



## Effect of probiotic and prebiotic in the formulation and elaboration of sausage as an alternative of consumption

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### ABSTRACT

In Ecuador, sausages are one of the most commonly used in typical foods, but traditionally these have only been beef, pork and chicken, which in turn do not contain any health benefits, on the contrary, their limited consumption is recommended. This research aims to provide the market with a new nutritional alternative that increases the variety of existing products with the difference that these sausages have an added value that are probiotics and prebiotics. To obtain a sausage formulation, variations were made in the percentages of the ingredients that were used, obtaining 3 treatments. The most popular treatment was determined by means of a sensory panel made up of 50 people, resulting in treatment 3 being the most accepted. A corresponding analysis was carried out whose result gave the presence of lactobacillus with a value of  $8.8 \times 10^8$ , proteins: 17.64% total fats: 26.48% cholesterol: 29.34 mg Carbohydrates: 0.82 mg / 100g dietary fiber: 3.17, the caloric intake of the sausage was 312 kcal, sodium: 495.23 mg / 100g, starch: 0.31% and according to the study of shelf life in refrigeration at 5 ° c the maximum time of consumption of the product is 22 days.

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### INTRODUCTION

In recent years diseases have increased as a significant cause of poor diet, as many of the consumer products have high fat, sugar, salt, etc. And this triggers diseases such as diabetes, hypertension and cancer. One of the foods that are less recommended for daily intake are sausages, since they have high values of sodium, carbohydrates and fats, and the constant use of these foods will contribute to health

problems. For this reason, studies have been carried out to make sausages that are recommended for daily consumption and provide health benefits, the first sausages to be processed are fermented-cured, and this group belongs to the salami (F.A.O., 2014). From a nutritional point of view, the suitability of fermented meat products as a vehicle for probiotic microorganisms can be integrated into a varied and balanced diet. In this sense, fermented "probiotic" meat products could be formulated with less salt and fat, which would qualitatively improve the nutritional characteristics of this type of product with functional claims (Rubio, 2014). There has also been a need to develop innovative products or modify those already on the market, with the aim of not only satisfying the needs of consumers but also providing benefits to consumer health. That is why we now have studied about agents that contribute to proper nutrition and digestion, the latter being a significant problem in our population since it triggers many serious diseases, such as cancer. Prebiotics are non-digestible ingredients in the diet that stimulate the growth or activity of one or more types

of bacteria in the colon. Probiotics are microorganisms that, when added as a supplement, favor the development of the microbial flora in the intestine. Polydextrose is insoluble prebiotic fibre, helping in its passage through the intestine to proper digestion and providing beneficial flora to the colonic flora. *Lactobacillus casei* is commonly used for the coagulation of milk when preparing fresh cheeses, this time it was used as a probiotic agent since it helps to restore and protect the intestinal flora and reduce pathogens. As a general objective, it was proposed to evaluate the effect of probiotics and prebiotics in the elaboration of chorizo as an alternative to meat and its acceptability. From there, it was introduced as specific objectives to establish 3 treatments for the elaboration of sausages, to execute a sensory panel to obtain chorizo of higher acceptability, to identify the presence of probiotic and prebiotic agents in the most accepted treatment, through an analysis of composition and nutritional value, to determine the useful life (percha) of the most acceptable treatment through microbiological, physical, chemical and sensory analysis. According to the INEN norm, carry out a market study to determine the acceptability of the most popular treatment in the south-west of the Guayas canton.

## MATERIALS AND METHODS

The research was of an experimental type, based on the development of a different product in which the design applied in this research was Design of blocks completely randomized with 3 treatments and 3 repetitions. The statistical evaluation of the data was performed through the analysis of variance, the comparison of averages made by the Tuckey test at 5% probability. A sensory tool was applied in order to obtain the most accepted sample, which was attended by 50 untrained panelists; these were chosen at random, and it was done three times for each treatment, an example of each of the 3 treatments was given, the results that the panelists issued were statistically evaluated according to what was established. When the evaluation of the 3 treatments was carried out, the organoleptic characteristics and attributes such as smell, colour, taste, texture were determined, and the acceptability of the best formulation was known. The elaboration of the product was carried out in the city of Guayaquil, in the citadel in the Universidad Agraria del Ecuador with GPS coordinates -2.131294, -79.883525.

