Use of Antibiotics in Animal Production and Its Impact on Human Health

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Abstract

Bacterial resistance is a natural phenomenon, however, accelerated by human kind due to the accentuated and indiscriminate use of antibiotic in human and animal health. The development of decontamination techniques and risk assessment of indiscriminate use are auxiliary tools for the control and prevention of “super bacteria”, originated from antimicrobial resistance. This research aimed to verify which the most used antibiotics are in animal health, the described and known decontamination methods in the literature, besides the impact of antimicrobial resistance in humans. The results demonstrated the need to carry out activities that involve actions of national and international institutions along with NGOs in order to supervise and control effectively the use of antibiotics. In general, it was possibly to verify a necessity for guidance to the use of antibiotics by means of the diagnosis of diseases, being necessary that the prescription should be prescribed exclusively by a veterinarian and respecting the grace period of the medicine in order to avoid damages to human health.

Keywords

Animal health, Antibiotics, Food safety, Antimicrobial resistance

Introduction

The production of data about bacterial resistance in livestock breeding and its correlation with human health highlight the phenomenon of cross-resistance. This fact is due to the relationship between antibiotics used in humans and those used in animal health. In addition, two main factors are responsible to accentuate this problem. First, economic interests focused on accelerated and large-scale production, in the agricultural sector and pharmaceutical industries. Second, the lack of knowledge of the producer, especially the small producers, and of the consumer related to constitution and the harmful effects of antibiotics on human health, when used incorrectly, can trigger allergic reactions and of deaths resulted from uncontrollable infections [1].

The introduction of antibiotics as growth promoters has occurred due to the expansion and need to increase food production, as the human population has grown exponentially in recent years, which reflected on the economic capacity of the planet, resulting in the scarcity of sufficient food resources to world population.

The use of antibiotics, in animals intended for human consumption, had as its main objective the therapeutic and prophylactic use in the control of bacterial infections. However, there was a deviation of this function, and it has been used in sub therapeutic doses, as growth promoters, a fact that contributed to the risk
of antimicrobial resistance. This risk arose because of grace period (period needed to reduce the levels of antibiotic doses used at values close to zero in animal tissues intended for consumption) has not been respected, especially in developing countries.

In 1998, the World Health Organization (WHO) alarmed the world about the impact of use of the quinolone class antibacterial in veterinary medicine due to Salmonella spp., Escherichia coli and Campylobacter spp. resistance. WHO considers the use of antibiotics in animal production as a rising risk to human health? Therefore, official institutions and consumer associations have been working to restrict the use of antibiotics as growth promoters in Europe [2].

The use of antibiotics in animal production

The possibility of a veterinary medicinal products to leave residues in products of animal origin depends on pharmacokinetic parameters (absorption, distribution, metabolism and excretion), which in turn are related to the physicochemical properties of the compound [3].

The presence of residues, besides the illegal use, leads to results out of conformity within the veterinary practice which are errors of the producer due to the lack of knowledge, poor identification of treated animals, grace period of drug not respected, general state of the animal's health, concomitant use of different drugs, use of a dosage greater than that recommended by the manufacturer, among others [3, 4].

In recent years, there has been an increase concern about the adverse effects caused by antimicrobial residues, as these drugs can cause changes in the intestinal microbiota of animals, resulting in microbial resistance [3]. In an in vivo study, Brewer et al. [5] demonstrated that the use of small concentrations of antibiotics resulted in the selection of bacteria, causing bacterial resistance.

Livestock production is growing fast in the agricultural sector. In the last decades, production and consumption of animal products have largely increased [6]. In a future, this increase is expected to continue in order to meet the high demand for livestock products such as meat, milk, eggs, and fish, especially in industrialized countries [7-9].

According to the European Commission (EC) about Regulation of the Council Directive 96/23 [10], residues of veterinary medicinal products can come from antimicrobial, anti-helminthic, anti-inflammatory, anabolic steroid sedative compounds, among others.

Subsequent to the introduction of antibiotics in human medical treatment in 1940, they were introduced into bird and pig production as growth promoters. In the mid-1950s, it was discovered that antibiotics when administered in animal feed in a dose lower than the therapeutic dose increased the productivity [11].

