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Antimicrobial Resistance of Bacteria in Food

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1. Introduction

Antibiotics are a major tool utilized by the healthcare industry to fight bacterial infections; however, bacteria are highly adaptable organisms, able to develop resistance to antibiotics. Consequently, decades of antibiotic use, or rather misuse, have resulted in bacterial resistance to many modern antibiotics. This resistance can cause significant danger and suffering for many people with common bacterial infections, which were once easily treatable with this type of medication [1]. Antibiotics are widely used in human and veterinary medicine as well as in agriculture for the treatment of infections, to improve growth and for animal prophylaxis, which can generate a selection of multiresistant bacteria. However, it is not fully understood how widespread antibiotic-resistant bacteria are in agricultural settings. The lack of such surveillance data is especially evident in dairy farm environments.

Over the past 6 decades, the introduction of new classes or modifications of antimicrobials has been marched slowly but surely by the development of new bacterial resistance mechanisms. Since the first reports, different studies have demonstrated that increases in antimicrobial resistance among both pathogenic and commensal bacteria can be observed after introduction of antimicrobials [2]. Therefore in this chapter I will discuss some of the research in which they reported the presence of antibiotic-resistant bacteria that are of importance in foods.

2. *Campylobacter*

Campylobacter was identified as a human diarrheal pathogen in 1973. *Campylobacter* is a major cause of disease in humans and poultry around the world and *Campylobacter* was, is the most frequently diagnosed bacterial cause for human gastroenteritis in the United States and throughout the world. Most cases of *Campylobacter* infections do not require antimicrobial treatment, being clinically mild and self-limiting [4]. Macrolides are considered the first choice drug for *C. jejuni* and *C. coli* enteritis, erythromycin and ciprofloxacin are the drugs of choice for treatment of human campylobacteriosis and fluoroquinolones are also used. Contaminated food is the usual source of human infection; therefore, the presence of antimicrobial-resistant strains in the food chain has raised concerns that the treatment of human infections will be compromised. Most disease in humans is associated with the consumption of contaminated poultry or cross-contamination with other foods [4].

This section provides a review of resistance prevalence in *C. jejuni* and *C. coli* from food. In this study, was investigated the prevalence of resistance to erythromycin and ciprofloxacin

in *Campylobacter* isolates recovered from turkey carcasses at two processing plants. Ciprofloxacin and erythromycin resistance in *Campylobacter* recovered from processed turkey occurred more frequently among *C. coli* than *C. jejuni*. Molecular subtyping in this study provides further information about the relationships between antimicrobial-resistant *Campylobacter* at processing level [5]

The antimicrobial resistance profiles of *Campylobacter* isolates recovered from a series of samples of retail food (n = 374) and humans (n = 314) to eight antimicrobial compounds were investigated. High levels of resistance in isolates of *C. jejuni* were observed for ceftiofur (58%), ampicillin (25%) and nalidixic acid (17%) with lowest levels observed for streptomycin (7.9%) and chloramphenicol (8.3%). A total of 80% of isolates of *C. jejuni* were resistant to human ceftiofur, while 17% were resistant to ampicillin and nalidixic acid, 8.6% to streptomycin and 4.1% to chloramphenicol. Antimicrobial resistance of clinical relevance, such as erythromycin, ciprofloxacin and tetracycline were 6.7, 12 and 15% respectively for all food isolates and was similar to the corresponding prevalence of resistance observed in human isolates, where 6, 4%, 12 and 13, respectively, were found to be resistant. Comparisons of strains of *C. jejuni* at each site showed a high degree of similarity although some regional variations exist. Comparison of the total populations of *C. jejuni* and *C. coli* showed minor differences, with *C. jejuni* strains resistant to ampicillin and ceftiofur. Patterns of multidrug resistance showed some profiles common to the human strains and clinical [6].

