



Incidence of Microbial Contamination in Pearl Millet Based Gruel Used as Weaning Food during Traditional Cooling

Adamu, S. D.
Home Economics Department,
Federal College of Education, Zaria, Kaduna State.

ABSTRACT

The incidence of microorganisms in pearl millet-based gruel during traditional cooling was assessed. The total aerobic counts of the gruel after traditional cooling were evaluated. The result indicated that the samples of gruel produced from varieties of eight millet cultivars (WA-14, WA-18, WA-21, WA-25, WA-26, WA-27, WA-29, and WA-32) were contaminated during cooling. Results also indicated that the total bacterial count of unmalted millet-based gruel after cooling for the eight millet cultivars ranged from 1.17 to 1.88 CFU/ML while its mold count ranged from 0.47 to 1.98 CFU/ML. Also, the total bacterial count of malted millet-based gruel for the eight millet cultivars after cooling ranged from 1.17 to 1.96 CFU/ML while the mould count ranged from 0.30 to 0.95 CFU/ML. The microorganism isolated from the gruel produced were; *Enterococcus faecalis*, *Escherichia coli*, *Aspergillus niger*, *Aspergillus flavus*, and *Schizosaccaromyces pombe*. Although some of these microorganisms are pathogenic they occurred at relatively low frequencies. Hence, there is a need to practice strict hygiene during this process.

ARTICLE INFO

Article History

Received: November, 2022
Received in revised form: March, 2023
Accepted: April, 2023
Published online: June, 2023

KEYWORDS

Millet; Microorganism; Cereal; weaning food; Flour

INTRODUCTION

Cereal grain is the major crop grown in the tropical region of Africa and other ecologically similar regions of the world. Most cereals have great potentials and contain nutrient as; protein, carbohydrate, fat, mineral matter, vitamins and other accessory food factors (Singh and Sosulki, 1985).

The roles played by these nutrients are for growth and replacement of energy required by the body. This function is not restricted to any type of nutrients; as carbohydrate, fat and protein can all provide energy and their values in this respect are measured in units or calories (Mccance and Widdowson, 1978).

Production of Cereal Grains

Cereal supply the most calories per acre among food crops, and can be safely stored for a longtime, processed into many acceptable products (Hulse et al., 1980). They are adapted to variety of soils and climatic

conditions. They can be cultivated both on a large scale with a small amount of labour, and on a small scale by human labour. They are excellent source of inexpensive protein, certain minerals and vitamins. More than two-third of the world's cultivated arable land are used in the cultivation of grain crops. Most developing countries rely on cereals as food sources. Cereals provide more than half of the calories consumed by humans and in most developing countries they provide over two-third of the total food. The major cereal grains of the world are: wheat, barley, rice, maize, sorghum and millet (Okaisabor, 1980).

In Nigeria, these grains are consumed directly or in a modified form as a major item of diet/flour, oil, bran e.t.c. these grains especially millet is processed into various traditional foods which include; tuwo, sinasin, kunun zaki, akammu, fura and burabusko (Nkama, 1993).

Corresponding author: Adams, S. D.

✉ regence2000@yahoo.com

Home Economics Department, Federal College of Education, Zaria.

© 2023. Faculty of Tech. Education, ATBU Bauchi. All rights reserved

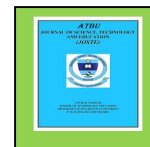


Table1: Output of Some Selected Cereal Crops in Nigeria ['000]

CROPS	2009	2010	2011
MILLET	4771	5120	5172
SORGHUM	6543	7014	7123
WHEAT	213	207	209
RICE	1800	2200	2450
MAIZE	11870	13956	14345

Source: FAO, 1991.

Millet

Millet originated in the semi arid savannah Sahel zones of Africa where many of the wild relatives can be found. The total world production of millet is put at approximately forty million (40 million) metric tons on some 55.9 hectares or 0.58 metric tones per hectare. The leading countries in the production of millet are; India, China and Nigeria respectively (FAO, 2009).

Millet is an important and warm temperature cereal crop, which avoid drought

and the only crop that thrive well in drier parts of the world. It is a highly palatable and nutritious grain crops (Ihekoronye and Ngoddy, 1985). It is commonly grown on sandy and free draining soil. In the sudan-sahel savannah, short duration types of millet require about 220mm of water well distributed during the month of august. The long duration cultivars may require 400-800mm of water over a period of three and half months (Agboola, 1979).

Table2: Projected Demand and Production Target of Millet (2010-2020) ['000]

Year	Demand	Production
2010	5228	5128
2011	5408	5196
2012	5416	5182
2013	6969	6699
2014	7705	7168
2015	8015	7795
2016	8815	8081
2017	9185	8501
2018	9306	8660
2019	9409	8765
2020	9815	8962

Source: FAO, 2009.

