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## Enriched cultures of lactic acid bacteria from selected Zimbabwean fermented food and medicinal products with potential as therapy or prophylaxis against yeast infections

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## ABSTRACT

**Objective:** To investigate the antifungal activity of crude cultures of putative strains of lactic acid bacteria (LAB) from a selection of Zimbabwean traditional and commercial food/medicinal products against yeasts (strains of environmental isolates of *Candida albicans* and *Rhodotorula* spp.).

**Methods:** Cultures of putative LAB from our selection of fermented products were enriched in de Man, Rogosa and Sharpe and isolated on de Man, Rogosa and Sharpe agar.

**Results:** The crude microbial cultures from the products that showed high antifungal activities (zone of inhibition, mm) were as follows: supernatant-free microbial pellet (SFMP) from an extract of *Melia azedarach* leaves [(27.0 ± 2.5) mm] > cell-free culture supernatants (CFCS) from Maaz Dairy sour milk and Mnandi sour milk [approximately (26.0 ± 1.8/2.5) mm] > CFCS and SFMP from Amansi hodzeko [(25.0 ± 1.5) mm] > CFCS from *Parinari curatellifolia* fruit [(24.0 ± 1.5) mm], SFMP from *Parinari curatellifolia* fruit [(24.0 ± 1.4) mm] and SFMP from mahewu [(20.0 ± 1.5) mm]. These cultures also showed high tolerance to acidic conditions (pH 4.0 and pH 5.0). However, culture from WAYA LGG (shown elsewhere to harbour antimicrobial activities) showed no antifungal activity. The LAB could have inhibited yeasts by either competitive exclusion or the release of antimicrobial metabolites.

**Conclusions:** Our cultures of LAB from a selection of Zimbabwean fermented products, especially *Ziziphus mauritiana* and fermented milk products have great potential for use as antifungal probiotics against yeast infections. Studies are ongoing to determine the exact mechanisms that are employed by the putative LAB to inhibit *Candida albicans*.

## 1. Introduction

With the ongoing attrition of cultures in Southern Africa in favour of western lifestyles, fermented products have remained central to diets in the region. Traditional fermented products are widely consumed in Zimbabwe, especially by the rural folks. Examples of traditional fermented products that are consumed in Zimbabwe include fermented maize porridges [mutwiwa

(Shona)/ilambazi lokubilisa (Ndebele)], fermented milk products [sour milk (English)/mukaka wakakora/hodzeko (Shona)/amasi (Ndebele)], non-alcoholic cereal-based beverages (mahewu, tobwa and mangisi), alcoholic beverages from sorghum or millet malt [chi 'seven days' (colloquial)/mupeta/doro remasese/chikokiyana/chi 'one day' (colloquial) (Shona)/utshwala (Ndebele); masawu beer (Shona)], distilled spirits (kachasu), fermented fruit mashes (makumbi)[1], a cake made from fermented fruits of *Parinari curatellifolia* (*P. curatellifolia*) [chambwa (Shona)][2]. A number of commercial/modern fermented products are also available in the country, especially sour milk brands including Lacto sour milk (Dairibord, Zimbabwe), Probrand sour milk (Probrand, Zimbabwe), Vuka vuka cheese (Kefalos, Zimbabwe), Kefalos Yoghurts (Kefalos, Zimbabwe), Lancewood sour cream (Lancewood, George, South Africa), Amasi hodzeko (CBS Foods, Zimbabwe), Limpopo sour

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milk, Mnandi sour milk (Zimbabwe), and alcoholic beverages, particularly traditional sorghum beer and Chibuku sorghum opaque beer (Delta Zimbabwe Limited).

Probiotic microorganisms have been shown to have prophylactic and therapeutic effects against gastrointestinal diseases, particularly diarrhoea. With a high HIV prevalence rate (15.2% in the 15–49 years age group; 2015 data)[3], erratic outbreaks of diarrhoeal diseases[4,5] and high prevalence of sexually transmitted infections[6] that collectively co-occur with poverty[7], the use of complementary diets and medicines has remained central to maintaining wellness and treatment of diarrhoea among many residents of Zimbabwe.

