RESEARCH Open Access

Oxytetracycline residues in bovine muscles, liver and kidney tissues from selected slaughter facilities in South Western Uganda

Pheonah Kebirungi^{1,5*}, Anthony Nyombi², Timothy Omara^{3,4}, Christopher Adaku⁵ and Emmanuel Ntambi⁵

Abstract

Background: Due to high disease burden and poor animal health services in Uganda, administration of antimicrobials particularly oxytetracycline (OTC) is often done by farm owners and workers without any prescription. This results in misuse of OTC with consequent high chances of antibiotic residues and antimicrobial resistance hence posing public health threat. The degree of public health threat from OTC use is not well established due to limited published data on antibiotic residues and usage in livestock production in Uganda. This study comparatively determined OTC residue levels in 318 samples of bovine muscles, liver and kidney tissues from Kiruhura, Mbarara and Ntungamo districts of South Western Uganda during dry and wet seasons.

Results: The results revealed that the overall OTC residues positivity levels was 74.84% while the district wise rates were 56.88%, 84% and 84.52% for Kiruhura, Mbarara and Ntungamo, respectively. The mean OTC residue levels in bovine muscles, liver and kidney tissues were above the recommended maximum residue limits of 200, 600 and 1200 μ g/kg, respectively as established by FAO/WHO. Of the collected samples, 72.41% (236/318) had OTC residues in concentrations above the recommended maximum residue limits. Wilcoxon signed rank test results showed that change in the seasons did not cause any significant changes in the liver OTC residue levels for all the districts, though this was significant for muscles from Kiruhura and Mbarara districts. Unacceptably high OTC levels were found in the muscles, liver and some kidney samples: Kiruhura muscles and liver samples had mean OTC concentrations of 1094 ± 378 μ g/kg and 1094 ± 198 μ g/kg, Mbarara muscles, liver and kidney samples had mean OTC mean concentrations of 1094 ± 163 μ g/kg, 1140 μ g/kg and 1140 μ g/kg in muscle and liver tissues.

Conclusions: The results of this study indicated that there are unacceptably high OTC residue levels in bovine tissues consumed in South Western Uganda. This poses a public and veterinary health threat to consumers of these bovine tissues.

Keywords: Oxytetracycline, Antimicrobial residues, Uganda, Cattle corridor, Antibiotic misuse

Full list of author information is available at the end of the article

Background

In Uganda, livestock production is an important subsector of agriculture contributing 4.2% to the total Gross Domestic Product (UBOS 2017). The major livestock in Uganda include cattle, sheep, goats, pigs, rabbits and poultry. Economically, cattle are regarded as the most important with significant contributions to the agricultural sector and socio-economic wellbeing of the farming communities in Uganda (Okello et al. 2021). Over 90%



^{*}Correspondence: kebirungi_pheonah@yahoo.com

¹ National Animal Disease Diagnostics and Epidemiology Centre, Ministry of Agriculture, Animal Industry and Fisheries, P.O. Box 513, Entebbe, Uganda

of the cattle herds in Uganda are owned by smallholder mixed farmers and pastoralists who rear them for milk, meat and other products (Nalubwama et al. 2019). Livestock pests and diseases caused by protozoans (39.3%), bacteria (21.4%), viruses (17.1%) and helminths (11.1%) (Byaruhanga et al. 2017; Okello et al. 2021) are the major constraints to cattle production systems in Uganda. Among the livestock diseases, tick-borne diseases are the most challenging to cattle production systems which accounts for 75.4% of losses in cattle (Byaruhanga et al. 2015; Tayebwa et al. 2018; Fuente et al. 2019). In order to maintain health and productivity in animal production, antibiotics are used for treatment of livestock diseases, as animal feed additives and to promote growth in animals (Ndoboli et al. 2019; Nayiga et al. 2020).

Tetracyclines are broad spectrum antibiotics which are widely used in animal production to prevent and treat animal diseases (Basulira et al. 2019). Oxytetracycline (OTC) is one of the affordable over-the-counter tetracyclines most commonly used by cattle keepers in rural households in African countries such as Tanzania (Caudell et al. 2017), Burkina Faso (Samandoulougou et al. 2015), Ethiopia (Agmas and Adugna 2018), Cameroon (Ngom et al. 2017) and Uganda (Basulira et al. 2019; Nayiga et al. 2020). In Uganda, OTC is being used against tick-borne diseases, mainly in areas that lie in the cattle corridor where tick resistance to acaricides has emerged (Vudriko et al. 2016). It has been reported that disease burden can vary between wet and dry seasons (Aalipour et al. 2013). Thus, there is a possibility that the rate of drug administration and amounts used may also vary between seasons (Bangar et al. 2015). In addition, the rate of metabolism and depletion/elimination of drugs from the body is contingent on weather and seasonal variations (Cerveny et al. 2021). This could be attributed to the body water content of the animal that affects drug metabolism and elimination, leading to deposition and prolonged stay of drug metabolites and residues in tissues of animals (Kok-Yong and Lawrence 2015).