Among the groups of antimicrobials used in animal production are: a) β-lactams (penicillins, cephalosporins, tetracyclines), b) phenicols (chloramphenicol, florfenicol and thiampenicol), c) aminoglycosides (neomycin, streptomycin), among others [3].

β-lactams are antibiotics that act inhibiting the transpeptidase by covalently binding to the D-alanyl-D-alanine terminal group from the bacterial cell wall peptidoglycan units. In 1928, penicillin was discovered, which is the co-product of Penicillium notatum metabolism. In animal production, benzylpenicillin is used to prevent or treat local and systemic infections. In mastitis cases, the administration is intramammary, but for decades, sub-therapeutic concentrations of this drug have been used as growth promoters. Benzylpenicillin presents health risk because it can induce practically all clinical forms of allergies. However, cephalosporin can be used in the treatment of mastitis and in endometriosis treatment, and it has a broad-spectrum because it is more resistant to the action of bacterial enzymes. Tetracyclines are routinely used in healthy cows to prevent infections [12]. One of the major problems in the use of tetracyclines is the development of bacterial resistance in intestinal biota [12].

Phenicols are a class of bacterial antibiotics that inhibit the synthesis of bacterial polypeptides. Chloramphenicol is a broad-spectrum antibiotic, predominantly active against pathogenic Gram-negative bacteria. This drug is prohibited in the United States, the European Union and Brazil, because despite being well accepted by animals, clinical use in humans can cause irreversible side effects, regardless of dose [12, 13]. According to ANVISA [12], florfenicol and thiampenicol are still used in the treatment of infectious diseases of cattle, pig and birds, but thiampenicol cannot be used in lactating cows.

In relation to aminoglycosides, the main drugs are streptomycin, gentamicin and neomycin. High doses of these drugs can cause ototoxic and nephrotoxic effects in humans and mammals. Neomycin is indicated for the treatment of mastitis and control of enteric infections, but due to of the adverse effects mentioned above, it should be used with caution, as according to the Joint FAO/WHO Expert Committee on Food Additives (JECFA) [14] the recommended daily intake (IDA) is 0.06 mg/kg body weight [3].

In veterinary medicine, antibiotics have been used against clinical and subclinical infectious diseases. In some countries, antibiotics have been used as antimicrobial growth promoters (AGPs) as well. To this end, they have been supplied in subtherapeutic doses to help livestock by improving growth rate, reducing mortality, and enhancing animal reproductive performance [15]. Antibiotics mostly employed in AGP applications include tetracyclines, ionophores, and penicillin’s [9, 16].

Global antibiotic consumption in livestock has been estimated as approximately 63,000 up to 240,000 metric tons yearly [17], which will increase due to the high consumption levels registered in emerging economies [17]. However, as far as sales of antimicrobials for food-producing species are concerned, a substantial decline has been observed in some countries [18]. Accordingly, this antibiotics overuse will
contribute to spreading antimicrobial resistance worldwide [9].

The indirect hazard occurs through the development of multidrug-resistant pathogenic bacteria that can be transferred to humans through residual contamination of fertilizers or even contamination of the soil with animal feces. So, residues of these drugs can be deposited in the soil, transferred to vegetables, and therefore likely to pose a high risk to human health [19, 20].

It is important to notice that there is a variation of risk perceptions both between nations and between different social groups has been studied and it is strongly impacted antibiotic use and policymaking [21, 22], in which, some countries decided to target antibiotic residues in food and milk, others decided to tackle agricultural antimicrobial resistance selection, and others decided to do nothing at all [23].

Antibiotic residues in product of animal origin

The Brazilian normative instruction number 13/04 (modified by normative instruction nº 44/15) from Ministry of Agriculture, Livestock and Food Supply (MAPA) approves the technical regulation about additions used in animal feed. It presents how to evaluate, register, commercialize and employ additives in animal feed, aiming to guarantee a satisfactory safety level for human, animal and environmental. This normative instruction defines as additives for animal feed products as any substance, microorganism or formulated product intentionally added to products intended to animal feed that can have or not nutritional value and can improve the performance of healthy animals or meet nutritional needs. However, additives must be used in the amount strictly necessary to obtain the desired effect [24].

