Fermented meat products

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Abstract

Fermentation is an ancient process that has been used to prevent foods and this process is widely practiced in the meat industry as a method of preparing and preserving meat. The bacteria, Lactobacillus, Staphylococcus and Micrococcus, play the most important role in this well known microbial process. Additionally, certain types of yeasts and molds are also used in production of some special fermented meat products. The meat products produced with this method are mostly approved in the meaning of consumers’ acceptance. Sucuk, Hungarian salami and Chorizo are the well known...
examples of this kind of products. Numbers of different parameters have to be controlled to obtain high quality fermented meat product. These parameters can be divided in two groups as intrinsic and extrinsic factors. In this chapter, fermentation practices in meat technology and the parameters that should be controlled to produce good quality fermented product are examined.

1. Fermentation

Fermentation is a term first used with regard to the foaming that occurs during the manufacture of wine and beer. The process dates back to at least 6,000 B.C. when the Egyptians made wine and beer by fermentation. From the Latin word *fermentare*, to cause to rise. The term “fermentation” is now used to refer to so many different processes that fermentation is no longer accepted for use in most scientific publications. Three typical definitions are given below:

i. A process in which chemical changes are brought about in an organic substrate through the actions of enzymes elaborated by microorganisms.

ii. The enzyme-catalyzed, energy-yielded pathway in cells by which “fuel” molecules such as glucose are broken down anaerobically. One product of the pathway is always the energy-rich compound adenosine triphosphate (ATP). The other products are of many types: alcohol, glycerol, and carbon dioxide from yeast fermentation of various sugars; butyl alcohol, acetone, lactic acid, and acetic acid from various bacteria; citric acid, gluconic acid, antibiotics, vitamin B_{12} and B_{2} from mold fermentation. The Japanese utilize a bacterial fermentation process to make the amino acid, L-glutamic acid, a derivative of which is widely used as a flavouring agent.

iii. An enzymatic transformation of organic substrates (feedstocks), especially carbohydrates, generally accompanied by the evolution of gas. A physiological counterpart of oxidation, permitting certain organisms to live and grow in the absence of air; used in various industrial processes for the manufacture of products such as alcohols, acids, and cheese by the action of yeasts, molds, and bacteria. Alcoholic fermentation is the best known example. Also known as zymosis. The leavening of bread depends on the alcoholic fermentation of sugars. The dough rises due to production of carbon dioxide gas that remains trapped within the viscous dough (1).

Although fermenting and drying are probably among the oldest methods of preserving and keeping foods, and practical experience
accumulated over several thousands years as well as the results of more than 50 years of research in this field are available, the complex nature of these processes, influenced by physical, chemical, biological and microbiological factors, still makes it necessary to continue this research (2).

Even though the biochemical details are out of this chapter, there are two major pathways for hexose (e.g., glucose) fermentation by lactic acid bacteria (LAB) that are the most important microorganisms for the production of fermented meat products (Figure 1). The transport and phosphorylation of glucose may occur as transport of free sugar and phosphorylation by an ATP-dependent glucokinase. Some LAB species use the phosphoenolpyruvate : sugar phosphotransferase system (PTS), in which phosphoenolpyruvate is the phosphoril donor. In either case, a high-energy phosphate bond is required for activation of the sugar (3).

2. Meat fermentation

Fermentation is a simple and inexpensive method for preservation of meat and meat products since antiquity. It has an added advantage of creating specific products with good aroma. Acid production (lowering the pH), H₂O₂ production and bacteriocins produced either singly or in combination by starter cultures are responsible for preventing the growth of food-borne pathogens and spoilage microorganisms in meat. Interest on the use of fermentation techniques in meat has been revived since there is now a restriction on the use of chemical additives for preservation of meat and meat products. Fermentation is a natural biological process that can be easily adopted in developing countries where refrigeration facilities are lacking (4). Meat fermentation is a complex biological phenomenon accelerated by the desirable action of certain microorganisms in the presence of a great variety of competing or synergistically acting species mainly acquired from meat (5). Dry-cured sausages are one of the oldest forms of preserving meat and are typical of Mediterranean countries with a dry climate (Spain, Italy, France, Portugal and Turkey). In contrast, smoke-cured sausages, or cooked sausages prevail in countries with a colder climate (6). In general, the qualitative characteristics of naturally fermented sausages are known to be largely dependent on the quality of the ingredients and raw materials, the specific conditions of the processing and ripening, and the composition of the microbial population. Since different genera and species, and even strains, have been shown to significantly affect the sensory traits of fermented sausages, the microbial ecology of fermented sausages has become of increasing interest over the last few decades (7).
Figure 1. Major fermentation pathways of glucose: (A) homolactic fermentation (glycolysis, Embden-Meyerhof-Parnas pathway); (B) heterolactic fermentation (6-phosphogluconate/phosphoketolase pathway). Selected enzymes are numbered: 1. Glucokinase; 2. fructose-1,6-diphosphate aldolase; 3. glyceraldehyde-3-phosphate dehydrogenase; 4. pyruvate kinase; 5. lactate dehydrogenase; 6. glucose-6-phosphate dehydrogenase; 7. 6-phosphogluconate dehydrogenase; 8. phosphoketolase; 9. acetaldehyde dehydrogenase; 10. alcohol dehydrogenase (3).
3. Microorganisms used in meat fermentation

Meat is mostly subjected to deterioration by the growth of several microorganisms. While these spoiling microorganisms are not acceptable in raw meat and meat products, certain types of fermentative microorganisms, especially the LAB, either exist in raw meat naturally (natural micro flora) or added by producers, are used for the production of fermented meat products. These desirable microorganisms added to the meat dough are called as starter cultures and they can be single species or the mix of certain microorganisms.

The microorganisms of importance during fermentation and maturation of fermented sausages are Gram positive and rod shaped, belonging to the genera *Lactobacillus*, *Micrococcus* and *Staphylococcus* (8). In some fermented sausages, particularly those produced in France, Spain and Italy, the sensory properties of the products are also influenced by the development of the surface flora, consisting of moulds and yeasts (9). The surface flora of many air-dried dry sausages consists mainly of moulds of the genus *Penicillium* and protects the products and gives them typical aromas (8).

Most fermented foods, including the major products that are common in the western world, as well as many of those from other sources that are less well characterized, are dependent on LAB to mediate the fermentation process (10). LAB enhance the physico-chemical properties of sausages and restrict the growth of some undesirable microorganisms. However, LAB have also been reported as major spoilage microorganisms in meat products. These types of microorganisms may cause slime and sour odour formation in sausages (11).

The broadly used microorganisms in starter culture preparations are given in Table 1.

All LAB produce lactic acid from hexose and since they lack functional heme linked electron transport chains and a functional Krebs cycle, they obtain energy via substrate level phosphorylation. The lactic acid produced may be L(+) or, less frequently, D(-) or a mixture of both. LAB are generally

<table>
<thead>
<tr>
<th>Lactic Acid Bacteria</th>
<th>Micrococcaceae</th>
<th>Yeasts*</th>
<th>Molds*</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Lactobacillus</em></td>
<td><em>Peptococcus</em></td>
<td><em>Debaryomyces</em></td>
<td><em>Penicillium</em></td>
</tr>
<tr>
<td><em>L.plantarum</em></td>
<td><em>P.acidilactici</em></td>
<td><em>D.hansenii</em></td>
<td><em>P.nalgiovene</em></td>
</tr>
<tr>
<td><em>L.sake</em></td>
<td><em>P.pentosaceus</em></td>
<td></td>
<td></td>
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<tr>
<td><em>L.carnosus</em></td>
<td><em>S.xylosus</em></td>
<td></td>
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<tr>
<td><em>L.pentosus</em></td>
<td><em>S.carnosus</em></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>M.varians</em></td>
<td><em>Micrococcus</em></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

*Yeasts and molds are used for surface applications.
mesophilic but can grow at temperatures as low as 5°C or as high as 45°C. Similarly, while the majority of strains grow at pH 4.0-4.5, some are active at pH 9.6 and others at pH 3.2. Strains are generally weakly proteolytic and lipolytic and require preformed amino acids, purine and pyrimidine bases and group B vitamins for growth (10).

Micrococcaceae species are used to enrich fermentative microorganisms during aging of the fermented meat products in order to enhance the colour stability of the cured-meat and prevent rancidity. The activity of this microbial group prevents the growth of spoiling microorganisms, decreases processing time and contributes to flavour development (13). It is generally accepted that micrococci participate in desirable reactions occurring during the ripening of dry fermented sausages, such as colour stabilisation, decomposition of peroxides, proteolysis and lipolysis. Some studies clearly show that the aroma of fermented meat products can be modulated by the presence of Staphylococcus spp. The relative importance of Micrococcaceae and lactic acid bacteria in aroma development has recently been reviewed and it is reported that S.xylosus and S.carnosus are able to produce esters and other important aromatic components from amino acids. These strains also prevent the formation of off-flavours, mainly by their high nitrate reductase and catalase activities (14).

The microorganisms used as starter culture should have some special effects on certain quality characteristics of fermented meat products. The colour of the product is affected by the drop in pH levels that is because the activity of LAB and likewise nitrate reduction, O₂ utilization and H₂O₂ decomposition effects of Micrococcaceae influence the colour. Also the colour of the products in which moulds and yeasts are used as starter culture is affected by H₂O₂ decomposition and O₂ utilization activities of these microorganisms as well. Aroma is another important quality feature of fermented meat products. It is directly affected by the starter cultures. LAB affect the aroma with producing acid while Micrococcaceae, moulds and yeasts have more efficiency on this property by decomposition of proteins, fats and delaying action on rancidity. LAB are the only genera responsible for the texture of the product with pH decreasing effect. However, in some products such as Hungarian Salami, moulds and/or yeasts are the microorganisms responsible for appearance, prevention from O₂ and light and they protect the product against drying. The textural or structural stability in market is also one of the main quality indicators for fermented meat products. It’s important that the product can keep its quality characteristics stable until consumption. Drop in pH, nitrate reduction and prevention of spoilage microorganisms in product are the main effects of LAB and Micrococcaceae on the market stability of product. Finally, nitrite decomposition effects of
LAB and *Micrococcaceae* are helpful for decreasing the level of chemical residues in the product (12). It is recommended that starter cultures for production of biogenic amines be tested in model media. Starter cultures can influence production of biogenic amines in dry fermented sausages by two ways: indirectly, by its ability to depress the growth of microorganisms with decarboxylase activity and, directly by its own decarboxylase activity (15).

4. Production of fermented meat products

There are hundreds of different formulations for fermented meat products all over the world. In every country, different names and different processes can be found and most of them are traditional for the country. Sucuk (Turkey), Hungarian Salami, Kantwurst (Austria), Lup cheong (China), Milano Salami (Italy), summer sausage (USA), salami aeros (Greece), Chorizo (Mexico, Spain), Salchichon (Spain), Fuet (Spain) and Pepperoni (Canada, USA) are the well known examples of fermented meat products.

Modern types of fermented dry sausage can be divided in two groups: sausages that are ripened over 4 weeks (leading to a firm texture with a mildly acidic, salty taste) or semi-dry sausages which are, depending on the product diameter, ripened for between 7 and 28 days and are less intensively dried (delivering a strongly acidic, salty, mild taste and a softer texture) (16). Fermented sausages, even though they are produced with different recipes and technologies, have the common properties of being formulated with lean meat and fat (at different proportions depending of the recipe), being able to be stored at room temperature and having a high microbial load (6).

Although most of the fermented meat products are produced from red meat, especially pork and beef, as reported by Alter et al. (17), consumers’ demand for low fat sausages has increased and there has been a concurrent increase in poultry sausage consumption. It was reported that in the year 2003, approximately 108,000 tones of poultry sausages were consumed in Germany, corresponding to 8.5% of the complete sausage consumption in that year (17).

Aquilanti et al. (18) investigated typical Italian salami Ciauscolo and reported that the physico-chemical analyses had showed a progressive drop in pH and water activity during ripening. The viable counts had revealed a dominance of LAB over coagulase negative cocci and yeasts. From the molecular identification of the isolates, the prevalence of *Lactobacillus curvatus*, *Lb.plan tarum* and *Staphylococcus xylosus* had been shown among the bacteria, while *Debaryomyces hansenii* had been the prevalent species among the yeasts, and it had been isolated throughout the whole ripening process. Minority species, namely *Rhodotorula mucilaginosa* and *Trichosporon brassicae*, had been also recovered from the meat batter (18).
Samelis et al. (5) conceded the virtual importance of starter cultures in eliminating health risks but they also reported that the replacement of the commercially available starter cultures containing species of low adaptation, i.e. \textit{Lb.\textit{pentosus}}, \textit{Lb.\textit{carnosus}} and pediococci, in traditional meat fermentations with 'wild' strains from the organisms governing the process, namely \textit{Lb.\textit{sake}}, \textit{S.\textit{xylosus}} and possibly nitrate-reducing \textit{S.\textit{saprophyticus}}, in order to develop new, company-specific starter cultures of high performance (5).

As a well known example for fermented meat products, sucuk, Turkish dry-fermented sausage, is produced in a large amount in various parts of Turkey (19). Although the natural microflora of meat is generally used as a starter culture, the predominant lactic organisms in Turkish style sausages are \textit{Lb.\textit{sake}}, \textit{Lb.\textit{plantarum}}, \textit{Lb.\textit{curvatus}} and \textit{Lb.\textit{brevis}} (8). Sucuk is produced in two stages; first stage is the fermentation either by added starter culture or by chance inoculation from the raw material or environment during the manufacturing. The second stage is the drying of the sucuk under the controlled (in commercial sucuk) or climatic condition (in traditional sucuk). Combination of these two stages is known as the ripening period. The main physical and chemical changes take place during this period. Desired physical quality attributes of sucsus (texture, colour, flavour, and odour) are mainly developed in this period (20). The basic formulation of Turkish sucuk can be written as follow;

\begin{itemize}
  \item 55 kg beef meat with 15\% fat (R3*)
  \item 30 kg beef meat with 3\% fat (R2*)
  \item 10 kg tail fat (L4*)
  \item 1.5 kg fresh garlic**
  \item 1 kg cumin
  \item 0.5 kg paprika (hot)
  \item 0.3 kg paprika (sweet)
  \item 0.3 kg allspice
  \item 0.4 kg dextrose***
  \item 0.5 kg black pepper
  \item 0.5 kg vegetable oil
  \item 2 kg NCS****
\end{itemize}

\begin{itemize}
  \item Starter culture
\end{itemize}

* Meat part codes according to classification of Koch, 1986 (21).
** Use of garlic can be changed according to seasons. In summer time \%1 fresh garlic addition is generally enough.
*** Requirements of the starter culture are determinative for the selection of monosaccharide type and quantity.
**** Nitrite Curing Salt: NaCl including 0.5\% NaNO_2
Also there are some special formulations for sucuk that include fenugreek, rosemary, cinnamon, white pepper or some other traditional spices. In Figure 2 general production process of sucuk is written and the keystones of the production are emphasized with details.

Another two well known fermented meat products are salami aeros and Hungarian salami. The basic formulations for Greek dry salami (salami aeros) can be written as follow (5);

76 kg pork meat  
24 kg pork back fat  
2.5 kg salt  
2.5 kg skim milk powder  
1.5 kg sugars (maltodextrin)  
0.3 kg mixed spices (black and red pepper, clove)  
0.1 kg garlic  
0.2 kg white wine  
0.02 kg NaNO₂  
0.02 kg NaNO₃  
0.06 kg Na-ascorbate  
Starter culture*  

* Natural microflora of meat is generally used as a starter culture.

Basic formulation for Hungarian Salami;

35 kg beef meat  
30 kg pork shoulder  
30 kg pork back fat  
2 kg NCS  
0.40 kg dextrose  
0.5 kg powdered garlic  
0.5 kg paprika (sweet)  
0.3 kg white pepper  
Starter culture*  

* Bacteria culture for fermentation and mold to ensure surface moldy structure.

5. Factors affecting the quality of fermented meat products

The growth of microorganisms, so of the starter cultures, depends on the existence of many different intrinsic and extrinsic factors. Since the ripening
Figure 2. General Production Process of Turkish Sucuk
process is the main step of the production of fermented meat products, these factors directly affect the final product quality. Table 2 shows the intrinsic and extrinsic factors that affect the microbial growth (22).

**Table 2.** Some intrinsic and extrinsic factors affecting microbial growth (22).

<table>
<thead>
<tr>
<th>Intrinsic Factors</th>
<th>Extrinsic Factors</th>
</tr>
</thead>
<tbody>
<tr>
<td>pH, acidity, acidulant identity, % buffering power</td>
<td>Temperature</td>
</tr>
<tr>
<td>Water activity and content, humectant identity</td>
<td>Relative humidity</td>
</tr>
<tr>
<td>Redox potential</td>
<td>Light intensity and wavelength</td>
</tr>
<tr>
<td>Presence of antimicrobials</td>
<td>Atmospheric gas composition and ratio</td>
</tr>
<tr>
<td>Identity and distribution of natural microbial floras</td>
<td>Packaging characteristics and interactions</td>
</tr>
<tr>
<td>Presence of physical structures</td>
<td>Processing characteristics and interactions</td>
</tr>
<tr>
<td>Availability of nutrients</td>
<td>Storage, distribution and display considerations</td>
</tr>
<tr>
<td>Colloidal form</td>
<td></td>
</tr>
<tr>
<td>Substrate surface to volume ratio</td>
<td></td>
</tr>
</tbody>
</table>

### 5.1. Intrinsic factors

#### 5.1.1. pH

Dried sausage, during manufacture, undergoes numerous chemical, biochemical and microbiological changes which impart its characteristic flavour. Among these reactions, acidification occurs chiefly as a result of the breakdown of sugars into lactic acid, and also of lipids into free fatty acids (23). Starter cultures used in dry sausage manufacturing process in the North of Europe produce sufficient amounts of lactic acid in order to decrease pH of meat to values 4.8-5.0. The low pH value is the basis for the safety as well as texture and colour of dry sausage (24). The acidification process of meat products should be slow, since a fast pH drop results in a massive protein denaturation that makes the product unacceptable (25).

In sucuk production, alike with other fermented products, the pH of the final product is dependent to the pH of raw material. Meat has a pH value between 5.4-5.8 is suitable for the production of sucuk. In the first 36 hours of ripening period, the pH should decrease to 5.3-5.4 and after 72 hours 4.9-5.0 interval is accepted (26). A pH value of 4.8 is a critical point for sucuk. Under this pH value some important quality defects can occur such as off-flavour and undesirable product texture. Traditionally produced and marketed sucuk should have a pH value between 5.1-5.2 and should not be over 5.4. Accordingly the sucuk with a pH of 5.8-5.9 is accepted as inconsumable (12, 27).

