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How to apply maritime pine bark extracts to extend the shelf-life of cured-smoked pork products?

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Abstract

The purpose of this work was to establish the experimental design for the application of maritime pine (*Pinus pinaster* Aiton subsp. *atlantica*) bark extracts, as a natural food additive with antimicrobial and antioxidant properties, in the production lines of two cured-smoked pork products, a salami-like product and smoked belly, aiming to extending their shelf-life. The process flow diagram of each product was drawn, describing, in detail, how the product will move during the process steps including the relationship between major components and how the material flows through the various steps and components. The production steps better adjusted to extract addition were selected based on products characteristics. The amount of extract to be added were also determined according to the desired antioxidant performance. The set of analyses to be performed during products shelf-life was defined. Sampling procedures and frequency for the subsequent analysis were established.

Keywords: Maritime pine; Pine bark extracts; Cured smoked meats

1. Introduction

Nowadays there is an increasing consumers demand for natural products without chemical additives. Many bioactive compounds extracted from different plant species are now considered by the food industry, mainly due to their antimicrobial and antioxidant activities, essential requirements for a more natural conservation.

The maritime pine, *Pinus pinaster* Aiton subsp. *atlantica*, is a very abundant tree in western Europe, being the main objective of its plantation the exploitation of wood for various industrial purposes, namely in paper manufacture, as a construction material or for furniture. Recently, maritime pine bark, due to its richness in polyphenols and flavonoids, it has been arousing the interest of the pharmaceutical industry for application in cosmetic products and as a food supplement [1]. Condensed tannins are the main polyphenols of maritime pine bark and exhibit strong antioxidant activity in vitro and in vivo [2, 3, 4, 5].

2. Materials and Method

The survey of the meat company's products, considering their characteristics, namely the different typology and the high fat content, and the high demand from consumers, were the criteria used for the selection of the products to be studied. Two products cured smoked pork products were chosen – a salami-like product "Chouriça" and smoked belly. The aim was to extend the current shelf-life of the selected products from 120 to 180 days, and from 90 to 150 days, of the "Chouriça" and of the smoked belly, respectively, based on the potential antioxidant properties of maritime pine bark extracts applied into products.

First, the process flow diagram of each product was drawn, describing, in detail, how the product will move during the process steps including the relationship between major components and how the material flows through the various steps and components. Secondly, the production steps better adjusted to extract addition were selected, based on products characteristics.

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It was investigated which process step was most suitable for the application of the extract, either from the point of view of industrial practicability, not causing additional time to the process, or from the point of view of maximizing the potential desired antioxidant effect by applying the most downstream step possible.

The amount of pine bark extract to be added were also determined according to the desired antioxidant performance, based on previous works using microwave assisted extraction technique.

The set of analyses to be performed during products shelf-life was defined and sampling procedures and frequency for the subsequent analysis were established.

3. Results and Discussion

3.1. Elaboration of the flow diagrams of smoked pork products production

The flow diagrams of the selected products, "Chouriça" and smoked belly are present in Figures 1 and 2, respectively. The orange frames in the diagrams signal the steps chosen for the application of pine bark extracts. For "Chouriça" two possible steps have been selected as the best or most appropriate, regarding the point of view of industrial practicability and of maximizing the desired antioxidant effect on product. The first step chosen was the end of the second maturation just before filing operation. The pine bark extracts here will be added dried to the mixture of meat and other ingredients before the second maturation; the second addition moment chosen was before the modified atmosphere packaging steps where the liquid extracts will be applied by brushing on the external surface of the "Chouriça".

In the smoked belly production, it was considered that the best moment to add the pine bark liquid extracts was after cutting the belly in portions and before packing. Thus, the extracts will be brushed over the smoked belly pieces.

3.2. What types of products properties need to be measured?

As the objective is to prolong the shelf life of the products, the analyses necessary to validate this extra time were selected. It was decided to carry out during all storage period, the following analysis: Moisture, water activity, pH, total antioxidant capacity by 3 methodologies: DPPH (2,2-diphenyl-1-picryl-hydrazyl-hydrate) free radical method, ABTS radical-scavenging assay and Oxygen Radical Antioxidant Capacity (ORAC) Assay. The analysis of variance (ANOVA) and the Tukey test will be used to determine statistically different values at a significance level of $p<0.05$. Microbiological parameters will be selected according the HPLS Guidelines for Assessing the Microbiological Safety of Ready-to-Eat Foods Placed on the Market [6] (*Mesophiles*, *Escherichia coli*, *Staphylococcus coagulase+*, *Clostridium perfringens*, spores of sulphite-reducing anaerobes, Yeasts and Moulds, *Listeria monocytogenes*; *Salmonella* spp.).