In Brazil, both biological and pharmaceutical veterinary products need prior authorization of MAPA to be marketed. Establishments that manufacture such products must be registered in a competent public agency [24].

The Maximum Residue Limits (MRL) are defined as the maximum concentration allowed of residues of veterinary products in food of animal origin that is legally allowed or recognized as safety for consumer’s health. The Brazilian normative instruction number 26/09 which provides technical regulation for the manufacture, quality control, commercialization and use of antimicrobial products in animals establishes MRLs established by the Codex Alimentarius [25] or in specific legislation and, in the absence of these, MRLs internationally recognized and authorized by MAPA can be accepted.

The Codex Alimentarius [25] is a commission formed by Food and Agriculture Organization of the United States (FAO) and World and Health Organization (WHO), which was created in 1962 to regulate trade in food of animal origin [26]. This commission also is the main regulatory agency that determines the MRL of substances for veterinary use in foods derived from an animal matrix. Table 1 shows the MRL for veterinary antibiotics in foods. The values presented in the table 1 vary for different species and limits for different tissues in the same animal such as muscle, liver, kidney and fat were considered, but MRLs values from milk and eggs were not considered.

Tian et al. [27], when reviewing about effect of thermal treatments on the degradation of antibiotic residues in food, noticed that the physicochemical composition of a food matrix as well as cooking methods and the addition of food additives are parameters that can influence the degradation of antibiotic residues. However, it must be emphasized that the reduction the antibiotics concentration caused by heat treatment results in an increase of the concentration of unknown by-products that can be harmful to health. Therefore, it is not safe to affirm that the antibiotics degradation by thermal treatments is effective in relation to food safety.

The impact of residues on human health

Antibiotics have benefited substantially public health for over 60 years and are also widely used in cattle destined to human consumption [28]. Since 1950s, the use of antibiotics in animal feed has become a worldwide trend, which triggered by the growing dependence on the agricultural industry to increase food production faster and more effectively, following population growth. So, more than 150 antibiotics are currently used in animal production for human consumption and 90% of these antibiotics are natural products of bacteria, fungi and semi-synthetic substances derived from modifications of natural compounds, and some of them are synthetic [29].

Currently about 80% of production animals destined for human consumption receive antibiotics, either to prophylactic or therapeutic function. Furthermore, the most widely used antimicrobials in the production animals designated to food consumption are β-lactams, tetracyclines, aminoglycosides, lincosamides, macrolides, and sulfonamides. However, the use of these antibiotics in animals intended for human consumption may leave residues or metabolites in meat, milk and eggs [30].

The residue may be from the antimicrobial itself or from its secondary metabolites. Such substances may be deposited, accumulated or stored in cells, tissues, organs or edible products of animal origin. The main purpose of the use of these drugs in animals is to prevent, control or treat animal diseases or to increase production. However, these residues can accumulate in humans over time and consumption, thereby increasing the selectivity of multi-resistant bacteria to these drugs widely used in human health [31].

Residues from antibiotics in animal origin food are one of the major causes of concern for human health. The consumption of these residues can lead to different disturbances such as direct toxicity, allergic reactions, carcinogenic effects (e.g.: sulfamethazine, 4-dedimethyl amino-4-oxo-tetracycline and furazolidone), mutagenicity, nephropathy (e.g.: gentamicin), hepatotoxicity, reproductive disorders, bone marrow toxicity (e.g.: chloramphenicol), allergy (e.g.: penicillin), and destruction of useful microbiota present in the gastrointestinal tract, especially children, elderly, pregnant and immune compromised [32, 33].
According to study performed by Wang and Tang [34], total quantity of annual antibiotics use, including medical and veterinary, reached 200,000 tons in whole world.

