Jana Publication & Research

BIOTECHNOLOGICAL POTENTIAL OF YEASTS ISOLATED FROM STREET HOT BEVERAGES FOR THE PRODUCTION OF BI...

07

BioTech

Institut Seni Indonesia Surakarta

Document Details

Submission ID
trn:oid:::1:3186783757

Submission Date
Mar 18, 2025, 2:28 PM GMT+7

Download Date
Mar 18, 2025, 3:58 PM GMT+7

File Name
IJAR-50703.docx

File Size
160.7 KB

9 Pages

4,977 Words

28,782 Characters

23% Overall Similarity

The combined total of all matches, including overlapping sources, for each database.

Filtered from the Report

- Bibliography
- Quoted Text

Match Groups

44 Not Cited or Quoted 14%
Matches with neither in-text citation nor quotation marks

23 Missing Quotations 9%
Matches that are still very similar to source material

0 Missing Citation 0%
Matches that have quotation marks, but no in-text citation

0 Cited and Quoted 0%
Matches with in-text citation present, but no quotation marks

Top Sources

19% Internet sources
17% Publications
7% Submitted works (Student Papers)

Match Groups

- **44** Not Cited or Quoted 14%
  Matches with neither in-text citation nor quotation marks
- **23** Missing Quotations 9%
  Matches that are still very similar to source material
- **0** Missing Citation 0%
  Matches that have quotation marks, but no in-text citation
- **0** Cited and Quoted 0%
  Matches with in-text citation present, but no quotation marks

Top Sources

19% Internet sources
17% Publications
7% Submitted works (Student Papers)

Top Sources

The sources with the highest number of matches within the submission. Overlapping sources will not be displayed.

1	Internet	pubs.ub.ro 2%
2	Student papers	Cavite State University 2%
3	Internet	onlinelibrary.wiley.com 2%
4	Internet	link.springer.com 1%
5	Internet	zenodo.org 1%
6	Internet	www.ajol.info 1%
7	Internet	www.ann.ugal.ro <1%
8	Internet	dent.ibnsina.edu.iq <1%
9	Internet	gsconlinepress.com <1%
10	Internet	pubmed.ncbi.nlm.nih.gov <1%

11	Internet	
fens.usv.ro		<1%
12	Internet	
docslib.org		<1%
13	Internet	
etd.aau.edu.et		<1%
14	Internet	
assets-eu.researchsquare.com		<1%
15	Internet	
recentscientific.com		<1%
16	Internet	
1library.net		<1%
17	Internet	
acervodigital.ufpr.br		<1%
18	Internet	
core.ac.uk		<1%
19	Internet	
irgu.unigoa.ac.in		<1%
20	Internet	
academicjournals.org		<1%
21	Publication	
"Biotechnology of Yeasts and Filamentous Fungi", Springer Science and Business ...		<1%
22	Publication	
"Biotechnology", Wiley, 1995		<1%
23	Publication	
Koua Atobla, Yatanan Casimir Blé, Mohamed Baguy Ouattara, Naka Touré, Dadié ...		<1%
24	Student papers	
Jawaharlal Nehru Technological University		<1%

25 Publication
Krystian Klimczak, Monika Cioch-Skoneczny. "Biotransformation of Hops-Derived ... <1%

26 Internet
docplayer.net <1%

27 Internet
journals.ashs.org <1%

28 Internet
mafiadoc.com <1%

29 Internet
repositorium.sdum.uminho.pt <1%

30 Publication
Hayet Djelal, Sofien Chniti, Monia Jemni, Amélie Weill, Walaa Sayed, Abdeltif Amr... <1%

31 Student papers
McGill University <1%

32 Publication
Charles Y. Tra Bi, Clémentine A. Kouakou-Kouamé, Florent K. N'guessan, Marcelli... <1%

33 Internet
www.journalijar.com <1%

34 Publication
K. C. M. Kouakou, D. B. C. Ebah, S. S. Guédé, G. A. Gbogouri. "Optimization of Proc... <1%

35 Publication
Kouamé Kohi Alfred, Yapi Yapi Eric, N'Cho Mathias, Bouatenin Koffi Maïzan Jean-P... <1%

36 Publication
Shruti Shukla, Juyeon Park, Jung Hyun Park, Jong Suk Lee, Myunghee Kim. "Devel... <1%

37 Internet
attitudepromo.iweventos.com.br <1%

38 Publication
"Fungi and their Role in Sustainable Development: Current Perspectives", Springe... <1%

39 Publication
A.K. Kouamé, M.J.-P.K. Bouatenin, T.N. Djéni, K.M. Dje. " Identification of hazards ... <1%

40 Publication
Arancibia, Mirari Y., M. Elvira López-Caballero, M. Carmen Gómez-Guillén, Marta F... <1%

41 Publication
Fernanda Corrêa Leal Penido, Carmen de Oliveira Goulart, Yara Cristina Fidelis Ga... <1%

42 Internet
dokumen.pub <1%

43 Internet
www.acarindex.com <1%

44 Publication
Balakumar, Sandrasegarampillai, and Vasanthy Arasaratnam. "Enhanced product... <1%

45 Publication
Ifeanyi A. Ndubuisi, Chioma O. Amadi, Tochukwu N. Nwagu, Y. Murata, James C. ... <1%

46 Publication
Lamine Samagaci, Honoré Ouattara, Sébastien Niamké, Marc Lemaire. "Pichia ku... <1%

47 Publication
Marcel Houngbédji, Pernille Johansen, Sègla Wilfrid Padonou, Noël Akissoé et al. "... <1%

48 Internet
dspace.nwu.ac.za <1%

49 Internet
www.mdpi.com <1%

50 Internet
www.researchsquare.com <1%

51 Publication
Walia, Abhishek, Preeti Mehta, Shiwani Guleria, and Chand Karan Shirkot. "Modifi... <1%

52 Publication
Huynh Xuan Phong, Preekamol Klanrit, Ngo Thi Phuong Dung, Mamoru Yamada, ... <1%

Journal Homepage: - www.journalijar.com
INTERNATIONAL JOURNAL OF ADVANCED RESEARCH (IJAR)
Article DOI: 10.21474/IJAR01/xxx
DOI URL: http://dx.doi.org/10.21474/IJAR01/xxx
ISSN NO. 2320-5407