The nutritional composition of final product will be determined following the Regulation (EU) No 1169/2011 on providing food information to consumers [7]. Sensory analyses of the final products were performed by a trained panel of 7 elements.

3.3. How much raw material is necessary to perform all the analytical determinations?

Eight and 9 sampling moments were considered, respectively for "Chouriça" and smoked belly and for all determinations mentioned above, samples will be analysed in triplicate. Thus, the quantities of "Chouriça" and smoked belly samples necessary for all assays during the storage period are shown in Tables 1 and 2, respectively.

Thus, it was concluded that the required amounts of "Chouriça" and smoked belly, to accomplish the study during storage, are respectively 33.948 kg and 15.488 kg. These amounts already include production losses that normally occur during the curing process of the meat plant (weight losses and defective products).

The microbiological quality, the nutritional composition and the sensory properties of the final products at current shelf-life and extended shelf-life limits (120 and 180 days for "Chouriça" and 90 and 150 days for smoked belly) will be evaluated. To accomplish these determinations 7 extra pieces of products are needed.

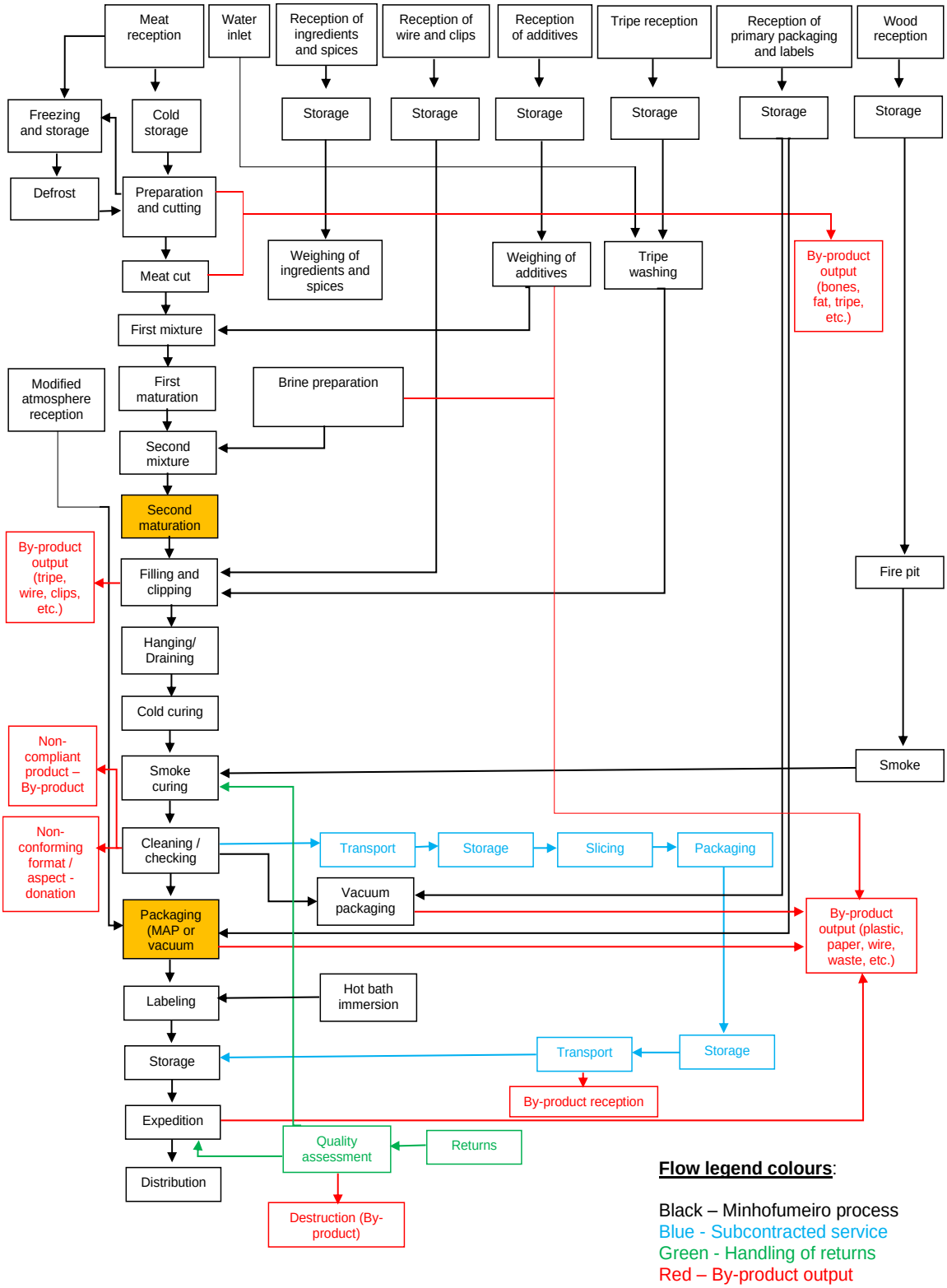

Figure 1. Flow diagram of "Chouriça" production (orange filled frames signal the steps for pine bark extracts addition)

