Elimination of cyanide from cassava roots grown in Congo, case of bitter variety NGAMANZA: kinetic and thermodynamic studies

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RESUME

Les études cinétique et thermodynamique sur l'élimination du cyanure dans les tubercules de manioc ont été menées en utilisant le processus de trempage simple. Une des variétés de manioc cultivées au Congo a été utilisée. La méthode argentométrique a été utilisée pour identifier et doser la quantité du cyanure extrait dans l'eau. Les constantes cinétiques indiquent que (a) des concentrations potentiellement toxiques de cyanure sont réduites aux limites tolérables en moins de trois jours de trempage, et (b) l'élimination est plus rapide quand l'échantillon frais est utilisé aux températures relativement hautes. Les valeurs des constantes d'équilibre et de l'énergie libre montrent que l'élimination du cyanure des tubercules de manioc est (a) un processus spontané et (b) contrôlé par le facteur entropique, en compensation du facteur enthalpique, l'élévation de la température favorisant efficacement l'élimination rapide du cyanure du manioc.

Mots-clé: Manioc, Elimination de cyanure, Etude thermodynamique et cinétique.

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ABSTRACT

Kinetic and thermodynamic studies on the elimination of cyanide from cassava roots have been undertaken using the soaking process. One variety of cassava grown in Congo has been used. The argentometric method was used to monitor and quantify the appearance of cyanide in water. Kinetic constants indicate that (a) potentially toxic concentrations of cyanide are reduced to safer limits in less than three days of soaking, and (b) the elimination is faster when fresh sample is used as raw materials at higher temperatures. The values of the equilibrium constants and free energy show that the elimination of cyanide from cassava roots is (a) a spontaneous process and (b) controlled by the entropic factor, in compensation with the enthalpic one, the elevation of the temperature promoting efficiently the fast elimination of cyanide from cassava.

Key words: Cassava, Elimination of Cyanide, Thermodynamic study and Kinetic study.

INTRODUCTION

The cassava (*Manihot Esculenta*) is the most important tuber cultivated in tropical zones. This tuber constitutes a major source of dietetic energy for more than 600 million people (1, 2, 3, 4, 5). It is considered to be the best source of carbohydrates (sugars) among the culture of basic commodities (5, 6, 7).

According to FAO, the cassava occupies the 4th place of the traditional cultures in the developing countries after rice, maize and corn. The cassava leaves are relatively rich in proteins, vitamins A and B (8,9). According to FAO and IFAD, cassava can be an important starting point for the growth in several developing countries if the diversification on the production and the commercial utility of this important tuber are improved.

The major problem is the presence of cyanide in the cassava which is a poison. Consumption of cassava containing cyanide can lead some times to a terrible nervous disease called "Konzo" and other diseases such as goitre, dwarfism, and cretinism (10, 11, 12, 13, 14). There is certainly in our opinion a relationship between the presence of cyanide in the cassava and these diseases.

Elimination of cyanide has been extensively investigated for different purposes (13, 14, 15, 16, 17, 18). Cyanide exposure is causing damage in the populations in almost all African countries using cassava as principal diet (4,11,12,13,14,19). Many surveys were undertaken to determine the cyanogenic potential of cassava (2,12,19). In Congo, the effects of cyanide exposure has been largely investigated (11,13). The increasing growth of populations combined with the lack of major diets other than cassava (cheaper diet) in the African continent poses a serious problem.

Agbor-Egbe *et al.* have highlighted the effectiveness of processing techniques is essential if cassava foods are to be produced on large scale (2)

The "Cassava processors" ("Moulin de manioc") transform the cassava roots into flour without changing the level of cyanide initially present in the cassava roots. There is a need for technology able to make flour from free cyanide cassava roots(7,8,15,16,21,22,23,24).

Developing countries are lacking adequate technology to eliminate cyanide in an industrial way, probably because physicochemical data required to dimension a reactor are missing. For instance, there is a relationship between elimination rate and reactor volume (20).

