

# ENHANCEMENT OF NUTRITIONAL QUALITY AND SAFETY IN RABBIT MEAT

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## ABSTRACT

Nutritional value and safety have gained great importance among the factors that determine meat quality. The close relationship between diet and health has led to changes in consumer habits, demanding products that meet their dietary and nutritional preferences. Rabbit meat is highly valued for its nutritional and dietary properties; it is a lean meat with a low-fat content and less saturated fatty acids and cholesterol than other meats. In this review, a short description of the nutritional value of rabbit meat is provided and the role of rabbit meat as a functional food is discussed. Rabbit meat consumption could be a good way to provide bioactive compounds to human consumers, since manipulation of rabbit's diet is very effective in increasing the levels of  $\omega$ 3 PUFA, CLA or Vitamin E. An overview of the possibilities to incorporate these bioactive compounds in rabbit meat is offered. Food safety is an important and essential aspect for consumers, especially on the meat sector since numerous crises have recently affected meat production. Bovine Spongiform Encephalitis (BSE) in bovine, high dioxin levels in chicken meat, the danger of increased spread of other infectious disease (e.g. Foot and Mouth disease, Avian influenza, etc.), as well as pathogens such as *Salmonella*, *Listeria monocytogens*, *Campylobacter* and *Escherichia coli* 0157:H7 have recently contaminated the European livestock and meat chains. Microbiological quality, presence of chemical residues, animal identification and traceability issues as well as the hazard analysis and critical control points system in rabbit meat production is discussed.

**Key words:** Meat, Nutritional quality, Safety, Rabbit.

## INTRODUCTION

Meat quality has been traditionally determined by sensory aspects, as their appearance, texture, as well as its aroma and flavour. However, at present the nutritional value and safety have gained great importance among the factors that determine meat quality. The nutritional role of meat is controversial because consumers generally consider that high ingestion of meat contributes to excess fat, cholesterol and saturated fatty acids, which are strongly linked to obesity and cardiovascular problems. The close relationship between diet and health has led to changes in consumer habits, demanding products that meet their dietary and nutritional preferences. Rabbit meat is highly valued for its nutritional and dietary properties; it is a lean meat with a low-fat content and less saturated fatty acids and cholesterol than other meats. Moreover, different strategies can be followed to increase beneficial meat components in order to obtain healthier rabbit meat, as will be discussed later. Food safety is an important and essential aspect for consumers, especially on the meat sector where numerous crises have recently affected meat production. These crises have led to a decline in consumer confidence and in subsequent economic losses for the meat industry. Microbiological quality, presence of chemical residues, animal identification and traceability issues, as well as the hazard analysis and critical control points system in rabbit meat production will be discussed.

## NUTRITIONAL QUALITY OF RABBIT MEAT

Recently, the nutritional value of rabbit meat has been reviewed by several authors (Combes, 2004; Dalle Zotte, 2004; Combes and Dalle Zotte, 2005; Hernández and Gondret, 2006), showing that rabbit meat has a high nutritional value compared with other meats.

The main components of meat, excluding water, are proteins and lipids. Rabbit meat is a lean meat rich in proteins of a high biological value and it is characterized by high levels of essential amino-acids (Dalle Zotte, 2004). Furthermore, meat is also an important source of highly available micronutrients, such as vitamins and minerals. Also, rabbit meat does not contain uric acid and has a low content of purines (Hernández, 2007). The information available on chemical composition of rabbit meat is extremely variable, especially regarding fat content, depending on the part of the carcass studied (Pla *et al.*, 2004) and also on the different productive factors (Dalle Zotte, 2002), especially feeding factors having a strong influence on the chemical composition of rabbit meat, in particular, on its lipid composition.

Rabbit meat is characterized by its lower energetic value compared with red meats (Dalle Zotte, 2004) due to its low fat content. Fat content varies widely depending of the carcass portion from 0.6 to 14.4% (fat from edible meat with intramuscular and intermuscular fat content) with an average value of 6.8% (Hernández and Gondret, 2006) with the loin being the leanest part of the carcass (1.2% of lipids). Fatty acid composition of rabbit meat is characterized by high polyunsaturated fatty acid content. The fatty acid composition of rabbit meat and its possible modification through diet will be discussed later on. The amount of cholesterol in rabbit meat is about 59 mg/100 g of muscle (Combes, 2004), lower values than those presented in meat from other species (61 mg in pork, 70 mg in beef, 81 mg in chicken) (Dalle Zotte, 2004).

