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Empirical Analysis of Hydrogen Cyanide in Streams used for Commercial Fermentation of Cassava

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Abstract: Ilaro, like many other regions in the western parts in Nigeria is famous for cassava processing. One of the major ways cassava is consumed in ilaro is through fermentation of the cassava into Fufu a staple food enjoyed by the indigenes of Ilaro. Ilaro is famous for its fufu production and it can be gotten from any area in the community. One of the most vital steps in fufu making is fermentation. The fermentation helps to detoxify the cassava of its cyanide content as well as break down the cassava to enable further processing. Cassava fermentation for fufu takes 4 to 7 days before further steps is taken. The water used for fermentation is gotten from the streams and discarded back in the streams continuously. The process of commercialized fermentation demandsa constant source of water in large quantities which is the main reason the streams in Ilaro are used for fermentation. Cyanide contamination of selected streams in Ilaro was studied as part of a bigger project aimed at setting background standard for contaminations of water, land and air in the Yewa South environment. A total of 15 samples were collected from the streams used for the cassava fermentation with five streams in the scope where 3 samples were collected from each streams. (points before activity, points after activity and points of activity) to better as certain the concentration of cyanide in the streams. From our research we were able to ascertain that cyanide concentration is below detectable limits in the points before activity and trace amounts below permissible guidelines by regulatory bodies such as WHO and US EPA were found in the points of activity and after activity. This study found that the streams are safe for domesticated use as well as the commercial fermentation of cassava but they should be monitored closely to avoid future contaminations as cyanide is a great risk to human health and in large quantities very fatal.
Keywords: Health Concern: Cyanide: Fermentation: Cassava: Streams: Nigeria.
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1.0 Background of the Study

Pollution is the introduction of contaminants into an environment, soil, food and water that causes instability, disorder, harm or discomfort to the ecosystem or living organisms (Gari, 2002). Sources of pollution can be natural or man-made. The major sources of pollution in urban areas are anthropogenic (human activities) while contamination from natural sources predominate in the rural areas (Hutchsinson & Meema, 1987). Cassava an edible tuber crop consumed in different forms by people in different parts of the world contains a very poisonous component cyanogenic glycosides, which can result in fatal cyanide poisoning if not properly detoxified by appropriate means (soaking, drying, boiling, frying etc.) (Alitubeera *et al., 2019*). Cyanogenic glycosides are a group of chemical compounds which occur naturally in over 2 000 plant species with at least 25 cyanogenic glycosides known to be found in the edible parts of plants. Cyanogenic glycosides alone are relatively non-toxic. However, as a result of enzymatic hydrolysis by beta-glucosidase following maceration of plant tissues as they are eaten, or by the gut microflora, cyanogenic glycosides are broken down to release hydrogen cyanide which is toxic to both animals and humans.

The potential toxicity of a cyanogenic plant depends primarily on its capacity to produce hydrogen cyanide (Food safety Focus, 2018). Cyanogenic glycosides are present in most plants because they act as a defense mechanism, which is evident as the yoccur at a significant rate only after plant tissues have been disrupted by herbivores, fungal attack, or mechanical means (Njoku *et al., 2018*). Cassava (*Manihotesculenta*), is an edible tuberous root that is resistant to drought, diseases, and pests, it is a major source of carbohydrates in tropical areas, the second most widely grown and consumed food in Nigeria in many different processed form. Cassava comes in two types (Sweet and Bitter Cassava). Sweet cassava roots contain less than 50 mg per kilogram of hydrogen cyanide on fresh weight basis, whereas that of the bitter variety may contain up to 400 mg per kilogram as a reason, sweet cassava roots can generally be made safe to eat by peeling and thorough cooking. However, bitter cassava roots require more extensive and complicated processing for detoxification (Ukonu et al., 2022; Ubreye Benjamin et al., 2022).

One of the traditional ways to prepare bitter cassava roots is by first peeling and grating the roots, and then prolonged soaking of the gratings in water to allow leaching and fermentation to take place, followed by thorough cooking to release the volatile hydrogen cyanide gas. Cutting the roots into small pieces, followed by soaking and boiling in water is particularly effective in reducing the cyanide content in cassava. Whilst fresh cassava requires traditional methods to reduce its toxicity, adequately processed cassava flour and cassava-based products have very low cyanide contents and are considered safe to use (Food safety Focus, 2018). Cyanide, a toxic contaminant, occurs naturally in most plants but has high concentration in cassava and bamboo shoot. It is released into the environment through volcanoes and natural biogenic processes from higher plants, bacteria, algae and fungi, biomass burning, discharges from, smoke industries, waste water treatment, tobacco smoke, wood smoke, from burning plastics, vehicular emission, inadequately processed cassava products etc. (Agency for Toxic Substances and Disease Registry 1997; Lasisi, 2018).