In Uganda, there is high involvement of quacks in the treatment of animals thereby allowing drug misadministration and noncompliance with withdrawal periods (Basulira et al. 2019; Musoke et al. 2021). Thus, drug residues remain in animal tissues at the time of slaughter. This practice may cause exposure to drug doses which can contribute to antimicrobial resistance problem. Globally, antimicrobial resistance is an increasingly serious public and veterinary health threat that requires immediate action by the concerned governing bodies (Tiseo et al. 2020). Without effective use of antibiotics, the success of treatment against bacterial infections, major surgeries and cancer chemotherapy is compromised. In addition, the cost of health care for patients

with resistant infections is higher than that of patients with non-resistant infections (FAO/WHO 2017). It is projected that by 2050 there will be more human deaths due to antimicrobial resistance than cancer, with majority being people living in poorer regions of Africa and Asia if no solutions for improper antimicrobial use in both animal and human health care are found (Groot and van't Hooft 2016).

As part of mitigation against antimicrobial resistance, there is a need for regular monitoring and testing of livestock products in order to establish safety of these products for human consumption. There is insufficient information in Uganda on the levels of OTC residues in cattle carcasses. In this study, we report for the first time the levels of OTC in bovine muscles, liver and kidney tissues of carcasses in South Western Uganda. The information from this study may inform regulatory bodies and guide decisions on livestock products.

Methods

Study area

The study was undertaken to establish the levels of OTC residues in slaughtered cattle carcasses in Ntungamo, Mbarara and Kiruhura districts of Southwestern Uganda (Fig. 1). These districts lie in the Ugandan cattle corridor, a South West-North East stretch characterized by high cattle population density and intensive use of antibiotics for treating veterinary diseases (Twongyirwe et al. 2019). The study was undertaken in the three districts due to variations in animal production practices and cultural differences among them.

Slaughter slabs and abattoirs considered in this study and their coordinates were Rubaare (-1.01152, 30.458193), Rwashameire (-0.839318, 30.13335), Nyakabare-Rukoni west (-0.961289, 30.458193), Rwentobo (-1.139585, 30.126516), Nyamunuka (-0.833597, 30.102735), Nyamitanga-Ruti (-0.624069, 30.631354) and Sanga (-0.494615, 30.908075).

Population size and sample size consideration

Twelve (12) slaughter slabs/abattoirs (four from each district) were selected using convenience sampling method. The sample size was determined using Kish's formula (Kish 1965):

$$N = Z^2 pq/d^2$$

where N= sample size, Z= score of confidence interval at 95% (CI=1.96), d=tolerable error (absolute precision) of 5%, p= prevalence of tetracycline residues in tissues and q=(1-p). Using a prevalence of 70% for OTC residues in beef as reported in a recent study from Uganda (Basulira et al. 2019), the sample size was calculated as

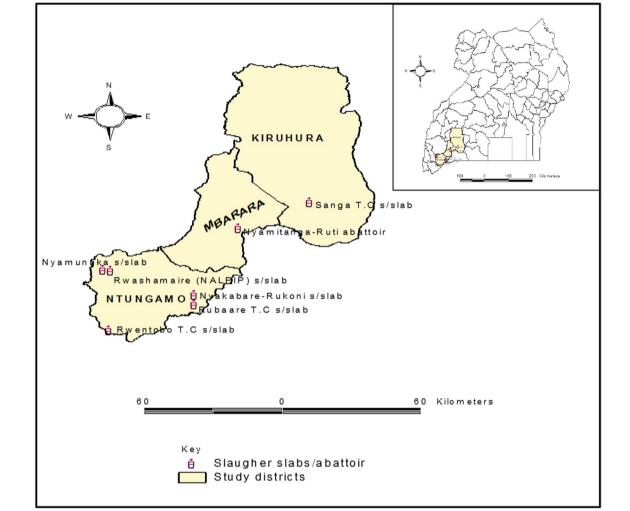


Fig. 1 Map showing the location of the study sites (slaughter slabs/abattoirs) in Ntungamo, Mbarara and Kiruhura districts. Inset is the map of Uganda showing the location of the selected districts

322. We obtained 318 samples of bovine muscles, liver and kidney tissues during the dry and wet seasons, representing 98.76% of the calculated sample size (Table 1).

Study design

Samples of muscle, liver and kidney tissues were collected weekly from carcasses in order to increase sample randomization. Only the slaughter slabs and abattoirs with a capacity of slaughtering more than twenty cattle per day were considered. The selection criteria for the slaughter

Table 1 Samples of bovine muscles, liver and kidney tissues collected during dry and wet season from South Western Uganda (N=318)

District	Muscles		Liver		Kidney		Sub-total	
	Dry season	Wet season						
Kiruhura	28	26	29	26	00	00	57	52
Mbarara	24	24	28	24	02	23	54	71
Ntungamo	21	20	21	20	02	00	44	40

slabs from which the samples were collected were further dependent on their geographical location that is, only those in peri-urban and urban centres were considered.

Two seasons were considered for sampling. These were dry season (September, October, January and February) and wet season (November, December and March) to study if there were any significant spatial differences in OTC residue levels for these two seasons in the districts.

Sample collection, transportation and storage

A total of 318 samples comprising of muscle, liver and kidney tissues were collected from the selected slaughter slabs/abattoirs. It was difficult to obtain kidney samples because of their high demand by consumers. Each sample collected weighed 200 g, was packaged in zip lock polythene bags and stored under ice in a cooler box prior to transportation to the laboratory where they were transferred into a freezer.