RESEARCH ARTICLE

BIOTECHNOLOGICAL POTENTIAL OF YEASTS ISOLATED FROM STREET HOT BEVERAGES FOR THE PRODUCTION OF BIOETHANOL FROM CASSAVA DOUGH WATER IN CÔTE D'IVOIRE

Manuscript Info

Manuscript History
Received: xxxxxxxxxxxxxxx
Final Accepted: xxxxxxxxxxxx
Published: xxxxxxxxxxxxxxx

Key words:-
Yeasts, cassava dough water, *Attiéké*, bioethanol.

Abstract

Attiéké is one of the dishes most consumed by the Ivorian population, made from fermented cassava dough. However, during this transformation, a significant amount of water, rich in nutrients, is released into the environment without further treatment, which poses numerous environmental problems. Thus, the general objective of this work is to valorize this cassava pulp water for conversion into bioethanol. Yeasts (*Candida kefyr*, *Candida rugosa*, and *K. ohmeri*) isolated from street hot drinks in Côte d'Ivoire were screened and used in a biotechnological application. The results showed that these three strains possess very interesting technological properties such as the ability to produce extracellular enzymes (pectinase, amylase, and cellulase). They exhibit good cell growth at 45 °C and can produce up to 5% ethanol. The fermentation of cassava dough waters from three municipalities in the Abidjan district (Côte d'Ivoire) has shown that these yeasts possess interesting technological abilities and can be proposed as potential starters.

Introduction:-

Scientifically known as *Manihot esculenta* Crantz, cassava is a dicotyledonous angiosperm that is a member of the Euphorbiaceae family. Its cultivation dates back to 1558, when the Portuguese introduced it to Africa from tropical America **(Bombily, 1995)**. Following wheat, rice, corn, potatoes, and barley as the world's six most important crops, cassava is a staple food for 800 million people globally, primarily in the world's poorest areas **(Djilemo, 2007)**. The main source of cassava's nutritional value is its roots, which are high in carbohydrates **(Ribeiros et al., 2019)**. Its high perishability, however, restricts its application and makes processing necessary to produce more stable goods and raise its market value **(Krabi et al., 2015)**.

A variety of meals, including *lafun*, *fufu*, *gari*, *chikwangué*, *agbélima*, and *attiéké*, are thus prepared from fermented cassava **(Assanvo et al., 2006 ; Padonou et al., 2010 ; Krabi et al., 2015)**. According to **Gnagne et al. (2016)**, *attiéké* is the most popular fermented cassava dish in Côte d'Ivoire. Cassava is used to make this steamed semolina, which has a light yellow or whitish hue and a slightly sour flavor **(Djeni et al., 2011)**. About 1,300,000 tons of attiéké are produced annually, according to estimates **(Assanvo et al., 2019)**. A series of sequential processes, including peeling, grinding, fermenting, pressing, sieving, drying, and steaming, are necessary to turn cassava into *attiéké* in an artisanal and empirical manner **(Krabi et al., 2015)**. The process of pressing is the one that enables the removal of a significant portion of the starch and water present in the cassava dough **(Akely, 2012 ; Ayawovi et al., 2016)**. Additionally, drainage makes it possible to get rid of cyanogenic substances. This operation includes the

Corresponding Author: Wilfried K. YAO
Address: yaowilfried22@gmail.com; Laboratory of Biotechnology, Agriculture and Valorization of Biological Resources, Faculty of Biosciences, Félix Houphouët-Boigny University, Abidjan, Côte d'Ivoire, 01 BPV 34 Abidjan 01

ISSN: 2320-5407 Int. J. Adv. Res. 7(6), XX-XX

cause pollution and produces unpleasant odors. Three hundred (300) liters of effluent are thought to be produced for every ton of cassava **(Costa et al., 2010)**. It would therefore be interesting for environmental preservation to recycle this pressing water. In addition, one of the methods for valorizing this press water is its transformation into bioethanol by yeasts. Indeed, this sugar-rich water could be an ideal substrate for fermenting yeasts **(De Andrade et al., 2016)**. Moreover, several research studies have already used yeasts for the recycling of wastewater in agro-industry **(Kassime et al., 2016 ; Kurcz et al., 2018 ; Ribeiro et al., 2019 ; Nicula et al., 2023)** and the production of bioethanol from industrial waste **(Boulal et al., 2010 ; Isla et al., 2013**). Thus, the objective of this study is to valorize cassava dough water with a view to its transformation into bioethanol by yeasts.

Materials and methods:-

Materials

Yeasts strains used in this study were previously isolated from street hot beverages made of tea and coffee collected in Cocody and Port-Bouet **(Atobla et al., 2021)** and identified by Maldi-Tof at the biobank laboratory of the Pasteur Institute of Côte d'Ivoire (Table 1). Yeasts strains were stored on Sabouraud chloramphenicol agar (Biokar Diagnostics, France), added with 30 % glycerol at -20 °C.