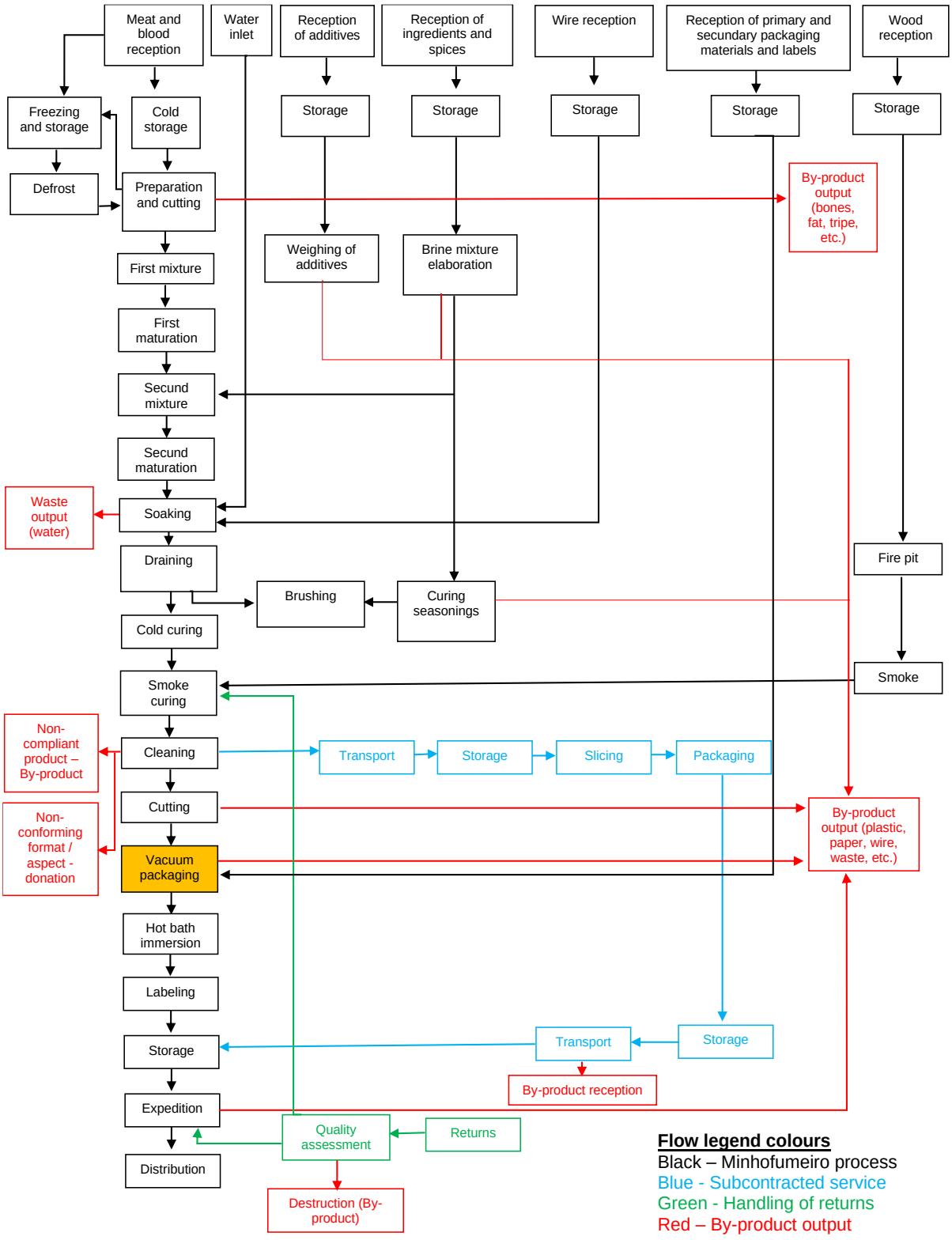

Figure 2. Flow diagram of smoked belly production (orange filled frames signal the steps for pine bark extracts addition)

