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Characterization and Screening of Lactic Acid Bacteria (LAB) and Yeast from Fermented Sorghum and Maize Products for use as Starter Culture

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Authors' contributions

This work was carried out in collaboration among all authors. All authors read and approved the final manuscript.

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ABSTRACT

This study focused on the isolation, Characterization and screening of lactic acid bacteria and yeast for use as starter culture in order to eradicate the problem of excessive microbial contamination and the presence of mycotoxins.
Lactobacillus plantarum and *Saccharomyces cerevisiae* were the predominant microorganisms isolated from the samples collected. The isolates were screened for their ability to produce enzymes and metabolites. Lactic acid bacteria produce a variety of antimicrobial compounds such as lactic acid, hydrogen peroxide, diacetyl on them as a natural competitive means to overcome other microorganism sharing the same niche. They were also screened for their ability to withstand some physiological stress like acid tolerance, temperature, salt concentration and antibacterial activity; the isolates produced significant values of enzymes and, antimicrobial metabolites. The antimicrobial activity of *Lactobacillus plantarum* is mainly attributed to the low pH due to Lactic acid production. The antimicrobial activity of the *Saccharomyces cerevisiae* appears to be mainly due to the competition with the other microorganisms. In addition, depletion of Oxygen and production of CO_2, competition for nutrients and the production of antimicrobial substances could have been responsible for the overall antimicrobial activity of both cultures. According to the results obtained, both lactic acid bacteria and yeast proved to be a good source of starter culture.

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1. INTRODUCTION

The sorghum grain is a flattened sphere with variable size and colours but made up of anatomical features called the pericarp-testa, the aleurone layer, the embryo and the starchy endosperm [1,2]. The pericarp is an outer protective layer making up to 5-6% of the kernel weight. It is a rich source of dietary fibre, minerals and vitamins. The endosperm is the storage tissue and the largest part of the kernel and also a rich source of both starch and protein. The relative proportion of protein and starch in the endosperm is the most important factor affecting grain hardness and density. Sorghum belongs to the grass family Gramineae. It is well adapted to the tropical weather of the African continent [3]. This makes sorghum one of the cereals which constitute a staple for people in sub-Saharan Africa [4]. Sorghum is consumed as porridge and as malted and distilled beverages in Africa and Asia and used as syrup, animal feed and ethanol production in the US and other developed countries [5]. Nigeria is the number one producer of sorghum in Africa followed in order by Sudan, Ethiopia, Somalia and Burkina Faso [6, 7].

Sorghum malt is used extensively in many African countries to produce local 'opaque' beer, although it is also used in modern breweries as adjuncts to produce 'Lager' beer. It's generally known that naked cereal grains such as sorghum and millet used in beer production are not protected by the presence of husks and are prone to mycotoxins contamination due to the presence of toxinogenic moulds [8].

Naturally occurring moulds grow easily on sorghum grains during malting or high moisture storage conditions which are the main stage of African opaque beer process production. The growth of moulds such as *Aspergillus flavus, Penicillium parasiticus, Fusarium graminearum, F. culmorum, F. roseum* and *F. moniliforme* on grains or during malting are known to elaborate aflatoxins, trichothecenes, fumonisins, Ochratoxin A and zearalenones, among other mycotoxins [9]. Most traditional malting process does not have any mechanism for controlling the microbial load during malting. Although in the industrial brewing sector a variety of chemical treatments are used. The chemical used include calcium hydroxide $Ca(OH)_2$, sodium hydroxide (NaOH) and Formaldehyde [9,10].

Lactic acid bacteria (LAB) are commonly defined as gram-positive, non-sporulating, catalase-negative, aero-tolerant, acid tolerant, nutritionally fastidious, strictly fermentative organisms that lack cytochromes and produce lactic acid as the major end product of carbohydrate metabolism [11]. LAB, in general, do not produce enzymes that can enhance malt modification. These organisms are, however, good protease producers and it has been demonstrated that proteolytic strains can increase the nitrogenous content of malt [9,12].

