Oxytetracycline and Procaine Penicillin Residues in Tissues of Slaughtered Cattle in Maiduguri, Borno State, Nigeria

Yaqub Ahmed Geidam1* · Hamidu Usman2 · Hassan Ismail Musa3 · Franca Anosike1 · Yemisi Adeyemi1

1 Department of Veterinary Medicine, University of Maiduguri, P.M.B. 1069, Maiduguri, Borno State, Nigeria
2 Department of Chemistry, University of Maiduguri, P.M.B. 1069, Maiduguri, Borno State, Nigeria
3 Department of Veterinary Public Health and Preventive Medicine, University of Maiduguri, P.M.B. 1069, Maiduguri, Borno State, Nigeria

Corresponding author: * ygeidam@yahoo.com

ABSTRACT

A study was conducted in Maiduguri to detect the presence of antibiotics (oxytetracycline and procaine penicillin) residues in some tissues of cattle slaughtered for human consumption. Thin layer chromatography was utilized to qualitatively screen tissue samples by running in parallel with reference standards of both antibiotics. Out of the total 285 tissues sampled, muscle produced the highest incidence rate (32.6%) of oxytetracycline residue followed by liver (5.0%) and then kidney (3.1%). Similarly, muscle tissue produced the highest incidence rate (15.7%) of procaine penicillin residues followed by liver (13.0%) and then kidney (8.3%). The presence of residues of these commonly used antibiotics in tissues is a pointer to a serious public health risk as these antibiotics are also used in humans and may result in the development of bacterial strains resistant to these antibiotics. This underscores the need for a national residue monitoring program in Nigeria in accordance with international regulations.

Keywords: antibacterial, antibiotics, kidney, liver, muscle
Abbreviations: rpm, revolutions per minute; TLC, thin layer chromatography

INTRODUCTION

Antibiotics are naturally occurring, semi-synthetic and synthetic compounds with antimicrobial activity that can be applied parentally, orally or topically (Kemper 2008). Presently in Nigeria, many antibiotics are widely used for preventing and treating several diseases, as well as for promoting growth in food-producing animals (Kabir et al. 2002, 2004). In Nigeria, cattle are considered to be an important source of protein to the ever-increasing human population. The population of cattle in Nigeria was estimated to be about 13.9 million (Bourn 2004). In Nigeria, legislation regarding drug use and veterinary drug residue control is lacking; basic facilities for determining of residues are also unavailable at the level of abattoirs, farms and markets (Kabir et al. 2004). Similarly, there is no specific residue monitoring program. However, there are scanty reports on veterinary drug residues and on the extent of the residue problem in Nigeria in which a limited survey in eastern Nigeria by Obuogbulam and Fidelis (1996) reported finding residues in two out of 24 cattle slaughtered in the region. Another study by Kabir et al. (2002) reported 7.4% in the north western region. A study was therefore carried out to screen animals slaughtered in Maiduguri, north-eastern Nigeria to determine the incidence of oxytetracycline and procaine penicillin residues in some tissues of cattle slaughtered as a preliminary means of detecting animals harbouring antibacterial drug residues. Antibiotics are not completely eliminated in animal organisms, as they are bioactive substances, acting highly effec-
tively at low doses and excreted after a short time of residence. Antibiotics are optimised with regard to their pharmacokinetics in the organisms: organic accumulation is, as in other pharmaceutics, objectionable and thus, they are excreted as parent compounds or metabolites (Kümmerer et al. 2000; Thiele-Bruhn 2003). The levels of antibiotics excretion depends on the active components, routes and periods of administration and the host animal involved; but these levels varies between 40-90% for tetracyclines and sulphonamides (Berger et al. 1986; Haller et al. 2001; Halling-Sørensen 2001).

MATERIALS AND METHODS

Study area

The study was carried out in Maiduguri, the Borno State capital, which is located at Latitude 11° 51’ N and Longitude 13° 10’ E in North-eastern Nigeria. The arid zone has rather austere climatic conditions with a hot dry season from late January to late June during which average daily peak temperatures, especially in April and May, are 34.4 to 37.8°C. The rainy season lasts from late June to mid September and provides an annual average of 46.3 cm rainfall. The cold north-easterly trade wind blowing across the Sahara desert in October to January brings with it cold and desiccant effects on the environment. Thus, nomadic herdsmen who own most of the cattle in the State are constantly on the move in most parts of the year in search of lush grazing ground and water. Fatigue imposed on already malnourished animals as a result of movement over long distances, and excessive heat load which is not easily shed, subject animals to severe stress, which in turn makes them succumb easily to infection by various agents and diseases that may necessitate treatment with antibiotics (Brísibe et al. 1996).