*Candida* species, *Rhodotorula mucilaginosa* and *Cryptococcus neoformans* are among the most frequent opportunistic yeast infections among HIV infected individuals[8]. Oral candidiasis has been shown to be one of the most common among all lesions among HIV-infected women (25%) who visited a health clinic in Zimbabwe in 2008[9]. Members of the genus *Rhodotorula*, frequently isolated in human environments, foods and beverages[10,11], have recently emerged as pathogens in HIV infected persons that present with meningitis[12,13]. *Rhodotorula* is one of the infrequent infections of the oral cavity, and can cause meningitis, endocarditis, ventriculitis, peritonitis, fungaemia, central venous catheter infection and keratitis[12]. While *Rhodotorula* is an uncommon opportunistic infection, it has emerged as a major cause of meningitis in HIV infected individuals[12]. With the high prevalence of HIV and the associated opportunistic infections that are frequently resistant to antibiotics, the discovery of novel complementary control measures is long overdue.

Despite the widespread use of fermented products in the country, the relative contributions of the complement of such foods to human health have remained unappraised. We sought to determine the antimicrobial activities of putative lactic acid bacteria (LAB) that are present in traditional and commercial fermented products, and a selection of environments against a strain of *Salmonella* spp., a strain of *Escherichia coli* and two cultures of yeasts [*Candida albicans* (*C. albicans*) and *Rhodotorula* spp.].

## 2. Materials and methods

### 2.1. Yeast isolates

Swab samples were collected from a female bathroom at Bindura University of Science Education. The samples were swabbed on potato dextrose agar (PDA), then the plates were incubated at 35 °C for 48 h. Isolation involved the streak plate procedure on PDA, simple staining with methylene blue and observation of peculiar oval shaped cells under a light microscope. The yeast isolates were identified using the conventional methods as described elsewhere[11,14]. Physiological and biochemical tests were used to characterise the putative yeast samples, including a gallery of fermentation of sugars, liquid assimilation of carbon compounds, assimilation of nitrogen compounds, growth at 25, 30, 37, 40, 42 and 45 °C, growth in vitamin free media, cycloheximide resistance, urease test and growth at high sugar concentrations. Following identification, isolates of *C. albicans* (Figure 1) and one of *Rhodotorula* spp. (Figure 2) were streaked on PDA slants and incubated under the same conditions as

above, following which they were stored under 4 °C until time of antimicrobial testing.



Figure 1. An isolate of *C. albicans* on PDA.

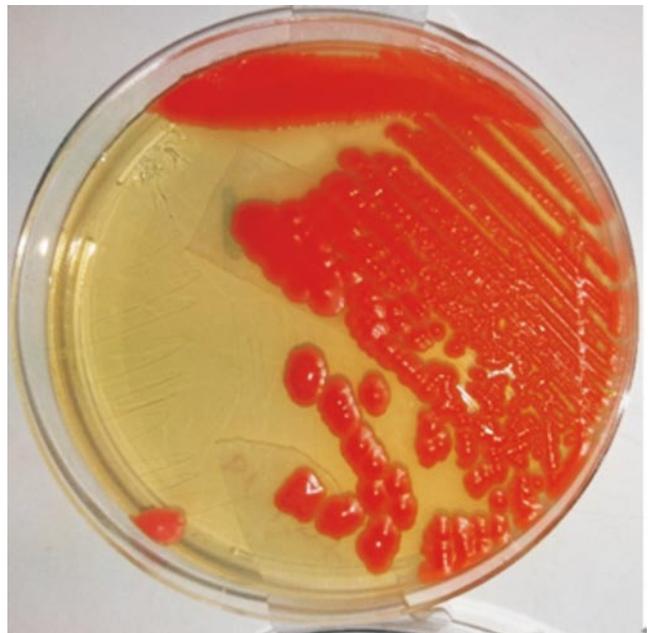


Figure 2. An isolate of *Rhodotorula* species on PDA.

### 2.2. Preparation of supernatant-free microbial pellets (SFMP) and cell-free culture supernatants (CFCS) of LAB

Our selection of fermented products, namely, commercial and traditionally fermented milk products, ‘chambwa’ cake, traditional and commercial sorghum beer, ‘masawu’ beer, rotten tomatoes and an extract from *Melia azedarach* (*M. azedarach*) plant (Table 1), were aseptically inoculated into de Man, Rogosa and Sharpe (MRS) broth medium (Oxoid, Basingstoke, United Kingdom). The MRS tubes were incubated in an anaerobic jar at 37 °C for 48 h. Each culture was neutralised to pH 7.0 by addition of 1 mol/L NaOH. The CFCS and SFMP were separated by centrifugation at 2000 r/min for 20 min. The pellet was washed twice with sterile ¼ Ringer’s solution

(Sigma-Aldrich, Gillingham, UK), re-suspended in 3 mL of Ringer's solution and stored at 4 °C until time of use. The CFCS were passed through 0.22 µm pore size filters and stored at 4 °C until time of use. A proportion of the CFCS in Ringer's solution was collected, and that of the SFMP was mixed with 10% sterile glycerol and both were stored at -20 °C for subsequent characterisation. As back-up, part of

the SFMP was streaked on MRS slants, incubated as described above (for LAB) and subsequently stored at 4 °C.