Table I: Yeast strains used in this study

Yeast strains	Strains code	Ecological niche	Isolation areas
Candida rugosa	IV5C2	Coffee	Cocody
Candida kefyr	V13T2	Tea	Port-Bouet
Kodamaea ohmeri	V3C5	Coffee	Port-Bouet

Methods

Effects of temperature and ethanol production by yeast strains

Effect of temperature on the growth of yeasts strains were analyzed in liquid medium as described **by Samagaci et al. (2014)**. A pure culture of yeasts was suspended in a sterile saline solution to give an optical density of 0.2 at 600 nm and then 200 µL of this suspension were added in 10 mL of Malt Yeast Glucose Peptone (MYGP) broth. The preparation is then incubated at different temperatures (30-45 °C) for 5 days. After incubation, the microbial growth was measured evert day by reading the optical density of culture with a spectrophotometer at 600 nm against a control. In parallel, the ethanol produced by the isolates was determined by removing a volume of 1.5 mL of each culture medium into Eppendorf tubes. Then the supernatant was collected after centrifugation at 10,000 rpm for 10 min at 4 °C and the ethanol content was determined using a vinometer **(Wendt, 2012)**. The ethanol content was determined by reading the graduation at the precise point where the liquid was stabilized **(Fers-Lidou et al., 2016)**.

Screening of hydrolytic enzymes by yeast strains

Hydrolytics enzymes production was carried out according to the method of **Samagaci et al. (2016)** on agar medium. First, yeasts strains were cultived in Malt Yeast Glucose Peptone (MYGP) broth, and optical density was read with a spectrophotometer at 600 nm after 24 h of culture. Then, a minimum medium containing 0.28% NH_4SO_4, 0.6% K_2HPO_4, 0.01% $MgSO_4$, 0.2% KH_2PO_4, 0.02% yeast extract and 1.7% agar was supplemented with different substrates at 1% polygalacturonic acid (PGA) for polygalacturonase, starch (for amylase), carboxymethylcellulose (CMC) for cellulase and Tween 80 for lipase, were used. Inoculation of the isolates was carried out described by **Yao et al. (2014)** and the petri dishes were incubated at 30°C during to 48h. After incubation, the clear zones around the wells, indicating hydrolytic activity were revealed with a solution of iodine and potassium iodide (5g potassium iodide + 1g iodine + 330mL distilled water) as described by **Soares et al. (1999)**.

After studying the technological properties of the yeasts, the isolates with the best potential were used for ethanol production from fermented cassava dough water.

Collect of cassava dough water

For the traditional production of Attiéké, several studies have described the production steps **(Krabi et al., 2015).** The water from the fermented cassava paste was collected during the mechanical pressing stage after fermentation. Thus, the grinded and fermented cassava was separated from its juice for several hours by compression with a mechanical press. A yellowish or whitish liquid is then collected in a container under the press. Cassava water recupered was stored in a cooler and sent to the laboratory for physico-chemical analysis. Samples from different communes (Yopougon, Cocody and Abobo) of Abidjan (Côte d'Ivoire) were separated into two 400 mL bottles.

ISSN: 2320-5407 Int. J. Adv. Res. 7(6), XX-XX

Unfermented cassava dough water characterization

Fresh unfermented dough waters were characterized by determining the following parameters: reducing sugars, pH, alcohol content and Brix.

Determination of reducing sugars of unfermented cassava dough water

Reducing sugars content was carried to the method of **Bernfeld (1955)**. To 200 µL of DNS solution was added 100 µL of fermented cassava dough water. The whole was heated in a boiling water bath for 5 minutes. After cooling, 1.7 mL of distilled water was added and then the absorbance of the solution is read at 540 nm, against a blank. A standard range was established with a stock glucose solution (1 mg/mL) in order to determine the amount of reducing sugars.

Determination of pH, ethanol and Brix degree of unfermented cassava dough water

pH of fermented cassava dough water was determined using a pH-meter by the method described by **AOAC (1990)**. Ethanol content in percentage was obtained using the method of **Wendt (2012)**. Soluble dry extract of cassava dough water was determined according to **AOAC (2005)**.

Biotechnological application of yeasts for the production of bioethanol

Fermentation was carried in Erlenmeyer flasks containing 200 mL of cassava fermented dough water sterile inoculated with a 2 mL bacterial suspension to OD 0.5 at 600 nm of *non-Saccharomyces* yeasts (Table I) and baker yeast *Saccharomyces cerevisiae* in pures cultures and incubated for 120 hours at 30 °C **(Dung et al., 2012)**. Every 24 hours, 10 mL of samples were removed for physico-chemical analyses such as pH, reducing sugar content, Brix degree, and ethanol amount.

Statistical analysis of data

All these analyses were carried out in triplicate. Mean and standard deviation were calculated. Analysis of variance (ANOVA) was performed using SPSS Statistics 20.0 software, and significant differences between means were determined using Duncan's test at the 5% level.

Results:-

Effect of temperature on the growth and ethanol production by yeasts strains

Effect of temperature on growth and ethanol production were carried in liquid medium and the results are reported in Table II. All isolates showed growth at all temperatures tested (30-45 °C). An increase in bacterial growth observed up to a peak of 35 °C (*C. rugosa* and *K. ohmeri*) and 40 °C (*C. kefyr*) and after these peaks, a decrease in microbial growth. *Candida kefyr* showed a good growth at the different temperatures tested with a relative growth above 50 %. Ethanol production correlates with the microbial growth of the different strains. The highest ethanol production observed at 30 °C with *K. ohmeri* produced the alcohol level of 5.2 %. However, *C. rugosa* and *C. kefyr* were able to produce ethanol at all temperatures tested compared to *K. ohmeri*, which did not produce ethanol at 45 °C.

