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Full Length Research Paper

Evaluation of ogi liquor from different grains for antibacterial activities against some common diarrhoeal bacteria in Southwest Nigeria

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The effect of raw ogi liquor from five varieties of grains; white maize, yellow maize, white guinea corn, red guinea corn and millet on some diarrhoeal bacteria; *Staphylococcus aureus, Shigella dysenteriae, Escherichia coli, Salmonella typhimurium* and *Enterobacter* species was investigated. The ogi liquors prepared using cold and hot water methods were effective in inhibiting the growth of most of the test organisms with zones of inhibition ranging from 4.0 - 14.0 mm. The growth inhibition mediated by these liquors, however, was not as wide as that of some of the antibiotics such as ciprofloxacin, gentamycin and ofloxacin, but in most cases superior to that of tetracycline, nitrofurantoin and cotrimoxazole.

Key words: Ogi liquor, antibacterial effects, diarrhoeal bacteria.

INTRODUCTION

Diarrhoea, which is an illness characterized by an increase in frequency and fluidity of stools is one of the most common diseases causing infant death in develop-ing countries (Walderman, 1998; Cheesbrough, 1994). Poor hygiene, consumption of contaminated foods and close proximity to animals all contribute to easy and frequent acquisition of pathogens that cause this condition which include *Escherichia coli*, *Salmonella* species, *Shigella* species, *Staphylococcus aureus*, *Clostridium difficile*, and *Campylobacter jejuni* among others (Prescott et al., 2005).

Diarrhoea, although self-limiting, may some times require antibiotic therapy. However, most of the aetiolo-gical agents especially bacteria have already developed resistance to most of the commonly employed antibiotics (Ashebir and Ashenafi, 1999). In addition, some of these antibiotics can also induce diarrhoea known as “antibiotic induced diarrhoea” (Cheesbrough, 1994; Marteau et al., 2002). Therefore it is necessary to find alternative means of treatment of this disease.

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Locally in some communities in southwestern Nigeria, uncooked ogi is normally administered to people having running stomach to reduce the frequency of stooling (Aderiye and Laleye, 2004). This prompted this investigation to determine whether uncooked ogi actually has antidiarrhoeic effect by specifically checking whether it has growth inhibitory activity on common bacteria that cause diarrhea and also to know whether the variety of grains used in preparing the ogi and the steeping method have any effect on the antibacterial activity.

MATERIALS AND METHODS

Bacteria

S. aureus, *Salmonella typhimurium*, *Shigella dysenteriae* and *E. coli* used in this study were collected from the Nigerian Institute of Medical Research (NIMR), Yaba, Lagos, Nigeria while *Enterobacter* species was collected from the State Specialist Hospital, Akure, Nigeria.

Grains

White maize (*Zea mays*), yellow maize (*Z. mays*, yellow variety), red guinea corn (*Sorghum vulgare*), white guinea corn (*Sorghum*

bicolor) and millet (*Pennisetum typhoideum*) purchased at a local market in Akure, Ondo State were used.

Preparation of ogi slurry

This was done according to the method of Odunfa and Adeleye (1985) with slight modifications. Two hundred grams of each variety of the cereal grains was weighed into different sterile small plastic pails with cover containing 300 ml sterile distilled water and steeped for 72 h at 28 ± 2^{o}C. After steeping, the water was decanted and the grains were wet-milled separately using properly washed grinding machine. The resulting pastes were sieved using different sterile muslin cloths, the filtrates collected into different sterile containers and allowed to settle for 3 days during which fermentation took place by the natural flora of the grains. For the hot water method, boiled water was used for steeping instead of cold water and the steeping duration was reduced to 24 h. The grains were also wet-milled, sieved and the filtrates were also allowed to settle for 72 h for fermentation to take place by the natural flora of the grains. The liquors of the fermented ogi slurries were then decanted, their pH was measured using Jenway pH meter and their titrable acidity was determined using AOAC method (1990) before they were tested for antibacterial activity against the test organisms.