The mineral fraction of rabbit meat is characterized by its low contents in sodium (49 and 37 mg/100 g for hind leg and loin, respectively) and iron (1.3 and 1.1 mg/100 g for hind leg and loin, respectively), while the phosphorus level is high (230 and 222 mg/100 g for hind leg and loin, respectively; Combes, 2004). Rabbit meat has a low zinc concentration (0.55 mg/100 g) and the copper concentration is quite similar to the meat of other species (0.03 mg/100 g) (Lombardi-Boccia *et al.*, 2005). Selenium levels in rabbit meat depend on diet, reported to vary between 9 µg/100 g (Díaz-Alarcon *et al.*, 1996) and 22 µg/100 g (Wiesner *et al.*, 1978).

Meat is an important source of B vitamins. Consumption of 100 g of rabbit meat contributes to 8% of daily Vitamin B<sub>2</sub>, 12% of Vitamin B<sub>5</sub>, 21% of Vitamin B<sub>6</sub>, and 77% of Vitamin B<sub>3</sub> requirements, and provides a fulfillment of the daily Vitamin B<sub>12</sub> requirement (Combes 2004). However, heat treatments alter Vitamin B contents (Lombardi-Boccia *et al.*, 2005). In addition, rabbit meat, as is true of other meats, contains only trace amounts of Vitamin A. Nevertheless, it should be noted that a high amount of this vitamin can be found in rabbit edible liver (Ismail *et al.*, 1992). Extra supplementation of Vitamin E in the diet (200 mg/kg) to improve the oxidative stability of the meat has led to an increase of almost 50% of Vitamin E in rabbit meat (Castellini *et al.*, 2000). Also, Vitamin E content is not affected by cooking treatment (Dal Bosco *et al.*, 2001).

## ENHANCEMENT OF NUTRITIONAL QUALITY OF RABBIT MEAT

### Rabbit meat and its role as functional food

In recent years, much attention has been paid to the influence of diet on human health and well-being. The primary role of diet is to provide sufficient nutrients to meet the nutritional requirements of an individual. There is now increasing scientific evidence to support the hypothesis that some foods and food components have beneficial physiological and psychological effects over and above the provision of the basic nutrients (Jones and Jew, 2007). Many traditional foods contain components with potential

health benefits. In addition to these foods, new foods are being developed to enhance or incorporate these beneficial components due to their health benefits or desirable physiological effects.

Consumer interest in the relationship between diet and health has increased the demand for information about functional foods. No universally accepted definition for functional foods exists. The International Life Science Institute (ILSI Europe) has proposed the following definition for functional foods: “A food can be regarded as functional if it is satisfactorily demonstrated to affect beneficially one or more target functions in the body, beyond adequate nutritional effects, in a way that is relevant to either an improved state of health and well-being and/or reduction of risk of disease. Functional foods must remain foods and they must demonstrate their effects in amounts that can normally be expected to be consumed in the diet: they are not pills or capsules, but part of a normal food pattern” (Diplock *et al.*, 1999). A functional food can be a natural food, a food to which a component has been added, or a food from which a component has been removed by technological or biotechnological means. It can also be a food where the nature of one or more components has been modified, or a food in which the bioavailability of one or more components has been modified, or any combination of these possibilities.

The nutritive value of meat has an increasing importance among the factors determining meat quality and consumer acceptability. Indeed, meat is a major source of proteins and essential amino-acids; it is a source of group B vitamins, minerals, and other bioactive compounds. However, meat is also a major source of saturated fatty acids and cholesterol and its consumption could be related to cardiovascular diseases, hypertension, obesity and diabetes (Valsta *et al.*, 2005). However, different strategies can be effectively used to increase or reduce bioactive compounds in order to produce functional meat and meat products (see Jiménez-Colmenero *et al.*, 2006 for a review). Rabbit meat, as it has been previously discussed, is a lean meat rich in proteins of high biological values, with highly unsaturated lipids, low cholesterol content, and noticeable quantities of linolenic fatty acid (C18:3  $\omega$ 3). Also, it displays a low content of sodium and a high content of phosphorus, and can be a good source of B vitamins (Hernández and Gondret, 2006). Most research conducted in recent years on rabbit meat quality has focused on incorporating bioactive compounds in meat for the benefit of human health. Moreover, rabbit meat consumption could become a good way to provide these bioactive compounds to human consumers, since manipulation of rabbit's diet is very effective in increasing the levels of  $\omega$ 3 PUFA (Hernández *et al.*, 2007; Nuchi *et al.*, 2007), CLA (Corino *et al.* 2002 and 2003), or Vitamin E (Castellini *et al.*, 1999). In addition, both selenium and iron are also responsive to dietary supplementation (Lynch and Kerry, 2000). These possibilities will be discussed in this review.