Cyanide exist in water as hydrogen cyanide (HCN) at PH less than 8 which is more harmful to aquatic life than the free cyanide ion. The cyanide toxic effect is due to its reaction with the trivalent ion in the cytochrome oxidase to inhibit electron transport and hence prevent the cells from consuming oxygen which leads to rapid impairment of the vital functions (Cacace *et al., 2007;* Ukonu et al., 2022; Ubreye Benjamin et al., 2022). Hyper baric oxygen (HPO), hydroxocobalimine,dicobalt-EDTA, IV dimethylaminophenol have been reported to be cyanide antidote which are effective in experimental and human cyanide poisoning (Lam & Lau, 2000). Exposure to small amounts of cyanide can be deadly regardless of the route of exposure. Cyanide is very poisonous; it stops cellular respiration by inhibiting an enzyme in mitochondria called cytochrome c. oxidase in the body. Water is a transparent and nearly colorless chemical substance that is the main constituent of Earth's streams, lakes, and oceans, and the fluids of most living organisms (Shumi et al., 2021; Chala et al., 2022; Enyi et al., 2022). The importance of water for human survival as well as many other sectors of the economy cannot be overemphasized. So, the contamination of sources of water should be of high priority due to the health risk associated with its contamination even as some anions are found in water are useful to human body, many others are harmful (Tayfur *et al.*, 2011).

Water is an essential part of daily human activities as it is used in almost all activities carried out by humans. Water is an essential source for detoxification of cassava to render it safe for consumption but with the detoxification of cassava, the water used becomes polluted and there is need for careful disposal to avoid contamination of major source of water which can lead to health risk for people in the area. Roots of cassava which are mainly starch are extremely rich in carbohydrates. According to Food and Agriculture Organization, cassava is the third most important source of calories in the tropics, after rice and corn (Shumi et al., 2021; Chala et al., 2022; Enyi et al., 2022). Cassava is consumed in various types of ways, which includes being eaten as whole root, grated root or root chips. In addition, it is prepared into flour which in turn can be used for cooking or production of cassava-based products such as breads, crackers, and puddings or beverages made with tapioca pearls, they can also be fermented and then cooked for consumption.

Cassava leaves are also eaten in some countries following extensive boiling. Apart from being used as human food, cassava products are also used as animal feed. (Food safety Focus, 2018). Fermented Cassava, processed product from cassava, popularly known as" Fufu" in Yoruba language is a staple food in Nigeria which contains essential and beneficial minerals needed for the body morphological processes. It is a rich source of carbohydrate, often referred to as the major fuel of the body tissues that releases energy needed by the body to function properly in its daily activities. Detailed studies of the fate and contents of cyanide in cassava, cassava bi-product and human beings as well has become a major task in research and there is continuous challenge to develop new methodology and optimize the already existing methodologies (Shumi et al., 2021; Chala et al., 2022; Enyi et al., 2022). Health education about cyanide poisoning from cassava and the need to adequately process cassava to reduce cyanogenic content must be conducted regularly by public health officials (Alitubeera *et al., 2019*).

Cyanide is a chemical compound that contains the cyano group -C≡N, which consists of a carbon atom triple bonded to a nitrogen atom. Cyanide could be a gas; hydrogen cyanide or a solid; potassium cyanide or sodium cyanide and they are fast-acting poisons that can be lethal (Ukonu et al., 2022; Owolabi et al., 2022). It's one of the fastest acting poisons known, and if a significant amount is ingested, it can lead rapidly to death of the person that ingested it. Upon ingestion, it binds to haemoglobin, which is a molecule in red blood cells responsible for carrying oxygen to the cells in our body. Haemoglobin then carries cyanide to the body's tissues, where it can bind to an enzyme called cytochrome oxidase. This enzyme is a vital tool cells require to make use of oxygen, and with cyanide bound to it, they are unable to do so (Chemistry of poisons, 2015).

In food processing, it is often necessary to carry out trace element analysis to ensure that harmful and non-essential elements are kept at low concentrations as much as possible (Eshiett et al., 2022; Osho & Haruna, 2022). Cyanide is toxic to human beings as it interferes with enzyme functions while some may have stimulatory effects. It must be noted that, the cyanide when consumed is highly toxic to human beings and aquatic life. Free cyanide forms react within few hours to days with almost any other chemical they are exposed to producing a wide variety of new compounds whose toxicity may be greater or less than the free cyanide itself (US Environmental Protection Agency, 1985). The increase of cyanide in water used for cassava fermentation can cause toxic effects for consumers and environment. The gravity of toxic effect depends on the nature, quantity, chemical form on body resistance and on synergetic or antagonistic effects of other chemical contaminants. Sometime in 2012, it was widely reported about a family who was nearly wiped out after eating food Prepared from fermented cassava flour in Ondo state (dailypost.ng 2012). Such food poisoning could result from pollution by known and unknown toxicants.