Table II: Effect of temperature on growth and ethanol production by yeasts strains isolated

Yeasts	Relative growth (%)				Ethanol production (%)			
	30 °C	35 °C	40 °C	45 °C	30 °C	35 °C	40 °C	45 °C
C. rugosa	33.5±0.76^{c}	100±0.60^{a}	80.2±0.94^{b}	11.3±0.9^{d}	4.14±0.21^{a}	3.5±0.44^{b}	2.2±0.09^{c}	2.0±0.06^{c}
C. kefyr	93.7±1.35^{b}	90.2±2.40^{b}	100±2.96^{a}	57.6±1.29^{c}	3.8±0.4^{b}	3.4±0.15^{b}	4.8±0.36^{a}	2.5±0.21^{c}
K. ohmeri	71.8±2.05^{b}	100±1.93^{a}	30.9±1.3^{c}	0.5±0.03^{d}	5.2±0.46^{a}	2.7±0.30^{b}	2.1±0.10^{c}	0±00^{d}

Data are represented as means±SEM (n=3). Mean with different letters in the same line are statistically different ($p<0.05$) according to Duncan's test

Hydrolytics enzymes production by yeast strains

The selection criteria based on the extracellular enzymatic activities used for the *non-Saccharomyces* yeasts, ensure the selection of the biotechnological potential yeast strains of interest in ethanol production. Investigation of

ISSN: 2320-5407 Int. J. Adv. Res. 7(6), XX-XX

hydrolytic activities showed that all these isolates are capables of producing amylase, cellulase polygalacturonase and pectinase on solid media. Concerning lipase, only *C. kefyr* strain was no able to degrade lipids (Table III).

Table III: Production of hydrolytics enzyms by yeasts strains

Yeasts	Enzymes				
	Amylase	Cellulase	Polygalacturonase	Pectinase	Lipase
C. rugosa	+	+	+	+	+
C. kefyr	+	+	+	+	-
K. ohmeri	+	+	+	+	+

(+) : presence of halo; (-) : absence of halo

Valorization trial of unfermented cassava dough water

Physico-chemical characterization of unfermented cassava dough water

Different samples of unfermented cassava dough water from three municipalities (Abobo, Cocody and Yopougon) in the district of Abidjan (Côte d'ivoire) were subjected to some physico-chemical analysis. The average values of these parameters are summarised in Table IV. The pH measurement revealed that not fermented cassava dough water is acidic with significant differences according to statistical analysis. It was on average 3.45±0.14 in Abobo, against 3.8±0.1and 3.65±0.14 in Yopougon and Cocody, respectively. The quantity of reducing sugars expressed was on average 0.302±0.02 g/100 mL in Yopougon, 0.488±0.03 g/100 mL in Cocody and 0.156±0.03 g/100 mL for Abobo. As for the dry extract, statistical analysis revealed that there was no significant difference in Brix values for samples from the municipalities of Yopougon and Abobo. Nevertheless, the values were ranged from 5.6±0.2 and 5±0.29 °B. The results in Table IV revealed that cassava dough water had an initial ethanol content. Alcohol content was higher in the juice from Abobo and Yopougon with average values of 1.71±0.1 and 1.65±0.13% respectively, while the Cocody sample had an ethanol content of 1.3±0.2%.

Table IV: Physico-chemical parameters of unfermented cassava dough water

Physico-chemical parameters	Yopougon	Cocody	Abobo
Reducing sugars (g/100 mL)	0.302±0.02^{b}	0.488±0.03^{a}	0.156±0.03^{c}
Brix (°)	5.5±0.10^{a}	5.0±0.29^{b}	5.6±0.20^{a}
pH	3.8±0.10^{a}	3.65±0.15^{ab}	3.45±0.14^{b}
Alcohol content (%)	1.65±0.13^{a}	1.3±0.20^{b}	1.71±0.10^{a}

Data are represented as means±SEM (n=3). Mean with different letters in the same line are statistically different ($p<0.05$) according to Duncan's test

Fermentation parameters of yeast strains for ethanol production from cassava dough water

The pH, Brix degree, and the amount of reducing sugars during the fermentation of cassava dough water by yeasts are summered in Table IV. Regarding the pH, its value changed, compared to initial value, which was between 3.45±0.01 and 3.8±0.0. Statistical analysis revealed significant differences in the parameters studied by municipality. The fermentation by yeasts of cassava dough water from the three municipalities resulted in a pH between 3.35±0.0 and 3.59±0.01 compared to the control (LBSC) which had a pH between 3.34±0.0 and 3.62±0.0 at the end of fermentation (120 h). As for reducing sugars, an increase in sugar content is observed both for the tests and for the control. At the beginning of fermentation, the content of reducing sugars ranges between 0.156±0.02 and 0.488± 0.04 g/100 mL. At the end of the 120 hours of fermentation, the content of reducing sugars reached values that oscillated between 0.359±0.03 and 0.759±0.05 g/100 mL for the trials and the control (LBSC). Finally, the Brix degree values at the end of the fermentation time (120 h) for the different trials and the control range between 2±0.0 and 4.2±0.0.

