



Cyanide, Nitrates and Nitrite Contents of Livestock Feeds in Umuahia, Nigeria.

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Abstract

The cyanide nitrate and nitrite contents in pig feeds from six different farms in Umuahia, Nigeria were assayed using spectrophotometric and enzymatic methods. Enzymatic analysis of the feed samples indicated mean total cyanide level of $59.30 \pm 3.72 - 361.90 \pm 11.2 \text{mg}^{-1} \text{DM}$. The mean cyanide content of cyanogenic plant materials used in the feed formulation ranged from $85.60 \pm 13.2 - 888.10 \pm 23.17 \text{mgkg}^{-1}$. Observation was made that the cyanide measured in these feed samples were higher than the recommended dietary cyanogens levels for both non-ruminants and ruminants; $100 \text{mg HCN equivalent kg}^{-1}$ for cassava ration. The mean levels of nitrate and nitrite ranged $25.5 \pm 2.76 \text{mg} - 285.5 \pm 12.70$ and $2.20 \pm 0.3 - 218.80 \pm 13.42 \text{mg kg}^{-1} \text{DM}$ respectively. The toxicological and nutritional significance of the results obtained are discussed.

Keywords: Cyanide, nitrate, nitrite, animal feeds.

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INTRODUCTION

The high demand for cereals by increasing human population and their use by millers for compounding livestock feed coupled with the need for livestock products have led to the use of unconventional feeds for animal production (Iyayi and Tewe, 1994). These unconventional feed materials include sorghum spent grains and wheat offals (by-products of sorghum and wheat malting respectively) as well as cassava peels. One of the factors that limits the use of these products as livestock feeds is their content of cyanogenic glycosides which on hydrolysis by endogenous β -glycosidase enzyme yield hydrogen cyanide (Conn, 1969) which is a potent poison (Coursey, 1973).

The use of cassava formulation for livestock feed dated back to 1903 when Tracy reported the use of this root crop for swine feeding. Despite overwhelming evidence from two decades of satisfactory performance of animals fed on cassava chips in European community countries, arguments still persist in the scientific circles as to the safety of cassava for sustainable livestock production (Tewe, 1994). The work of Osuntokun (1970) on Wistar rats substantiates the claim of the unsuitability of cassava as animal feed. His investigation revealed that when rats were fed "puru - puru" a cassava product for a period of 18 – 24 months, plasma thiocyanate levels were raised and rats developed ataxic and segmented demyelination similar to that which is found in patients suffering from Tropical Ataxic Neuropathy (TAN). Other studies (Charavanaparavana 1944; Ekpechi, 1967, 1969; Delange, and Ahiluwalia 1983) constitute strong circumstantial evidence which have continued to influence current thinking on the safety of cassava for human and livestock feeding.

Moreover, there is now an increasing evidence that thiocyanate, the main cyanide metabolite can lead to the promotion of N-nitrosamine formation in vivo which enhances carcinogenesis (Maduagwu and Umoh 1988; Okoh, 1992.). However, deaths reported in livestock fed forage sorghum products, fresh or processed cassava roots or leaves have been sporadic. In most instances, dietary cyanide exposure from cassava or sorghum has been implicated without the determination of the amount of cyanogens in the materials thus making the evidence circumstantial.

This paper describes the cyanide, nitrate and nitrite contents of pig feeds in six farms in Umuahia, Abia State-Nigeria.

MATERIALS AND METHODS

Feed Samples

Animal feed samples and the raw materials used for their formulation were collected from six different Pig Farms that are involved in semi-intensive animal production in Umuahia, Abia State-Nigeria. These included Michael Okpara University Farm, designated (A), Umunwanwa Farm (B), Ngodo Farm (C), Umuopara Farm (D), Ubakala Farm (E) and Afara Farm (F).

Feed composition.

Feed sample from farm A contained sorghum spent grains (25%), wheat offals (25%), soybean cake (15%), fish meal (2%), palm kernel meal (25%), bone meal (3%), Oyster shell (2%), salt (0.25%), vitamin min-Premix (2.5%), methionine (0.1%) and Cysine (0.1%). Feed sample from Farm B is made up of 80% fermented spent grains and 20% other constituents while feed sample from Farm C contained 75% wheat offals and 25% palm kernel. Feed sample from Farm D contained cassava peels (50%), palm kernel cake (25%) and sorghum spent grains (25%). Feed samples

from Farms E and F were cassava peels (50%), palm kernel cake (25%) and soybean meal 25% for E and sorghum spent grains (>90%) for F.