### Fatty acid composition

As Hernández and Gondret (2006) state, rabbit meat fat comprises mostly saturated fatty acids (SFAs) and polyunsaturated fatty acids (PUFAs), with percentages around, 36.9%, and 34.6% of total fatty acids in the hind leg, respectively. Monounsaturated fatty acids (MUFAs) are less represented (about 28.5%) (Table 1). The most ubiquitous fatty acids are oleic (C18:1), palmitic (C16:0), and linoleic (C18:2) acids, showing percentages higher than 20% of total fatty acids. Altogether, rabbit meat has a high ratio of PUFA to SAT fatty acids (0.75 and 0.85 for the loin and the meat of hind leg, respectively; Alasnier *et al.*, 1996; Ramírez *et al.*, 2005). Among the PUFAs, linoleic (C18:2) and linolenic (C18:3) are essential fatty acids because animal organisms are unable to synthesize them. Linoleic acid is the precursor of  $\omega$ 6 family of PUFA, while linolenic acid serves the same function for the  $\omega$ 3 family, especially for eicosapentaenoic (EPA) and docosahexaenoic (DHA) fatty acids. A minimum intake of combined EPA and DHA of 500 mg/day is recommended for human cardiovascular health (ISSFAL, 2004). The amount of linoleic fatty acid is about ten times greater in rabbit meat than in beef and lamb and around double than the quantity reported for pork meat (Enser *et al.*, 1996). The amount of linolenic acid is also remarkably abundant in rabbit meat (3%, Hernández and Gondret, 2006) in comparison with those reported in other meats (1.37 in lamb, 0.70 in beef and 0.95 in pork; Enser *et al.*, 1996). However, rabbit meat has a very low amount of EPA and DHA (Ramírez *et al.*, 2005). The  $\omega$ 6: $\omega$ 3 ratio reaches high values, 7 (Dal Bosco *et al.* 2004) or 11 (Ramírez *et al.*, 2005) for the loin and the meat of hind leg, respectively. Therefore, increasing  $\omega$ 3 fatty acid and

consequently decreasing the  $\omega 6:\omega 3$  ratio up to 5 is an interesting goal to improve the nutritional value of rabbit meat for human benefits (ISSFAL, 2004).

**Table 1:** Least squares means and standard errors of fatty acids content in rabbit leg meat (mg/100 g of meat)

Fatty acids	Mean $\pm$ s.e.
C10:0 (Capric)	3.19 $\pm$ 1.01
C12:0 (Lauric)	6.27 $\pm$ 0.68
C14:0 (myristic)	67.1 $\pm$ 2.82
C16:0 (palmitic)	712 $\pm$ 24.6
C16:1 <i>cis</i> $\omega 7$ (palmitoleic)	78.0 $\pm$ 5.16
C16:1 <i>cis</i> $\omega 9$	9.36 $\pm$ 0.36
C17:0 (Margaric)	16.9 $\pm$ 0.63
C17:1 (Heptadecenoic)	6.74 $\pm$ 0.58
C18:0 (stearic)	185 $\pm$ 5.88
C18:1 $\omega 9$ (oleic)	635 $\pm$ 24.3
C18:1 $\omega 7$ (vaccenic)	34.9 $\pm$ 1.32
C18:2 $\omega 6$ (linoleic)	777 $\pm$ 33.2
C18:3 $\omega 3$ ( $\alpha$ -linolenic)	81.2 $\pm$ 4.81
C20:1 (icosaenoic)	9.96 $\pm$ 0.73
C20:2 $\omega 6$ (eicosadienoic)	12.8 $\pm$ 0.58
C20:3 $\omega 6$ (eicosatrienoic)	6.68 $\pm$ 0.54
C20:4 $\omega 6$ (arachidonic)	45.4 $\pm$ 1.24

Adapted from Ramírez *et al.*, 2005

It is well known that rabbits, and other non-ruminants, are able to incorporate dietary fatty acids into adipose and muscle tissue lipids. Therefore, fatty acid composition is strongly affected by dietary lipid composition. The effects of various dietary fat sources have been the subject of many experiments in recent years. The addition of vegetable fat compared to animal fat sources in the diet leads to differences in rabbit meat quality, especially regarding fatty acid composition of the tissues and meat flavour (Oliver *et al.*, 1997; Hernández *et al.*, 2000). For instance, sensory test panels attributed a higher “liver” taste to animals fed with an animal diet, while meats of animals fed with a vegetable diet had a higher “aniseed” or “grass” flavour. However, no differences between groups were found for the texture parameters evaluated.