Antimicrobial resistance was evaluated in *Campylobacter* spp isolated of beef cattle in four commercial feedlots in Alberta (Canada). All calves were given chlortetracycline and oxytetracycline in food, and most animals (93%) were injected with long-acting oxytetracycline. A total of 1586 *Campylobacter* strains were isolated, these consist of *Campylobacter coli* (n = 154), *Campylobacter fetus* (n = 994), *Campylobacter jejuni* (n = 431), *Campylobacter hyointestinalis* (n = 4), and *Campylobacter lanienae* (n = 3) which were recovered and characterized [4]. Increases in the prevalence of strains resistant to tetracycline and doxycycline (56 to 89%) of *C. coli*, *C. fetus* and *C. jejuni* were observed [4]. Increased resistance to erythromycin was also found in strains of *C. coli* in the three episodes of isolation. Most isolates of *C. fetus* recovered were resistant to nalidixic acid and a relatively small number of multi-drug resistant strains were recovered. Widespread use of antimicrobial agents in meat production and possible horizontal transfer of mobile genetic elements with resistance determinants among bacteria *Campylobacter* and other taxa emphasized [4].

Campylobacter has become the leading cause of zoonotic enteric infections in developed and developing countries worldwide. Epidemiological and microbial studies show that poultry is the most important source for quinolone-susceptible and quinolone-resistant *Campylobacter* infections in humans. Trend over time for macrolide resistance show stable low rates in most countries, and macrolides should remain the drugs class of choice for *C. jejuni* and *C. coli* enteritis. However, macrolide resistance is emerging in some countries and needs to be monitored.[7].

3. *Salmonella*

Salmonella spp. is widely distributed in nature, colonizing a range of animal hosts. *Salmonella* entérica is recognized as one of the most common bacteria causes of food borne

diarrheal illness worldwide. It had been estimated that annually there are about 1.3 billion cases of acute gastroenteritis due to nontyphoidal salmonellosis, resulting in 3 million deaths. In industrialized countries food animals are the main reservoir for human infections, the majority of which originate from contaminated meat products and eggs. It is very important the issue of antibiotic resistance of *Salmonella* spp, which has been investigated as its ecology and pathogenesis

Substantial effort has been made to disclose the genetic means by which *Salmonella* spp has evolved to resist antimicrobials. Acquired resistance arises by two ways: by mutations in chromosomally encoded genetic elements and by acquisition of exogenous mobile resistance genes by plasmids, integrons and transposons. Both mechanisms can lead to rapid changes of a bacterial populations, horizontal genes transfer appears to be most important in the evolutions of salmonella resistance.[8] In this section I summarize some examples which show the presence antimicrobial resistance *Salmonella* spp in food.

Burgos et al. isolated and identified enteric bacteria in the soil of dairy farms and found that enteric bacteria from dairy farm soil are resistant to multiple drugs and carriers of antibiotic resistance plasmids. This suggests that the surface layer of farm land plays an important role, as it is an environment that can be a reservoir for the development of bacterial resistance against antibiotics [3].

In another study undertaken in Alberta during 1996 and 1999, 209 strains of *Salmonella*, obtained from food animals were isolated and 17 antimicrobial drugs were tested and , 11.8% of strains were positive for resistance. These strains were commonly resistant to tetracycline (35.4%), streptomycin(32.5%), sulfamethoxazole (28.7%), ticarcillin (27.3%) and ampicillin (26.8%)[9]. *Salmonella* enterica serovar Heidelberg frequently causes foodborne illness in humans. The authors compared the prevalence of *Salmonella* serotype Heidelberg in a sampling of 20,295 meats, including chicken breast ,ground turkey, ground beef and pork ribs, collected between 2002 and 2006 a total of 298 *Salmonella* serovar Heidelberg isolates were recovered, representing 21.6% of all *Salmonella* serovars from retail meats. One hundred seventy-eight (59.7%) were from ground turkey, 110 (36.9%) were from chicken breast, and 10 (3.4%) were from pork chops; none was found in ground beef. One hundred ninety-eight isolates (66.4%) were resistant to at least one compound, and 49 (16.4%) were resistant to at least five compounds. Six strains (2.0%), all ground turkey, were resistant to at least nine antimicrobial agents. The greatest resistance in isolates from poultry was to tetracycline (39.9%), followed by streptomycin (37.8%), sulfamethoxazole (27.7%), gentamicin (25.7%), kanamycin (21.5%), ampicillin (19.8%), amoxicillin-clavulanate (10.4%) and ceftiofur (9.0%). These data indicate that *Salmonella* serovar Heidelberg is a common serovar in retail poultry meat and includes widespread clones of multidrug-resistant strains [10].