#### 5.1.2. Meat type

The quality of raw material directly affects the properties of final product. Although normal meat is always the first choice of producers in the production, according to financial cares the PSE (pale, soft, exudative) or
DFD (dark, firm, dry) meats can be used. PSE meats with high water content are more suitable than DFD meats for fermented meat products. The higher the water content of the raw material, the faster the water releasing in the ripening chamber. However the mix of PSE and normal meat are mostly used as raw material in fermented meat products.

In traditional sucuk production, water buffalo or calves are used in Turkey. However the population of water buffalos has been decreasing in last decades, and the main raw material has changed to beef meat significantly (26). Lamb meat is also used in sucuk production, especially in traditional products, as well. However the special odour of lamb meat limits the usage in industrial scale.

5.1.3. Fat content

The fat content in most of the fermented meat products is between 25 to 45% in order to fulfill consumer expectations in different countries. This value, for example, should be less than 40% for Turkish fermented sucuk and 35% for traditional Greek sausages according to Food Legislations of these countries (27, 28).

Fat acts as a reservoir for flavour compounds and contributes to product texture and juiciness. In raw sausages the granulated fat helps to loosen the sausage mixture and this aids the continuous release of moisture from the inner layers of the product. For traditional Greek sausages, fat level significantly affects the processing and quality characteristics especially weight loss, brine concentration, water activity, colour, consistency, odour and the taste of the product (28).

Products such as sucuk contains high fat, which is visible when the product is sliced. At the beginning of the production the sucuk dough is prepared from fresh meat and fat; and its initial fat content is generally around 10–20%. However, the fat content of the product increases to 30–40% after drying (29). Sheep tail fat is generally used in sucuk production. Beef tallow and beef kidney fat from beef can also be used in the production (26).

5.2. Extrinsic factors

In the dry sausages industry, ripening is nowadays considered to be one of the most important stages of the integrated supply chain needed to ensure that the end products have the final requirements in terms of quality and safety standards (30). Temperature, relative humidity and air flow rate in the ripening room have determinative effects on the manufacturing practices.

5.2.1. Temperature

In the production process of almost all fermented meat products there are four important steps; preparation, curing, fermentation and storing.
Temperature has a key role in every step. According to the requirements of microorganisms, the producer has to choose right temperature values to support (in curing and fermentation steps) or prevent (in preparation and storing steps) microbial growth.

The temperature directly affects pH, water activity, microbial growth and texture of fermented sausages. There is a strong correlation between temperature and ripening as well as drying (31).

5.2.2. Relative humidity

The moisture content of the final product should be 40% at most for sucuk (27). The conditions of the ripening chamber have significant effect on the moisture loosing characteristics of the product. Choice of meat type, grinding the meat and fat to desirable particle size, choice of casing type and success in stuffing are the other important keystones for controlling moisture of the product. Ripening chamber is adjusted to have 95% relative humidity at the beginning of sucuk production and finally it reaches 75% that allows the products to have moisture content of 40% at the end of the ripening process.

5.2.3. Air flow

It is essential to control the temperature, the humidity and the velocity of the air flows surrounding the sausages, in order to ensure homogeneous conditions to all products under ripening. Flow ripening chambers are used in industrial applications to maintain these controlled conditions. These chambers, which are among the most widespread used for industrial applications, have a main upward air flow that is cyclically moved along the cell floor, ensuring the required average ripening homogeneity (30).

5.3. Other factors

5.3.1. Additives

Additives used in the production of sucuk are mainly antimicrobials, antioxidants, flavouring and colouring compounds. Commercially used antimicrobials in sucuks are nitrite, nitrate, sorbic acid, benzoic acid, citric acid and their salts, while antioxidants are ascorbic acid and α-tocopherols (32).

Since the 1950s fermented sausages has contained at least 0.3–3% added carbohydrates. Sugars (glucose and occasionally lactose or saccharose) are usually included for the industrial manufacture of fermented meat products and during fermentation and ripening, lactic acid bacteria convert sugars to lactic acid (16, 33).

5.3.2. Equipment

The grinders, mixers, fillers and sub-parts of these and the other equipments have primary importance for the quality of the product. Besides
the high quality raw material and good manufacturing practices, the equipments play determinative role in the production. Producer has to pay attention to select correct equipments according to the requirements of the production. In the production of fermented meat products, grinders, mixers and fillers are the main equipments.

The cross sectional appearance is one of the most important quality characteristic for fermented meat product and this property is directly related to the plates of the grinder and the capacity and quality of the mixer. If the grinder and mixer do not ensure optimum grinding and mixing conditions, producer can not control the temperature of the product in grinding or mixing process and so he can not reach the desirable mosaic cross sectional appearance in the final product. It is not only the sectional view problem but also it may cause taste problems according to the effects in ripening period. Just as explained in the production process of the sucuk in previous section, the fat should be added as frozen to provide mosaic structure and to prevent the covering action of the fat on meat particles. Uncontrolled grinding can cause a rapid increase in the dough temperature and melting of the fat. So it affects the structure, the fermentation and so the whole process.

Similarly, the filler machine has to accommodate some expected features in the production. Air hole in the final product is the main defect that occurs mostly in filling stage. If the filler does not ensure fully vacuumed conditions while filling, air hole formation will be inevitable result that affects the product quality.

5.3.3. Casing type

Natural, collagen, fibrous or cellulose casings can be used in the production of fermented meat products. Process conditions should be arranged according to casing type in the meaning of filling, ripening temperature and time. When the air-dried small intestine from cattle is used as a natural casing in fermented product such as sucuk, bratwurst, kielbasa or Italian sausage, the casing should have enough hygienic quality.

5.3.4. Storing conditions

Storage conditions significantly affected the microflora, pH, weight loss, brine concentration, water activity and the production of certain compounds such as biogenic amines (15, 28). The colour of the product is also affected by these conditions. For example, Papadima et. Al. (28) reported that yellowness of the traditional Greek sausages has affected during storage according to the conditions of storage room (28).

According to their research on sausage poličan, Czech dry fermented meat product produced from beef and pork meat, Komprda et al. (15)
suggested that consumers should not store dry fermented sausages at room temperature. It is reported that hazardous amounts of biogenic amines can arise in a product not stored in refrigerated conditions. Legislators are recommended to establish limits for tyramine and total biogenic amines content in dry fermented meat products. Tyramine is quantitatively most important biogenic amine in fermented sausages and the presence of other amines can potentiate the negative effect of tyramine on human health (15).

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Microbiological investigation of halal butchery products and butchers’ premises

C Little, I Gillespie, J de Louvois, R Mitchell

Summary: Halal butchers’ premises were investigated as they had not been represented in a recent study of butchery products and butchers’ premises conducted by the Local Authorities Coordinating Body on Food and Trading Standards and the PHLS. This study examined 183 raw prepared meats and 212 environmental samples from 105 halal butcher premises. Only raw meats were prepared on 97 of the premises visited; and the types of meat prepared on the remaining eight premises was not specified. Four halal butchers sold cooked meats prepared elsewhere. Salmonella spp. and Campylobacter spp. were detected in 12 (7%) and 52 (28%) of the 183 raw meat products, respectively. Five raw prepared meats (3%) contained both salmonella and campylobacter. Vero cytotoxin producing Escherichia coli O157 was isolated from a raw meat product that also contained campylobacter. No cooked meat products were available for collection. The physical separation of raw and unwrapped cooked meat products in premises that prepared raw and sold cooked meats was not recorded. Apron cloths were the most heavily contaminated environmental samples examined; hygiene indicator microorganisms indicated an increased risk of cross contamination. Managers in 85 premises had received no food hygiene training and 88 premises had no hazard analysis system in place. Improvements are needed to reduce the risk of cross contamination.


Keywords:
campylobacter
environmental microbiology
Escherichia coli O157
hygiene
Islam
meat
meat products
salmonella

Introduction
Halal meat is meat that has been slaughtered according to Islamic law, in which the animal is killed by a transverse cut to the throat, rather than by the usual European method of rendering the animal unconscious by stunning and then killing it by bleeding. Genuine halal meat is sold from registered shops and is often eaten within 24 hours of slaughter. Little information is available about the microbiological quality of meat and meat products from halal butchers. They were poorly represented in a recent study of butchery products and butchers’ premises conducted by the Coordinated Food Liaison Group of the Local Authorities Coordinating Body on Food and Trading Standards (LACOTS) and the PHLS. The LACOTS/PHLS study focused on premises that handle both raw and cooked meats; halal butchers usually prepare raw meats only. Contaminated meats bought from halal butchers have been linked to outbreaks and sporadic cases of food poisoning, including an outbreak of Salmonella wangle infection in England in 1992, which affected 210 people, eight of whom were admitted to hospital (PHLS Communicable Disease Surveillance Centre (CDSC), unpublished data). The source of infection was found to be chicken prepared by a halal butcher who sold raw and cooked meats. The same butcher had been implicated in an outbreak of Salmonella wangle infection, linked to halal meats in 1982 (CDSC, unpublished data).

For comparison the protocol and parameters examined were the same as those used in the LACOTS/PHLS study. Two raw and two cooked meat products (if available) and up to five environmental samples were collected from each premises. Raw meat samples were examined for the presence of Escherichia coli O157, Salmonella spp., and Campylobacter spp., and E.coli were enumerated.
Cooked meat samples were examined for these parameters and, in addition, coliforms, *Staphylococcus aureus*, and the aerobic plate count (APC) were determined to indicate hygiene and levels of contamination. The presence of *E. coli* O157 and the enumeration of coliforms, *E. coli*, and *S. aureus* in environmental samples were used to indicate hygiene and levels of contamination.

**Methods**

**Sample collection**

This study took place in April and May 1998 and involved local authorities and laboratories in England and Scotland. A standardised protocol and reporting system was used. All samples were collected by staff from local environmental health departments in accordance with the Food Safety Act 1990, Code of Practice No 7. Butchery products (approximately 150g) included raw meat, minced or diced, prepared on the premises and, if available, cooked meat sliced on the premises. Environmental areas sampled on the premises included chopping surfaces for raw and for cooked meats, weighing balances used for raw and/or cooked meats, blades used to slice cooked meat, and apron cloths (used for wiping surfaces, equipment, and utensils). Surfaces were sampled and the samples were processed as described previously. All environmental sampling materials were supplied by Technical Services Consultants Ltd (Heywood, United Kingdom).

Observations and responses to enquiries were recorded on a standard form that included details on the Inspection Rating Category of the premises, Confidence in Management score (*Food Safety Act 1990, Code of Practice No. 9*), manufacturing activities on the premises, documentation of a hazard analysis system, food hygiene training received by staff, and - if applicable - the degree of physical separation of raw meat and unwrapped cooked meat products and other ready-to-eat foods.

**Sample examination**

Samples were examined as described previously. All isolates of salmonella, campylobacter, and *E. coli* O157 were sent to the PHLS Laboratory of Enteric Pathogens (LEP) for further characterisation.

**Results**

Samples were submitted by 20 local authority food liaison groups (17 in England and three in Scotland), representing 32 local authorities, for examination by 12 laboratories (see Acknowledgements). A total of 395 butchery products and environmental samples were collected and examined from 105 halal butchers’ premises.

**Microorganisms isolated from halal butchery products and premises**

**Raw meat products**

A total of 183 raw meat products were examined; 89 (49%) raw minced meat, 63 (34%) raw chopped/diced meat, 15 (8%) other raw meat products (whole chickens, chicken thighs and/or breasts, giblets), and 16 (9%) unspecified raw meats. Most of the raw meat samples were of lamb (70; 38%), mutton (56; 31%), or chicken (42; 23%). Raw meat products containing salmonella or campylobacter were collected from 40 of the 105 butchers’ premises. Five raw meat products (chicken, mutton) obtained from four different premises contained both salmonella and campylobacter. *E. coli* O157 vero cytotoxin (VT) 2 phage type 2 (PT2) was detected in one sample of raw minced lamb that also contained campylobacter. *E. coli* O156 VT 1 was detected in one raw mutton product (table 1).

Salmonellas were detected in 12 raw meat products (table 1) – raw lamb/mutton (6), raw chicken (5), and raw beef (1). Five different serotypes were isolated from the 12 positive samples. The commonest isolates were *S. enteritidis* phage type (PT) 4 (4) and *S. heidelberg* (4). One raw chicken sample contained *S. hadar* and *S. virchow*. *S. typhimurium* definitive phage type (DT) 104 isolated from a raw mutton sample was the current epidemic strain, resistant to ampicillin, chloramphenicol, streptomycin, sulphonamides, and tetracyclines (R-type: ACSSuT). Two *S. virchow* PT47 isolates were resistant to ampicillin, chloramphenicol, sulphonamides, tetracyclines, trimethoprim, and furazolidone (R-type: ACSSuTTmFu).

Campylobacter spp. were detected in 52 raw meat products (table 1), most in raw chicken (50%; 21/42) or raw lamb/mutton (23%; 9/26). Thirty of the 52

---

**TABLE 1** Incidence of foodborne pathogens in raw meat products from halal butchers’ premises

<table>
<thead>
<tr>
<th>Raw meat type</th>
<th>Product number</th>
<th>Salmonella sp (%)</th>
<th>Campylobacter sp (%)</th>
<th>Verocytotoxin-producing <em>Escherichia coli</em> O157 (%)</th>
<th>non-O157 (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mince</td>
<td>89</td>
<td>4 (4)</td>
<td>20 (22)</td>
<td>1</td>
<td>–</td>
</tr>
<tr>
<td>Chopped/diced</td>
<td>63</td>
<td>6 (10)</td>
<td>21 (33)</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Other*</td>
<td>15</td>
<td>2 (13)</td>
<td>8 (53)</td>
<td>–</td>
<td>–</td>
</tr>
<tr>
<td>Not specified</td>
<td>16</td>
<td>–</td>
<td>3 (19)</td>
<td>–</td>
<td>1 (6)</td>
</tr>
<tr>
<td>Total</td>
<td>183</td>
<td>12 (7)</td>
<td>52 (28)</td>
<td>1 (1)</td>
<td>1 (1)</td>
</tr>
</tbody>
</table>

* whole chickens, chicken thighs and/or breasts, giblets
† *E. coli* O157 VT2 PT2
‡ *E. coli* O156 VT1
campylobacters isolated from raw meat samples were further characterised, 21 of which were C. jejuni and nine C. coli. A wider range of serotypes of C. jejuni were identified than of C. coli (table 2). About a third (11/30) were untypable, however, of which there were six different phage types (table 2).

**Cooked meat products**

None of the 105 premises visited prepared cooked meats on the premises. Four sold cooked meats on the premises, but no samples were available at the time of collection.

**Environmental samples**

A total of 212 environmental samples were examined – 103 from raw meat chopping surfaces, 50 from weighing balances used for raw meats, 29 apron cloths, 14 from bandsaws (used exclusively for cutting raw meats), seven from other raw meat surfaces (display cabinet, refrigerator shelf, mincer, cloth for skinning chickens), four from raw meat slicer blades, three from cooked meat slicer blades, and two from cooked meat chopping surfaces.

None of the 212 environmental samples yielded E. coli O157. Apron cloths were the most heavily contaminated with coliforms, E. coli, and S. aureus, indicating a reservoir of bacteria and a cross contamination hazard. S. aureus was found in two of the 29 apron cloth samples, with counts ≥ 10⁴ cfu/mL cloth eluent. Coliforms and E. coli were present at counts ≥ 10⁴ cfu/mL cloth eluent in nine and seven apron cloths samples, respectively. Only one other sample, from a raw meat chopping surface, yielded a high level of coliforms (≥ 10⁸ cfu/cm²) (table 3).

**Halal butchers’ premises in relation to food hygiene practice and the Pennington Group report recommendations**

The survey found the following:

- Premises were classified according to the Food Safety Act 1990 (Code of Practice No.9). Premises that pose a greater risk to the consumer are inspected more frequently than those with a lesser risk. Six of the 105 premises sampled had an inspection rating category A (minimum frequency of inspection every six months), 27 B (every year), 52 C (every 18 months), three D (every two years), zero E (every three years), one F (every five years), and no category was recorded for 16.

- The food hygiene performance of the management was scored using the Confidence in Management/Control Systems scoring system described in the Food Safety Act 1990 (Code of Practice 9). One of the premises achieved a score of zero, which indicates a high degree of confidence, one scored five (moderate confidence), 47 scored ten (some confidence), 24 scored 20 (little confidence), and nine scored 30 (no confidence). At 23 premises the sampling officer did not record the scores.

- Ninety-seven of the premises did not require approval under the Meat Product (Hygiene) Regulations 1994 as they were used for the preparation of raw meats only. The approval status of the eight remaining premises was not recorded as the report did not specify whether raw meat products only or both raw and cooked meat products were prepared.

- Five out of the 105 premises, as well as preparing and selling raw meats, sold cooked meats or other ready-to-eat foods (salami, cheese) supplied from elsewhere. Additional comments from sampling officers indicated that seven premises also sold unwrapped fresh produce, herbs, and spices. The degree of physical separation of raw meat and unwrapped cooked meat products, the use of display cabinets, refrigerators, equipment, utensils, staff, and method of serving in these premises was not recorded.

- Meats stored on the counter or in displays were kept at temperatures of ≤ 8°C in 66 premises, 8°C in 12, and in 27 the temperature was not recorded.

- Managers in 85 premises had not received food hygiene training; in 19 he/she had received training, and in one this information was not given. Fourteen of those with training had attended a basic six hour course in food hygiene training; the type of training for the remaining five was not specified. The Pennington Group Report recommends that all staff of premises handling both raw and cooked meats should have received basic training in food hygiene and that at least one person in each butchers’ business should be trained to intermediate level. Only one of the managers from the four premises that sold both raw and cooked meats had received food hygiene training (basic six hour course).

- The Pennington Group report recommends documentation of hazard analysis as a requirement of licensing arrangements pending hazard analysis and critical control points (HACCP) implementation. Three of the 105 premises had a documented hazard analysis system in place, and

### TABLE 2 Sero- and phage types of *Campylobacter* isolated from raw meat products

<table>
<thead>
<tr>
<th>Campylobacter sp</th>
<th>Serotype</th>
<th>Phage type (number of isolates)</th>
</tr>
</thead>
<tbody>
<tr>
<td>C. jejuni</td>
<td>HS2</td>
<td>1 (2)</td>
</tr>
<tr>
<td></td>
<td>HS5</td>
<td>1 (1), 44 (1)</td>
</tr>
<tr>
<td></td>
<td>HS11</td>
<td>1 (2), 18 (1), 39 (1)</td>
</tr>
<tr>
<td></td>
<td>HS18</td>
<td>2 (2)</td>
</tr>
<tr>
<td></td>
<td>HS45</td>
<td>1 (1)</td>
</tr>
<tr>
<td></td>
<td>HS50</td>
<td>35 (1)</td>
</tr>
<tr>
<td></td>
<td>HS60</td>
<td>2 (1)</td>
</tr>
<tr>
<td></td>
<td>HS rough</td>
<td>35 (1)</td>
</tr>
<tr>
<td></td>
<td>Untypable</td>
<td>6 (1), 33 (2), 34 (1), 35 (1), 44 (2)</td>
</tr>
<tr>
<td>C. coli</td>
<td>HS9</td>
<td>44 (1)</td>
</tr>
<tr>
<td></td>
<td>HS24</td>
<td>1 (1)</td>
</tr>
<tr>
<td></td>
<td>HS56</td>
<td>7 (1), 44 (2)</td>
</tr>
</tbody>
</table>

* RDNC, reacted with the phage set but did not conform to a recognised phage type.
a further 10 had a hazard analysis system that was undocumented. Eighty-eight premises had no system for hazard analysis, and for four this information was not given. One of the four premises that sold raw and cooked meats had a documented hazard analysis system.

