Detection of β- Lactam antibiotics (Penicillin and Amoxicillin) residues in Goat milk

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Abstract: The use of antibiotics may result in drug residues and the promotion of drug resistant strain of organisms. This study evaluated penicillin and amoxicillin residues in goat milk sourced from Ibadan city in South Western Nigeria. A total of one-hundred and sixty-six goat milk samples from Red Sokoto and West African breeds of goats were analysed using the high performance liquid chromatography method. All the samples were positive for the presence of penicillin and amoxicillin residues. The mean penicillin and amoxicillin residues was 0.2823 ± 0.1227 and 0.1228 ± 0.0571ppm for Red Sokoto (RS) and 0.2572 ± 0.0194 and 0.1076 ± 0.0058ppm for West African dwarf (WAD) breed of goats respectively. Hence, values obtained were in line with Maximum residue limit of (4μg/L) set by the European Union regulations or Food and Drug Administration (U.S. FDA), of 5μg/L for both antibiotics. However, there was no significant difference at p< 0.05 in the means of the penicillin residues in both breeds, but a reverse was the case with amoxicillin residues. Thus, the goat milk are safe for consumption. None the less, it calls for close monitoring to prevent values from increasing in the near future due to indiscriminate use of these antibiotics.


Key words: Antibiotic residues, Penicillin, Amoxicillin, Goat milk

Introduction
Milk obtained from lactating goats is a richer source of animal protein as compared to cow milk, to mean farmers; goat is referred to as a poor man’s cow. Peasant farmers that cannot afford raising a cow can rear several goats as a source of livelihood and for consumption. These goats are reared within the household, being fed majorly with wastes and exposed to various hazards the environment can afford. They in turn serve as ready supply of; raw milk for consumption, processing of other milk products and are easily slaughtered for meat. Thus, it is important to evaluate the constituent of goat milk, since most farmers in Nigeria fall into this category, in other to ensure its safety to the health of the populace.

Antibiotic residues in milk are of great concern to dairy farmers, milk processors, regulatory agencies, and consumers. They emanate from the wide use of antibiotics in dairy animal management for the treatment of disease and as dietary supplements. They may be administered orally, as feed additives or directly by injection, incorrect use of these pharmacologically active drugs causes residues to remain in edible parts. The presence of antimicrobial drug residues in milk can provoke allergic reactions in some hypersensitive individuals as reported by Dewdney et al. (1991) and Dayan (1993) and may induce resistant populations of bacteria that do not respond to treatments commonly used for human illnesses (Nijsten et al., 1993; Van den Bogaard and Stobberingh, 1999). Drug residues also alter the processing qualities of raw milk by inhibiting starter cultures used in the preparation of cheese and other fermented dairy products as presented by Brady and Katz, (1988) or indicate that the milk may have been obtained from an animal with a serious infection (Schenck, Callery, 1998; Choma et al., 1999; Phillips et al., 2004). Pasteurization and other forms of heat treatment eliminate pathogenic microorganisms but have limited or variable effects on drug residues (Moats, 1988).

Penicillin and amoxicillin are classified among the β-lactam antimicrobials including ampicillin, cloxacillin and hetacillin. Penicillins are divided into four classes ranging from narrow to broad range of effectiveness, based on their ability to kill various types of bacteria. Amoxicillin, is a semi-synthetic broad-spectrum penicillin, it is acid-stable, but not penicillinase (or beta-lactamase)-stable.

Penicillin is the antimicrobial for which consultation is most frequently sought through Food Animal Residues Avoidance Databank (FARAD) and is one of the most commonly detected drug residues in tissue and milk. Allergic reactions to foods containing residue concentrations of penicillin are rare and are almost always dermatogetic reactions (Sundlof, 1989). There are, however, reports of anaphylactic reactions developing after consumption of food containing
penicillin residues (Lindemayr et al., 1981). Moats (1988) reported that pasteurization only reduces penicillin residues by approximately 10% to 20%, and penicillin can persist at concentrations that can adversely affect the growth of starter cultures for fermented dairy products (Suhren, 1996).

Although many extra label penicillin products have been approved for cattle, swine and sheep, none are approved for goats. When used IM in sheep at the label dose of approximately 6,600 U/kg, an 8- or 9-day slaughter withdrawal time is required, however data regarding tissue depletion after extra label PPG administration in small ruminants do not exist. Limited plasma data regarding IV administration of penicillin to sheep and goats suggest that penicillin serum half-lives in those species are similar to or shorter than that of cattle (Bengtsson et al., 1997; Schadewinkel-Scherkl, 1991).

However, after IM administration of only 10,000 to 12,000 U of PPG/kg (4,545 to 5,454 U of PPG/lb), measurable residues in goat milk samples persisted through 72 hours (Zeng et al., 1996).

This limited and conflicting data in sheep and goats do not allow FARAD to make recommendations for extra label withdrawal intervals with confidence. Practitioners choosing to treat off-label with PPG should consider testing milk for residues of penicillin prior to marketing.

The presence of drug residues in milk supplies and products is of public health implications and are perceived by consumers as undesirable (McEwen et al., 1991; Bencini and Pulina, 1997). Today, it is the general consensus that even slight traces of antibiotics in milk and food for human consumption should not be tolerated (Jepsen, 1990). Allergic reactions in highly sensitive consumers and potential carcinogenicity, mutagenicity, teratogenicity and long-term toxic effects of the residues of all classes of antibiotics which were presented in a report by Epstein (2000) are of public health concern.

