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Effect of Harvest and Cook Processing on Cyanides Content of Cassava Cultivars and Cassava-Based Dough Consumed In Lomé, Togo

[1]T. Tchacondo, [1]S.D. Karou, [1]E. Osseyi, [1]A. Agban, [1]L.M. Bawa, [1]A.R. Atcha, [2]K. Soumana, [2]K. Assignon, [2]A. Kpemoua and [1]C. De Souza
[1]Ecole Supérieure des Techniques Biologiques, Alimentaires et(ESTBA-UL), Université de Lomé, Togo
[2]Institut Togolais de Recherche Agronomique (ITRA), Lomé, Togo

Abstract: Cassava is a staple food for many populations, but it can be a source of poisoning due to its content of cyanide (CN). This study aimed to evaluate the influence of harvest and cook processing on the reduction of cyanide content in cassava-based foods. Ten cassava cultivars were collected from the National Collection in Togo. The CN content was measured to examine the influence of the following factors: cultivars, time of harvest, drying, grinding, boiling flour and the mixture of flour with corn flour. The results showed significant variation of CN content in the range of 375±3.32 and 27±14.28 mg/kg, within cultivars. The maximum CN content was obtained when the crop was harvested during the dry season (375.3±3.32 mg/kg) and the minimum in the rainy season (93.05±2.35 mg/kg). A circadian variation was also observed, with high concentration in the morning (67.3±1.75 mg/kg) and low concentration in the evening (50.55±4.32 mg/kg). For the same cultivar, the cyanide content of fresh tuber was at least two times greater than that of the dried one and five times higher than that of the dough. The mixture of cassava flour with corn flour significantly reduced the CN content of the dough. But this decline was stabilized for corn flour above 40%. Thus, by acting on the various studied factors, the amount of cyanide in cassava-based food can be sufficiently reduced.

Key words: Cassava cultivar, cyanide, dough, harvest time

INTRODUCTION

Manihot esculenta Crantz., commonly named cassava, is a plant cultivated for its tuberous roots. The plant adapts to various climates and its harvest can be delayed for a long time. This makes cassava, a very important food reserve during the dearth season, shortage or famine (FAO, 2000). Indeed the plant contributes to solve food insecurity problems and poverty in rural areas in Africa. Nutritionally, it is a major source of calories in the form of starch.

In Togo, cassava is consumed in various forms and it is the second most commonly consumed tuber after yam (Agbobli *et al*., 2007). However, the roots of the cassava contain cyanogenic glycosides that can potentially release cyanide ions (Cliff *et al*., 2011). These cyanides are known by far to be toxic for human. In fact, cyanide (CN) in the body, directly or indirectly interferes with the functioning of certain organs and enzymes (Okolie and Osagie, 1999; Rocha-e-Silva *et al*., 2010). Goitre, cretinism, tropical ataxic neuropathy or spastic paralysis are among the diseases attributed to the toxic effects of CN diets, dominated by root of cassava non or poorly treated, and of low protein content (Cliff *et al*., 2011).

Therefore, the elimination of CN is very crucial in cassava processing (Nambisan, 2011). Traditional methods proposed for this application have required long time treatment (Lü *et al*., 2009). However, during famine, these methods are ignored by the population and Cassava is not treated prior to processing and consumption (Bradbury and Denton, 2010; Mlingi *et al*., 2011; Cliff *et al*., 2011). In this way, CN content in the food only depends on cooking method, exposing consumers to CN intoxication (Nambisan, 2011).

The present study aimed to evaluate CN content in several cassava cultivars. Furthermore, CN rates were evaluated in function of harvest time and cooking method.