The objective of this study is to provide some missing physicochemical data such as the rate of extraction of the hydrocyanic acid from the cassava root, the values of equilibrium constants and free energy. To our knowledge, there are no kinetic or thermodynamic data useful to industries willing to design reactors able to reduce cyanide levels or speed the cyanide elimination process(8, 24, 25, 26).

The medical field will benefit from these data, because the adequate technology will help decrease the risks of cyanide exposure for the regular consumption of cassava (5,11,12). Knowing the exact number of days for soaking is an important parameter for improving the economic viability of the process and the quality of the end product (2,24). In fact, Ihedioha *et al.* have demonstrated that viscosities of flours decreased with increasing duration of holding the root in water; the quality of bread declined with increasing delay in processing root to flour (8).

EXPERIMENTAL TECHNIQUES

Cassava samples, namely Ngamanza, has been used as received. They were graciously given to us by the interpreter of the PSA (Food Security Project), financed by CARITAS Belgium, at MENKAWU round the tray of Bateke.

Tubers had the age of eleven months and have been kept fresh during all the period of manipulations.

A thermostat stove, a pH-meter, and Whatman papers n ° 3, a steam distillation installation as well as a digital balance were used.

For analyses, reagents and solutions used are: the anhydrous sodium carbonate 5%, the picric acid pure for analysis, caustic potash 0,5 N pure for analysis, the ammonia 25%, the nitrate of silver ($AgNO_3$) 0,01 M and pure concentrated hydrochloric acid for analysis.

PRINCIPLES OF THE METHOD

Qualitative method

Cyanogen quantification experiments were performed in triplicate using the argentometric method (27). The pH of the distilled water used for soaking was adjusted to $pH < 4$ by adding a few drops of HCl. After soaking the cassava, the hydrocyanic acid released by enzymatic hydrolysis drained by the steam was recovered in alkaline solution and titrated by argentometry method:

$$[Ag(CN)_2]^- + Ag^+ \rightleftharpoons Ag[Ag(CN)_2]^-$$

Because the ion $(Ag(CN)_2)^-$ formed has tendency to combine with the ion Ag^+ with each addition of $AgNO_3$ to form a precipitate according to the following equilibrium,

$$[Ag(CN)_2]^- + Ag^+ \rightleftharpoons Ag[Ag(CN)_2]$$

we added NH_4OH in excess to make the complex $Ag(Ag(CN)_2)$ soluble. The end of the reaction is reached when all the CN^- ions are binded. At this point, the amount of Ag^+ in excess reacts with I^- ions which are present (we added KI) in the reaction medium. Because AgI is insoluble in NH_4OH, the precipitate of AgI is identified as a cloud (whitish or yellowish) in the medium.

Quantitative method

Ten 250 ml-flasks were prepared. Each flask contains 50 g of the selected variety of cassava and distilled water has been used to fill up the flasks. After seven hours, 100 ml of the solution of the first flask were distilled by steam distillation to recover HCN released by hydrolysis. The second sample undergoes steam distillation after 14 hours and so on. Every seven hours, a steam distillation is performed for three days.

The distillate is recovered in another 100 ml flask containing 5 ml of 0.75 M NaOH. Distillation was stopped when about 10 ml of distillate are obtained and then 5 drops of NH_4OH and 2 drops of KI are added before titration with $AgNO_3$ 0.1M.

The titration is made on black bottom in order to observe clearly the opalescence due to the formation of AgI. Distilled water has been used as reference.

The difference between the volume (in ml) of $AgNO_3$ used for the titration of the sample and the volume (in ml) used for the reference indicates the amount of CN^- in the sample.

RESULTS AND DISCUSSION

Experiences succeeded to the following results that present values of concentrations of HCN in the fresh cassava soaked at different temperatures (Table 1).

Kinetic parameters determination

It has been established that the elimination phenomenon of cyanide behaves as a first order and the kinetic constant (**k**), which is a measure of kinetic activity of water, has been calculated from the classical relation of formal kinetic:

$$\ln\left(\frac{a}{a-x}\right) = kt \qquad (1)$$

or

$$x = a(1-e^{-kt}) \qquad (2)$$

where:

a is the initial extractable concentration in cyanide; **x** its conversion (amount extracted in water) and **t** is the time expressed in hours.