Sample analysis

Sample preparation

Samples were prepared and analyzed at the National Animal Disease Diagnostics and Epidemiology Centre, Ministry of Agriculture, Animal Industry and Fisheries, Uganda. A validated analytical method was used in sample processing and analysis. Samples were removed from freezer and allowed to attain room temperature (19-25 °C). The samples were sliced into small pieces and later homogenized in a commercial blender for 2 min. Weighed 5 g of each homogenized sample was transferred into a propylene centrifuge bottle (50 ml). McIlvaine-Ethylene Di-Tetraamine (EDTA) buffer solution (10 ml) was added into the propylene centrifuge bottle and the sample was further homogenized using an ultrasonic homogenizer (model 150, V/T Biologics, INC) for 1 min. The mixture was centrifuged for 10 min at 3500 rpm and the supernatant was collected into a clean centrifuge bottle (50 ml). The sample extraction procedure was repeated to obtain a second supernatant. The resultant supernatants were pooled together and filtered using qualitative Whatmann filter paper prior to solid phase extraction.

The filtrate was cleaned and purified using immunoaffinity columns (200 mg, 6 ml) Waters HLB oasis cartridges. The solid phase extraction cartridges were pre-conditioned using distilled water (3 ml) followed by methanol (2 ml). The sample was washed using distilled water (5 ml) and the extract was eluted from the immunoaffinity columns using 0.01 M methanol-oxalic acid solution (2 ml) into clean labelled beakers. The sample extract was finally mixed using a vortex agitator and further filtered through 0.45 μm nylon syringe filters into

high-performance liquid chromatography (HPLC) amber glass vials (2 ml) ready for HPLC analysis.

HPLC analysis of samples

The analysis and quantification of OTC residues in the sample extracts were done using a HPLC (Perkin Elmer, Flexar) equipped with a constant flow pump and a variable wavelength ultraviolet detector set at 350 nm and at a flow rate of 1.5 ml/min. Elution of the OTC was done using isocratic reverse phase separation mode on a Perkin Elmer Brownie Bio C18 (5 mm, 150 × 4.6 mm) column using methanol, acetonitrile and oxalic acid mixture (1:3:6) v/v, pH 3.85 as the mobile phase. Measured 20 µL of the analyte/sample extract was injected in duplicate to obtain peak areas of positive samples corresponding to retention times of OTC reference standard. For determination of OTC, a blank and OTC standard solutions (20 μL) of varying concentrations at maximum residue limits (MRLs): 0.5, 3.0, 4.0 and 5.0 µg/kg were injected into the HPLC equipment and their peak areas corresponding to retention time of the reference standard were obtained for a calibration curve that was used in quantification of OTC residues in the samples.

Statistical analysis

Qualitative data were analyzed using Stata statistical software (version 10, Stata Corp) in order to compare residue levels in different tissues and seasons for the districts. Quantitative data was analysed using SPSS (version 20, IBM Inc) for Wilcoxon signed rank tests in order to establish if there were any significant differences in OTC residue levels in the muscle, liver and kidney tissue samples for dry and wet seasons for the three districts. All tests were performed at 5% level of significance.

Results

Oxytetracycline positivity rate of the samples

In total, 318 samples were collected and 238 (74.84%) had detectable levels of OTC residues. Of the 318 samples, 236 samples (72.41%) had OTC residues in concentrations above the recommended MRLs. From Kiruhura, muscles (67.8%) and liver (58.62%) samples were positive for OTC residues (Table 2) though no sample for kidney was obtained. For Mbarara, muscles (75%), liver (85.71%) and kidney tissues (100%) were positive for OTC residues. For Ntungamo, muscles (76.19%), liver (76.19%) and kidney (100%) tissues were positive for OTC residues.

For the wet season, muscles (46.15%) and liver tissues (53.85%) from Kiruhura, muscles (87.5%), liver (87.5%) and kidney samples (82.61%) from Mbarara, and muscles (95%) and liver (90.0%) tissues from Ntungamo were positive for OTC residues.

Table 2 Proportion of muscle, liver and kidney samples from Kiruhura, Mbarara and Ntungamo districts of South Western Uganda that contained OTC residues during the dry season (n = 155) and wet season (n = 143)

Season	District	Positive samples (% positive)	Muscle (% positive)	Liver (% positive)	Kidney (% positive)
Dry season	Kiruhura (<i>n</i> = 57)	36 (63.15)	19/28 (67.86)	17/29 (58.62)	NA*
	Mbarara ($n = 54$)	44 (81.48)	18/24 (75.00)	24/28 (85.71)	2/2 (100.00)
	Ntungamo ($n = 44$)	34 (77.27)	16/21 (76.19)	16/21 (76.19)	2/2 (100.00)
Wet season	Kiruhura ($n = 52$)	26 (50.00)	12/26 (46.15)	14/26 (53.85)	NA
	Mbarara $(n=71)$	61 (85.91)	21/24 (87.50)	21/24 (87.50)	19/23 (82.61)
	Ntungamo ($n = 40$)	37 (92.50)	19/20 (95.00)	18/20 (90.00)	NA