In contrast to LAB, the use of fungi as starter cultures has not been extensively studied. The majority of researchers have added fungi to improve the microbial stability of malt, and the direct enhancement of malt modification by such cultures is still a relatively unexplored field [12]. Precautions should be taken when choosing a potential candidate, as various fungi are known to produce toxigenic substances. In addition, the spores of certain fungi might be allergens or cause lung disease in workers of malt houses, especially immune-compromised individuals [9]. Boivin & Malanda [13], demonstrated how *Geotrichum candidum* could effectively restrict undesirable fungi and mycotoxins production when added during the malting process. The antimicrobial activity of the *Saccharomyces* spp. could be attributed mainly to competition with the other microorganism and also confirm that the antimicrobial effect of the yeast *Geotrichum candidum* when added during steeping of barley grain was due to competition with other microorganisms [13]. Laitila et al. [14] added *Wickerhamomyces anomalus* (synonym *Pichia anomala*) with antagonistic activity against moulds, to the malting process.

The main aim of this research is to isolate and screen lactic acid bacteria (lab) and yeast from fermented sorghum and maize products for use as a starter.

2. METHODOLOGY

2.1 Sample Collection

Samples were collected from Mokola, Bodija, and Ojoo in Ibadan, Oyo State, where spontaneously fermented cereal products are produced and sold. Already produced spontaneously fermented Ogi-baba was purchased transported in ice packs to the laboratory.

2.2 Isolation of Lactic Acid Bacteria

Lactic Acid Bacteria were isolated from ogi-baba samples using the pour plate technique as described by Vennel et al. [15]. Serial dilutions up to 10^{-9} were prepared in test tubes [16]. Each dilution was made using peptone water, prepared by dissolving one gram of peptone reagent into 100mL of distilled water which were then sterilized. One millilitre of each sample was then taken using sterile 1.0mL pipettes and homogenized in 9mL of peptone water. 1ml aliquots from 10^{-4}, 10^{-6}, 10^{-8} dilutions from the different samples were plated out by mixing with 20mL of molten PCA and also in MRS media in sterile Petri dishes. Each serial dilution was made in duplicate. The plates were swirled gently to enhance an even distribution of the inoculums throughout the medium and left to solidify.

2.3 Characterization of the Isolates

Identification was on the basis of the presence and characteristics of microscopic and macroscopic examination, physiological, morphological and biochemical tests.

2.4 Biochemical Identification Tests

The biochemical tests on lactic acid bacteria (LAB) isolates were conducted according to standard protocols [16-24].

2.4.1 Characteristic properties of the LAB isolates and assay for enzyme activity

The isolates were screened for enzyme production (amylase, lipase, and protease) according to Hugh & Leifson [21].

2.4.2 Antibacterial activities of LAB isolates

The antimicrobial activity was assayed using the agar well diffusion method described by Schillinger & Lucke [25]. The indicator strains used includes *Escherichia coli, Klebsiella* sp*, Salmonella* sp. and *Campylobacter* sp.*,* were collected from Food Microbiology Department, University of Ibadan, Ibadan, Oyo State. The test bacteria were incubated in nutrient broth at 37°C for 24 hrs, approximately 10^5 – 10^7 cfu/mL of the bacteria to be tested for sensitivity (indicator bacteria) were inoculated (1%) into 20 mL of nutrient agar and poured into petri dishes.

For the growth of LAB strain, MRS broth was used. Ten millilitres (10mL) of broth was inoculated with each species of LAB and were incubated at 35°C for 48hrs. After incubation, a cell-free solution was obtained by centrifuging z\the culture, followed by filtration of the supernatant through a filter. Wells of 4.0 mm in diameter were bored in the nutrient agar plate by using the broad end of Pasteur's pipette. Then 10 mL of cell free supernatants was filled in 4mm diameter sealed wells cut in the nutrient agar. Once solidified, the dishes were stored for 2 hrs in a refrigerator. The inoculated plates were incubated for 24hrs at 37°C, and the diameter of the inhibition zone was measured with callipers in millimetres [26]. LAB strains with inhibition zones of 0.5 – 4mm, 5-9mm, and 10-15mm were considered as weak inhibition, strong inhibition and very strong inhibition respectively.

2.4.3 Quantitative determination of antimicrobial compound produced

The antimicrobial compound tested for were lactic acid, acetic acid and hydrogen peroxide production which were done according to Oyewole [27].

2.4.4 Characterization of yeast isolates

2.5 Biochemical Tests

The biochemical tests on yeast were conducted according to [24, 28].