MATERIALS AND METHODS

Plant materials: This study was conducted in the Department of Food Science and Technology - Agro alimentary Industries, ESTBA-UL of University of Lomé, Togo, from January to July 2010. Ten cassava cultivars, Manihot esculenta Crantz, namely 96/1632-ITRA-TG, 96/0409-ITRA-TG, Ganavé-ITRA-TG, Sorade (Local), 312/524-ITRA-TG,96/1642-ITRA-TG, Som/51/03-ITRA-TG, 96/0603-ITRA-TG, 95/0166-ITRA-TG and Gbaze

Corresponding Author: Karou D. Simplice, Ecole Supérieure des Techniques Biologiques Alimentaires et (ESTBA-UL), Université de Lomé UL, BP 15.15-Lomé, Togo,

Table 1: Cyanide content of ten cassava cultivars

	Methods	
Cultivars	Silver Nitrate HCN (mg/kg)*	Bradbury Kit HCN (mg/kg)
96/1632-ITRA-TG	375.3±3.32	200-400
96/0409-ITRA-TG	185.4±16.49	100-200
Ganavé-ITRA-TG	170.1±11.45	100-200
Sorade (Local)	145.8±14.28	100-200
312/524-ITRA-TG	122.4±8.25	100-200
96/1642-ITRA-TG	118.8±9.35	100
Som/51/03-ITRA-TG	100.8±17.36	50-100
96/0603-ITRA-TG	95.4±3.12	100
95/0166-ITRA-TG	81±5.40	100
Gbaze Kouté (Local)	27±14.28	20–30

*: Each value is the average of 3 assays followed by standard deviation

kouté (Local) were purchased from the Togo National Collection. All cultivars were from 8 to 10 months.

Determination of toxicity index of cultivars: The toxicity index was referred to as the concentration of cyanogenic glycosides expressed in milligram of cyanide (CN) per kg of sample. This was determined by two methods: The method described by Yamashita *et al.* (2010) and the method using the kit of Bradbury *et al*. (1999). The results were expressed as mean of three assays followed by standard deviation. Data were analyzed using ANOVA test and the statistical significance was set at $p<0.05$. Harvested cassava tubers were washed, peeled, cut into small pieces and crushed. Toxicity index was determined first on fresh crushed tubers for all cultivars to select the most toxic cultivar. Further analyses were performed on this selected cultivar. Indeed, the toxicity was assayed in function of harvest season. Thus, cultivar samples were harvested in dry season (December - January) and in rainy season (April - May). Circadian variation of cyanide content in the cultivar was also assayed. For this, two sets of harvest were made in April during three days, in the morning (7am - 8am) and in the afternoon (15pm - 16pm). The effect of mill-drying was evaluated by comparing the CN content of fresh tubers with CN content of milled dried tuber. Each assay was conducted trice and the averages were calculated.

Determination of toxicity index of dough: Harvested cassava tubers cut into small pieces as mentioned above were dried in sun and milled. This is a traditional method called, "Atchorolé", which is widely used by the local population during the lean season or famine. It allows in less than 8 hours of sun-drying to have flour or cassava paste. The dough samples were prepared by soaking 20 g of cassava flour alone or a mixture of cassava flour and corn flour purchased from marketplace in 150 ml of boiling water. The incidence of boiling was then determined by measuring CN content in the experimental dough prepared with cassava flour alone. Corn flour was randomly purchased in Lomé marketplace. Cyanide content was first determined on dough made of this four alone. Togolese people prefer the dough prepared from a mixture of corn flour and cassava. In order to monitor the incidence of the corn four on the reduction of CN content, serial mixtures were made by varying the amount of one or other flour to check the incidence of the corn flour on the final CN content of the dough. CN content was determined on the experimental dough made from these mixtures.

