

Full Length Research Paper

## Comparative antimicrobial potentials of amidun obtained from yellow and white maize varieties on some diarrhoea causing microorganisms

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This study determines the comparative antimicrobial potentials of amidun “supernatant solution of fermented maize mash” from yellow and white maize varieties on some diarrhoea causing microorganisms. The pH of the two amidun varieties were determined, their antimicrobial activities was also determined using agar well diffusion. Disc diffusion assay was also done using commercially produced antibiotics. Generally, amidun from the two maize varieties had acidic pH, with mean value of 3.6 for amidun from white maize and 4.0 for amidun from yellow maize. Both maize varieties inhibited growth of the tested microorganisms but at slightly different concentrations. The mean zones of inhibition of amidun from white maize varieties (9.5 mm) was higher than zones of inhibition by yellow maize varieties (9.2 mm) on *Salmonella abaeetuba* ATCC 35460, but with no significant ( $p>0.05$ ) difference. Zones of inhibitions given by amidun from the two maize varieties were the same (9 to 12 mm) on *Escherichia coli* ATCC 25922 and *S. abaeetuba* ATCC 35460 (7 to 12 mm). Although, the highest concentration obtained was not as high as what was observed with commercial antibiotics. This study showed that the effect produced by amidun from the two maize varieties is not significantly different ( $p>0.05$ ). Hence, the variety of maize used in this study had no significant ( $p>0.05$ ) effect on the antimicrobial activity of amidun

**Key words:** Antimicrobial, amidun, agar well, antimicrobial assay.

### INTRODUCTION

Maize or corn (*Zea mays*), is the most important cereal crop in sub-Saharan Africa and, with rice and wheat, it is one of the three most important cereal crops in the world. Maize is high yielding, easy to process, readily digested,

and cheaper than other cereals. It is also a versatile crop; growing across a range of agro ecological zones. Every part of the maize plant has economic value: the grain, leaves, stalk, tassel, and cob can all be used to produce

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a large variety of food and non-food products (Purseglove, 1992; Osagie and Eka, 1998). One of such food prepared from maize is “ogi”, fermented cereal porridge made from maize and produced using simple processing methods. The “ogi” porridge is very smooth in texture and has a sour taste reminiscent of that of yoghurt (Banigo and Muller, 1972). The traditional preparation of “ogi” include soaking of corn kernels (maize grains) in cold water for 2 to 3 days, washing several times with clean water, wet milling to paste and sieving to remove bran, hulls and germ (Akinrele, 1970; Odunfa, 1985). The pomace retained on the sieve is discarded and used as animal feed, while the filtrate is fermented for 2 to 3 days to yield “ogi”, which is a sour, starchy sediment with water on top. The top water is called “omi-eko” or “omikan” or omidun. Ogi colour depends on the colour of the maize grain used, yellow or milk white maize grain (Onyekwere et al., 1989). “Ogi” an acid fermented cereal mash made from maize or corn is traditionally produced by soaking the maize/corn grains in cold or warm water for 24 to 72 h to allow for fermentation before it is wet milled, sieved and soured. Sedimentation of the soured “ogi” for 48 to 72 h gives rise to omidun (Dada and Muller, 1983; Odunfa and Adeleye, 1985; Onyekwere et al., 1989; Teniola and Odunfa, 2001). Omidun has been traditionally found to be of medicinal importance in the South-Western part of Nigeria. It is used to soak bark of the root of some plants to treat not only fever and malaria, but is popularly used as solvent for herbal extraction, dish stain removal and as insect killers. Omidun has been used in the extraction of antimicrobial agents from some leaves such as *Bryophyllum pinnatum* and *Kalanchoe crenata*. The extracts were found inhibitory against some Gram-negative organisms (Aibinu et al., 2007). Information from indigenes also claims that omidun is popularly used in the control of diarrhoea. Series of work has been done on the microbial and therapeutic values of omidun. Omidun has been reported by Falana et al. (2011) to contain some microorganisms including *L. plantarum*. They also reported that these microorganisms have antimicrobial efficacy against some pathogenic microorganisms including *Escherichia coli* (Falana et al., 2012). However, information on comparative effects of omidun from white and yellow maize varieties is also essential. Hence, this research focused on comparative effects of omidun from yellow and white maize varieties on two tested pathogenic microorganisms.