Recently, *Salmonella* Enterica subsp. enterica serovar *Saintpaul* has been increasingly observed in several countries, including Germany. However, the pathogenic potential and epidemiology of this serotype are not very well known. Fifty-five isolates of *S. turkey saintpaul* Germany and Turkey food products isolated from 2000 to 2007 were analyzed using an antimicrobial agent, organic solvents, and disinfectant susceptibility testing, detection of determinants of resistance, plasmid profiles, pulsed-field gel electrophoresis (PFGE) and hybridization experiments[11].The pattern of resistance was observed for ampicillin, amoxicillin-clavulanate, gentamicin, kanamycin, nalidixic acid, streptomycin,

spectinomycin, and several third-generation cephalosporins (including ceftiofur and cefoxitin. This study revealed that a multiresistant *S. saintpaul* line Saintpaul is widespread in turkeys and turkey products in Germany.[11]

In Denmark, Skov, M et al, compared 8144 *Salmonella* isolates collected from meat imported or produced, as well as the Danish patients. Isolates from imported meat showed a higher rate of antimicrobial resistance, including resistance to multiple drugs, which were isolated from domestic beef. Isolates from humans showed resistance rates lower than those found in imported meat. These findings suggest that programs to control resistant *Salmonella* spp. are a worldwide problem [12]

A study in Vietnam shows that enteric bacteria in samples of raw foods contain a set of mobile genetic elements and the transfer of antibiotic resistance can easily occur between similar bacteria. This study was undertaken to examine the contamination rate and molecular characteristics of enteric bacteria isolated from a selection of food sources in Vietnam [16]. One hundred and eighty raw food samples were tested; 60.8% of the samples were from meat and 18.0% of samples of shellfish contaminated with *Salmonella* spp. More than 90% of all food sources contained *Escherichia coli*. The isolates were selected for antibiotic resistance against 15 antibiotics, and 50.5% of *Salmonella* isolates and 83.8% of isolates of *E. coli* were resistant to at least one antibiotic[13]. Isolates were screened for the presence of mobile genetic elements that confer resistance to antibiotics. Fifty-seven percent of *E. coli* and 13% of *Salmonella* isolates were found to contain integrons, and some isolates contained two integrons Plasmids were also detected in the 23 *Salmonella* isolates resistant to antibiotics and 33 isolates of *E. coli*. One hundred thirty-five *Salmonella* isolates and 76% of *E. coli* isolates contained plasmids of 95 kb, and some isolates contained two large plasmids. Conjugation experiments showed the successful transfer of all or part of the phenotypes of antibiotic resistance among isolates of *Salmonella* and *E. coli* contaminated food. The results show that enteric bacteria in raw food samples from Vietnam contain a set of mobile genetic elements and the transfer of antibiotic resistance can easily occur between similar bacteria[13]

Another study in Vietnam, was undertaken to examine the levels of *Salmonella* in samples of raw foods, including chicken, beef, pork and shellfish to determine their antibiotic resistance. A total of 180 samples were collected and analyzed, we obtained 91 isolates of *Salmonella*. Sixty-one percent of meat and 18% of shellfish samples were contaminated with *Salmonella* spp. The susceptibility of all isolates to a variety of antimicrobial agents was tested, and resistance to tetracycline, ampicillin / amoxicillin, nalidixic acid and streptomycin sulfafurazole was found in 40.7%, 22.0%, 18.7%, 16.5% and 14.3% of the isolates, respectively. Resistance to enrofloxacin, trimethoprim, chloramphenicol, kanamycin, and gentamicin was also detected (8.8 to 2.2%). About half (50.5%) of the *Salmonella* isolates were resistant to at least one of the antibiotics.[14]

