# AN ATTEMPT TO USE PROBIOTIC IN THE PRODUCTION OF DRY FERMENTED PORK SAUSAGE

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*Abstract*—**The purpose of the present studies was to assess the survival probiotic bacteria Lactobacillus casei ŁOCK 0900 in dry fermented sausage and to investigate the effect of their addition on physical-chemical properties and sensory quality of dry fermented sausage. The count of probiotic bacteria in dry fermented sausage after ripening and after 11 weeks of storage was ≥ $10^6$ cfu/g, hence the product satisfied one of the requirements set for probiotic food. There is a possibility of using a strain of probiotic bacteria in dry fermented sausage but the product needs to be improved considering its sensory quality. An addition of probiotic bacteria to the batter caused intensification of oxidation processes in the product, which showed in an increased oxidation-reduction potential, a higher TBARS index and an increased proportion of the oxidated form of myoglobin with brown colour during the storage.**

*Index Terms*—**dry fermented sausage, probiotic.**

## I. INTRODUCTION

In recent years, a considerable interest of the consumers in functional food, including probiotic food, has been observed (Holzapfel & Schillinger, 2002). Fermented meat products constitute an exclusive group of fermented dry meats, with a long used-by date (up to 6 months), consisting of the top quality meat-fat raw materials (Kołożyn-Krajewska & Dolatowski, 2009). Health benefits resulting from the consumption of probiotics encourage broadening the assortment of products containing these beneficial microorganisms. The purpose of the present studies was to assess the survival of probiotic bacteria *Lactobacillus casei* ŁOCK 0900 in dry fermented sausage and to investigate the effect of their addition on the physical-chemical properties and sensory quality of dry fermented sausage.

## II. MATERIALS AND METHODS

The raw material for the production of raw fermented sausage was pork meat of class I from the ham, and back pork fat obtained in industrial conditions from ecologically bred pigs of Wielka Biała Polska breed with the life weight of 120-130 kg. The raw material was obtained from the carcasses chilled for 48 hours aftet the slaughter. The meat was cured using the curing mixture in the quantity of 2.8% (2.8 g/kg salt, 150 mg/kg sodium nitrate, 100 mg/kg sodium nitrite) for 72 hours at the temperature 2-4°C, it was mixed with ground fat with an addition of glucose, probiotic Lactobacillus casei ŁOCK 0900, spices (garlic, black pepper).

Fibrous casings were filled with the batter. The products were subjected to depositing, and next 3-weeks' ripening at the temperature of 18-19°C and humidity of 80-90%. The following research variants were applied: control product(C); product with an addition of Lactobacillus casei ŁOCK 0900 ($2x10^8$ cfu/g), glucose 0.6% (LC). After the ripening process, sausages were cold smoked. Next, the products were vacuum packed and stored in cold conditions for 11 weeks. The obtained products were examined immediately after the process of ripening and after storage.

The following were marked in physical-chemical tests:

- Acidity – measuring the value of pH using a digital pH/conductometer CPC-501 (Elmetron) and a combined electrode ERH-111 (Elmetron).
- Redox potential – according to the method by Ahn & Nam (2004) using a combined electrode ERPt-13 with a digital measurer CPC-501 (Elmetron).
- The content of myoglobin (Mb), metmyoglobin (MetMb), oxymyoglobin ($MbO_2$) was established by mans of a spectrophometer Nicole Evolution 300 BB by Thermo Elektron Corporation according to the methodology provided by Krzywicki (1982).

- TBARS index was determined measuring the value of number TBARS [mg MDA/kg] according to the method by Pikul, Leszczyński & Kummerow (1989). The intensity of the colour appearing in the reaction of dimalone aldehyde with 2-thiobarbituric aid was measured spectrophotmetrically at the wave length of 532 nm with a spectrophotometer Nicole Evolution 300 (Thermo Elektron Corporation).