The adaptation of microorganisms to antibiotics and antimicrobial resistance have increased because of the excessive and incorrect use of that class of drugs in human and animal health. The regular presence of heavy metals in animal manure further increases the abundance of antibiotic resistance in bacterial populations by co-selection [35]. In addition, the World Health Organization (WHO) classify as being of broad therapeutic use in human medicine some of antibiotics that are used to promote growth in pigs, birds and cattle. However, long-term exposure may promote the development of genetic variations of microorganisms and the production of antibiotic resistant genes, fact that results in the evolution of resistance of pathogens and bacteria [36, 37].

As there is currently no development of new antibiotics, antimicrobial resistance can generate a high proliferation of "super bacteria" and in the future diminish/eliminate the chances of effective treatment of diseases from those multi-resistant pathogens. In addition, those micro-organisms can be transported from foods, mainly from animal sources, to the consumer through the impulsive consumption of these agricultural products and the irresponsible use of growth promoters in the production of animal origin products [36].

**Measures of prevention and control of antibiotic residues**

There is great concern related to the presence of antibiotic

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Table 1: Maximum residue limits for residues of veterinary drugs in foods, according Codex Alimentarius.

<table>
<thead>
<tr>
<th>Antibiotic</th>
<th>Animal Production Species - Maximum Residue Limits (µg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Cattle</td>
</tr>
<tr>
<td>Amoxicillin</td>
<td>4’/ 50&lt;1,5</td>
</tr>
<tr>
<td>Avilamycin</td>
<td>4’</td>
</tr>
<tr>
<td>Benzylpenicillin</td>
<td>4’/ 50&lt;1,5</td>
</tr>
<tr>
<td>Cefotiofur</td>
<td>100&lt;1/ 1000&lt;1/ 2000&lt;1/ 600&lt;2</td>
</tr>
<tr>
<td>Chloramphenicol</td>
<td>*</td>
</tr>
<tr>
<td>Chlorotetracycline/Oxytetracycline/Tetracycline</td>
<td>100&lt;1/ 200&lt;1/ 600&lt;1200&lt;3</td>
</tr>
<tr>
<td>Colistin</td>
<td>50&lt;1/ 150&lt;2,5/ 200&lt;3</td>
</tr>
<tr>
<td>Danofloxacin</td>
<td>100&lt;1/ 200&lt;1/ 400&lt;3</td>
</tr>
<tr>
<td>Dihydrostreptomycin/Streptomycin</td>
<td>200&lt;1/ 600&lt;2&lt;1/ 100&lt;3</td>
</tr>
<tr>
<td>Erythromycin A</td>
<td>-</td>
</tr>
<tr>
<td>Flumequine</td>
<td>500&lt;1,5/ 1000&lt;1/ 3000&lt;4</td>
</tr>
<tr>
<td>Furazolidone</td>
<td>*</td>
</tr>
<tr>
<td>Gentamicin</td>
<td>100&lt;1/ 200&lt;1/ 500&lt;2</td>
</tr>
<tr>
<td>Lincomycin</td>
<td>150&lt;3</td>
</tr>
<tr>
<td>Monensin</td>
<td>2’&lt;1/ 100&lt;1&lt;3</td>
</tr>
<tr>
<td>Narasin</td>
<td>15&lt;1,5/ 50&lt;3</td>
</tr>
<tr>
<td>Nitrofurazol</td>
<td>*</td>
</tr>
<tr>
<td>Olaquindox</td>
<td>*</td>
</tr>
<tr>
<td>Pirlimycin</td>
<td>100&lt;1,5&lt;1/ 400&lt;1/ 1000&lt;4</td>
</tr>
<tr>
<td>Ronidazole</td>
<td>*</td>
</tr>
<tr>
<td>Sarafloxacin</td>
<td>-</td>
</tr>
<tr>
<td>Spectinomycin</td>
<td>200&lt;1/ 500&lt;1/ 200&lt;2&lt;1/ 500&lt;3</td>
</tr>
<tr>
<td>Spiramycin</td>
<td>200&lt;1,5&lt;1/ 300&lt;1&lt;1/ 600&lt;2</td>
</tr>
<tr>
<td>Tilmicosin</td>
<td>100&lt;1,5&lt;1/ 300&lt;1&lt;1/ 1000&lt;3</td>
</tr>
<tr>
<td>Tylosin</td>
<td>100&lt;1,2,5&lt;3</td>
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</tbody>
</table>

* Insufficient data were available or there was a lack of data to establish a safe level of residues or its metabolites in food representing an acceptable risk to consumers, significant health concerns were identified. For this reason, authorities should prevent residues of these antibiotics in food. This can be accomplished by not using these antibiotics in food producing animals.

