Assessment of cadmium (Cd) residues in organs and muscles of slaughtered pigs at Nsukka and environs in Enugu State, Nigeria

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Pork is a major source of protein to man and widely consumed in Enugu State. The present study was undertaken to ascertain the presence and levels of cadmium in muscles and organs of slaughtered pigs at Nsukka agricultural zone. From a total of 160 slaughtered pigs, liver, kidney and muscle samples of each pig were collected and processed for the detection of cadmium residue using atomic absorption spectrophotometer. A prevalence rate of 76.25% was recorded for cadmium residue in Nsukka agricultural zone. There is a strong association (P<0.0001) between occurrence of cadmium residue and the source of samples. The mean concentrations of cadmium in liver (0.041, 0.035 and 0.056 mg/kg) and kidney (0.041, 0.035 and 0.056 mg/kg) from different sources (Nsukka, Orba and Obollo-Afor, respectively) were significantly higher (p<0.05) than what was obtained in muscles and also significantly higher (p<0.0001) than their specific maximum permissible limits (MPL). The levels of cadmium in few samples that exceeded the maximum permissible levels may pose human health threat to pork consumers in the study area.

Key words: Cadmium, kidney, liver, muscle, pig, slaughter house.

INTRODUCTION

Meat and meat products form an important part of human diet. In many African countries, as well as in Nigeria, internal organs (liver, kidneys, heart, and lungs) are sold and consumed as a cherished food source. The risk of
heavy metal contamination in meat is of great concern for both food safety and human health because of the toxic nature of these metals at relatively minute concentrations (Santhi et al., 2008). Heavy metals such as cadmium are naturally occurring elements in the earth’s crust, and thus direct or indirect exposure to them from natural sources is inevitable especially for animals that are not intensively reared. Animals reared on contaminated pasture or fed with contaminated feed become a source of heavy metal residues in edible animal products (meat, fish, egg, milk). Indiscriminate dumping of waste materials on land and water bodies, illegal mining of ores, painting of animals' houses, and methods of processing slaughtered pigs have been incriminated in habitual contamination of animals and their products with heavy metals (Bala et al., 2012). Water bodies located near the abattoir often get contaminated with these hazardous substances through bad abattoir practices, improper management and supervision of abattoir activities. Pigs may drink water from ponds, streams, rivers and other possible contaminated water sources.

Cadmium is the most abundant, naturally occurring element; it was discovered in early 19th century and widely distributed in air, soil, water and plants (Bernard, 2008). Some other sources include cigarette smoking, industrial and agricultural chemicals and contaminated food products. Cereals and vegetables are said to be the major sources in food because of their high consumption rate, followed by meat and offals. It is found in the environment mainly associated with zinc and to a lesser extent with lead and copper. Exposure to cadmium has been associated with hepatic injury, lung damage, hypertension nephrotoxicity, osteoporosis, neurotoxicity, genotoxicity, teratogenicity, and it has been classified as a human carcinogen Group 1 (Huff et al., 2007; Gallagher et al., 2008).

The polluted meats from the edible animal products exposed to heavy metals in the environment are sold in the market for human consumption. It is therefore imperative that this study be carried out with the major aim to investigate the possible presence and prevalence of cadmium residues in organs and muscles of slaughtered pigs in the study area and also, to determine its level (concentration) in the tissues.

MATERIALS AND METHODS

Study area

The study was done in Nsukka agricultural zone of Enugu State, South East Nigeria. Nsukka agricultural zone has three major slaughter houses located at: Nsukka urban with map coordinates of 6°51’24"N and 7°23’45"E; Orba with a map coordinates of 6°51’0"N and 7°27’0"E and Obollo-Afor with coordinates of 6°N and 7°E (Figure 1). Nsukka has a total land area of about 17.5 sq mi (45.38 km²), and has an elevation of 1,810ft (522 m) with a population of 309,633 (National Population Census (NPC), 2006).

Study design

The research work was a four month cross sectional survey and laboratory analysis of post slaughter matrix samples from slaughtered pigs, to determine the presence and concentration of Cadmium.