4. *Escherichia coli*

E. coli is a bacterium, which very easily and frequently exchanges genetic information through horizontal gene transfer (e.g. by conjugation, transformation or transduction) with other related bacteria, such as other *E. coli* strains, *Salmonella*, *Shigella*. Therefore, *E. coli* strains may exhibit characteristics that have been acquired from a wide variety of sources.A

recent review describes the population structure of commensal *E. coli*, the factors involved in the spread of different strains, how the bacteria can adapt to different niches, and how a commensal life style can evolve into a pathogenic one (Tenaillon et al., 2010). All humans and animals carry *E. coli* in their intestines as they are part of the normal gut flora and usually harmless. However, there are several types of *E. coli* strains that may cause gastrointestinal illness in humans. These strain types can be divided into several pathogroups. These strain types can be divided into several pathogroups, resistant to ampicillin, amoxicillin/clavulanic acid, piperacillin/sulbactam, piperacillin/tazobactam, cefuroxime, etc. The strain carries plasmid-borne *bla*CTX-M-15 and a *bla*TEM-1 genes. An *E. coli* O104:H4 with a MLST ST678 was previously observed about 10 years ago in Germany in a Haemolytic Uremic Syndrome (HUS) case (Mellmann et al., 2008), the STEC O104:H4 outbreak strain shows an unusual combination of virulence factors of STEC and EAEC which has only been reported sporadically in humans before (Morabito et al., 1998) [16].

Another study analyzed the prevalence of *Escherichia coli* O157 in patients with diarrhea and surface water of some selected sources in Zaria (Nigeria), was evaluated of susceptibility to antibiotics and plasmid profiles of 184 isolates of *E. coli*, obtained from water samples of 228 and 112 diarrheal stool samples (collected from children <15 years) using standard methods. The most active antibiotics were gentamicin, chloramphenicol, and fluoroquinolones. Seventy-nine (42.9%) of 184 *E. coli* isolates were resistant to four or more antibiotics. The Multidrug Resistance (MDR) was higher among water isolates than clinical isolates. Of the 35 MDR isolates (20 of which were O157 strains), 22 (62.9%) harbored plasmids, all of which not less than 2.1 kb in size. Among the 20 strains of *E. coli* O157, only seven (35.0%) contained multiple plasmids. An *E. coli* O157 isolated from the aquatic system contains two plasmids resistant to seven drugs, including ampicillin, cefuroxime, ciprofloxacin, cotrimoxazole, nalidixic acid, nitrofurantoin and tetracycline. Loss of plasmids correlated with the loss of resistance to antibiotics (mutant) strains selected on tetracycline (50 mg / mL) in nutrient agar plates [17].

The role of animal-based foods as vehicles for antibiotic-resistant bacteria has also been studied. One study on chickens fattening evaluated the incidence and distribution of antibiotic resistance in 197 commensal *Escherichia coli* strains. The effects of supplementation with antimicrobial agents approved bambarmycin, penicillin, salinomycin and bacitracin or a combination of salinomycin more bacitracin. All isolates showed some degree of resistance to multiple antibiotics and resistance to tetracycline (68.5%), amoxicillin (61.4%), ceftiofur (51.3%), spectinomycin (47.2%), and sulfonamides (42%). These data demonstrate that the multidrug resistance of *E. coli* can be found in broilers, regardless of antimicrobial growth promoters used.

Water can also be an important vehicle for transmission of bacteria durability antibiotics, so I quote a study that was performed on wastewater from a plant to produce antibiotics which characterized the population of bacteria in surface waters of production plant of oxytetracycline (OTC). Found high levels of TBT in the wastewater (WW) and the antibiotic was still detectable at 20 Km (RWD), with undetectable levels in the water upstream (RWU). A total of 341 bacterial strains, most identified as Gammaproteobacteria. The most of the isolates (94.2% and 95.4% respectively WW and RWD) had tet (A) gene and it was the most common (67.0%), followed by tet (W), tet (C), tet (J), tet (L), tet (D), tet (y) and tet (K) (in the range between 21.0% and 40.6%). The authors propose that the strong selective pressure

imposed by high concentrations of TBT contributes to the widespread dissemination of resistance genes and other genes of tetracycline resistance to antibiotics, possibly through mobile genetic elements[15]

5. *Enterococcus*

Enterococci colonize the gastrointestinal tract of the oral cavity and vaginal tract of humans and most animals [18]. The emergence of antimicrobial resistance represents the greatest threat to the treatment of human enterococcal infections. Enterococci are intrinsically resistant to a number of antimicrobial agents normally used to treat infections caused by gram-positive bacteria. The enterococci have a remarkable ability to acquire new mechanisms of resistance and to transfer resistance determinants by way of conjugation [19]. In this section I focus in a review of antimicrobial resistant enterococci strains isolated from some food.