Sample collection

A total of 285 tissues comprising of muscle (89), liver (100) and kidney (96) of cattle routinely slaughtered at Maiduguri abattoir were screened for the presence of some antibiotics at slaughter. Fresh tissue samples, which included kidney, liver and muscle tissues of cattle were collected from Maiduguri Monday market and Maiduguri abattoir and transferred into sterile vials and transported to the laboratory on ice and processed immediately. The samples were analyzed for the residues of oxytetracycline and procaine penicillin.

Sample processing

Five grams each of muscle, kidney and liver tissues were measures and mixed with 5 ml of ethanol (May and Baker Laboratory Reagents Ltd, England). The samples were crushed and squeezed into fine particles in a Chinese mortar. The solvent was later transferred to 15 ml Falcon centrifuge tubes and centrifuged at 7000 rpm for 10 min. The clear supernatant was transferred into fresh glass test tubes and evaporated to dryness. After drying, the deposits were dissolved in 0.2 ml ethanol and made ready for TLC examination as described by Tajjick et al. (2002). For comparison of extracted residues with standard antibiotics [oxytetracycline, obtained from Sigma (St. Louis, MO, USA) and procaine penicillin (Sishui Xier-kang, Shandong, China)], working concentrations of standard antibiotics were prepared by dissolving 0.1 g of each powder in 4 ml methanol (Thangadu et al. 2002). The rationale for the choice of these antibiotics for the study is that they are the most commonly used antibiotics in the study area.

Preparation of silica plate

Glass plates of 20 x 20 cm dimensions were washed in acetone bath. For each plate, 2 g of silica gel F256 (Marik, Germany) was mixed in 5 ml of distilled water and shaken thoroughly to produce fine paste. Clean glass plates were coated with silica pastes by thin layer chromatography (TLC) gel spreader system (Shandon, UK) of 0.25 mm thickness. The plates were then activated at 120°C for 2 h prior to use as described by Boyer (1993).

Pointing, running and digestion

About 0.5 g of the samples was dissolved in 20 ml of methanol, an aliquot of which was spotted on the TLC plates. Treated plates were transferred to already equilibrated TLC tank containing acetone-methanol (1: 1) as the solvent system. The plates were removed from the tank after reaching the solvent front, chromatograms were then observed on UV light at 250 nm (Thangadu et al. 2002).

Evaluation of the thin layer chromatogram

The thin layer chromatogram was evaluated qualitatively based on a multistage distribution process as described by Tajjick and Shoshre (2006). This was achieved by parallel running with the reference antibiotics standards. Later, photometric evaluation was comparatively performed directly on the layers.

RESULTS AND DISCUSSION

Table 1 shows the distribution of oxytetracycline in the tissues of slaughtered cattle by organ. Of all the organs sampled, muscle tissue produced the highest incidence rate of 32.6% followed by liver (5.0%) and then kidney (3.1%). Of the 285 samples screened, 37 (13.0%) of them were positive for the presence of oxytetracycline in the sampled tissues. The high positive samples of oxytetracycline could be as a result of indiscriminate use of drugs in animals as buttressed by Kabir et al. (2002).