## MATERIALS AND METHODS

This study was carried out at the Department of Microbiology, Federal University of Agriculture, Abeokuta.

### Collection of omidun samples and tested microorganisms

White and yellow maize varieties were obtained from a market in

Abeokuta. The maize varieties were steeped separately for 72 h, wet milled, sieved and fermented for 48 h at 30±2°C by the maize natural microflora. The test organisms, *E. coli* ATCC 25922 and *S. abaetetuba* ATCC 35460, were typed cultures obtained from Lagos State University Teaching Hospital (LUTH), Idi Araba, Lagos, on Nutrient Agar slants and were taken to the laboratory for confirmation. Each was re-cultured on fresh nutrient agar plates for purity.

### pH determination

The pH of omidun samples from white maize varieties and yellow maize varieties was determined using Extech model 30451 pH meter with a reference glass electrode.

### Antibiotics sensitivity pattern of the test organisms

This was done using disc diffusion method of Ochei and Kolhatkar (2008). One millilitre of 18 h broth culture of each of the test organisms was transferred into sterile Petri-dishes (different organism per plate) using sterile syringe. Each plate was then overlaid with 20 ml nutrient agar, swirled carefully for even distribution of the organisms within the agar and allowed to gel. The procedure was done in triplicate for each omidun sample and the mean zone of inhibition was obtained for each triplicate. Standard commercial antibiotics disc was distributed on the seeded agar plates and incubated at 37°C for 24 h. The diameter of the zones of inhibition around the antibiotics were measured and recorded.

### Agar well antimicrobial assay

This was done using agar diffusion method. One milliliter of 18 h broth culture of each of the test organisms was transferred into sterile Petri dishes (different organism per plate) using sterile syringe. Each plate was then overlaid with 20 ml nutrient agar, swirled carefully for even distribution of the organisms within the agar and allowed to gel. Wells were bored in the agar with the aid of a sterile cork borer. Different omidun samples (0.1 ml) were put into each well and 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 were measured and recorded.

### Statistical analysis

The pH and diameter of zones of inhibition of the varieties of omidun samples were subjected to one-way analysis of variance (ANOVA) and Duncan Multiple Range Test to separate the means and it was determined at the 5% probability level using SPSS 16.0 for Windows (SPSS, 2007).

## RESULTS

### pH of omidun from white and yellow maize varieties

Omidun samples obtained from white maize varieties had pH values ranging from 2.9 to 4.2, while omidun samples obtained from yellow maize varieties had pH values ranging from 3.4 to 4.5. There was a significant difference ( $p < 0.05$ ) between the mean pH values of omidun from

**Table 1.** pH of omidun obtained from white and yellow maize varieties.

Sample code	pH of white maize	Sample code	pH of yellow maize
1	3.7 ±1.4	16	3.4 ±0.6
2	2.9±1.4	17	3.6±0.2
3	3.1±0.1	18	3.6±0.2
4	3.9±1.3	19	3.6±0.3
5	3.3±0.4	20	4.0±0.7
6	4.0±0.2	21	4.9±1.3
7	3.3±0.5	22	4.0±0.3
8	4.2±0.1	23	4.2±0.3
9	4.2±0.3	24	4.1±0.2
10	3.4±0.5	25	4.3±0.4
11	3.5±0.7	26	3.6±0.8
12	3.1±0.2	27	3.7±0.2
13	3.0±0.7	28	4.2±0.9
14	3.6±0.5	29	4.0±0.1
15	4.1±0.2	30	4.5±0.1
Mean	3.6±0.4	Mean	4.0±0.4

Mean values within a column with the same letter are not significantly different ( $p>0.05$ ).

white maize varieties (3.6) and that of omidun from yellow maize varieties (4.0).