The reception and selection were used low-moisture pork and beef, pork fat (was washed out under running water and immersed in a germicidal (chlorine) solution), probiotic bacteria and prebiotics. The

meat was minced with a 5 mm disc, the pork with a 12 mm one and the fat in cubes of 25 mm, the meat and the fat are passed through a mill, the meats and fat are mixed, they are added the salts, the condiments and the ice until obtaining a homogenous mass and finally the probiotic and prebiotic bacteria. The dough is allowed to stand in refrigeration for 24 hours. This stage is also known as aging, and in it, the maturation reactions of the dough are developed. The mass is stuffed into a narrow 30 mm pork casing; a (10 mm) nozzle was used for filling, the stuffed casings are tied. They are then hung on hooks and washed with drinking water to remove the mass residues adhered to the surface of the casing. The chorizos were transferred to a pre-drying chamber for 6 to 8 hours at room temperature. During this stage, the ripening reactions of the dough are presented. The chorizos were stored in refrigeration at 4 ° C, until the moment of sale (Senasa, 2000). In order to obtain an ideal formulation that meets the requirements of the INEN 1338 standard and also to be liked for consumption, it was started by establishing the list of ingredients and additives that are part of the product to be made. Variations were made in the percentage of condiments, oregano and salt, looking for this to affect the organoleptic characteristics of the chorizo in order to have a product with 3 alternatives in taste. In turn, 3 different formulations of a mixture of probiotic and prebiotic agents were applied to each treatment, respectively (Table 1).

For the determination of the treatment with greater sensory acceptance, a group of 50 untrained people was formed, to whom the parameters that would be evaluated in relation to sausage with prebiotics and probiotics should have been indicated. Each panelist was assigned a rating table of 4 attributes, colour, taste, texture and smell. Each attribute was rated on a scale of 5 to 1, corresponds from very good to very bad respectively.

## RESULTS AND DISCUSSION

The registered data were grouped and analyzed in a statistical way through the application of a statistical analysis of variance. In order to determine the product with the most significant sensory acceptance, the tukey test was applied, which allowed comparing the averages obtained and through the resulting (average) values, the product of better sensory characteristics was determined.

By means of the rating of averages, we can point to treatment 3, with the highest acceptance in the texture with a value obtained of 4.72, followed by treatment 1 with 3.92 and leaving treatment 2 as the one

**Table 1: Chorizo formulation**

| Ingredients   | Treatment 1 | Treatment 2 | Treatment 3 |
|---------------|-------------|-------------|-------------|
| Pork Meat     | 50%         | 50%         | 50%         |
| Beef          | 20%         | 20%         | 20%         |
| Pork fat      | 18%         | 18%         | 18%         |
| Salt          | 5%          | 1%          | 1%          |
| Ice           | 6,32%       | 6,32%       | 6,32%       |
| Condiments    | 3,16%       | 1,16%       | 1,16%       |
| Oregano       | 2,16%       | 3,16%       | 3,16%       |
| Phosphate     | 0,05%       | 0,05%       | 0,05%       |
| Ascorbic acid | 0,06%       | 0,06%       | 0,06%       |
| Prebiotics    | 0,10%       | 0,11%       | 0,15%       |
| Probiotics    | 0,10%       | 0,09%       | 0,05%       |
| Nitrite       | 0.05%       | 0.05%       | 0.05%       |
| TOTAL         | 100%        | 100%        | 100%        |