Although reports on antibiotics residues in milk and milk products did not eliminate the antibiotics especially with products that had undergone heat processing like cheese, evaporated milk and yoghurt (O’Keefe and Kennedy, 2008). The presence of residues of these antibiotics in some milk products at a level exceeding the MRLs of FAO/WHO (Joint Food and Agricultural Organisation (1998) in report presented by the Expert Committee on Food Additives Standards is probably due to injudicious use of antibiotics in the treatment of infections in animals and lack of adherence to withdrawal period before milking. Dina and Arowolo (1991) reported the widespread misuse of veterinary drugs in Nigeria due to inadequate monitoring and prescription by untrained personnel. The objective of this study is to evaluate levels of antibiotics (penicillin and amoxicillin) residues in goat milk samples.

**Materials and methods**

**Sampling**

One hundred and sixty-six goat milk (66 West African dwarf and 100 Red Sokoto goats) samples were randomly collected from different locations in Ibadan, Oyo-State. The samples were collected using sterile universal bottles, and transported on ice to the food hygiene laboratory, Department of veterinary public health and preventive medicine, University of Ibadan, Samples were stored at -20°C until analysis.

**Antibiotic residue analysis of samples**

High performance liquid chromatography standard methods were used for antibiotic residue analysis. Penicillin- and amoxicillin were analyzed according to Shaikh and Moats (1993) and Wenhong Luo et al. (1997) respectively.

**Results**

The mean levels for penicillin and amoxicillin red Sokoto goats were 0.2823 ± 0.0123 and 0.1228 ± 0.0057 respectively, while the corresponding values for the West African dwarf goats were 0.2572 ± 0.0194 and 0.1076 ± 0.0058. In the both breeds the amoxicillin levels were significant different (p<0.05) (table 1). The values for penicillin was significantly (p<0.05) higher in both breeds of goat than amoxicillin (figure 1).

**Discussion**

Both breeds presents higher values of penicillin when compared to amoxicillin, this is expected, due to the fact that penicillin is more accessible in Ibadan, South -Western Nigeria where the study was conducted. Penicillin is the first drug of choice in the treatment of bacterial infections in these animals, it is most often used as an extra labelled drug. This could also account for the closeness in the values observed in both breeds, thus, the mean residue of penicillin was not significantly different at P<0.05 level in both breeds. However, the reverse was the case in the mean residue of amoxicillin.

The antibiotics quantification of the goat milk samples was limited to Penicillin and Amoxicillin due to unavailability of other β - lactam antibiotic standards at the time of study.

The result reveals that all samples analysed was contaminated with these antibiotics to an extent. Reports on the antibiotic residue levels of the breeds in this study is scarce, the findings in this study was therefore compared with reports from cow milk. The
values of penicillin in this study were remarkably lower than maximum residues level of Penicillin G (up to 6240±550μg/L) reported in bovine raw milk (Ghidini et al., 2003). These values were within maximum limits of 4μg/L set by the European Union regulations for both antibiotics as presented by Ghidini (2002), 5μg/L as reported by FAO/WHO (1998), 10μg/L as reported by Holstage et al. (2002) and 3.0μg/L as presented by Junqueira and Brito (2006).

Mean Amoxicillin residues in the raw goat milk in the two breeds of goat is also below the ranged values of (8.5±1 to 53.7±2.3μg/L) reported by Cozzani et al. (2005). However, the result of this present study is in line with the MRL’s of 4.0μg/L by Ghidini et al. (2003) and 10.0μg/L by Holstage et al. (2002).

Table 1: Showing Mean antibiotics residues in µg/L

<table>
<thead>
<tr>
<th>Breed of goat</th>
<th>Antibiotics</th>
<th>Mean ± standard error of mean (µg/L)</th>
<th>MRL(µg/L)</th>
<th>Level of significance at P&lt;0.05 with equal variance assumed</th>
</tr>
</thead>
<tbody>
<tr>
<td>Red Sokoto (RS)</td>
<td>Penicillin</td>
<td>0.2823 ± 0.0123</td>
<td>4 µg/L</td>
<td>0.430</td>
</tr>
<tr>
<td></td>
<td>Amoxicillin</td>
<td>0.1228 ± 0.0057</td>
<td>4 µg/L</td>
<td>0.005</td>
</tr>
<tr>
<td>West African Dwarf (WAD)</td>
<td>Penicillin</td>
<td>0.2572 ± 0.0194</td>
<td>4 µg/L</td>
<td>0.430</td>
</tr>
<tr>
<td></td>
<td>Amoxicillin</td>
<td>0.1076 ± 0.0058</td>
<td>4 µg/L</td>
<td>0.005</td>
</tr>
</tbody>
</table>

MRL (Maximum Residue Limit) are as defined by the European Union Regulations (EU)

*Total number of milk samples is 166

**Conclusion**

All the 166 samples analysed were contaminated with penicillin and amoxicillin-antibiotic residues at levels within limit required by international standards. Thus, the goat milk are regarded as safe for consumption. However, there is need for close monitoring so as to prevent values from increasing in the near future due to drug abuse (i.e. indiscriminate use of drugs).

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**Competing interests**

The authors declare that they have no competing interests.
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