A preliminary study on mycotoxin contamination in red meat from registered abattoirs in South Africa

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Abstract

The frequency of some major mycotoxins in marker tissues (liver and kidney) and in muscle tissue of slaughter pigs and cattle, obtained from registered abattoirs in South Africa, was studied. Samples of each three bovine carcasses were obtained from two abattoirs, and samples of three porcine carcasses were from a third abattoir. All samples originated from animals from subsistence farming. All samples were analysed for aflatoxins (AFB1, AFB2, AFG1, AFG2, deoxynivalenol (DON), ochratoxin A (OTA) and zearalenone (ZEN) using immunoaffinity chromatography extract cleanup and high-performance liquid chromatography (HPLC). At a limit of quantification (LOQ) of 1 μg/kg (individual AFs, 100 μg/kg (DON), 1 μg/kg (OTA) and 20 μg/kg (ZEN)), no mycotoxins were detected in any of the samples.

Keywords: Mycotoxins; Meat; Biotransformation; Food security

Introduction

In South Africa, research on mycotoxins has mostly been done on maize and maize products, sorghum and peanuts (Shephard et al. 2010; Ncube et al. 2010; Janse van Rensburg 2011). Some studies on the occurrence of mycotoxins in animal feed, namely compound feed, feed for poultry and cattle as well as pelleted dog, have also been published (Changwa et al. 2018; Dutton et al. 2012; Mwanza and Dutton 2014; Njobeh et al. 2012; Singh et al. 2017). Mokubedi et al. (2019), in a survey of which included 105 samples of poultry feed, found that positive results for 16 different mycotoxins.

In addition to animal feed, mycotoxins could be analysed in indicator tissues (liver, kidney) and in biological fluids, such as milk and blood (Cimbalo et al. 2020). Using tissue samples as indicator of mycotoxin exposure has several benefits including the accumulation effect of the mycotoxin (Voss et al. 2007). Some toxins, even when fed at low concentrations, persist in the liver and kidneys (Voss et al. 2007). In South Africa, Mwanza and Dutton (2014) found milk both produced by dairy farms from rural subsistence and more sophisticated commercial farms to be contaminated at mean levels of 0.15 μg/kg and 0.14 μg/kg, respectively. Although, except of aflatoxins and OTA, little or no carry-over of mycotoxins from feed into bovine and porcine tissues have been found so far, feeding stuff as available in subsistence farming may contain exceedingly high levels of mycotoxins. Therefore, even low transfer rates of these toxins may be relevant for food safety and animal health (Gallo et al. 2015; Völkel et al. 2011) To date, no study has investigated the occurrence of mycotoxins in mycotoxin marker organs (liver and kidneys) and muscle tissue from bovine and porcine animals from registered abattoirs in South Africa.

Materials and methods

Meat carcasses used in the study for mycotoxin screening were bought at registered abattoirs in South Africa. All carcasses originated from free ranging animals of subsistence rural farmers that were not fed controlled feed and that had been slaughtered during the summer of 2017. Three porcine carcasses were collected from the Vhembe District in Limpopo, and three bovine carcasses were obtained from the Vhembe District and Transkei region of South Africa, respectively. Four subsamples, consisting of each one kidney and liver sample, and two meat tissue samples (chuck and loin/thin flank), each weighing 1 kg, were obtained from each carcass. All samples were transported overnight in cooler boxes with ice packs to the University of Pretoria and frozen at − 18 °C for 3 days and thawed overnight at room temperature until analyses. The chuck and loin/thin flank meat samples were deboned, cubed using a knife and minced. The liver and kidney samples were dissected to remove thick veins and tubes, then cubed and minced. All bovine subsamples were halved to create a total of 48 test samples of 24 identical pairs. One of each bovine sample pair and all porcine samples were cubed and minced then packed in airtight freezer bags, labelled and frozen at − 18 °C for storage and transportation to the analytical laboratory. The other 12 bovine samples were cooked in water at 180 °C until an internal temperature of 70 °C of each respective sample was reached. The cooked samples were weighed, cubed, minced, packed in airtight freezer bags, labelled and frozen at − 18 °C for 7 days until analyses. The delay in analyses was due to logistical arrangements to ensure the integrity of the samples.

