The Levels of Antinutritional Factors in *Moringa Oleifera* and *Vernonia Amygdalina* Leaves Found in Some Part of Plateau State, Nigeria

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Abstract

In this research work, various materials were used and they include; apparatus such as fume cupboard, measuring cylinder, spatula, centrifuge, pipette and reagent such as Na$_2$CO$_3$, CaCl$_2$, H$_2$SO$_4$, HCl, AgNO$_3$. The nutrient composition of *V. Amydalina* are: phytate 11.9 ± 0.01 (mg/100g), oxalate 244.02 ± 0.57 (mg/100g), tannins 1.28 ± 0.50 (mg/100g), alkaloid 1.66 ± 0.01%, HCN 2036.00 ± 0.58 (mg/100g) and *M. oleifera* are: phytate 10.58 ± 0.01 (mg/100g), oxalate 334.33 ± 0.67 (mg/100g), tannin 8.19 ± 0.01 (mg/100g), alkaloid 1.72 ± 0.01% and HCN 3998.30 ± 0.49 (mg/100g). These results showed that *V. Amydalina* leaves could be a bio resources for Zn as a result of the low level of phytate. But they are not Cu bioavailable resources due to the high level of oxalate. Hence, people are encouraged to utilize *V. amygdalina* and *M. oleifera* leaves as a good source of micronutrient particularly those prove to be bioavailable.

Introduction

Antinutrient refers to substance that reduces the nutrient utilization by binding with the mineral to form complexes that are readily indigestible. Antinutrients are found at Some level of antinutrients may be found in almost all food for many reasons. Nowadays, the levels may be reduced in modern crops due to process domestication (Geo, 2008). Anti-nutrients in food are responsible for deleterious effects related to the absorption of nutrients and micronutrients. However, some anti-nutrients may exact some beneficial effect (Welch and Graham, 2004). The basis of these anti-nutrients elicits harmful and beneficial biological active compounds which,
in recent times, revealed their functions in several biological compounds (Igile, 1996). It is generally recognized that plant hold antinutrients obtained from pesticides, fertilizers and chemicals with occur naturally (Igile, 1996). Secondary metabolites types of antinutrients are recognized to contain high biological activity (Zenk, 1991). Phytate, alkaloids, oxalate, saponins, flavonoids, hydrogen cyanide were shown to be present.

Moringa is being harvested for its high nutritious content. Both the edible leaves and flowers may be used as a source of food and medicine and may also be used as cosmetic oil or forage for livestock. Moringa have the height of about 5 to 10cm (Padayachee and Bajinath, 2012). Through research, the moringa was found to contain many essential nutrients, for instance, vitamin, amino acid, beta-carotene, antioxidant, anti-inflammatory nutrient and omega 3 and 6 fatty acids (Kasolo et al., 2010).

**Materials and Methods**

**Collection and Preparation of Plant Materials**

The leaves samples of *Moringa oleifera* and *Vernonia amygdalina* were obtained from farm. The leaves were planted and harvested, destalked and washed with a clean cold water. The leaves were dried under a sun shade and the dried leaves were pulverised in a porcelain mortar, and the sample were stored in plastic container. The powdered samples were used for the analysis.

**Methods**

Various methods were reported by different researchers for the quantification of the antinutritional factors in green vegetable leaves. Oxalates were determined according to Underwood and Day (1986) procedure, phytate, Maret and Sandstead (2006) method, tannins, (Aletor, 1993) procedure, alkaloids, Henry (1973) method and cyanogenic glucosides, alkaline titration method of AOAC (1990).

**Phytate Content Determination**

The method reported by Maret and Sandstead (2006) was adapted for phytate quantification. Powdered sample (4g) were soaked in 100 cm³ of 2% HCl (w/v) and allowed to stand for over 3 hours before filtration. From the filtrate, 25 cm³ was taken and placed in a conical flask, 5 cm³ of 0.3% NH₄SCN (aq) and 53.5 cm³ of distilled water were mixed together and titrated against standard FeCl₃ (aq) having 0.00195g Fe/cm³ and observed the formation of brown-yellow color which may persist for 5 minutes. Blank was treated in a similar manner.

**Determination of Hydrogen Cyanide**

The methods proposed by AOAC (1990) were adapted. 100 cm³ of the leaves has undergone steam-distillation using sodium hydroxide solution (NaOH). KI solution was used for treatment the distillate. The distillate was titrated against 0.02N AgNO₃ solution. The end point was recorded immediately when the color changed to more turbid solution. Determination of HCN content was evaluated by measuring 1ml of 0.02N AgNO₃ as equivalent of 1.08mg hydrogen cyanide.

**Oxalate Determination**

Day and Underwood (1987) proposed a method for determination of oxalate. The leaves sample should be (1g) and put inside a 100cm³ flask. 75cm³ of 3MnH₂SO₄ was measured, placed into the same conical flask and stirred for about 1 hour. The filtration of the solution was carried out using a Whatman No 1 filter paper. 25cm³ of the filtrate was measured and titrated over 0.05N potassium permanganite (KMnO₄) solution till the appearance of pale-pink color. 1ml of 0.05m KMnO₄ was used to calculate the oxalate content.