### 2.3. Isolation and characterisation of putative LAB

Crude culture of putative LAB was streaked on MRS agar (Oxoid,

**Table 1**

Description of products used as sources of potential antifungal LAB.

Abbreviation used	Local name of product	English/Latin name	Uses	Source	Known microbial composition/Description
MAH	Homemade mahewu	–	Traditional beverage	Mutoko, Zimbabwe; consumed around Zimbabwe	Naturally fermented beverage made from sorghum
SMH	Hodzeko sour milk	Sour milk	Traditional beverage/relish	Mutoko, Zimbabwe; consumed around Zimbabwe	Naturally fermented cows' milk
MAE	Fermenting <i>M. azedarach</i> extract	Chinaberry tree, pride of India, bead-tree, Cape lilac, syringa berry tree, Persian lilac, Indian lilac	Traditional medicine against sexually transmitted diseases	Goromonzi, Zimbabwe; used around Zimbabwe	An extract from <i>M. azedarach</i>
ZMB	Masawu beer	Jujube beer	Lightly alcoholic/non-alcoholic beverage	Mutoko, Zimbabwe; consumed around Zimbabwe	Alcoholic beverage made from fruits of <i>Z. mauritiana/Ziziphus jujuba</i> (L.) Lam. beer
FZMF	Masawu fruit	Jujube fruits	Traditional food/wild fruit	Mutoko, Zimbabwe; consumed around Zimbabwe	<i>Z. mauritiana/Ziziphus jujuba</i> (L.) Lam./Jujube fruits
PCFC	<i>P. curatellifolia</i> fruit cake	Chambwa (Shona), cake from Hissing tree fruit/Mobola plum cake (English)	Cake (food)	Mutoko, Zimbabwe; consumed in Mashonaland East Province (primarily Mutoko), Zimbabwe	A cake made from an extract from <i>P. curatellifolia</i> Planch. ex Benth. fruits, finger millet powder and water
DTOM	Decomposing tomatoes	Tomatoes	Food	Mutoko, Zimbabwe; frequently used around Zimbabwe	Rotten tomatoes ( <i>Solanum lycopersicum</i> )
KCC	Kefalos cheddar cheese	Commercial cheese	Cheese/food	Manufactured by Kefalos, Harare, Zimbabwe	A cheese made from cows' milk fermented using an unknown microbial culture
LSC	Lancewood sour cream	Sour cream	Sour cream/food	Manufactured by Lancewood, George, South Africa, sold in Zimbabwe	Cream made from cows' milk fermented using an unknown microbial culture
AHSM	Amasi hodzeko	Sour milk	Beverage/relish (food)	Manufactured by CBS Foods, Harare, Zimbabwe	Sour milk product made from cows' milk fermented using an unknown microbial culture
LLSM	Limpopo sour milk	Sour milk	Beverage/relish (food)		Sour milk product made from cows' milk fermented using an unknown microbial culture
MDSM	Mnandi sour milk	Sour milk	Beverage/relish (food)		Sour milk product made from cows' milk fermented using an unknown microbial culture
TOB	Traditional opaque beer	chi 'seven days' (colloquial)/mupeta/doro remasese/chikokiyana/chi 'one day' (Shona)/utshwala (Ndebele)	Alcoholic beverage (opaque beer)	Mutoko, Zimbabwe	A traditional opaque beer brewed primarily from sorghum and maize
PCF	<i>P. curatellifolia</i> fruit	Hissing tree, Mobola plum (English) Mubuni, Muchakata, Muhacha, Muisha (Shona), Umkhuna (Ndebele)	Food	Mutoko, Zimbabwe	Fruits consumed in Zimbabwe when ripe as fruits or used to make products such as 'chambwa' cake in Mutoko area of Zimbabwe
MDL	Maaz Dendairy Lacto sour milk	–	Beverage/relish (food)	Kwekwe, Zimbabwe	Sour milk product made from cows' milk fermented using an unknown microbial culture
KVC	Kefalos Vuka vuka cheese	–	Cheese/food	Harare, Zimbabwe	A cheese made from cows' milk fermented using an unknown microbial culture
CHOB	Chibuku opaque beer	'Super beer' in Zimbabwe; doro remasese/hwahwa (Shona); utshwala (Ndebele)	Alcoholic beverage (opaque beer)	Harare, Zimbabwe	A commercial opaque beer brewed primarily from sorghum and maize, manufactured by Delta Corporation, Harare, Zimbabwe
WAYA LGG	WAYA LGG	Antimicrobial probiotic product	A commercial probiotic anti-diarrheal product	Available from Medis d.o.o., Slovenia and internationally	An antimicrobial probiotic product available from Medis d.o.o., Slovenia, containing <i>L. rhamnosus</i> LGG, which is sold internationally with claims that it has antimicrobial activities against agents of gastroenteritis infections
PL	Prolife	–	A commercial probiotic anti-diarrhoeal product	Manufactured by Jadran-Galenski Laboratorij d.d., Rijeka, Croatia	A probiotic product containing $2.6 \times 10^8$ living bacterial cells/mL ( <i>Bacillus coagulans</i> , <i>Lactobacillus acidophilus</i> , <i>Streptococcus thermophilus</i> and <i>Lactobacillus bulgaricus</i> , <i>Bifidobacterium bifidum</i> )
PJ	Probio Junior	–	A commercial probiotic anti-diarrhoeal product	Manufactured by Fidimed, Trzin, Slovenia	Containing $1 \times 10^9$ CFU per bag, namely, <i>Lactobacillus casei</i> , <i>L. rhamnosus</i> , <i>Streptococcus thermophilus</i> , <i>Bifidobacterium breve</i> , <i>Lactobacillus acidophilus</i> , <i>Bifidobacterium infantis</i> and <i>Lactobacillus bulgaricus</i>
BG	BioGaia	–	A commercial probiotic anti-diarrhoeal product	Manufactured by BioGaia AB, Stockholm, Sweden	Containing $2 \times 10^9$ CFU/mL, sunflower oil, medium chain triglyceride oil and <i>Lactobacillus reuteri</i> DSM 17938 ( <i>Lactobacillus reuteri</i> Protectis)