3. RESULTS AND DISCUSSION

The biochemical properties of the Lactic acid bacteria (LAB) and yeast isolates from Ogi-baba are shown in Tables 1 and 2 respectively. Most of the LAB isolates were identified as *Lactobacillus plantarum* while the rest were *Lactobacillus fermentum, Pediococcus acidilactici, Lactobacillus brevis, and Leuconostoc mesenteroides.* Majority of the yeast isolates were identified as *Saccharomyces cerevisiae* while the rest were *Candida tropicalis and Kluyveromyces marxianus.* Their sugar fermentation patterns of the LAB isolates are presented in Tables 3. Table 4 and 5 shows the Antagonistic activity (mm) of LAB and yeast respectively on some selected spoilage microorganisms.

Fig. 1 shows the percentage occurrence of LAB isolates with *Lactoacillus plantarum* making up to 40% of the isolates, *Lactobacillus fermentum* was 27% of total isolates, *Leuconostoc mesenteroides* was 10%, Lactobacillus brevis

was 13% and *Pediococcus acidilactici* was 10% of the total LAB isolates. Fig 2 shows the percentage occurrence of Yeast isolates with *Saccharomyces cerevisiae* at 57%, *Candida tropicalis* at 29% and *Kluyveromyces marxianus* at 14% of total yeast isolates.

The Lactic Acid Bacteria isolated from the Ogi-baba samples collected were *Lactobacillus plantarum*, *Lactobacillus fermentum, Pediococcus acidilactici, Lactobacillus brevis,* and *Leuconostoc mesenteroides*. The yeast isolated were *Saccharomyces cerevisiae, Candida tropicalis* and *Kluyveromyces marxianus. Lactobacillus plantarum* and *Saccharomyces cerevisiae* were the most predominant microorganisms isolated in the Ogi-baba, this is in agreement with the report of Kayodé, Odunfa & Adeyele, Lyumugabe et al. [26, 29, 30].

All the Lactic acid bacteria isolates produced varying zones of inhibition against spoilage bacteria and moulds. Most of them produced large zones of inhibition against *Salmonella* and *Klebsiella.* The LAB strains also produced varying zones of inhibition against the mycotoxigenic spoilage moulds. *Lactobacillus fermentum* CLB11 and CLB9 produced the highest zone of inhibition (26mm) on *Salmonella* spp and *Campylobacter* spp respectively after 48 hrs of incubation. *Saccharomyces cerevisiae CYT1* produces varying zone of inhibition ranging from 5 – 14 mm on the selected spoilage microorganisms except *Klebsiella* spp which was recorded zero zone. *Salmonella, S.aureus and A.flavus* also recorded zero zone to some of the isolates, Deak & Beuchat [28] reported similar results.Various mechanisms have been suggested to be responsible for the inhibitory effects of the bacteria on fungal growth, such as nutritional competition, secondary metabolites, pH or combinations of these mechanisms [31].

The inhibitory effects of fermentative organisms, particularly LAB, on spoilage and food poisoning organisms are well documented [28]. The various factors contributing to the antimicrobial activity of LAB are low pH due to the production of organic acids, hydrogen peroxide, ethanol, diacetyl, depletion of nutrients and microbial competition [32-34], [13] and Vanne et al. [15] showed that the growth of toxigenic storage fungi could be restricted by LAB *in vitro*. Manga & Oyeleke [17] reported a *Lb. plantarum* that was able to inhibit the growth of *A. flavus* but felt the effect was due to a combination of acidity and microbial competition. LAB are generally known to produce lactic acid during fermentation. They are even classified as heterofermentative or homofermentative based on their lactic acid production pattern. This explains the drop in pH in the steep treatments. The treatment with LAB starter had a faster drop in pH than those without LAB as a starter.

