

Beef's Antibiotics Residues Determination from Arjona and Magangué Municipalities in Bolívar, Colombia

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Abstract

The objective of this investigation was to determine the amount of residues of Penicillin G, Sulfadimidine and streptomycin in beef from two beneficiary plants in the municipalities of Arjona and Magangué, department of Bolívar-Colombia. For this, 10 samples were obtained from each provenance site, for a total of 20 samples for the investigation. The samples were packed in labeled polyethylene bags and refrigerated at a temperature between 3°C and 5°C. The quantification of residues was carried out through high performance liquid chromatography (HPLC). The results showed that only residues of penicillin G over pass the minimum amounts for antibiotics imposed by the authorities, while residues of sulfadimidine and streptomycin are below what is allowed.

Keywords: HPLC, Penicillin G, Sulfadimidine, Streptomycin, Drugs

1. Introduction

Meat can be considered as a valuable food since it is composed of different nutrients

among which are proteins, minerals, fats, vitamins, small amounts of carbohydrates, among other bioactive components and given its composition can be part of a balanced diet [1]. The consumption of beef per capita in the world in 2016 was 6.5 kg / habitant, while consumption in Colombia for the same year was 18.6 kg /habitant [2, 3]. Unfortunately, antibiotic residues have increased significantly due to the indiscriminate and usual use of antibiotics in clinical practice. This type of medication is commonly used to prevent and treat diseases in animals to improve animal production [4].

Antibiotics are used in medical treatment for humans and animals. The main antibiotics used in animal husbandry include penicillins, sulfonamides and tetracyclines. The administration of these drugs to animals is normally done through feeding, which leads the preservation of these residues in the animal tissues that will be consumed [5-8]. Drug residues in food are a major concern for public health in many countries, especially when most food sales ignore official quality assurance channels. Consumers are well aware that their food supply is free from contamination by herbicides, pesticides, medicines or antibiotics due to the fact that they can cause serious health risks, causing allergic reactions, carcinogenicity and promotion of the spread of bacterial resistance to antibiotics used in medicines for human use [4].

When there is an application of an antibiotic to an animal, it is metabolized, which helps its elimination and detoxification. Although there is elimination of antibiotics in the organism of the animals to which they have been supplied, residues of said drugs are created in foods of animal origin, these residues are generally pharmacological substances. These substances are located in variable areas, some of the preferred sites are muscle tissue, fat, liver and kidneys [9, 10]. Due to the problem of veterinary residues plus the need to protect public health, it was necessary for the sanitary authorities to establish maximum residue limits (LMR) both for the different animal species destined for human consumption and for the different edible tissues of these (muscles, liver, kidneys, fat, milk, honey and eggs) [10, 11]. Due to the few quantitative studies of antibiotic residues in beef in Colombia, it is important to conduct research on this topic. For which the objective of this study was to determine the amount of antibiotic residues in beef from the municipalities of Arjona and Magangué, department of Bolívar (Colombia).

2. Methodology

2.1. Sampling

For this research, two municipal benefit plants (PBM) were established, one belonging to the municipality of Arjona and the other to Magangué, both located in the department of Bolívar, each having a daily average of 75 cattle slaughtered. Ten samples of skeletal muscles were taken in the diaphragmatic area of freshly

slaughtered cattle carcasses for each PBM selected, with dimensions no greater than 5 cm in length and 3 cm in width. At the moment of taking the samples, only the sex of the animal was distinguished. The samples were packed in new and properly labeled polyethylene bags, then refrigerated at temperatures between 3 °C and 5 °C and then transported to the laboratory.

2.2. *Samples preparation*

Samples preparation was carried out according to the methodology of Acevedo et al., [12], 10 grams of beef were added with 5% methanol, these were subjected for 30 minutes to a microfiltration to ensure complete dilution of the antibiotic (Cole Parner 8853 ultrasound bath). Next, it was centrifuged (Becton Dickinson Centrifuge, USA) for 10 min at 5000 rpm. The supernatant is taken and transferred to a beaker containing 1 ml of Carrez I solution and Carrez II solution. This mixture was stirred and centrifuged for 10 minutes, the supernatant was passed through a C-18 solid phase extraction cartridge, previously conditioned. The extraction of the analyte in the cartridge was carried out by elution with 5 ml of methanol. The methanol solution was filtered and injected into the chromatography equipment. All samples were prepared in triplicate.