*Z. mauritiana*: *Ziziphus mauritiana* Lam.; *L. rhamnosus*: *Lactobacillus rhamnosus*.

Basingstoke, United Kingdom), following which the plates were incubated in an anaerobic jar at 37 °C for 48 h. Isolated colonies that showed different characteristics were purified on MRS agar then incubated as described above. After 48 h incubation, colonies were stained following the classic Gram staining protocol. All isolates that were deemed typical of LAB were collected and inoculated on slants of MRS agar and in MRS broth. The slants and broth tubes were incubated under the same conditions as above, following which the former was stored at 4 °C and the latter at -20 °C in 10% sterile glycerol.

#### 2.4. Acid tolerance

The acid tolerance assay was tested in accordance with a protocol described by Lee *et al.*[15] with modifications. Briefly, each crude or LAB isolates (SFMP) were re-suspended in sterile Ringer's solution (in triplicate) and concentrations were adjusted to an optical density (OD) of 0.2 at 620 nm using a Biobase EL 10B Microplate Reader (Jinan, China) (equivalent to a concentration lying between 7 to 8 log CFU/mL). The acid tolerance test was studied in MRS with pH adjusted to 4, 5 and 7. Each broth containing tube was inoculated with 1 mL of each culture. 100 µL of each culture was inoculated in the MRS broth tubes incubated anaerobically at 37 °C for 18 h. After incubation, growth of the cultures was measured at 620 nm using Biobase EL 10B Microplate Reader (Jinan, China). Tolerance to acidic condition was estimated by comparing log 10 OD values after exposure to acidic (pH 4.0 and pH 5.0) and normal (pH 7/control) conditions. The following equation was used[15]:

$$\text{Acid tolerance} = [(\text{Log OD (620 nm after 18 h of exposure)}) - \text{Log OD (620 nm at 0 h of exposure)}] / \text{Log OD (620 nm at 0 h of exposure)} \times 100$$

