



# Report of the Scientific Committee of the Spanish Agency for Consumer Affairs, Food Safety and Nutrition (AECOSAN) about histological methods for differentiating between meat preparations and meat products

## Section of Food Safety and Nutrition

Elena Alonso Lebrero, José Manuel Barat Baviera, María Pilar Conchello Moreno, Ramón Estruch Riba, María Antonia Ferrús Pérez, Guillermina Font Pérez, Susana Guix Arnau, Arturo Hardisson de la Torre, Ángeles Jos Gallego, Ascensión Marcos Sánchez, Amelia Marti del Moral, Olga Martín Belloso, María Aránzazu Martínez Caballero, Alfredo Palop Gómez, Gaspar Pérez Martínez, José Luis Ríos Cañavate, Gaspar Ros Berrueto, Jesús Ángel Santos Buelga, Jesús Simal Gándara, Josep Antoni Tur Marí

## Technical Secretary

Vicente Calderón Pascual

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## Working group

Gaspar Ros Berrueto, Jesús Ángel, Santos Buelga  
(Coordinators)  
José Manuel Barat Baviera  
María Pilar Conchello Moreno  
Jesús Simal Gándara

## Abstract

The distinction between meat preparation and meat product is carried out using different histological methods.

The Section of Food Safety and Nutrition of the Scientific Committee of the Spanish Agency for Consumer Affairs, Food Safety and Nutrition (AECOSAN) has reviewed the information available about five histological methods, their analysis protocol and the relevant scientific literature. It has concluded that no scientific literature has been found that defines the criteria or characteristics of the appropriate analytical methods for determining the loss of histological characteristics of fresh meat. Likewise, there are methods that do not outline objective criteria for categorisation, they depend entirely on the subjectivity of the analyst and are therefore not very reliable. Some of the methods allow for objective quantification of the results and, although they depend on the experience of the analysts for the microscopic examination of the samples, there are well-defined interpretation criteria. However, the uncertainty of the methods is not provided, nor has a validation of them been carried out in accordance with the generally accepted guidelines (for example Standard UNE-EN ISO/IEC 17025:2005).

At present, these methods for histological differentiation are considered as a good starting point for establishing the transition from meat preparation to meat product. However, in order to determine their validity, it is necessary to continue to work to identify robust methods that allow for this assessment and/or quantification of the histological modifications of muscle in its transformation into meat and subsequent loss of structure, not only with micrographic methods, but rather with other complementary methods that objectively indicate the denaturation of meat protein.

## Key words

Meat preparation, meat product, histological methods.

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## 1. Introduction

Regulation (EC) No 853/2004 of the European Parliament and of the Council, of 29 April 2004 (EU 2004a) laying down specific hygiene rules for food of animal origin establishes, among others, the following definitions:

- Meat preparations: Fresh meat, including meat that has been reduced to fragments which has had foodstuffs, seasonings or additives added to it or which has undergone processes insufficient to modify the internal muscle fibre structure of the meat and thus to eliminate the characteristics of fresh meat.
- Meat products: processed products resulting from the processing of meat or from the further processing of such processed products, such that the cut surface shows that the product no longer has the characteristics of fresh meat.

Establishing this differentiation is of great importance given that, depending on the type of meat derivative, the hygiene criteria applied or the use of authorised additives is very different. In the specific case of additives, the use of phosphates (E338, E341, E343, E450-452) is limited in meat preparations to breakfast sausages, Finnish salted Christmas ham, burger meat with a minimum vegetable and/or cereal content of 4 %, mixed with meat, *Kasseler*, *Bräte*, *Surfleisch*, *toorvorst*, *šášlōkk*, and *ahjupraad* and nitrites (E249-250) are limited to marinated pork loin, skewered diced pork, marinated pork cheek, marinated pork rib, *Kasseler*, *Bräte*, *Surfleisch*, *toorvorst*, *šášlōkk*, *ahjupraad*, *kielbasa surowa*, *biała kielbasa*, *kielbasa surowa metka* and *tatar wołowy (danie tatarskie)*. Nitrates (E251-252) are not authorised in any meat preparations. In meat products, in turn, phosphates and nitrites are generally authorised and, with meat products that are not treated with heat, it is also possible to use nitrates (EU, 2008).

