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Enterobacteriaceae in Pork Meat: Causal Agents of Public Health Problems

Enterobacterias en Carne de Cerdo: Agentes Causales de Problemas de Salud Pública

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SUMMARY

Pork meat is one of the most consumed products worldwide, and pathogenic microorganisms in pork, such as Enterobacteriaceae, represent a public health risk, causing *foodborne diseases*. Enterobacteriaceae in pork meat processing is an indicator of poor sanitation management. *Escherichia coli* (*E. coli*) and *Salmonella* spp. are the most prevalent bacteria. Different studies report that their high percentage and the multidrug resistance found are an alarming risk to consumers´ health.

Keywords: Enterobacteriaceae, Pork, Public health.

RESUMEN

La carne de cerdo es uno de los productos más consumidos a nivel mundial, y los microorganismos patógenos en la carne de cerdo, como las Enterobacterias, representan un riesgo para la salud pública, provocando *enfermedades de transmisión alimentaria*. Las Enterobacterias en el procesamiento de la carne de cerdo son un indicador de una gestión sanitaria deficiente. *Escherichia coli* (*E. coli*) y *Salmonella* spp. son las bacterias más prevalentes. Diferentes estudios indican que su porcentaje alto y la multifarmaco-resistencia son un riesgo alarmante para la salud de los consumidores.

Palabras clave: Enterobacterias, Carne de cerdo, Salud pública.

INTRODUCTION

Meat is one of the main food sources for the human population worldwide. It is an important source of protein, essential amino acids, zinc, iron, phosphorus, and vitamin B, providing benefits to adults (cell repair and regeneration) and children and adolescents (growth and development) (Bohrer, 2017). The consumption of meat is in high demand globally. However, consumption varies depending on the region due to the influence of various factors such as gender, rural or urban origin, educational level, and age of consumers (Estevez-Moreno *et al*., 2021). Another important factor is income, which in the case of the Mexican population, is a determining aspect influencing the selection of chicken, pork, or beef meat (Huerta-Sanabria *et al*., 2018).

The production and consumption of chicken, pork, beef, lamb, and mutton have grown, reaching record highs in 2021, with an annual meat production and consumption of 335,275 and 334,975, respectively. Mexico is among the countries with the highest meat protein production and consumption. Chicken meat is the most consumed (4,166 tons), followed by pork (2,217 tons) and beef (1,723 tons) (OECD-FAO, 2022).

On the other hand, one of the world's biggest problems facing meat production and distribution is its susceptibility to bacterial contamination. This situation is related to deficiencies in hygiene and poor handling practices during processing and distribution, which cause public health problems such as gastroenteritis and diarrhea, among others. The Center for Disease Control and Prevention (CDC) reports more than 250 foodborne diseases (FBD), mainly caused by bacteria (CDC, 2021). Microbial spread in meat occurs during the slaughtering process, mainly Enterobacteriaceae, a large family of gram-negative bacteria, such as *Salmonella*, *Escherichia coli* (*E. coli*), *Klebsiella pneumoniae* (*K. pneumoniae*), and *Yersinia*, among others (Rönnqvist *et al*., 2018; Peruzy *et al*., 2021). Bacteria, such as *Staphylococcus aureus* (*S. aureus*), *Pseudomonas*, *Brochothrix*, *Carnobacterium photobacterium* (*C. photobacterium*), *Listeria*, among others, have also been frequently detected in meat processing (Peruzy *et al*., 2019).

Enterobacteriaceae cause FBD (Mladenović *et al*., 2021), mainly gastrointestinal illnesses (Guzmán *et al*., 2017). In recent years there has been infections with strains resistant to one or more antibiotics, which has complicated treatment, aggravating the disease, and even leading to the death of patients. For this reason, the World Health Organization (WHO) considers bacterial or multidrug resistance as one of the ten main health problems worldwide (WHO, 2019).

Different techniques have been established to detect Enterobacteriaceae, such as traditional microbiological methods involving bacterial colony counting and molecular techniques, such as the polymerase chain reaction (PCR), for rapid and more accurate detection (Cauchie *et al*., 2020). Several studies have shown that Enterobacteriaceae are a recurrent problem in the production and commercialization of meat products, representing an important public health risk. Therefore, this study analyzed the presence of Enterobacteriaceae in pork meat and its potential risk as a causal agent of diseases.

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We searched scientific articles in the PubMed, ScienceDirect, and Google Scholar databases for information and data on food safety issues in pork meat, regarding their production and consumption, bacterial pathogens presence, the prevalence of Enterobacteriaceae, and their impact on public health. The search was restricted to publications in English and Spanish. The literature review was carried out from March to July of 2022. The literature search was undertaken using the keywords: Enterobacteriaceae, meat, pork, antimicrobial resistance, and public health.