#### Antimicrobial sensitivity result of commercial antibiotics

Some commercial antibiotics were tested for antimicrobial activity against the two gastro enteric bacteria (Table 2). The antibiotics showed varying degree of activity to the tested microorganisms. There was a significant difference ( $p<0.05$ ) in the activity of the tested microorganisms. Ciprofloxacin, Azithromycin, Cefuroxime and Ceftriazone were effective against the tested microorganisms by showing diameters of zones of inhibition greater than 14 mm. The two organisms showed resistance to Gentamicin, Tetracycline and Amoxicillin with diameters of zones of inhibition less than 14 mm. Augmentin was effective against *E. coli* ATCC 25922, but *S. abaetetuba* ATCC 35460 was resistant to it.

#### Comparative inhibitory effect of omidun from white and yellow maize varieties on the tested microorganisms

Similar values of zone of inhibition (ranging between 9 and 12 mm) were obtained for omidun samples from white maize varieties and yellow maize varieties on *E. coli* ATCC 25922; also 7 to 12 mm was obtained for omidun samples from white and yellow maize varieties on *S. abaetetuba* (Tables 3 and 4). The type of maize from

which omidun was prepared did not significantly ( $p>0.05$ ) affect the antimicrobial activity exhibited on the gastro enteric bacteria. Though, the mean zones of inhibition of omidun from white maize on *E. coli* ATCC 25922 was 10.6 mm, while it was 10.7 mm on *S. abaetetuba* ATCC 35460, the difference was not significant ( $p>0.05$ ). Also, the mean antimicrobial activity shown by omidun from yellow maize on *E. coli* ATCC 25922 and *S. abaetetuba* ATCC 35460 was 9.5 and 9.2 mm, respectively (Figure 1).

#### DISCUSSION

This study has revealed that omidun samples obtained from white maize and yellow maize varieties showed varying pH (Table 1). The mean pH obtained for omidun from yellow maize varieties (4.0) was significantly higher than that of omidun obtained from white maize (3.6) varieties. This is similar to what was obtained by Omemu et al. (2007) during the souring period of "ogi". Therefore, the acidic pH of omidun samples from the two maize varieties could be partly responsible for the zones of inhibition obtained in this study, this is because most bacteria are acid intolerant (Steinkraus, 1997) and cannot grow at low pH except a few such as the lactic acid bacteria (LAB) (Savadogo et al., 2006). LAB has also been reported as predominant in fermented cereal products by many authors (Omemu, 2011; Opere et al., 2012).

The issue of antimicrobial resistance of pathogens to antibiotics have necessitated the development of antimicrobial agents from other sources (Williams, 2000)

**Table 2.** Inhibitory effects of antibiotics on *E. coli* ATCC 25922 and *S. abaetetuba* ATCC 35460.

Antibiotics	Zone diameter (mm) against <i>E. coli</i> ATCC 25922	Zone diameter (mm) against <i>S. abaetetuba</i> ATCC 35460
Ciprofloxacin	21±0.0	20±1.4
Azithromycin	20±0.0	20±0.0
Cefuroxime	17±0.4	16±0.1
Ceftriazone	17±0.0	14±0.3
Augmentin	15±0.5	10±0.2
Amoxycillin	6±0.0	8±0.1
Tetracycline	4±0.0	6±0.4
Gentamicin	4±0.0	4±0.0

Mean values within a column with the same letter

**Table 3.** Inhibitory effect of omidun from white maize on test organisms.

Sample code	Zone diameter (mm) against <i>E. coli</i> ATCC 25922	Zone diameter (mm) against <i>S. abaetetuba</i> ATCC 35460
1	12±1.4	10±0.7
2	12±0.7	12±0.7
3	10±1.4	8±0.7
4	9±0.7	7±1.4
5	10±1.4	8±0.7
6	10±0.7	10±1.4
7	12±0.7	10±1.6
8	9±2.1	8±0.2
9	12±0.7	12±0.9
10	10±1.4	12±0.2
11	10±1.4	9±1.4
12	12±0.9	10±0.1
13	12±1.4	10±1.4
14	10±0.7	8±1.4
15	9±2.8	9±1.4

Mean values within a column with the same letter are not significantly different ( $p>0.05$ ).

and that is one of the reasons for this research work. In the present study, similar zones of inhibitions (9 to 12 mm against *E. coli* ATCC 25922 and 7 to 12 mm against *S. abaetetuba* ATCC 35460, respectively) were obtained for the studied omidun samples from the two maize varieties (Tables 3 and 4). Hence, the type of maize from which omidun was prepared did not significantly ( $p>0.05$ ) affect the antimicrobial activity exhibited on the tested gastro enteric bacteria.

