2. Microbiological parameters and quantitative analytical methods used in this study.

3. RESULTS

The microbiological tests conducted on the sample units of durum wheat flour used for the preparation of fresh pasta have shown that all indicators of microbiological quality (Total Mesophilic Aerobic Bacteria, *Enterobacteriaceae*, Yeasts and Molds), show that the average values are inside the limits indicated by the regulations, therefore this flour is suitable in terms of microbiological quality as raw materials for the preparation of pasta.

The microbiological analysis carried out on the samples of fresh pasta subjected to the first method of pasteurization (100°C x 80'') packaged in MAP have shown that the indicators of microbiological safety (*E. coli*, Clostridia sulphite reducing, *Salmonella* spp and *L. monocytogenes*), that indicate the possible presence of microbiological risk because potentially pathogenic, are resulted always absent, during the 21-day monitoring (Figure 1).

Figure 1. Microbiological quality and shelf life of the fresh pasta subjected to the first method of pasteurization (100°C x 80'') packaged in MAP.

While the monitoring of the microbiological indicators of hygiene, that indicate contamination and deteriorating of the food (Total Mesophilic Aerobic Bacteria, *Enterobacteriaceae*, *Staphylococcus*, Yeasts and Molds) have showed early (7th day) values very close to the maximum limits indicated by the regulations.

By performing on the fresh pasta the second method of pasteurization (102°C x 97''), we found the absence of microbiological safety indicators and an improvement of the average values of the microbiological parameters related to the monitoring of hygiene microbiological indicators. In particular, it should be emphasized the absence of Yeasts and Molds to 21 days from the packaging (Figure 2).

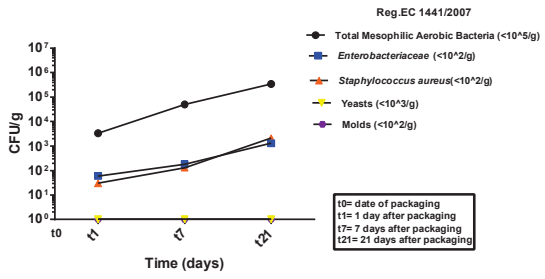

Figure 2. Microbiological quality and shelf life of the fresh pasta subjected to the second method of pasteurization (102°C x 97'') packaged in MAP.

In samples of fresh pasta subjected to the third method of pasteurization (102°C x 100'') and packaged in MAP, the microbiological controls have detected again the absence of microbiological safety indicators (*E. coli*, Clostridia sulphite reducing, *Salmonella* spp and *L. monocytogenes*) and a improvement more sensitive of values of the microbiological parameters related to monitoring of hygiene (Total Mesophilic Aerobic Bacteria, *Enterobacteriaceae*, *Staphylococcus*, Yeast and Molds), with results significantly inside of the regulatory limits.

It must be noted, in particular, such with the optimizing the heat treatment the growth of Yeast and Molds is inhibited until 21 days from the packaging, but also that of the *Enterobacteriaceae* at least until 7th day (Figure 3).

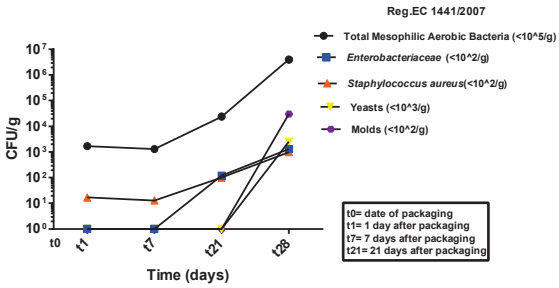

Figure 3. Microbiological quality and shelf life of the fresh pasta subjected to the third method of pasteurization (102°C x 100'') packaged in MAP.

To establish accurately the shelf life of fresh pasta we performed microbiological analysis to 28 days from the packaging. The microbiological controls have highlighted values of the microbiological counts on plate over the limits indicated by the regulations for all microbial groups considered.

4. CONCLUSIONS

With "durability" or "shelf-life" of a food is defined the time interval within which the food in storage conditions defined, it retains the characteristics of safety for the using and the typical characteristics organoleptic and sensory.

Determine exactly, the commercial life of a product, is of fundamental importance for the food company that must, as required, in European Community Regulations, offer the consumer a guarantee of quality and safety of its products for the entire duration of the shelf-life (Article 3 Reg. EC 2073/2005), that the company itself states.

Thanks to this study:

1) the quality and microbiological safety of fresh

pasta typical of Alto Tammaro has been improved;

2) the "shelf-life" of fresh pasta typical of Alto Tammaro has been established to 21 days , using the method of pasteurization (102°C for 100''), at refrigeration temperature of 4°C with MAP packaging.

5. REFERENCES

AIDEPI (Associazione delle Industrie del Dolce e della Pasta Italiane) 2013.

Regulation EC 852/2004.
Regulation EC 2073/2005.
Regulation EC 1441/2007.

ACKNOWLEDGMENTS

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