CYANOGENIC PLANTS OBSERVED IN SOKOINE UNIVERSITY AGRICULTURE FARM, MOROGORO, EASTERN TANZANIA

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SUMMARY

Identification of plants with cyanogenic potential and determination of their cyanogen content qualitatively and quantitatively was carried out in Sokoine University of Agriculture (SUA farm. The two studies involved field survey to identify the presence of these plants in the farm where livestock graze. The plants identified were Cassava plant, *Cynodon plectostachyus, Cynodon dactylon, Sorghum spp, Zea mays, Eucalyptus spp, Acacia spp* and bamboo plant. Some leaves, seeds, cassava tubers and their peels were collected for cyanide analysis in qualitative and quantitative scales using picrate paper and isonicotinate 1,3-dimethyl barbiturate methods respectively. All of the identified plants proved to contain cyanogenic glycosides except the *Acacia* spp and bamboo plant. With these results it is concluded that plants with cyanogenic potential exist in SUA area. Under normal conditions they are not hazardous but under certain conditions such as drought and rapid growth in early rains, they become potentially cyanogenic. The analysis of cyanide in the leaves needs further investigation in order to provide better methods which will give accurate results.

INTRODUCTION

Cyanide, hydrocyanic acid (HCN) or prussic acid are terms relating to the same toxic principle. HCN is a colourless or pale blue liquid or gas which has the odour of bitter almonds. It is one of the quickest acting poisons available to mammals. It reacts readily with ferric (trivalent) iron of the cytochrome oxidase forming a cyanide -cytochrome complex which is quite stable. Since iron is maintained in the trivalent state, electron transport stops and cellular respiration is brought to a halt. Thus cyanide causes tissues anoxia. Cyanide is present in inorganic salts or plants as naturally occurring glycoside or as free hydrocyanic acid (Salkowski and Penny, 1994).

Cyanogenic plants, like other poisonous plants, grow together with pastures in the plant communities and hence putting grazing livestock at some risk. There are extensive areas of natural pastures existing in the tropical and subtropical regions of Africa, Asia, Latin America and Australia; these support large herds of cattle used for various purposes including dairy and beef. However the productivity of these herds is generally low when expressed on per capita or per hectare basis (Butterworth, 1985). This is because the presence of poisonous plants limits the use of pastures for livestock production.

The presence of poisonous plants has been linked to massive losses of livestock which occur in areas where animals are traditionally kept under extensive conditions. Such areas are frequently denuded by droughts, overstocking and uncontrolled fires (Colegate and Dorling, 1994). Devastating outbreaks of plant poisoning have been reported to have occurred in the North Western Cape Province of South Africa during 1926 and 1927 whereby about 600,000 sheep died of plant-induced photosensitization and over one million were killed in 1929 and 1930 by Geigeria spp (Kellerman et al., 1988). Other outbreaks have been reported occurring throughout the year but more frequently in late winter, spring and early summer (Colegate and Dorling, 1994).

There is very little information on poisonous plants available in Tanzania. Few poisonous plants were identified in Mtwara and Lindi, the southern regions of Tanzania (Otaru et al., 1986). Such plants include: *Albizia adianthifolia, Schenia racemoce and Setaria megaphyla.* Around Sokoine University of Agriculture (SUA) campus some poisonous plants which were thought to be associated with impairment of animal health have been identified (Mosha, 1985). Such plants include: *Lantana camara, Ricinus communis, Solanum incanum* and some *mycotoxins.*

Due to scarcity of information on poisonous plants in Tanzania, losses emanating from cyanide toxicity have been difficulty to quantify.

This work was initiated in order to provide information on the existing cyanogenic plants within SUA. The specific objectives were to identify cyanogenic plant poisoning within SUA farm and to determine the amount of cyanide present in the plants on a qualitative basis at various stages to growth. Cassava tubers and peels were also analyzed quantitatively. The information obtained might shed light on how best to tackle the problem of poisonous plants within SUA and Tanzania as a whole.

MATERIALS AND METHODS

Site:
The Sokoine University of Agriculture (SUA) farm in Morogoro district of Tanzania was the site of the study. The farm is 1725 feet above mean sea level and it is located within the Latitude 06° 50' 20"S and Longitude 37° 39' 20"E. The farm covers an are of 2,300 ha. There are two main seasons in the year - the wet season and the dry season. The wet season extends between December and May with short rains being received in November and December and the long rains in the period March to May. The mean rainfall during the wet season is 696 mm. This is about 83% of the total annual rainfall. The long rainy season is the main crop growing season. The dry season extends from July to November with an average of 17% of the annual rainfall being received during this period.