#### 2.5. Antifungal activity

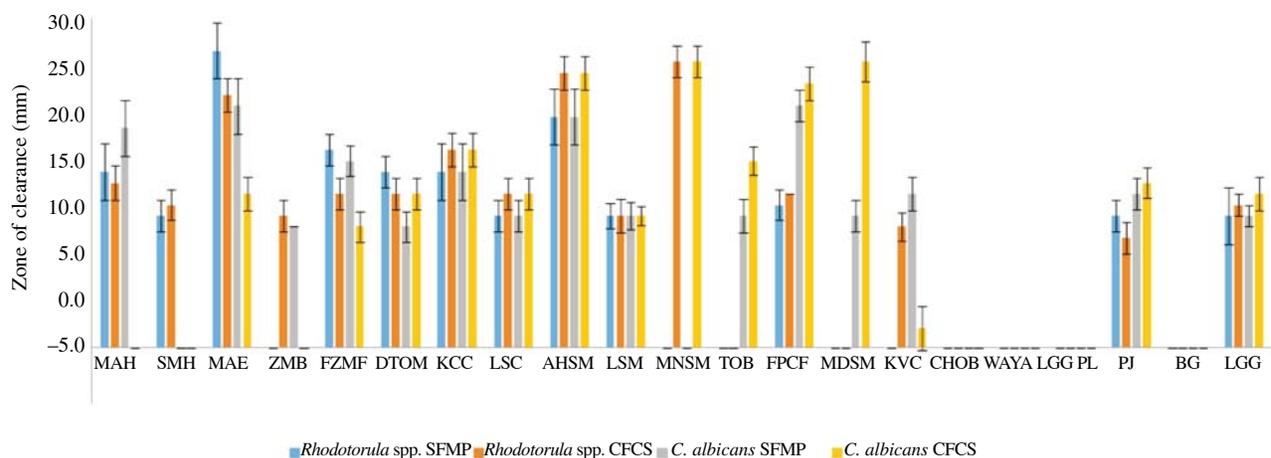
A strain of *Rhodotorula* spp. and *C. albicans* was inoculated into potato dextrose broth and incubated as described above. After 48 h, absorbance of the yeast cultures was adjusted to a McFarland

equivalent of 0.2 at 620 nm in a Biobase EL 10B Microplate Reader (Jinan, China). The adjusted cultures were swabbed on PDA. Wells were then drilled into the agars using an auger (9 mm). Then, 60 µL of CFCS or pellet cultures from the fermented products were introduced into triplicate wells. The plates were dried for 30 min at room temperature and then incubated aerobically at 37 °C for 48 h. Antimicrobial activities were evaluated in terms of the zones of clearance as follows: inhibition zones greater than 20 mm, 10 to 20 mm, and less than 10 mm were considered strong, intermediate, and low inhibitions, respectively.

### 3. Results

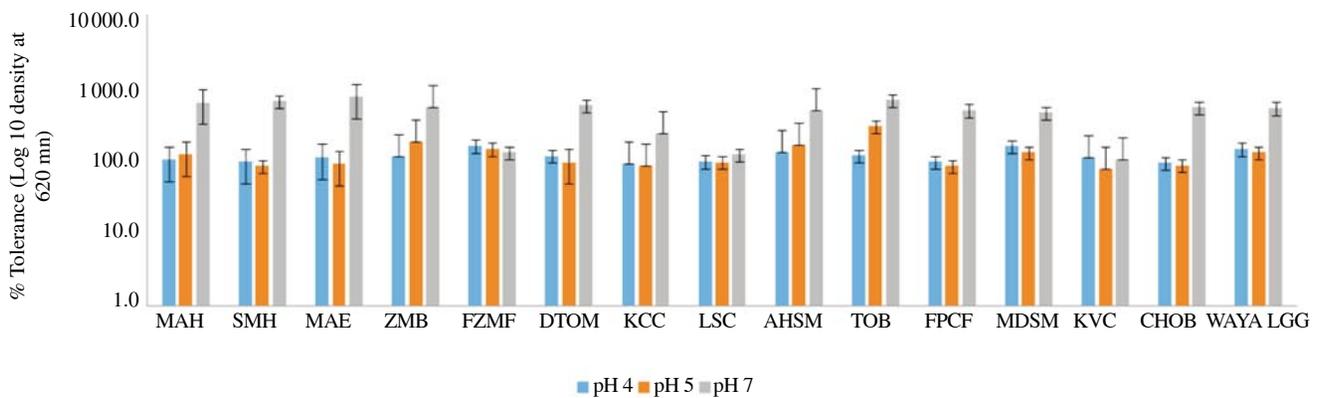
As shown in Figures 1 and 2, *C. albicans* and *Rhodotorula* species were successfully isolated on PDA which supported the growth of yeast. Putative LAB from a fermented extract of *M. azedarach*, Amansi hodzeko sour milk, Mnandi sour milk (MNSM), fermenting *P. curatelifolia* fruits (FPCF) and Maaz Dendairy sour milk showed 'strong inhibition' against a strain of *Rhodotorula* spp. and one of *C. albicans*. SFMP and CFCS from the same products above (AME, AHSM, MNSM, FPCF, MDSM) and Kefalos cheddar cheese had significantly greater inhibition than those from control antimicrobial products (WAYA LGG, Prolife, Probio Junior and BioGaia) and a pure culture of *L. rhamnosus* LGG strain ( $P < 0.05$ ). Washed LAB cells from mahewu (a lightly-alcoholic beverage), *M. azedarach*, *Z. mauritiana* beer, fermenting *Z. mauritiana* fruit, a cake made from fermented fruits of *P. curatelifolia* and Kefalos Vuka Vuka cheese showed significantly greater inhibitory activity against *C. albicans* than CFCS; and those from *M. azedarach*, fermenting *Z. mauritiana* fruit and *P. curatelifolia* fruit cake had significantly greater inhibitory activity than CFCS against *Rhodotorula* spp. ( $P < 0.05$ ) (Figure 3).