The observed zones of inhibition may be due to LAB in 'ogi', which cannot be ruled in omidun. LAB had been known to produce antimicrobial substances such as organic acids, diacetyl, hydrogen peroxide and bacteriocin (Mathieu et al., 1993; Bonade et al., 2001) and has been reported to be a potential host for the production of therapeutic recombinant protein.

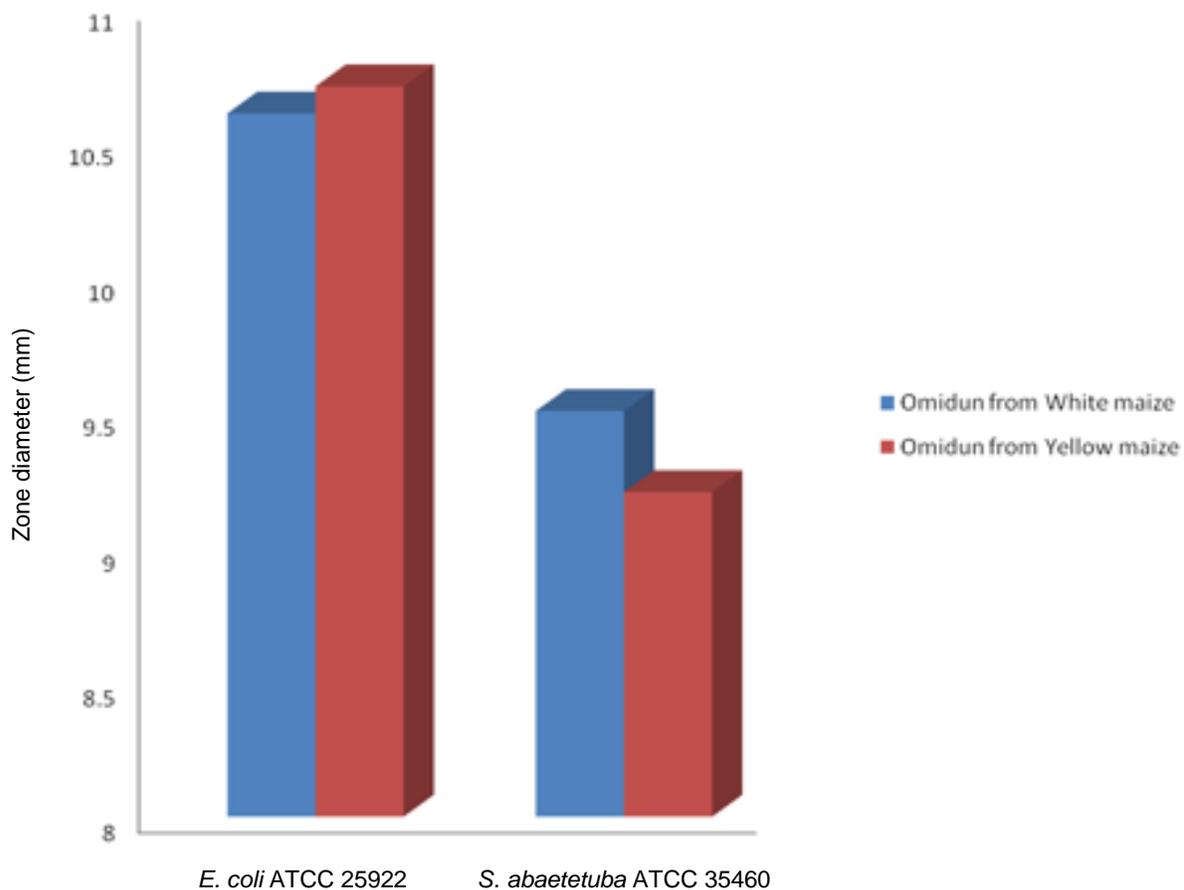
Although, omidun from white maize varieties showed slightly higher zones of inhibition (10.6 mm against *E. coli* ATCC 25922 and 10.7 mm against *S. abaetetuba* ATCC 35460), than omidun from yellow maize varieties (9.5 mm against *E. coli* ATCC 25922 and 9.2 mm against *S. abaetetuba* ATCC 35460). However, the varying zones of inhibition were not significant ( $p>0.05$ ) and may be attributed to their varying TTA as obtained by Omemu et al. (2007) during the souring process of "ogi". It might also be attributed to the varying microbial composition of the omidun samples (Odufa and Adeleye, 1985).