Prevalence of Enterobacteriaceae in pork meat

Several studies have demonstrated the presence of Enterobacteriaceae in pork meat samples (Table 1). In the United States, *E. coli* was detected with a prevalence of 12 % and *Salmonella* spp. with almost 6 % (Mollenkopf *et al*., 2011); years later, *E. coli* was reported with a value of 18 % (Scheinberg *et al*., 2017). While, in the northeast from Mexico, in the state of Tamaulipas, an investigation reported *E. coli* contamination in pork meat with a high prevalence (50 %) (Martínez *et al*., 2018).

Salmonella spp. had a prevalence of 32 % in Romania (Mihaiu *et al*., 2014), and 56 % (Arcos-Ávila *et al*., 2013), and 71.4 % in Colombia (Rondón-Barragán *et al*., 2015). Salmonella has been detected with a prevalence of 22 % in samples of ground pork in the central region from Mexico (Villalpando-Guzmán *et al*., 2016). A recent study in southern Brazil detected *Salmonella enterica* (*S. enterica*) in this same type of meat with a prevalence of 17 % (Kich *et al*., 2020). In contrast to these results, in Mexico city reported *Salmonella* spp. with a prevalence of only 2 % (Gutiérrez *et al*., 2020).

Antimicrobial resistance in pork meat

Due to the high manufacture and administration of antibiotics in the production process of livestock animals (in 2010, more than 63,000 tons were used, and by 2030, more than 105,000 tons is estimated), antimicrobial resistance has rapidly increased and become a global public health threat (Van Boeckel *et al*., 2015; Elshamy and Aboshanab, 2020).

The presence of antibiotic-resistant bacteria in pork meat is a serious problem. Bacterial strains with resistance to different groups of antibiotics are shown in Table 2. In Europe, *E. coli* and *Salmonell*a strains multiresistant to β-lactams were detected in samples from a processing company in Germany, that was supplied by slaughterhouses from Poland, Belgium, and Spain. Therefore, the authors suggested that the cause of such resistance is due to a very extensive processing and distribution chain (Schill *et al.*, 2017). In Latin America, *E. coli* strains resistant to five antibiotics (ampicillin, tetracycline, nalidixic acid, chloramphenicol, and cotrimoxazole) were isolated in pork samples from markets in Lima, Peru (Ruiz-Roldán *et al*., 2018).

Impact on public health

According to WHO estimates, in 2015, FBD (caused by bacteria, viruses, parasites, toxins, and chemicals) triggered various outbreaks worldwide, sickening more than 600 million people annually and killing around 420,000. On the American continent, more than 77 million people fall ill annually, and around 9,000 die from consuming contaminated food. Among the food pathogens that endanger health, the Enterobacteriaceae responsible for diseases are mainly *Salmonella* (salmonellosis) and *E. coli*, which cause gastrointestinal problems such as nausea, vomiting, abdominal pain, and diarrhea. Other signs are fever and headache (WHO, 2018; Nastasijevic *et al*., 2020).

Enterobacteriaceae infections from pork meat consumption represent a serious worldwide health problem for consumers. In the United States, chicken and pork meat consumption cause most salmonellosis infections (Bonardi,

Tabla 1. Prevalencia de Enterobacterias en carne de cerdo de diferentes países.
Table 1. Prevalence of Enterobacteriaceae in pork from different countries.

Bacteria	Prevalence (%)	Type of sample	Place of origin	Country
E. coli	12.2	Meat	Retail markets	United States (Mollenkopf *et al*., 2011)
Salmonella spp.	5.8			
Salmonella spp.	32.21	Meat	Production and retail markets	Romania (Mihaiu *et al*., 2014)
Salmonella spp.	56.0	Meat	Slaughter	Colombia (Arcos-Ávila *et al*., 2013)
Salmonella spp.	71.4	Meat	Slaughter	Colombia (Rondón-Barragán *et al*., 2015)
Salmonella spp.	22.5	Ground beef	Retail markets	Mexico (Villalpando-Guzmán *et al*., 2016)
E. coli	18	Meat	Slaughter	United States (Scheinberg, 2017)
E. coli	50.8	Ground beef	Retail markets	Mexico (Martínez *et al*., 2018)
Salmonella spp.	2.7	Tenderloin	Retail markets	Mexico (Gutiérrez *et al.,* 2020)
Salmonella enterica	17.2	Meat	Slaughter	Brazil (Kich *et al*., 2020)

Tabla 2. Resistencia antimicrobiana de Enterobacterias provenientes de carne de cerdo.
Table 2. Antimicrobial resistance of Enterobacteriaceae from pork.