- Adequate safety procedures were in place to ensure food safety in 68 of the 105 premises, as judged by the sampling officer, in 23 premises they were not, and in 14 this was not recorded. Significantly more raw meat products collected from premises judged not to have adequate safety procedures in place were contaminated with *Campylobacter* spp. (14/33) than from those with safety procedures in place (33/126) (p<0.01).

### Discussion

Campylobacters and salmonellas are the commonest pathogens implicated in food poisoning in the United Kingdom. Vero cytotoxin producing *E. coli* O157 (VTEC O157) is less common but has emerged as an important pathogen because of its virulence and low infectious dose. All three pathogens are known to colonise the intestines of farm animals and may contaminate meat of cattle, sheep, and poultry at the time of slaughter. Pathogenic microorganisms are therefore inherent constituents of raw meat and its products. Subsequent handling and processing of raw meat products such as comminution may spread contamination. Over a quarter of raw meat products collected from halal butchers premises were

### TABLE 3 Microbiological results of 212 environmental samples from halal butchers’ premises

<table>
<thead>
<tr>
<th></th>
<th>Not detected</th>
<th>&lt;10&lt;sup&gt;2&lt;/sup&gt;</th>
<th>10&lt;sup&gt;2&lt;/sup&gt;-&lt;10&lt;sup&gt;3&lt;/sup&gt;</th>
<th>10&lt;sup&gt;3&lt;/sup&gt;-&lt;10&lt;sup&gt;4&lt;/sup&gt;</th>
<th>10&lt;sup&gt;4&lt;/sup&gt;-&lt;10&lt;sup&gt;5&lt;/sup&gt;</th>
<th>10&lt;sup&gt;5&lt;/sup&gt;-&lt;10&lt;sup&gt;6&lt;/sup&gt;</th>
<th>10&lt;sup&gt;6&lt;/sup&gt;-&lt;10&lt;sup&gt;7&lt;/sup&gt;</th>
<th>≥10&lt;sup&gt;7&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw meat chopping surfaces (n=103)&lt;sup&gt;a&lt;/sup&gt;</td>
<td>S. aureus</td>
<td>81&lt;sup&gt;†&lt;/sup&gt;</td>
<td>20</td>
<td>2</td>
<td>6</td>
<td>1</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Coliforms</td>
<td>58&lt;sup&gt;†&lt;/sup&gt;</td>
<td>30</td>
<td>8</td>
<td>6</td>
<td>1</td>
<td>5</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em></td>
<td>65&lt;sup&gt;†&lt;/sup&gt;</td>
<td>26</td>
<td>7</td>
<td>5</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em> O157</td>
<td>103</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Raw meat slicer blade/bandsaw (n=18)&lt;sup&gt;b&lt;/sup&gt;</td>
<td>S. aureus</td>
<td>18&lt;sup&gt;†&lt;/sup&gt;</td>
<td>7</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Coliforms</td>
<td>13&lt;sup&gt;†&lt;/sup&gt;</td>
<td>12</td>
<td>5</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em></td>
<td>16&lt;sup&gt;†&lt;/sup&gt;</td>
<td>7</td>
<td>5</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em> O157</td>
<td>18</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Weighing balance used for raw meats (n=50)&lt;sup&gt;c&lt;/sup&gt;</td>
<td>S. aureus</td>
<td>42&lt;sup&gt;†&lt;/sup&gt;</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Coliforms</td>
<td>33&lt;sup&gt;†&lt;/sup&gt;</td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em></td>
<td>38&lt;sup&gt;†&lt;/sup&gt;</td>
<td>7</td>
<td>5</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em> O157</td>
<td>50</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Other raw meat surfaces (display cabinet, refrigerator shelf, mincer, cloth for skinning chickens, n=6)&lt;sup&gt;d&lt;/sup&gt;</td>
<td>S. aureus</td>
<td>6&lt;sup&gt;†&lt;/sup&gt;</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Coliforms</td>
<td>2&lt;sup&gt;†&lt;/sup&gt;</td>
<td>1</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em></td>
<td>6&lt;sup&gt;†&lt;/sup&gt;</td>
<td>7</td>
<td>5</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em> O157</td>
<td>7</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Cooked meat chopping surface (n=2)&lt;sup&gt;e&lt;/sup&gt;</td>
<td>S. aureus</td>
<td>1&lt;sup&gt;†&lt;/sup&gt;</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Coliforms</td>
<td>1&lt;sup&gt;†&lt;/sup&gt;</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em></td>
<td>1&lt;sup&gt;†&lt;/sup&gt;</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em> O157</td>
<td>2</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Cooked meat slicer blade (n=3)&lt;sup&gt;f&lt;/sup&gt;</td>
<td>S. aureus</td>
<td>3&lt;sup&gt;†&lt;/sup&gt;</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Coliforms</td>
<td>2&lt;sup&gt;†&lt;/sup&gt;</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
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<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em></td>
<td>2&lt;sup&gt;†&lt;/sup&gt;</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em> O157</td>
<td>3</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td>Apron cloth (n=29)&lt;sup&gt;g&lt;/sup&gt;</td>
<td>S. aureus</td>
<td>20&lt;sup&gt;†&lt;/sup&gt;</td>
<td>3</td>
<td>2</td>
<td>2</td>
<td>2</td>
<td>1</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td>Coliforms</td>
<td>4&lt;sup&gt;†&lt;/sup&gt;</td>
<td>5</td>
<td>3</td>
<td>8</td>
<td>2</td>
<td>4</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em></td>
<td>6&lt;sup&gt;†&lt;/sup&gt;</td>
<td>6</td>
<td>5</td>
<td>5</td>
<td>2</td>
<td>3</td>
<td>1</td>
</tr>
<tr>
<td></td>
<td><em>E. coli</em> O157</td>
<td>29</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
<td>1</td>
</tr>
</tbody>
</table>

- a: cfu/cm<sup>2</sup>
- b: cfu/swab
- c: cfu/mL
- †: lower limit of detection 20 cfu/cm<sup>2</sup>, swab or mL
- ‡: lower limit of detection 3 cfu/cm<sup>2</sup>, swab or mL

Notes:
- A hazard analysis system was not documented for 10 premises.
- Eighty-eight premises had no system for hazard analysis.
- One of the four premises that sold raw and cooked meats had a documented hazard analysis system.
contaminated with campylobacter and 7% with salmonella. One sample of raw meat yielded VTEC O157 and another VTEC O156. Not all VTEC isolates are enterohaemorrhagic, and many VTEC strains isolated from animal species - for example, E. coli O156 – appear not to cause human disease\(^7\),\(^8\). The percentages of raw meat samples contaminated with salmonella (7%), campylobacter (28%), and VTEC O157 (0.6%) reported here are considerably higher than in the previous study, in which halal butchers’ premises were not represented (4%, 0.6%, 0.1%, respectively)\(^7\). The large numbers of campylobacters and salmonellas in these raw prepared meats could reflect contaminated supply, cross contamination, and/or poor hygiene practices. Legal action brought against slaughterhouse businesses is published monthly by the Ministry of Agriculture, Fisheries and Food (MAFF)\(^9\).

The Pennington Group report recommends that hazard analysis procedures should be documented, staff trained in food hygiene, and raw meat and unwrapped cooked meat products physically separated and wherever possible handled by separate staff\(^7\). Most of the halal butchers visited during this study prepared and sold raw meat only, but some also sold cooked meats and/or unwrapped fresh produce, herbs, and spices. Two thirds of the premises had adequate temperature controls for displaying meats. Only one of the five premises that sold both raw and cooked meats had a documented hazard analysis system, and its manager was the only one to have received formal training in food hygiene. Hazard analysis and food hygiene training are areas that require improvement to reduce the risk of cross contamination with foodborne pathogens in halal butchers’ premises. Overall, halal butchers’ premises were classified as posing a lower risk to the consumer than premises in the previous study\(^7\), as most handled raw meats only, and were therefore inspected less frequently. Confidence in management scores for halal butchers’ premises were less favourable than in the previous study, indicating the need for more frequent inspections\(^8\).

The results from this study suggest that raw meat products from halal butchers’ premises could be a potential source of human infection. It is therefore vital that meat producers, slaughterhouses, butchers’, retails, and consumers apply the basic rules of hygiene to prevent these (like other raw meats) from contaminating other foods, minimise bacterial growth (by proper refrigeration), and ensure that any bacterial pathogens present are destroyed (by thorough cooking) before the meat is eaten.

Acknowledgements
We thank the staff in the environmental health departments in England and Scotland who collected samples for this study and all the staff in public health laboratories at Epsom, Ipswich, Middlesbrough, Newcastle, Portsmouth, Preston, Shrewsbury/Telford, Southampton, and Stoke, the PHLS London Food, Water and Environmental Unit, Dundee City Council Scientific Services, and Glasgow Scientific Services, who performed the microbiological examinations. We thank LEP for characterising the strains of salmonella and campylobacter, and Lilian Hucklesby for entering the data.

References
MEAT PROCESSING HYGIENE

Principles of meat processing hygiene and regulatory practices (incl. GHP and HACCP)

Meat processing hygiene is part of Quality Management (QM) of meat plants and refers to the hygienic measures to be taken during the various processing steps in the manufacture of meat products. Regulatory authorities usually provide the compulsory national framework for food/meat hygiene programmes through laws and regulations and monitor the implementation of such laws. At the meat industry level, it is the primary responsibility of individual enterprises to develop and apply efficient meat hygiene programmes specifically adapted to their relevant range of production.

Operations in meat processing plants comprise the manufacture of value-added meat products from primary products of meat origin and non-meat origin. There are three principles of meat hygiene, which are crucial for meat processing operations.

- Prevent microbial contamination of raw materials, intermediate (semi-manufactured) goods and final products during meat product manufacture through absolute cleanliness of tools, working tables, machines as well as hands and outfits of personnel.
- Minimize microbial growth in raw materials, semi-manufactured¹ goods and final products² by storing them at a low temperature.
- Reduce or eliminate³ microbial contamination by applying heat treatment at the final processing stage for extension of shelf life of products (except dried and fermented final products, which are shelf-stable through low a_w and pH)⁴

¹) Semi-manufactured goods must be stored refrigerated during production breaks and resting periods. Processing steps, such as cutting, grinding, comminuting, mixing, filling, smoking and cooking take place under climatized conditions or ambient temperatures. Ambient temperatures are hygienically acceptable as long as these processing phases are of short duration or when product temperatures are rising as a result of the processing.
²) In some final products low pH or low a_w also serve to contain microbial growth in combination with or in replacement of refrigeration.
³) Elimination of contamination only in fully sterilized (canned) products.
⁴) For some food products useful, but in the meat industry not commonly used, are other methods for food preservation, such as irradiation and high hydrostatic pressure treatment.
The above three principles guide meat hygiene programmes in the further processing of meat (see also Fig. 452). However, meat processing hygiene is more complex. In particular, the hygienic treatment of meat before reaching the processing stage is of utmost importance for the processing quality of the meat. Failures in slaughter hygiene, meat cutting and meat handling/transportation and in the hygiene of by-products and additives will all contribute to quality losses and deterioration of the final processed meat products.

Highly contaminated raw meat is unsuitable for further processing. Final products made from hygienically deficient raw meat materials are unattractive in colour, tasteless or untypical in taste with reduced shelf life due to heavy microbial loads (see page 353, 356). Moreover, there is also the risk of presence of food poisoning microorganisms, which can pose a considerable public health hazard (see page 357).

In the light of growing consumer consciousness as well as regionalization and globalization in trade, quality conscious meat plants need internal quality control/quality management schemes not only for the final
products but also for the raw materials and the various processing steps.

Such Quality Management Schemes (QM) have technical and hygienic components. Technical aspects encompass product composition, processing technologies, packaging, storage and distribution. Details on the manufacturing practice for each individual group of meat products are included in the chapters on processing technology (see page 103 - 212). For the sanitary quality and safety related to meat processing, two useful schemes¹ can be applied known as

- Good Hygienic Practices (GHP) and
- Hazard Analysis and Critical Control Point (HACCP) Scheme.

Both schemes are not verbally laid down in codes ready to be used for the various purposes in the meat sector although some generic examples can be accessed in handbooks or via internet. Factory and production specific versions need to be established and compiled by taking into account official laws and regulations as well as recommended codes of practice.

**Good Hygienic Practices (GHP)**

Good Hygienic Practices/GHP follows general hygienic rules and applies recognized hygienic principles² as well as laws and regulations issued by the competent authorities, referring to meat and meat products, equipment, premises and personnel. GHP schemes are not factory specific, they apply to all types of meat plants. They are intended to establish and maintain acceptable hygienic standards in relevant meat operations. There is more emphasis on slaughter hygiene in GHP schemes for slaughterhouses and more emphasis on meat processing hygiene in GHP schemes for meat products manufacturing enterprises. However in principle, GHP schemes remain interchangeable for similar types of meat plants.

¹ There are a number of additional specialized norms and standards for auditing purposes in meat/food industries in use, some of them with regional scope and mostly with links to GHP and HACCP.

² The FAO/WHO Codex Alimentarius Commission has issued a new CODE OF HYGIENIC PRACTICE FOR MEAT in 2005 (CAC/RCP 58-2005). In addition to relevant laws and regulations by the competent authorities, this recommended international Code of Practice provides a suitable platform for the development of official or individual meat hygiene programmes.
GHP for **meat processing plants** refers principally to:

- Appropriate functional plant layout and sanitary design of equipment
- Raw materials that meet hygiene quality standards
- Processing methods that allow safe handling of food
- Appropriate waste and pest control measures
- Appropriate sanitation procedures (cleaning and disinfection)
- Compliance with potable water criteria
- Functional cold chain
- Regular examination of health and personal hygiene of staff
- Regular training of staff on hygiene requirements

**Hazard Analysis and Critical Control Point Scheme (HACCP)**

HACCP are **factory** and **product specific** strictly sanitary control schemes that shall prevent, detect, control and/or reduce to save levels **accidentally occurring hazards** to consumers’ health. Despite GHP in place, accidental hazards cannot be ruled out and may occur at any processing step of the individual meat product. Specifically for **meat processing plants**, such hazards may be provoked by failures such as:

- batches of incoming raw meat materials with abnormal tissues or heavy contamination,
- breakdowns in refrigeration,
- failure in cooking/sterilization operations,
- abnormal pH or aw in raw or finished products,
- errors in levels of application of curing salts and other additives,
- technical problems in sealing of vacuum packages or cans with the risk of recontamination.

HACCP schemes serve as additional **alarm systems** in the interest of consumer protection to prevent such problems occurring.

The revolutionary idea of HACCP is to implement control measures that focus on prevention rather than relying on end-product-testing. All relevant possible hazards in the entire production chain, from primary production to consumption of each individual product, must be identified and measures taken for their prevention. In case potential hazards should occur, they can be **detected, contained or eliminated at any stage**.

Plant personnel have a key role to play and must be trained in hazard detection and elimination. For practical purposes, those possible hazards may be listed on **specific templates** for confirmation of presence or absence during routine controls. **Specific control mechanisms**, in the
first place of physical, chemical and visual nature (temperature, pH, visual check etc.), are installed at selected control points to detect such potential hazards. These control mechanisms are designed to deliver most results almost instantly and allow immediate intervention during the processing phase of food/meat products.

The need for immediate action within HACCP systems excludes microbiological control (of raw materials, semi-fabricated products, tools, equipment, and premises) as a directly applicable control measure. Microbiological control takes hours or days to obtain the results, which does not allow corrective interventions during the usually short manufacturing period. However, this does not mean that microbiological control is worthless for HACCP. Routine microbiological control carried out within the framework of GHP is an extreme helpful tool also for HACCP as its results will demonstrate the efficiency of the HACCP-system. Hygienically acceptable microbiological test results are an indicator of the proper functioning of the meat plant’s HACCP scheme.

HACCP\textsuperscript{1} is not a scheme for the assessment and improvement of the general hygienic status of a meat plant. HACCP is not designed to further raise hygienic standards. Excellent conditions as applicable for GHP-conform plants must already be in place. GHP is a prerequisite requirement for the introduction of HACCP.

The misconception still exists that HACCP is intended to raise levels of general hygiene in meat plants with low hygienic standard. HACCP is not workable where plant layout/structure, equipment and/or processing methods do not comply with good hygienic standards.

One important point to distinguish HACCP from GHP is that GHP describes process requirements and practices incl. personal hygiene of staff to ensure safety of food. The individual product is not specifically targeted. Unlike GHP, HACCP always focuses on the individual product. As technologies vary from product to product, it is obvious that separate HACCP approaches are required for each category of products.

\textsuperscript{1) More detailed information on HACCP see boxes on pages 344-348.
HAZARD ANALYSIS CRITICAL CONTROL POINT (HACCP)

What is HACCP?

Internal sanitary related control and monitoring system in food plants with the aim of preventing/minimizing or eliminating health hazards to consumers. HACCP identifies, evaluates and controls hazards, which are significant for food safety. The characteristics of HACCP are:

- Potential for immediate prevention measures before or during production to counteract suspected or emerging health risks
- Exclusively aimed at health risks to consumers

Food plant internal control procedures based on HACCP principles have become an obligation worldwide in many countries with advanced food industries. HACCP procedures are imposed on relevant food plants by the competent authorities, whose task is to assess and evaluate the correct application and conduct of HACCP. The food plants themselves are responsible for the proper implementation of HACCP, such as monitoring of sensory, physical and chemical parameters during production and immediate intervention in case of emerging health risks and recording of results.

Requirements for introduction of HACCP schemes are yet different from region to region. In a number of countries (e.g. EU, US) meat plants in general have to comply with HACCP, whereby for smaller plants or such specializing in limited activities or products, simplifications or exceptions exist. In some other parts of the world, HACCP schemes are not yet commonly introduced. However, it can be anticipated that such plants involved in regional or global distribution of food will also be obliged to comply with HACCP principles.

Basic elements of HACCP in meat processing plants

- Every single meat product with product specific technology requires a specifically designed individual HACCP scheme.
- As a precondition for implementing HACCP concepts, hazard analysis and risk assessment referring to meat plant specific processing methods or products, have to be carried out.
- Critical control points (CCPs) have to be identified, critical limits be established and monitoring systems properly implemented.

