

contain Chl a and b. That HHDB does not may be due to FDB probably being killed rapidly by frost before translocation or chemical alteration of the Chl, whereas in a hot-house, under humid conditions, these processes would have taken place by the time the bract becomes dried material. The relatively high content in Chl b of FDB is different from the ratio usually found in Chl-containing tissues (Allen, 1966; Comar and Zscheile, 1942; Robinson, 1967; Shlyk, 1971; Strain and Svec, 1966) and in green bract; this may have implications for the function of cotton bract and of Chl b, which is presently thought to be biosynthesized from Chl a (Shlyk, 1971).

The waste obtained from ginning cotton is often made into pellets which are used as feed for animals. This waste material contains bract, and bract has been shown to contain chlorophyll, which can be readily converted to pheophytin. It has been suggested that pheophytin may bind Cu, making it unavailable for utilization by cattle, etc., and therefore accounting for the observations that the Cu status of cattle falls during the summer grazing period and improves during winter, even in diets that contain less Cu (Mills, 1964). Copper pheophytin has been shown to be readily formed (Mills, 1964) after treatment of aqueous acid suspensions of grass with ionic Cu. The isolated product is stable even in the presence of sulfide and chelating agents and decomposes appreciably to ionic Cu only below pH 3.2. It is not known whether chlorophyll and magnesium can be released from dried bract by the surface of lung tissue as it can from plant tissue under the physiological conditions of the rumen. In addition, the effect of pheophytin, magnesium, or chlorophyll on lung tissue is not presently known nor has any connection between these materials and byssinosis been investigated. On the other hand, magnesium poisoning in humans has been reported (Wacker and Parisi, 1968). Since FDB contains chlorophyll and pheophytin, which are highly reactive substances, it is suggested that it may be worthwhile to

investigate whether these materials can be released from bract by lung surface tissue and if so to investigate the effect of these compounds on lung surface lining to determine if these materials can contribute to the acute response of byssinosis.

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## Rapid Field Method for Evaluating Hydrocyanic Toxicity of Cassava Root Tubers

A simple and rapid method for field evaluating the hydrocyanic toxicity of cassava roots is described. The method uses the blue copper-ben-

zidine reaction, is reliable, and can be performed in a few minutes in the field.

An important part of the crop diversification program at the Cocoa Research Centre (CEPEC) is the selection of new cultivars for cassava (*Manihot esculenta* Crantz). However, the considerable variation in the HCN toxicity levels of cassava root tubers has led to the development of a quick field method for evaluating this toxicity, especially in view of the large number of selections being studied. The toxicity level is often further complicated by the prevailing ecological conditions and the management system used for the cultivation of cassava.

The conventional method of using sodium picrate paper strips is tedious and requires some laboratory equipment which is difficult to take to the field. In addition, the 3 hr needed for the full chemical reaction to become apparent does not lend itself to field conditions, where a quick yet accurate response is preferred.

An adaption of the benzidine blue test (Feigl, 1954), which is accurate down to 0.25  $\mu$ g of HCN, gave good results when employed for field work with cassava root tubers. No interfering substances are present. The results are reported in the present paper.

## MATERIALS

For the experimental work, squares (3 cm  $\times$  3 cm) of Whatman No. 1 filter paper, a dropper flask (all glass eyedropper), and a sharp knife were used.

## REAGENTS

The reagent used is made up of two parts: solution A consists of 1.43 g of cupric acetate  $\cdot$  H<sub>2</sub>O dissolved in 500 ml of distilled water, while solution B consists of 275 ml of saturated benzidine acetate solution mixed with 225 ml of

**Table I. Relation of Color Intensity and Yield of HCN for Cassava Root Tubers<sup>a</sup>**

Variety	Yield of HCN in ppm by AOAC method	Degree of toxicity	Color intensity by benzidine blue method
Palmeiras	377.5	Poisonous	Intense blue
CEPEC 62	313.0	Poisonous	Strong blue
IAC 780	200.0	Poisonous	Medium blue
Itapecuru	68.5	Poisonous	Weak blue
Engole boi	41.0	Tolerable	Very weak blue
CEPEC	27.0	Innocuous	Almost no color

<sup>a</sup> Few examples of 22 HCN determinations.

distilled water. Solution B should be stored in a dark flask.

Since benzidine acetate is not easily found, weigh 2 g of pure benzidine and warm up with 5 ml of a 50% aqueous solution of acetic acid. Add 500 ml of distilled water, agitate for 15 min, let stand for 5 min, and decant. Use this as saturated benzidine acetate solution.

The reagent to be used in the field is made up from equal parts of solutions A and B. Preferably this final reagent should be made up on the day of use, since it can not be kept for more than a week.

#### PROCEDURE

The cassava root tubers were either examined in the field or taken to the laboratory, depending on the distance. A drop of the reagent was put in the center of the filter paper square, and as it soaked into the paper, the blot became dome shaped. The filter paper was placed over a slice of cassava root of at least 1 cm thickness so that the blot's dome remained without contact with the surface of the slice. The dome functions as a trap for the HCN gas.

The speed at which the reaction occurs and the color intensity are proportional to the amount of HCN present in the root slice. To quantify the HCN toxicity, the speed of the reaction, the halo around the blot, and the color intensity (termed as either strong, medium, weak, or uncol-

ored) are all observed. The entire test is completed in 3 min.

#### RESULTS AND DISCUSSION

In order to evaluate the practicability of the above method for cassava root tubers, 22 different cultivars were also tested for HCN using the alkaline determination method (AOAC, 1970). The correlation between the two methods was extremely good (Table I), indicating the relationship between the amount of HCN found according to the AOAC test and the color intensity as developed from the benzidine blue method.

It is said by some (Albuquerque, 1969) that 500 ppm of HCN in plant material is potentially lethal, whereas 50 ppm is innocuous. These values are naturally liable to change according to the analytical method used for their determination, government regulations, and the presence or absence of active enzymes (present in cassava) which liberate the HCN during some food processing procedures. Kingsbury (1964) suggested that more than 20 mg per 100 g may be potentially dangerous, and this certainly may be true for some dry seeds. In the case of cassava root tubers, the benzidine blue test would appear to be an excellent method for evaluating HCN levels, as it is quick, reliable, and can be easily performed when necessary in the field itself.

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#### Correction

#### IDENTIFICATION OF THE GAS CHROMATOGRAPHIC DIELDRIN AND ENDRIN PEAKS BY CHEMICAL CONVERSION

In this article by D. W. Woodham, C. D. Loftis, and C. W. Collier [*J. Agr. Food Chem.* **20**(1), 103 (1972)], in Figure 5, tracings (a) and (b) should be reversed.