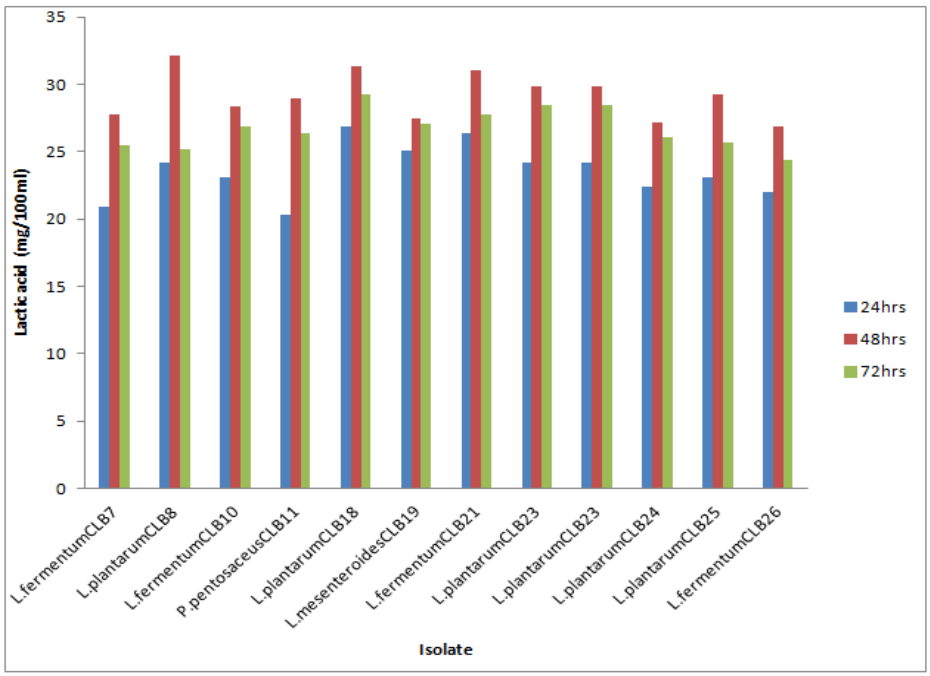

Fig. 1. Production of lactic acid by LAB isolated from spontaneously fermented Ogi-baba

Table 1. Characteristics of the Yeast strains isolated from spontaneously fermented Ogi-baba

Isolate code	Cell Morphology	Spore Test	Catalase test	Oxidase test	Arginine Hydrolysis	Gelatin liquefaction	Urease	Starch Hydrolysis	Growth at 25oC	Growth at 45oC	Growth at pH 3.9	Growth at pH 8.5	Growth at 4.0% NaCl	Growth at 8.0% NaCl	Glucose	Galactose	Maltose	Sucrose	Lactose	Melibiose	Sorbitol	Mannitol	Raffinose	Xylose	Fructose	Probable Identity
CLY 1	Round,flat, smooth and cream coloured	+	+	-	-	-	-	-	+	-	+	-	+	+	+	+	+	+	-	-	-	+	+	-	+	*Saccharomyces cerevisiae*
CLY 2	Oval,flat and whitish	+	+	-	-	-	-	-	+	-	+	+	+	+	+	+	-	+	+	_	+	+	-	-	+	*Candida tropicalis*
CLY 3	Round, raised, white coloured, in pairs	+	+	-	-	-	-	-	+	-	+	-	+	+	+	+	-	+	-	_	-	-	+	-	+	*Kluyveromyces marxianus;*
CLY 4	Round,flat, smooth and creamy	+	+	-	-	-	-	-	+	-	+	-	+	+	+	+	+	+	-	-	+	+	+	-	+	*Saccharomyces cerevisiae*
CLY 5	Round,smooth and cream coloured	+	+	-	-	-	-	-	+	-	+	-	+	+	+	+	+	+	-	-	-	+	+	-	+	*Saccharomyces cerevisiae*
CLY 6	Oval,flat and whitish	+	+	-	-	-	-	-	+	-	+	+	+	+	+	+	-	+	+	_	+	+	-	-	+	*Candida tropicalis*
CLY 7	Round,smooth and cream coloured	+	+	-	-	-	-	-	+	-	+	-	+	+	+	+	+	+	-	-	-	+	+	-	+	*Saccharomyces cerevisiae*

Key: + = Positive reaction, - = Negative reaction

Table 2. Biochemical and Physiological Tests for LAB Isolates from Spontaneously Fermented Ogi-Baba