Sampling technique and sample collection

One (Nsukka) out of the three agricultural zones in Enugu State was randomly selected. The three major slaughter houses (Nsukka urban, Orba and Obollo-Afor) were purposively selected. Stratified random sampling was used to select pigs from each slaughter house assigning them into female and male sex strata and systematic random sampling was used to select 1 in 3 pigs slaughtered from each group, twice a week for four months. A total of 480 fresh samples of liver, kidney and muscle from 160 slaughtered pigs were collected between the months of June, 2014 and September, 2014. Eighty (80) pigs were sampled from Nsukka urban since it has a higher slaughter capacity than the other two. Forty (40) pigs each were sampled from Orba and Obollo-Afor slaughter houses since they have the same slaughter capacity. Age was determined using teeth eruption and wearing. About 50g each of liver and muscle samples and a whole kidney of each selected slaughter pig was packed in sterile polythene bags, labeled and sent to Veterinary Public Health and Preventive Medicine, University of Nigeria, Nsukka for freezing pending analysis. The frozen samples were transported in a cold chain to Springboard Research laboratory, Awka Anambra State, Nigeria, for chemical analysis. Information on the method of processing and the type of materials used was collected by observation and pictures were taken.

Sample processing

Digestion of sample (Dry digestion)

Liver, kidney and muscle samples were dried in the oven at 45°C. After drying, individual sample was crushed into fine powder using mortar and pestle, and 1.0 g of the fine powdered sample was weighed into porcelain crucible and ignited in a muffle furnace at 500°C for 6 to 8 h. The samples were then removed from the furnace and allowed to cool in desiccators, and weighed again. 5 cm cube of 1 M Trioxonitrile (V) acid (HNO₃) solution was added to the left-over ash and evaporated to dryness on a hot plate and returned to the furnace for re-heating at 400°C for 15 to 20 min until perfect grayish-white ash was obtained. The samples were then allowed to cool in desiccators. 15 ml (cm³) hydrochloric acid (HCL) was then added to the ash to dissolve it and the solution was filtered into 100 cm³ volumetric flask. The volume was made to 100 cm³ with distilled water.

Analysis

Cadmium (cd) residues were tested for in the digested liver, kidney and muscle under specified condition using Atomic Absorption Spectrometer (AAS). The procedure was done according to the
manufacturer (AA-6800, Shimadzu Atomic Absorption Spectrophotometer) (Szkoda and Zmudzki, 2005).

**Stock standard solution:** Cadmium, 100 mg/L. Dissolve 1,000 g of cadmium metal in a minimum volume of (1+1) HCl, Dilute to 1 liter with 1% (v/v) HCL.

**Light sources:** Hollow cathode lamps were used for cadmium.

**Data analysis and presentation**

The data generated from the study were statistically analyzed using both SPSS version 17 and GraphPad Prism Statistical software version 5.02 (www.graphpad.com). Gaussian distribution of data sets was tested for, using D’agostino Omnibus Normality test before choosing the most appropriate statistical tests. Chi square analysis was used to determine if there is an association between the occurrence of cadmium residue and the source of samples and type of organ. Analysis of variance and post hoc test were performed to determine if there is significance difference in the mean concentrations of cadmium among various age groups. One–sample t test was used to determine if there is a significant difference in the mean concentration of cadmium in the organs and their specific maximum Permissible Limit (MPL). The alpha value of significance was set at the probability level of < 0.05.

**RESULTS**

**Prevalence of cadmium residue in slaughtered pigs in Nsukka agricultural zone**

Out of a total of 160 pigs sampled, 122 (76.25%) were positive while 38 (23.25) were negative for cadmium residue and from 480 organs sampled (160 each of liver, kidney and muscle from the 160 pigs), 262 (54.58%) were positive for cadmium residue (Table 1).

**Distribution of cadmium in slaughtered pigs in the three slaughter houses**

Forty (50%) out of the 80 pigs sampled from Nsukka slaughter house were positive for cadmium residue, 34 (85%) of 40 pigs in Orba and 38 (95%) of 40 In Obollo Afor slaughter houses were positive for cadmium residue (Figure 2). There was a strong association (p<0.0001) between occurrence of cadmium and the source of samples.

**Organ distribution of cadmium in slaughtered pigs**

In Figure 3, the presence of cadmium was recorded in 86 (53.75%) of liver, 96 (60%) of kidney and 80 (50%) of muscle samples. There is no association (p=0.1926) between occurrence of cadmium and the organ type.

**Comparison of the number of positive samples and the mean concentrations of cadmium in the different organs from different sources with their specific MPLs**

Table 2 shows that out of 160 samples of each organ...
Table 1. Prevalence of cadmium residue in slaughtered pigs in Nsukka agricultural zone.