Analytical Procedure

Cyanide determination.

Cyanide content of each feed sample was extracted according to the method of O'Brien *et al* (1991) and their cyanogens contents determined spectrophotometrically using exogenous β -glucosidase (Ikediobi *et al* 1980).

Nitrate and nitrite determination.

Nitrite content of the samples was determined spectrophotometrically by the method of Follet and Ratcliff (1963) and nitrate after reduction to nitrite using cadmium column based on the same method.

Statistical analysis: Student's t-test was used for statistical comparisons.

RESULTS AND DISCUSSION.

The mean total cyanide content of the feed samples ranged from $59.30 \pm 3.72 - 361.90 \pm 15.9 \text{ mgkg}^{-1} \text{ DM}$ (Table 1). The feed sample D (from Umuopara Farm) contained the highest level of mean total cyanide; $361.90 \pm 15.9 \text{ mgkg}^{-1} \text{ DM}$ while the least of mean cyanide $59.30 \pm 3.72 \text{ mgkg}^{-1} \text{ DM}$ was measured in feed sample C from Ngodo Farm (Table 1). The cyanide levels in these feeds reflect the percentage of cyanogenic plant materials that make up the rations. The levels of mean cyanide content of cassava peels, sorghum spent grains and wheat offals that make up high percentage of the feed samples were 888.10 ± 23.17 , 127.9 ± 13.07 , and $85.6 \pm 1.32 \text{ mgkg}^{-1} \text{ DM}$ respectively (see Table 1). For example, while the feed sample from Umuopara Farm contained 50% cassava peel, 25% palm kernel cake and 25% spent grains (ie, 75% of the feed materials are from cyanogenic

plants), the feed samples C 75% wheat offals and 25% palm kernel cake.

The cyanide content of the cassava peels used as component of these feeds contained about $888.1 \pm 23.17 \text{ mg kg}^{-1} \text{ DM}$ while sorghum spent grains contained $127.90 \pm 13.07 \text{ mg kg}^{-1} \text{ DM}$ (see Table 2).

There was a statistically significant difference ($p < 0.01$) between the levels of cyanide content of feed sample C and the others. Feeds sample A which is from Michael Okpara University Farm contained $197.50 \pm 10.3 \text{ mgkg}^{-1} \text{ DM}$ cyanide. This value is higher than the recommended cyanide level for animal rations; less than 100 mgkg^{-1} (Wood, 1992). Levels of dietary cyanide as low as $40-60 \text{ mg HCN equivalent kg}^{-1}$ in fermented cassava product called "puru-puru" have been reported to cause neurological abnormalities in humans and rats (Osuntokun, 1970). Elevated serum thiocyanate and reduced thyroxin have also been reported in pigs fed cassava peel-based diets containing $96 \text{ mg HCN equivalent kg}^{-1}$ (Tewe, 1994) while elevated placenta thiocyanate transfer and elevated milk thiocyanate were reported from cassava diets containing $500 \text{ mg HCN equivalent kg}^{-1}$ (Tewe and Maner, 1981).

The most consistent report on the levels of dietary cyanogens tolerated in animal feeds is that from European Community countries where cassava has been used in livestock rations for the past two decades. Wood (1992) stated that a level of $100 \text{ mg HCN equivalent kg}^{-1}$ was the highest, which could be tolerated in cassava chips imported into European Community countries. This report agrees with earlier report of Bolhuis (1954) which gave a level below $50 \text{ mg HCN equivalent kg}^{-1}$ as safe. According to Bolhuis, $50 - 100 \text{ mg HCN equivalent kg}^{-1}$ was set as moderately toxic and above $100 \text{ mg HCN equivalent kg}^{-1}$ as dangerously toxic. Based on the experience from European Community countries; it has

been suggested that a level of cyanogens below 50mg HCN equivalent kg^{-1} should be the target for cassava based rations. Thus, the cyanide contents of the feed samples under study could be said to be quite high. It is note worthy that the animals in most of the Farms in this study are rarely given veterinary care. Thus, poisoning from cyanogens even if it occurs are not taken into account.