All samples were custom-analysed by the SGS Agri Food laboratory (Cape Town, South Africa), which uses routine analytical methods for the main mycotoxins as regulated, for example, by the European Union. Mycotoxins were tested quantitatively using VICAM test kits and high-performance liquid chromatography (HPLC). All samples were respectively tested for the prevalence of aflatoxin (AF) B_1, B_2, G_1, G_2, deoxynivalenol (DON), ochratoxin A (OTA) and zearalenone (ZEN), respectively. Aflatoxin was tested with AflaTest WB which is a quantitative method for the detection of aflatoxin B1, B2, G1 and G2 without the use of chloroform or methylene chloride. First samples were grinded and weighed. Then samples were mixed with an extraction solution (salt, methanol and water), blended and filtered. The extract was then applied to the AflaTest WB column bound with specific antibodies to which the aflatoxin bound. The column was washed to rid the immunoaffinity column of impurities. Last methanol was passed through the column to remove the aflatoxin from the antibodies and the methanol solution was injected into an HPLC system (VICAM

2018a). Deoxynivalenol was tested with DONtest WB, a quantitative method to test samples for the presence of deoxynivalenol in parts per million. Samples were prepared by mixing with an extraction solution, blended and filtered. The extract was then applied to the DONtest WB column which contained deoxynivalenol antibodies to which the mycotoxin bound. The column was washed to rid the immunoaffinity column of impurities. An eluting solution was passed through the column to remove deoxynivalenol from the antibodies, and this eluting solution was then injected into and HPLC system (VICAM 2018b). Ochratoxin was tested with OchraTest WB, a quantitative method for the detection of ochratoxin A in a variety of commodities. Samples were prepared by mixing with an extraction solution followed by blending and filtering. The extract was then added to the OchraTest WB column which contains ochratoxin antibodies. Ochratoxin then bound to these antibodies. The column was then washed to rid the immunoaffinity column of impurities. Methanol was passed through the column to remove the ochratoxin from the antibodies and the methanol was injected to the HPLC system (VICAM 2018c). Zearalenone was tested with ZearalaTest WB, a quantification method for the detection of zearalenone using an HPLC. The samples were prepared by mixing with an extraction solution (90% acetonitrile and 10% water), blending and filtering. The extract was applied to the ZearalaTest WB column which contained antibodies to which the zearalenone bound. The column was washed to rid of impurities. Methanol was passed through to column to release the zearalenone from the antibodies and the methanol was injected to the HPLC system (VICAM 2018d). The limit of quantification (LOQ) and limit of detection (LOD) was 1 µg/kg for AFB_1, AFB_2, AFG_1, and AFG_2 and 4 µg/kg for total AF, 100 µg/kg for DON, 1 µg/kg for OTA and 20 µg/kg for ZEN.

Results and discussion

None of the samples tested was positive for mycotoxins. Similar results have been obtained by the national monitoring and evaluation programme of the Department of Agriculture, Land Reform and Rural Development (DALRRD). This programme continuously and routinely monitored bovine, ovine, poultry and porcine tissue, liver and kidney samples country wide for the prevalence of the mycotoxins OTA, AF and ZEN. The meat samples are either pooled or single-sampled at random by a state veterinarian in the respective districts of sampling. Analysis is performed by the Agricultural Research Council-Onderstepoort Veterinary Institute (ARC-OVI) laboratory according to routine procedures. The data from DALRRD is currently unpublished and the sampling plan from DALRDD was random and sample count limited due to financial constraints. In order to verify their negative test results, a study was proposed with a more complete sampling plan and an independent laboratory. Home-grown maize samples collected from the Vhembe district as well as in the Centane region, formerly known as the Transkei, have shown over many seasons to be contaminated with various mycotoxins, and thus, the decision was made to sample in those two regions for this project (Mngqawa et al. 2016; Shephard et al. 2013).

The absence of mycotoxins in any of the samples in the current study could have several reasons. If none of the animals was fed highly contaminated feed short before slaughter, carry-over rates and tissue half-life time of mycotoxins may be too low to allow their detection with the methods used in this study. One exception would be OTA, which has relevant carry-over and long half-life time in pigs and therefore can persist over a considerable time period in blood and kidneys (Duarte et al. 2011). Further, this study did not include mycotoxin primary or secondary metabolites and reaction products such as aflatoxin M1, aflatoxin DNA, glucuronides and cleavage products.

For example, in bovines, aflatoxin is partly degraded in the rumen, followed by absorption in the intestines after which it is metabolised by the liver. Similarly, the transfer rates of this toxin into edible tissue of pigs and poultry is very low due to a rapid pre-systemic and hepatic metabolism (Fink-Gremmels and van der Merwe 2019).

Even though no mycotoxins were detected in any animal samples in the current study, as well as by the DALRRD monitoring programme, it is still of importance that South African animal feed, mycotoxin metabolic organs (liver and kidney) and meat tissue are screened on a regular, continuous basis. This will help to ensure livestock health, which in turn contributes to food security and to human health.

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Ethics declarations

Ethical clearance, EC005-17, for the project was approved by the Animal Ethics Committee of the University of Pretoria on 27 February 2017. The authors declare that this study had no competing interests or conflicts of interest.

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