Similarly, Table 2 shows the distribution of procaine penicillin in the tissues of slaughtered cattle by organ. Of all the organs sampled, muscle tissue produced the highest incidence rate of 15.7% followed by liver (13.0%) and then kidney (8.3%). The presence of these veterinary drug residues in animal tissues is considered a public health hazard. Penicillin in animal tissues was reported to have caused severe anaphylactic reaction in a consumer (Teh and Rigg 1992) apart from the known fact of causing the development of drug resistance. The high positive samples of oxytetracycline could be as a result of indiscriminate use of this drug as both prophylaxis and therapeutic means of disease control in animals (Kabir et al. 2002, 2004). Furthermore, Kemper (2008) enumerated the three risks derived from immediate appliance of antibiotics as environmental contamination with original substances or derivatives, the indirect impact on health via resistant micro-organisms and the direct organic damage. Additionally, the influences on the biotic environment are a matter of concern (Kemper 2008). The main interest regarding the use of antibiotics in human and animal treatment is the development of resistant bacterial strains representing a health risk to humans and animals. Especially, the application of veterinary antibiotics to food animals is supposed to enhance the selection for

<table>
<thead>
<tr>
<th>Organ</th>
<th>Positive (%)</th>
<th>Negative (%)</th>
<th>Total (%)</th>
<th>Incidence rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Muscle</td>
<td>29 (32.6)</td>
<td>60 (67.4)</td>
<td>89 (31.3)</td>
<td>32.6</td>
</tr>
<tr>
<td>Liver</td>
<td>5 (5)</td>
<td>95 (95)</td>
<td>100 (35.0)</td>
<td>5.0</td>
</tr>
<tr>
<td>Kidney</td>
<td>3 (3.1)</td>
<td>93 (96.9)</td>
<td>96 (33.7)</td>
<td>3.1</td>
</tr>
<tr>
<td>Total</td>
<td>37 (13.0)</td>
<td>248 (87.0)</td>
<td>285 (100)</td>
<td>13.0</td>
</tr>
</tbody>
</table>

Table 2 Distribution of Procaine penicillin in the tissues of slaughtered cattle by organ.

<table>
<thead>
<tr>
<th>Organ</th>
<th>Positive (%)</th>
<th>Negative (%)</th>
<th>Total (%)</th>
<th>Incidence rate (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Muscle</td>
<td>14 (15.7)</td>
<td>75 (84.3)</td>
<td>89 (31.3)</td>
<td>15.7</td>
</tr>
<tr>
<td>Liver</td>
<td>13 (13)</td>
<td>87 (87)</td>
<td>100 (35.0)</td>
<td>13.0</td>
</tr>
<tr>
<td>Kidney</td>
<td>8 (8.3)</td>
<td>88 (91.7)</td>
<td>96 (33.7)</td>
<td>8.3</td>
</tr>
<tr>
<td>Total</td>
<td>32 (12.3)</td>
<td>250 (87.7)</td>
<td>285 (100)</td>
<td>12.3</td>
</tr>
</tbody>
</table>

n= number of samples tested
strains resistant to antibiotics used in human medicine. Transmission of these strains might be performed via direct contact with animals or via the food-chain to the consumers. As antibiotic resistance protects antibiotic-producing organisms from their own products, and other originally susceptible organisms from competitive attack, it is as ancient as antibiotics. Studies have shown that excretion rates of tetracyclines are dependent on the substance, the mode of application, the excreting species and time after administration, but it has been shown to vary between 40 and 90% for tetracyclines and sulphonamides (Berger et al. 1986; Haller et al. 2001; Halling-Sørensen 2001). Misuse of antibiotics by farmers and veterinarians alike, in addition to causing residues in edible tissues, is also contributing to the development of microbial drug resistance and the spread of resistant bacteria, including those with serious public health consequences within and across national borders (Levy et al. 1976; Lyons et al. 1980; Wolfgang 1998; EU Scientific Steering Committee 1999).

REFERENCES


ECC Council (1986) Council directive concerning the examination of animals and fresh meat for the presence of residues (86/469/ECC). Official Journal of the European Community L275, 36-45


Halling-Sørensen B (2001) Inhibition of aerobic growth and nitrification of bacteria in sewage sludge by antibiotic agents. Archive of Environmental Contamination Toxicology 40, 451-460


Levy SB, Fitzgerald GB, Macone AB (1976) Spread of antibiotic resistance plasmids from chicken to chicken and from chicken to man. Nature 260, 40-42


Teh WL, Rigg AS (1992) Possible penicillin allergy after eating chicken. Lan- guage ii, 1632

Thangadu SSK, Shukia SK, Anjanegulu Y (2002) Separation and detection of certain β-lactams and fluoroquinolone antibiotic drugs by thin layer chromatography. Analytical Sciences 18, 97-100