2.3. *Antibiotics to be determined*

The antibiotics that were determined were: Sulfadimidine, Penicillin G and Streptomycin, according to the methodologies presented by Kishida and Furusawa [13], Kukusamude et al., [14] and Edder et al., [15], respectively.

2.4. *Chromatography conditions*

Separations were developed on a Phenomenex Synergi Polar-RP Column 250X4.6 mm column (Phenomenex, USA). The mobile phase consisted of a mixture of 85% sodium monobasic phosphate (NaH₂PO₄) 20 mM, 15% acetonitrile (ACN) at a flow of 1.5 mL / min and a detection wavelength of 210 nm. The injection volume of the sample was 20 µL. The equipment used was a liquid chromatograph (BAS SS60) equipped with a ternary pump and manual injection and a UV-VIS detector (BAS) of variable wavelength.

3. Results

In Table 1 you can see the HPLC results for the different antibiotics analyzed according to the municipal benefit plant and additionally compare the levels of the residues with the maximum allowed by the standard [16] for each type of medicine.

Table 1. Analysis of antibiotic residues in beef in $\mu\text{g/Kg}$

Antibiotic	Arjona PBM		Magangué PBM		LMR
	Males	Females	Males	Females	
Sulfadimidine	31,32±2,51 ^a	50,33±2,52 ^b	40,3±1,32 ^c	58,92±2,51 ^d	100 $\mu\text{g/Kg}$
Penicillin G	4,17±0,29 ^a	9,59±1,53 ^b	5,60±1,47 ^a	11,34±1,53 ^b	4 $\mu\text{g/Kg}$
Streptomycin	201,73±5,32 ^a	281,60±6,66 ^b	195,43±2,82 ^a	265,45±3,51 ^c	600 $\mu\text{g/Kg}$

As seen in Table 1, the Magangué PBM for sulfadimidine antibiotics and penicillin G has more waste than the Arjona PBM, while the latter had a higher concentration of streptomycin. Regarding the concentration in males and females, it can be noted that the females in both PBM had higher concentration in all antibiotics, presenting a statistically significant difference ($p < 0.05$) between sexes. The presence of more waste in females than in males can be due to the use of antibiotics in diseases that occur in females such as mastitis and metritis [17]. In addition, although in this table there is a statistically significant difference between males and females, only the limit is surpassed in both cases for penicillin G. Table 1 shows the existence of sulfadimidine residues in the samples of the two PBMs but these do not exceed the maximum residue limits. In the same way in this table you can notice that there is a significant difference between the two samples and between sexes, which may be due to the use of larger amounts of this medicine. Medications with sulphonamides are popular antibacterial agents for cattle used as growth promoting agents, and are also used against bacterial and coccidial infections, although their residues in meat products are a potential danger to human health, since residues of these medications could cause hypersensitivity reactions [18-20].

Although most β -lactams (such as penicillin G) are excreted rapidly through urine [21], Table 1 shows that the penicillin content is low with respect to the other drugs evaluated, this exceeds the maximum residue limits in both PBM. Some of the causes that can bring the consumption of meat with residues penicillin are diarrhea, edema, urticaria, allergic reactions among other symptoms that are not fatal [22]. This type of medication is used for the treatment of bacterial infections of bovines raised for the production of meat and/or milk [23]. Among other indications, penicillin G administered parenterally has been used for the treatment of mastitis, arthritis and respiratory infections [24]. Streptomycin has been reported some allergic reactions and anaphylactic reaction by consumption of meat with this type of waste [19, 20, 25].

One of the reasons for the presence of antibiotics in the meat can be the incorrect prescription of antimicrobials by the doctor, the therapeutic misuse of these agents such as the case of buying medicines in unauthorized establishments (pet stores, farms livestock, among others), the use without these, or the use of antimicrobials not authorized in the veterinary sector [10]. Meats contaminated with antibiotic residues, if used to obtain fermented products, may present abnormal fermentations and failures of the starter cultures [26, 27]. In addition, antibiotic

residues could even alter the development of the contaminating microbial flora, which could lead to the masking of some pathogens during the bacteriological control of food [28]

4. Conclusion

The presence of antimicrobials in the samples taken indicates that the animals are often exposed to these veterinary drugs weeks or even days before being taken to the municipal benefit plants and that there is a lack of vigilance towards the withdrawal times of the drugs before sending the animals to slaughter.

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