This article focuses on the determination of the levels of free cyanide and total cyanide concentrations in some selected streams used for the commercial fermentation of Cassava (Fufu) in Ilaro Yew South Ogun State Nigeria by comparing the results obtained in this study with Nigerian Standard for Drinking Water Quality (NSDWQ), USEPA (US Environmental Protection Agency) and the World Health Organization (WHO) permissible levels of free cyanide and total cyanide in surface waters (Ukonu et al., 2022; Owolabi et al., 2022). These Streams are the major sources of water and are used for different purposes by humans. People who are involved in cassava fermentation for fufu production get their source of water from the streams and the by-product Cyanide is often times disposed back into these water bodies (Eshiett et al., 2022; Osho & Haruna, 2022). This informed our investigation to determine concentration of cyanide in the streams and as well to create awareness where needed so that activities leading to these possible contaminations be stopped as much work has been done on the streams used for the commercial fermentation of cassava in Ilaro, it lightened our research interests on the streams.

Picture 1: Stream used for Cassava Fermentation

Picture 2: Cassava Fermentation Waste kept beside the stream

Picture 3: Stream used for Cassava Fermentation

Picture 4: Woman Sifting out Fermentation waste for disposal

2.0 Materials of the Study

***2.1 Study Area*:** Ilaro, is a town in Ogun State Nigeria. Ilaro town houses about 30,000 people that are hospitable, mostly with high sense of humor, willing and ready to welcome investor, mechanized farmers and tourists. Ilaro is the headquarters of Yewa south local government, now known as Yewa land which replaced Egbado division of the former Western state and later became a part of Ogun state of Nigeria. Ilaro town is about 50 km from Abeokuta, the Ogun state capital, and about 100 km from Ikeja, the capital city of Lagos state. Other neighboring towns include Ajilete, Oke-Odan Owode, Papa-Alanto and Imasayi. The dialet spoken in Ilaro as the headquarters of Yewa/Egbado people is the Egbadodialet (http://ilarotown.com/our_history.html, 2017).

2.2 Sample Collection: In Nigeria there are two main peak seasons for cassava processing in a year, the first which starts in April and ends in June while the second commences in August and lasts till November. A total of 10 samples of water used for cassava fermentation was collected from five different streams as this were the major streams used for the fermentation process (Odo Double Crown, Odo Oke Ela, Odo Garage, OdoIdonji Odo Powerline) with three samples from each streams i.e. site from before activity, site of activity and site after activity) in Ilaroyewa local government Ogun state Nigeria. These areas were chosen for the case study because these streams are used primarily for commercial fermentation process of cassava. The samples were collected in a container covered with aluminium foil so as to filter light at 490 nm and below and prevent photodecomposition of cyanide complexes. To avoid further contamination during sampling, all containers were thoroughly cleaned and rinsed prior to use. The sample containers were prewashed with acids, water, and metal-free detergents. Samples were brought to room temperature prior to analysis.

2.3 Reagents: Rhodanine indicator – it was freshly prepared by dissolving 20 mg of p-dimethylamino-benzal-rhodanin, in 100 mL of acetone. Standard silver nitrate solution (0.0192N), AgNO3 – was prepared by crushing approximately 5 g of AgNO3 and drying to constant weight at 40 °C. An amount of 3.2647 g of dried AgNO3 was dissolved in 1 litre of water.

3.0 Research Methodology

There are several methods for the analysis of cyanide species directly in a matrix or following a pretreatment. The methods include titrimetric, spectrophotometric, potentiometric, ion-selective electrode (ISE), ion chromatography – high performance liquid chromatography (IC-HPLC), indirect AAS, gas chromatographic and infrared spectrometric. Of all these methods, Spectrophotometric method was used in this analysis due to the fact that equipment required for the alternate methods were not available (Onwuka, 2005).

3.1 Preparation of stock solution of cyanide: 0.40mg/L stock solution of cyanide was prepared by dissolving 1g of KCN with distilled water in 1000ml volumetric flask and made up to the mark with distilled water.