The HACCP scheme is subdivided into seven consecutive steps (“principles”). Through these seven HACCP principles a practical approach is provided to identify potential significant hazards to consumers’ health and to take relevant corrective actions:
1. Hazard analysis and risk assessment

The first principles requires initially the **exact description** of the products to be fabricated, including product composition, texture/structure, processing details (such as degree of comminuting, additives, filling, heat treatments), packaging and if applicable chemical and microbiological criteria.

Once the characteristics of each product are detailed, potential hazards to consumers’ health during processing are identified. Hereunder, a summary listings of hazards are given, from where those hazards likely to be associated with the fabrication of a specific meat product can be identified.

**Examples for hazards in meat processing**

*Biological hazards*: Parasites (causing zoonotic diseases), bacteria (causing food poisoning/food borne infections and intoxications), moulds (mycotoxins causing food borne intoxications), viruses (causing food borne infections) (see page 357)

*Physical hazards*: Rests of unwanted materials (glass, bone fragments, animal teeth/in case of processing head meat, metal fragments such as sausage clips, broken knife blades, needles, plastics, stones)

*Chemical hazards*: Contaminants (heavy metals, PCB’s, chemical solvents, cleaning and disinfection compounds)  
Residues (veterinary drugs, feed additives, pesticides)  
Food additives with risk of overdoses (nitrate/nitrite, chemical preservatives)

2. Identification of Critical Control Points (CCP)

A CCP is defined as any point or procedure in a specific food system, where loss of control may result in an unacceptable health risk. CCPs can be located at any point along the production line of a specific meat product, where biological, physical and chemical hazards may occur and where such risks can be controlled and/or eliminated. CCPs should only be established, where firm methods for control and monitoring can be applied.

CCPs must be used only for purposes of product safety. They should not be confused with control points that do **not** control safety and where loss of control does **not** lead to unacceptable health risks, e.g. reduced or strong water binding capacity of meat, knives of grinders or choppers with reduced cutting capability, mechanical problems in portioning sausages or can fillings etc. Moreover, issues of meat plant hygiene routinely covered by GHP and which are not product specific, are normally **not** CCPs. Such examples are:

- **Potable water outlets**,  
- **Hot water container** for tool disinfection (“sanitizers”),  
- **Cleaning** and **disinfection** equipment, chemicals and methods.  
- **Sanitation measures** (e.g. periodic cleaning and disinfection of meat cutting boards)  
- **Personal hygiene**
Specific preventive measures to avoid cross contamination (e.g. plant internal transports of raw materials and finished products must not cross each other)

Specific food handling procedures (e.g. meat containers must not directly be placed on the floor, but on stands, pallets etc.)

Suggested control points directly related to meat processing and therefore suited for the establishment of CCPs are:

- unloading bay for raw materials (meat and non-meat ingredients),
- cold storage rooms,
- meat cutting and preparation facilities,
- facility for handling non-meat additives,
- meat comminuting units (grinders, bowl choppers etc.),
- filling equipment and casings,
- heat treatment facilities (smokehouses, cooking vats, autoclaves),
- packaging equipment and materials (including canning),
- cold store for final products,

It is up to the individual meat processing plant to decide, at which points in the processing line CCPs should be established. This will vary from meat plant to meat plant, depending on plant lay-out equipment, type of products and also on previously experienced accidentally occurred shortcomings.

3. Establishment of Critical Limits for each CCP

Critical limits correspond to the extreme (highest and lowest) values acceptable from the point of view of product safety. This does not always imply that a numerical value has to be fixed. Monitoring may also be based on visual observation, e.g. dirt/faecal contamination of meat, changes to untypical colour, changes in product structure or texture. Besides such sensory parameters, numerical critical limits must be specified for each objective control measure at each CCP. Criteria often used include temperature, time, moisture level, pH, and water activity.

Examples

Visual check of damage to packaged incoming raw materials (rejection in case of severely damaged packages of meat materials or additives)

Visual check of contamination of raw materials (meat, fat). Discolouration (rejection of meat or fat in severe cases), meat potentially contaminated with food poisoning agents (e.g. minimal dirt contamination to be trimmed off, critical dirt or fecal contamination leads to rejection of the meat)

Temperature control of meat derived from slaughterhouses/cutting plants (e.g. \( \leq +4^\circ C \))

pH of incoming meat (e.g. \(< 6.0 \) for pork, \(< 5.7 \) for beef)

Visual check during meat cutting and grading (e.g. to separate and discard unsuitable meat tissues such as those containing parasites, abscesses, etc.)
Moisture content expressed as \( a_w \) (refers mainly to dry fermented products which should not be packaged or marketed if moisture content keeps above a certain level)

Additives (some products require a certain salt level for better stability in hot environments; nitrite levels should be high enough to inhibit bacterial growth but below toxic levels; the same applies to chemical preservatives)

Control of pasteurization parameters (ensure sufficient cooking, measured as core temperatures in products, e.g. 74°C)

Control of sterilization temperature and time for canned products (e.g. ensure that desired F-values are reached, e.g. F value 4 in fully sterilized canned products)

Visual appearance and texture of final products (greenish discolouration and slimy surfaces as signs of microbial growth, mould growth on surfaces of dried sausages)

4. Establishment of a monitoring system for each CCP

Monitoring is the regular/periodic measurement or observation at a CCP to determine whether a critical limit or target level has been met. The monitoring procedure must be able to detect loss of control at the CCP. Monitoring at CCPs should deliver results rapidly in order to enable corrective action during processing. Lengthy analytical testing is not practicable in the context. Hence most of the testing for critical limits listed in (3) is visual, physical and to some extent chemical. The slower microbiological testing (see also page 331) does not allow immediate corrective action.

Physical and chemical pattern to be instantly measured or monitored in meat processing lines include:

- **Temperature**
- **Time** limits see No. 3
- **pH**
- **Moisture**

5. Establishment of corrective actions

Corrective actions are those actions to be taken either when monitoring results show that

- a CCP has deviated from its specified critical limit or target level or
- when monitoring results indicate a trend towards loss of control

Action taken must reduce to safe level or eliminate the actual or potential hazard identified.

Corrective actions are for example

- **Reject** incoming meat with too high internal temperatures
- **Adjust** temperature for refrigerated storage and transport of meat
- **Remove** with clean knives minimal visual contamination of meat surface, **reject** heavily contaminated meat
- **Adjust** cooking and sterilization parameters (temperature/time)
Reject meat with too high pH
Adjust quantity of curing substances (level of nitrite, nitrite curing salt should contain 99.5% common salt and 0.5% nitrite)
In case of dry fermented products: If $a_w$ of processed products is too high, stop packaging in water vapour impermeable packages

Products with suspected hygienic deficiencies have to be separated from other products. Additional treatments may have to be applied, e.g. additional heat treatment in case of undercooking. Final judgement (if fit or unfit for consumption) has to be made by responsible, competent persons. Interventions at CCPs are carried out based on instant observation of hygienic failures/shortcomings. Corrective actions should be documented in the HACCP written records.

6. Establishment of verification procedures

Procedures are needed to ensure that the HACCP system is working correctly. Particular attention must be given to the monitoring frequency, which may be daily or several times a day or more frequently. Checks on the persons doing the monitoring should be done regularly as well as calibration of instruments used.

Established critical limits can be revalidated (changed) in the light of new developments. The system as a whole for individual products has to be reviewed in case of introducing changes in the processing technology such as changes in raw materials, product composition, processing equipment or packaging systems.

Test results derived from GHP routine quality control, in particular microbiological analysis, are valuable supplementary information within the HACCP system, support the verification process and prove the practicability of HACCP.

7. Establishment of documents and records

Documents and records must be produced commensurate with the nature and size of the food business to demonstrate the application of principles 1-6. These documents serve for the competent authorities to evaluate the efficacy of the HACCP procedure carried out at the plant. Records also help to trace causes of problems that were encountered during past production.

This documentation includes amongst others

- Certification on receipt of raw meat materials and non-meat ingredients documenting supplier compliance with processor’s specifications
- CCP determinations (for each product)
- Critical limits set and results achieved for each CCP (including possible deviations from critical limits and corrective actions)
- Modifications introduced to the system in the light of changes of technology or other developments
HACCP in small meat processing plants

The rather complex HACCP approach including identification of critical control points and measurement and interpretation of test results, demonstrates the difficulties in introducing HACCP schemes in small food or meat processing enterprises. Comprehensive test systems would require a multidisciplinary approach, as well as knowledge of microbiological, chemical and physical hazards, technical processes and operation of equipment. This is available in large industries but generally not in small- to medium-scale enterprises. Flexibility should be given in these situations for simplified approaches, if HACCP schemes are to be introduced in small food businesses. Competent authorities tend to accept these views. In plants dealing with limited numbers of products or technologies, these simplified approaches can even go so far as to use GHP schemes instead of HACCP. It is obvious that in such cases GHP approaches may be more practical and less cost-intensive than HACCP.

Two examples for preparation of HACCP plans (see page 350, 351)

These are summary plans, which need to be expanded in more detail if adapted for relevant meat plants, depending on the plant layout, equipment and processing technology. Potential hazards, which are indicated as physical, chemical and biological, would have to be specified in detail according to the listings given on page 344. The majority of the potential hazards are “biological”, which mostly refer to microbiological risks. This corresponds with the aim of HACCP, which is prevention of health hazards to consumers. Health hazards through food are mostly caused by microbiological activity, which can be prevented if properly controlled.

The first example (cured cooked ham) is a product which is heat treated during manufacture and hence was stabilized microbiologically to a certain extent, but requires refrigerated storage. The second example refers to a meat product, which does not undergo heat treatment during processing (fresh frozen beef burger) and therefore remains particularly sensitive from the hygienic point of view.

Due to the nature of the two products, periodic microbiological tests are recommended in the framework of GHP. Periodic microbiological testing is particularly important for the product “Fresh Frozen Beef Burgers” to be marketed raw. Microbiological test results can be incorporated in HACCP. They are not a means for immediate intervention in ongoing productions (microbiological tests take too long to use their results for immediate action), but rather in the verification procedure, which serves to prove whether the HACCP system is working. Microbiological results
are a means to confirm the efficiency of the meat plant internal HACCP system, when it can be proved that the established limits were not exceeded.

The Critical Control Points (CCPs) indicated are examples for the establishment of CCPs. It is up to the processing plant to increase or decrease their number according to the plant specific risk assessment.

Table 17: HACCP plan for Cured Cooked Ham

<table>
<thead>
<tr>
<th>CCP</th>
<th>Biological</th>
<th>Physical, chemical, biological</th>
<th>Physical, chemical, biological</th>
<th>Physical, chemical, biological</th>
<th>Physical, chemical, biological</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reception of raw meat materials</td>
<td>Physical, chemical, biological</td>
<td>Red meat color, pH not above 6.2 (DFDI), no visual defects of meat/fat/skin surfaces, core temperature ≤4°C</td>
<td>Check purchase specification. Inspection by random sampling of appearance, odour, temperature and pH</td>
<td>Trim surface if only few minor visible contaminations or remaining hairs. Reject delivery, if other target levels not met</td>
<td>Physical characteristics of meat received, certificate of sanitary status and origin of meat. Meat temperature recordings.</td>
</tr>
<tr>
<td>Storage in reception chiller</td>
<td>Biological</td>
<td>Chiller temperature ≤4°C</td>
<td>Periodic temperature control</td>
<td>Minor temp. deviation: Adjust temperature. Major temperature deviation: Reject meat</td>
<td>Temperature/time recordings</td>
</tr>
<tr>
<td>Cutting, deboning, trimming</td>
<td>Biological</td>
<td>Room temperature +10°C, meat temperature ≤+7°C. Absence of alterations in meat such as abscesses, purulent or blood infiltrations</td>
<td>Meat temperature control. Check for meat alterations and abnormal tissues</td>
<td>Further cooling if meat temperature too high. Reject / discard entire meat parts with alterations such as abscesses, purulent/blood infiltrations</td>
<td>Record meat temperature. Record accidental findings</td>
</tr>
<tr>
<td>Evaluation and weighing of non-meat ingredients</td>
<td>Chemical</td>
<td>Nitrite content in curing salt ≤0.6% (if curing salt mix done by operator). Curing salt free of impurities. No impurities in other non-meat ingredients</td>
<td>Check storage conditions of nitrite salt, exact weighing of nitrite portion (if mix done by operator). Curing salt quality check. Check other non-meat ingredients for impurities</td>
<td>Adjust weight of nitrite portion correctly or use freshly mixed curing salt. Replace other non-meat ingredients</td>
<td>Records of status and expiration dates of non-meat ingredients. Results of weighing nitrite portions</td>
</tr>
<tr>
<td>Preparation and injection of curing brine</td>
<td>Physical, chemical biological</td>
<td>Brine temperature at injection 2±4°C</td>
<td>Check brine temperature</td>
<td>No utilization of curing brines failing temperature and purity requirements</td>
<td>Record conditions encountered</td>
</tr>
<tr>
<td>Tumbling</td>
<td>Biological</td>
<td>Room temperature ≤+4°C, time ≤ 8 hours</td>
<td>Check temperature/time</td>
<td>Adjust room temperature if too high</td>
<td>Temperature/time recordings</td>
</tr>
<tr>
<td>Packaging, moulding</td>
<td>Biological</td>
<td>Cleanliness of synthetic materials, tightness of enclosure by clip or seal</td>
<td>Check quality of materials and clipping/sealing</td>
<td>Reject unsuitable synthetic bags, correct clipping/sealing failures</td>
<td>Record on packaging material, equipment</td>
</tr>
<tr>
<td>Cooking</td>
<td>Biological</td>
<td>Internal cooking temperature (core temperature) ≤+70°C. Temperature of cooking media +78°C</td>
<td>Check core temperature by electronic temperature measurement</td>
<td>Increase cooking temperature or prolong cooking time until required core temperature is reached</td>
<td>Record temperature of production batch. Record any deviation in temperature</td>
</tr>
<tr>
<td>Cooling (in water)</td>
<td>Biological</td>
<td>Cooling to ≤+15°C core temperature in ice water</td>
<td>Check core temperature / time. Check cooling water temperature Add ice if cooling water temperature too high</td>
<td>Time/temperature record of cooling period</td>
<td></td>
</tr>
<tr>
<td>Storing (chiller)</td>
<td>Biological</td>
<td>Temperature of cooling room ≤+4°C</td>
<td>Check temperature daily</td>
<td>Adjust temperature as the case may be</td>
<td>Record of cold chain temperature</td>
</tr>
</tbody>
</table>

CCP = Proposed Critical Control Point

1) pH to be measured at topside (Musc. gracilis)
2) Alternatively: check meat and decide on further utilization for processing into hygienically less sensitive products.
Table 18: HACCP plan for Fresh Frozen Beef Burger

<table>
<thead>
<tr>
<th>HACCP PLAN</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Product:</strong> Fresh Frozen Beef Burgers (extended, with salt and spices, vacuum packed)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Process steps</th>
<th>Hazard</th>
<th>Target level/ Critical limit</th>
<th>Monitoring Procedure</th>
<th>Corrective action if standards are not met</th>
<th>Records</th>
</tr>
</thead>
<tbody>
<tr>
<td>Reception of raw meat materials (beef, boneless) CCP</td>
<td>Physical, chemical, biological</td>
<td>Internal meat temperature (\leq+4^\circ C), red meat colour, fresh. Slightly acidic odour, no visible contamination, no discoloration, not slimy, no other defects</td>
<td>Check purchase specification. Inspection of meat surfaces by random sampling. Check internal meat temperature</td>
<td>Reject delivery, if target levels not met</td>
<td>Physical characteristics of meat received, certificate of sanitary status and origin of meat. Meat temperature recordings</td>
</tr>
<tr>
<td>Storage in reception chiller</td>
<td>Biological</td>
<td>Room temperature (\leq+4^\circ C). Meat internal temperature (\leq+4^\circ C)</td>
<td>Temperature control of chilling room and meat (internal)</td>
<td>Minor temperature deviation: Adjust chiller temperature. Major temperature deviation: Reject meat</td>
<td>Temperature/time recordings of chiller. Temperature recordings of meat</td>
</tr>
<tr>
<td>Weighing and composition of non-meat ingredients</td>
<td>Physical, chemical</td>
<td>Visibly clean non-meat ingredients (common salt, no curing salt to be used)</td>
<td>Check salt, spices and extenders for impurities</td>
<td>Reject suspected batches of non-meat ingredients</td>
<td>Record of status and expiration dates for non-meat ingredients</td>
</tr>
<tr>
<td>Prepare meat for grinding, effect grinding</td>
<td>Biological</td>
<td>Room temperature (\leq+10^\circ C). Period from delivery of meat from chiller to pass through grinder maximum 20 minutes. Meat free of grossly abnormal tissues and post-dressing contamination</td>
<td>Check period of product flow. Check for abnormal tissues and post-dressing contamination</td>
<td>Improvement in product flow. Discard meat parts with abnormal tissues, post dressing contamination</td>
<td>Product flow/temperature recording</td>
</tr>
<tr>
<td>Mixing of meat with ingredients CCP</td>
<td>Biological</td>
<td>No further increase of contamination. Room temperature (\leq+10^\circ C). Period from grinding to completion of mixing/blending maximum 30 minutes. Temperature of meat/meat ingredients mix (\leq+10^\circ C)</td>
<td>Check period of product flow. Check mix temperature</td>
<td>Minor deviations: Adjust time/temperature regime. Major deviations: Reject batch</td>
<td>Product flow/temperature recording</td>
</tr>
<tr>
<td>Patty moulding</td>
<td>Biological</td>
<td>Carry out immediately after mixing. No significant product temperature increase</td>
<td>Temperature/time control</td>
<td>Increase process speed. Return mix to chiller if no immediate moulding process</td>
<td>Product flow/temperature recording</td>
</tr>
<tr>
<td>Freezing CCP</td>
<td>Biological</td>
<td>Blast freezer at (-35^\circ C)</td>
<td>Temperature control</td>
<td>Adjust freezer temperature</td>
<td>Record blast freezer temperatures</td>
</tr>
<tr>
<td>Packaging</td>
<td>Biological</td>
<td>Clean packaging materials</td>
<td>Check packaging failures</td>
<td>Adjust packaging machine in case of insufficient vacuum packaging</td>
<td>Results of packaging</td>
</tr>
<tr>
<td>Freezer storage</td>
<td>Biological</td>
<td>Temperature of storage freezer (-18^\circ C) to (-30^\circ C)</td>
<td>Continuous temperature check</td>
<td>Rise of temperature: immediate identification and correction of temperature problems, transfer to alternative storage freezer if long-term problem</td>
<td>Continuous freezer temperature records</td>
</tr>
</tbody>
</table>

CCP = Proposed Critical Control Point

\(^{1}\) Alternatively: Check meat and decide on further utilization for processing into hygienically less sensitive products.