Table V presents the ethanol production of yeasts strains from cassava dough waters originating from three municipalities (Abobo, Cocody, and Yopougon) in the Abidjan district. The results indicated an increase in ethanol concentration throughout fermentation for both the tests and the control (LBSC) which is a strain of *Saccharomyces*

cerevisiea. Ethanol production for all strains and the control (LBSC) ranged from 5.8±0.02 to 10.4±0.03 %. This value of 10.4±0.0 % ethanol was obtained with the *K. ohmeri* strain for the sample from the Cocody municipality. The results of co-culture and monoculture gave the same results compared to the control, which provided lower values for the studied parameters.

Table IV: Average pH, reducing sugars and brix of cassava dough water at the end of fermentation

Municipalities	Strains	Reducing sugars (g/100 mL)	pH	Brix
Abobo	LBSC	0.359±0.03^{c}	3.36±0.0^{b}	2.0±0^{c}
	C. rugosa	0.479±0.02a^{b}	3.41±0.03^{a}	2.3±0.1^{a}
	K. ohmeri	0.421±0.06^{b}	3.37±0.02^{b}	2.2±0.05^{b}
	C. kefyr	0.441±0.01^{b}	3.40±0.03^{a}	2.0±0.06^{c}
	Mix	0.504±0.02^{a}	3.35±0.0^{b}	2.0±0^{c}
Cocody	LBSC	0.759±0.05^{a}	3.34±0.0^{d}	3.3±0.1^{b}
	C. rugosa	0.679±0.04^{bc}	3.46±0.0^{a}	3.6±0.12^{a}
	K. ohmeri	0.631±0.02^{cd}	3.38±0.04^{c}	3.4±0.11^{b}
	C. kefyr	0.741±0.04^{ab}	3.40±0.01^{b}	3.1±0.07^{c}
	Mix	0.594±0.03^{d}	3.45±0.02^{a}	3.2±0^{bc}
Yopougon	LBSC	0.637±0.05^{a}	3.62±0.0^{a}	4.0±0^{b}
	C. rugosa	0.438±0.06^{c}	3.52±0.02^{b}	3.6±0.06^{c}
	K. ohmeri	0.515±0.03b^{c}	3.48±0.05^{c}	4.2±0.20^{a}
	C. kefyr	0.714±0.02^{a}	3.59±0.01^{a}	4.1±0.1^{ab}
	Mix	0.546±0.06^{b}	3.54±0.0^{b}	4.0±0.1^{ab}

LBSC: Baker's yeast *Saccharomyces cerevisiae;* MIX: *C. rugosa*, C. *kefyr* and *K. ohmeri*. The parameters studied were analysed by municipality. Data are represented as means±SEM (n=3). Mean with different letters in the same column are statistically different ($p<0.05$) according to Duncan's test.

Table V: Changes in ethanol concentration during fermentation of cassava dough water

Municipalities	Strains	Ethanol production (%)				
		24 h	48 h	72 h	96 h	120 h
Abobo	LBSC	2.7±0.04^{e}	3.4±0.03^{d}	4.5±0.00^{c}	5.4±0.04^{b}	5.8±0.02^{a}
	C. rugosa	2.6±0.05^{e}	4.4±0.04^{d}	5.2±0.07^{c}	6.6±0.04^{b}	7.3±0.06^{a}
	K. ohmeri	2.7±0.00^{e}	4.8±0.05^{d}	5.8±0.04^{c}	6.9±0.02^{b}	7.8±0.03^{a}
	C. kefyr	2.5±0.04^{e}	3.8±0.02^{d}	4.6±0.03^{c}	5.7±0.04^{b}	6.8±0.02^{a}
	Mix	2.5±0.03^{e}	3.6±0.02^{d}	4.0±0.05^{c}	5.2±0.03^{b}	6.5±0.04^{a}
Cocody	LBSC	2.8±0.02^{e}	3.8±0.05^{d}	5.4±0.03^{c}	6.2±0.07^{b}	7.0±0.01^{a}
	C. rugosa	2.7±0.01^{e}	4.1±0.02^{d}	5.7±0.01^{c}	6.7±0.03^{b}	7.5±0.05^{a}
	K. ohmeri	3.2±0.03^{e}	5.6±0.01^{d}	7.6±0.05^{c}	9.2±0.04^{b}	10.4±0.02^{a}
	C. kefyr	3.0±0.03^{e}	5.2±0.03^{d}	6.9±0.08^{c}	8.1±0.01^{b}	9.4±0.05^{a}
	Mix	2.8±0.05^{e}	4.4±0.01^{d}	5.7±0.06^{c}	6.1±0.04^{b}	7.7±0.05^{a}
Yopougon	LBSC	2.2±0.04^{e}	4.2±0.04^{d}	4.8±0.05^{c}	5.9±0.06^{b}	7.0±0.04^{a}
	C. rugosa	2.6±0.04^{e}	5.1±0.03^{d}	7.7±0.06^{c}	8.7±0.02^{b}	9.5±0.06^{a}
	K. ohmeri	2.0±0.05^{e}	4.3±0.04^{d}	5.3±0.01^{c}	6.0±0.05^{b}	7.3±0.05^{a}
	C. kefyr	2.4±0.03^{e}	4.8±0.04^{d}	7.5±0.08^{c}	8.3±0.06^{b}	9.5±0.02^{a}
	Mix	2.6±0.02^{e}	4.5±0.06^{d}	6.9±0.06^{c}	8.5±0.04^{b}	9.3±0.05^{a}

LBSC: Baker's yeast *Saccharomyces cerevisiae;* MIX: *C. rugosa*, C. *kefyr* and *K. ohmeri*. The parameters studied were analysed by municipality. Data are represented as means±SEM (n=3). Mean with different letters in the same line are statistically different ($p<0.05$) according to Duncan's test.