Generally, the high antimicrobial activity of omidun from white maize (Figure 1) was not as high as what was observed in most of the commercially produced antibiotics, an indication that commercial antibiotic (orthodox medicine) was more effective against the

**Table 4.** Inhibitory effect of omidun from yellow maize on test organisms.

Sample code	Zone diameter (mm) against <i>E. coli</i> ATCC 25922	Zone diameter (mm) against <i>S. abaeetuba</i> ATCC 35460
16	10±1.4	8±0.7
17	12±0.7	10±1.1
18	9±1.4	8±1.4
19	9±0.7	9±0.8
20	10±0.7	10±1.4
21	12±0.1	10±1.4
22	10±0.1	10±0.5
23	12±0.1	12±1.4
24	12±0.9	10±0.2
25	10±0.5	8±0.7
26	10±0.0	8±0.4
27	10±0.2	10±0.7
28	12±0.2	10±1.2
29	10±1.2	7±0.9
30	12±0.1	8±1.4

Mean values within a column with the same letter are not significantly different ( $p>0.05$ ).



**Figure 1.** Mean inhibitory effect of omidun from white maize and yellow maize on *E. coli* ATCC 25922 and *S. abaeetube* ATCC 35460.

tested microorganisms than amidun samples. Antimicrobial activities of amidun samples may not be due to the action of a single active compound found in them and some of which might have been detoxified by the tested microorganisms. Moreso, disparity between activities of the two varieties of amidun and standard antimicrobial drug may be due to the mixture of bioactive compounds present in amidun samples compared to the pure compound contained in the standard antibiotics (Gatsing et al., 2010). However, many consumers may still prefer the use of LAB or their antibacterial compounds (such as amidun) to control food-borne pathogens and spoilage organisms. Many researchers such as Ogunbanwo et al. (2004) reported that, unlike synthetic chemicals, cells of LAB which might be present in amidun samples have no known adverse effects.

This work shows that both amidun obtained from white and yellow maize varieties have potential therapeutic properties, but this property was slightly higher in amidun from white maize varieties than in amidun from yellow maize varieties, although with no significant difference ( $p > 0.05$ ) and this might mean that amidun from white maize varieties possess slightly higher antimicrobial components more than amidun obtained from yellow maize variety.

## Conclusion

The findings in this research work suggest the use of amidun, preferably the one obtained from white maize variety in the control of conditions caused by the tested microorganisms. However, further work is highly recommended in order to isolate, identify and quantify the active metabolite present in amidun samples from the two maize varieties (white and yellow).

## Conflict of Interests

The authors have not declared any conflict of interests.