Determination of antibacterial activity of the prepared ogi liquors

This was done using agar diffusion method of Onifade and Kolawole (1999). One milliliter of 18 h broth culture of each of the test organisms was taken using a sterile syringe and placed into sterile petridishes (different organism per plate). Each plate was then overlaid with 20 ml nutrient agar, carefully swirled to allow even distribution of the organisms within the agar and were allowed to gel before 6 wells (8 mm in diameter) were bored in the agar with the aid of a sterile cork borer. The ogi liquors from the grains were put into the wells; 0.1 ml per well and different type for different well, sterile distilled water was used as control. The plates were incubated at 37 C for 24 h. The diameter of the zones of inhibition around the wells containing the liquors was determined and recorded.

Antibiotic sensitivity pattern of the test organisms

This was done as above except that instead of making wells on the already seeded plates and pouring in the prepared liquors, standard commercial antibiotics disc was placed on the seeded agar plates before the plates were incubated at 37 C for 24 h.

Isolation and identification of microorganisms present in the liquors

The different ogi liquors prepared from the different grains were streaked onto different nutrient agar (NA) and potato dextrose agar (PDA) plates. The plates were incubated at 37 C (for bacterial growth on NA) and room temperature of 28 ± 2^{o}C (fungal growth on PDA) for 24 h and 72 h respectively. Isolates were identified according to Holt et al. (1994).

RESULTS

The ogi liquors used in this study, irrespective of mode of steeping of the grains, had antibacterial activities against most of the test bacteria except *Enterobacter* sp. whose growth was not inhibited by the ogi liquor obtained from the grains steeped using cold water method and *S. aureus* whose growth was not inhibited by ogi liquor from the grains steeped using hot water (Table 1). The ogi liquor from millet steeped in cold water had the highest inhibitory effect on the growth of *S. typhimurium* (9.0 mm) while ogi liquor from white guinea corn and red guinea corn steeped in hot water gave the highest inhibition of 14.0 mm against *E. coli*. For *S. aureus*, ogi liquor from white guinea corn steeped in cold water gave the highest inhibition (14.0 mm) on the organism while for *S. dysenteriae*, ogi liquor from white guinea corn steeped in hot water gave the highest inhibition (9.0 mm). Ogi liquor from yellow maize steeped in hot water on the other hand gave the highest inhibition (13.0 mm) against *Enterobacter* sp.

For the antibiotic sensitivity assay, the antibiotic that had the highest inhibitory effect on the growth of *S. typhimurium* is gentamycin; it gave an inhibitory zone of 20.0 mm (Table 1). Antibiotics such as amoxycillin, augmentin, colistin, nalidixic acid, nitrofurantoin and cotrimoxine did not inhibit the growth of this organism. For *E. coli* and *Enterobacter* sp., ciprofloxacin gave the highest growth inhibition of these organisms with zones of inhibition of 48.0 mm and 50.0 mm, respectively. For *S. aureus* and *S. dysenteriae,* ofloxacin on the other hand gave the highest inhibition of the growth of these organisms with zones of inhibition of 36.0 mm and 29.0 mm, respectively. Although tetracycline inhibited the growth of all the test organisms, this inhibition was not superior to that mediated by the ogi liquors used (Table 1).

The pH of the ogi liquors used ranged from 5.02 - 5.45, while the titrable acidity (% lactic acid) ranged from 13 - 22% (Table 2). From the ogi liquors, organisms such as *Lactobacillus* sp., *Saccharomyces cerevisiae* and *Candida krusei* were isolated and identified.