Microbiological analyses were conducted using an automatic system to measure the count of microorganisms - TEMPO® (Biomerieux, France). Original tests TEMPO® LAB, used to determine the count of bacteria of lactic acid in food products, were used for microbiological markings. The results are given in colony forming units in one gram of product (cfu/g). The applied system of microbiological markings made it possible to achieve the levels of reliability similar to standards NF ISO 15214 (1998) and recommendations of the Compendium for Microbiological Research of Food of the American Health Association (2004). Sensory analyses were conducted by means of the Qualitative Descriptive Analysis - QDA (ISO 13299.2, 1998).

## III. RESULTS AND DISCUSSION

After ripening, the count of lactic acid bacteria (LAB) was stabilized at the average level of 8.83 log cfu/g in the control and 9.04 cfu/g in the sample with probiotic.

During the storage, the count of the studied microorganisms stayed at a stable level and was about 8.93 log cfu/g in the control. On the other hand, the average count of LAB in the product containing probiotic was, on average, about 9.21 log cfu/g. In the study of Aro, Nyam-Osor, Tsuji, Shimada, Fukushima & Sekikawa (2010), the survival of LAB in fermented sausage was at a similar level. Results concerning the survival by probiotic bacteria of lactic acid at the level higher than $10^6$ cfu/g, which ensures the probiotic character of the product. Fig. 1. presents pH value of the examined sausage samples directly after ripening and in the 11th week of storage.

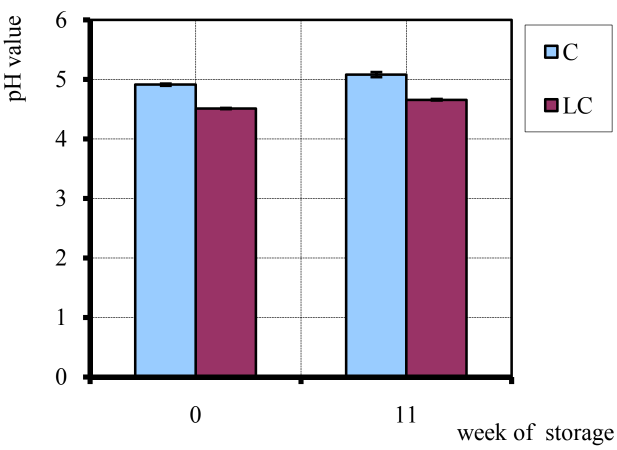


**Figure 1**. pH value in dry fermented sausage in 0 and 11 weeks of cold storage (C – control; LC – sample containing Lactobacillus casei ŁOCK 0900; n = 3)

In all samples the value of pH increased in the final stage of storage. Lower pH values in fermented sausage inoculated with L. casei ŁOCK 0900 are a result of transformations of glucose caused by microorganisms of lactic fermentation. As a consequence, this leads to the formation of mainly lactic acid. Similar results of pH value were obtained by Herrero, Ordonez, Romero de Avila, Herranz, de la Hoz & Combero (2007), who – in their market studies of fermented sausage – observed fluctuations of pH value between 4.21 and 6.17.

The redox potential significantly differed for all samples. Its higher values were found in the samples with an addition of probiotic as compared to the control, both at the beginning and at the end of storage, which can point to the domination of oxidation in the system. The highest value of the redox potential (Fig. 2A) was found in the sample with an addition of probiotic (about 365 mV) in the 11$^{th}$ week of storage. The value of the redox potential higher in comparison to the control can be caused by the metabolic activity of microorganisms producing hydrogen peroxide as a side product. The value of the redox potential in the control sausage fell by about 10mV during the storage.

The studies found out higher values of the oxidation index in the sample with an addition of probiotic bacteria as compared to the control both after the ripening and after the storage. A higher value of TBARS index can point to advanced transformations of autoxidation of lipids contained in the sample as a result of complex transformation of hydroperoxides becoming the source of short-chain volatile secondary products such as aldehydes, ketones, which give a characteristic aroma to the products. A part of lipid hyperoxides can undergo transformations into cyclical endoperoxides, which are precursors to malone aldehyde.