^{*}Not applicable as no samples were available

Oxytetracycline residue concentrations in the samples during dry season

As shown in Table 3, 19 of the muscle samples from Kiruhura district which were positive for OTC had a mean OTC concentration of $1094\pm378~\mu g/kg$ while 17 of the liver samples had OTC residues with a mean concentration of $967\pm198~\mu g/kg$. Both mean OTC residue levels were above the MRLs of 200 and 600 $\mu g/kg$, respectively (FAO/WHO 2014). For Mbarara, 18 muscle samples had OTC residues with a mean concentration of $668\pm163~\mu g/kg$. Out of the liver samples collected, 24 had OTC residues with a mean concentration of $3778\pm1140~\mu g/kg$. On the other hand, 2 kidney

samples had OTC residues with a mean concentration of 12,576 \pm 1630 µg/kg, which was tenfold the maximum residue limit of 1200 µg/kg (FAO/WHO 2014). Both the muscle and liver samples had very high mean OTC residue levels, which were above the recommended MRLs (FAO/WHO 2014). For Ntungamo district, 16 muscle samples had OTC residues with a mean concentration of 586 \pm 123 µg/kg, which is above the recommended MRLs (200 µg/kg). For liver tissues, 16 samples had OTC residues with a mean concentration of 5194 \pm 1463 µg/kg which is above the maximum residue limit of 600 µg/kg. Two kidney samples collected had OTC residues with a mean concentration of 969 \pm 187 µg/kg. This mean

Table 3 Oxytetracycline residue concentrations (µg/kg) in muscles, liver and kidney samples from Kiruhura, Mbarara and Ntungamo districts of South Western Uganda during the dry and wet seasons

Season	District	OTC residues	Muscle	Liver	Kidney
Dry season	Kiruhura	Mean±SD	1094±378	967±198	NA*
		Range	33-1505	129-2616	NA
		Percentage positive samples above MRLs	67.86%	58.62%	NA
	Mbarara	$Mean \pm SD$	668±163	3778 ± 1140	$12,576 \pm 1630$
		Range	55-2273	21-26,226	6587-18,565
		Percentage positive samples above MRLs	75.00%	85.71%	100%
	Ntungamo	$Mean \pm SD$	586 ± 123	5194 ± 1463	969 ± 187
		Range	21-3189	25-56,590	738-1201
		Percentage positive samples above MRLs	76.19%	76.19%	0.00%
Wet season	Kiruhura	$Mean \pm SD$	381 ± 159	926 ± 291	NA
		Range	21-1237	112-4365	NA
		Percentage positive samples above MRLs	46.15%	53.85%	NA
	Mbarara	$Mean \pm SD$	1585 ± 447	1367±379	1366±341
		Range	10-6304	49-3616	77–7073
		Percentage positive samples above MRLs	87.50%	87.50%	82.61%
	Ntungamo	$Mean \pm SD$	679±175	744 ± 302	NA
		Range	29-1394	47-3799	NA
		Percentage positive samples above MRLs	95.00%	90.00%	NA
FAO/WHO limit (FAO/WHO 2014)			200	600	1200

 $SD: Standard\ deviation, MRLs:\ Maximum\ residue\ limits.\ ^*NA = Not\ applicable\ as\ no\ samples\ were\ obtained.\ Mean\ values\ in\ bold\ are\ above\ the\ FAO/WHO\ limit\ samples\ were\ obtained.\ Mean\ values\ in\ bold\ are\ above\ the\ FAO/WHO\ limit\ samples\ were\ obtained.\ Mean\ values\ in\ bold\ are\ above\ the\ FAO/WHO\ limit\ samples\ were\ obtained.\ Mean\ values\ in\ bold\ are\ above\ the\ FAO/WHO\ limit\ samples\ were\ obtained.\ Mean\ values\ in\ bold\ are\ above\ the\ FAO/WHO\ limit\ samples\ were\ obtained.\ Mean\ values\ in\ bold\ are\ above\ the\ FAO/WHO\ limit\ samples\ were\ obtained.\ Mean\ value\ samples\ obtained.\ Mean\ value\ obtained.\ Mean\ value\ samples\ obtained.\ Mean\ value\ obtained.\ Mean\ value\$

was below the recommended maximum residue limit of $1200 \mu g/kg$ (FAO/WHO 2014).

Oxytetracycline concentrations in the samples during the wet season

Out of 26 muscle samples collected from Kiruhura, 12 had OTC residues with a mean concentration of $381 \pm 159 \,\mu\text{g/kg}$ which is above the recommended maximum residue limit of 200 µg/kg. For liver samples, 14 out of 26 had OTC residues with a mean concentration of $926\pm291~\mu g/kg$ which surpassed the maximum residue limit of 600 µg/kg (FAO/WHO 2014). No kidney samples were collected during the wet season from Kiruhura (Table 3). For Mbarara, 21 muscle samples had OTC residues with an average concentration of $1585 \pm 447 \mu g/$ kg which exceeded the recommended maximum residue limit of 200 µg/kg. For liver samples, 21 had OTC residues with a mean concentration of $1367 \pm 379 \ \mu g/$ kg which is above the recommended maximum residue limit of 600 µg/kg (Table 3). Nineteen (19) kidney samples had OTC residues with a mean concentration of 1366 ± 341 µg/kg. This was above the recommended maximum residue limit of 1200 μg/kg (FAO/WHO 2014). For Ntungamo district, 19 muscle samples had OTC residues with an average concentration of $679 \pm 175 \, \mu g/kg$, which is above the maximum residue limit of 200 μg/ kg. On the other hand, 18 liver had OTC residues with a mean concentration of 744 ± 302 µg/kg. This also exceeded the maximum residue limit of 600 µg/kg (FAO/ WHO 2014). No kidney samples were collected during the wet season from Ntungamo district.