The distinction between meat preparation and meat product is made by companies in the sector using various histological methods in order to establish this differentiation and, currently, there are no valid criteria for accepting these methods.

Given that the differentiation can result in a greater use of food additives and, therefore, have a significant influence on the exposure of consumers to additives, and on food safety, it is necessary to find out which histological differentiation methods are acceptable in a way that there are clear criteria that allow the official control authorities as well as the production sector, to ensure proper use of food additives.

To that end, the Section of Food Safety and Nutrition of the Scientific Committee of the Spanish Agency for Consumer Affairs, Food Safety and Nutrition (AECOSAN) has been requested to evaluate the different histological methods available in Spain for differentiating between meat preparations and meat products.

To this effect, AECOSAN has requested information from different laboratories about the methods that they use to differentiate between meat preparations and meat products (Alkemi, S.A., Aquimisa, S.L., Histalim, Prehistol and Visavet), all based on histological techniques.

## 2. Definitions, regulations and guidance on fresh meat and meat derivatives

Annex I of Regulation (EC) No. 853/2004 establishes the following definitions (EU, 2004a):

### 1. Meat

- 1.1. Meat: the edible parts of the animals referred to in points 1.2 to 1.8, including blood.
- 1.2. Domestic ungulates: domestic bovine (including *Bubalus* and *Bison* species), porcine, ovine and caprine animals, and domestic solipeds.
- 1.3. Poultry: farmed birds, including birds that are not considered as domestic but which are farmed as domestic animals, with the exception of ratites.
- 1.4. Lagomorphs: rabbits, hares and rodents.
- 1.5. Wild game: –wild ungulates and lagomorphs, as well as other land mammals that are hunted for human consumption and are considered to be wild game under the applicable law in the Member State concerned, including mammals living in enclosed territory under conditions of freedom similar to those of wild game, and –wild birds that are hunted for human consumption.
- 1.6. Farmed game: farmed ratites and farmed land mammals other than those referred to in point 1.2.
- 1.7. Small wild game: wild game birds and lagomorphs living freely in the wild.
- 1.8. Large wild game: wild land mammals living freely in the wild that do not fall within the definition of small wild game.
- 1.10. Fresh meat: meat that has not undergone any preserving process other than chilling, freezing or quick-freezing, including meat that is vacuum-wrapped or wrapped in a controlled atmosphere.
- 1.13. Minced meat: boned meat that has been minced into fragments and contains less than 1 % salt.
- 1.14. Mechanically separated meat (MSM): the product obtained by removing meat from flesh-bearing bones after boning or from poultry carcasses, using mechanical means resulting in the loss or modification of the muscle fibre structure.
- 1.15. Meat preparations: Fresh meat, including meat that has been reduced to fragments which has had foodstuffs, seasonings or additives added to it or which has undergone processes insufficient to modify the internal muscle fibre structure of the meat and thus to eliminate the characteristics of fresh meat.

### 7. Processed products

- 7.1. Meat products: processed products resulting from the processing of meat or from the further processing of such processed products, such that the cut surface shows that the product no longer has the characteristics of fresh meat.