Different vegetable oil sources have first been used in rabbit diets to increase the level of lipid unsaturation (see Dalle Zotte, 2002 for a review). The dietary use of linseed oil has been proposed by many authors as a way to raise the content of  $\omega 3$  PUFA (Bernardini *et al.*, 1999; Dal Bosco *et al.*, 2004; Colin *et al.*, 2005; Hernández *et al.*, 2007). Dal Bosco *et al.* (2004) studied the synergistic effect of dietary  $\alpha$ -linolenic acid and vitamin E on the oxidative stability and nutritional and eating characteristics of fresh and stored rabbit meat. This study confirmed the ability of rabbit to synthesize long chain PUFA (EPA and DHA) from the dietary precursor, leading to an increase in  $\omega 3$  PUFA content of the meat of rabbits consuming the  $\omega 3$  diet, without any alteration of oxidative stability and sensory quality of the meat. Hernández *et al.* (2007) studied the fatty acid composition of rabbit leg meat from animals fed with three diets enriched with 3% animal fat, 3% linseed oil and 3% sunflower oil, respectively. These authors found higher percentages of linolenic and linoleic fatty acids in the meat of the animals fed with the diet enriched with linseed oil and sunflower oil, respectively, than in the animals fed with the diet enriched with animal fat. An increase of the long chain polyunsaturated fatty acid (EPA and DHA) as well as a decrease of the  $\omega 6:\omega 3$  ratio (mean value of 1.31) was also achieved with the diet enriched with linseed oil. Recently, some studies pointed out the possibility of manipulating  $\omega 3$  PUFA content in rabbit carcasses during the early growth period (Castellini *et al.*, 2004; Muñoz *et al.*, 2004). These studies showed that dietary maternal  $\omega 3$  PUFA are secreted in the milk, allowing  $\omega 3$  enrichment in the young tissues.

Grass-based diets can also modify fatty acid composition of rabbit meat. Forrester-Anderson *et al.* (2006) suggested that grass-based diets fed to rabbits reared outdoors on pasture altered the fatty acid profile, enhancing  $\omega 3$  fatty acid content. Pla *et al.* (2007) also found that meat of hind-leg of organic

source rabbits was poorer in monounsaturated and richer in polyunsaturated fatty acids than meat from conventional rabbits. Other strategies for specifically increasing long chain fatty acids such as EPA and DHA are based on the dietary use of fish oils or algae (Bryhni *et al.*, 2002 in pigs; Bou *et al.*, 2004 in chicken; Nute *et al.*, 2007 in lamb). A high increase of long chain polyunsaturated fatty acids can be achieved feeding rabbits with diets enriched with fish oils. However, high levels of lipid oxidation (Nuchi *et al.*, 2007), lower growth and impair carcass and meat quality, depending of the fish oil used (Navarrete *et al.*, 2007).

Lipid oxidation is a major non-microbial factor responsible for the quality deterioration of muscle foods. It leads to discoloration, higher drip-loss, and the development of off-odours and off-flavours (Monahan, 2000). The dietary manipulation of tissue lipid composition to produce meat with a high content of PUFA could reduce the oxidative stability of meat-products and have negative effects on meat quality. The level of oxidation after refrigerated storage of rabbit leg meat has been studied by Hernández *et al.* (2008). These authors found that oxidation products evaluated by measuring peroxide value (PV) and TBARS (TBA-reactive substances) were not very high in rabbit meat, although both oxidation parameters increase with storage time. However, when rabbits were fed with a diet enriched with 3% linseed oil, rabbit meat showed higher TBARS values after 5 *post-mortem* days of refrigerated storage (Hernández *et al.*, 2007), although values were well below for apparent rancidity (Campo *et al.*, 2006). Nevertheless, diets were supplemented with 100 ppm of  $\alpha$ -tocopherol. Dietary inclusion of 8% linseed produced higher susceptibility to lipid oxidation (TBARS) of both fresh and frozen (-20 °C for 3 or 6 months) meat batters for hamburger production (Bianchi *et al.*, 2006).

### ~~Conjugated linolenic fatty acid~~

Conjugated ~~linolenic fatty~~ acid (CLA) consists of a group of geometric and positional isomers of linoleic acid. It has potential nutritional benefits for humans, because it has anti-obesity (Lin *et al.*, 1995) and anti-carcinogenic activities (Ip *et al.*, 1996); it is also able to ameliorate diabetes (Housseknecht *et al.*, 1998), and it has a protective effect against atherosclerosis in rabbits (Lee *et al.*, 1994). Naturally occurring CLA originates mainly from bacterial isomerisation or/and biohydrogenation of PUFA in the rumen and the mammary gland (Griinari and Bauman, 1999). Food sources originated from ruminants are known to have markedly higher CLA concentration than those from monogastric animals (Schmid *et al.*, 2006). Rabbits are able to recycle part of the end microbial fermentation products through cecotrophy, so that the CLA retained in their meat might be higher than in other non-ruminant species (Gómez-Conde *et al.*, 2006).

