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Detoxification Effect of Fermentation on Cyanide Content of Cassava Tuber

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ABSTRACT: Cassava is a staple food for approximately 800 million people in tropical countries. The tuber which comprises mainly starch also contains high concentration cyanogenic glycosides which give rise to the hydrocyanic acid by enzymatic hydrolysis. The purpose this research therefore is to investigate the detoxification potential of fermentation, on the toxic (cyanide) content of cassava tuber. The fermentation process is achieved by soaking freshly peeled cassava tuber and the fermentation of grated cassava tuber hence, two fermentation treatments. This fermentation treatments were carried out at intervals of 2 days by using 20g of the sample portions of the fresh cassava tuber (UM 8082), which initially contained as high as 160.46mg/HCN/kg of the tuber. The pH values of the medium were measured daily to correlate it with on-going events. The Knowles and Watkin's method of steam distillation technique was used to analyze the HCN (Hydrogen cyanide). Based on the experimental results obtained, there was a remarkable drop or reduction of this toxic principle in the tuber. Prolonged period of fermentation (5 to 6 days) and favourable pH medium 4.0 to 4.5) was found to effect tremendously HCN removal. In fact, 94.7% reduction that is 8.45 Mg/kg and 81.3% reduction that is 28.70mg/kg was achieved for the soaked and grated cassava tubers respectively. These values being below the recommended value 30mg/kg for safe cyanide level in food-stuff indicates that fermentation is an effective (and economic) method for detoxification of cassava tuber for human consumption. ©JASEM

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Cassava species are generally classified as sweet or bitter according to low or high cyanide concentration in their root parenchyma tissue. Bitter species contain high concentrations of glycosides distributed throughout the tissue, which forms the poisonous hydrocyanic acid on hydrolysis. This is also present in sweet cassava but in much smaller quantities Bolhius (1954). A broad range of cyanide concentration has been found among several cassava species and most of them fall within the intermediate range between the sweet and bitter species Bolhius (1954).

Onwueme, (1978), showed the concentration of hydrocyanic acid in cassava tubers ranges from 10 to 490mg/kg although this depends on variety. Bitter variety has cyanogenic acid levels of about 300mg/kg in fresh tuber, while the sweet variety has cyanogenic acid level of about 400mg/kg in fresh tuber.

The tuber is processed into garri, tapioca and cassava flour for human consumption, while the leaves are cooked and eaten especially in Sierra Leone. Sweet cassava tubers can also be eaten boiled. They can be fed raw or billed to pigs, goats, horses and cattle. The main industrial use of cassava is in the production of starch and alcohols Komolafe (1981). Cassava could be a raw material for the production of fumigants, since HCN it produces on hydrolysis is an active fumigant Prince (1985).

MATERIALS AND METHOD

Steam distillation method was used to separate the hydrocyanic acid from the prepared sample. The steam distillation was used because the HCN is a compound of relatively low volatility with boiling point of 25.7^0C, hence it can be obtained by co-distilling it `with water.

The distillation was carried out by generating steam from a boiling flask and then forcing it into another flask containing the sample to be distilled. The steam is used to heat up the sample, the distillate is condensed and collected. This method for extraction of cyanide content of a sample was based on Knowle and Watkins method (1960).

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RESULTS AND DISCUSSION

The initial total hydrogen cyanide content of the freshly peeled cassava tuber i.e. immediately after its harvest is shown in Table i. This shows that the sample is indeed a high cyanide cassava variety. To obtain the various concentrations in mg/kg at the specified periods of 2 days, 2 titrations were done in order to obtain an average value hence increasing accuracy.

Table i shows a substantial reduction of the hydrogen cyanide content with days of fermentation. It also shows that after two days of fermentation of the grated cassava tuber, there was a 41.7% reduction of HCN level of the cassava sample. Further reduction was recorded on the fourth and sixth day respectively with a 55% and 81.3% reduction. 36% reduction after two days of soaking the freshly peeled cassava tuber was observed with 88.4% and 94.7% reduction after four and six days respectively. The various pH of the medium with days was shown in Table ii. Generally, there was a decrease in pH value with days indicating that the medium becomes more acidic with days of fermentation.

Cassava contains the cyanogenic glucosides linamarine and lotaustralin. These cyanogenic glucosides are hydrolyzed to the corresponding cyanohydrin and glucose by the endogenous enzyme linamarase when cellular damage occurs de Bruijn, (1973): Nartey, (1978). Cyanohydrin breakdown non enzymatically at a rate dependent upon pH, temperature and length of time, (Cooke (1978): Oke (1983), with their stability increasing at acidic pH values. A second enzyme, hydroxynitrile lyase, may also contribute to cyanohydrin breakdown into HCN and Acetone Conn, (1969).

It should be remembered that HCN is a gas, (boiling point 25.7°c) that evaporates in the air at ambient temperature once produced. Hence, the Knowle and Watkin's method of determining the cyaninde content of the sample only accelerates the liberation HCN into the air. The HCN determined in the experiment after the heat treatment represented the residual HCN content of the cassava tube. In the fermentation of grated cassava roots, after 2 days of fermentation 41.7% total cyanide content has been hydrolyzed. This is against 36.8% reduction for the grated root and could be explained by considering that cellular damage due to grating brought both cyanogenic glycosides and endogenous enzyme linamarase into contact, hence the greater hydrolysis. In the soaked tuber, the root has not softened which implies that microbial induces root retting. At this stage the pH of the medium is around 5.0 which show that organic acids produced by corynobacterium in manihot are at the very initial stage.