1 Muscle; 2 Liver; 3 Kidney; 4 Milk; 5 Fat/Skin.
According to Nascimento et al. [39], the presence of antibiotics, mainly penicillin, can lead to allergies in the most susceptible individuals and, consequently, episodes of asthma, digestive disorders, urticarial and even anaphylactic shock.

In the following topics, some prevention and control measures will be presented for antibiotic residues.

### Irradiation or ionizing radiation

Irradiation has been reported to convert non-degradable pollutants into degradable ones. For example, tetracycline and oxytetracycline, which could be degraded to different degrees at different radiation intensities. This technique has also been used to decontaminate groundwater [36].

Irradiation or ionizing radiation is a technology that is based on passage of a beam of electrons and gamma rays through the sample to decontaminate it. This technique is considered a fast and reliable disinfection and decontamination process, besides being a sterilization technique compatible with most of the materials, among them matrices of animal origin for human consumption. Such technique has been reported to be effective for transformations of non-degradable persistent organic pollutants into degradable products [40, 41].

Irradiation of electron beams in organic pollutants is a technique frequently used for the purification of groundwater [40, 42-45]. In those studies, electron beam irradiation was able to degrade chloramphenicol, tetracycline and oxytetracycline to varying degrees and at different intensities of radiation. In addition, Alsager et al. [46] studied the destruction of commonly used veterinary antibiotics, such as amoxicillin, doxycycline, and ciprofloxacin in water and various food commodities (milk, chicken meat, and eggs) by ionizing gamma irradiation. Their results indicate that this technology can be used to minimize considerable environmental and economic impact of buildup antimicrobial resistance against antibiotics.

The products resulting after the degradation of chloramphenicol were tested for microbial toxicity showed no antimicrobial effects [44]. A study performed by Cho [47] reports the use of electron irradiation for the treatment of tetracycline in pig manure. In this study, the efficiency of the degradation and intermediate formation of artificial contaminants in pig manure was tested. The efficiency of degradation increased with the increase of the radiation intensity and with this it was also evaluated the formation of tetracycline after the radiolytic degradation.

Therefore, electron irradiation was considered to be a highly effective technique for the degradation of antibiotics, especially tetracycline. However, this technique can be expensive for large-scale use. Future studies should focus on standardizing the radiation procedure and the intensity to be applied on a wide range of antibiotics.

### Biological remediation technology

Biological remediation technology has also been reported to convert non-degradable to degradable pollutants. The phytoremediation potential of some woody plants is partially effective in the removal of sulfonamides.

Phytoremediation is studied in order to find better conditions of inoculation of some solubilization bacteria, such as *Pseudomonas* sp. and *Bacillus* sp. because these bacteria are nitrogen fixers. In addition, along with nitrogen fixation with antioxidant properties, low molecular weight organic acids should be also used to supply energy required for plants after the stress from antibiotic residual overload, as well as to have a nutrient supply to the inserted microorganisms, promoting absorption and degradation of antibiotic products contained in the soil. There are reports that plant sediments contaminated with fluoroquinolone were phytodegraded [36].

### Biosensors

The first article on the development of a biosensor for the detection of antibiotic residues (amphotericin B and nystatin) was published in 1979 [48]. In prevention and control area of antibiotic residues, a method that has been drawing attention in the field of antibiotic screening in foods, are biosensors. The biosensor instruments consist of two main components: a biorecognition element, which is an element that recognizes the target analyte, and a transducer that converts the recognition event into a measurable signal [49]. Gaudin [50] has written a review on biosensor development for the screening of antibiotic residues, she stated that this technology has a very broad reach in industries such as pharmaceuticals, health, agriculture products and food.