## REFERENCES

- Aibinu I, Adenipekun E, Odugbemi T (2007). Emergence of Quinolone Resistance among *Escherichia coli* strains isolated from Clinical infections in some Lagos state Hospitals in Nigeria. *Nig. J. Health Biomed. Sc.* 3(2):73-78.
- Akinrele IA (1970). Fermentation studies on maize during the preparation of a traditional African starch-cake food. *J. Sci. Agric.* 21(12):619-625.
- Banigo EOI, Muller HG (1972). Manufacture of ogi (a Nigerian fermented cereal porridge): Comparative evaluation of corn, sorghum and millet. *Can. Inst. Food Sci. Technol. J.* 5 (4):217-221.
- Bonade JA, Dagnan AJ, Garver MJ (2001). Production of helveticin from *Lactobacillus helveticus*. *Lett. Appl. Microbiol.* 33:153-158.
- Dada LO, Muller HG (1983). The fate of aflatoxin B1 in the production of ogi, a Nigerian fermented sorghum porridge. *J. Cereal Sci.* 1(1):63-70.
- Falana MB (2011). Microorganisms associated with supernatant solution of fermented maize mash (Omidun) from two varieties of maize grains. *Researcher* 3(7).
- Falana MB, Bankole MO, Omemu AM, Oyewole OB (2012). Antimicrobial potentials of some selected Microorganisms associated with supernatant solution of fermented maize mash amidun from two varieties of maize grains. *Afr. J. Microbiol. Res.* 6:4095-4101.
- Gatsing D, Nkeugoauapi CFN, Nkah BFN, Kuate JR, Tchouanguép FM (2010). Antibacterial activity, bioavailability and acute toxicity evaluation of the leaf extract of *Alchornea cordifolia* (Euphorbiaceae). *Int. J. Pharmacol.* 6:173-182.
- Mathieu F, Sudirman S, Rekhif N, Milliere JB, Lefebvre G (1993). Mesenterocin 52 a bacteriocin produced by *Leuconostoc mesenteroides* subspecies mesenteroides FR 52. *J. Appl. Bacteriol.* 74: 372-379.
- Ochei JO, Kolhatkar AA (2008). *Medical Laboratory Science Theory and Practice*. Tata McGrawHill Publishing Company, New York. pp 603.
- Odunfa SA, Adeyeye S (1985). Microbiological changes during the traditional production of ogi baba, a West African fermented sorghum gruel. *J. Cereal Sci.* 3(2):173-180.
- Odunfa SA (1985). African fermented foods. *In: Wood, B.J.B. (Ed.), Microbiology of Fermented Foods, Vol. 2*. Elsevier Science, London and New York, pp. 155-199.
- Ogunbanwo ST, Sanni AI, Onilude AA (2004). Effect of bacteriocinogenic *Lactobacillus* sp. on the shelf life of *fufu* a traditional fermented cassava product. *World J. Microbiol. Biotechnol.* 20:57-63.
- Onyekwere OO, Akinrele IA, Koleoso OA (1989). Industrialization of ogi fermentation. *Industrial. Indig. Fermented Foods* 33:329-362.
- Omemu AM, Oyewole OB, Bankole MO (2007). Significance of yeast in the fermentation of maize for Ogi production. *Int. J. Food Microbiol.* 24:571-576.
- Omemu AM (2011). Fermentation dynamics during production of ogi, a Nigerian fermented cereal porridge. *Report Opin.* 3(4):8-17.
- Opere B, Aboaba EO, Ugoji OO, Iwalokun ABA (2012). Estimation of Nutritive Value, Organoleptic Properties and Consumer Acceptability of Fermented Cereal Gruel (OGI). *Adv. J. Food Sci. Technol.* 4(1):1-8.
- Osagie AU, Eka OU (1998). Nutritional Quality of Plant Foods. Postharvest Research Unit, University of Benin, Benin. pp. 34 -41.
- Purseglove JW (1992). *Tropical Crops: Monocotyledons*. Longman Scientific and Technical, New York, pp. 300-305.
- Savadogo A, Outtara CA, Bassole IH, Traore SA (2006). Bacteriocins and lactic acid bacteria - A mini review. *Afr. J. Biotechnol.* 5(9):678-683.
- SPSS (2007). *SPSS 16.0 for Windows*. SPSS Inc. Chicago, IL.
- Steinkraus KH (1996). *Handbook of Indigenous Fermented Foods*, 2<sup>nd</sup> ed, Marcel Dekker, Inc, New York.
- Steinkraus KH (1997). Classification of fermented foods: Worldwide review of household fermentation technique. *Food Control* 8: 311-317.
- Teniola OD, Odunfa SA (2001). The effects of processing methods on the level of lysine, methionine and the general acceptability of ogi processed using starter cultures. *Int. J. Food Microbiol.* 63:1-9.