**Table 2: Sensory rating summary**

|             | Texture | Color | Odor | Tast |
|-------------|---------|-------|------|------|
| Treatment 1 | 3,92    | 3,78  | 3,74 | 3,92 |
| Treatment 2 | 3,6     | 3,58  | 3,56 | 3,6  |
| Treatment 3 | 4,72    | 4,4   | 4,64 | 4,72 |

with the least acceptance with a range of 3.60. It was observed that the treatment 3 with the highest average sensory acceptance in the colour parameter once the analysis of variance was performed with a mean of 4.40, followed then by treatment 1 with 3.78 and leaving the final treatment 2 with a value of 3.58. Also treatment 3, with the highest acceptance in the parameter of odor with a value obtained of 4.64, followed by surgery 1 with 3.74 and leaving treatment 2 as the least accepted with a range of 3.56, and the highest average sensory acceptance in the flavor parameter once the analysis of variance was performed with a mean of 4.72, followed by treatment 1 with 3.92 and leaving treatment 2 with a value of 3.60 at the end (Table 2).

It can be clearly seen that treatment 3 obtained the highest acceptance on the part of the sensory panel made up of 50 people. Determined so that treatment 3 must be the product to which the respective chemical, microbiological and nutritional physical analyzes are carried out.

To determine the content of probiotic bacteria that a food must contain in order to be recognized as a functional food, we proceeded to investigate the national and international regulations resulting in that only ranks have been specified for dairy products. In the (INEN 2395, 2012) INEN 2395 standard, an amount of  $10^6$  CFU / g is indicated, and in

the Central American technical standard, the quantity of  $1 \times 10^6$  CFU / g is indicated. Although there is no specification for sausages, it should be noted that the presence of lactobacilli was positive, with a value of  $8.8 \times 10^8$ .

The analysis was performed to determine the amounts of Proteins, Total Fat, Cholesterol, Carbohydrates, Dietary fibre (Polydextrose), Energy, Sodium and Starchas detailed:

#### Proteins

According to the INEN 1338 standard for meat products, the minimum value of proteins in meat products is 14%. The sausage obtained a value of 17.64, satisfying this requirement satisfactorily.

#### Total fat

According to the norm INEN 056: 2013, the entire fat content ranges between 20 to 40%. The resulting value was 26.48% complying with the specification.

#### Cholesterol

According to studies at Harvard University, a maximum of 200 mg is recommended. 100 grams of the sausage contains 29.34 mg.

#### Carbohydrates

Meats do not provide carbohydrates. However, the sausages have values due to the ingredients that make it up. A value of 0.82 mg / 100G was obtained.

### Dietary Fiber (Polydextrose)

The fibre content is 3.17; it should be noted that there are no specifications for fibre in the regulations.

### Energy

The caloric intake of the sausage was 312 kcal per 100 grams of the product.

### Sodium

According to the INEN 1336 standard for meat products, sodium salts have a maximum dose of up to 1500 mg/kg. The resulting value of the analysis was 495.23 mg / 100g

### Starch

The content of starch varies according to the type of meat product, from absence to 6% maximum. The value obtained was 0.31%

As one of the objectives of the project, it was decided to determine shelf life through microbiological analysis of chorizo with probiotics and prebiotic. The microbiological analyses were determined based on the indications of the norm 1338: 2012 for meat products; This standard indicates that Mesophilic Aerobes, Escherichia coli, Staphylococcus aureus, Salmonella must be analyzed.

The sample was kept under refrigeration at a temperature of 5 ° C. The microbiological analyzes indicated by the INEN 1338 standard for meat products were performed, obtaining results that were evaluated under the comparison of the maximum limits of acceptance of each microorganism as indicated in the regulations for meat products. The results of the microbiological analyses of the third analysis, carried out 22 days after the reception of the sample, indicate that the maximum microbiological limit is still being met, which suggests that the example can continue with the analysis.