**Remarks:** In the processing of this product there is no heat treatment included to reduce microbial contamination. The necessary heat treatment immediately prior to consumption, which is not part of the manufacturing process, is the only relevant measure to control potential contamination with pathogenic microorganisms. In order to minimize the risk of pathogenic microorganisms, special advice on the handling of the products before heat treatment and on the intensity of heat treatment must be available on the package.

During processing, the nature of the product requires periodic microbiological testing as part of GHP and HACCP verification. Microbiological testing of ground meat should take place once a week or more frequently in cases of suspected hygiene failures. Microbiological testing of finished mixes containing meat/non-meat ingredients mixes can be done on case-to-case basis.
The impact of microbial contamination on meat and meat products (Fig. 458)

Meat hygiene serves to minimize the impact of undesirable microorganisms and chemical residues on meat. While residue control is primarily the task of the competent authorities, control of microbial contamination is the responsibility of meat plants in the first place. Meat plant management and staff should therefore possess sufficient knowledge about impact of microorganisms on food and of basic rules on how to prevent or minimize microbial contamination (Fig. 453, 454, 455).

Microorganisms of relevance with regard to meat hygiene include parasites, moulds, bacteria and viruses. Within these groups bacteria play the most important role. Therefore, the focus of meat plant internal hygiene measures is mainly on bacteria, while moulds and viruses play a minor role but disinfection measures must also target them. The incidence of parasites should normally pose no major problems in meat which has passed meat inspection, or if efficient internal pest control programmes or measure are in place.

How does bacterial contamination of meat occur?

In live animals, the muscle meat is virtually sterile. However other parts of the animal such as skins, hooves and intestines contain enormous numbers of bacteria. Depending on the slaughter hygiene, these bacteria find their way to the carcass or “contaminate” the meat during slaughterhouse operations. Skinning, scalding, evisceration, dressing and carcass transport are common contamination points. Most bacteria reach the carcass via butchers’ hands, tools, contact with equipment or through water, air, etc. The bacterial contamination of meat is not stopped after slaughtering. It is ongoing during the operations following the slaughter process, such as meat cutting and meat processing (Fig. 452).

It is quite normal and unavoidable to find bacterial counts of “total plate count” (TPC, see page 335) of the order of several thousands per cm² on meat surfaces in commercial slaughtering and meat handling. However, the principle must be to keep bacterial counts as low as possible through adequate hygienic measures. Total plate count numbers exceeding 100,000 per gram (10⁵ per cm²) on fresh meat are not acceptable and alarm signals and meat hygiene along the slaughter and meat handling chain must be urgently improved (Table 19).
### Table 19: Recommended microbiological criteria for fresh meat

<table>
<thead>
<tr>
<th></th>
<th>Good microbiological standard</th>
<th>Critical microbiological condition</th>
<th>Not acceptable</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total plate count(^1) per cm(^2)</td>
<td>Less than 10000 &lt;10(^4)</td>
<td>Between 10000 and 100000 &gt;10(^4) - &lt;10(^5)</td>
<td>More than 100000 &gt;10(^5)</td>
</tr>
<tr>
<td>Enterobacteriaceae(^2) per cm(^2)</td>
<td>&lt;100</td>
<td>&gt;100 - &lt;1000</td>
<td>&gt;1000</td>
</tr>
</tbody>
</table>

---

1) “Total plate count” is the total number of bacteria comprising all microbial groups (page 336).

2) “Enterobacteriaceae” is a specific bacterial group, which indicate contamination by faecal and related materials (page 339).
Meat spoilage through micro-organisms

Meat spoilage bacteria will grow if temperatures are not kept in the cooling (-1°C to +4°C) or freezing (below -1°C) range. Not all bacteria which contaminate meat will behave in the same way. Some may multiply already at temperatures at around 10°C, others at higher temperatures, for example 30°C. Most bacteria can optimally grow in the range between 30°C and 37°C (Fig. 456 and Fig. 457). Some may attack the protein portion of the meat resulting in the production of very unpleasant putrefactive odours, others may break down carbohydrate components in particular in processed meats causing intensive sour taste or acidity. Others may attack the fats, producing rancidity (Fig. 458; table 20). These various bacterial impacts result in meat spoilage or decomposition. Spoilage of meat and meat products causes serious financial losses for the meat industries as such products, due to their sensory changes exposed through unpleasant smell and taste are unfit for human consumption. But spoiled meat, if accidentally ingested, is usually not the cause for illness in consumers.

Fig. 456: Growth of microorganisms on meat (starting from same initial bacterial loads/approx. 1000 per gram meat, but different storage temperatures, 0°C, 5°C, 10°C, 15°C). At 20°C spoilage on the second day at 0°C spoilage after more than 20 days.
Fig. 457: Growth of microorganisms on meat (starting from different initial bacterial loads/100, 10,000 and 500 million per gram, but same storage temperature (+5°C)).

Fig. 458: Impact of bacteria on meat

- **Meat spoilage**
  - **Putrefaction** (breakdown of protein)
  - **Slime**
  - **Sourness** (production of lactic acid)
  - **Discoloration**
  - **Rancidity** (breakdown of fats)

- **Food/meat poisoning**
  - **Infection** (ingested with contaminated food, bacteria multiply and produce toxins in consumer’s organism, cause illness)
  - **Intoxication** (microorganisms - bacteria, moulds - multiply in contaminated food and produce toxins, ingestion of toxins by consumer, causes illness)
<table>
<thead>
<tr>
<th>Phenomenon</th>
<th>Microorganisms</th>
</tr>
</thead>
<tbody>
<tr>
<td>Putrefaction</td>
<td>Pseudomonas (&quot;Cold room flora&quot;), Proteus, Clostridium (Fig. 459)</td>
</tr>
<tr>
<td>Souring</td>
<td>Lactobacillus, Enterococcus, Pediococcus (&quot;Lactic acid bacteria&quot;)</td>
</tr>
<tr>
<td>Fermentation¹</td>
<td>Yeasts (Saccharomyces), Enterobacteriaceae, Lactic acid bacteria</td>
</tr>
<tr>
<td>Turbidity (cloudy brine in meat juice)</td>
<td>Lactic acid bacteria, Enterobacteriaceae (e.g. vacuum packed meat, sausage slices)</td>
</tr>
<tr>
<td>Greenish discoloration</td>
<td>Lactic acid bacteria (Fig. 461)</td>
</tr>
<tr>
<td>Slime formation</td>
<td>Pseudomonas, Streptococcus, Enterobacteriaceae (on open meat), Lactic acid bacteria (on vacuum packed meat and meat products), Yeasts (on raw fermented products such as raw hams) (Fig. 460)</td>
</tr>
<tr>
<td>Rancidity of fats</td>
<td>Mainly due to presence of oxygen, but certain microorganisms are also capable of causing fat deterioration.</td>
</tr>
<tr>
<td>Mould growth</td>
<td>Penicillium, Aspergillus, Mucor (Fig. 462.)</td>
</tr>
</tbody>
</table>

¹ This refers to undesirable fermentation processes. For some meat products (raw-fermented hams and sausages) controlled fermentation is wanted and necessary (see page 124 and 177).
Meat poisoning through micro-organisms

Harmful microbes may have little adverse effect on carcasses or meat in terms of visible alterations and spoilage (smell and taste), but can have severe negative effects on consumers called food or meat poisoning. After consumption of meat contaminated with food poisoning bacteria, food poisoning results in severe illness with consumers needing intensive and costly medical treatment.

The impact of food poisoning bacteria, depending on the species of microorganisms, is either as a

- food borne infection or
- food borne intoxication.

Bacteria that cause food borne infections, must first multiply to high infectious numbers in rich protein foods such as meat and have to be ingested by consumers. They cause sickness through microbial metabolic substances i.e. toxic substances released by the living microorganisms inside the human digestive tract. The best known examples of food borne infections are those caused by Salmonella bacteria (Fig. 463). In some instances relatively high numbers of bacteria are needed to make people severely sick. For example, it is estimated that $10^5/g$ of Salmonella bacteria are needed in ingested food to cause Salmonellosis. In other cases, for example in the case of a recently emerged very pathogenic form of the normally harmless E.coli bacteria (entero-pathogenic form, mostly type O157 H7 residing in faecal material, on skin of animals), only a few hundred bacteria per gram food can cause severe illness with gastro-intestinal symptoms and fever and even death.
Microorganisms causing **food borne intoxications** produce and release the poison during their multiplication in the food. Upon ingestion by consumers of such food, which was heavily intoxicated outside the human body, severe gastro-intestinal food poisoning symptoms (**vomiting, diarrhea, abdominal pain, fever**) occur.

Food borne intoxications are frequently caused by *Staphylococcus aureus* (Fig. 464, 467, 468). These bacteria are present in purulent wounds and frequently in the respiratory system of healthy people. When they get into meat, which is not sufficiently refrigerated, they multiply rapidly and produce toxins, which cause severe gastro-intestinal symptoms only a few hours after ingestion by consumers. Another bacteria, *Cl. botulinum*, in the absence of oxygen e.g. in canned food or deep layers of raw-fermented hams, is capable of producing one of the strongest toxins known. Intoxication, if not treated immediately, can be fatal to consumers.

Bacteria are the most common food poisoning microorganisms. Apart from bacteria, moulds can also play a role in the incidence of food poisoning.

**Fig. 463: Food infection by Salmonella**
Deficient toilet hygiene, human carrier of Salmonella contaminates food (minced meat)
Moulds (Fig. 465) are sometimes found on the surface of meat products after prolonged storage. Growth of moulds (see page 124) on meat can have two undesirable effects. Firstly, strong growth of moulds can spoil the affected meat parts. Secondly, and this is a more serious issue, certain types of moulds produce toxins which are released into the food. If consumed in food or feed they can, in the long term, have carcinogenic effects.

**Aflatoxins** are strongly carcinogenic, in particular hepatotoxic, i.e. cause liver cancer through long term impact (Aflatoxin = toxin of *Aspergillus flavus*). **Ochratoxin** is strongly nephrotoxic, i.e. it causes kidney disease, in particular kidney enlargement and kidney failure (Ochratoxin = toxin of *Penicillium vividicatum*).
Viruses were always suspected to cause food infections. In the last years it has been shown that in particular the Norovirus group can be responsible for food infections with similar, mainly gastro-intestinal symptoms, as bacterial food infection agents.

Table 21: Major meat poisoning organisms

<table>
<thead>
<tr>
<th>Organism</th>
<th>Type of Infection</th>
</tr>
</thead>
<tbody>
<tr>
<td>Salmonella</td>
<td>Food borne infection</td>
</tr>
<tr>
<td>E. coli (enteropathogenic type)</td>
<td>Food borne infection</td>
</tr>
<tr>
<td>Listeria monocytogenes</td>
<td>Food borne infection</td>
</tr>
<tr>
<td>Campylobacter jejuni</td>
<td>Food borne infection</td>
</tr>
<tr>
<td>Yersinia enterolytica</td>
<td>Food borne infection</td>
</tr>
<tr>
<td>Staphylococcus aureus</td>
<td>Food borne intoxication</td>
</tr>
<tr>
<td>Clostridium botulinum</td>
<td>Food borne intoxication</td>
</tr>
<tr>
<td>Mycotoxin producing moulds</td>
<td>Food borne intoxication</td>
</tr>
<tr>
<td>Norovirus</td>
<td>Food borne infection</td>
</tr>
</tbody>
</table>

Good Hygienic Practices in meat processing

Microbial meat spoilage or food poisoning through meat can be prevented if the microbial load/bacterial contamination, which occurs during slaughtering and meat handling, is kept as low as possible. The key for achieving this is strict meat hygiene including an uninterrupted cold chain throughout the entire meat production and handling chain.

Meat hygiene is a complex field, based on regulations by competent authorities and meat plant internal hygiene programmes, to be supervised by the plant management (see page 341). Those programmes will only be successful if meat plant staff are familiar with and active in observing basic hygiene requirements. In order to facilitate the application of hygiene requirements, it has proven useful to differentiate between:

a. Personal hygiene
b. Slaughter and meat processing hygiene
c. Hygiene of slaughter and meat processing premises
d. Hygiene of slaughter and meat processing equipment
The topics a-d are of equal significance. Negligence in any of the four areas may give rise to hazards, which can cause economic losses and affect consumers’ health.

Some key requirements for meat processing plants are listed below. More detailed hygiene requirements are laid down in national regulations and in international codes, such as FAO/WHO CODEX ALIMENTARIUS Code of Hygienic Practice for Meat (CAC/RCP 58-2005). Guidelines on slaughter hygiene or meat transport and storage hygiene are not included hereunder. However, as meat is the primary material for processed meat products, the application of hygienic practices in slaughterhouses and throughout the cold chain is equally important. Principles of sanitation of premises and equipment are described in a separate chapter (page 369).

Principles of personal hygiene

- Wear clean protective clothes (Fig. 405, 406)
- Washing hands before starting work (Fig. 466)
- Repeatedly washing hands during work
- No finger rings, watches, bracelets
- Access to production areas with working clothes only
- Cleaning/disinfection of hands/tools/clothes if there was contact with highly contaminated subjects or abnormal animal parts likely to contain pathogens.
- Fresh wounds through knife cuts etc. must be covered by a water tight bandage. Workers with purulent wounds are not allowed to work with meat. (Risk of spread of Staph. aureus bacteria, see Fig. 464, 467, 468).
- Strict toilet hygiene must be observed (removal of apron, hand washing and hand disinfection). Toilets must be kept clean and must not have direct access to production areas. (Risk of spread of Salmonella, see Fig. 463).
- Periodic medical examination of staff
Ideally meat cutting/deboning should be carried out in climatized rooms (approx. +10°C) with low air humidity. Meat should be brought in progressively and not accumulate on work tables.

If visual contamination of manufacturing meat occurred, do not try to wash it off but remove it with knives by cutting off superficial meat parts in the case of minor contamination. Discard the meat in case of heavy contamination.

Do not hose down floor and wall areas or equipment next to meat processing operations or final products with a power hose. (Risk of contamination by aerosol/droplets, see Fig. 469).

Basic hygiene of meat processing

Fig. 467: Fresh non purulent wound, to be protected by impermeable bandage.

Fig. 468: Purulent wound, working with meat prohibited.

Fig. 469: Cleaning with pressurized water must be avoided in rooms where meat is present.
Never take meat pieces, which accidentally had contact with the floor or other contaminated surfaces, back onto working tables or into meat processing machines (Fig. 470).

Containers for meat, fat, or semi-or fully processed meat products must not be placed directly on the floor but on hygienic stands, pallets etc. (Fig. 471).

**Hygiene of meat processing premises**  
(*Hygienic requirements for lay-out and construction of slaughterhouse and meat processing buildings*)

Meat processing facilities must meet the following basic hygienic standards in order to ensure and maintain clean and hygienic working conditions:

- Adequate rooms for personnel must be available including sections for changing clothes and for personal hygiene.
- Wall windows must be positioned at a sufficient height from the floors in order to allow profound washing and disinfection of floors and walls. Wall windows for processing plants must be at their lowest part at least 2 m high, for easy cleaning.
high over floor level. Window frames should be of non-corrosive material e.g. aluminium or similar and must not be painted (Fig. 472).

- Walls in all rooms, where meat and by-products are handled, must have smooth and easily washable surfaces up to a minimum height of 2 m in processing plants. Walls should preferably be covered with wall tiles or at least with washable paint (Fig. 472, 475).

- Floors in the mentioned sections must be impermeable for water and reasonably smooth for good cleaning, but anti-slip for workers safety. They are usually made of fat-resistant concrete. Additional covering by epoxy substances or floor ceramics are possible (Fig. 473, 475).

- In order to facilitate proper cleaning, the junction between floor and walls must be coved, i.e. rounded (not rectangular), which can be achieved by extending the floor concrete up to an height of 10-50 cm alongside the walls. If the concrete layer alongside the wall is...
sufficiently thick (approx. 10-20 cm), it serves also as shock absorber and protects the walls against damage by transport vehicles, such as trolleys, fork lifts etc. Appropriate coves at wall-floor junctions can also be achieved by using special curved wall tiles (Fig. 475).

- All wet rooms must have floor drains, which should be covered by non-corrosive metal plates or grills (Fig. 473). The covers should be easily removable for proper cleaning of the drains. Drain sinks must be of the siphon type (anti-smell).

- Provisions must be made to channel waste water from hand-wash facilities, cool room evaporators, tool sterilizers, etc. by means of water pipes or similar directly into effluent drains without contaminating the floor.

- Rooms for meat processing should have sufficient ventilation. Air conditioning is only required in meat cutting/deboning rooms (10 - 12°C).

- Supply systems for electrical wiring and pipes for hot and cold water as well as for compressed air should not hamper cleaning operations and be out of reach of possible dirt contamination (Fig. 478). Insulations for hot water pipes must have smooth surfaces and be washable.

- Openings for ventilation must be bird- and insect-proof.
Hygiene of meat processing equipment  
(Hygienic requirements for design and construction of machinery, working tables and tools)

In production lines in the meat industries equipment and hand-tools should be used, which enable workers to perform all operations according to Good Hygienic Practices. It is the responsibility of the meat plant management to provide adequate equipment for all working places. For equipment manufactures, directives have been issued as to proper design and construction of meat processing equipment. Designs must allow easy and profound cleaning and avoid any accumulation of difficult to remove organic matters (negative examples see Fig. 476, 477, 479, 480).

As a principle in modern meat industries it is commonly accepted that tools and surfaces in contact with meat should be made of food grade stainless steel or synthetic materials. **Stainless steel** must be used for working tables, meat hooks (at least their parts contact in meat), blades of knives, saws, cleavers and axes. All parts of machinery in contact with meat, fat, sausage mixes and meat ingredients must be of stainless steel such as frozen meat cutter, grinder, meat mixer and tumbler, meat emulsifier, sausage stuffer, brine injector etc. The bowls of bowl cutters are nowadays also mostly made of stainless steel. All the stainless steel parts must be smooth, easily accessible for cleaning and without hidden spaces, where particles of meat materials may accumulate (Fig. 481, 482).
Galvanized steel or food-grade aluminium are useful materials in the meat industries as they are non-corrosive. Those materials should however not be in direct contact with meat, as they are not sufficiently smooth or may release unwanted substances. But they are very suitable materials for overhead rails and supporting structures, working platforms and frames for tables and machinery (Fig. 481).

Food grade synthetic materials are used for many types of meat containers and for handles of knives and other hand tools, for cutting boards and some internal parts of meat processing equipment such as washers, parts of valves etc. (Fig. 483, 484, 485, 486, 487).
In summary it can be stated that Good Hygienic Practices in meat processing requires efforts by both management and staff.

- It is the duty of the plant management to procure investments in **good quality premises and equipment** and in continuous **plant and equipment maintenance**.
- For the meat plant staff it is an obligation to observe during all meat processing operations **relevant hygienic rules**.