Cultures of LAB from fermented *Z. mauritiana*, Amansi hodzeko sour milk, and Maaz Dendairy sour milk showed similar acid tolerance (greater or insignificantly lower) to pH 4.0 as WAYA LGG, while fermented *Z. mauritiana*, Amansi hodzeko sour milk, Maaz Dendairy sour milk and *Z. mauritiana* (ZMD) showed similar acid tolerance to pH 5.0 as WAYA LGG (Figure 4).



**Figure 3.** Inhibition of putative isolates of *C. albicans* and *Rhodotorula* species by washed pellet cultures and CFCS of LAB from samples of fermented food/beverages enriched in MRS broth.

Inhibition was classified as follows: strong (mean clearance zone  $\geq 20$  mm), intermediate (mean clearance zone lying between 10 and 20 mm) and low (mean clearance zone lying between 10 and 20 mm).



**Figure 4.** Acid tolerance of crude cultures of LAB from samples of fermented food/beverages after exposure for 18 h in MRS broth. Tolerance to acid was considered satisfactory if the culture grew as much as (the same/insignificantly lower than) or better than a culture of WAYA LGG in MRS.

#### 4. Discussion

Antimicrobial activities of SFMP and CFCS from a selection of fermented products showed variable activities, with those from a fermented extract of *M. azedarach*, Amansi hodzeko sour milk, MNSM, FPCF and Maaz Dendairy sour milk which showed 'strong inhibition' against a strain of *Rhodotorula* spp. and one of *C. albicans*. Additionally, SFMP and CFCS from AME, AHSM, MNSM, FPCF, MDSM and Kefalos cheddar cheese exhibited significantly greater inhibition than those from control antimicrobial products (WAYA LGG, Prolife, Probio Junior and BioGaia) and a pure culture of *L. rhamnosus* LGG strain ( $P < 0.05$ ). Washed LAB cells from mahewu (a lightly-alcoholic beverage), *M. azedarach*, *Z. mauritiana* beer, fermenting *Z. mauritiana* fruit, a cake made from fermented fruits of *P. curatelifolia* and Kefalos Vuka vuka cheese showed significantly greater inhibitory activity against *C. albicans* than CFCS; and those from *M. azedarach*, fermenting *Z. mauritiana* fruit and *P. curatelifolia* fruit cake had significantly greater inhibitory activity than CFCS against *Rhodotorula* spp. ( $P < 0.05$ ) (Figure 3). Antimicrobial activity of LAB isolated from fermented foods has also been reported in studies on LAB strains isolated from fermented foods elsewhere, particularly tarhana[16], katak[17] and kefir[18]. Katak is a Bulgarian dairy product with strong inhibition activity against fungal growth, particularly mycelial growth[17]. Kefir, a fermented milk product with origins in Northern Ossetia within the Caucasus Mountains region, is known to harbour strong antimicrobial activities similar to those of ampicillin, azithromycin, ceftriaxone, amoxicillin, and ketoconazole[18].

To the best of our knowledge, the current report is first to show antimicrobial activities of fermented products in Zimbabwe, let alone the direct inhibition of yeasts by non-replicating cells of LAB *in vitro*.

CFCS of cultures from Amasi hodzeko sour milk, MNSM, Maaz Dendairy sour milk and a control isolate of *L. rhamnosus* GG (ATCC 53103) showed significantly greater inhibitory activity than washed cells against the two yeasts ( $P < 0.05$ ), implying the LAB that were cultured from these products released metabolites with high antimicrobial activity against the yeasts. Cell free supernatants of cultures from the fermented dairy products above are thought to

have exerted their antimicrobial activities through the modulation of pH and activities of microbial metabolites such as bacteriocins that accumulate during fermentation[19].