It follows that while plotting **x** versus **t** from the least-squares method applied the non linear regression, one obtains curves whose slopes give the constants **k** and the half extraction times were calculated by the traditional relation $\frac{0,693}{k}$.

In figure 1 and in table II indeed, it can be seen the evolution of cyanide concentration versus soaking time, the speed constants and the extraction halftimes corresponding to the moiety of the total extractable concentration of cyanide from cassava.

It can be observed from these results that the quasi-total elimination of cyanide from cassava takes place after a soaking time of 65 hours at 29 °C, close to 52 hours at 32 °C, 46 hours at 34 °C, 43 hours at 36 °C against only 40 hours at 38 °C. The temperature plays a major role in the elimination of cyanide and in the time management of this elimination.

It can be noted that the extraction halftime decreases with the increase of the temperature while the speed constant increases with the temperature.

It can be seen therefore that at ambient temperature, the elimination of extractable cyanide requires three days of soaking, whereas at 38 °C the extraction requires two days only for the total elimination.

Thermodynamic parameters determination

The elimination of cyanide from cassava in aqueous medium is an equilibrium between two phases (the solid phase constituted of cassava, and the liquid one, constituted of water) with the equilibrium constant of partition of cyanide between two phases. The cyanide released from the cassava solid goes through the aqueous medium by diffusion; therefore the elimination of cyanide will be limited by the phenomenon of diffusion and the enzymatic action. The enzymatic action inhibits at $pH \leq 4$, but only the diffusion phenomenon controls the reaction of elimination of cyanide.

The distribution equilibrium is governed by this relationship:

$$\frac{1}{K_{app}} = \frac{1}{K_{true}} + X_{CN_{sol-bound}} \frac{1}{C_{CN_{aq}}} \quad (3)$$

either

$$\frac{a}{X_{CN_{aq}}} = 1 + \frac{1}{K_{true}} + X_{CN_{sol-bound}} \frac{1}{C_{CN_{aq}}} \quad (4)$$

with

$$K_{app} = \frac{C_{CN_{aq}}}{C_{CN_{sol}}},\ K_{true} = \frac{X_{CN_{aq}}}{X_{CN\,sol-free}} \text{ and}$$

$$C_{CN_{aq}} = X_{CN_{aq}} \quad (5)$$

$$C_{CN_{sol}} = X_{CN_{sol\ free}} + X_{CN_{sol-bound}} \quad (6)$$

where

$C_{CN_{sol}}$ is the concentration of cyanide in the solid phase; $X_{CN\,sol-free}$ is the actual concentration of free cyanide in the solid phase; $X_{CN_{sol-bound}}$ its actual concentration bound in the solid phase; $X_{CN_{aq}}$ represents the actual concentration of cyanide in the aqueous phase; K_{app} and K_{true} are respectively the experimental and true equilibrium constants, the later being the ratio of only free entities in the two phases.

Gotten values are represented by figure 2 at different temperatures and are deduced from tables 1a to 1e according to the relation (4). Intercepts of these curves give K_{true}, and it can be noted in table 3 that K_{true} increases with the increase of the temperature and this shows that the extraction is better done at high temperature.

While carrying these constants according to their corresponding temperatures, according to the relation of Van't Hoff:

$$\ln K_{true} = -\frac{\Delta G^0}{RT} = \frac{\Delta S^0}{R} - \frac{\Delta H^0}{R} \cdot \frac{1}{T} \quad (7)$$

results of figure 3 below are deduced, which are presented in table 4.

The values of ΔG^o calculated are listed in table 4. Standard deviation has been used to estimate our measures. These values are negative for all the samples ($\Delta G^o \prec 0$), that means that the process of partition of cyanide between the solid phase and the aqueous phase is spontaneous.