DISCUSSION:-

ISSN: 2320-5407 Int. J. Adv. Res. 7(6), XX-XX

According to **Coelho et al. (2020)**, cassava plays a key role in the food production and economies of several countries worldwide. Due to its starch content, alcoholic fermentation of cassava dough water could add value to cassava. In our study, we used yeasts such as *Candida rugosa*, *Candida kefyr*, and *Kodamea ohmeri* from street hot beverages. Various yeasts species *non-Saccharomyces* were often isolated from Africa foods and beverages fermentations such as *Candida lusitania*, *Cryptococcus laurentii*, *Candida kefyr*, *Issatchenkia orientalis*, *Kazachstania unisporus*, *Rhodotorula mucilaginosa*, *Candida pararugosa* **(Mogmenga et al., 2019)**.
It is important that the microorganisms to be used in the bio-industry are capable of withstanding the constraints imposed by these environments **(Rezki-Bekki, 2014)**. Temperature has an influence on ethanol production and growth yeasts. Indeed, our results are in agreement with those of **Phong et al. (2019)** who showed a decrease in ethanol concentration when the temperature of the fermentation increased. In addition, these authors showed that these yeasts were able to grow at temperatures between 40 and 42°C. According to **D'Amore et al. (1989)**, high temperatures caused the intracellular ethanol of yeast cells to accumulate and halt its growth, which in turn slowed the yeast's fermentation activity and reduced the amount of ethanol produced. Moreover, more, the growth of *non-Saccharomyces* species belonging to the genera *Kloeckera*/*Hanseniaspora* and *Candida* is generally limited to the first few days of fermentation, because of their weak ethanol tolerance. However, quantitative studies on grape juice fermentation have shown that *Kloeckera apiculata* and *Candida stellata* can survive at significant levels (up to 10^6-10^7 CFU/mL) during fermentation, and for longer periods than thought previously **(Pardo et al., 1989)**.

Moreover, the study also showed that the tested yeasts strains exhibit the ability to synthesize enzymes of interest. This ability of the studied strains to synthesize enzymes of the hydrolase class is a significant advantage. Reaserch by **Strauss et al. (2001)** reported that *non-saccharomyces* yeasts (*Candida stellata*, *Kloeckera apiculata*) were capable of producing amylase, pectinolytic enzymes, and cellulase. Furthermore, the work of **Escribano et al. (2017)** revealed that *non-Saccharomyces* yeasts strains isolated from different oenological origins such as *Torulaspora deldrueckii*, *Debaromyces hansenii*, *Metchnikowia pulcherina* and *Cryptococcus* sp. were capables of producing both cellulase and pectinolytic enzyms. This activity was also found in the work of **Bouatenin et al. (2016)**. The use of these strains as starters could improve the optimization of the fermentation process but would also contribute to the rapid and significant production of organic acids and volatile compounds necessary for the production of aroma and flavor for attiéké production **(Djeni et al., 2008)**. The ability of these strains to produce these extracellular enzymes could play a role in the softening of cassava dough and the hydrolysis of starch into free sugar.

Regarding the valorization trial of the pressing juice from fermented cassava dough, three strains (*C. rugosa*, *C. kefyr*, and *K. ohmeri*) exhibiting a good alcohol production rate and possessing the ability to synthesize sought-after enzymes were selected. The aim of this trial was to evaluate the fermentation potential of these strains while valorizing the pressing juices of fermented cassava dough. In African countries, most industrial or traditional residues are discarded into nature and constitute a source of environmental pollution. Several studies on bioconversion processes using yeasts such as *Saccharomyces cerevisiae*, *Candida utilis*, *Candida tropicalis*, *Rhodotorula muciloginosa* to obtain value-added products from industrial residues have been conducted **(Mogmenga et al., 2019)**.

The pH of the cassava dough water after fermentation was lower than the original pH, which may suggest that the fermentation process was successful. Work of **Ribeiro et al. (2019)** obtained an acidic pH for cassava wastewater in Brazil, which is consistent with our results. The yeasts transformed glucose into alcohol and other intermediate compounds, such as organic acids, throughout the fermentation process, which may be the cause of this pH drop **(Dung et al., 2012)**. Cassava is rich in reducing sugars and starch. However, starch is the main macromolecule in cassava **(Ribeiro et al., 2019)**. During fermentation, the extracellular enzymes produced by yeasts will hydrolyze the starch into simple sugars like glucose, which in turn will undergo fermentation into ethanol. This fermentation will lead to a decrease in the Brix degree and an increase in the amount of reducing sugars. The amount of ethanol produced in our study is similar to those obtained in the work of **Dung et al. (2012)**, which range from 5.83 to 7 %.

Several research studies in the literature have used yeast co-culture for bioethanol production. These studies have shown a good cohabitation of these strains accompanied by a good ethanol production yield **(De Bari et al., 2013 ; Karagöz & Özkan, 2015 ; Lujan-Rhenals et al., 2015)**. All the technological properties of these yeasts make them good candidates for the alcoholic fermentation of various substrates.

Conclusion:-

ISSN: 2320-5407 Int. J. Adv. Res. 7(6), XX-XX

Characterization of these yeasts strains indicates that autochthonous non-*Saccharomyces* yeasts could have a positive impact on the ethanol productions that contribute to biotechnology application. This study showed that the isolated yeasts are capable of producing extracellular enzymes for the degradation of starch and other substrates contained in cassava paste water. They exhibit good growth and ethanol production at high temperatures (45 °C) in a liquid medium. The biotechnological application of these yeasts in alcoholic fermentation has demonstrated that these strains possess characteristics that make them potential starters.