At the end of the elaboration of the sausage with the content of probiotics, bromatological analyses were carried out in which a carbohydrate content of 0.82 mg was determined in 100 grams of the sample; this value is relatively low because the meat does not contain carbohydrates. The INEN 1336 standard does not indicate a range for this parameter. The contribution of Dietary Fiber was 3.17%, with an energy contribution of 312 kcal. The Sodium content was 495.23 mg / 100g, exceeding the limit established in the INEN 1336 standard. Although these values are not related to the specifications in the INEN 1336 standard, they can be compared with the results of other work related to sausages (Ramirez *et al.*, 2001). Regarding carbohydrates, they are not usually foods with a very

remarkable content in the sausages, only in the case that they carry flours or starches added will increase this value. Considering its mineral content, the one that stands out above all others is sodium, a component of the salt added in all of them as a flavouring and curing and preservation method.

In a formulation of 2000 grams of sausages, a mixture of 1 g of probiotics and 3 g of prebiotic was used. To determine its contribution to the nutritional content of the product, an analysis of probiotics was performed, which gave a result of  $8.8 \times 10^8$ . The content of probiotics could not be related to values of references for sausages because the INEN 1336 Standard for meat products does not make indications for the use of this microorganism in meat products. But nevertheless, the use of probiotics is justified according to what is mentioned in the NTE (INEN 2395, 2012) : (INEN.056, 2011) Fermented kinds of milk; Probiotic microorganism. "Live microorganism, which supplied in the diet and ingested in sufficient quantity exerts a beneficial effect on health, beyond the nutritional effects."According to the studies carried out on the product (chorizo with probiotics and prebiotics), it is granted a shelf life in refrigeration at 5 ° C for 22 days. This analysis was based on the maximum time of consumption of the product. This analysis guarantees the viability of making sausages with probiotic contents because the use of these microorganisms is not usually used in meat products, they have always been always intended for incorporation into dairy foods as mentioned (Cabezas, 2003).

In general, probiotics are found in dairy products because it is easier to make these good bacteria grow in these types of products. Probiotic bacteria are characterized, among many other issues, because they take their food from milk sugar, lactose, initially producing lactic acid, responsible for the acid taste of fermented milk. In the same way, they are able to multiply and maintain themselves inside our intestines, acting as protectors of our intestinal mucosa and facilitating the proper functioning of this section of the digestive tract, with the consequent improvement in the absorption of nutrients from food.

### CONCLUSION

The treatment 3 obtained the highest acceptance from the sensory panel made up of 50 people with the following grades texture 4.72, colour 4.4, smell 4.64 and taste 4.72. Determined so that treatment 3 must be the product to which the respective chemical, microbiological and nutritional physical analyzes are carried out.

In a formulation of 2000 grams of sausages, a mixture of 1 g of probiotics and 3 g of prebiotic was used. An analysis of probiotics was carried out, which gave a result of  $8.8 \times 10^8$ . There are no specifications for the contents of probiotics in sausages. We proceeded to investigate the national and international regulations resulting in that only ranges for dairy products have been specified whose range is  $10^6$  CFU/g.

As a result of the nutritional analysis, the following values were obtained: Proteins: The sausage gained an amount of 17.64% Total fats: According to the INEN 056: 2013 standard, the total fat content ranges from 20 to 40%. The resulting value was 26.48% complying with the specification. Cholesterol: The sausage contains 29.34 mg Carbohydrates: A value of 0.82 mg / 100G Dietary Fiber was obtained: The fibre content is 3.17. Energy: The caloric intake of the sausage was 312 kcal per 100 grams of the product. Sodium: The value resulting from the analysis was 495.23 mg / 100g Starch: The value obtained was 0.31%.

The analysis of shelf life through microbiological analysis of chorizo with probiotics and prebiotic. *Mesophilic Aerobios, Escherichia coli, Staphylococcus aureus, Salmonella* were analyzed, it is concluded a shelf life in refrigeration at 5 ° C for 22 days.

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