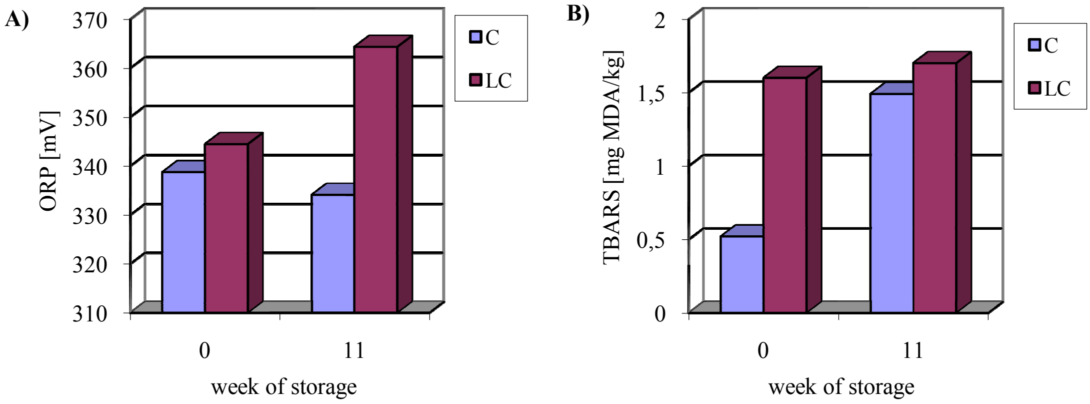


**Figure 2**. The redox potential (ORP) (A) and index TBARS (B) in ripening sausage in 0 and 11th weeks of cool storage (C – control; LC – sample containing Lactobacillus casei ŁOCK 0900; n = 3)

This is confirmed in the studies on the percentage proportion of oxidized myoglobine, whose highest proportion was observed in the sample with an increased addition of probiotic bacteria directly after ripening (Tab.1). A decreased amount of metmyoglobin and increased content of myoglobin after 11 weeks of storage were observed both in the control and in the sample with an addition of probiotic. Studies on meat so far (Greene, 1969) point out that the process of oxidation of oxymyoglobin and myoglobin take place through fat oxidation. This process probably takes place under the effect of free radical or peroxides, which can directly react with myoglobin during oxidation, causing its oxidation.

**Table 1**. Percentage content of Mb, MetMb, MbO2 in ripening sausage in 0 and 11 week of cool storage (C – control; LC – sample containing Lactobacillus casei ŁOCK 0900; n = 3)

| Sample | | Mb [%] | | MetMb [%] | | $MbO_2$ [%] | |
|---|---|---|---|---|---|---|---|
| Week of storage | | 0 | 11 | 0 | 11 | 0 | 11 |
| C | $\bar{x}$ | 17.53 | 16.53 | 74.53 | 70.24 | 7.93 | 13.23 |
| | *SD* | 1.04 | 5.04 | 2.31 | 0.65 | 3.18 | 5.67 |
| LC | $\bar{x}$ | 8.10 | 20.74 | 82.50 | 77.55 | 9.40 | 1.71 |
| | *SD* | 0.36 | 1.54 | 3.55 | 1.33 | 3.48 | 0.99 |