Bacteria	Resistant to	Country
E. coli	GEN, TGC, OFX, LEV.	Slovakia (Gajdošová *et al*., 2011)
E. coli	AMC, AMP, PIP, CEC, COX, CXM, IMP, APR, GEN, NEO, SPT, STR, TOB, CMP, CIP, ENR, COL, DOX, CXT.	Germania (Schwaiger *et al*., 2012)
E. coli Proteus vulgaris Klebsiella pneumoniae Enterobacter cloacae	AMP, PIP, CAZ, CXM, CTX.	Spain (Ojer-Usoz *et al*., 2013)
Salmonella spp.	TET, FFC, AMP, CMP, AMC, EFT, STX, TMP, GEN, CIP.	Colombia (Bermúdez & Rincón 2014)
Salmonella spp.	SXT, STR, SPT, TET, AMP, TMP	Thailand (Sinwat *et al*., 2015)
E. coli	AMP, AMC, CXT, CMP, STR, KAN, GEN, SXT, TMP, TET, CIP.	Czech Republic (Skočková *et al*., 2015)
E. coli	CXT	Cuba (Marrero-Moreno *et al*., 2017)
E. coli Escherichia fergusonii E. cloacae Proteus mirabalis P. vulgaris	GEN, CEP, CTX, CIP, AMP, CAZ, CMP. AMP CTX, CAZ, AMP. CEP, CTX, TGC, AMP, CMP, COL. CEP, TGC, AMP, CMP, COL.	Germany (Schill *et al*., 2017)
E. coli	AMP, TET, NAL, CIP, CMP.	Peru (Ruiz-Roldán *et al*., 2018)
E. coli	AMP, AMC, PIP, EPF, TET, GEN, TOB.	Thailand (Lugsomya *et al*., 2018)

β-lactamicos: amoxicilina (AMC), ampicilina (AMP), piperacilina (PIP), cefaclor (CEC), cefoxitina (COX), cefuroxima (CXM), imipenem (IMP), cefotaxima (CXT), ceftiofur (EFT), cefalosporina (CEP). **Cloranfenicol**: cloranfenicol (CMP) **Aminoglicosidos**: apramicina (APR), gentamicina (GEN), neomicina (NEO), espectinomicina (SPT), estreptomicina (STR), tobramicina (TOB), kanamicina (KAN), florfenicol (FFC). **Fluoroquinolones**: ciprofloxacina (CIP), enrofloxacina (ENR), doxiciclina (DOX). **Antimetabolitos**: sulfametoxazol (SXT), trimetoprima (TMP). **Tetracliclinas**: tetracliclina (TET). **Quinolonas**: lovofoxacina (LEV), ofloxacina (OFX), ácido nalidíxico (NAL). **Glicilciclina**: tigeciclina (TGC). **Ureidopenicilinas**: ceftazidime (CAZ). **Polimixina**: colistina (COL).

β-lactams: amoxicillin (AMC), ampicillin (AMP), piperacillin (PIP), cefaclor (CEC), cefoxitin (COX), cefuroxime (CXM), imipenem (IMP), cefotaxime (CXT), ceftiofur (EFT), cephalosporin (CEP). **Chloramphenicol**: chloramphenicol (CMP). **Aminoglycosides**: apramycin (APR), gentamicin (GEN), neomycin (NEO), spectinomycin (SPT), streptomycin (STR), tobramycin (TOB), kanamycin (KAN), florfenicol (FFC). **Fluoroquinolones**: ciprofloxacin (CIP), enrofloxacin (ENR), doxycycline (DOX). **Antimetabolites**: sulfamethoxazole (SXT), trimethoprim (TMP). **Tetracycliclines**: tetracyclicline (TET). **Quinolones**: lovofoxacin (LEV), ofloxacin (OFX), nalidixic acid (NAL). **Glycylcycline**: tigecycline (TGC). **Ureidopenicillins**: ceftazidime (CAZ). **Polymyxin**: colistin (COL).

2017). According to Tran *et al*. (2018), *Salmonella enterica* (*S. enterica*) and some *E. coli* strains from pigs are major intestinal pathogens. In Germany, public health authorities investigated salmonellosis outbreaks in 2013 and 2014. Based on a trace-back analysis, they detected *Salmonella muenchen* (*S. muenchen*) in a pig breeding farm and considered it a probable source of contamination. The investigation suggested that intoxications were caused by consuming raw pork and pork products, traditional foods in some regions of Germany (Schielke *et al*., 2017).