<table>
<thead>
<tr>
<th>Status</th>
<th>No of Pigs (%)</th>
<th>No of Organ types (%)</th>
<th>Total</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>Liver</td>
<td>Kidney</td>
</tr>
<tr>
<td>Positive</td>
<td>122 (76.25)</td>
<td>86</td>
<td>96</td>
</tr>
<tr>
<td>Negative</td>
<td>38 (23.75)</td>
<td>74</td>
<td>64</td>
</tr>
<tr>
<td>Total</td>
<td>160</td>
<td>160</td>
<td>160</td>
</tr>
</tbody>
</table>

Figure 2. Source/Location distribution of cadmium in slaughtered pigs.

Sample source

<table>
<thead>
<tr>
<th>Sample source</th>
<th>Number</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nsukka urban</td>
<td>40</td>
</tr>
<tr>
<td>Orba</td>
<td>30</td>
</tr>
<tr>
<td>Obollo</td>
<td>30</td>
</tr>
</tbody>
</table>

Processing of slaughter pigs in some of the slaughter houses

Figures 5a to c show different ways slaughter pigs are processed in the slaughter houses. In Figure 5a, old tyre is used to light fire for singeing the pigs. Figure 5b shows the direct use of petrol on the skin of the slaughtered pigs for faster burning and singeing at Orba and Obollo.
Table 2. The number (%) of organs of slaughter pigs with mean concentration above MPL.

<table>
<thead>
<tr>
<th>Sample type</th>
<th>No sampled</th>
<th>Maximum Permissible Limit (MPL)</th>
<th>No (%) below</th>
<th>No (%) above</th>
</tr>
</thead>
<tbody>
<tr>
<td>Kidney</td>
<td>160</td>
<td>156 (97.5)</td>
<td>4 (2.5)</td>
<td></td>
</tr>
<tr>
<td>Liver</td>
<td>160</td>
<td>154 (96.25)</td>
<td>6 (3.75)</td>
<td></td>
</tr>
<tr>
<td>Muscle</td>
<td>160</td>
<td>126 (78.75)</td>
<td>34 (21.25)</td>
<td></td>
</tr>
<tr>
<td>Total</td>
<td>480</td>
<td>436 (90.83)</td>
<td>44 (9.17)</td>
<td></td>
</tr>
</tbody>
</table>

Figure 3. Organ distributions of cadmium.

Table 3. Mean and SEM organ concentration of cadmium in slaughtered pigs according to Age range.

<table>
<thead>
<tr>
<th>Age range of pigs</th>
<th>Organ mean and standard error of the mean (SEM) concentration (mg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Kidney</td>
</tr>
<tr>
<td>0-1</td>
<td>0.2301±0.0916&lt;sup&gt;a&lt;/sup&gt;</td>
</tr>
<tr>
<td>2-3</td>
<td>0.1419±0.0896&lt;sup&gt;b&lt;/sup&gt;</td>
</tr>
<tr>
<td>4-5</td>
<td>0.0174±0.0080&lt;sup&gt;c&lt;/sup&gt;</td>
</tr>
<tr>
<td>≥6</td>
<td>0.0131±0.0068&lt;sup&gt;d&lt;/sup&gt;</td>
</tr>
</tbody>
</table>

Values within same column with different superscripts are statistically different at P < 0.05. SEM: Standard Error of mean.

slaughter houses and Figure 5c shows the use of old plastic bottle to aid the fire for singeing at Orba slaughter house.

**DISCUSSION**

The 76.25% prevalence of cadmium in the study area seems significant and indicative of high exposure of pig consumers to cadmium residue. The prevalence is at par with 65% prevalence recorded by Oladipo and Okareh (2015) in slaughter goats at Ibadan central abattoir but slightly differs from 100% prevalence recorded by Bala et al. (2012) at Sokoto. The parity between the prevalence of cadmium residue in pigs from Ibadan central abattoir andNsukka urban abattoir in this study which recorded 50% prevalence could be likened to the fact that both are located in urban areas and students are taken to those abattoirs for meat inspection and teaching indirectly creating awareness, because of their proximity to the
University, Orba and Obollo Afor slaughter houses are interiorly located and so are exposed to more mundane singeing practices and higher rate of exposure to cadmium due to the processing materials or methods (fueling of wood with kerosene, fuel, plastic and tyre) used in singeing the slaughtered pigs as also reported by Ekenma et al. (2014). Also, awareness of the dangers of such practices is low as such exposure to cadmium due to such practices may be due to ignorance on the part of the butchers. Hence, there is a strong association between the occurrence of cadmium residue and location/source of samples.