From a medical point of view, the resistance of enterococci to vancomycin, teicoplanin and streptogramins is of special interest. In the case of vancomycin, there are identical types of groups of *vanA* of enterococci genes from fecal samples of animals, pet food, hospital patients, people in the community and water samples, these resistance genes may contaminate humans through the food chain [20]. Also, it was found that the most frequently isolated species are *Enterococcus faecium* (32.61%), followed by *E. faecalis* (21.74%), with high levels of resistance to streptomycin and gentamicin. These results confirm the presence of enterococci within the community having susceptibility profiles similar to those of strains found in hospital. [21].

A study at the Faculty of Pharmaceutical Sciences, University of Sao Paulo, Brazil, reported that 52.5% of the samples of raw and pasteurized milk, meat, cheese and vegetables were positive for enterococci; the most contaminated being the meat and the cheese. *E. faecium* was the predominant species, followed by *E. faecalis*, *E. gallinarum* and *E. casseliflavus*. Virulence genes were found and resistance to gentamicin, tetracycline and erythromycin in *E. faecalis* and three strains of *E. faecium* were resistant to vancomycin [22]. From the strains resistant to antibiotics, 72.4% of *E. faecalis* were able to form biofilm and 13.8% to adhere to Caco-2, which shows a virulent capacity of these types of enterococci [22].

The importance of ready-to-eat food (RTEF) and the Antibiotic Resistance (AR) gene flow has been assessed. RTEF are consumed frequently and may play a role in the acquisition of the determinants of AR in the human digestive tract. The study by Macovei et al, evaluated three RTEFs (chicken salad, a chicken burger and carrot cake) which were taken as samples from five fast food restaurants five times in the summer and 5 in the and winter. The overall concentrations of enterococci during the two seasons were similar (10^3 CFU / g), the most prevalent were *Enterococcus casseliflavus* (41.5% of isolates) and *Enterococcus hirae* (41.5%) in winter and *Enterococcus faecium* (36.8%), *E. casseliflavus* (27.6%) and *Enterococcus faecalis* (22.4%) in summer. In winter isolates were resistant mainly to tetracycline (50.8%), ciprofloxacin (13.8%) and erythromycin (4.6%). In summer isolates were resistant mainly to tetracycline (22.8%), erythromycin (22.1%), and kanamycin (13.0%). The most common gene was *tet (M)* (35.4%) Genotyping of *E. faecalis* and *E. faecium* with pulsed-field gel electrophoresis revealed that the food contamination likely originated from various sources and is not clonal [23].

Another study from the southwestern United States, which characterized the profiles of antibiotic resistance of enterococci isolated from fresh produce harvested, found that of 185 Enterococci isolates, 97 (52%) were *Enterococcus faecium*, 38 (21%) were *Enterococcus faecalis*, and 50 (27%) were other Enterococcus species. Of clinical significance in humans is the fact that strains of *E. faecium* had a much higher prevalence of resistance to ciprofloxacin, tetracycline and nitrofurantoin than *E. faecalis*. 34% of the strains had multiple patterns of drug resistance, excluding intrinsic resistance. These data may help to elucidate the role of food in the transmission of antibiotic-resistant strains in human populations [24].

Additionally, one hundred and five VanA of the Glycopeptide-resistant enterococci (GRE) isolated from, human; animal, and food, were studied for genetic variability and molecular markers. The presence of indistinguishable *vanA* elements, mostly plasmid-borne, and virulence determinants in different species and PFGE-diverse populations suggested that all GRE might be potential reservoirs of resistance determinants and virulence traits transferable to human-adapted clusters. [25].

Research undertaken in Turkey has assessed vancomycin resistance and antibiotic resistance profiles of enterococci in different types of food purchased in local markets. Of a total of 200 samples, 50% had high levels of enterococci contamination, the greater resistance being found in samples of cream cheese. Only 4 strains were identified as resistant to vancomycin and identified as *E. faecalis*, from chicken. The results of this study emphasize the urgency of preventive measures to be taken to control antibiotic use on farms [26].

