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

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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 µg/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 µg/kg and 967 ± 198 µg/kg; Mbarara muscles, liver and kidney samples had mean OTC mean concentrations of 668 ± 163 µg/kg, 3778 ± 1140 µg/kg and 12,576 ± 1630 µg/kg, respectively while Ntungamo samples had mean OTC concentrations of 586 ± 123 µg/kg and 5194 ± 1463 µg/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

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%...
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):

\[ N = \frac{Z^2pq}{d^2} \]

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
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 Dry season | Muscles Wet season | Liver Dry season | Liver Wet season | Kidney Dry season | Kidney Wet season | Sub-total Dry season | Sub-total 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-Ethylen 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 \( (n = 57) \) | 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 \( 1094 \pm 378 \) µg/kg while 17 of the liver samples had OTC residues with a mean concentration of \( 967 \pm 198 \) µg/kg. Both mean OTC residue levels were above the MRLs of 200 and 600 µg/kg, respectively (FAO/WHO 2014). For Mbarara, 18 muscle samples had OTC residues with a mean concentration of \( 668 \pm 163 \) µg/kg. Out of the liver samples collected, 24 had OTC residues with a mean concentration of \( 3778 \pm 1140 \) µg/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 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  |
|-----------|--------------|---------------|---------|--------|---------|
|           |              | Mean ± SD     |         |        |         |
| Dry season| Kiruhura     | 1094 ± 378    | 967 ± 198 | NA     |
|           | Mbarara      | 668 ± 163     | 3778 ± 1140 | 12,576 ± 1630 |
|           | Ntungamo     | 586 ± 123     | 5194 ± 1463 | 969 ± 187 |
| Wet season| Kiruhura     | 381 ± 159     | 926 ± 291 | NA     |
|           | Mbarara      | 1585 ± 447    | 1367 ± 379 | 1366 ± 341 |
|           | Ntungamo     | 679 ± 175     | 744 ± 302 | NA     |

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
was below the recommended maximum residue limit of 1200 µg/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 ± 159 µg/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 ± 291 µg/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 ± 447 µg/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 ± 379 µg/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 ± 175 µg/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 µg/kg, 600 µg/kg and 1200 µg/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 Godoua 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 ± 5.52 µ/kg, kidney: 9.47 ± 3.24 µ/kg and liver: 12.73 ± 4.39 µ/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 Olatoye 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.

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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.

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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.

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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.

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