Conjugated linoleic acid has also received a great deal of attention as a supplement in rabbit feed. Dietary CLA supplementation is an effective tool for increasing, in a dose dependent manner, the amount of CLA in intramuscular lipids of rabbits with *cis*-9,*trans*-11 being the predominant isomer (Lo Fiego *et al.*, 2005; Petacchi *et al.*, 2005). In addition to the beneficial effect of CLA in human health, CLA can favorably modify body composition (Corino *et al.*, 2002; 2003) due to its potential to increase lean tissue deposition in various species (Dunshea *et al.*, 2005). Rabbit growth performance and carcass characteristics at standard slaughter weight (2.5 kg, 76 d) were not affected by diets supplemented with 0.25 or 0.5% CLA. However, CLA supplementation reduced perirenal fat weight at heavy slaughter weight (3.1 kg), and lowered concentration of serum triglycerides and total cholesterol (Corino *et al.*, 2002). Regarding the chemical composition of rabbit meat, a significant decrease in meat lipid content was evident only when rabbits fed diet with a high supplementation level of CLA (0.5%) were considered at heavy slaughter weight (3.1 kg) (Corino *et al.*, 2003).

### Antioxidants

There has been an increasing interest in the use of antioxidants in rabbit feed formulas because the dietary manipulation of tissue lipid composition to produce meat with a high PUFA content could decrease meat oxidative stability.

Hernández and Gondret (2006) reviewed the use of vitamin E in the diet. Vitamin E is commonly used in animal feeds as an indispensable component of biological membranes with stabilizing properties and a high antioxidant activity. Vitamin E is the generic term used to describe at least eight naturally occurring compounds that exhibit the biological activity of  $\alpha$ -tocopherol (Morrissey *et al.*, 2000). In recent years, different studies have examined the effects of dietary extra supplementation with vitamin E on the deposition of  $\alpha$ -tocopherol in tissues, on meat quality characteristics, and on oxidative stability and the shelf life of rabbit meat. Several authors (López-Bote *et al.*, 1997; Castellini *et al.*, 1999) have shown that the deposition of  $\alpha$ -tocopherol in rabbit muscle is very efficient and has a strong relationship with the supplementation level used in the diet. Dietary  $\alpha$ -tocopheryl acetate supplementation has been found to stabilize color of raw meat (Corino *et al.*, 1999), even after refrigerated storage (Dalle Zotte *et al.*, 2000). Vitamin E has also been effective in reducing lipid oxidation during refrigerated and frozen storage of meat (Castellini *et al.*, 1999; Lo Fiego *et al.*, 2004). In addition, Vitamin E supplementation increases the oxidative stability of cooked rabbit meat (Castellini *et al.*, 1999), whatever the different cooking methods studied (Dal Bosco *et al.*, 2001). Also, a high  $\alpha$ -tocopherol level improves some physical traits of meat, reducing shear values and increasing water holding capacity (Castellini *et al.*, 1998). The effect of dietary synergetic supplementation of Vitamins C and E have been also investigated, leading to an increase in the vitamin content and reducing the oxidation of the lipids (Castellini *et al.*, 2000; Lo Fiego *et al.*, 2004). Different natural ways to improve the oxidative stability of rabbit meat have also been studied. For example, rabbit lipid oxidative stability was improved by increasing the level of oats in rabbit diet (López-Bote *et al.*, 1998). Coni *et al.* (2000) also verified the antioxidant efficiency of extra virgin olive oil and oleuropein, an olive oil biophenol, in rabbit plasma and isolated low density lipoproteins (LDL). However, it seems that oleuropein did not reduce meat susceptibility to oxidation (Paci *et al.*, 2001).

Meat contains several natural antioxidants such as catalase, superoxide dismutase and glutathione peroxidase (GSH-Px). Studies on meat of several species (Pradhan *et al.*, 2000) indicate that endogenous antioxidant enzymes could potentially delay the onset of oxidative rancidity in refrigerated stored meat. Indeed, GSH-Px could have an important role controlling lipid oxidation due to its high activity in rabbit meat when compared to other species (Hernández *et al.*, 2002). There are other endogenous antioxidants such as histidine-containing dipeptides, carnosine and anserine, but contents vary according to anatomical location and species (Decker, 2000).