RESULTS

Toxicity index of cassava cultivars: The concentration of cyanide (CN) of ten cultivars namely 96/1632-ITRA-TG, 96/0409-ITRA-TG, Ganavé-ITRA-TG, Sorade (Local), 312/524-ITRA-TG, 96/1642-ITRA-TG, Som/51/03-ITRA-TG, 96/0603-ITRA-TG, 95/0166-ITRA-TG and Gbaze kouté (Local), was determined using the two methods (Table 1). Silver nitrate method yielded CN contents ranging from 375.3±3.82 to 27±14.28 mg/kg. The cultivar 96/1632-ITRA-TG appeared to be very toxic, with a CN content of 375.3±3.82 mg/kg. Globally, the highest amounts of CN (CN>100 mg/kg) were recorded with cultivars 96/1632-ITRA-TG, 96/0409-ITRA-TG, Ganavé-ITRA-TG, Sorade (Local), 312/524-ITRA-TG, 96/1642-ITRA-TG and Som/51/03-ITRA-TG, while the cultivar Gbazé kouté (local) displayed the lowest cyanide amount (27±14.28 mg/kg). Variance analysis revealed a significant difference between cultivars ($p<0.01$).

The second method did not allow determining a fix value of CN in the sample. Results are expressed in term of range. As found in the first method the highest amount was obtained with the same cultivar (96/1632-ITRA-TG). Similarly the lowest amount of CN was found in Gbazé kouté (local). There was a good correlation between the maximal values of CN content of cultivars ($r = 0.9416$), showing that the two methods are unambiguous ways to give good predication of CN content in the analyzed samples. The percentage reduction was determined using the CN content of cassava dough as maximal CN content i.e., 0% reduction.

Table 2: Cyanide contents in function of several parameters

	Morning	Afternoon
Time of harvest		
CN Content (mg/kg)	67.03±1.75	50.53±4.32
Season of harvest	Dry season	Rainy season
CN Content (mg/kg)	375.3±3.32	93.05±2.35
State of the sample	Fresh tubers	Dry tubers
CN Content (mg/kg)	375.3±3.32	133.2±2.27

Values are the average of three experiments followed by standard deviation

As the silver nitrate yields fix values, this method was used for further analysis. These analysis were performed with the cultivar 96/1632-ITRA-TG that was found to have high CN content. Table 2 displays the cyanide content in function of different parameters including the state dry or fresh, the harvest season and time of harvest of the samples. According to our results, tubers harvested in the morning contained more CN than those harvested in the afternoon. The elimination rate of CN from morning to evening was 24.62% ($p<0.02$). These data indicate that the timing of harvest is important in reducing the rate in the CN cassava. The amount of CN in samples harvested in the dry season was higher than that harvested in the rainy season. The percentage loss of cyanide from the dry season to rainy season was 75.21% ($p<0.04$). Fresh tubers were found to contain more CN than dried tubers, thus drying cassava resulted in a decreased rate of 64.5% ($p<0.02$).

Toxicity index of dough: Cyanides determined in the flour and cassava dough yielded 133.2±2.27 and 71.4±3.8 mg/kg, respectively. Indeed, boiling cassava flour resulted in 53.6% decrease of CN amount ($p<0.027$). Dough prepared with corn flour alone displayed CN content of 1.71±0.31 mg/kg. Cyanides determined in function of corn flour amount are presented in Fig. 1. The results indicated that the reduction in CN content was a function of corn flour amount in the dough. Good correlation was obtained between these two variables ($r = 0.8854$).