Isolate code	Gram stain	Spore Test	Motility	Morphology	Catalase test	Oxidase test	Arginine Hydrolysis	Gas Production from MRS Broth	MR Test	VP Test	Nitrate Reduction	H2S production	Starch Hydrolysis	Growth at 10°C	Growth at 45°C	Growth at pH 3.9	Growth at pH 8.5	Growth at pH 9.6	Growth at 4.5% NaCl	Growth at 6.5% NaCl	Growth at 8% NaCl
CLB1	+	-	-	Rod	-	-	+	+	+	-	-	-	-	+	+	+	-	-	+	+	-
CLB2	+	-	-	Rod	-	-	-	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB3	+	-	-	Rod	-	-	-	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB4	+	-	-	Rod	-	-	+	+	+	-	-	-	-	+	-	+	+	+	+	-	-
CLB5	+	-	-	Rod	-	-	-	+	+	-	-	+	+	-	+	+	+	-	+	+	-
CLB6	+	-	-	Cocci	-	-	+	-	+	-	-	-	-	+	+	+	-	-	+	+	-
CLB7	+	-	-	Rod	-	-	-	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB8	+	-	-	Rod	-	-	-	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB9	+	-	-	Rod	-	-	+	+	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB10	+	-	-	Rod	-	-	+	+	+	-	-	-	-	+	-	+	+	+	+	-	-
CLB11	+	-	-	Rod	-	-	+	+	+	-	-	-	-	+	+	+	-	-	+	+	-
CLB12	+	-	-	Rod	-	-	+	+	+	-	-	-	-	+	+	+	-	-	+	+	-
CLB13	+	-	-	Rod	-	-	+	+	+	-	-	-	-	+	+	+	-	-	+	+	-
CLB14	+	-	-	Rod	-	-	-	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB15	+	-	-	Rod	-	-	-	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB16	+	-	-	Rod	-	-	-	+	+	-	-	-	-	-	+	+	+	-	+	+	
CLB17	+	-	-	Rod	-	-	+	+	+	-	-	-	-	+	-	+	+	+	+	-	-
CLB18	+	-	-	Rod	-	-	-	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB19	+	-	-	Rod	-	-	-	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB20	+	-	-	Rod	-	-	+	+	+	-	-	-	-	+	+	+	-	-	+	+	-
CLB21	+	-	-	Rod	-	-	-	+	+	-	-	-	-	-	+	+	-	-	+	+	
CLB22	+	-	-	Rod	-	-	+	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB23	+	-	-	Cocci	-	-	+	-	+	-	-	-	-	+	+	+	-	-	+	+	-
CLB24	+	-	-	Cocci	-	-	+	-	+	-	-	-	-	+	+	+	-	-	+	+	-
CLB25	+	-	-	Rod	-	-	+	+	+	-	-	-	-	+	+	+	-	-	+	+	-
CLB26	+	-	-	Rod	-	-	-	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB27	+	-	-	Rod	-	-	-	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB28	+	-	-	Rod	-	-	-	-	+	-	-	+	+	+	+	+	-	-	+	+	-
CLB29	+	-	-	Rod	-	-	+	+	+	-	-	-	-	+	-	+	+	+	+	-	-
CLB30	+	-	-	Rod	-	-	+	+			-	-	-	+	+	+	-	-	+	+	-

Key: + = Positive reaction, - = Negative reaction

Table 3. Carbohydrate fermentation pattern of LAB isolates from spontaneously fermented Ogi-Baba