## SAFETY OF RABBIT MEAT

The safety of meat has been at the forefront of societal concerns in recent years. Numerous crises including Bovine Spongiform Encephalitis (BSE) in bovine, high dioxin levels in chicken, the danger of increased spread of other infectious disease (e.g. Foot and Mouth disease, Avian influenza, etc.), as well as pathogens, such as *Salmonella*, *Listeria monocytogens*, *Campylobacter* and *Escherichia coli* 0157:H7, have recently contaminated the European livestock and meat chains. Major meat safety issues and related challenges include microbial pathogens, food additives and chemical residues, and animal identification and traceability (Sofos, 2008).

### Microbiological quality of rabbit meat

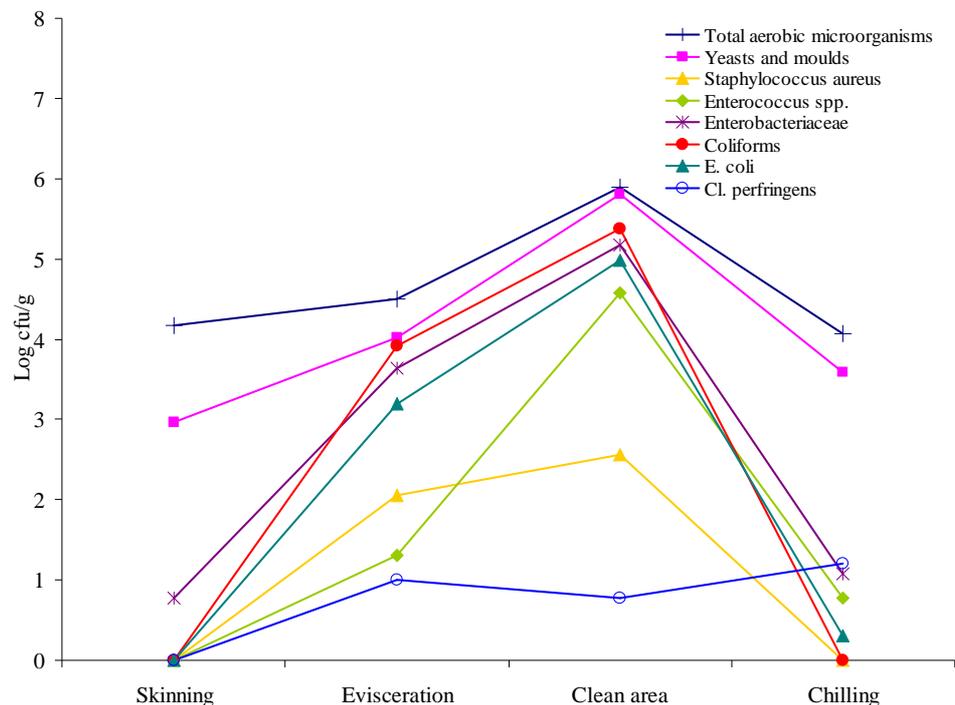
Safety and shelf life of meat are limited by microbial growth. Dominant contaminants on carcasses and packed rabbit meat are *Pseudomonas*, lactic acid bacteria, yeasts and *Brochothrix thermosphacta*, according to Rodríguez-Calleja *et al.* (2004) with total bacteria counts between 4.01-4.96 log cfu/g. It is established that microbial levels of 6-7 log cfu/g are critical for the spoilage of meat. Rodríguez-Calleja *et al.* (2005a) studied the self life of rabbit carcasses, overwrapped with oxygen-permeable film and stored at 3°C, over 8 days. Shelf life according to both appearance and odor was estimated at 6.8 days reaching the aerobic plate counts values of 8 log cfu/g. In fact, after 5 days of storage, most of the carcasses already showed some softening and the counts of these bacteria were about 7 log cfu/g. Other authors estimated shelf life of rabbit carcasses in 3 days at 4°C (Bobbitt, 2002). These

differences could be explained by differences in initial microbial counts, since a high initial contamination of meat reduces product shelf life (Gil *et al.*, 1998). In addition, it is possible to increase the shelf life of rabbit meat using modified atmospheres (Berruga *et al.*, 2005) or irradiation (Badr, 2004; Rodríguez-Calleja *et al.*, 2005b).

Microbial ecology of rabbit meat could also be affected by different feeding programs; some components of the feed could play a specific role on the growth rate of some microbial groups. Vannini *et al.* (2003) showed that a dietary supplementation of whole linseeds limited the growth rate of several microbial groups (except psychrotrophic bacteria) with a consequent increase in meat shelf life. In addition, dehydrated alfalfa meal at high percentages in the diet seems to also have an inhibiting effect on microbial growth in rabbit meat products (Vannini *et al.*, 2002).

The slaughtering process may cause extensive contamination of muscle tissue with a vast range of micro-organisms. Some of these micro-organisms come from the animal intestinal tract and others from the environment in contact with the animals before or during slaughter. López *et al.* (2002) have studied the evolution of the most important contaminant and pathogen biota on carcasses during the slaughter process of rabbits (Figure 1).