Acknowledgements:-

The authors thank street beverage vendors. This work was supported by ASCAD (Academy of Sciences, Arts, Cultures of Africa and African Diasporas) funds.

References:-

1. Akely, P.M.T. (2012). Influence of controlled fermentation, pressing, and mechanized grating of cassava (*Manihot esculenta* Crantz) on the physicochemical and sensory characteristics of "*attiéké*". [Doctoral Thesis]. Nanguy Abrogoua University, Abidjan (Côte d'Ivoire), 160p.
2. AOAC (1990). Official methods of analysis of the Association of Official Analytical Chemists, 15th edition, volume 1, Washington DC, 771 p.
3. AOAC (2005). Official methods of analysis of the Association of Official Analytical Chemists, 17th Edition Washington, DC.
4. Assanvo, J.B., Agbo, G. and Farah, Z. (2019). Surveys on the Consumption of *attiéké* (Traditional and a commercial, *garba*) in Côte d'Ivoire. J. Food Res., 8: 23-29.
5. Assanvo, J.B., Agbo, G.N., Behi, Y.E.N., Coulin, P. and Farah, Z. (2006). Microflora of traditional starter made from cassava for *attiéké* production in Dabou (Côte d'Ivoire). Food control, 17: 37-41.
6. Atobla, K., Blé, Y.C., Ouattara, M.B., Touré, N., Dadié, A. and Niamké, S. (2021). *Candida* spp. associated with hot beverages of coffee and tea sold on street in Côte d'Ivoire. Afr. J. Microbiol. Res., 15: 476-481
7. Ayawovi, E., Regina, K., Antoine, A.A. and Niamké, S. (2016). Towards added value *attieké* production in Côte d'Ivoire using *Bacillus* spp. as starters. Turk. J. Agric. Food Sci. Techno., 4: 1077-1084.
8. Bernfeld, P. (1955). Amylases, alpha and beta. Method. Enzymol., 1: 149-158.
9. Bombily, M.S. (1995). Cassava: an important food resource in Guinea. In: Agbor E., Brauman A., Griffon D., Trèche S., eds.: Orstom. Paris. Food processing of cassava. 56-61 - UNCTAD, (2012). Produced sheet: Cassava. Available at: http://www.unctad.info/en/Infocomm/Products-AAACP/PRODUCT-SHEET---Cassava/ (on 08/01/12)
10. Bouatenin, K.M. J.-P., Djéni N.T., Kakou, A.C., Menan, E.H. and Djé, K.M. (2016). Optimization of α-amylase production by microorganisms isolated from traditional cassava ferments from three *attiéké* production areas in Côte d'Ivoire. Eur. Sci. J., 12: 259-272.
11. Boulal, A., Benali, B., Moulai, M. and Touzi, A. (2010). Processing date waste from the Adrar region into bioethanol. Renew. Energy. Rev., 13: 455-463.
12. Coelho, E., Ballesteros, L.F., Domingues, L., Vilanova, M. and Teixeira José A. (2020). Production of a distilled spirit using cassava flour as raw material: chemical characterization and sensory profile. Molecules, 25:1-13.
13. Costa, S.G.V.A.O., Nitschke, M., Lépine, F., Déziel, E. and Contiero, J. (2010). Structure, properties and applications of rhamnolipids produced by *Pseudomonas aeruginosa* L2-1 from cassava wastewater. Process Biochem., 45: 1511-1516.
14. D'Amore, T., Celotto, G., Russell, I. and Stewart G.G (1989). Selection and optimization of yeast suitable for ethanol production at 40 °C. Enzyme Microb. Technol., 11: 411-416.
15. De Andrade, C.J., Barros, F.F., De Andrade, L.M., Rocco, S.A., Luis Sforça, M., Pastore, G.M. and Jauregi, P. (2016). Ultrafiltration based purification strategies for surfactin produced by *Bacillus subtilis* LB5A using cassava wastewater as substrate. J. Chem. Technol. Biotechnol., 91: 3018-3027.
16. De Bari, I., De Canio, P., Cuna, D., Liuzzi, F., Capece, A. and Romano, P. (2013). Bioethanol production from mixed sugars by *Scheffersomyces stipitis* free and immobilized cells, and co-cultures with *Saccharomyces cerevisiae*. New Biotechnol., 30: 591-597.
17. Djeni, N.T., N'guessan, K.F., Dadié, A.T. and Djé, K.M. (2008). Impact of different levels of a traditional starter on the fermentation of cassava dough for *attiéké* production. Food, 2: 155-151.
18. Djeni, N.T., N'guessan, K.F., Tokan, D.M., Kouame, K.A. and Djé, K.M. (2011). Quality of *attieke* (a fermented cassava product) from the three main processing zones in Côte d'Ivoire. Food Res. Int., 44: 410-416.