Variations in oxytetracycline concentrations in the samples during the dry and wet seasons

For samples from Kiruhura, OTC residues for muscles $(p\!=\!0.023)$ during dry and wet seasons were statistically different while the reverse was true for liver samples $(p\!=\!0.475)$. For samples from Mbarara, the concentrations of OTC residues during the dry and wet seasons were statistically different only for the muscles (muscles, $p\!=\!0.024$ and liver, $p\!=\!0.059$). For Ntungamo district, no statistically significant variations (muscles, $p\!=\!0.159$ and liver, $p\!=\!0.232$) in OTC concentrations were obtained for both tissue samples.

Discussion

This study revealed that 74.84% (238/318) of the samples collected and analyzed had detectable levels of OTC. Furthermore, 74.21% (236/318) of the analyzed samples had OTC residue levels above acceptable MRLs of 200 μ g/kg, 600 μ g/kg and 1200 μ g/kg (FAO/WHO 2018). These observations were expected since muscle, liver and the kidneys are the major storage and excretory organs for

tetracyclines and are parenchymatous in nature (Warner et al. 1990). OTC is a lipophobic first-generation tetracycline which is usually not metabolised in animal tissues (Raykova et al. 2021). The occurrence of its residues in animal-derived foods have been reported in other developing countries. For example, Ethiopian studies (Agmas and Adugna 2018; Myllyniemi et al. 2008) indicated OTC residues prevalence of 97.67%, 93.8%, 37.5% and 82.1% in bovine muscles, liver and kidney samples in levels above FAO/WHO maximum limits. In Tanzania, OTC residues were detected in 71.1% of the muscle, liver and kidney tissues sampled from slaughterhouses and butchers, with 68.3% of them being above the acceptable regulatory levels (Kimera et al. 2015). Another study that was carried out in Dodoma in Tanzania showed that 35% of the samples contained OTC residues in levels above the maximum residue limit of 200 µg/kg (Mgonja et al. 2017).

On the other hand, lower positivity rates and concentrations of OTC in bovine tissues than obtained in this study were previously reported. For instance, Ngom et al. (2017) reported an OTC positivity rate and mean concentration of 1.49% and 240 µg/kg (range 0 to 22,000 µg/ kg), respectively in bovine carcasses from Maroua and Godola towns of Cameroon. In Nigeria, Olufemi and Agboola (2009) reported a prevalence rate of 54.44% for OTC residues with 34.44% of these being above the FAO/WHO MRLs. The mean residue concentrations for the positive samples were 51.8 µg/kg for muscles, 197.7 µg/kg for liver and 372.7 µg/kg for the kidney samples which are lower than the levels found in this study. In the neighbouring Kenya, a prevalence of 44% for OTC residues in bovine muscles, liver and kidney samples was reported with mean residue concentrations of 790 µg/kg, 1090 μg/kg and 1380 μg/kg, respectively (Muriuki et al. 2001). Another report from South West Nigeria (Olatoye and Ogundipe 2013) reported the presence of OTC with mean residue concentrations of 1324.7 ± 148.0 , 856.6 ± 118.0 and 651.7 ± 101.3 µg/kg in bovine kidney, liver and beef samples, respectively with 8.7%, 8.6% and 11.2% of these being above the MRLs. In the same year, a significantly lower mean OTC residue concentrations (muscles: $16.17 \pm 5.52 \, \mu/kg$, kidney: $9.47 \pm 3.24 \, \mu/kg$ and liver: $12.73 \pm 4.39 \,\mu/kg$) were reported in the same region of Nigeria (Adesokan et al. 2013). Findings from a study on rural and urban origin beef carcasses in Uganda showed that 6.7% of the samples had unacceptable levels of tetracyclines (Basulira et al. 2019). However, this low positivity rate could be because of the large number of samples considered and the fact that it employed Randox screening method as compared to this study that used a confirmatory HPLC for the detection and quantification of OTC residues. The foregoing study used a screening method and highlights the reason why this

study investigated OTC residues in bovine tissues using a more accurate, specific and confirmatory chromatographic (HPLC-ultraviolet) technique. Overall, the levels of OTC residues in the tissues followed the order muscles>liver>kidneys, which is in agreement with a previous observation (Adesokan et al. 2013).