Such efforts will result in good storage life of attractive meat products with desirable appearance, flavour and taste.
Impact of *Halal* and Non-halal Slaughtering on the Microbiological Characteristics of Broiler Chicken Meat and Sausages

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Abstract    The *halal* (permissible) rules of slaughter are based on Islamic law. The animal has to be alive and healthy, a Muslim has to perform the slaughter in the appropriate ritual manner, and the animal's throat must be cut by a sharp knife severing the carotid artery, jugular vein and windpipe in a single swipe. Blood must be drained out of the carcass. The main objectives of this study were to compare between the quality characteristics of chicken slaughtered according to Islamic rules (*halal*) and those slaughtered according to non-Islamic rules (non-halal). Twenty hydroid strains of broiler chickens were used and their weights ranging between 1.5-1.75 kg, these chickens were divided into two groups; the first group were slaughtered according to Islamic rule, while according to non-Islamic rule. Sausage was prepared from both types of meat. Microbiological methods were used to analyze chicken meat as well as sausage. The results showed that coliforms were not detected in *halal* chicken meat, while the non-halal meat contained $3.0 \times 10^3$ c.f.u./g. Moreover, the *E. coli* were found in relatively large number in non-halal meat ($4.0 \times 10^4$ c.f.u./g) in contrast to halal meat which did not contain any *E. coli* cells. The halal meat sausage samples were not contaminated with either Coliforms, *E. coli* or Salmonella, while the non-halal meat sausage contained $15.0 \times 10^5$, $23.3 \times 10^4$ and $15.0 \times 10^4$ c.f.u./g of Coliforms, *E. coli* and Salmonella, respectively. It is highly recommended to follow the Islamic rule in slaughtering poultry and to apply hazard analysis and food hygiene to reduce the risk of cross contamination with foodborne pathogens in poultry farms.

Keywords  *Halal* slaughtering, Chicken, Meat, Sausage

1. Introduction

Sudan has a very good potential to be world major player in poultry products exports, a lot of local and international investors are starting new poultry business in Sudan. This development in poultry sector is a part of Sudan huge economic development (http://news.sudanvisiondaily.com/details.html?rsnpid=206028) [1].

An efficacious way of preventing food-borne human diseases is to monitor the microbiological quality of poultry meat and meat products during production, storage and distribution. According to Fries [2], the microflora of poultry is transferred from the primary production sites to production lines, and further, by subsequent contamination. Microflora of crude chicken meat is heterogeneous and originates from slaughtering premises, operators’ hands, equipment and outfit, and water and air [3].

Processed raw poultry meat naturally harbors bacteria. Most of the bacteria are responsible for the spoilage of poultry meat. However, poultry products can harbor food borne pathogens, from which salmonella stereotypes, *Clostridium jejuni*, *Listeria monocytogen*, *Clostridium perfringens* and *Staphylococcus aureus* [4]. Therefore, poultry and poultry products ranks first or second in foods associated.

*Halal* meat essentially is meat that Muslim is allowed to eat according to Islamic law. The laws require that only certain types of meat can be eaten and that meat must be prepared in a certain way, it is also essential that halal foods is not prepared with non-halal food as there is a risk of cross contamination if a chef accidentally uses the same knife to cut the different types of meat with for example.

According to Qur'an and Islamic law some substances are wrong for people to eat, whether by their nature in the way in which they have been treated or butchered, so all halal meat is prepared according to strict guidelines [5]. Allah's name should be pronounced over the meat as thanks during the slaughter process, any animal slaughtered in another idol's name can be never being halal.

Processing of poultry meat involves conversion of raw poultry carcasses into value added products e.g.
reconstructed products, cold cuts or breaded products. Further processing of poultry meat are improving juiciness and flavor, shelf life and water holding capacity [6].

Sausages are a category of processed meat. They are minced processed meat and/or comminuted meat, which may be combined with other foods, and are encased or formed into discrete units. They do not include meat formed or joined into the semblance of cuts of meat. They often used as substitutes for meat flesh. They have a maximum of 50% fat-free meat flesh [7]. Natural and artificial casings are used as forms and containers for sausages. The casings bind and protect the sausage mixture as well as the expansion of the sausage.

The objective of the present study was to compare between the microbiological qualities of broiler chicken slaughtered according to Islamic and non-Islamic rules as well as to produce sausage from the two types of chicken meat and evaluation of its microbiological quality.

2. Materials and Methods

2.1. Collection of Samples

Twenty broilers chicken were collected from Wad Almajzoob farm (central Sudan). These chickens are hydroid strains and their weights ranging between 1.5 - 1.75 kg. Each sample was taken immediately after slaughtering in a sterilized container, and transported under aseptic conditions to the Meat Technology Laboratory at the Department of Food Science and Technology of the University of Gezira.

2.2. Methods of Slaughtering

2.2.1. Halal Slaughtering

The chickens were divided into two groups; each group contained ten broilers chicken. The first group was slaughtered according to Islamic rule (Halal slaughtering).

The Halal slaughtering method started with holding the right foot, the feet of the broiler chicken was held. With the left foot, the wings were held down. The boiler chicken was given a drink water so that it relaxed as by this time it was under a lot of stress. The feathers were picked off from the front of the neck so that the knife did not have to cut through the feathers and takes too long. The process started at the beginning by reciting Allah's name and Tasmiyah (Bismillah Allahu Akbar).

2.3. Preparation of Meat Samples

Poultry meat samples were prepared for analysis, these samples included: Halal and Non-halal chicken meat samples. The meat samples were transported immediately to the Department of Food Science and Technology laboratory pending.

2.4. Manufacture of Sausage

Chicken sausage products from halal chicken meat (HCS) and non-halal chicken meat (NHCS) were prepared at the laboratory using standard method adopted by the sausage manufacturer in the Sudan. In this method:

Three kg of minced chicken meat were mixed with the ingredients which included: 150 gm wheat flour, 150 gm of vegetable protein (chickpea), 4gm of coriander, 4 gm of shamar, 2gm of chili powder, 2gm of pepper, 2gm of cinnamon, 2gm canella, 2gm of nutmeg, 3 gm of garlic and 10 gm of salt. All these spices had been ground before addition to the recipe. The minced meat and ingredients were mixed in an electric mincing machine, then the mixture was transported to the sausage casing machine, in which the minced meat was enforced into previously prepared cleaned sheep intestine casings, the product was formed into finger-like forms of about 5-7 cm in length.

2.5. Preparation of Serial Dilution

For preparation of serial dilution, 10 grams meat sample was shaken thoroughly with 90 ml sterile distilled water to give $10^{-1}$ dilution. Asset of 6 tubes containing 9ml sterile distilled water was prepared and 1ml of the suspension was transferred to the first tube of the dilution series. This was repeated up to the dilution $10^{-7}$ and 1 ml of the suspension was transferred to the first tube of the dilution series. This was repeated up to the dilution $10^{-7}$.

2.6. Microbial Analysis

The different microbiological characteristics of chicken meat product (sausages) were carried out according to Harrigan and McCane [9] methods. These methods included:

2.6.1. Total Viable Count

One ml aliquots from suitable dilution were transferred aseptically into sterile Petri dishes. To each dilution, 10–15 ml of melted and cooled (42°C) plate count agar were added. Inoculums was mixed well with the medium and allowed to solidify. The plates were then incubated at 37°C for 24 hours. The total viable count was calculated by the standard formula:

$$\frac{S}{S+D} \times P \times C = TCFU$$

2.6.2. Yeast and Mould Count

From suitable dilution 0.1 ml samples was aseptically
surface plated on to Potato Dextrose Agar medium (PDA) with 40 ppm Chlor amphenicol added to inhabit bacterial growth. The plates were incubated at 25°C - 28°C for 48 hour as described by Harrigan and Mac Can [9]. The counts were presented as colony forming units per gram (cfu/g).

2.6.3. Coliform Test

One ml of sample was plated onto (MacConky Agar) media. The plates were incubated at 37°C for 48 hours and the counts were presented as colony forming unites per gram (cfu/g).

2.6.4. E.coli Test

Plates showing positive coliforms were subjected to the confirmed test using Brilliant green bile lactose broth in test tubes with Durham tubes. The test tubes were then incubated at 44°C for 48 hours. Each confirmed positive tube was sub cultured into E.C. broth medium and then incubated at 44.5°C for 24 hours. Tubes showing any amount of gas production were considered to be positive.

2.6.5. Salmonella Detection

Ten grams of sample were weighted aseptically and mixed well with 100 ml sterile nutrient broth. This was incubated at 37°C for 24 hours. Then 10 ml were drawn aseptically and added to 100 ml selenite broth. The broth was then incubated at 37°C for 24 hours. Then with a loopful streaking was done on dried Bismuth sulphite agar plates. The plates were then incubated at 37°C for 72 hours.

Black metallic sheen discrete colonies indicated the presence of salmonella. A confirmatory test was carried out by taking a discrete black sheen colony and sub culturing it in a Triple sugar iron agar tubes.

Production of black colour at the bottom of the tube confirms the presence of salmonella.

3. Results and Discussion

The study has taken into consideration all the samples of poultry meat and sausage made from poultry meat that arrived at the laboratory during the period from 01/8/2013 to 10/30/2013.

The microbiological safety and quality of poultry meat are equally important to producers, retailers and consumers, and both involve microbial contaminants on the processed product. Two quite different groups of microorganisms are relevant: on the one hand certain foodborne pathogens, and, on the other, organisms that are generally harmless to human health, but, being psychrotrophic, are able to multiply on the product during chill storage. Spoilage results mainly from ‘off’-odour development, and product shelf-life is determined both by the number of spoilage organisms present initially and the temperature history of the product at all stages of production and subsequent storage and handling [10]. For chill-stored poultry, Viehweg et al. [11] demonstrated that virtually all the odorous substances found at spoilage could be attributed to microbial growth and metabolism.

Friedhoff et al. [12] have described the use of simple microbiological criteria, including aerobic mesophilic colony counts, Enterobacteriaceae counts and in some instances, enumeration of yeast, performed on samples taken during processing in small businesses to verify good manufacturing practices. This verification through monitoring was found to be an attractive alternative to the examination of end products.

Contamination of poultry meat with foodborne pathogens remains an important public health issue, because it can lead to illness if there are malpractices in handling, cooking or post-cooking storage of the product. In developed countries, foodborne illness causes human suffering and loss of productivity, and adds significantly to the costs of food production and healthcare. It is also a possible cause of mortality, which is even more of a problem in developing regions, where the health status of many individuals is already compromised.

The microbiological characteristics of halal and non halal poultry meat and sausage prepared from that meat are indicated in Tables (1 and 2).

<table>
<thead>
<tr>
<th>Sample</th>
<th>Total Viable Count (c.f.u/g)</th>
<th>Total Yeast and Mould (c.f.u/g)</th>
<th>Coliforms (c.f.u./g)</th>
<th>E. coli (c.f.u/g)</th>
<th>Salmonella (c.f.u/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HM</td>
<td>2.3 x 10⁴</td>
<td>9.1x10³</td>
<td>Nil</td>
<td>Nil</td>
<td>Nil</td>
</tr>
<tr>
<td>NHM</td>
<td>13.1 x10⁴</td>
<td>61.1x10³</td>
<td>3.0 x 10⁵</td>
<td>4.0 x 10⁵</td>
<td>4.3x10⁴</td>
</tr>
</tbody>
</table>

HMS: Halal meat sausage
NHMS: Non-halal meat sausage

<table>
<thead>
<tr>
<th>Sample</th>
<th>Total Viable Count (c.f.u/g)</th>
<th>Total Yeast and Mould (c.f.u/g)</th>
<th>Coliforms (c.f.u./g)</th>
<th>E. coli (c.f.u/g)</th>
<th>Salmonella (c.f.u/g)</th>
</tr>
</thead>
<tbody>
<tr>
<td>HM</td>
<td>15.6 x 10⁴</td>
<td>51.0 x 10⁵</td>
<td>Nil</td>
<td>Nil</td>
<td>Nil</td>
</tr>
<tr>
<td>NHM</td>
<td>84.0 x10⁴</td>
<td>91.3 x 10⁵</td>
<td>15.0x10⁵</td>
<td>23.3 x 10⁴</td>
<td>15.0 x 10⁴</td>
</tr>
</tbody>
</table>

HMS: Halal meat sausage
NHMS: Non-halal meat sausage
3.1. Microbiological Characteristics of halal and Non-Halal Poultry Meat

The Microbiological characteristics of halal and non halal poultry meat is shown in Table (1). Almost half of the tested samples of poultry show a low contamination and only a few samples had high microbe contamination. The total viable count of halal poultry meat (2.3 x 10^4 c.f.u./g) was less than that of non-halal poultry meat which was 13.1 x 10^4 c.f.u./g. Also the yeast and mould count of non-halal poultry meat (61.1x10^4 c.f.u./g) exceeded that of halal poultry meat which was 9.1x10^3 c.f.u./g. On the other hand, coliforms were not detected in halal poultry meat, while the non-halal meat contained 3.0 x 10^7 c.f.u./g of coliforms. Moreover, the E. coli were found in relatively large number in non-halal meat (4.0 x 10^4 c.f.u./g) in contrast to halal meat which did not contain any E.coli cells. Salmonella spp. were not detected in halal meat while it was found in large numbers in non-halal meat (4.3x10^4 c.f.u./g). Little et al., [13] detected Salmonella spp. and E.coli in 7% and 0.6% of the 183 raw meat products he tested, respectively.

Generally, the microbial analysis indicated that halal slaughtering method resulted in lowering the various microbial loads of poultry meat. These data, however, coincide with that reported in the literature[14] [15] whereas they are shown to be much higher with respect to that recently found by Teldeschi [16] in samples of meat from chicken and products derived from chicken.

The levels of viable bacteria recovered from non-halal meat samples in this study are similar to those found in a Canadian baseline study of poultry carcasses slaughtered in federally inspected abattoirs in 1997 to 1998 and in the USDA baseline study conducted in 1994 to 1995. The majority of samples had coliform bacteria and generic E. coli at levels of 11 to 100 CFU/cm^2, which is also the range for coliforms found in the USDA baseline study for the majority of the samples. The level of E. coli isolated from non-halal meat, however, was higher than the levels reported in the previous Canadian and USDA baseline studies in which most samples had E. coli in the range of 1 to 10 CFU/cm^2.

Coliform bacteria are one of the most important indicator organisms that are most commonly used to ensure food safety. Coliform bacteria include a large group of many types of bacteria that occur throughout the environment. They are common in soil and surface water and may even occur on your skin. Large numbers of certain kinds of coliform bacteria can also be found in waste from humans and animals. Most types of coliform bacteria are harmless to humans, but some can cause mild illnesses and a few can lead to serious waterborne diseases.

Specific types of coliform bacteria may be tested for, especially after a total coliform bacteria test is positive. These subgroups of coliform bacteria include fecal coliform and Escherichia coli or E. coli. Fecal coliform bacteria are specific to the intestinal tracts of warm-blooded animals, including humans, and thus require a more specific test for sewage or animal waste contamination. E. coli is a type of fecal coliform bacteria commonly found in the intestines of animals and humans. A positive E. coli result is much more serious than coliform bacteria alone because it indicates that human or animal waste is entering the water supply. There are hundreds of strains of E. coli. Although most strains are harmless and live in the intestines of healthy humans and animals, a few strains can produce a powerful toxin and can cause severe illness and death.

Contamination of poultry carcasses and parts with salmonella is well documented and data are available for many parts of the world [4] [17], although inter-country comparisons are not usually possible, because of differences in sampling and methods of testing. Most salmonellas found on poultry meat are non-host-specific and are considered capable of causing human food poisoning. Salmonellae survive well in the environment. Also, growth only occurs under conditions of high moisture, reduced oxygen and an environmental temperature above 30ºC. The organisms are particularly sensitive to drying and the effects of freezing and thawing, which can cause a 1 - 2 log reduction in the level of contamination on poultry meat.

It has been reported that E. coli O157 and Salmonella pathogens are known to colonise the intestines of farm animals and may contaminate meat of cattle, sheep, and poultry at the time of slaughter. Pathogenic microorganisms are therefore inherent constituents of raw meat and its products. Subsequent handling and processing of raw meat products such as comminution may spread [18][19].

3.2. Microbiological Characteristics of Sausages Prepared from halal and Non-Halal Poultry Meat

Meat products may be contaminated with microorganisms from meat handlers, who carry pathogenic microorganism during the processes of manufacturing, packing and marketing. Improper cooking, refrigeration or storage may lead to meat borne illness [20]. Foodborne pathogens are the leading causes of illness and death in developing countries costing billions of dollars in medical care medical and social costs.

The microbiological characteristics of sausages prepared from halal and non halal poultry meat is shown in Table (2). The total viable count of halal meat sausage (HMS) and non-halal meat sausage (NHMS) was 15.6 x 10^4 c.f.u./g and 84.0 x10^4 c.f.u./g, respectively. On the other hand, the total yeast and mould count of HMS and NHMS was 51.0x10^4 and 91.3x10^4(c.f.u./g), respectively. The results in Table (4.2) indicted that the HMS samples were not contaminated with either Coliforms, E. coli or Salmonella, while NHMS samples contained 15.0x 10^5 c.f.u./g 23.3 x 10^5 c.f.u./g and 15.0 x 10^3 c.f.u./g of Coliforms, E. coli and Salmonella, respectively. This indicates that halal slaughtering method resulted in lowering microbial viable counts of halal meat sausage samples. The large counts of different microbial groups in these raw prepared sausages could reflect contaminated supply, cross contamination, and/or poor hygiene practices. However, the microbial contamination...
may be added or reduced at different stages of processing of hot sausage. Dowdell and Board [21] carried out a microbiological survey of British Fresh Sausage and reported the presence of coliform bacteria. Also, Sachindra et al. [22] who isolated coliforms from raw and cooked sausage concluded that cooking process reduce the microbial counts substantially in the sausage.

Some studies showed that the augment of the additives, onion, garlic meal, pepper and E vitamin can decrease the microbial agents in cooked sausage products [23][24][25].

4. Conclusions

Generally, the microbial analysis indicated that halal slaughtering method resulted in lowering the various microbial loads of poultry meat although the raw meat as well as its product sausage were contaminated with various microbial groups which could reflect contaminated supply, cross contamination, and/or poor hygiene practices. However, the contamination data obtained in the present study are in line with or even less than those reported in the cited literature and fall within the limits set by national legislation.

It is highly recommended to apply the basic rules of hygiene to prevent raw poultry meat from contaminating other foods.

ACKNOWLEDGEMENTS

The authors express their gratitude and thanks to the staff of the Department of Food Science and Technology of Gezira University for presenting technical assistance.