<b>Table 1.</b> Definitions contained in Regulation (EC) No 853/2004	
Meat	The edible parts of the animals referred to in points 1.2 to 1.8, including blood.
Fresh meat	Meat that has not undergone any preserving process other than chilling, freezing or quick-freezing, including meat that is vacuum-wrapped or wrapped in a controlled atmosphere.
Minced meat	Boned meat that has been minced into fragments and contains less than 1 % salt.
Mechanically separated meat (MSM)	The product obtained by removing meat from flesh-bearing bones after boning or from poultry carcasses, using mechanical means resulting in the loss or modification of the muscle fibre structure.
Meat preparations	Fresh meat, including meat that has been reduced to fragments which has had foodstuffs, seasonings or additives added to it or which has undergone processes insufficient to modify the internal muscle fibre structure of the meat and thus to eliminate the characteristics of fresh meat.
Meat products	Processed products resulting from the processing of meat or from the further processing of such processed products, such that the cut surface shows that the product no longer has the characteristics of fresh meat.

**Source:** (EU, 2004a).

Likewise, the European Commission has published a guidance document on the implementation of certain provisions of Regulation (EC) No 853/2004 (EC, 2009). Section 5.9 referring to meat preparations and meat products lists their definitions (outlined previously in this section) and also outlines that the definitions of “processing”, “unprocessed products” and “processed products” are established in article 2, paragraph 1, letters m), n) and o) respectively, of Regulation (EC) No 852/2004 (EU, 2004b), and they are applicable to all of the food products, including meat (Table 2).

<b>Table 2.</b> Definitions contained in Regulation (EC) No 852/2004	
Processing	Any action that substantially modifies the initial product, including heating, smoking, curing, maturing, drying, marinating, extraction, extrusion or a combination of those processes.
Unprocessed products	Foodstuffs which have not undergone a treatment, including products which have been, for example, divided, parted, severed, boned minced, skinned, ground, cut, cleaned, trimmed, husked, milled, chilled, frozen, deep-frozen or thawed.
Processed products	Foodstuffs resulting from the processing of unprocessed products. These products may contain ingredients that are necessary for their manufacture or to give them specific characteristics.

**Source:** (EU, 2004b).

The document also indicates that all meat products are included in the definition of “processed products”. However, meat preparations may be included under the definition of “unprocessed products” or that of “processed products”. For example, a meat preparation shall be included in the definition of “processed products” if the actions mentioned in the definition of “processing” that are applied, are not sufficient to completely modify the internal structure of the muscular fibre of the meat and if the cut surface shows that the product has the characteristics of fresh meat.

The definitions of “meat preparations” and “meat products” (among other definitions of Regulation (EC) No 853/2004 (EU, 2004a)) are deliberately broad given the innovative nature of meat technology. However, it is necessary for the products to be included under a specific definition and that they are created in accordance with the relevant hygiene requirements. Furthermore, the standards regarding the criteria for selling, additives and labelling refer to said definitions, and it is therefore essential to harmonize their application.

Products can be put into the market before the processing results in a complete modification of the meat/internal structure of the muscle fibre. In such cases, the degree to which the characteristics of the fresh meat have disappeared at the time when the product is sold, will determine the definition that it is included under. If the characteristics of fresh meat do not disappear completely, the product will be considered for inclusion under the definition “meat preparations”. If, on the contrary, the characteristics of fresh meat disappear completely, it will be considered for inclusion under the definition “meat products”.

Therefore, the following clarifications can be made:

- Meat that has been chopped up includes minced meat. Consequently, minced meat to which food products, condiments or additives have been added, comes under the definition of meat preparations.
- Fresh meat that has been completely marinated through to the centre is included under the definition of “meat products”, as the definition of “processing” includes marinating. This process produces the denaturation of the proteins of the muscle fibre, which constitutes a modification of the internal muscle fibre structure and the elimination of the characteristics of fresh meat on the cut surface.
- Fresh meat that has not been marinated completely to the centre is included under the definition of “meat preparations”, as the modification of the internal muscle fibre structure is not complete and the cut surface still has the characteristics of fresh meat.
- Meat that is salted or cured, even completely, that is sold during the initial maturation stage, and that has not been subjected to any other processing, for example cooking or drying, is included under the definition of “meat preparations”, as it maintains the characteristics of fresh meat. When the product is subjected to new processes, such as drying, that result in the elimination of the characteristics of fresh meat, it will be included under the definition of “meat products”.
- Meat subjected to fast frying, which stays raw in the centre, is included under the definition “meat preparations”, as the heat processing has been insufficient to modify the internal

muscle fibre structure of the meat, and thus to completely eliminate the characteristics of the fresh meat. Consequently, the cut surface still has the characteristics of fresh meat.