**Tannins Determination**

0.1g of the powdered leaves was placed inside 100cm³ conical flask and 350cm³ of distilled water added (Aletor, 1993). The flask was gently heated to boiling for 1 hour, filtered hot and the filtrate was collected in a 50cm³ volumetric flask. The residue was washed severally, and distilled water was used
top the volume of the combined solution. To 1, 2, and 3cm³ of the standard tannic acid and 10cm³ of the sample solution in a 50cm³ volumetric flasks, 2.5cm³ of Folin-Denis reagent and 10cm³ of Na₂CO₃ solutions has been added and distilled water was used to top the volume of the solution. The flask was allowed to stand for 20 minutes after which optical density was measured at 760nm.

Results

Table 1: Antinutrient in the Moringa oleifera leaves

| Antinutrients   | Composition (Mg/100g) |
|-----------------|------------------------|
| Oxalate         | 334.33±0.67            |
| Phyate          | 10.58±0.01             |
| Tanins          | 8.19 ±0.01             |
| Alkaloids       | 1.72±0.01              |
| Hydrogen cyanide| 3998.30±0.49           |

Table 2: Antinutrients in the Vernomia amygdalina leaves

| Antinutrients   | Composition (Mg/100g) |
|-----------------|------------------------|
| Oxalate         | 334.33±0.67            |
| Phyate          | 10.58±0.01             |
| Tanins          | 8.19 ±0.01             |
| Alkaloids       | 1.72±0.01              |
| Hydrogen cyanide| 3998.30±0.49           |

Discussion

From table 1 and 2 values are calculated as Mean ± SEM (n=3)

From the Table 1 and 2 above, the Oxalate content of Vernomia amygdalina and Moringa oleifera, 244.02±0.57 mg/100g and 339.33±0.67 mg/100g were high compare to 202.50 ± 6.50 mg/100g reported for A. viridis leaves (Umar et al., 2007). Studies revealed that antinutrients were observed to chelate several elements like Zn, Fe and Ca with strong binding affinities and therefore diminishes their bioavailability. High levels of some antinutrients e.g oxalate and phytates in human food are considered undesirable. High level of oxalate, decreases Cu bioavailability by binding with it to form insoluble salt (Umar et al., 2007).

The susceptibility to hydrogen cyanide toxicity in ruminants is higher than non-ruminants. Hydrogen cyanide
cyanide (HCN) is converted to thiocyanate (SCN) by detoxification of the absorbed hydrogen cyanide in the liver by the enzyme Rhodanese. Cytochrome oxidase could be inhibited by excess cyanide ion. Thus stopping ATP formation, while the tissues deprived of energy with lead to rapid death. For sheep and cattle, the sufficient limit to cause death is is 2.0 to 4.0mg/kg body weight (Kumar Jha et al., 2013). Cyanide can cause goitrogenic effects due to During thiocyanate production by detoxification, CN can cause an effect called goitrogenic (Kumar Jha et al., 2013). Hydrogen cyanide (HCN) content of V. Amydalina and M. oleifera, 2036.00±0.58 mg/100g and 3993.30±0.49 mg/100g were very high compare to 13.07 ± 2.38 mg/100g reported for A. viridis (Umar et al., 2005), (Whitney and Rolffes, 2010) reported HCN content in some raw leaves such as Celosia argentea (200 mg/100g), Tartinum triangulare (75mg/100g) and Celosia laxa (300 mg/100g). These showed that the level of HCN were above the permissible range for human consumption, hence will lead to serious health problem. Neurological disorder and gastrointestinal disorder are all cause by alkaloids. Chaconine, solanine and glycoalkaloids found in potato and Solanum spp. Were reported to be haemolytically active and are harmful to fungi and human beings. Gastrointestinal and neurological disorders are the toxicological consequences of potato glycoalkaloids involved more especially when the doses is above 20 mg/100g sample (Kumar Jha et al., 2013). Alkaloids content of V. Amydalina and M. oleifera, 1.66±0.01 % and 1.72±0.01 % were very low compare to 20 ± 6.5% reported for Solanum spp (Saeto, 2008). These showed that the alkaloid content is safe for consumption.

Conclusion
It has been revealed that, on the general basis, plants contained anti-nutrients obtained x pesticides, fertilizer and various acquiring chemicals that are found in nature. It is also known that high level of these antinutritional factors leads to bad effects upon the bioavailability of many minerals when consumed. However, it can be inferred from the results above that, V. Amydalina and M. oleifera leaves could be a bioresources for Zn, because of the low value of phytate content. But they are not Cu bioavailable resources due to the high level of oxalate. Hence, populace are encouraged to utilize V. Amydalina and M. oleifera leaves as a good sources of micronutrient particularly those proves to be bioavailable.

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Conflict of interest.
No conflict of interest declared.

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