# ArtiSaneFood

Innovative Bio-interventions and Risk Modelling Approaches for Ensuring Microbial Safety and Quality of Mediterranean Artisanal Fermented Foods

# Definition of the prototype artisanal products



Partner: UCO







#### Cheese

Based on the flowcharts provided by the partners, a general flowchart of cheese production was elaborated (Figure 1).



**Figure 1.** General Flowchart of Cheese Production. Steps which are not mandatory depending on cheese type (red colour); Critical control points (<> shaped).

#### Description of cheese manufacture production steps

The main steps of cheese production are briefly described below. Operations such as milk filtration before storage or skimming of milk prior to fermentation were not included in the general flowchart, as they were less reported in the selected products.

#### 1. Reception of raw milk

The milk used for cheese production in the present project can be from different species: cow (Squacquerone di Romagna, Camembert de Normandy), goat (Transmontano, Jben, Klila), sheep (Manchego) or it can be a mixture of milk types from more than one species, e.g., goat and sheep (Katiki). At the reception, milk is usually submitted to a series of determinations for quality verification before being used for cheese production.

#### 2. Storage of raw milk

After reception of raw milk in processing environments, it is stored under refrigeration prior to cheesemaking, usually at  $\leq$  6 °C for a maximum of 24 h. This step is considered as a critical control point since failures in temperature control during storage can enable microbial proliferation, leading to milk spoilage or early milk fermentation.

#### 3. Pasteurization

Raw milk has been used for cheese production following traditional manufacture processes. However, for safety reasons and to comply with current microbiological criteria, some cheese elaborations have applied a pasteurization process in milk for cheese production. In the ArtisaneFood project we have both raw-milk (Transmontano, Jben, Klila, Manchego, Camembert) and pasteurized-milk cheeses (Katiki, Squacquerone di Romagna) depending on partner country, therefore, pasteurization was referred as "not mandatory" (red colour, Figure 1), although it can be considered a critical control point when applied. In cases the milk undergoes pasteurization, the process is conducted in a properly designed equipment, most commonly at 72 °C for 15-20 seconds. After pasteurization, milk is cooled down before following the production process steps.

#### 4. Transfer into tanks

After storage or pasteurization, milk is transferred to tanks, where the appropriate environmental conditions are set to kick-off the fermentation process.

#### 5. Fermentation kick-off

Once the target environmental conditions have been set, the fermentation process starts. Fermentation can be either starter-assisted (Katiki, Transmontano, Squacquerone di Romagna, Manchego and Camembert cheeses) or promoted by the autochthonous microbiota of milk (Jben and Klila cheeses), i.e., spontaneous fermentation. Starter cultures added for cheesemaking generally belong to the Lactic Acid Bacteria (LAB) group. Therefore, the environmental conditions may promote the growth of starter cultures or the autochthonous fermenting microbiota, starting the conversion of lactose into lactic acid.

#### 6. Coagulation

Once the cheesemaker has gauged that sufficient lactic acid has been produced during fermentation, rennet is added to cause the casein to precipitate. Rennet usually contains the enzyme chymosin which converts  $\kappa$ -casein to para- $\kappa$ -caseinate (the main component of cheese curd) and glycomacropeptide, which is removed together with the cheese whey. In this step, milk in the liquid state is converted into curd, having a gelled structure. As the curd is formed, milk fat is trapped in a

casein matrix. After adding the rennet, the milk is left to form curds after coagulation. This step is a critical control point.

#### 7. Cutting

Cutting the curd is an essential step in the cheesemaking process, as it increases the surface area for draining whey from the curd. Some recipes advise cutting the curds in larger pieces to produce a cheese with higher moisture, while others make cutting into smaller pieces to produce a drier cheese.

#### 8. Whey draining

After cutting the curd, the cheese whey must be released. Whey is a nutritious medium for bacterial growth, and its presence may lead to spoilage. The cheesemaker must, therefore, remove most of the whey to dehydrate the curd. There are several ways to separate the curd from the whey, which is controlled by the cheesemaker.

#### 9. Moulding, pressing and mould removal

The purpose of the moulding and pressing steps is to produce cheese with a desired size and shape. These steps aid on the removal of free whey, air, and spaces between curd pieces, leading to the consolidation of cheese. The moulds usually are left open on one or both sides and may allow the smooth drainage of whey. The intensity of pressure applied, the duration of pressing as well as other variables as pressing temperature depend on the desired characteristics of cheese. After pressing, the moulds are removed.

#### 10. Salting

The cheese can be salted by immersion in sodium chloride (NaCl) solution, i.e. brine, or by the application of NaCl on cheese surface, i.e. dry salting. NaCl concentrations applied depend on cheese type. After salting in brine solutions, some cheeses can be submitted to a drying period before maturation (Jben, Klila, Camembert de Normandy). Salting is a critical control point.

#### 11. Maturation

Following salting, cheeses must be kept under specific environmental conditions (temperature, relative humidity) to develop their biochemical and physical characteristics. Maturation conditions depend on cheese type. This step is a critical control point.

#### 12. Packaging

The packaging operation appears to be optional in cases cheeses are sold in local markets. However, it was included in the general flowchart since it is a relevant operation to avoid cheese contamination during distribution and retailing, by separating the external environment from the food matrix. Packaging materials used vary among producers.

#### 13. Storage

The cheeses are stored under conditions recommended by the cheesemakers before consumption, during their shelf-life. The shelf-life vary according to the cheese-type having hard cheeses a more extended shelf-life period due to the more limiting water activity values than soft or semi-soft cheeses.