Sample Collection:
The samples included plant parts such as leaves, tubers and seeds from various plants within the farm. The plants sampled include those known to be cyanogenic and also those known to be toxic but with unknown toxic principle; others were those plants which were evergreen throughout the year and were suspected to be poisonous. Leaf sample collection was done during young stage before flowering. Collection of the various samples was done during wet and dry seasons. The samples were analysed in the laboratory on the same day and immediately after collection.

Laboratory methods
For qualitative determination of cyanide in plant parts the "picrate paper" method as described by Osweiler et al. (1995) was used. Strips of moistened picrate paper were suspended over crushed sample in a water bath which was heated to 30-35°C. A well marked turning of the suspended picrate paper into brick-red colour within a few minutes indicated presence of cyanide in the sample. For quantitative test of cyanogenic material in plants, the method of Essers et al. (1993) was used. The methods allows quantification of HCN by means of colourimetric reactions using isonicotinate/1,3- dimethyl barbiturate as reagent. In order to determine the cyanogenic potential of the root or processed product the glucosidic and intermediate cyanogens must be converted to free cyanide. This is achieved by enzymatic hydrolysis of cyanogenic glycosides and the manipulation of pH within the assay system in order to convert cyanohydrins into hydrogen cyanide which can then be quantitatively determined.

RESULTS

The field survey confirmed the presence of the following cyanogenic plants within SUA farm:-

(a) *Maniholt esculenta* crantz (eassava)
(b) *Cynodon plectostachyus*
(c) *Cynodon dactylon*
(d) *Sorghum* spp
(e) *Zea mays* (maize)
(f) *Eucalyptus* spp
(g) *Acacia* spp
(h) Bamboo plant

The results of the picrate test for samples of plants mentioned above are shown in Table 1. From the table it is seen that the cassava plant, *Cynodon dactylon* and *Sorghum* spp changed the picrate paper to brick-red. *Zea mays* and *Eucalyptus* spp. plants showed very little change of colour to brown. *Acacia migrescens* and bamboo leaves did not change the colour of the picrate paper at all.

Table 2 shows the results of the quantitative analysis of cassava tubers and peels. Cassava leaves were not quantitatively analyzed for cyanogens because the methods of analysis does not give reliable results for leaves (Mlingi, Tanzania Food and Nutrition Centre, personal communication).

DISCUSSION

According to the picrate paper test a change to brick-red colour on the suspended picrate paper within a few minutes indicates the presence of cyanide in the heated plant sample. A mild reaction in one to several hours shows that the plant sample contains insignificant amounts of cyanogens while the absence of colour change indicates that the plant sample doesn't contain cyanide (Osweiler et al, 1985). Cassava rhizome peels, tubers and leaves changed the picrate paper to brick-red in a few minutes and hence contained cyanide. The cassava plant is a herbaceous semi-shrubby perennial whose tubers and leaves are edible by man and livestock. So the cyanide needs to be removed before the tubers or leaves are eaten by man or livestock. *Cynodon plectostachyus* and *Cynodon dactylon* both of which are stoloniferous and rhizomatous perennials change the picrate paper to brick-red in a few minutes implying that they contain cyanide. Similarly leaves of sorghum from young plants were positive with the picrate test and hence contained cyanide. This implies that young sorghum plants are not suitable livestock fees. Sorghum seeds however did not contain cyanide. Young leaves from *Zea mays* and *Eucalyptus* spp. tested sparingly positive for cyanide particularly during the dry season. This is probably because they concentrate the cyanide during the dry season.

Legend for table 1; Strips of moistened picrate paper were suspended over crushed plant samples in water bath heated to 30-35°C. Turning of picrate paper into brick-red monitored. +++=positive, change to brick-red in 3-5 minutes, ++=positive; change in 5-10 minutes, +=positive; change in 10-15 minutes, ±= sparingly positive; change to brick-red in 15-20 minutes.