Based on this preliminary finding, we hypothesise that the LAB in our selection of fermented products may exert their antimicrobial activities via (i) direct inhibition of yeasts (antagonism), perhaps through competitive exclusion, (ii) release of antimicrobial metabolites, or (iii) quorum sensing based exclusion. A recent study has shown that *Lactobacillus* species possess a strain specific spectrum and mechanism of inhibitory activities against different moulds based on various factors such as release of organic acid such as lactic acid, hydrogen peroxide production and protein compounds[16]; it is however important to isolate specific strains from samples to determine their mechanism of inhibition *in vitro*. The inhibition of fungi by LAB from different environments including fermented milk products has been shown elsewhere[16,17,20,21].

Our results also showed similar acid tolerance of LAB cultures obtained from fermented *Z. mauritiana*, Amansi hodzeko sour milk and Maaz Dendairy sour milk to pH 4.0 compared to that of WAYA LGG. Further, LAB cultures from fermented *Z. mauritiana*, Amansi hodzeko sour milk, Maaz Dendairy sour milk and *Z. mauritiana* beer showed similar acid tolerance to pH 5.0 compared to that of WAYA LGG. Acid tolerance in LAB strains isolated from fermented foods has been reported in other studies[22].

Based on our findings, we report high antimicrobial activities of a crude supernatant-free microbial culture obtained from a fermented extract of *M. azedarach* showing significant inhibition, compared to control antimicrobial cultures (WAYA LGG, PL, PJ and BG) against *Rhodotorula* spp. and *C. albicans* strains ( $P < 0.02$ ). Further, CFCS from *M. azedarach* (natural product), Amansi hodzeko sour milk (commercial Zimbabwean product) and Mnandi sour milk (commercial Zimbabwean product) had significant inhibitory effect against a strain of *Rhodotorula* spp. ( $P < 0.02$ ), while Amansi hodzeko sour milk, Mnandi sour milk and fermenting *P. curatelifolia* fruits (a natural product) showed significant effect against a strain of *C. albicans* ( $P < 0.04$ ). Similarly, crude cultures of LAB from fermented *Z. mauritiana*, Amansi hodzeko sour milk and Maaz Dendairy sour milk showed reasonably high tolerance

to pH 4, and those from these three cultures and one from *Z. mauritiana* beer had reasonably high tolerance to pH 5, which were comparable to that of WAYA LGG. Cultures of LAB that are present in a selection of fermented Zimbabwean products can be useful as therapy/prophylaxis against yeast infections that are frequent in HIV infected individuals in Zimbabwe and Southern Africa.

Our collection of crude LAB obtained from two Zimbabwean natural products, namely, a fermenting extract from *M. azedarach* and fermenting *P. curatelifolia* fruits, and two commercial products, namely, Amansi hodzeko sour milk and Mnandi sour milk could be useful in the management of yeast infections. The LAB containing fermented milk and plant products can be useful in functional foods that can help in the management of yeast infections that are frequent among immunocompromised individuals, especially people living with HIV, in Southern Africa. Our selection of LAB cultures has been shown to maintain viability under acidic conditions that are found in the human/animal stomach (average pH 4.0). Further studies are underway to establish the mechanisms by which consortia of LAB from our selection of Zimbabwean fermented products exert their antimicrobial activities. Studies on the population dynamics and viability under various conditions *in vitro* and *in vivo* are also underway.

### Conflict of interest statement

We declare that we have no conflict of interest.