Isolate code	Xylose	Raffinose	Sorbitol	Galactose	Maltose	Mannitol	Arabinose	Ribose	Lactose	Sucrose	Inositol	Glucose	Fructose	Trehalose	Cellobiose	Probable Identity
CLB1	+	+	+	+	+	-	-	-	+	+	-	+	+	+	-	*Lactobacillus fermentum*
CLB2	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB3	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB4	-	-	-	+	+	-	+	-	+	+	-	+	+	-	-	*Lactobacillus brevis*
CLB5	-	+	-	-	+	-	-	+	+	+	+	+	+	-	+	*Leuconostoc mesenteroides*
CLB6	+	-	-	+	-	-	+	+	+	+		+	+	-	+	*Pediococcus acidilactici*
CLB7	+	+	+	+	+	-	-	-	+	+	-	+	+	+	-	*Lactobacillus fermentum*
CLB8	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB9	+	+	+	+	+	-	-	-	+	+	-	+	+	+	-	*Lactobacillus fermentum*
CLB10	-	-	-	+	+	-	+	-	+	+	-	+	+	-	-	*Lactobacillus brevis*
CLB11	+	+	+	+	+	-	-	-	+	+	-	+	+	+	-	*Lactobacillus fermentum*
CLB12	+	+	+	+	+	-	-	-	+	+	-	+	+	+	-	*Lactobacillus fermentum*
CLB13	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB14	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB15	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB16	-	+	-	-	+	-	-	+	+	+	+	+	+	-	+	*Leuconostoc mesenteroides*
CLB17	-	-	-	+	+	-	+	-	+	+	-	+	+	-	-	*Lactobacillus brevis*
CLB18	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB19	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB20	+	+	+	+	+	-	-	-	+	+	-	+	+	+	-	*Lactobacillus fermentum*
CLB21	-	+	-	-	+	-	-	+	+	+	+	+	+		+	*Leuconostoc mesenteroides*
CLB22	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB23	+	-	-	+	-	-	+	+	+	+		+	+	-	+	*Pediococcus acidilactici*
CLB24	+	-	-	+	-	-	+	+	+	+		+	+	-	+	*Pediococcus acidilactici*
CLB25	+	+	+	+	+	-	-	-	+	+	-	+	+	+	-	*Lactobacillus fermentum*
CLB26	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB27	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB28	-	+	+	+	+	-	+	+	+	+	-	+	+	+	+	*Lactobacillus plantarum*
CLB29	-	-	-	+	+	-	+	-	+	+	-	+	+	-	-	*Lactobacillus brevis*
CLB30	+	+	+	+	+	-	-	-	+	+	-	+	+	+	-	*Lactobacillus fermentum*

Key: + = Positive reaction, - = Negative reaction

Table 4. Antagonistic activity (mm) of LAB isolated from spontaneously fermented ogi-baba against Food spoilage organisms using 24 and 48 hours' metabolite

Isolate Code	*E.coli*		*Klebsiella*		*Salmonella*		*Campylobacter*		*Probable Identity*
	24hr	48hr	24hr	48hr	24hr	48hr	24hr	48hr	
CLB1	14	20	13	20	10	21	15	17	*Lactobacillus fermentum*
CLB2	12	15	14	18	7	20	15	16	*Lactobacillus plantarum*
CLB3	14	13	16	18	12	16	14	16	*Lactobacillus plantarum*
CLB4	10	15	14	15	12	14	12	10	*Lactobacillus brevis*
CLB5	12	18	17	16	14	20	16	15	*Leuconostoc mesenteroides*
CLB6	12	20	16	20	19	23	15	15	*Pediococcus acidilactici*
CLB7	16	22	17	20	14	25	16	20	*Lactobacillus fermentum*
CLB8	18	23	14	23	19	20	15	20	*Lactobacillus plantarum*
CLB9	15	18	15	20	16	22	14	26	*Lactobacillus fermentum*
CLB10	10	18	12	19	22	20	10	24	*Lactobacillus brevis*
CLB11	18	23	16	22	10	26	17	19	*Lactobacillus fermentum*
CLB12	12	22	15	20	16	23	17	25	*Lactobacillus fermentum*
CLB13	12	21	16	25	20	25	12	21	*Lactobacillus plantarum*
CLB14	18	20	14	18	14	24	10	20	*Lactobacillus plantarum*
CLB15	20	21	16	20	14	21	9	21	*Lactobacillus plantarum*
CLB16	15	20	12	21	12	25	10	18	*Leuconostoc mesenteroides*
CLB17	16	21	15	19	14	25	12	20	*Lactobacillus brevis*
CLB18	12	21	14	19	15	21	12	16	*Lactobacillus plantarum*
CLB19	16	20	15	18	15	25	14	18	*Lactobacillus plantarum*
CLB20	14	16	12	14	12	20	12	20	*Lactobacillus fermentum*
CLB21	10	22	14	18	12	20	13	10	*Leuconostoc mesenteroides*
CLB22	14	20	13	18	12	21	11	15	*Lactobacillus plantarum*
CLB23	12	25	12	23	15	19	11	17	*Pediococcus acidilactici*
CLB24	15	18	15	22	13	19	13	16	*Pediococcus acidilactici*
CLB25	10	20	13	21	12	22	12	16	*Lactobacillus fermentum*
CLB26	10	23	14	20	10	21	13	19	*Lactobacillus plantarum*
CLB27	15	21	11	21	12	17	12	17	*Lactobacillus plantarum*
CLB28	14	19	13	20	9	18	12	17	*Lactobacillus plantarum*
CLB29	13	18	16	22	13	20	12	19	*Lactobacillus brevis*
CLB30	10	24	14	21	9	21	11	18	*Lactobacillus fermentum*