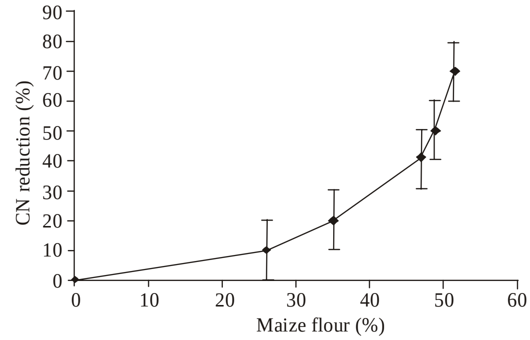

Fig. 1: Cyanide reduction in the dough in relation with the amount of corn flour

DISCUSSION

Cyanogenic glucoside content of cassava cultivar is decisive in its nutritional quality (Nyirenda *et al*., 2011). Two methods were used to determine the accurate concentrations of cyanide samples of 10 cultivars of cassava consumed in Togo. The assays yielded concentrations ranging from 27 to 375 mg/kg of fresh tuber. Except Gbazé Kouté (local), all cultivars could be considered as rich in CN, with levels above 50 mg/kg (Nambisan, 2011). The choice of cultivar may therefore be crucial for the quality of cassava root based foods. To eliminate the CN some treatments are proposed in the literature (Ojo and Deane, 2002; Lü *et al*., 2009). However, for cyanide rich varieties, other criteria should be explored (Hamel, 2011; Mlingi *et al*., 2011; Nambisan, 2011). According to the literature, in plants, biological phenomena such as the synthesis of certain substances often occur according to circadian rhythms and/or seasonal. Thus, the time of harvest was considered to examine its influence on the CN content of fresh cassava. Our results indicated that the concentration of CN varies according to season and harvest time for the same cultivar. Harvest conducted during the rainy season and in the afternoon would significantly reduce the rate of CN in the cassava products. The seasonal variation of CN content may be explained by the fact that, in the rainy season, the cyanogenic glycosides soluble in water, are partially released into the soil (Silvestre and Arraudeau, 1983). This property is also used in the technique of soaking for the elimination of cassava CN (Bradbury and Denton, 2010; Mlingi *et al*., 2011).

For food flour, cassava must be harvested at the right moment, peeled, crushed and dried. However, it is shown that in case of damage of organ, CN is released from the cyanogenic glucoside through the action of linamarase. The HCN is evaporated as a result of drying (Sornyotha *et al*., 2010). Thus, it was interesting to see the degree of decrease in the rate of CN in the processing of fresh cassava into flour. The results obtained with the technique "*atchorolé*" are in agreement with the literature. There was a significant decrease in the concentration of CN with a loss of 64.5% after 8 h of drying. However, the rest of CN in the flour (133.2 mg/kg) is above the required standard of 10 mg/kg of dry flour (Onabolu *et al*., 2001). Thus, the technique for obtaining "*atchorolé*" flour should be improved. Indeed for a good removal of cyanide, it takes more than 8 hours of drying (FAO, 1991).

The effect of cooking on the cyanide content was also evaluated. The boiling resulted in a decrease of CN of about 53.6%. According to Bradbury and Denton (2010), warm water favors the elimination of HCN vapor. The decrease in CN can be explained by the formation of HCN under the action of linamarase on the cyanogenic glycoside especially at the beginning of the cooking when the temperature is still low to inactivate enzymes. Some authors reported that chronic ingestion of cassava, especially if it is not associated with other foods rich in sulfur amino acids, exposes the consumer to cyanide poisoning (Dufour, 2011). In fact, there are traditional and modern methods to best reduce the rate of cyanide (Nhassico *et al*., 2008; Nambisan, 2011; Hamel, 2011). But in times of scarcity or famine, farmers, because of the application time from 24 h to several days, ignore these methods (Nyirenda *et al*., 2011; Cliff *et al*., 2011). For simple and faster solutions, consumers in Togo combine corn flour, and flour obtained from "Achorolé" technique for the preparation of dough. This is benefic since corn is a cereal rich in sulfur amino acid (Ojo and Deane, 2002). Our results indicate that the rate of CN decreases with the increase in corn flour. This is expected because it is a dilution; but this decline in the rate of CN becomes insignificant when the amount of corn flour exceeds 40% of the mass of the mixture of flour used for preparing the dough. Thus, the decrease in the rate of CN may also be due to the effect of transulfurases on some chemical elements in corn including sulfur amino acids. In fact some thiocyanates from CN are formed via this reaction, thus reducing the rate of CN in the dough (Oluwole *et al*., 2002). However, at high levels, corn meal can mask and prevent formation and release HCN. This could explain the insignificant reduction of cyanide.

CONCLUSION

The present study revealed that the majority of cassava cultivars consumed in Togo could be toxic because their CN content. The residual CN content in food varies in function of the season and the time of harvest, and cook processing. The issue of the study demonstrated that, by acting on the various studied factors, the amount of cyanide in cassava-based food can be sufficiently reduced.

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