3.2 Calibration curve for cyanide determination: 0.40mg/L stock solution of cyanide was diluted to prepare 0.02mg/L, 0.04mg/L, 0.06mg/L, 0.08mg/L and 0.10mg/L standard solutions of cyanide. A blank solution was also prepared (Onwuka, 2005). The absorbance of each concentration was measured at 490nm using a Novaspec model 4049UV/VIS spectrophotometer. The calibration curve for the cyanide determination was obtained by plotting absorbance against concentrations of the standard cyanide solutions. Then the graph factor obtained from the plot was used to calculate the cyanide content of the samples.

3.3 Preparation of Alkaline picrate solution: Alkaline picrate solution was prepared by dissolving 1g of picrate and 2g of sodium carbonate in a volume of minimally warm water in 100ml volumetric flask and made up to the mark with distilled water (Esser *et al., 1996*).

3.4 Determination of cyanide using UV/Visible spectrophotometer: 5ml of the sample filtrate were put in a corked test tube and 4 ml of the alkaline picrate were added, and the solution was incubated in a water bath for 5 minutes. After colour development (reddish brown colour), the absorbance of the corked test tube was read on a Novaspec model 4049uv/visible spectrophotometer at 490nm which is the wavelength of maximum absorption (λmax) of cyanide and this procedure was repeated for each sample. The absorbance of a blank solution containing 1ml distilled water and 4 ml alkaline picrate solution was also read and extended it on the calibration graph (Esser *et al., 1996*).

3.5 Data Collection: The data for this research work was obtained through primary and secondary source. Primary source was gotten from the results of analysis done on the water sample while secondary sources was gotten from internet, conferences and journals proceedings, magazines, books and other archival sources of informations.

3.6 Data Analysis: Microsoft excel version 2011 was used to calculate the Mean Standard Deviation of the concentrations of cyanide in the streams. The descriptive statistics are presented in Table 1 and Table 2 showing the standards of WHO (World Health Organization), Nigerian Standard for Drinking Water Quality (NSDWQ)and EPA (United States Environmental Protection Agency).

4.0 Results from Data Analysis

Sampling point	Site Before activity	Site of activity	Site after activity
Odo Double Crown	BDL	0.02 ± 0.01	0.01 ± 0.01
OdoOke Ela	BDL	0.03 ± 0.01	0.02 ± 0.01
Odo Garage	BDL	0.04 ± 0.01	0.02 ± 0.01
OdoIdonji	BDL	0.05 ± 0.01	0.01 ± 0.01
OdoPowerline	BDL	0.03 ± 0.01	0.01 ± 0.01

Table 1: Result from Analysis

Keys: BDL=Below Detectable Limit

Agencies	Free cyanide (mg/L)
NSDWQ	0.1
USEPA	0.2
WHO	0.2

Table 2: The WHO, NSDWQ and EPA Standard values for Cyanide
Source: Guidelines for Drinking Water (2017) & NSDWQ for Drinking Water (2015)

5.0 Discussions and Conclusions

From the table 1 as shown above, when compared with the Standards in table 2 shows that cyanide concentration levels are below detectable and/or permissible limits. From the streams, point before activity shows below detectable limits and this can be understandable since little to no activity occurs at this point as it is before the point where fermentation occur. The site of activities shows trace amount of cyanide (0.02 ± 0.01, 0.03 ± 0.01, 0.04 ± 0.01, 0.05 ± 0.01, 0.03 ± 0.01) which when compared with standards are below the permissible limits this is possible as it is a flowing streams most of the waste water from fermentation processes are discarded back in the streams which will normally flow downwards. For the site after activity trace amount of cyanide were also detected (0.01 ± 0.01, 0.02 ± 0.01, 0.02 ± 0.01, 0.01 ± 0.01, 0.01 ± 0.01) which are also below permissible limits of regulatory bodies this goes to indicates the streams are flowing back to a source therefore leading to the minute discovery of cyanide. Cyanide enters air, water and soil from both natural and anthropogenic activities. Most cyanide in surface water will form hydrogen cyanide which would eventually be evaporated. Some cyanide in water are mostly transformed into less harmful chemicals by microorganisms (Jaszczak *et al., 2017)*. In this study cyanide levels in the streams were below Regulatory body's permissible guidelines. Based on our results it seems that the fermentation process has no major contamination effect on the streams as they're flowing but nevertheless, monitoring of the streams for their cyanide concentration level should be continuous for environmental safety. As even as the concentration limits were in trace amounts within the standard limits, the fermentation process may contaminate the underground water in long term effect or might end up contaminating the end point of the streams. Therefore, regular estimation of cyanide concentration as well as other contaminants should be carried out as well as tracing of the end points of the streams to really ascertain the environmental health risk associated with the process.

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7.0 Reference of the Study

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