The positive value of the standard enthalpy ($\Delta H^o \succ 0$) shows that the phenomenon is endothermic, and therefore favoured by the liberation of the cyanide from linamarin in cassava when there is elevation of the temperature, and the positive value of the standard entropy ($\Delta S^o \succ 0$) is probably ascribed to the neat structure around cyanide in cassava compared to another medium in which it is solvated, that means in water.

Indeed, cyanide in cassava is in a neat state; it is necessary to provide a certain quantity of energy to loose it from this milieu, and its passage in water is due to an increase of disorder caused by an increase of volume in the aqueous medium even though it gets ready to a solvatation. The general spontaneous reaction shows that entropic factor ($-T\Delta S^0$) compensates the enthalpic one (ΔH^o) for explanation of this phenomenon.

CONCLUSION

The kinetic data obtained from this variety of cassava show that use of fresh cassava as raw materials is the best option to achieve maximum efficiency.

Thermodynamic data such as equilibrium constants and free energy were calculated. The kinetic approach is done in order to determine the optimal time of total elimination of extractable free cyanide from cassava, which changes with the temperature as it has been discussed.

The thermodynamic study reveals that the yield of extractable free cyanide from cassava is function of the temperature; the process is spontaneous in the studied temperatures range and its evolution is governed by the entropic factor.

The kinetic and thermodynamic data may be useful to industries willing to manufacture reactors for rapid cyanide elimination from cassava and results obtained could be essential if cassava foods are to be produced on a larger scale and the processing techniques are to be modified without causing adverse effects to human health so as to maintain consisting and organoleptic qualities.

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EXES

1. Concentration of the cyanhydric acid in the cool cassava

e ambient temperature (29 °C)

N° of the sample	Soaking time	Volume of $AgNO_3$ in ml				Concentration of HCN extracted in mg/kg
		1st test	2nd test	3rd test	Average	
1	7 hours	0,06	0,06	0,05	0,056	11,11 ± 0,05
2	14 hours	0,09	0,10	0,09	0,093	18,60 ± 0,05
3	21 hours	0,14	0,16	0,15	0,150	30,00 ± 0,05
4	28 hours	0,17	0,18	0,17	0,174	34,60 ± 0,05
5	35 hours	0,18	0,18	0,19	0,184	36,60 ± 0,05
6	42 hours	0,19	0,19	0,19	0,191	38,00 ± 0,05
7	49 hours	0,23	0,24	0,25	0,241	48,00 ± 0,05
8	56 hours	0,26	0,28	0,29	0,277	55,20 ± 0,05
9	63 hours	0,27	0,29	0,28	0,281	56,00 ± 0,05
10	70 hours	0,27	0,28	0,29	0,281	56,00 ± 0,05

2 °C

N° of the sample	Soaking time	Volume of $AgNO_3$ in ml				Concentration of HCN extracted in mg/kg
		1st test	2nd test	3rd test	Average	
1	7 hours	0,15	0,15	0,15	0, 154	30,64 ± 0,05
2	14 hours	0,20	0,21	0,21	0,211	42,10 ± 0,05
3	21 hours	0,27	0,27	0,27	0,273	54,50 ± 0,05
4	28 hours	0,30	0,30	0,30	0,305	60,80 ± 0,05
5	35 hours	0,32	0,32	0,32	0,324	64,60 ± 0,05
6	42 hours	0,36	0,36	0,36	0,358	71,30 ± 0,05
7	49 hours	0,44	0,44	0,44	0,438	87,30 ± 0,05
8	56 hours	0,49	0,49	0,49	0,493	98,20 ± 0,05
9	63 hours	0,50	0,50	0,50	0,504	100,28 ± 0,05
10	70 hours	0,50	0,50	0,50	0,504	100,28 ± 0,05