This study revealed that the number of samples positive for OTC were higher, with high percentage of violative residue levels when compared to that obtained in some countries from which such studies have been reported. For example in Colombia (Correa et al. 2018a), all the 60 animal carcases evaluated had OTC residues and 6% of these exceeded the MRLs set by Colombian standards body. These high levels of OTC residues in the tissue samples could be due to exposure of the animals to antibiotics weeks or even days before slaughter, unauthorized use of the antimicrobials, over use of the drug, inadequate knowledge of the farmers and/or failure to apply instructions on the drug label (Agmas and Adugna 2018; Correa et al. 2018a; 2018b). The differences in the OTC levels recorded in our study and previous studies in Uganda and other countries could be due to differences in the sample sizes considered and the analytical techniques used for the detection and quantification of OTC residues. In addition, this could be due to differences in the climatic conditions, ages and breeds of the cattle slaughtered as well as the drug administration and withdrawal practices among farmers in the different countries (Agmas and Adugna 2018; Basulira et al. 2019; Cerveny et al. 2021; Musoke et al. 2021). This emphasizes the need for using more robust and precise analytical techniques, obtaining adequate number of samples, collecting information on drug administration practices, breeds and ages of the slaughtered animals as well as the climate statistics of the study areas to allow for comparison between studies.

This study has exposed a potentially serious public health threat for consumers of bovine meat products in Kiruhura, Mbarara and Ntungamo districts. During this study however, it was observed that there was no regulation and control of the value chain from the veterinary offices of the districts and other relevant regulatory institutions on where these products are consumed. Therefore, high chances could be that these animal products may end up in other parts of the country where they are consumed hence public health threat due to OTC residues in studied tissues. As such, regulatory bodies should conduct regular monitoring and surveillance on the use of antibiotics on the farms and also educate and enforce adherence to drug withdrawal periods before slaughter of animals in order to ensure suitability of animal products for both local consumption and international trade requirements.

On comparing the OTC levels in the different tissues during dry and wet seasons, this study found that seasonal variations did not play a significant role in OTC accumulation in bovine tissues. For samples from Kiruhura and Mbarara, the concentrations of OTC residues during the dry and wet seasons were statistically different for the muscles but the reverse was true for liver samples. For Ntungamo district, no statistically significant variations in OTC concentrations were observed for both muscles and liver samples. These findings indicate that OTC was used by farmers regardless of seasons for all the districts because in both dry and wet season, the OTC residues were found in the analyzed tissues of cattle slaughtered from the three districts. This could probably be due to the fact that animal diseases presents a challenge to farmers which could have prompted the use of OTC in both seasons (Byaruhanga et al. 2015). These results are in partial agreement with the findings of Olatove and Ogundipe (2013) who unravelled that OTC levels in bovine tissues (kidney, liver and beef) of carcasses from some cities of South West Nigeria varied significantly between the wet and dry seasons. However, the presence of such high levels of OTC residues in these organs indicated that the withdrawal periods were not being adhered to, and that the products were not safe for human consumption. The World Health Organization confirmed that antibiotic resistance is an international health concern, therefore measures should be taken to mitigate the emergence and spread of resistant bacteria through misadministration of both human and veterinary drugs (Okocha et al. 2018; Tiseo et al. 2020).

Conclusions

This study indicated that the overall OTC residues positivity levels was 74.84% while the district wise rates were 56.88%, 84% and 84.52% for Kiruhura, Mbarara and Ntungamo, respectively. Of the collected samples, 72.41% (236/318) had OTC residues in concentrations above the recommended MRLs. The levels of OTC residues in the tissues followed the order: muscles > liver > kidneys. Overall, these high concentrations present public and veterinary health threats. Seasonal variations in OTC residue levels were only significant for bovine muscles sampled from Kiruhura and Mbarara districts.

Abbreviations

HPLC: High-performance liquid chromatography; MRLs: Maximum residue limits; OTC: Oxytetracycline.

Acknowledgements

The authors are grateful to the District Veterinary Officers and Staff of the slaughter slabs and abattoirs from the three study districts. We are grateful to the Ministry of Agriculture, Animal Industry and Fisheries, Uganda for sample processing and analysis. We further acknowledge the contribution of

the entire staff of Chemistry Department, Mbarara University of Science and Technology (Uganda) during the design of this study.

Authors' contributions

PK, CA and EN designed the study. PK collected samples and performed the analytical work. CA and EN supervised the study. PK analyzed the collected data and wrote the first draft of the manuscript. AN, TO, CA and EN reviewed the manuscript. All the authors revised and approved the final manuscript.

Funding

This research received no external funding.

Availability of data and materials

The raw data supporting the conclusions of this study are available from the corresponding author upon request.

Declarations

Ethics approval and consent to participate

This study was approved by the Research Ethics Committee of Mbarara University of Science and Technology, Mbarara, Uganda.

Consent for publication

Not applicable.

Competing interests

The author declares that there is no conflict of interest regarding the publication of this paper.

Author details

¹ National Animal Disease Diagnostics and Epidemiology Centre, Ministry of Agriculture, Animal Industry and Fisheries, P.O. Box 513, Entebbe, Uganda. ² Cranfield University, Shrivenham Campus, Defence Academy of the United Kingdom, Swindon SN6 8LA, UK. ³ Department of Chemistry and Biochemistry, School of Sciences and Aerospace Studies, Moi University, P. O. Box 3900-30100, Eldoret, Kenya. ⁴ Africa Center of Excellence II in Phytochemicals, Textile and Renewable Energy (ACE II PTRE), Moi University, P. O. Box 3900-30100, Eldoret, Kenya. ⁵ Department of Chemistry, Faculty of Science, Mbarara University of Science and Technology, P.O. Box 1410, Mbarara, Uganda.