On the third day, the pH had decreased further to about 4.5 showing increasing conversion of cassava starch to organic acids. On the fourth day, the pH of the medium further to drops to 4.1 and this condition favours hydrolysis of cyanoglycosides to free HCN. This is why in the fermentation of a tuber, the total (initial) cyanide content dropped sharply to 88 4% after 4 days implying that 11.6% residual content is present. For the grated root, 55.0% drop in total cyanide content was noted on the 4th day.

On the sixth day of HCN determination in both samples, 94.7% reduction of the initial total cyanide level in the soaked tuber was recorded while 81% reduction for the grated root. The least pH value was also noted accordingly, i.e. 4.0. The soaked root has been completely retted (softened) by the microbial growth. This induces leaching of the cyanogens from the roots into the water. The lower percentage reduction of the total cyanide for grated roots may be explained by noting that microbial organisms are not directly involved in linamarin hydrolysis. This has been confirmed by the work of Giraud (1993): where a lactobacillus plantarum isolated with high linuamarin activity was inoculated into a grated root fermented and there was no clear improvement in linamarin reduction. This implies that grating is the important process in bring about linamarin hydrolysis. The pH on the day of harvest is 6.5 and increased only very slightly' to 6.6 on the following day. On the second day it has fallen sharply to 5.0 implying that fermentation has just stated. A steady fall to 4.5 and 4.1 recorded on the third and fourth day respectively, showing that liberation of HCN and CO_2 is very high. The lowest value was recorded on the fifth day.

In both cases, fermentation reduces the bound and free cyanide, however, much reduction is accomplished with time. The works of Bourdoux et al (1982), Cooke and Maduagwu (1978) and Akinrele *et al*., (1962) further vindicates the accuracy of this work.

Bourdoux *et al*, (1982) found that soaking cassava roots for one day deer the cyanide content from 108.2 to 59.5ppm (part per million) implying reduction and finally to 2.9 (ppm) in 5 days implying about 97% and 94% detoxification. Correlating the above, with

this work, it implies that results approximately concur, 97% and 94% respectively. Cooke and Maduagwu, (1978), on the other hand noted a negligible decrease in bound cyanide after 4 hours.

The work of Akinrele *et al.*, (1962), shows that the HCN content of a freshly peeled grated root decreased from 104ppm to 52ppm showing 50% reduction in 5 days. This value is also quite close to the calculation or residual HCN content after 4 days; a value of 55% reduction.

The higher value of the initial total cyanide content for the freshly peeled root over that of the grated peeled root might be due to the fact that grating may have caused some hydrolysis of cyanide to free HCN which is liberated as gas into the air. From table i, it could be seen that after 6 days of fermentation, the HCN content of both soaked and grated cassava were 8.45mg HCN/kg and 28.71HCNmg/kg respectively. This shows that this unit process (Fermentation) is quite harmless to man, especially that of the soaked process.

However, Akinrele *et al.*, (1962) noted that the acceptable safe level of residual cyanide should not exceed 30ppm. i.e, 30mg HCN/kg. This shows that further treatments must be carried out for the grated root, if the toxicity of cyanide must be over looked. These include heating (roasting) at temperature higher than the decomposition temperature reported for linamarase, 72^0C (Joachin and Pandiltesekere, 1944) and linamarin, 150^0C Ceriglelli, (1955). This means that at such high temperature, though the linamarase, is destroyed, the linamarin is decomposed by heat and so this is an effective method of detoxification.

Conclusion And Recommendations: In the final analysis, the results so obtained showed that fermentation is an effective way of detoxifying cassava tuber. This is enhanced with longer period (time) of fermentation about 5/6 days. However, to safeguard against cyanide poisoning the residual cyanide level in any foodstuff should not exceed 30mg/HCN/kg, signifying that further detoxification might be done on the grated root to eliminate any fear of cyanide poisoning.

The above information shows that prolonged fermentation of freshly soaked and grated cassava tuber is an excellent detoxification process/pathway for cyanides in fresh cassava tuber. This inexpensive process makes fermentation the most economic option available.

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Table 1 Results of grated and soaked cassava tuber

For grated cassava tuber				For soaked cassava tuber		
Days	HCN mg/kg	% Retention	% Reduction	HCN mg/kg	% Retention	% Reduction
0	53.71	100	0	160.46	100	0
2	89.52	58.3	41.7	101.35	63.2	36.8
4	69.25	45.0	55.0	18.58	11.6	88.4
6	28.71	18.7	81.30	8.45	5.3	94.7

. **Table 2;** pH value and mean temperature of medium with days

Days	pH value	Mean temperature
0	6.5	25˚C
1	6.6	25˚C
2	5.0	25˚C
3	4.5	25˚C
4	4.1	25˚C
5	4.0	25˚C
6	4.0	25˚C

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