#### Sausage

Based on the flowcharts provided by the partners, a general flowchart accounting with the main steps of the production of fermented sausages was elaborated (Figure 2).



Figure 2. General Flowchart of Sausage Production. Steps which are not mandatory depending on sausage type (red colour); Critical control points (<> shaped).

#### Description of sausage manufacture production steps

The steps of the general flowchart depicted in Figure 2 are briefly described below.

#### 1. Reception of raw meat

The meat used for sausage production in the present project can be pork (Alheira de Mirandela, Nomboulo, Emilia-Romagna salami, Salchichón) or sheep (Dry Merguez, Merguez). As the meat is frequently referred as the main source of contamination of fermented sausages, meat providers may be certified and able to assure the quality of raw meat.

#### 2. Storage of raw meat

After reception of raw meat in processing environments, it is stored under refrigeration prior to sausage production, usually at  $\leq 8$  °C for a maximum of 24 h. This step is considered as a critical control point since failures in temperature control during storage can enable microbial proliferation in meat.

#### 3. Cutting

Meat and fat are cut into pieces with a cutting machine. The aim of this step is to facilitate integration of the raw ingredients Cutting is a critical control point.

#### 4. Mincing

The meat and fat are finely divided into smaller pieces using a meat grinder. This step is not applied in all production processes, for example for Noumboulo sausage. Mincing is a critical control point.

#### 5. Seasoning and addition of other ingredients

Seasoning is the process of adding salts, herbs, or spices to the meat to enhance the flavour or for other technological reasons. Their addition is also justified as antimicrobial agents or as antioxidants. In this step, starter cultures or other ingredients such as vegetables (Salchichón), wine (Emilia-Romagna salami) or sugars to start the fermentation process can be added.

#### 6. Cooking

This step is applied for the production of "Alheira de Mirandela" sausage. It consists of the cooking of the mixture, with addition of bread. It is a critical control point.

#### 7. Homogenization

In this step, the ingredients are mixed in order to obtain a homogeneous mixture.

#### 8. Storage

Step prior to stuffing, also referred as surface curing (Noumboulo sausage), Drainage (Dry Merguez sausage), vacuum-storage (Emilia-Romagna sausage) and maturation (Merguez, Salchichón). This step is a critical control point.

#### 9. Stuffing

The mixture is stuffed into natural or artificial casings from different calibre depending on the product.

#### **10.** Fermentation

Fermentation was not included in the flow diagrams provided by the partners as a separate step. However, generally a fermentation step with specific environmental conditions are reported for fermented sausages. Fermentation can be either starter-assisted or promoted by the autochthonous microbiota of meat, i.e., spontaneous fermentation. Bacteria belonging to the Lactic Acid Bacteria group, coagulase-negative staphylococci (CNS), as well as yeasts may be used as starter cultures to produce fermented sausages. Therefore, the environmental conditions may promote the growth of starter cultures or the autochthonous fermenting microbiota, starting the fermentation of sugar into lactic acid.

#### 11. Maturation

Fermented sausages must be kept under specific environmental conditions (temperature, relative humidity) to dry and develop their biochemical and physical characteristics. Maturation conditions depend on fermented sausage type. This step is a critical control point.

#### 12. Smoking

Smoking is applied in fermented sausages to impart flavour and to avoid the growth of moulds on their surface. Among the selected products, it is applied to produce "Alheira de Mirandela" and Noumboulo sausage. The conditions of smoking application vary among sausages. This step is considered a critical control point.

#### 13. Packaging

The packaging operation appears to be optional for fermented sausages, but it was included in the general flowchart since it is a relevant operation to avoid contamination during distribution and retailing, by separating the external environment from the food matrix. Packaging materials used vary among producers.

#### 14. Storage

The sausages are stored under conditions recommended by the producers before consumption, during its shelf-life. The shelf-life vary according to the sausage-type.

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## Prototype artisanal fermented foods









# Portugal - IPB

- PDO 'Alheira de Mirandela' 'Alheira de Vinhais' sausage
- PDO 'Serrano Cheese'



## PGI 'Alheira de Mirandela' 'Alheira de Vinhais' sausage





Ratio pork:poultry (kg): 1:1 Ratio meats:water (kg): 1:2





Critical Control Point

### PDO 'Queijo de Cabra Transmontano' cheese



FOOD



> Critical Control Point

# Spain - UCO

- Spanish Iberian raw-cured sausage "salchichón"
- PDO 'Manchego' cheese



### Spanish Iberian raw-cured sausage "salchichón"







### PDO 'Queso Manchego' cheese



# Italy - UNIBO

- PDO 'Squacquerone di Romagna' cheese
- Emilia-Romagna salami



### PDO 'Squacquerone di Romagna' cheese











# France - CNIEL

 PDO 'Camembert de Normandie' cheese



### PDO 'Camembert de Normandy' cheese







# Greece - AUA

- Noumbulo sausage
- PDO 'Katiki Domokou' cheese



### Noumboulo sausage















# Morocco - UIZ

- Moroccan Jben cheese
- Merguez sausage











### Merguez sausage





# Algeria - UO

- Algerian Jben cheese
- Klila cheese



### Algerian Jben cheese















# Tunisia – ISBT-UMA

- Dry Merguez sausages
- Kaddid sheep meat
- Leben milk









### Kaddid sheep meat





Proportion of ingredients (w/w): Raw meat (90 %), fat (7.5 %), salt (1.5 %), spices (0.5 %)



> Critical Control Point

## Leben milk (buttermilk)