19. Djilemo, L. (2007). Cassava flour (*Manihot Esculenta* Crantz) non-fermented: The future of cassava cultivation in africa. In: international cassava workshop, Abidjan, Côte d'Ivoire, June 4-7, 2007.
20. Dung, N.T.P., Thanonkeo, P. and Phong, H.X. (2012). Screening useful isolated yeasts for ethanol fermentation at high temperature. Int. J. Appl. Sci. Technol, 2: 65-71.
21. Escribano, R., Lucia Gonzalez-Arenzana, L., Patrocinio Garijo, P., Berlanas, C., Lopez-Alfaro, I., Lopez, R., Gutierrez, A.R. and Santamaria, P. (2017). Screening of enzymatic activities within different enological *non-Saccharomyces* yeasts. J. Food Sci. Technol., 56: 1555-1564.
22. Fers-Lidou, A., Berger, O. and Montchamp, J.-L. (2016). Palladium-catalyzed allylation/benzylation of H-phosphinate esters with alcohols. Molecules, 21: 2-15.
23. Gnagne, G.B.A.A., Koffi, E.K., Assanvo, J.B. and Soro, S. (2016). Influences of freezing and drying of *attiéké* on its physicochemical and organoleptic characteristics. Int. J. biol. chem. sci., 10: 808-819.
24. Isla, M.A., Comelli, R.N. and Seluy, L.G. (2013). Wastewater from the soft drinks industry as a source for bioethanol production. Bioresour. Technol., 136: 140-147.
25. Karagöz, P. and Özkan, M. (2015). Ethanol production from wheat straw by *Saccharomyces cerevisiae* and *Scheffersomyces stipitis* co-culture in batch and continuous system. Bioresour. Technol., 153: 286-293.
26. Kasmi, M., Chatti, A., Hamdi, M. and Trabelsi, I. (2016). Eco-friendly process for soft drink industries wastewater reuse as growth medium for Saccharomyces cerevisiae production. Clean. Techn. Environ. Policy., 18: 2265-2278.
27. Krabi, E.R., Assamoi, A.A., Ehon, A.F., Bréhima, D., Niamké, L.S. and Thonart, P. (2015). Production of *attiéké* (couscous made from fermented cassava) in the city of Abidjan. Eur. Sci. J., 11: 277-292.
28. Kurcz, A., Błazejak, S., Kot A.M., Bzducha-Wrobel, A. and Kieliszek, M. (2018). Application of industrial wastes for the production of microbial single-cell protein by fodder yeast *Candida utilis*. Waste Biomass Valor., 9: 57-64.
29. Lujan-Rhenals, D.E., Morawicki, R.O., Gbur, E.E. and Ricke S.C. (2015). Fermentation of soybean meal hydrolyzates with *Saccharomyces cerevisiae* and *Zymomonas mobilis* for ethanol production. J. Food Sci., 80: 1512-1518.
30. Mogmenga, I., Somda, M.K., Ezeogu, L.I., Ugwuanyi, J. and Traoré, A.S. (2019). Yeasts biotechnologies application and their involving in african traditional fermented foods and beverages. Int. J. Adv. Res., 7: 44-62.
31. Nicula, N.-O., Lungulescu, E.-M., Rîmbu, G.A., Marinescu, V., Corbu, V.M. and Ortansa Csutak O. (2023). Bioremediation of Wastewater Using Yeast Strains: An assessment of contaminant removal efficiency. Int. J. Environ. Res. Public Health., 20: 2-22.
32. Padonou, S.W., Nielsen, D.S., Akissoe, N.H., Hounhouigan, J.D., Nago, M.C. and Jakobsen, M. (2010). Development of starter culture for improved processing of *Lafun*, an African fermented cassava food product. J. Appl. Microbiol., 109:1402-1410.
33. Pardo, I., Garcia, M.J., Zuniga, M. and Uruburu, F. (1989). Dynamics of microbial populations during fermentation of wines from the Utiel-Requena region of Spain. Appl. Environ. Microbiol., 55: 539-541.
34. Phong, H.X., Klanrit, P., Dung, N.T.P., Mamoru Yamada, M. and Thanonkeo, P. (2019). Isolation and characterization of thermotolerant yeasts for the production of second-generation bioethanol. Ann. Microbiol., 69:765-776.
35. Rezki-Bekki, M.A. (2014). Production of metabolites by yeasts: characterisation and identification of aromas and alcohols. Thesis, Departement of Nanotechnology, University of Oran, Algeria, 161 p.
36. Ribeiro, J.E.S., Sant'Ana, A.M.S., Martini, M., Sorce, C., Andreucci, A., de Melo, D.J.N. and Da Silva, F.L.H. (2019). *Rhodotorula glutinis* cultivation on cassava wastewater for carotenoids and fatty acids generation. Biocatal. Agric. Biotechnol., 22: 1-8.
37. Samagaci, L., Ouattara, H., Niamké, S.and Lemaire, M. (2016). *Pichia kudrazevii* and *Candida nitrativorans* are the most well-adapted and relevant yeast species fermenting cocoa in Agneby-Tiassa, a local Ivorian cocoa producing region. Food Res. Int., 89: 773-780.
38. Samagaci, L., Ouattara, H.G., Goualié, B.G., Niamké, S.L. (2014). Growth capacity of yeasts potential starter strains under cocoa fermentation stress conditions in Ivory Coast. Emir. J. Food Agric., 26: 861-870.
39. Soares, M.M.C.N., Da Silva, R. and Gomes, E. (1999). Screening of bacterial strains for pectinolytic activity: characterization of the polygalacturonase produced by *Bacillus* sp. Rev. Microbiol., 30, 299-303.
40. Strauss, M.L., Jolly, N.P., Lambrechts, M.G. and Van Rensburg, P. (2001). Screening for the production of extracellular hydrolytic enzymes by *non-Saccharomyces* wine yeasts. J. Appl. Microbiol., 91: 182-190.
41. Wendt, T. (2012). Analysing wine at school. Sci School, 24: 47-52.

42. Yao, W., Ouattara, G.H., Doué, G., Goualié, G.B., Gisèle A. Koua, A.G. and Niamké, S. (2015). Emphasis on Techno-functional properties of *Bacillus* strains involved in ivorian cocoa fermentation towards their use as potential starter. Br. Microbiol. Res. J., 10: 1-10.