In addition, a study led by Martins et. al in Portugal whereby a total of 983 strains of enterococci were isolated from sewage sludge and effluent waste. These were tested against 10 different antibiotics. Multiresistance was found in 49.4% of the strains. Only 3.3% and 0.6% were resistant to ampicillin and vancomycin, respectively. However, observed 51.5% resistance to rifampicin, tetracycline 34.6%, 24.8% and 22.5% for erythromycin, nitrofurantoin. These results indicate that the use of antibiotics has created a large pool of resistance genes and the processes of wastewater treatment do not prevent the spread of resistant enterococci in the environment [27].

Different species of enterococci can frequently be isolated from environmental samples such as soil, water, plants or animal raw products.. In a study led by M.T. Tejedor Junco in 2009, isolated 78 strains of enterococci, from alfalfa (*Medicago sativa*) plant samples, drip irrigated with conventional water and a secondary effluent. *E. faecalis* (10.2%), *E. faecium* (2.6%), *E. hirae* (5.1%), *E. casseliflavus* (2.6%) and *E. mundtii* (79.5%) were isolated, In They found that all strains of enterococci, were susceptible to glycopeptides, penicillin and ampicillin. They did not detect strains with high level resistance to aminoglycosides. [28].

Additionally, the products supplied for feeding animals have been widely studied because they are potential vehicles for transmission of resistant bacteria. This has been demonstrated in a study conducted in 2006 in Portugal where 1137 enterococci strains and 163 *Escherichia coli* strains were recovered from 89 poultry feed samples, where 69.1% of enterococci isolates obtained from broiler feed were resistant to tetracycline and *E. coli* were resistant to ampicillin, tetracycline and streptomycin in 22.9%, 27.6% and 19.0% respectively. These data allow us to infer that the animal feed is a significant source of antibiotic resistant bacteria, thus leading to their introduction in the farm environment. The Poultry feed is at the start of the food safety chain, and might serve as a source of antimicrobial resistant bacteria present in poultry meat [29].

Some antibiotics had been used as growth promoters in Europe for several years, creating strains of resistant *E. faecium*. Resistant bacteria, have been isolated from samples of sewage, animal stools, meat products, samples from community and clinical samples from different European populations. Glycopeptide-resistant *E. faecium* (GREF), can be found in hospitals and outside them, and in the food chain by contaminated meat products. This suggests that the origin of these strains are other sources outside the hospital, probably for commercial ranching. Thus, to prevent the spread of antibiotic resistant strains such as enterococci, or transferable resistance genes, prudent use of antibiotics is necessary in human medicine and veterinary and in the animal husbandry [20].

There are no available studies about strains of enterococci resistant to antibiotics, isolated from Colombian food. It has been reported to a clinical level that the rate of vancomycin resistance in isolates of *Enterococcus* species by 2004 was about 7% in Brazil - Pan-American Health Organization (PAHO). In Colombia the problem is raised to similar levels and is a common source of nosocomial infections [1].

6. *Staphylococcus aureus*

S. aureus is one of the most important human and veterinary pathogens, and the epidemiology, pathology and antimicrobial resistance of this bacterium has been studied intensively in innumerable studies. *S. aureus* was one of the first bacteria in which the development of antimicrobial resistance (penicillin) was observed [2]. In human medicine, methicillin resistance is not only observed among *S. aureus*, but is also prevalent among other staphylococci as, *S. intermedius*, *S. epidermidis*, *S. hominis*, etc, have been isolated from animal sources in different studies and have been demonstrated that they strains have mec A gene and are Methicillin resistant bacteria. [4]

In the United States, during 1992–2003, the number of health care-associated infections due to MRSA increased from 35.9% to 64.4% and in UK, death certificates increased by 39% [30]. In Colombia, the CA-MRSA (community-acquired, or community-associated *Staphylococcus aureus*) increased from 1% in 2001 to 5.4% in 2006 [31].

While environmental MRSA transmission has been investigated, transmission through food products has not received enough attention [32, 33]. However, Normmano et al. established the presence of *S. aureus* strains that harboured the mecA gene isolated from food samples such as bovine milk, mozzarella cheese, and pecorino cheese [33]. Other than information from that report the current prevalence of resistant bacteria in food matrices and levels of MRSA consumer exposure risks remain unknown.