Received: 8 December 2021 Accepted: 12 January 2022 Published online: 25 January 2022

References

- Aalipour F, Mirlohi M, Jalali M (2013) Prevalence of antibiotic residues in commercial milk and its variation by season and thermal processing methods. Int J Environ Health Eng 2(1):41
- Adesokan H, Agada C, Adetunji V, Akanbi I (2013) Oxytétracycline and penicillin-G residues in cattle slaughtered in southwestern Nigeria: implications for livestock disease management and public health. J S Afr Vet Assoc 84(1):5
- Agmas B, Adugna M (2018) Antimicrobial residue occurrence and its public health risk of beef meat in Debre Tabor and Bahir Dar. Northwest Ethiopia Vet World 11(7):902–908
- Bangar YC, Dohare AK, Kolekar DV, Avhad SR, Khan TA (2015) Seasonal variation in morbidity pattern in cattle by log-linear model approach. J Appl Anim Res 43(3):283–286
- Basulira Y, Olet SA, Alele PE (2019) Inappropriate usage of selected antimicrobials: comparative residue proportions in rural and urban beef in Uganda. PLoS ONE 14(1):e0209006
- Byaruhanga C, Oosthuizen MC, Collinsa NE, Knobela D (2015) Using participatory epidemiology to investigate management options and relative importance of tick-borne diseases amongst transhumant zebu cattle in Karamoja Region. Uganda Prevent Vet Med 122(3):287–297
- Byaruhanga J, Tayebwa DS, Eneku W, Afayoa M, Mutebi F, Ndyanabo S, Kakooza S, Okwee-Acai J, Tweyongyere R, Wampande EM, Vudriko P (2017)

- Retrospective study on cattle and poultry diseases in Uganda. Int J Vet Sci Med 5(2):168–174
- Caudell MA, Quinlan MB, Subbiah M, Call DR, Roulette CJ, Roulette JW, Roth A, Matthews L, Quinlan RJ (2017) Antimicrobial use and veterinary care among agro-pastoralists in Northern Tanzania. PLoS ONE 12(1):e0170328
- Cerveny D, Fick J, Klaminder J, McCallum ES, Bertram MG, Castillo NA, Brodin T (2021) Water temperature affects the biotransformation and accumulation of a psychoactive pharmaceutical and its metabolite in aquatic organisms. Environ Int 155:106705
- Correa DA, Castillo PMM, Martelo RJ (2018a) Beef's antibiotics residues determination from Arjona and Magangué municipalities in Bolívar. Colombia Contemp Eng Sci 11(34):1695–1702
- Correa DA, Margarita P, Castillo PMM (2018b) Quantification of oxytetracycline residues in samples of beef from the municipality of Arjona-Bolivar (Colombia). Contemp Eng Sci 11(45):2249–2255
- FAO/WHO (2014) Report of the Codex Committee on Residues of Veterinary Drugs in Foods. https://www.bing.com/search?q=FAO%2FWHO+ (2014)+Report+of+the+Codex+Committee+on+Residues+of+Veter inary+Drugs+in+Foods.&cvid=8324fc75cf8e4be5811abb2412318d94& aqs=edge..69i57j69i58.919j0j9&FORM=ANAB01&PC=U531
- FAO/WHO (2017) Report of the Twenty-third Session of the Codex Committee on Residues of Veterinary Drugs in Foods. Houston, Texas, United States of America.
- FAO/WHO (2018) Maximum Residue Limits (MRLS) And Risk Management Recommendations (RMRS) For Residues Of Veterinary Drugs In Foods, CX/MRL 2–2018.
- Fuente J, Contreras M, Kasaija PD, Gortazar C, Ruiz-Fons JF, Mateo R, Kabi F (2019) Towards a multidisciplinary approach to improve cattle health and production in Uganda. Vaccines 7(4):165
- Groot MJ, van't Hooft KE (2016) The hidden effects of dairy farming on public and environmental health in the Netherlands, India, Ethiopia, and Uganda, considering the use of antibiotics and other agro-chemicals. Front Public Health 4:12
- Kimera ZI, Mdegela RH, Mhaiki CJ, Karimuribo ED, Mabiki F, Nonga HE, Mwesongo J (2015) Determination of oxytetracycline residues in cattle meat marketed in the Kilosa district. Tanzania Onderstepoort J Vet Res 82(1):911
- Kish L (1965) Survey sampling. Wiley, New York
- Kok-Yong S, Lawrence L (2015) Drug distribution and drug elimination. In: Basic pharmacokinetic concepts and some clinical applications. IntechOpen, pp 99–116
- Mgonja F, Mosha R, Mabiki F, Choongo K (2017) Oxytetracycline residue levels in beef in Dodoma region. Tanzania Afri J Food Sci 11(2):40–43
- Muriuki FK, Ogara WO, Njeruh FM, Mitema ES (2001) Tetracycline residue levels in cattle meat from Nairobi salughter house in Kenya. J Vet Sci 2(2):97–101
- Musoke D, Namata C, Lubega GB, Kitutu FE, Mugisha L, Amir S, Brandish C, Gonza J, Ikhile D, Niyongabo F, Ng BY, O'Driscoll J, Russell-Hobbs K, Winter J, Gibson L (2021) Access, use and disposal of antimicrobials among humans and animals in Wakiso district, Uganda: a qualitative study. J Pharm Policy Pract 14:69
- Myllyniemi AL, Rannikko R, Lindfors E, Niemi A, Bäckman C (2008) Microbiological and chemical detection of incurred penicillin G, oxytetracycline, enrofloxacin and ciprofloxacin residues in bovine and porcine tissues. Food Addit Contam 17:991–1000
- Nalubwama S, Kabi F, Vaarst M, Kiggundu M, Smolders G (2019) Opportunities and challenges for integrating dairy cattle into farms with certified organic pineapple production as perceived by smallholder farmers in Central Uganda. Org Agr 9:29–39
- Nayiga S, Kayendeke M, Nabirye C, Willis LD, Chandler CIR, Staedke SG (2020) Use of antibiotics to treat humans and animals in Uganda: a crosssectional survey of households and farmers in rural, urban and peri-urban settings. JAC-Antimicrob Resist 2(4):dlaa082
- Ndoboli D, Lukuyu B, Wieland B, Grace D, Roesel K (2019) The use of antimicrobials to boost livestock productivity at Ugandan piggery and poultry farms and implications for public health. Paper presented at the Seventh All Africa Conference on Animal Agriculture, Accra, Ghana, 29 July–2 August 2019.
- Ngom RRBV, Garabed RB, Rumbeiha WK, Foyet HS, Schrunk DE, Shao D, Pagnah AZ (2017) Penicillin-G and oxytetracycline residues in beef sold for human consumption in Maroua. Cameroon Int J Food Contam 4:17