These authors found that there was an increase of micro-organism counts during evisceration process, especially enteric micro-organisms, so an improvement of this process is required in order to reduce the final contamination of carcasses. After carcass chilling, microbial counts were reduced to a great extent. Nevertheless, final counts of total aerobic micro-organisms and yeasts and moulds were still high. *Listeria monocytogenes*, *Salmonella spp.* and *Campylobacter spp.* were not found in all steps of the slaughter process. *Staphylococcus aureus* was present during evisceration. However, after chilling this micro-organism was not present.



**Figure 1:** Evolution of microbial counts during rabbit slaughter process (from Hernández and Gondret, 2006)

### Chemical contaminants

Foods of animal origin play an important role in determining the exposure of human beings to contaminants either of biological or chemical origin. Chemical contaminants are compounds that have a potential risk for consumers. They are present in relatively small amounts, usually due to human

activities. In vivo, the main ways in which animals can accumulate harmful substances are poisons and the addition or voluntary administration of substances with pharmacological action (antibiotics, compounds with hormonal action and tranquilizers) or insecticides. An accidental contamination of food can be produced; water and air can be contaminated by heavy metals, halogenated hydrocarbons and other persistent pesticides, or by cleaners and disinfectants. Moreover, pollution or harmful substances can be formed during meat processing and storage.

POPs is the acronym for a group of persistent organic pollutants framed within the Stockholm Convention (Stockholm Convention on Persistent Organic Convention (POPs) 2004; available from <http://www.pops.int/>). These pollutants represent a relevant and growing concern for human beings due to their physical-chemical properties, bioaccumulative behaviour in lipid tissues, and possible toxicological effects (Brambilla *et al.*, 2008).

Recently, the presence of some of these POPs, such as dioxins, has been studied in rabbit (Ábalos *et al.*, 2007). In fact, a European Project, entitled “Feeding Fats Safety” (Food-CT-2004-07020) was developed in the 6<sup>th</sup> EC Framework Program. The aim of this project was to study different aspects of animal nutrition in order to preserve animal health and to produce safe and good quality meat products. Rabbits and chickens were the species used in this project. The ability of different fats to modify the lipid composition of meat, the effect of the level of fat oxidation and the toxicological or physiological effects of certain fat degradation compounds and undesirable contaminants (dioxins, polycyclic aromatic hydrocarbons) have been studied. Ábalos *et al.* (2007) studied the presence of PCDD/Fs (polychlorodibenzo-*p*-dioxins and dibenzofurans) and DL-PCBs (“dioxin-like” polychlorinated biphenyls) in rabbit and chicken meat samples from animals fed fish oil spiked feed at different levels of contaminants. Three different levels of contaminants under the maximum quantity allowed by the EU Directive were tested. The profile of PCDD/Fs in chicken samples from the three different treatments resembled the profile previously observed in the corresponding feeds, in general, the levels of the different compounds increased when increasing their amount in the feed. However, in rabbit meat samples, completely different bioaccumulation behavior was observed when compared to chicken samples. The profile of PCDD/Fs in rabbit meat did not correspond to that in present feeds. In fact, there were no significant differences in PCDD/F toxicity among rabbit samples from the three different treatments. For DL-PCBs, the profile was similar between feeds and meat samples, both in chickens and rabbits.

The level of polycyclic aromatic hydrocarbons (PAHs) in rabbit tissues and their rate of transfer from feed has also been studied by Devier and Budzinski (2007) in the frame of the European Project: “Feeding Fats Safety” (Food-CT-2004-07020). PAHs are a group of chemicals that are formed during the incomplete burning of coal, oil, gas, wood, garbage, or other organic substances, such as tobacco and charbroiled meat. There are more than 100 different PAHs. Some of them are known or suspected carcinogens, and are linked to other health problems. In this study, rabbits were fed with diets containing three levels of PAHs (153, 81 and 32 ng PAHs/g ww). The most interesting results were that PAH metabolites were not transferred from feed to meat, liver or plasma, even with highly contaminated feeds, being excreted in urine and bile.

The presence of heavy metals (Cu, Pb, Cd and Hg) in rabbit meat was studied by Skřivanová *et al.* (2002b). These authors pointed out that negligible concentration of heavy metals was found in rabbit meat, concluding that rabbit meat produced in Czech Republic was safe from the point of view of the heavy metal environmental presence. Skřivanová *et al.* (2001) showed that feeding supplemental copper at 150 mg/kg (far over the amount allowed by the EU Directive) improved performance and decreased mortality of rabbits. However, the copper was accumulated in liver, although no contamination of meat was observed (Skřivanová *et al.*, 2002a).