Nowadays the assessment of the activity of an antibiotic is crucial to the successful outcome of antimicrobial therapy; however, the development of resistance both in human and animal bacterial pathogens has been associated with the extensive therapeutic use of antimicrobials or with their administration as growth promoters in meat production [34, 35].

In order to achieve the detection of sensitivity or resistance of *Staphylococcus aureus* strains, various techniques have been described, such as the employment of a cefoxitin 30 ug disc, using semiconfluent inoculums and overnight incubation at 35°C, resulting in a sensitivity of 100% and a specificity of 99%. In this way, disc diffusion remains the method of choice for

routine screening for methicillin resistance, when the technical or economic capabilities are absent in the microbiological laboratories [36]

In addition, the most widely used molecular typing method for the study of local and global epidemiologies of MRSA is pulsed field gel electrophoresis (PFGE). This method has been used to identify MRSA clones that have a particular ability to cause major outbreaks [37, 38].

An unpublished study developed by the “Laboratorio de Ecología Microbiana y de Alimentos” (LEMA at the Universidad de los Andes), in Colombia genotyped the MRSA strain detecting *mecA* gene isolated from food samples circulating in Bogotá. Positive strains were genotyped for the identification of clonal groups using pulsed field electrophoresis (PFGE). 5 of the 149 strains were confirmed to have the *mecA* gene, indicating the presence of the SCC cassette. The electrophoretic pattern obtained by PFGE for these strains has revealed that 4 (80%) of the 5 strains belong to the Chilean clone, with 100% genetic similarity; this clone has been associated with 65% of infections associated with health care. This is the first evidence of the presence of MRSA in food in Colombia; nevertheless, this study is not published yet but is in the process of submission to publication[39].

Furthermore, a review of the resistance of gram-positive cocci in Colombia shows how our neighbors, Ecuador and Venezuela have a lower rate of resistance to that identified in our country at that time (25% vs. 47%) for coagulase-negative staphylococci-hospital in 2004 to the Pan American Health Organization (PAHO), where the frequency of *S. aureus* resistant to methicillin is much higher, with maximum values for Bogotá 60% and 70% for the 2001 to 2003 period. [40]. Reyes. J. et al, investigated the resistance profiles and mechanism of macrolide resistance in isolates of *Streptococcus pneumoniae* (1679), *Staphylococcus aureus* (348), coagulase-negative staphylococci (CoNS) (175), and *Enterococcus* spp. (123) from Colombian hospitals. The prevalence of macrolide resistance is low in Colombian pneumococci and high in MRSA (cMLS_B-type). [41].

7. Lactic acid bacteria (LAB)

For several decades, studies on the selection and spread of antibiotic resistance have focused mainly on clinically relevant species. However, recently several researchers have suggested that commensal bacteria such as lactic acid bacteria (LAB) may act as reservoirs of genes resistance to antibiotics similar to those found in human pathogens [42]. The main threat associated with these bacteria is that these resistance genes can be transferred to pathogenic bacteria [43].

Genes that confer resistance to tetracycline, erythromycin and vancomycin have been detected and characterized in *Lactococcus lactis*, enterococci and, recently, in lactobacilli isolated from fermented meat and milk products [1]. One example of the this resistance is the presented by lactobacilli, *pediococci* and *Leuconostoc* spp. which have been reported to have a high natural resistance to vancomycin, a property that is useful to separate them from other Gram-positive bacteria [44].

Thirty-one strains of *Lactobacillus delbrueckii* subsp. *bulgaricus* as components of yoghurt cultures showed intrinsic resistance towards mycostatin, nalidixic acid, neomycin, polymyxin B, trimethoprim, colimycin, sufamethoxazol and sulphonamides. Susceptibilities

to cloxacillin, dihydrostreptomycin, doxycycline, furadantin, novobiocin, oleandomycin, oxacillin and streptomycin were prominent while kanamycin and streptomycin susceptibilities varied [45]. This has raised the discussion of new issues concerning the safety of probiotics in relation to the nature of the procurement and distribution of antimicrobial resistance genes [46].