REFERENCES


The Taxonomy and Physiology of Fungi

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1. General characteristics of fungi

- **Eukaryotic**
- Cell structure superficially similar to plant cells, however:
  - Lack photosynthetic pigments
  - The cell wall contains generally chitin but not cellulose
- Grow both as single cells (yeasts) or as mycelia or hyphae
1.1. The taxonomic position of fungi among other eukaryotes

- According to the recent genomic data superficially very diverse eukaryotic organisms can actually be divided into six major phylogenetic “Supergroups”
- Fungi, together with animals, belong to ”Opisthokonta” supergroup (eukaryotes with polar flagella)
1.2. The classification of fungi

- A major taxonomic revision is underway (see the previous slide)
- Several former fungi have been now transferred to either animal or plant kingdoms (former Myxomycetes now amoebas and "Oomycetes" or "Water moulds" are related to brown algae
- The modern phylogenetic classification is shown on right
1.3. Chytridiomycota

- Produce motile, asexual zoospores and motile gametes
- Water organisms
- Some species plant pathogens
1.3.2. Zygomycota

- Form **zygosporangia** during the sexual reproduction
- Mainly terrestrial moulds
- Chitosan as the cell wall polymer
- Form asexual **mitos pores** (in specific sporangia) and chlamydospores (fragmentation products of the hyphae)
- The asexual and sexual reproduction cycles shown on right
- Species of the genus *Mucor* important in certain biotechnological applications (organic acid production)
1.3.3. Ascomycota

- Most of the common moulds belong to this phylum
- Also *Saccharomyces* yeasts are considered as (pseudo)ascomycetes
- The sexual spores are formed in specialised "sacs" (asci)
- Produce also asexual conidiospores in specific conidiophores
- Ascomycetes that do have sexual life cycle are called *Fungi imperfecti* or *Deuteromyces*
  - Penicillium
  - Aspergillus
  - Asexual yeasts form a single artificial genus *Candida*
- The life cycle of a typical ascomycet shown on the right
1.3.4. Basidiomycota

- The common "mushrooms" as well as certain plant pathogens ("rusts")
- The life cycle is shown on the right
1.3.5. A pragmatic classification of microfungi (moulds and yeasts) for clinical purposes

- "Dimorphics" indicate that the fungus can grow in certain conditions as mycelium and in another conditions as separated cells (yeasts)
- "Opportunistic" means that these organisms can cause opportunistic infections
- "Dermatophytes" indicate fungi that colonize the skin and may cause infections
1.4. Fungal physiology and ecology

- All are heterotrophs (often called as saprophytes)
- Most have aerobic, respiratory metabolism, some (for example yeasts) are also able to grow fermentatively
- Generally require high $a_w$, but also some xerophytic fungi are known
- Generally favour mesophilic conditions and neutral pH, but again pH tolerant species are also found
- Many types of secondary metabolites produced, some so called mycotoxins
1.4.1. Mycotoxins

A. Some important mycotoxins of *Penicillium* and *Aspergillus*

- Aflatoxins are haepatotoxic and carcinogenic mycotoxins produced by *Aspergillus*
- Several types, $B_1$ the most toxic, $M_1$ the metabolite found in milk
- Ochratoxins are kidneytoxic (pigs especially vulnerable), genotoxic and potentially carcinogenic toxins produced by *Penicillium* and *Aspergillus* species

Fig. 4. Chemical structure of ochratoxin A (OTA).
1.4.2. Mycotoxins

B. *Fusarium*-toxins

- Trichotecenes
  - T2 toxin (top right), deoxynivalenol (DON)
  - T2 toxin causes damage to skin and mucous membranes, DON liver toxic (animals specially sensitive)
- Zearalenone (middle right)
  - Hormone-like activity
- Fumonisins (bottom right)
  - Very common in maize
  - Potentially carcinogenic
  - Horses very sensitive
1.4.2. *Saccharomyces cerevisiae*

- Reproduces by budding
- The sexual and asexual life cycles shown top right (mating types a and α)
- 16 chromosomes, also nuclear plasmids often present
- The mating type switching a well known model for cellular differentiation
1.4.3. *Stachybotrus cartarum*

- A mould belonging to Fungi imperfecti
- Favours moist environments with high cellulose content
- Associated with “sick” buildings
- Produces a variety of mycotoxins
- “Sick building” syndrome
1.4.4. *Trichophyton*

- A mould belonging to Fungi imperfecti
- Forms macro- and microconidia
- In soils and on skin (athletes’ foot, other dermal diseases)
1.5. Things to remember

• Fungi are eukaryotic micro-organisms forming traditionally a separate kingdom, now included in the supergroup Opisthokonta (related to animals)
• Grow as mycelia or, in case of yeasts, as single cell, found in many types of environments, mostly aerobic, although anaerobic metabolism sometimes possible (yeasts)
• Zygomycota, Ascomycota and Basidiomycota are the three phyla comprising the higher fungi
• Moulds (filamentous fungi) and yeasts important both in biotechnological applications, in ecology and in health and disease
• The secondary metabolism of fungi produces a wide variety of substances, some toxic
References

5. Introduction to the Microbiology of Food Processing Small Plant News Guidebook Series United States Department of Agriculture Food Safety and Inspection Service/
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Elements of Food Microbiology

Microbial Metabolism
Energy metabolism

• The food-associated micro-organisms are **heterotrophs**, meaning that they require organic carbon compounds both as a carbon source and for energy production. Sugars are typical energy sources, but also fats and proteins serve this function.

• The energy metabolism can be aerobic (based on cellular respiration) or anaerobic (or fermentative).

• The micro-organisms can be divided according to their energy metabolism into the following:
  – Obligate aerobes (only aerobic metabolism: many bacteria, molds)
  – Facultative anaerobes (both respiratory and fermentative metabolism possible: many bacteria, molds)
    • Aerotolerant organisms: only fermentative metabolism, tolerate oxygen (i.e. lactic acid bacteria)
  – Obligate anaerobes (only fermentative metabolism, do not tolerate oxygen: many bacteria)
Energy Metabolism
The production of pyruvate

- Pyruvic acid ("Brenztraubensäure") is the central metabolite of the energy metabolism
- Micro-organisms have three basic metabolic routes to pyruvic acid:
  - Glycolysis or Fructose-1,6-bisphosphate pathway (the most common)
  - Pentose phosphate pathway
  - 2-Keto-3-desoxy-6-phosphogluconate pathways (KDPG-route, only in some bacteria)
Energy Metabolism

High Energy Compounds and Reducing Molecules

- Adenosine triphosphate (ATP) is the central high energy compound to store and release metabolic energy (top figure).
- NaD(P) and its reduced form NAD(P)H₂ (lower figure) are central in the main cellular oxidation/reduction reactions.
- A general rule: Energy-yielding reactions oxidative, reductions often (but not always) energy consuming.
Energy Metabolism

**Glycolysis**

- Two molecules of pyruvic acid is formed from one molecule of glucose.
- Two molecules of ATP (a high energy compound) and two molecules of NADH$_2$ (a reducing compound) are formed.
Energy Metabolism
Pentose Phosphate Pathway

• A complicated cycle, in which transketolases and transaldolases are central:
• The net reaction: $3 \times \text{glucose} \rightarrow 2 \times \text{Fructose-6-P} + 3 \text{CO}_2 + \text{glyceraldehyde 3-P}$
• Glyceraldehyde-3-P will be further metabolized as in glycolysis
• Additional products include $2 \ \text{NAD(P)H}_2$ and biosynthetic intermediates

2-Keto-3-desoxy-6-phosphogluconate(KDPG) pathway

• Only in some bacteria
• Produces 2 pyruvates, 1 ATP, 1 NAD(P)H$_2$, and 1 NADH$_2$ per glucose processed
Energy Metabolism

The aerobic metabolism of pyruvic acid

The Citric Acid Cycle

- A multienzyme complex pyruvate decarboxylase turns pyruvic acid into acetylcoenzyme-A (Ac-CoA) and CO₂
- Ac-CoA joins the citric acid cycle by reacting with oxaloacetate forming citric acid
- The complicated cycle finally returns to oxaloacetate, liberating 2 CO₂, 3 NADH₂, FADH₂ and 1 ATP
  - FADH₂ another molecule involved in oxidation/reduction
Energy Metabolism

The aerobic metabolism of puryvic acid

The respiratory chain

• In the respiratory chain the reduced NADH$_2$ and FADH$_2$ formed in the pyruvate synthesis and in the citric acid cycle donate their electrons in successive oxido-reductions to oxygen, which is the final electron acceptor.

• The schematic presentation of the respiratory chain is the following:
  – Reduced NADH$_2$ and FADH$_2$ → [the sulphur-iron proteins of the membrane, kinones (coenzyme-Q) and cytochromes] → oxygen
  – In a complete respiratory chain each NADH$_2$ produces 3 ATP and FADH$_2$ 2 ATP
  – Many bacteria have their own versions of the respiratory chain (i.e. *Escherichia coli* lacks cytochrome-c, leading to a negative outcome in the oxidase test)

• The theoretical energy balance per one molecule of glucose is 38 ATP
Energy Metabolism
Anaerobic alternatives

- Pyruvic acid is the central molecule in many of the anaerobic fermentations.
- In the fermentation, the balance between oxidation and reduction is preserved by reducing some of the intermediates formed during the oxidative phase.
- As examples, ethanol- and lactic acid fermentations on the right.
Energy metabolism
Examples of other types of fermentations

- *Bifidobacterium*-fermentation: \(2 \times \text{glucose} \rightarrow 2 \times \text{Lactic acid} + 3 \times \text{Acetic acid}\)

- Propionic Acid Fermentation: \(3 \times \text{Lactic acid} \rightarrow 2 \times \text{propionic acid} + \text{Acetic acid} + \text{CO}_2\)

- *Enterobacteriaceae* – fermentations
  1. *Escherichia coli*-fermentations: end products lactic acid, formic acid, succinic acid
  2. *Enterobacter*-fermentations: end products asetoin and 2,3-butandiol (neutral compounds)

- *Clostridium*-fermentations: many end products, such as butyric acid, acetic acid, acetone, ethanol \(\text{CO}_2, \text{H}_2\) etc.
Secondary metabolism

• In addition to energy metabolism and biosynthetic activities many micro-organisms have also secondary metabolism.
• Examples of secondary metabolites are pigments, volatiles, antibiotics, toxins etc.
• Different mycotoxins (aflatoxins, ochratoxins, patulin, trichotocenes, fumonisin etc) produced by different molds, are typical harmful food-associated microbial secondary metabolites, as well as toxins produced by several food poisoning bacteria (*Staphylococcus, Clostridium, Bacillus* *ym*)
Other microbial metabolites

• Many catalase negative bacteria produce hydrogen peroxide (causing discoloration of foods)
• Many other small molecular weight compounds can be formed as a result of microbial enzymatic action
• Of particular interest regarding food are biogenic amines produced by microbial amino acid decarboxylases (histamine, tyramine, etc)
References


5. Introduction to the Microbiology of Food Processing Small Plant News Guidebook Series United States Department of Agriculture Food Safety and Inspection Service/

Food risks and hazards related to microorganisms and parasites

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1. Microorganisms – a general introduction

• Microorganism is a general term of taxonomically very heterogenous group of unicellular organisms that can not be observed with an unaided eye

• Three main groups are generally recognized:
  – Bacteria or prokaryotes
    • Proper bacteria
    • Archebacteria (often extremophilic organisms)
  – Protists (eukaryotes: fungi, microalgae, protozoa)
  – Viruses
1.1. The general features of different microbial cell types

- Typical bacterial cells (top right) have dimensions of few microns, have their DNA free in the cytoplasm and many of the metabolic functions occur on the cell membrane.
- Typical eukaryotic cells (middle right) have dimensions of up to tens of microns, have their DNA within a nuclear envelope and several cellular organelles that are responsible for metabolic functions.
- Viruses (bottom right) have dimensions of tens to hundreds on nanometers and consist of a nucleocapsid and nucleic acid (double or single stranded DNA or RNA). Obligate intracellular parasites.
1.2. Microorganisms in health and disease

• By far most bacteria and protists are either beneficial or harmless for humans and the environment
• A typical human being harbours appr. $10^{14}$ microbial cells (mostly bacteria), or ten times the number of somatic cells
• The studies on the importance of human microbiota in maintaining the normal health are stille in their infancy – a challenging prospect
• In contrast, the role of microorganisms as causative agents of infectious diseases has been the focus of attention since the times of Louis Pasteur
2. Microorganisms and Food

• Microorganism-based processes have been utilized for millennia to produce healthy and wholesome food
  – Fermented milks and other dairy products (cheeses etc.)
  – Sourdoughs and other bakery products
  – Salami-type of sausages and cured meat
  – Sauercraut, pickled cucumber, olives, kimchi & other types of fermented vegetables and fruit
  – Wine and beer

• However, microorganisms of all kinds (bacteria, fungi, protozoa, viruses) can also cause health problems as spoilage organisms and food-associated* pathogens

* "Food" in this context includes also water
2.1. The prevalence of microbial food poisonings

- According to the Finnish Food Safety Authority (Evira) the number of food poisoning incidents in Finland in 1975 – 2009 is annually 40 – 80 and the number of persons affected appr. 83 000!
- In 2009 alone the number of incidents was 55 and the number of diseased 1661
- The most common bacteria associated with food poisonings in 2009 were: *Clostridium perfringens* and *Salmonella*; the most common virus was water-associated norovirus (affecting 1386 people!)
- Less common but important food poisoning microorganisms include: *Listeria monocytogenes*, *Bacillus cereus*, *Staphylococcus aureus*, pathogenic *Escherichia coli*, *Campylobacter jejuni*, *Clostridium botulinum*, hepatitis A virus and certain protozoa (Giardia, Cryptosporidium)
- Annually the number of diseased persons is hundreds of thousands in the EU and tens of millions globally
2.2. How do the microorganisms cause food poisonings?

• The microorganisms can:
  – Cause an intestinal or systemic infection
  – Produce different toxins
  – Both cause an infection and a toxin-based poisoning
  – Produce biogenic amines (histamine, tyramine etc produced by many bacteria in proteinacious foods)

• Typical microbial toxins include:
  – Enterotoxins (typically proteins)
  – Various small molecular weight compounds
3. Food poisoning - associated bacteria

3.1. *Clostridium*

- Clostridia are strictly anaerobic, endospore forming Gram-positive rod shaped bacteria
- Endospored often at the other end of the cell
- Usual habitats soil, marine and lake sediments, gut
- Anaerobic metabolism and thermoresistant spores make canned foods especially vulnerable to clostridial contamination
3.1.1. *Clostridium perfringens*

- Found in soil, decaying plant material, water sediments and in the gastrointestinal tract
- High optimum temperature (37 – 43 °C)
- Strong gas formation and production of H₂S typical
- Causes common food poisonings (typically from meat dishes) with diarrhoea, headache and fever as main symptoms
- Infective dose $10^6 – 10^7$ cells/g food
- The symptoms start within 24 h and cease within 48 h from eating
3.1.2. *Clostridium botulinum*

- Divided into four physiological groups (I, II, III and IV)

<table>
<thead>
<tr>
<th>Properties</th>
<th>I</th>
<th>II</th>
<th>III</th>
<th>IV</th>
</tr>
</thead>
<tbody>
<tr>
<td>Toxin type</td>
<td>A,B,F</td>
<td>B,E,F</td>
<td>C,D</td>
<td>G</td>
</tr>
<tr>
<td>Proteolysis</td>
<td>yes</td>
<td>no</td>
<td>weak</td>
<td>no</td>
</tr>
<tr>
<td>Saccharolysis</td>
<td>no</td>
<td>yes</td>
<td>no</td>
<td>no</td>
</tr>
<tr>
<td>Temperature optimum</td>
<td>35 – 40 °C</td>
<td>18 – 25 °C</td>
<td>40 °C</td>
<td>37 °C</td>
</tr>
<tr>
<td>Lowest tolerated pH</td>
<td>4.6</td>
<td>5.0</td>
<td>?</td>
<td>?</td>
</tr>
<tr>
<td>Causes poisonings in</td>
<td>humans</td>
<td>humans</td>
<td>animals (birds)</td>
<td>?</td>
</tr>
<tr>
<td>Toxin genes</td>
<td>chromosomal</td>
<td>chromosomal</td>
<td>prophage</td>
<td>plasmid</td>
</tr>
<tr>
<td>Close species</td>
<td><em>C. sporogenes</em>, <em>C. putrificum</em></td>
<td><em>C. sporogenes</em>, <em>C. beijerinckii</em></td>
<td><em>C. haemolyticum</em></td>
<td><em>C. subterminale</em></td>
</tr>
</tbody>
</table>
3.1.2.1. The mechanism of action of botulinum toxins

Structure and activation of toxins

The neurotoxic action (inhibition of acetylcholine release in the neuronal synapsis)
3.2. *Salmonella*

- *S. enterica* the "important" species
- Gram-negative, facultatively anaerobic rods, many species produce hydrogen sulphide
- pH range 4.0 – 9.0, temperature range 5 – 45 °C
- Belong to Enterobacteriacea and are typical intestinal bacteria of humans and animals
- Present also in waters as a result of faecal contamination
3.2.1. The serology of *Salmonella*

- The Kauffman-White system, which is based on the combination of sera against O-antigens (bacterial LPS) and flagellar antigens (H-antigens) and capsular antigens (Vi), is used to differentiate *Salmonella* into thousands of different serotypes or serovars.

- The serovars are nowadays indicated as follows: *Salmonella enterica* subsp. *enterica* serovar Montevideo, or shorter *Salmonella* Montevideo.