4 °C

N° of the sample	Soaking time	Volume of $AgNO_3$ in ml				Concentration of HCN extracted in mg/kg
		1st test	2nd test	3rd test	Average	
1	7 hours	0,25	0,24	0,25	0,252	50,18 ± 0,05
2	14 hours	0,34	0,32	0,33	0,329	65,60 ± 0,05
3	21 hours	0,40	0,40	0,40	0,400	79,00 ± 0,05
4	28 hours	0,44	0,44	0,42	0,437	87,00 ± 0,05
5	35 hours	0,46	0,46	0,47	0,465	92,60 ± 0,05
6	42 hours	0,52	0,53	0,52	0,525	104,60 ± 0,05
7	49 hours	0,64	0,63	0,63	0,636	126,60 ± 0,05
8	56 hours	0,70	0,71	0,71	0,709	141,20 ± 0,05
9	63 hours	0,72	0,72	0,73	0,726	144,56 ± 0,05
10	70 hours	0,72	0,73	0,72	0,726	144,56 ± 0,05

d) at 36 °C

N° of the sample	Soaking time	Volume of AgNO$_3$ in ml				Concentration of HCN extracted in mg/kg
		1st test	2nd test	3rd test	Average	
1	7 hours	0,28	0,28	0,28	0,283	56,39 ± 0,05
2	14 hours	0,36	0,36	0,36	0,361	71,81 ± 0,05
3	21 hours	0,43	0,43	0,42	0,428	85,20 ± 0,05
4	28 hours	0,46	0,48	0,47	0,468	93,21 ± 0,05
5	35 hours	0,49	0,50	0,48	0,496	98,81 ± 0,05
6	42 hours	0,55	0,55	0,55	0,556	110,82 ± 0,05
7	49 hours	0,67	0,67	0,67	0,667	132,79 ± 0,05
8	56 hours	0,86	0,85	0,87	0,860	171,25 ± 0,05
9	63 hours	0,88	0,86	0,87	0,869	172,93 ± 0,05
10	70 hours	0,86	0,87	0,88	0,869	172,93 ± 0,05

e) at 38 °C

N° of the sample	Soaking time	Volume of AgNO$_3$ in ml				Concentration of HCN extracted in mg/kg
		1st test	2nd test	3rd test	Average	
1	7 hours	0,32	0,31	0,30	0,314	62,60 ± 0,05
2	14 hours	0,46	0,46	0,46	0,466	92,90 ± 0,05
3	21 hours	0,53	0,51	0,52	0,522	103,90 ± 0,05
4	28 hours	0,65	0,66	0,64	0,653	130,10 ± 0,05
5	35 hours	0,81	0,81	0,78	0,805	160,30 ± 0,05
6	42 hours	0,89	0,91	0,90	0,902	179,48 ± 0,05
7	49 hours	0,95	0,96	0,97	0,962	191,59 ± 0,05
8	56 hours	1,00	1,00	1,03	1,011	201,30 ± 0,05
9	63 hours	1,00	1,01	1,02	1,011	201,30 ± 0,05
10	70 hours	1,01	1,01	1,01	1,011	201,30 ± 0,05

Table 1a-b show that the concentration of cyanide increases over time in water and becomes constant after 56 hours, being enhanced by the temperature. Results obtained at 38° C show that maximum extractable quantity of cyanide in this variety of cassava is about 201 mg per kg of fresh matter.

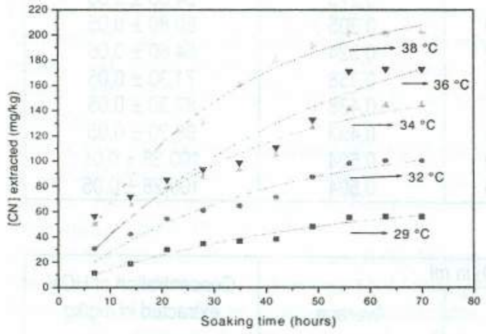
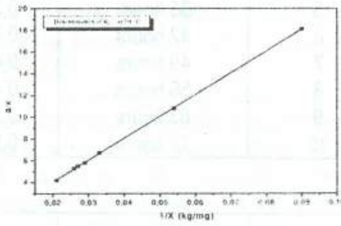