Since these animals depend solely on plant protein, it is necessary that such feed diets be enriched with protein supplements. The need to supplement cassava diets with methionine and cysteine has been demonstrated for monogastric species (Maner and Gomez, 1973); the role of these supplements in cyanide detoxification for ruminants has been demonstrated (Tewe 1981).

Table 1: The Cyanide contents of compounded pig feeds and some plant materials used for their formulation

Feed Sample	Mean non-glucosidic cyanide conc. (mg/kg DM).	Mean glucosidic cyanide Conc. (mg/kg DM).
Farm A	79. 70 \pm 5. 30	197. 50 \pm 10. 30.
Farm B	92. 40 \pm 7. 3	105. 1 \pm 8. 91
Farm C	55. 90 \pm 5. 6	59. 3 \pm 8. 91
Farm D	118. 6 \pm 12. 7	361. 9 \pm 15. 9
Farm E	42. 4 \pm 3. 31	144. 9 \pm 11. 2
Farm F	218. 8 \pm 13 42	44. 9 \pm 5. 0
Cassava peels	561. 3 \pm 18 74	888. 10 \pm 23. 17
Wheat offals	54. 2 \pm 6. 32	85. 60 \pm 13. 2
Sorghum spent grain	55. 1 + 6. 32	127. 9 + 13 07.

Note: Each value is $\times \pm$ s.d calculated from five (5) determinations.

The mean levels of nitrate and nitrite measured in these feed samples ranged from 25.5 ± 2.76 – 285.5 ± 12.70 and 2.2 ± 0.3 – 218.8 ± 13.42 mg kg^{-1} respectively. The lethal dose of sodium nitrite for pigs is 88mg kg^{-1} body weight (Jones, 1988) and doses of 48 – 77mg kg^{-1} cause moderate to severe but total methemoglobinemia (Norton and Campbell 1990). Nitrite is known to interact with haemoglobin, forming methemoglobin by oxidation of ferrous iron to ferric state, preventing or reducing the ability of blood to transport oxygen, a condition known as methemoglobinemia (Philips, 1971; Bruning-Farn and Kaneene, 1993; Jones 1993).

Nitrite is also involved in the formation of nitrosamines, compounds known to be carcinogenic, mutagenic, embryopathic and teratogenic in experimental animals (Gray et al; 1979). The ingestion of 8 – 15g of nitrate can cause severe gastroenteritis with abdominal pain, blood in stool and urine, weakness and collapse. Chronic ingestion of smaller doses can cause dyspepsia, mental depression and headache (Magee, 1983). Though pigs are highly susceptible to nitrite poisoning, the results from this study indicate no possible acute effect on the animals fed on them.

Table 2: Nitrate and nitrite contents of pig feeds in Umuahia, Nigeria

Feed sample	Mean NO_3 Conc. (mg/kg DM)	Mean NO_2 Conc. (mg/kg DM)
Farm A	266.60 ± 9.7	12.20 ± 1.32
Farm B	262.22 ± 11.70	173.0 ± 8.71
Farm C	154.3 ± 9.3	101.10 ± 7.4
Farm D	25.5 ± 2.76	2.20 ± 0.3
Farm E	102.22 ± 11.3	54.4 ± 3.51
Farm F	285.5 ± 12.7	218.8 ± 13.42

Note: Each value is $\bar{x} \pm s.d$ determined from 5 determinations.

In conclusion, the results from this study indicate that the unconventional feeds mainly cassava peels, sorghum spent grains and wheat offal being used in some animal farms in Umuahia, Abia State, Nigeria contain high levels of cyanide, nitrate and nitrite. The levels of these compounds in animal feed should be of much concern considering their toxicological implications for both animals and humans who consume these animals.