Table 1: Qualitative cyanide content of some plants in SUA farm in the wet and dry season

Plant species/part	Wet	Dry
1. rhizome, Manlhot esculent crantz	+++	+++
a) Peels	++	++
b) Tuber	+++	+++
c) Leaves		
2. Cynodon plectostachyus	+++	+++
a) Young leaves	+++	+++
b) Old leaves	-	-
c) Dry leaves		
3. Cynodon dactylon	-	++
a) Fresh leaves	-	-
b) Dry leaves		
4. Sorghum spp		
a) Leaves - young	+	+
plants	+	-
b) Leaves before	-	-
flowering	-	-
c) Leaves at flowering and after		
d) Seeds		
5. Zea mays		
a) Leaves - young	±	+
plants	-	-
b) Leaves at	-	-
flowering and after	-	-
c) Young seeds (undried)		
d) Dry seeds		
6. Eucalyptus spp		
a) Young leaves	-	±
b) Seeds	-	-
7. Acacia migrescens	-	-
a) Leaves		
8. Bamboo tree		
a) Leaves	-	-

Table 2: Cyanogen content of fresh root and peels of cassava* (mg CN equiv./kg)

Sample type	Free Cyan ogens	Inter media te Cyan ogens	Glycos ides	Total Cyan ogens°
Fresh root	2.7	7.9	83.6	94.2
Fresh peel	6.0	27.5	1173.6	1207.1

Legend: *Values are means of duplicates
° Total cyanogens consists of free cyanide, intermediate cyanogens and glycoside cyanide.

Although bamboo plant is known to contain the cyanogenic glycoside p-glucosyloxy-mandelonitrile in the young leaves (Lundqnist, 1992), it appears that the one existing within SUA does not contain cyanogen. This is probably due to the soil types which does not favour the synthesis of the glycoside in this plant. *Acacia* spp (Fabaceae) i.e. *A. arioloba* (camel thorn), *A. sieberana* and *A. caffra* (hook thorn) are known to be associated with prussic acid poisoning (Kellerman et al., 1988). That is to say they are potentially cyanogenic. However, the acacia plants existing within SUA area i.e. *A. nigrescens* and *A. polyacaritha* tested negative with the picrate paper. This suggests that the plants are not cyanogenic; probably these two species do not synthesize glycoside at all or may be SUA conditions influenced by the soil type, rainfall and humidity do not favour HCN accumulation in the plant.

The *Eucalyptus* spp existing within SUA area has shown insignificant change of colour in the picrate test. This suggests that the cyanogen content of the plant is insignificant. This is in contrast to what has been said in the literature that *Eucalyptus cladocalyx* (sugar gum) is potentially cyanogenic (Kellerman et al. 1988; Lundquist, 1992) and that cyanide poisoning of goats had resulted from the ingestion of folliage of the tree.

The total cyanogen of fresh cassava peels was about twelve times greater than that of fresh cassava roots (Table 2). Qualitatively cyanogenic glycosides were not uniformly distributed in the various tissues of the cassava plant. The highest concentration was found in the peel of the tuber followed by the leaves and the lowest in the central pith of the tuber (Table 1). These results agreed with those obtained by Gondwe (1974) in his experiment in which he found that the concentration of cyanogenic glycoside in the tuber increases from centre outwards (Gondwe, 1974). Other authors (Mlingi, 1995; Wood 1965) have also described that the HCN content of the peel is higher than that of the flesh. It is suggested that most of the cyanogenic glycoside in cassava (linamarin) is stored in the peel after it has been synthesized in the leaves.

Young leaves contain higher levels of cyanogenic glycoside than the older ones. The intensity of colour change of the picric acid impregnated paper was deep brick-red when compared to that of the older ones. Young plants are therefore potentially more cyanogenic than older ones. This can be due to fast growing of the leaves which have more nitrogen for chlorophyll pigment synthesis while at the same time nitrogen is one of the principal precursors for the synthesis of cyanogenic glycoside. The rapidly growing cyanogenic plants have been described to have the highest amount of cyanogenic glycosides when compared to older plants (Lundquist, 1992; Mlingi, 1995).

The cassava leaves tested for cyanide during dry season contained qualitatively higher amount of glycosides than those tested during wet season and so were the rhizome tubers and the peels. Probably the higher cyanide content in the cassava leaves, tubers and peels during dry season is a result of concentration. As with cassava the two species of Cynodon tested had high concentration of cyanogenic glycosides in the young leaves probably for the same reasons.

In sorghum and maize the highest concentration of the glycosides was found in leaves in young shoots. No cyanogens were detected in seeds. These results agree with findings by other authors (Kellerman et al., 1988; Osweiler et al., 1985).

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