Figure 1. Evolution of cyanide concentration from Ngamanza cassava versus soaking time at different temperatures.

a. at the ambient temperature (24° C)



b. at 32° C

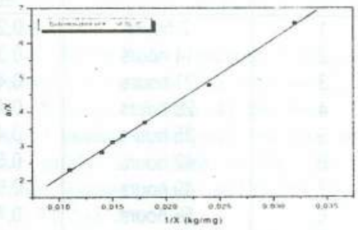
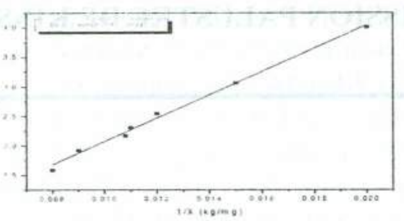


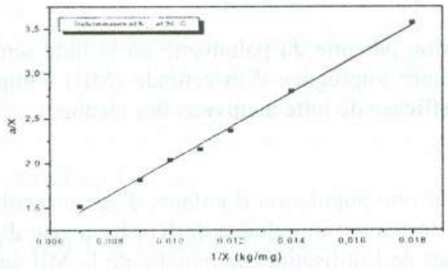
Table 2. Extraction speed constants and halftimes at different temperatures

Soaking temperature (°C)	Speed constants (h^{-1})	Extraction halftime (h)
29° C	0,0213 ± 0,0014	32,5352
32° C	0,0271 ± 0,0087	25,5719
34° C	0,0297 ± 0,0063	23,3333
36° C	0,0319 ± 0,0047	21,7241
38° C	0,0341 ± 0,0046	20,3222

c. at 34° C



d. at 36° C



e. at 38° C

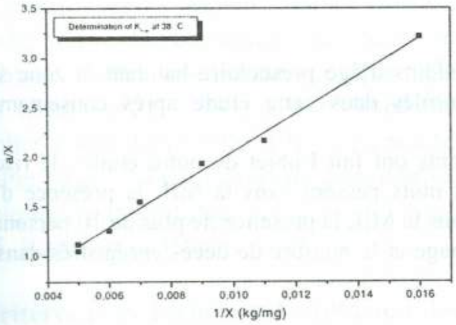


Figure 2. Variations of $\frac{a}{X_{CN_{aq}}}$ versus $\frac{1}{X_{CN_{aq}}}$ at different temperatures.

Table 3. Values of true distribution constants at different temperatures

Soaking temperature (K)	K_{true}
302,15	1,036
305,15	1,058
307,15	1,114
309,15	1,148
311,15	1,193

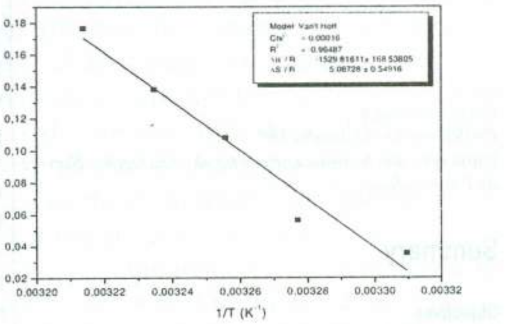


Figure 3. Evolution of the true equilibrium constant with the temperature according to the relation of Van't Hoff.

Table 4. Values of ΔG^o, ΔH^o and ΔS^o for cyanide elimination from NGAMANZA cassava at atmospheric pressure

ΔG^o, ΔH^o in kilojoules by mole, ΔS^o in kilojoules by mole kelvin

T (°C)	ΔG^o	$-T\Delta S^0$	ΔH^o
302,15	- 0,05 ± 0,0001	12,77 ± 1,38	- 12,72 ± 1,40
305,15	- 0,17 ± 0,0001	12,89 ± 1,39	
307,15	-0,26 ± 0,0001	12,98 ± 1,40	ΔS^o
309,15	-0,34 ± 0,0001	13,06 ± 1,41	0,042 ± 0,004
311,15	-0,43 ± 0,0001	13,15 ± 1,42	