References

- Bolhuis, G. G.** (1954). The toxicity of cassava root. *Netherland Journal of Agricultural Science* **2**: 176 – 185.
- Bruning-Fann, C. S., Kaneene, J. B.** (1993). The effects of nitrate, nitrite and N-nitro-compounds on human health; a review. *Vet. Human. Toxicol.* **35**:521 – 538.
- Charavanaparavana, C.** (1944). Studies in manioc and lima beans with Special reference to the utilization as harmless food. *Tropical Agriculture* **100** (3):14 – 168.
- Conn, E. E.** (1969) Cyanogenic glycosides. *J. Agric. Fd Chem.* **17**: 519
- Coursey, D. G.** (1973). Cassava as food. Toxicity and technology. In chronic cassava toxicity. Proceeding of an international workshop, London, 1973 Ottawa Canada Inter develop. Res. Center. 1973. pp 27 – 36.
- Delange F and Ahiluwalia R.** (1983). Cassava toxicity and thyroid research and public health issues. Ottawa Canada, Inter Develop. Res. Center. Monogr. 1983 10 RC – 207e.
- Ekpechi, O. L** (1973). In chronic cassava toxicity (Nestel, B. L and R. Macintyre Eds) 10RC 010e, Ottawa, Canada 29 – 30 Jan. 1973
- Ekpechi, O. L** (1969). Pathogenesis of endemic goiter in Eastern Nigeria. *Br. J. Nutr.* **21**: 537 – 45.
- Follet, M. J. and Ratcliff, P. N** (1963). Determination of nitrite and nitrate in meat products. *J. Sci. food Agric.* **14**:138 – 144.
- Gray, J. I; Irvine, D. M, Kakuda, Y** (1979). Nitrate and N-nitrosamines in cheese *J. Food Product* **42**:263 – 272.
- Ikediodi, O. C, Onyia, G. O. Eluwa, C. E.** (1980). A rapid and inexpensive enzymatic assay for total cyanide in cassava (*Manihot esculenta* Crantz). And cassava products. *Agric. Biol. Chem. Japan.* **44** (12):2803 – 2809.
- Iyayi, E.A and Tewe, O.O** (1994) Cassava feeding in small holder livestock units. *Acta Horticulturae* **375**:261-269.
- Jones, T.** (1993). Poison: nitrate/nitrite. In pract. **15**, 146 – 147. Maduagwu, E. N. and Umoh I. B (1988). Dietary thiocyanate and N-nitrosation in vivo in the wistar rats. *Ann Nutr. Metab.* **32**:30 – 37.
- Magee, P. N.** (1983). Nitrate. In Environmental Aspect of Cancer. The role of macro and micro components of food; Wynder E. L, Leveille, G. A., Weisburger, J. H., Livinstone G. E., Eds; food Nutrition press: Westport, C. T, 1983.; pp 198 – 210.
- Maner, J. H and Gomez G.** (1973). Implications of cyanide toxicity in animal feeding studies using high cassava rations in chronic cassava toxicity: Proceedings of an interdisciplinary workshop, London, England, 29 – 30 January 1973. IDRC 010e p 113 –120.
- Norton, J.H. and Campbell R.S.F.** (1990); *Vet Bull.* **60**: 1137

O'Brien, G. M., Taylor, A. J., and Poulter, N. H. (1991) Improved Enzymatic assay for cyanogens in fresh and proceeded. *J Sci. Food Agric.* **56**, 277 – 289.

Okoh, P. N. (1992). The metanoliclate of cyanide carbon in animals. *Nig. J. Phs. Sci.* **8**(1 – 2) :1 – 9.

Osuntokun, B. O., (1970). Cassava diet and cyanide metabolism in Wister rats. *British Journal of Nutrition* **24**: 377.

Philips, W. E. J. (1971). Naturally occurring nitrate and nitrite in foods in relation to intantic methaemoglobinaemia. *Food Cosmet. Toxicol.* **9**, 219 – 228.

Tewe, O. O. (1981). Supplementation of cassava/urea based rations with elemental sulphur for sheep and goat: effect on growth, ruminal, serum and urinary thiocyanate

concentrations. *Nutrition Reports International* **24** (6): 1263 – 1269.

Tewe, O. (1994). Indices of cassava safety for livestock feeding. *Acta Horticulture* **375**, 241 – 249.

Tewe, O. O., and Maner, J. H (1981). Performance and patho- physiological changes in pregnant pigs fed cassava diets containing different levels of cyanidde. *Research in Veterinary Science* **30**: 147 – 151.

Tracy, S. M., (1903). Cassava. USDA Farmers Bulletin, 167. Washington, 31pp.

Wood, J.F. (1992) Quality aspects of tradable cassava products including problems of aldustration in Roots, Tubers, Plantain and Bananas in animal feeding (Ed. D. Machin and S. Nyvold) FAO Animal production and health paper, 95: 67-80.