Antibiotics are a special case within the subject of chemical contaminants. The use of antibiotics in animal production has led to a great reduction of the infectious diseases of the animals and a consequent reduction of the risk of transmission of infectious agents to consumers. Therefore, the use of antibiotics has led to an increase in the safety of the food chain. However, the presence of these

residues in meat could be harmful to consumers because antibiotic residues in low concentrations encourage the development of microbial resistance, which could have serious consequences (Chander *et al.*, 2007). Antibiotics should be removed in time to ensure their elimination from the tissues. The legislation for pharmaceutical and veterinary products established a withdrawal period of antibiotics of 28 days for fattening rabbits, limiting the addition of antibiotics to the first days of fattening. The use of antimicrobials in rabbit production has been reviewed by Badiola *et al.* (2007).

### **Animal identification and traceability issues**

Traceability is the more comprehensive concept of tracking the movement of identifiable products through the marketing chain. The main objective of traceability is to minimize any adverse health effects by a quick and complete recall. For an adequate recall, it is necessary that all food products and all of the ingredients used in producing the food are traceable at all stages of production, both processing and distribution. A review of rabbit meat traceability was provided by Cavani and Petracci (2006). Animal identification is an effective tool for meat traceability. Current livestock identification systems (ear-tags, tattoos, etc.) are not completely efficient and offer certain inconveniences. Transponders could be a way of monitoring live rabbits to improve traceability systems if its cost is reduced in the future (Crimella *et al.*, 2005). For example, the spread and then containment of BSE in various countries made the issue of animal identification and traceability a reality and was an important challenge for its adoption by various organizations and health authorities. Animal identification and meat traceability are not themselves an issue of food safety, animal disease prevention, quality assurance, or country-of-origin labelling. However, they may be important components of such programs. The need for traceability has also arisen by consumer concerns over food authenticity. It is well known that organic foods, protected designation of origin (PDO) and protected geographical indication (PGI) products and other labels are steadily gaining in popularity. Other categories that consumers are demanding is traditional, regional, and handmade products, which are claimed to be safer because they are more natural. If one wants to keep consumers' confidence by use of such labels, the early detection of any kind of fraud must be made possible.

### **Hazard Analysis and Critical Control Points**

The Hazard Analysis and Critical Control Points system (HACCP) is a preventive system used to safely produce food in the agricultural sector. This system is based on prevention rather than inspection. The HACCP principles are applicable to all stages of production, including the production of raw materials, preparation, handling, processing, and distribution and consumption system (Brown *et al.*, 2000).

HACCP is based on the following seven principles:

- Principle 1: Conduct a hazard analysis.
- Principle 2: Determine the critical control points (CCPs).
- Principle 3: Establish critical limits for each critical control point.
- Principle 4: Establish critical control point monitoring procedures.
- Principle 5: Establish corrective actions.
- Principle 6: Establish verification procedures.
- Principle 7: Establish record-keeping and documentation procedures.

One of the main critical control points in the slaughter of rabbits is the evisceration since it can produce gastrointestinal tract ruptures, producing an important microbiological contamination of the carcass. There is some information about HACCP system for slaughterhouses in rabbits (Tantiñá *et al.* 2000). In table 2, the main stages in rabbit slaughter and the potential hazards associated are listed.

**Table 2:** Main stages and associated hazards in the slaughter of rabbits

Stage	Hazards
<b>Ante mortem</b>	
Animal production (farms)	1. Infectious and parasitic diseases 2. Chemical residues 3. Mechanical injuries 4. Injuries when rabbits are introduced in cages
Loading, transportation, reception and unloading	5. Deficiencies in transport: trucks, cages, density, travel
Ante mortem inspection	6. Deficient inspection
Stunning and hanging	7. Incorrect immobilization and stunning 8. Incorrect hanging
<b>Dirty area: slaughter, skinning, evisceration</b>	
Bleeding	9. Incomplete severing of blood vessels 10. Incomplete bleeding
Feet and ear removal and skinning	11. Carcass contamination by deficient handling practices and deficient cleaning of utensils and automated cutting machines
Evisceration	12. Fecal material release due to rupture of gastrointestinal tract (microbiological contamination)
Skinning of hind legs	13. Carcass contamination by deficient handling practices and deficient cleaning of utensils and automated cutting machines
<b>Clean area</b>	
Post-mortem inspection	14. Deficient inspection
Refrigeration	15. Insufficient refrigeration
Packaging	16. Carcass contamination by deficient handling practices

Adapted from Tantiñá et al. (2000).

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