- The three main serovars of *S. enterica* are Typhimurium, Enteritidis and Typhi.
3.2.2. Pathogenicity of *Salmonella*

- Cause typhoid and paratyphoid fever as well as less severe intestinal infections (enteritis)
- The cellular mechanism of pathogenicity is very complex
- Specific pathogenicity islands in the genome identified as well as pathogenicity plasmids
- The genes associated in the pathogenicity islands are thought to be involved in the intracellular invasion to host macrophages, while the plasmid-associated genes are required for the killing of invaded macrophages
3.2.3. *Salmonella* as a food/feed problem

- In Finland alone more than 2000 cases of *Salmonella* enteritis annually (however, the vast majority foreign origin)
- The serotypes often same as with domestic animals
- The incubation time appr 3 days, the symptoms diarrhoea, stomach cramps, fever
- The patients shed bacteria in their faeces for appr two weeks
- Regarding the animal feed and animal products the situation in Finland very good compared to Central and Southern Europe
3.3. *Listeria monocytogenes*

- Gram-positive, non-sporeforming facultatively anaerobic rod shaped bacterium
- Present in soils, waters and sediments, and in the gastrointestinal tract
- Common in domestic animals (ruminants)
- Temperature range for growth is $< 4 – 37 \, ^\circ\mathrm{C}$, and the minimum tolerated pH appr. 3.5, tolerates NaCl up to 25%
- Can exist either as a saprophytic bacterium or as a pathogen
3.3.1. Pathogenesis of *L. monocytogenes*

- Human pathogenesis
  - Intestinal infections
  - Invasive infections (listeriosis)
    - Septicemia
    - Meningitis
    - Intrauterine infections of pregnant women
      - Abortions and stillbirths
      - Meningitis in newborn

- At least 13 pathogenic serotypes known, but most common ones are 1/2a, 1/2b, and 4b.
3.3.2. Foodborne infections

• The exceptionally wide temperature and pH range as well as the salt tolerance of *L. monocytogenes* together with its ubiquitous presence in the environment make *L. monocytogenes* a serious food pathogen

• The food poisoning outbreaks can have a high rate of fatalities(sometimes 20 – 30 % of the cases)

• Symptoms range from mild, flue-like syndroms to gastrointestinal disorders, which may precede the more serious invasive infections

• Immunocompromised persons, very old and very young especially vulnerable

• Incubation time from week to a month
3.3.3. Risk foods

- Milk and dairy products (soft cheeses)
- Tubers and vegetables (contaminated with soil)
- Vacuum packed meat and fish products
- Sausages of salami type
3.4. *Bacillus cereus*

- Gram-positive, endospore forming aerobic bacteria common in soil
- Endospores are bacterial survival forms (seen as bluish dots inside the cells in the picture on right) that can tolerate heat (up to 100 °C) and germinate in suitable conditions
3.4.1. *B. cereus* food poisonings

- The contaminated foods mainly containing flour or rice, puddings, sauces and stews
- The spores tolerate the heat treatment, and germinate while the foods are subsequently stored at ambient temperature
- Two types of poisonings.
  - Diarrhoeal poisoning (common in Europe and North-America) caused by proteinaceous enterotoxins
  - Emetic poisoning (common in Far-East) caused by the emetic toxin (cereulide)
- The symptoms start within few hours of the meals and are usually over in 24 hours
3.4.2. *B. cereus* enterotoxins

- Proteins that maybe present already in the contaminated food, however probably mainly produced in the gut by the ingested bacteria

<table>
<thead>
<tr>
<th>Toxin</th>
<th>Subunits and sizes</th>
</tr>
</thead>
<tbody>
<tr>
<td>Hemolysin BL</td>
<td>3 components (46, 38 ja 37 kDa)</td>
</tr>
<tr>
<td>Non-haemolytic enterotoxin Nhe</td>
<td>3 components (45, 39 ja 105 kDa)</td>
</tr>
<tr>
<td>Cytotoxin K</td>
<td>35 kDa</td>
</tr>
</tbody>
</table>
3.4.3. *B. cereus* emetic toxin (cereulide)

- A cyclic **dodecadepsipeptid**, (1,2 kDa), which consists of three identical subunits (D-O-Leu-D-Ala-L-O-Val-L-Val)
- Thermoresistant
- Stimulates the *Vagus afferent*-nerve and causes the vomiting reflex
- The cytotoxicity is based on the ability to form pores in the cellular membrane leading to influx of ions (K+) and destruction of the membrane potential
3.5. *Staphylococcus aureus*

- Facultatively anaerobic Gram-positive, catalase negative cocci, tolerates bile acids and 6.5% NaCl, produces yellow pigment
- Present regularly on the skin and on the mucous membrane of the nose (20 – 50% symptomless carriers)
3.5.1. *S. aureus* as a pathogen

- Produces several toxins that cause tissue damage or systemic poisonings
- These include Toxic shock syndrome toxin (TSST-1, associated with an epidemic of serious poisonings due to *S. aureus* grown in a specific brand of tampons), epidermolytic toxins, leucocidins and haemolysins
- Before the time of antibiotics often a cause of fatal septicaemias
- Methicillin resistant *S. aureus* an emerging threat
- Common causes of mastitis in cows
3.5.2. *S. aureus* and food poisonings

- Caused by *S. aureus* enterotoxins formed during the growth of the bacterium in the contaminated food
- Typical contaminated foods are meats, ham, cheese, creams and puddings
- The symptoms start within 1 -6 hours after eating the food, and include vomiting, diarrhoea and abdominal cramps, and cease within 24 hours
3.4.3. Staphylococcal enterotoxins

- At least 11 different enterotoxins: SEA, SEB, SEC, SED, SEE, SEG, SEH, SEI, SEJ, and SEK known (there are additionally three subtypes of SEC)
- SEB (on the right) the best characterized
- The molecular weights 25 – 29 kDa, thermostable and resistant to trypsin
- So called pyrogenic superantigens
- Detection in food based on immunological techniques
3.4.4. Genetics of *S. aureus* virulence

- Both the enterotoxin genes as well as the gene for TSST-1 toxin are associated with "phage-related chromosomal island" or bacterial viruses that have integrated themselves as parts of the bacterial genome
3.5. Pathogenic *Escherichia coli*

- Gram-negative, facultatively anaerobic, oxidase negative rods
- *Escherichia coli* present in the intestines of humans and animals usually as harmless commensals
- Five types of pathogenic *E. coli* are known
  - Enteroaggregative *E. coli* (EAggEC)
  - Enteroinvasive *E. coli* (EIEC)
  - Enteropathogenic *E. coli* (EPEC)
  - Enterotoxigenic *E. coli* (ETEC)
  - Enterohaemorrhagic *E. coli* (EHEC)
3.5.1. EHEC strains

- Relatively acid tolerant (may survive hours at pH of appr. 3)
- Temperature limits for growth 8 – 42 °C
- The prototype strain EC O157:H7 (O and H refer to O and H antigens as with *Salmonella*).
- Like EPEC strains produce attachment-effacement lesions, but affect the large intestine
- Produce Shiga- or Shiga-like enterotoxins (Stx1 and Stx2), which are very similar to those produced by *Shigella dysenteriae* (the causative agent of dysentery)
- The enterotoxins often associated with prophages
3.5.2. Diseases caused by EHEC

• Haemolytic-Uremic syndrome (HUS)
  – Haemolytic anemia, acute renal failure

• Haemorrhagic colitis (HC)
  – Bloody diarrhoea, abdominal cramps, vomiting etc
  – Incubation time appr. 4 days, duration up to 7 days
  – Aftereffects may include Guillain-Barré syndrome (a paralytic disorder of the nervous system)
3.5.3. Typical contaminated foods and sources of contamination

• Meat and meat products, unpasteurized milk, poultry, and occasionally seafood (shrimp and fish)

• Because of the association with meat (beef) cattle is suspected to be a reservoir of EHEC

• Weaned calves have a higher prevalence than adult cattle (underdeveloped rumen)
3.6. *Campylobacter jejuni*

- Gram-negative, microaerophilic intestinal bacterium, common in poultry
- Can cause severe enteritis via contaminated water and food
- Guillain-Barré syndrome affecting the peripheral nervous system may also occur
4. Water-associated pathogenic viruses

- Norovirus
  - RNA viruses belonging to the family *Calicoviridae*
  - The most common cause of water borne outbreaks in many countries
  - Persons with O-blood group specially susceptible
  - Detection by PCR

- Hepatitis A
  - A single-stranded RNA-virus belonging to the family of *Picornaviridae*
  - Infection via polluted water and seafood

- Hepatitis A
5. Food/water-associated protozoa

- **Giardia**
  - Anaerobic, flagellated protozoa, with infectious cysts as survival forms
  - Giardiasis may last 2-4 weeks with various gastrointestinal symptoms (diarrhoea, cramps, nausea)
  - "Nokia-outbreak" in Finland a couple of years ago

- **Cryptomonospora**
  - Forms oocysts as survival forms
  - Infects especially (but not exclusively) HIV-positive persons
6. Multicellular parasites
6.1. Tapeworms (*Cestoda*)

- Flat, segmented worms (sizes vary between few mm to several meters) inhabiting the intestinal tract or internal organs
- Important families *Diphyllolotrium* (*D. latum* on right) *Echinococcus*, *Taenia*
- Prevention of *D. latum*
  - High salt content
  - Sufficient heat treatment
  - Freezing < 20 °C for 24 h
- *Taenia saginata*, cows intermediate hosts, situation in Kazakhstan?
6. Multicellular parasites

6.2. Nematodes (Nematoda)

- Fish-mediated parasites Anisakis simplex (right) and Pseudoterranovia decipiensi

- Trichinella causes a severe but luckily relatively rare human disease (infestation of muscles)
  - Bad quality meat (pork, beef, game etc) source of infection
7. Moulds (filamentous fungi)

- Moulds are a very large and heterogenous group of fungi, infesting cereals, nuts, beans and peas, fruit, juices and jams, dairy products etc.
- Mycotoxins, or toxic secondary metabolites are very common and can cause a health hazard to humans or animals
- Tens of different mycotoxins are known; most important ones are aflatoxins, ochratoxins, and different *Fusarium* toxins.
7. Moulds (filamentous fungi)

7.1. The most common mycotoxin producers

- The most common mycotoxin producers belong to genera *Penicillium*, *Aspergillus* (top right) and *Fusarium* (bottom right)
- The form of conidiophores or structures containing the asexual spores an important taxonomic criterion
7. Moulds (filamentous fungi)

7.2. Some important mycotoxins of *Penicillium* and *Aspergillus*

- Aflatoxins are haepatotoxic and carcinogenic mycotoxins produced by *Aspergillus*
- Several types, $B_1$ the most toxic, $M_1$ the metabolite found in milk
- Ochratoxins are kidneytoxic (pigs specially vulnerable), genotoxic and potentially carcinogenic toxins produced by *Penicillium* and *Aspergillus* species
7. Moulds (filamentous fungi)

7.2. Fusarium- toxins

- Trichotecenes
  - T2 toxin (top right), deoxynivalenol (DON)
  - T2 toxin causes damage to skin and mucous membranes, DON liver toxic (animals specially sensitive)
- Zearalenone (middle right)
  - Hormone-like activity
- Fumonisins (bottom right)
  - Very common in maize
  - Potentially carcinogenic
  - Horses very sensitive
8. Algal toxins

- Produced by dinoflagellates and cyanobacteria
- Marine algal toxins (such as saxatoxin, right) can accumulate in the food chain and cause food poisonings (clams and shellfish!)
9. Prion diseases (Bovine Spongiform Encephalitis)

- Bovine spongiform encephalitis (BSE, "mad cow disease") is a prion disease caused by faulty folding of certain proteins associated with the bovine central nervous system.
- The use of tissues from slaughtered animals as feed started an epidemic in cows in the UK in 1990’s.
- Also humans, who had consumed meat of diseased animals developed a similar type of disease (Creutzfeld-Jacob Disease); 28 people died of this condition in the UK in 2000.
- The incident triggered in the EU very strict control measures to contain the disease, and they apparently have been successful.
9. Conclusions: Protect and survive

• Most food/water-borne microbial and parasitic risks can be managed with relatively simple actions
  – Good personal hygiene (”wash your hands each time you visit the toilet”)
  – Proper handling of the foodstuffs (cold chain, heat treatments, avoidance of cross contamination)
  – Responsible primary production (animal health and welfare)
  – Good practices in food manufacturing (HACCP-system!!!)
References


5. Introduction to the Microbiology of Food Processing Small Plant News Guidebook Series United States Department of Agriculture Food Safety and Inspection Service/

Food Legislation in the European Union (EU)

1. What is the EU?
1.1. The European Union (EU) – Facts and Figures

- 27 member states, 17 of which share the common currency (euro, €)
- Population appr. 495 million
- GDP (€12,710 billion in 2010)
1.2. The Brief History of the EU

- Started as "The European Coal and Steel Community in 1950
- Gradually deepening political and economic cooperation (Maastricht treaty in 1993 a landmark of the modern EU)
- Rapid expansion with many new member states since 1990’ies
- The aim to ensure the free movement of goods, capital and people in the EU
- Not a Federal State, not a federation of states, but something between
1.3. Decision making in the EU

• The main actors:
  – European Council
  – Council of the European Union
  – European Parliament
  – European Commission

• The Justus Lipsius Building in Brussels, Belgium, the seat of the Council of the European Union
1.3.1. European Council

- Convenes 2-4 times per year
- Consists of heads of states or governments
- Decides on the political direction and priorities of the EU
- Donald Tusk the current President
- No legislative powers
1.3.2. Council of the European Union

- Consists of national ministers of the member states
- Legislative powers together with the European Parliament
- Directs the economic and foreign policy of the EU
- Decision made by Qualified Majority (see the figure on the right)
1.3.3. European Parliament

- Directly elected by voters every 5 years
- The members do not represent their countries but political parties and groups
- Pass the European laws jointly with the Council of the European Union
- Scrutinizes the working of the Commission
- Decides (together with the Council) on the EU budget
1.3.4. Commission

- The executive body ("European Government")
- One Commissioner from each of the member states (not national representatives), each responsible for a specific policy area
- The current President Jean-Claude Juncker
- The only EU body that can initiate new legislation
- The Commission has several departments or Directorates - General (GD)
- The most important DG in matters related to food and food safety is the DG for Health and Food Safety (SANTE)
1.3.4.2. Standing Committee on the Food Chain and Animal Health (SCFCAH)

- Assists SANCO (and Commission) in the decision making
- Consists of national representatives of the member states
- Decision by Qualified Majority (as in the Council of European Union)
1.3.4.3. SANTE-Associated Agencies

- European Food Safety Authority
- European Medicines Agency
- European Centre for Disease Control
- Community Plant Variety Office
- Executive Agency for Health & Consumer
1.3.4.4. European Food Safety Authority (EFSA)

- Established in 2002 to conduct independent risk assessment in matters related to food and feed
- Situated in Parma, Italy
- The risk assessment is done by Scientific Panels consisting of known experts in the specific aspects of food and feed safety

EFSA Scientific Panels

- Risk assessment and scientific assistance
- Animal health and welfare (AHAW Panel)
- Biological hazards (BIOHAZ Panel)
- Biological monitoring
- Contaminants (CONTAM Panel)
- Dietary and chemical monitoring
- Plant health (PLH Panel)
- Scientific assessment support
- Scientific evaluation of regulated products
- Feed (FEEDAP Panel)
- Nutrition (NDA Panel)
- Food ingredients and packaging (ANS Panel, CEF Panel)
- GMO (GMO Panel)
- Pesticides (PPR Panel)
1.4. Landmarks in the EU Legislation on Food Safety

• The White Paper on Food Safety (2000)
• Initiatives:
  – To establish EFSA
  – To harmonize the EU food legislation
• Subsequently a solid body of EU laws on food safety have been passed, and will be discussed during the other lectures
1.5. The legal instruments of the EU

Directives
- A Directive requires member states to achieve a particular legislative goal
- It does not stipulate, how this goal is achieved
  - Each member state can introduce the requirements of a directive into her own legislation according to her own legal traditions and practices

Regulations
- A Regulation becomes immediately enforceable as law in all member states simultaneously
References

5. Introduction to the Microbiology of Food Processing Small Plant News Guidebook Series United States Department of Agriculture Food Safety and Inspection Service/
Training course for undergraduates (Syllabus)
on the 2015 - 2016 academic year in the discipline
"Hygiene and microbiology of food products "

1. General information

 Faculty Technologies and Bioresources
 Code and name of the specialty 06M073500 - Food safety
 Course, semester 1 year, 1st semester
 Cycle of discipline Elective
 Number of credits 2
 Place of employment 226 aud. 203aud.
 Lecturer Atte von Wright, Dr., Professor
 Teachers Atte von Wright, Dr., Professor

2. Prerequisites and postrequisites

Prerequisites: Microbiology
Postrequisites: Implementation of the HACCP system for food businesses,
 risk management, genetically modified foods

3. Course Objectives

The aim of study - to give students an understanding of the causes of food safety

Lectures
1. The genetics of bacteria and their phages
2. Elements of Food Microbiology
3. The Taxonomy and Physiology of Fungi
4. Prokaryotic Cell and Bacterial Taxonomy
5. Food risks and hazards related to microorganisms and parasites
6. Food Legislation in the European Union (EU)

Seminars
1. Meat processing hygiene – 4 hour
2. Fermented meat products - 4 hour
3. Microbiological investigations of Halal butchery products and butcher’ premises – 4 hour
4. Impact of Halal and Non-halal Slaughtering on the Microbiological Characteristics of Broiler
Chicken Meat and Sausages – 4 hour

As a result of studying the course graduate must:
• Be aware of risk classification;
• learn how to conduct a quantitative risk assessment;
• be able to conduct a sensitivity analysis, verification of stability, the construction of a
 simulation model;
• be able to calculate probabilistic risk criteria;
• be able to make recommendations to reduce the risk;
• Master praktiches-kiwi techniques to inform management decisions under conditions of
uncertainty and risk.

4. Distribution of working time student

<table>
<thead>
<tr>
<th>In total, (volume)</th>
<th>Classroom training</th>
<th>Field work -90 h.</th>
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</thead>
<tbody>
<tr>
<td></td>
<td>Lectures</td>
<td>Laboratory</td>
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<tr>
<td>2 credits</td>
<td>50 min/ clas.</td>
<td>100 min/ clas.</td>
</tr>
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</table>
5. Course Description:

6 Politics of course
- Do not miss classes without good reason, in the absence of a valid reason for more than 3 classes, you need to work;
- Actively participate in the learning process;
- Perform all tasks at a sufficient level and deliver them in a timely manner;
- All intermediate certification must pass in time, or to work;
- Must perform independent work as a single project.

7. References
5. Introduction to the Microbiology of Food Processing Small Plant News Guidebook Series United States Department of Agriculture Food Safety and Inspection Service.

8 Timetable for implementation and delivery tasks in the discipline

<table>
<thead>
<tr>
<th>Types of Work</th>
<th>Items</th>
<th>rating</th>
<th>percentage %</th>
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<tbody>
<tr>
<td>Class work, including .:</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Lecture</td>
<td>visit, note-taking activity</td>
<td>100</td>
<td>100</td>
</tr>
<tr>
<td>Practical / seminars</td>
<td>visit, activity, homework, calculation and graphic tasks, etc.</td>
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<td>100</td>
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</table>

Independent wook

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<th>Schedule (weeks)</th>
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</thead>
<tbody>
<tr>
<td>Proposal</td>
<td>issuing assignments</td>
<td>delivery of the work</td>
<td>Credit s</td>
</tr>
<tr>
<td></td>
<td>1st week</td>
<td>2nd week</td>
<td>100</td>
</tr>
</tbody>
</table>

The final control (exam)
In the of study, oral, 26 questions

The final grade

The final grade of the student in each discipline is given by:
where ИО - the final score of the discipline;  
PK1 and PK2 - landmark points for control 1 and 2;  
Э - Assessment Exam (on a 100-point scale).  
The final grade is put in the gradebook and student transcript in alphabetical and numerical terms.

Alphabetic system of evaluation of educational achievements of students

<table>
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<tr>
<th>A summary of letter system</th>
<th>Digital equivalent points</th>
<th>% content (points on a 100-point scale)</th>
<th>Based on the traditional system</th>
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<tbody>
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<tr>
<td>A-</td>
<td>3,67</td>
<td>90-94</td>
<td></td>
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<tr>
<td>B+</td>
<td>3,33</td>
<td>85-89</td>
<td>good</td>
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<tr>
<td>B</td>
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