- Meat subjected to fast frying that is marinated to the centre or completely cured before being fried is included under the definition of “meat products”, as the marinating or complete salting process eliminates the characteristics of fresh meat.
- Meat that has been completely fried, but which must be cooked before consumption, is included under the definition of “meat products”, as the frying has modified the internal muscle fibre structure of the meat to the point where the characteristics of fresh meat have disappeared and the cut surface no longer has the characteristics of fresh meat.

<b>Table 3. Summary of clarifications</b>		
<b>Fresh meat</b>		
Meat that has been chopped up including minced meat	Minced meat that food products, condiments or additives have been added to.	Meat preparations
<b>Marinades</b>		
Fresh meat that has been marinated completely through to the middle	The definition of “processing” includes marinating. This process produces the denaturation of the proteins of the muscle fibre, which constitutes a modification of the internal muscle fibre structure and the elimination of the characteristics of fresh meat on the cut surface.	Meat products
Fresh meat that has not been marinated completely through to the middle	The modification of the internal muscle fibre structure is not complete and the cut surface still has the characteristics of fresh meat.	Meat preparations
<b>Salted or cured</b>		
Meat that is salted or cured, even completely,	that is sold during the initial maturation stage, and that has not been subjected to any other processing, for example cooking or drying, is included under the definition of “meat preparations”, as it maintains the characteristics of fresh meat.	Meat preparations
	When the product is subjected to new processes, such as drying, that result in the elimination of the characteristics of fresh meat, it will be included under the definition of “meat products”.	Meat products
<b>Frying</b>		
Meat subjected to fast frying, that remains raw in the middle	The heat process has been insufficient to modify the internal muscle fibre structure of the meat, and thus to completely eliminate the characteristics of the fresh meat. Consequently, the cut surface still has the characteristics of fresh meat.	Meat preparations
Meat that is subjected to fast frying that is marinated through to the centre or completely cured before being fried	The marinating or complete salting process has eliminated the characteristics of fresh meat.	Meat products
Meat that has been completely fried, but which must be cooked before consumption	The frying has modified the internal muscle fibre structure of the meat to the point where the characteristics of fresh meat have disappeared and the cut surface no longer has the characteristics of fresh meat.	Meat products

On the other hand, the AECOSAN has published an explanatory note on the use of additives in meat derivatives (AECOSAN, 2015), point 4 of which outlines that meat derivatives not subjected to processing are those that have been created with fresh meat, including meat that has been chopped or minced, to which other food products, condiments or additives have been added. This group includes, but is not limited to fresh llonganissa, sausage, *butifarra* and *chorizo*.

This type of meat derivatives has not been treated, therefore there is no processing, which means they are classified as “meat preparations”.

This consideration as “meat preparations” should be extended to all meat derivatives that use the term “fresh” in their name, as by definition “meat products” show on the cut surface that the product no longer possesses the characteristics of fresh meat.

Therefore, products such as fresh *chorizo*, fresh *chistorra*, fresh sausage, llonganissa, *butifarra*, etc., are “meat preparations” and can only include among their ingredients additives from group 8.2 (meat preparations).

The labelling of these meat derivatives should include indications from the manufacturer regarding the storage temperature during the marketing of the product in order to guarantee its remains a fresh product during its useful life, as well as indications for handling the product and consumption recommendations (for example, do not consume raw, consume completely cooked, etc.). These indications should be respected until the product is purchased by the consumer.