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### References

- [1] Gadaga TH, Mutukumira AN, Narvhus JA, Feresu SB. A review of traditional fermented foods and beverages of Zimbabwe. *Int J Food Microbiol* 1999; **53**(1): 1-11.
- [2] Nyakupfuka A. *Global delicacies: diversity, exotic, strange, weird, relativism*. Bloomington: Balboa Press; 2013.
- [3] Chingwaru W, Vidmar J. Thirty years on with an HIV epidemic in Zimbabwe (1985–2015). *HIV AIDS Rev* 2016; **15**: 26-32.
- [4] Maponga BA, Chirundu D, Gombe NT, Tshimanga M, Shambira G, Takundwa L. Risk factors for contracting watery diarrhoea in Kadoma City, Zimbabwe, 2011: a case control study. *BMC Infect Dis* 2013; **13**: 567.
- [5] Polonsky JA, Martínez-Pino I, Nackers F, Chonzi P, Manangazira P, Van Herp M, et al. Descriptive epidemiology of typhoid fever during an epidemic in Harare, Zimbabwe, 2012. *PLoS One* 2014; **9**(12): e114702.
- [6] Gonese E, Mapako T, Dzangare J, Rusakaniko S, Kilmarx PH, Postma MJ, et al. Within-gender changes in HIV prevalence among adults between 2005/6 and 2010/11 in Zimbabwe. *PLoS One* 2015; **10**(7): e0129611.
- [7] Pascoe SJ, Langhaug LF, Mavhu W, Hargreaves J, Jaffar S, Hayes R, et al. Poverty, food insufficiency and HIV infection and sexual behaviour among young rural Zimbabwean women. *PLoS One* 2015; **10**(1): e0115290.
- [8] Kaur R, Dhakad MS, Goyal R, Bhalla P, Dewan R. Spectrum of opportunistic fungal infections in HIV/AIDS patients in tertiary care hospital in India. *Can J Infect Dis Med Microbiol* 2016; **2016**: 2373424.
- [9] Chidzonga MM, Mwale M, Malvin K, Martin JN, Greenspan JS, Shiboski CH. Oral candidiasis as a marker of HIV disease progression among Zimbabwean women. *J Acquir Immune Defic Syndr* 2008; **47**(5): 579-84.
- [10] Kot AM, Błazejak S, Kurcz A, Gientka I, Kieliszek M. *Rhodotorula glutinis* – potential source of lipids, carotenoids, and enzymes for use in industries. *Appl Microbiol Biotechnol* 2016; **100**(14): 6103-17.
- [11] Misihairabgwi JM, Kock L, Pretorius E, Pohl C, Zvauya R. Characterisation of yeasts isolated from traditional opaque beer beverages brewed in Zimbabwean households. *Afr J Microbiol Res* 2015; **9**(8): 549-56.
- [12] Deepa A, Nair BJ, Sivakumar T, Joseph AP. Uncommon opportunistic fungal infections of oral cavity: a review. *J Oral Maxillofac Pathol* 2014; **18**(2): 235-43.
- [13] Mohd Nor F, Tan LH, Na SL, Ng KP. Meningitis Caused by *Rhodotorula mucilaginosa* in HIV-infected patient: a case report and review of the literature. *Mycopathologia* 2015; **180**(1-2): 95-8.
- [14] Barnett JA, Payne RW, Yarrow D. *Yeasts: characteristics and identification*. Cambridge: Cambridge University Press; 2000.
- [15] Lee HK, Choi SH, Lee CR, Lee SH, Park MR, Kim Y, et al. Screening and characterization of lactic acid bacteria strains with anti-inflammatory activities through *in vitro* and *Caenorhabditis elegans* model testing. *Korean J Food Sci Anim Resour* 2015; **35**(1): 91-100.
- [16] Kivanc M, Kivanc SA, Pektas S. Screening of lactic acid bacteria for antifungal activity against fungi. *J Food Process Technol* 2014; **5**: 310.
- [17] Tropcheva R, Nikolova D, Evstatieva Y, Danova S. Antifungal activity and identification of lactobacilli, isolated from traditional dairy product “katak”. *Anaerobe* 2014; **28**: 78-84.
- [18] Bourrie BC, Willing BP, Cotter PD. The microbiota and health promoting characteristics of the fermented beverage kefir. *Front Microbiol* 2016; **7**: 647.
- [19] Parada JL, Caron CR, Medeiros AB, Soccol CR. Bacteriocins from lactic acid bacteria: purification, properties and use as biopreservatives. *Braz Arch Biol Technol* 2007; **50**(3): 512-42.
- [20] Mokoena MP, Mutanda T, Olaniran AO. Perspectives on the probiotic potential of lactic acid bacteria from African traditional fermented foods and beverages. *Food Nutr Res* 2016; **60**: 29630.
- [21] Sellamani M, Kalagatur NK, Siddaiah C, Mudili V, Krishna K, Natarajan G, et al. Antifungal and zearalenone inhibitory activity of *Pediococcus pentosaceus* isolated from dairy products on *Fusarium graminearum*. *Front Microbiol* 2016; **7**: 890.
- [22] Kabore D, Sawadogo-Lingani H, Dicko MH, Diawara B, Jakobsen M. Acid resistance, bile tolerance and antimicrobial properties of dominant lactic acid bacteria isolated from traditional ‘maari’ baobab seeds fermented condiment. *Afr J Biotechnol* 2012; **11**(5): 1197-206.