Although it has been reported that cadmium residue accumulates more in kidney and liver (Nwude et al., 2010; El-Salam et al., 2013) as also detected in this work owing to the fact that kidney and liver are organs of biotransformation and detoxification, but no association was found between the type of tissue and the occurrence of cadmium residues. The non association could be likened to the singeing practices which makes accumulation of cadmium in the muscle almost as high as in the internal organs (kidney and liver). Accumulation in internal organs (kidney and liver) occur due to oral exposure (consumption of cadmium contaminated food and water). Animals, especially free range pigs are exposed to heavy metals in our local environment through scavenging in open waste or refuse dumps, and polluted drinking water (Obiri-Danso et al., 2008; Okoye and Ugwu 2010). Okoye and Ugwu (2010) also reported high levels cadmium in soils from Enugu State which they claimed could serve as a source of heavy metals in animals grazing in such areas of the State. Although the rate of occurrence of cadmium residue is more for the more interiorly located sites, Nsukka recorded a higher range value and mean concentration of cadmium than the other locations (Orba and Obollo). This may be attributed to the higher level of industrial activities (mechanics and automobiles engines) observed in Nsukka than Obollo Afor and Orba.

The higher levels of cadmium recorded in organs in the present study is similar to what was reported by Bala et al., 2012 in pigs slaughtered at Nasarawa State. The number of positive samples (2.5 and 3.75%) out of 160 samples of each organ, with concentrations higher than their respective MPL as recommended by the EC (2011) for kidney (1.0 mg/kg) and liver (0.5 mg/kg) are small and the mean concentrations of the two organs in the different locations are significantly lower than their respective MPLs. The number of muscle samples with concentrations higher than its MPL is high at 21.25% but the mean concentration for both Nsukka and Orba slaughter houses are significantly lower than the MPL. There is no significant difference in mean concentration of cadmium in muscle samples from Obollo and its MPL; again, this could be due to the processing method recorded in this study of the use of petrol, rubber and plastic to singe the pigs as shown in the pictures. The results from this study implies that Cd accumulates more in kidney compared to other organs and is in agreement with some other studies (Iwegbue, 2008; Rahimi and Rokni, 2008; Bala et al., 2012).

The result shows that the mean concentrations of cadmium decreased as the age increased, mean cadmium concentrations in age range 0 to 1 in kidney
(0.2301 mg/kg), liver (0.0907 mg/kg) and muscle (0.0679 mg/kg) was significantly higher than the age range 2 years and above. The reason for the variation in age is because younger pigs have immature metabolic rate, innate curiosity and active calcium absorption mechanism. Generally, mean values for cadmium in all the organs and
muscles of pigs slaughtered in the study area were below the European commission and WHO recommended maximum permissible level except in the muscle of age range 0 to 1 year, which had higher mean concentration than its MPL, but the difference is not statistically significant. The fact that the mean concentration of the muscle tissue is higher than its MPL, although not significant, it may still pose serious public health threat to consumers considering the high rate of exposure recorded in this study. Consumption of cadmium has also been reported to have a known bio-importance in human biochemistry and physiology and consumption even at very low concentrations can be toxic (Nazir et al., 2015).

Conclusion

It is clear from this study, that heavy metals bioaccumulate in different concentrations in organs and muscles of pigs. The level of cadmium in pigs slaughtered at Nsukka, Orba and Obollo Afor varied with majority of the samples falling below the MPL. It could also be concluded that the cadmium residue accumulates more in kidney and liver than the muscle although higher mean concentration was seen in muscle tissue. Younger pigs are more prone to cadmium residue accumulation than the older ones. The potential risk posed by cadmium bioaccumulation and toxicity may continue to increase unless adequate environmental control measures are put in place and enforced by public health authorities. Therefore, to protect public health and ensure food safety; routine monitoring measures for heavy metal residues should be put in place and enforced by the government to guarantee food safety for consumers.

Conflict of Interests

The authors have not declared any conflict of interests.

REFERENCES