Table 5. Antagonistic activity of Yeast strains isolated from spontaneously fermented ogi-baba as by zones of inhibition (mm)

Isolate code	*A.flavus*	*A.niger*	*A.fumigatus*	*E.coli*	*Salmonella*	*S.aureus*	*Klebsiella*
Saccharomyces cerevisiaeCYT1	11	12	5	14	10	8	-
Candida tropicalisCYT2	-	9	8	13	8	6	7
Candida tropicalisCYT3	6	7	-	14	6	-	8
Saccharomyces cerevisiaeCYT4	7	8	6	10	8	5	7
Kluyveromyces marxianusCYT5	-	6	5	9	-	-	5
Candida tropicalisCYT6	5	7	-	12	9	8	8

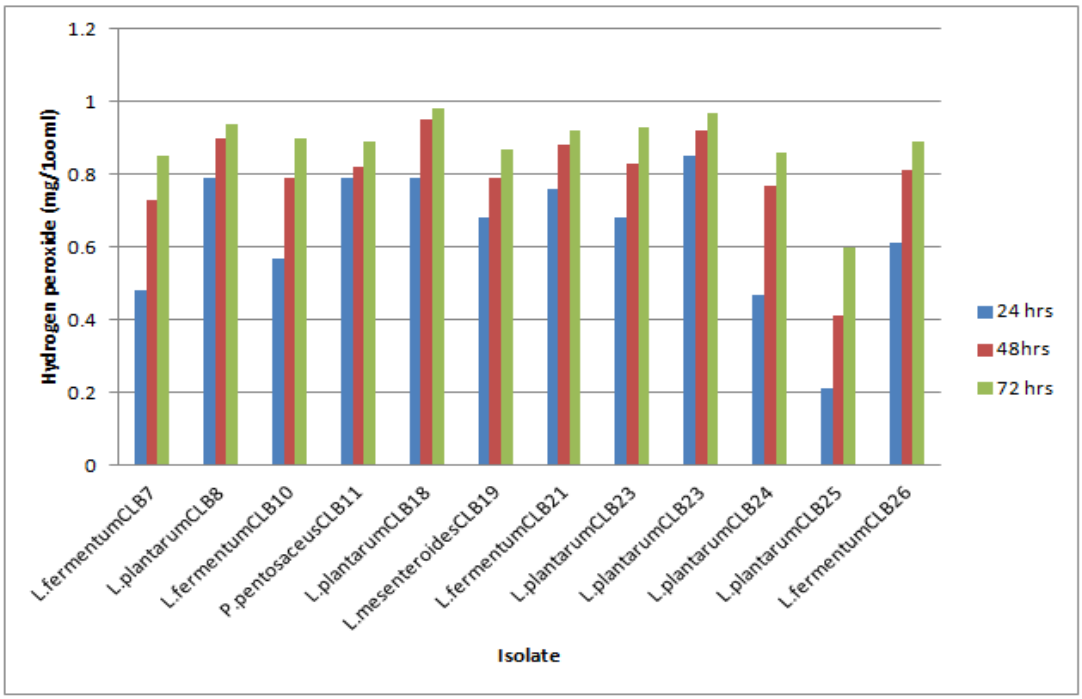

Fig. 2. Hydrogen Peroxide production by LAB isolated from spontaneously fermented Ogi-baba

4. CONCLUSION

In conclusion, this study focused on the isolation and screening of lactic acid bacteria and yeast for use as starter culture for improvements in the production of sorghum malt in order to eradicate the problem of excessive microbial contamination and the presence of mycotoxins. According to the results obtained, both lactic acid bacteria and yeast proved to be a good source of starter for this purpose.

5. RECOMMENDATION

Based on the results of this research, it is hereby recommended that biological control of moulds and enteric bacteria using cultures of Lactic acid bacteria and yeast as starter culture should be adopted during the steeping stage of malting.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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