- Okello WO, MacLeod ET, Muhanguzi D, Waiswa C, Shaw AP, Welburn SC (2021) critical linkages between livestock production, livestock trade and potential spread of human African Trypanosomiasis in Uganda: bioeconomic herd modeling and livestock trade analysis. Front Vet Sci 8:611141
- Okocha RC, Olatoye IO, Adedeji OB (2018) Food safety impacts of antimicrobial use and their residues in aquaculture. Public Health Revs 39(1):1–22
- Olatoye I, Ogundipe G (2013) Quantitative analysis of oxytetracycline residue in beef and chicken meat from cities of Southwest Nigeria. Bull Animal Health Prod Afr 61(1):39–48
- Olufemi OI, Agboola EA (2009) Oxytetracycline residues in edible tissues of cattle slaughtered in Akure, Nigeria. Int J Food Saf 11:62–66
- Raykova M, Corrigan D, Holdsworth M, Henriquez F, Ward A (2021) Emerging electrochemical sensors for real-time detection of tetracyclines in milk. Biosensors 9(7):232
- Samandoulougou S, Ilboudo AJ, Bagre TS, Tapsoba FW, Savadogo A, Scippo M, Traore AS (2015) Screening of antibiotics residues in beef consumed in Ouagadougou. Burkina Faso Afr J Food Sci 9(6):367–371
- Tayebwa DS, Vudriko P, Tuvshintulga B, Guswanto A, Nugraha AB, Gantuya S, Batiha GE, Musinguzi SP, Komugisha M, Bbira JS, Okwee-Acai J, Tweyongyere R, Wampande EM, Byaruhanga J, Adjou Moumouni PF, Sivakumar T, Yokoyama N, Igarashi I (2018) Molecular epidemiology of Babesia species, Theileria parva, and Anaplasma marginale infecting cattle and the tick control malpractices in Central and Eastern Uganda. Ticks Tick-Borne Dis 9(6):1475–1483
- Tiseo K, Huber L, Gilbert M, Robinson TP, Van Boeckel TP (2020) Global trends in antimicrobial use in food animals from 2017 to 2030. Antibiotics 9(12):918
- Twongyirwe R, Mfitumukiza D, Barasa B, Naggayi BR, Odongo H, Nyakato V, Mutoni G (2019) Perceived effects of drought on household food security in South-western Uganda: Coping responses and determinants. Weather Clim Extremes 24:100201
- UBOS (2017) Statistical Abstract. Uganda Bureau of Statistics. Kampala, Uganda Vudriko P, Okwee-Acai J, Tayebwa DS, Byaruhanga J, Kakooza S, Wampande E, Omara R, Muhindo JB, Tweyongyere R, Owiny DO, Hatta T, Tsuji N, Umemiya-Shirafuji R, Xuan X, Kanameda M, Fujisaki K, Suzuki H (2016) Emergence of multi-acaricide resistant Rhipicephalus ticks and its implication on chemical tick control in Uganda. Parasit Vectors 9:4
- Warner G, Bartels H, Klare P (1990) Detection of inhibitors in animals from normal kills. Svensk-Veterinartidning 19:664–665

Publisher's Note

Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

Submit your manuscript to a SpringerOpen journal and benefit from:

- ► Convenient online submission
- ► Rigorous peer review
- ► Open access: articles freely available online
- ► High visibility within the field
- Retaining the copyright to your article

Submit your next manuscript at ▶ springeropen.com