Several biological recognition elements can be used, such as enzymes, antibodies, microbial cells, among others, which have been used in the manufacture of biosensors [51]. Enzymes and antibody/antigen affinity are the widely used recognized elements for sorting of residues of veterinary drugs [52].

Biosensors are classified by their biorecognition element or their transducer type being optical, electrochemical and mass-based transducer the most popular and common biosensor methods for the detection of antibiotic residues. Another type of biosensor, piezoelectric, has very few reports in the literature regarding the use of it for detecting residues of antibiotics and other food contaminants [51]. A microbial biosensor consists of a transducer in association with viable or non-viable immobilized microbial cells. Cellular biosensors have been developed for the detection of antibiotic residues [53], beta-lactam antibiotics [54], quinolones [54], chloramphenicol and quinolones [55]. In addition, Tian et al. [27] developed a yeast whole-cell biosensor regulated by two promoters capable of detecting genotoxic compounds. The same authors affirm that the use of yeast cells as biosensors is increasingly important for the analysis of a variety of biologically toxic chemical compound because they provide higher throughput, strong resistance to the cytotoxicity of the chemical compounds tested, lower consumption of the compound used and are sensitive to a broad spectrum of genotoxins.
Electrochemical method

According to Santos [56], dairy farms should be encouraged to create waste control programs, beginning with the development of treatment protocols for the diseases that most frequently appear in lactating cows. It is necessary that besides to the protocols, there are records of the treatments performed with information on the number of the animal, duration of treatment, routes of administration, grace periods, doses and medications used [57].

The treatment of cows with antimicrobial agents, both for external and internal use, requires the establishment of time required for the drug to be completely eliminated [58]. This fact, however, can lead to problems of propagation of antibiotic resistant microorganisms to soil and water. Therefore, it is essential to develop a suitable technology for the degradation of antibiotics in the milk used, as is the case of the electrochemical processes, which are being used for the treatment of residual water and the decomposition of some compounds [59, 60].

Kitazono et al. [61] verified the selective degradation of the tetracycline antibiotics present in the milk, using an electrochemical method. Electrochemical oxidation is a physical-chemical method used for the degradation of chemical substances. This treatment can be applied in the treatment of residual water and decomposition of some compounds. In this study, the authors investigated the electrochemical oxidation of tetracycline residues in milk using active and inactive anodes in different electrolytes. According to the research, as the electrochemical treatment consumes a lot of energy, this study aimed the development of a selective oxidation method for the degradation of tetracyclines in milk. The highest antibiotic degradation rate found by the authors was achieved by means of an inactive anode and a NaCl electrolyte.

Oxygen adsorbed on the surface of the anode and oxidation using sodium hypochlorite could increase the degradation of the tetracyclines in the milk, however, the organic components of milk affected the removal of the antimicrobial. So far, electrochemical oxidation has only been carried out in lab scale, but according to the authors, it is an effective method that could be used for the degradation of antimicrobials in milk, helping to minimize contamination of the environment, since milk with residues of antibiotics should be discarded.

Miyata et al. [62] investigated the electrochemical oxidation of tetracyclines to treat residual water from cattle, and the method proved to be effective in degrading the antibiotic in question. Tetracycline concentrations in the aqueous solutions were reduced from 100 mg/L to less than 0.6 mg/L during 6 hours of electrochemical treatment using a Ti/IrO2 anode with Na2SO4 electrolyte. The concentration of oxytetracycline in residual water from 100 mg/L to less than 0.7 mg/L was also reduced by the same treatment.

Conclusions

Caution should be exercised in the use of antibiotics and in the description of methods of antimicrobial resistance, since factors other than administration to production animals should be considered. Another important factor to consider is that antibiotics were strong allies in the increase of animal production in Brazil and in the world. However, the indiscriminate use of antibiotics should be controlled and monitored in human and animal health, since the emergence of “super bacteria” can seriously affect human health due to the lack of development of new antibiotics. Studies of methodologies for the degradation of antibiotic residues in products of animal origin are necessary, since there has been an increase in the use of these drugs in animal production over the years.

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