Starter cultures of *Lactobacillus*, *Weissella* and *Bifidobacterium* of African and European origins were studied for their susceptibility to antimicrobials. Ouoba et al, evaluated and compared on its investigation to 24 antimicrobial, variations were observed and high levels of intrinsic resistance were found among the species studied. These authors confirmed the ability of *Lb. reuteri* from Africa to transfer by conjugation the gene *erm(B)*, (resistance to erythromycin) to enterococci in vitro experiments [47]. Finally, they also identified a higher prevalence of phenotypic resistance to aminoglycosides in isolation from Europe. This is corroborated by recent publications in which they documented the transfer of macrolide resistance in *Enterococcus* from *Lactobacillus* in vivo [48].

Pan L. et al, reported the presence of higher MICs (Minimal Inhibitory Concentration) in 14 of 202 strains of LAC, isolated from Chinese fermented food. 14 strains reported the presence of multi-resistance and the presence of genes *tet (M)* and *erm (B)*, *mefA* and *aphA3* located on plasmids or chromosome. They found that lactic acid bacteria resistant to antibiotics are widespread among traditional Chinese fermented food and the incidence of this resistance was dependent on raw materials and manufacturing area of food; thus, the incidence of LAC resistance isolated during fermentation of sausages is much higher than that presented in the fermentation of vegetables. The results presented in these studies indicate the possible role of LAC as reservoirs for dissemination of antibiotic resistance in food and environment[49].

In Colombia there are some reports about antimicrobial resistance of bacterias responsible for human diseases, but its unknown how frequent is the presence of these bacterias in food and its importance in human illness. The following are some comments about the problem of antimicrobial resistance in Colombia. Currently in Colombia, we have data based on antibiotic resistance of nosocomial strains, usually in intensive care units (ICU). This highlights the lack of a comprehensive study of this aspect in the Industrial and Food Process. Data record of 4008 set out the isolates from ICUs in 2003, 4,004 in 2004 and 4304 in 2005, where the most frequent were, in order: *S. aureus*, *E. coli*, *P. aeruginosa*, *K. pneumoniae*, *A. baumannii* and *E. cloacae*. There was a statistically significant decrease in the number of isolates of *A. baumannii*. Salient issues that expose this study is the high resistance to ciprofloxacin (CIP) from *E. coli*, which is used as a marker, because once a gram-negative bacteria is resistant to this antibiotic it must be considered resistant to other quinolones. The resistance phenotype of this organism to third generation cephalosporins suggests the production of beta-lactamases of extended spectrum (ESBL) during the three year study. [50]. Of all the bacteria studied, *A. baumannii* showed the highest rates of multidrug resistance. Therefore, although there was a significant decrease in the number of isolates of this bacterium, in ICU this is a major concern. Among all prevalent in this study enterobacteria, *E. cloacae* could have the most ability to select resistance, including the carbapenems, to produce very high amounts of AmpC and close porins [50]. However,

recent studies suggest that the prevalence of vancomycin resistance by the isolation of *Enterococcus faecium* found in South America is low (only 6%) compared to that presented in the United States [51]. Finally, according to the findings of this study, it is urgent to continue working with the national surveillance network of the resistance of both the hospital and those pathogens resistant organisms from food or water, in order to assist in the control of this public health problem of great importance [51].

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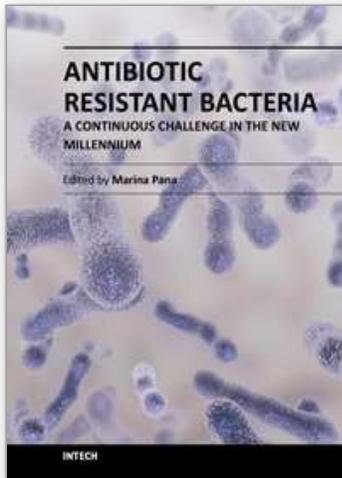
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Antibiotic-resistant bacterial strains remain a major global threat, despite the prevention, diagnosis and antibiotherapy, which have improved considerably. In this thematic issue, the scientists present their results of accomplished studies, in order to provide an updated overview of scientific information and also, to exchange views on new strategies for interventions in antibiotic-resistant bacterial strains cases and outbreaks. As a consequence, the recently developed techniques in this field will contribute to a considerable progress in medical research.

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