DISCUSSION

The growing resistance of microorganisms to antimicrobial agents is becoming a serious concern in the treatment of infectious diseases. As a result, efforts are being made to develop antimicrobial agents from local sources for better chemotherapeutic effects (Gills, 1992). From this study, almost all the test organisms were inhibited by the raw ogi liquor prepared from the different grains used, irrespective of the mode of steeping. This shows that raw ogi liquor has bioactive components, which had inhibitory activity on these organisms. One of such could be bacteriocins which are proteinaceous antimicrobial compounds that are inhibitory towards sensitive organisms (Tagg et al., 1976; Ogunbanwo et al., 2003) and which act by destroying the bacterial membrane (Savadogo et

Table 1. Inhibitory effect of ogi liquor from the different grains and antibiotic sensitivity pattern on the growth of selected common diarrhoeal bacteria.

Type of grains/Antibiotics	Method of steeping	Diameter of zones of inhibition (mm)				
		Salmonella typhimurium	*Escherichia coli*	*Staphylococcus aureus*	*Shigella dysenteriae*	*Enterobacter* species
Maize (white)	Cold	4.0	7.0	13.0	8.0	0.0
	Hot	4.0	8.0	0.0	6.0	4.0
Maize (yello	Cold	3.0	5.0	12.0	8.0	0.0
	Hot	7.0	6.0	0.0	4.0	13.0
Guinea corn (white)	Cold	7.0	5.0	14.0	6.0	0.0
	Hot	8.0	14.0	0.0	9.0	7.0
Guinea corn (red)	Cold	6.0	9.0	12.0	6.0	0.0
	Hot	8.0	14.0	0.0	5.0	6.0
Millet	Cold	9.0	10.0	10.0	4.0	0.0
	Hot	8.0	6.0	0.0	6.0	7.0
Gentamycin		20.0	24.0	26.0	2.0	28.0
Ofloxacin		10.0	32.0	36.0	29.0	42.0
Ciprofloxacin		7.0	48.0	25.0	28.0	50.0
Tetracycline		4.0	8.0	5.0	4.0	1.0
Amoxycillin		0.0	14.0	0.0	0.0	16.0
Colistin		0.0	22.0	0.0	0.0	0.0
Augmentin		0.0	18.0	0.0	0.0	0.0
Nalidixic acid		0.0	0.0	0.0	0.0	8.0
Nitrofurantoin		0.0	0.0	0.0	0.0	0.0
Cotrimoxine		0.0	0.0	0.0	0.0	0.0

Table 2. The pH and total titrable acidity of the ogi liquors used in this work.

Types of grains used	Method of steeping	pH	Total titrable acidity (%)
Maize (white)	Cold	5.09	20
	Hot	5.40	14
Maize (yellow)	Cold	5.02	22
	Hot	5.31	15
Guinea corn (white)	Cold	5.30	17
	Hot	5.45	13
Guinea corn (white)	Cold	5.32	15
	Hot	5.34	14
Millet	Cold	5.35	14
	Hot	5.16	16

al., 2006). This could be a possibility because lactobacilli which have been reported by Lindgren and Dobregosz (1990) and Brink et al. (1994) to produce various compounds such as organic acids, diacetyl, hydrogen peroxide and bacteriocin during lactic acid fermentation were actually isolated from the liquors. Other bioactive component that may likely be present therefore in the liquor is lactic acid which is normally determined as titrable acidity and expressed as the % lactic acid content. From this investigation, the total titrable acidity observed which ranged from 13 - 22% points to the fact that lactic acid was actually present in the liquors. In addition, the low pH of the liquors (5.02 - 5.45) (Table 2) could also be partly responsible for the inhibition because most bacteria cannot grow at low pH except few such as the lactic acid bacteria. These could be some of the factors responsible for the inhibition of the test organisms by the ogi liquors used.

This study has been able to confirm that uncooked ogi liquor from different varieties of grains has antibacterial

activity against common bacteria that cause diarrhoea. It is, therefore, suggested that people having diarrhoea should drink raw ogi liquor to treat the infection especially in rural areas where they might not have access to medical attention. This will save many lives especially infants' in rural communities before they get to the hospital for proper medication.

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