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Abstract

The goal of this work consists in adapting local products from Lanhouin to the expectations, tastes and regulatory requirements of European markets. To meet the European markets requirements, two different strategies were applied; one consists to improve the fermentation conditions by adding cultures of *Lactobacillus plantarum* and *Staphylococcus xylosus* as biopreservative organisms and the second consists to improve the storage and the shelf life by using different packaging methods. The improved Lanhouin obtained and presented in forms of powder and cube, packaged in a plastic bag (Type Walovac 90 B) can be stored at ambient temperature $(30 \pm 2 \,^{\circ}\text{C})$ up to 90 days without microbial problem.

1. Introduction

The goal of this work consists in adapting local products from Lanhouin to the expectations, tastes and regulatory requirements of European markets. To meet the European markets requirements: i) marinating system has been done during the ripening step to avoid high microbial proliferation and to limit the production of biogenic amines which are toxics, ii) *L. plantarum* and *S. xylosus* were used as biopreservative agents during fermentation, and iii) shelf life has been increased by improving the packaging of the end-product.

In deliverables 3.2.2.3 and 3.3.3, collections of technological bacteria have been done for Lanhouin. The level of technological bacteria was high for Coagulase Negative *Staphylococcus* (CNS). Even if the technological bacteria such as lactic acid bacteria were not dominant in Lanhouin, the biopreservation by adding cultures together with packaging have been tested to check if the quality and shelf life of the product could be improved. In this case, strains of *L. plantarum* and *S. xylosus* have been tested.

In this deliverable (D3.3.2), we report only results concerning the impact of starter culture for biopreservation. The impact of other factors (marinating, oven drying...) on physico-chemical

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and nutritional characteristics and toxicological indicators will be reported in the deliverable D3.3.5.

2. Methodology

Lanhouin was manufactured according to the flow diagram Figure 1, established by UAC (Benin). More details will be given in Deliverable 3.3.5. For the adaptation to the European market, one of the strategies was based on the addition of culture *L. plantarum* and *S. xylosus* to improve the safety, and the other one was based on the packaging to improve the storage and shelf life of the product.

Fresh fishes (*Pseudotolithus* sp) were bought at Cotonou seaport (Benin). The Fresh fish was washed, scaled, gutted, beheaded and washed before filleting. The fish flesh was separated from skin and bones and marinated for 4 hours. The marinated fish was ground using à blender (Blender, USA).

The biopreservation treatment was made during the fermentation step of Lanhouin. The marinated and ground flesh fish was inoculated with mixture of cultures of *L. plantarum* and *S. xylosus* and homogenized in aseptic conditions. For the mixed starter cultures, the suspensions of different cultures were mixed equally and 1ml of the final mixed culture inoculated into 100 g of marinated fish flesh (this gave approximately a concentration of 10⁶ cells/g of fish flesh). The ingredients were mixed under aseptic conditions and the inoculated fish mixtures were packed in batch of 150 g into 250 sterilized jar. The inoculated fish mixtures were then incubated at 35°C for 36 h. Samples were taken at 0, 6, 12, 24 and 36 h of fermentation for microbiological and physico-chemical analyses. In total, five samples, each representing one fermentation time, were fermented at two different fermentation trials. Similarly, one non-inoculated sample was used as control at each fermentation time.

For the validation of the shelf life, two samples were prepared: one with starter as described above and another without any starter (control). At the end of the fermentation period, the fermented ground fish flesh samples were oven dried at 65°C for 12 h, packaged in a plastic bag (Type Walovac 90B) and stored at 30 ± 2 °C up to 90days.

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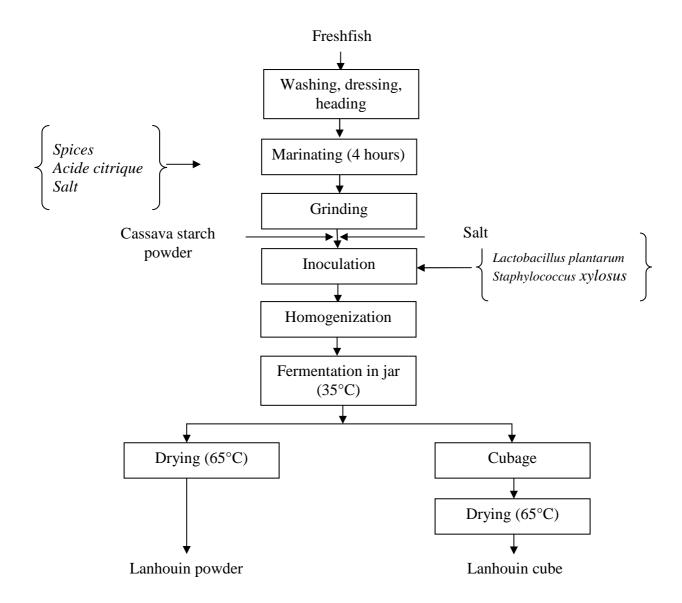


Figure 1: Process flow diagram for improved Lanhouin

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Table 1: Ingredients used to produce improved Lanhouin with starter

	Fish flesh fermented without	Fish flesh fermented
	inoculum (control)	with starter
	10 /100	10. (100
Sodium chloride (NaCl)	10 g/100 g	10g/100 g
Citric acid	1.0 g/100 g	1.0 g/100 g
Citile acid	1.0 g/100 g	1.0 g/100 g
3 Spices	$0.5 \times 3 \text{ g/}100 \text{ g}$	0.5 × 3 g/100 g
•		
Cassava starch powder	5 g/100 g	5 g/100 g
Dianuacamustian stantan		
Biopreservation starter		
Lactobacillus plantarum	ø	1ml/100g
· · · · · · · ·		
Staphylococcus xylosus		

3. Results and discussion

For the control sample (LPW), fermented fish without starter, the total viable counts (TVC) and the level of CNS were low even after a long time of storage. The drying at 65°C is certainly responsible of these low counts. For the sample inoculated with starter (LPS), the ratio TVC/LAB after 90 days of storage at 30 ± 2 °C was inferior to 100 as recommended by the Federation des Entreprises du Commerce et de la Distribution (FCD, 2009) (Figure).

No pathogens were detected in both samples; yeast and molds counts were less than 1 Log CFU/g for all samples.

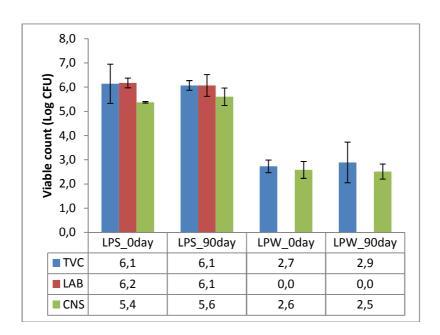


Figure 2: Microbiological status of dried end-product obtained from two fermentation trials

LPS _0: Lanhouin powder obtained after oven drying of ground fish flesh fermented with starter (0 day of storage)

LPS_90: Lanhouin powder obtained after oven drying of ground fish flesh fermented with starter (90 days of storage)

LPW_0: Lanhouin powder obtained after oven drying of ground fish flesh fermented without starter (0 day of storage)

LPW_90: Lanhouin powder obtained after oven drying of ground fish flesh fermented without starter at 90 day of storage

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Conclusion

The Lanhouin powder or cube inoculated with the biopreservative starters or not inoculated can be easy to handle for consumer, with a shelf-life of 90 days when packaged in a plastic bag. However, these preliminary experiments should be completed by sensory test to decide if addition of biopreservative starters is useful or not on this aspect.