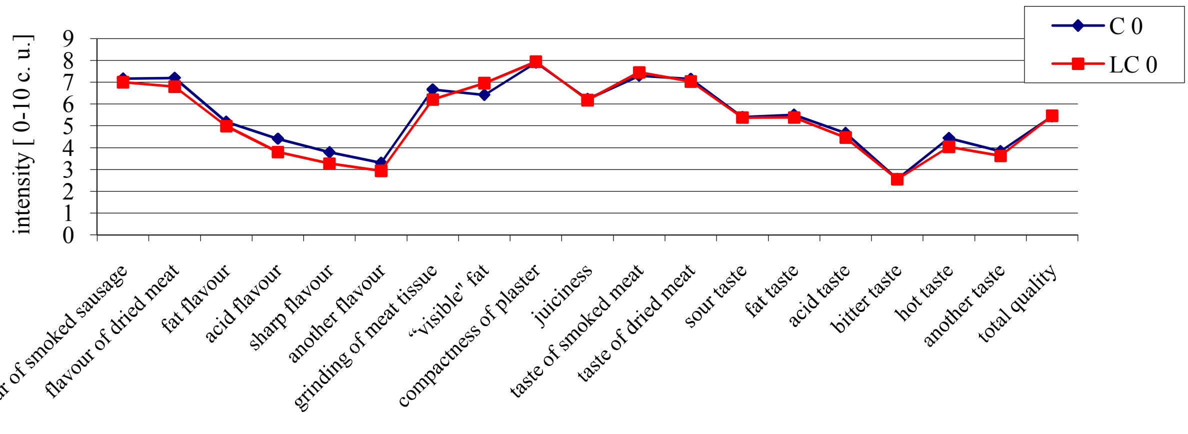


**Figure 3**. Sensory evaluation of the fermented sausage produced with probiotic culture Lactobacillus casei ŁOCK 0900 and glucose (LC0) and the control (C0) after ripening process; o.- odour, int.- intensity, f.- flavor

Mean results of sensory assessment of products directly after ripening are presented in figure 3, whereas the data on the stored products can be found in figure 4. The studied markers of flavour, taste and appearance were characterized by different intensity. At the beginning of storage, the total quality was evaluated at the level of 5.31 – 5.46 c.u.

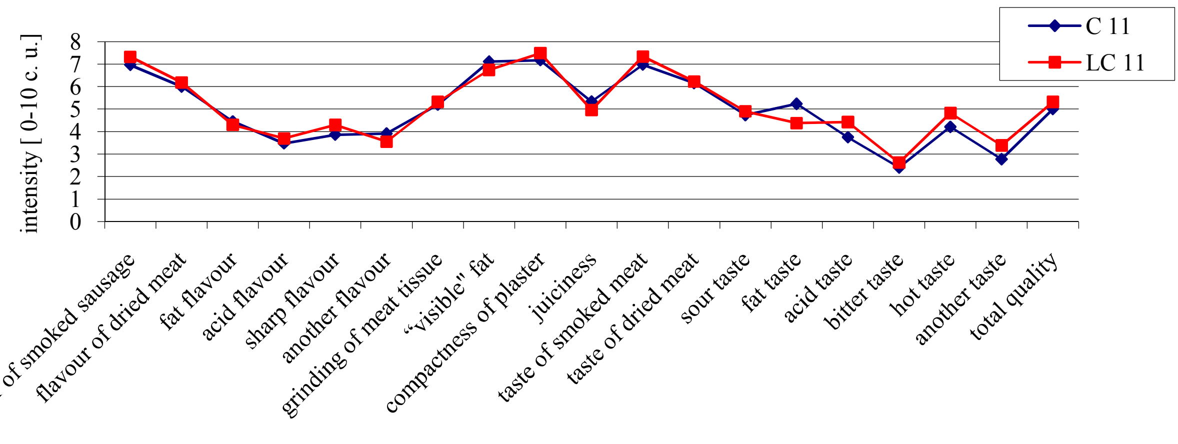


**Figure 4**. Sensory evaluation of the fermented sausage produced with probiotic culture Lactobacillus casei ŁOCK 0900 and glucose (LC11) and the control (C11) after storage; o.- odour, int.- intensity, f.- flavour

The studied markers of flavour, taste and appearance were characterized by different intensity. At the beginning of storage, the total quality was evaluated at the level of 5.31 – 5.46 c.u. Storing products in cool conditions had only a slight effect on a decrease of this marker. C.u. value of about 5 (in the scale of 0-10 c.u.) testifies to the quality accepted by the assessors; however, before being introduced into the market, the product should be improved in respect of its sensory properties.

## IV. CONCLUSION

The count of probiotic bacteria in raw fermented sausage directly after ripening as well as after 11 weeks of storage was $\geq 10^6$ cfu/g, therefore the product met one of the requirements for probiotic food.

There is a possibility of using a strain of probiotic bacteria in raw fermented sausage but the product needs to be improved as for its sensory quality.

An addition of probiotic bacteria to the batter caused intensification of oxidation processes in the product, which was reflected in an increased redox potential, an increased TBARS index and, at the same time, a higher proportion of the oxidized form of myoglobin with brown colour during the storage.

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