### 3. Considerations regarding the analytical methods

The analytical methods have to be suitable for the test that is carried out. Preferably, methods published as standards should be used (international, national, regional). The non-standardised methods (developed by the laboratory itself) should be validated beforehand (that is to say, confirm, through examination and the contribution of objective evidence, that they comply with the particular requirements for a specific intended use). For the validation, it is advisable to use one or several of the following techniques (ISO/IEC, 2005):

- Calibration using reference materials.
- Comparison with results obtained using other methods.
- Inter-laboratory comparison.
- Systematic assessment of the factors that influence the result.
- Assessment of the uncertainty of the result.

It is necessary to have an analysis method based around determining the histological characteristics of the meat derivative in order to be able to determine if the internal muscle fibre structure has been modified and, therefore, if the characteristics of fresh meat have been eliminated (EU, 2004a).

In order for the analysis method to be interpretable by all of the interested parties (analysis laboratory, client, administration, official control) it would be necessary to assign quantitative values to the samples, establishing thresholds that make it possible to categorise meat derivatives and the uncertainty of measurements. Furthermore, it could include repeatability/reproducibility



limits, robustness to external influences (for example in the case of frozen samples), as well as other values that are commonly used to validate analytical methods.

Regulation (EC) No 853/2004 (EU, 2004a) does not outline at what point the internal muscle fibre structure is considered to have been completely modified and when the characteristics of fresh meat have been eliminated. Therefore, the methods can have flexibility in the interpretation of the analytical results. However, this flexibility has to be clearly established in the specifications of the method. That is to say, there must be objective values and confidence intervals that make it possible to clearly differentiate between the different categories of meat derivatives.

## Conclusions of the Scientific Committee

The following conclusions have been reached following a review of the information available on the five methods evaluated, the protocols of the histological methods of existing analyses and the relevant scientific literature:

- It has not been possible to find scientific literature that defines the criteria or characteristics of suitable analysis methods for determining the loss of the histological characteristics of fresh meat.
- The analytical methods that are offered to the meat industries carried out microscopic analysis of samples of the meat derivative. The loss of striations and cell degeneration are generally considered to be the most suitable parameters for estimating the disappearance of the characteristics of fresh meat, which is consistent with the requirements of Regulation (EC) No 853/2004 (EU, 2004a).
- In order to consider that the product no longer possesses the characteristics of fresh meat (meat product) there needs to be a total or complete modification of the cut surface. If this is not the case, we can only talk of there being a substantial modification, therefore it would be a processed meat preparation. The application of unjustified correction methods in the validation of the method to estimate the total modification (for example 75 %) would not be compatible with the definition of the meat product.
- There are methods that do not outline objective criteria for categorisation, they depend entirely on the subjectivity of the analyst and are therefore not very reliable.
- Some of the methods allow for objective quantification of the results and, although they depend on the experience of the analysts for the microscopic examination of the samples, there are well-defined interpretation criteria. However, the uncertainty of the methods has not been clarified, nor has a validation of them been carried out in accordance with the generally accepted guidelines (for example Standard UNE-EN ISO/IEC 17025:2005; ISO/IEC, 2005).
- There are meat derivatives that can modify their characteristics during their useful life period. For example, it can be seen that fresh chorizo is difficult to categorise, even by using objective parameters. Some samples are considered to be meat preparations and others to be meat products. Given that the interpretation guides consider derivatives that bear the adjective "fresh" to be preparations, companies would need to be aware of these possible modifications, and be careful with the terms that they use on the labelling of these foods.

- At present, these methods for histological differentiation are considered as a good starting point for establishing the transition from meat preparation to meat product. However, in order to determine their validity, it is necessary to continue to work to identify robust methods that allow for this assessment and/or quantification of the histological modifications of muscle in its transformation into meat and subsequent loss of structure, not only with micrographic methods, but rather with other complementary methods that objectively indicate the denaturation of meat protein.

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