In The Netherlands, a study reported a strong association between salmonellosis and pork, considering that consuming pork contaminated with *Salmonella* increases the risk of intoxication in people treated with medications such as antibiotics or antacids. Additionally, salmonellosis contamination can also occur in workers from pig farms and slaughterhouses (Berends *et al*., 1998).

An investigation by Hernandez *et al.* (2011) showed data on salmonellosis cases (paratyphoid and other *salmonella*-derived diseases) in Mexico from 2000-2008. In 2000, 10,000 cases were reported, although, in the same year, they began to decrease; however, from 2002 to 2007, the cases increased to 12,000 and slightly decreased in 2008. In the case of shigellosis, by the end of 1999, there were 40,000 cases, with a significant decrease in the 2000-2008 period, reaching 10,000 cases; however, this number was still considered high according to those reported in the epidemiology bulletin of the Mexican Republic.

Good handling practices

Good handling practices and sanitary surveillance are important during the different stages of the production and distribution chain, to avoid pork meat contamination with bacterial pathogens harmful to humans (Tang *et al*., 2017). Meat can be contaminated from its origin due to animal diseases, medication residues, or when the microbial flora of the slaughtered animal comes into contact with the meat. It can also be contaminated by surfaces, equipment, utensils, water, and even the hands of workers (Nerin *et al*., 2016).

Slaughterhouses represent a strategic control point for achieving meat product safety (COFEPRIS, 2017, in Spanish). In the case of Mexico, the National Health, Safety and Food

Quality Service (SENASICA, in Spanish) and the Agriculture and Rural Development Secretariat (SADER, in Spanish), have as their objective the reduction in agricultural and livestock risks, among others, and to keep the surveillance of food contamination and agri-food quality of import and export products under strict control.

Breeding and animal care stage: At the start of pig breeding, feeding plays an important role in avoiding risks to animal and human health. In Mexico, the Official Mexican Standard, NOM-061-ZOO-1999, which states the "Animal health specifications of food products for human consumption", indicates which finished food products should be used for animal consumption. In the care of animals intended for human consumption, overexposure to antibiotics has potential adverse effects through direct toxicity to consumers and the generation of microbial resistance (Chen *et al*., 2019). Due to the presence of antimicrobial-resistant bacteria, the use of antimicrobials for animal growth promotion was banned in Europe and the United States, particularly those classified as critically important for treating human infections (Iriti *et al*., 2020). In Mexico, the specifications for the use of antibiotics in animals are described in the Official Mexican Standard NOM-064-ZOO-2000, "Guidelines for the classification and prescription of veterinary pharmaceutical products by the level of risk of their active ingredients." It establishes the technical and scientific criteria for the active ingredients used in the formulation of veterinary pharmaceutical products to avoid toxic effects on animals.

Slaughter and distribution stage: The pig slaughter process is commonly carried out in Federal Inspection Type (TIF, in Spanish) slaughterhouses, Health Secretariat Type (TSS, in Spanish) municipal slaughterhouses, or private slaughterhouses. TIF slaughterhouses are subject to stricter standards of hygiene regulation and cold chain integrity and are mainly used by large meat companies (OECD, 2018). However, the three slaughter sites must comply with the sanitary regulations for handling animal products stipulated in the NOM-194-SSA1-2004 standard, which has as its main objective the definition of the sanitary specifications that establishments must comply with if engaged in the slaughter and preparation of animals for supply, storage, transport, and sale of their products. In addition, meat product transportation and distribution chains must comply with NOM-024-ZOO-1995. This standard mentions the specifications and zoosanitary characteristics for the transportation of animals, their products, and by-products, and chemical, pharmaceutical, biological, and food products for use in animals or consumption by them. To this end, the Federal Commission for the Protection against Sanitary Risks (COFEPRIS, in Spanish) monitors the strategy for evaluating sanitary risks and management actions in slaughterhouses to reduce risks in meat products.

CONCLUSION

The increase in the worldwide production and consumption of pork meat in recent years have acquired greater relevance for the food industry because the industry must provide food free of pathogenic microorganisms. Enterobacteriaceae are important because they spread easily and contaminate pork meat, causing gastrointestinal diseases. In this study, about the prevalence of Enterobacteriaceae in pork meat shown that *Salmonella* and *Escherichia* are frequently reported in slaughterhouses and retail markets. Unfortunately, both generous of strains are reported with antimicrobial resistance to several groups of antibiotics, such as β-lactams, aminoglycosides, fluoroquinolones, and chloramphenicol. Therefore, the responsible use of antibiotics in veterinary swine care practices should adhere to the provisions of good management practices manuals and current regulations aimed at the proper use of antibiotics during the raising of animals for human consumption.

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