Table 1. Raw material necessary for the analytical methodologies of "Chouriça" during 180 days of storage

Chouriça						
Sampling times/days		**Nº samples**	**Samples weight (kg)**	**Losses during production (weight loss)**	**Losses during production (defective product)**	**Raw material needed**
t=0	0 dias	3	± 0.2 kg each	± 35 %	± 3 %	3 x 9 x 0.2= 5.4 kg
t=1	30	3				(7+7) x 0.2= 2.8 kg
t=2	60	3				5.4 + 2.8= 8.2 kg
t=3	90	3				35 + 3= 38 %
t=4	120	3 (+ 7)*				8.2 x 0.38= 3.116 kg
t=5	135	3				8.2 + 3.116= 11.316 kg
t=6	150	3				11.316 x 2= 22.632 kg[1]
t=7	165	3				22.632 + 11.316= **33.948 kg**[2]
t=8	180	3 (+ 7)*				

*Seven extra samples are necessary for microbiological, nutritional and sensory analysis of "Chouriça" at current shelf-life (120 days) and extended shelf life (180 days); [1]This amount is due to the tests in the two different steps that were considered; [2]Total quantity required including the control samples

Table 2. Raw material necessary for the analytical methodologies of smoked belly during 150 days of storage

Smoked Belly					
Sampling times/days		**Nº samples**	**Samples weight (kg)**	**Losses during production (defective product)**	**Raw material needed**
t=0	0	3	± 0.2 kg each	± 3 %	3 x 8 x 0.2= 4.8 kg
t=1	30	3			(7+7) x 0.2= 2.8 kg
t=2	60	3			4.8 + 2.8= 7.6 kg
t=3	90	3 (+ 7)*			7.6 x 0.03= 0.144 kg
t=4	105	3			7.6 + 0.144= 7.744 kg
t=5	120	3			7.744 kg + 7.744 kg= **15.488 kg**[1]
t=6	135	3			
t=7	150	3 (+ 7)*			

*Seven extra samples are necessary for microbiological, nutritional and sensory analysis of smoked belly at current shelf-life (90 days) and extended shelf life (150 days); [1]Total quantity required including the control samples

3.4. What is the quantity of pine bark extracts needed for the two meat products?

The calculation of the amount of pine bark extracts needed for this study, was based on the studies by Balzan et al. [8] in which a concentration of 0.75g of active compound/kg of product was used for extending the shelf-life of raw and cooked fresh pork sausages. In that study 0.75g of active compound were added and the extract has an active compound concentration of 65%. Thus, the quantity to add to 0.75= 0.75 x 0.35= 0.2625 g, i.e. 0.75 + 0.26≈ 1g/kg of product.

For "Chouriça", considering the first step, in which the dry extract will be added together with the ingredients in the second mixture, 11.316 kg of "Chouriça" are needed. Thus, the corresponding amount of extract to apply will be= 1g x 11.316 = 11.316g

For the smoked belly and for the second step considered in "Chouriça", where extracts will be applied by brushing on the external surface of the products before packaging, the extract will be added to a solution that will later be brushed on, its concentration is 1/1, that is, for the smoked belly, to

brush the 7.744 kg samples a volume of about 478 mL is necessary, therefore, 478 g of dry substrate are needed; to brush the 11.316 kg of "Chouriça", a solution of about 699 ml is needed, thus requiring 699 g of dry substrate.

Overall, a total of 1188.316 g of pine bark extracts, at minimum, is needed for testing both, smoked belly and "Chouriça".

3.5. What extraction technology will be used for obtention of pine bark extracts?

Microwave assisted extraction (MAE) compared to conventional Soxhlet, showed better performance, both in the extraction yield and total phenolic content of pine bark extracts. In addition, it was faster and more efficient than Soxhlet [9]. As the core of this study is based on the potential antioxidant activity of pine bark extracts due its phenolic content, MAE will be used for obtention of pine bark extracts.

4. Conclusion

The experimental design for the future application of maritime pine (*Pinus pinaster* Aiton subsp. *atlantica*) bark extracts in two cured smoked pork products aiming to extend the shelf life of these products, based on the antioxidant properties of the extracts, was elaborated. This included the elaboration of a detailed process flow diagram of each product including the relationship between major components and how the material flows through the various steps and components, the selection of the steps better adjusted to extract addition, the amount of extract to be added to each product and the set of analyses to be performed during products extended shelf-life was defined. Sampling procedures and frequency for the subsequent analysis were established.

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