Varieties of Millet

Approximately one third of the world's millet is grown in Africa, about 70% of which is grown in West Africa. The various types of millet varieties available are; *Penisetum Americana* (former name). *Penisetum typhoides* (early millet) or Pearl millet (or cocktail or bulbrush or bajra) is the most common millet. Another variety of millet is *Eleusine coracana* otherwise called proso, ragi or mania. Some lesser known food species of millet are; foxtail millet (*sotaria italica*), Japanese millet (*Echinochloa crusgali*), brown top millet (*Panicum ramosum*).

In Nigeria, two species are common, i.e. *Penisetum typhoid* (early millet) and *Penisetum meriva* (late millet).

Uses and Utilization

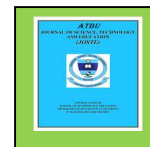
Millet is a staple food for millions of people in the dryer part of Africa and sub-tropical Africa. Air dried grain contains appropriately 12.4% water, 11.6% protein, 5% fat, 6.7% carbohydrate, 1.2% fibre and 2.7% ash. The gluten content varies from 8.8% - 16% (Onweme and Sinha, 1991). Millet is generally low in glycine and methionine. However, the nutritive value of millet is comparable to that of rice and wheat. The

Corresponding author: Adams, S. D.

✉ regence2000@yahoo.com

Home Economics Department, Federal College of Education, Zaria.

© 2023. Faculty of Tech. Education, ATBU Bauchi. All rights reserved



grain may be cooked like rice or grounded into flour to make porridge, cake or bread. Millet grains are also used to produce malt. Malted seed is an important source of beer used in the brewing industry for the production of alcoholic beverages. Millet is also used to produce many traditional food such as; Fura, Sinasin, Kunun zaki and to a large extent weaning food.

According to Kent (1983) over 400 million people rely on millet for their sustenance out of a total world production of 44 million millet grains. Some 85% was used for human food and 10% for feed (FAO, 1991).

Research Problem

The preparation of weaning foods by use of millet based gruel is common in most Nigerian communities. Most mothers starts giving their infant children this weaning food from the ages of 3 months and above. Their preparations in most cases are usually crude and lack conventional hygienic procedures; as such most of the time contamination of microbes occurs. This has led to frequent cases of diarrhea and cholera in children. Hence the need to study the traditional method of preparing these millet based weaning food has become imperative, in order to identify the likely microorganisms associated with the gruel.

Research Objectives

Based on the research problem identified, the objectives of the research were as follows:

1. To determine the total aerobic plate count of pearl millet based gruel.
2. To isolate and identify microorganisms associated with the various gruel.
3. To assess the distribution of these microorganisms isolated in each.

MATERIALS AND METHOD

Raw Materials

The millet used as raw material in this work was obtained from the institute of Agricultural Research, Ahmadu Bello University, Samaru-Zaria. These were WA-25, WA-26, WA-18, WA-32, WA-27, WA-14, WA-29 and WA-21. The post harvest conditions of

the millet were not known. The chemicals used were analytical grade and the methods used were standardized methods.

Materials;	Source
Hot air oven -	(Memmert/Germany)
Water bath -	(Memmert/Germany)
Autoclave -	(Express/Germany)
Incubator -	(Memmert/Germany)
Microscope -	(Olympus/Switzerland)
Colony counter -	(Gallenkamp/England)
Aluminium foil	
Cotton wool	
10% Commercial bleach	
50% Lactic acid	
70% Absolute alcohol	
Lactophenol mountain fluid	
Slides and cover slips	
Inoculating needle	
Industrial methylated spirit	
Dilution tubes	
Petri dishes	
Bacteriological pipette	
Test tube racks	
Beakers	
Wire loop	
Spirit lamp	
Scales	
pH meter	
Thermometer	
Refrigerator	
Hammer mill and swab stick	

Method

Sample Preparation

The millet grain used were steeped for 12-24 h and then sterilized with a bleach (parazon) containing 3.5% sodium hypochlorite. Then allowed to germinate for 72 h and harvested, dried polished, ground into powder form (Mousa et al., 1992).

The whole analysis was conducted in an aseptic condition while the surface used was sterilized with disinfectant (hypo bleach) whose concentration is 0.5%.

Culture Media Used

Manitol Salt Agar (MSA)
Nutrient Broth (NB)
Eusine Methylene Blue Agar (EMBA) – Levine
Deoxycholate Citrate Agar (DCA)
Nutrients Agar (NA) – Oxoid Ltd- England

Corresponding author: Adams, S. D.

✉ regence2000@yahoo.com

Home Economics Department, Federal College of Education, Zaria.

© 2023. Faculty of Tech. Education, ATBU Bauchi. All rights reserved

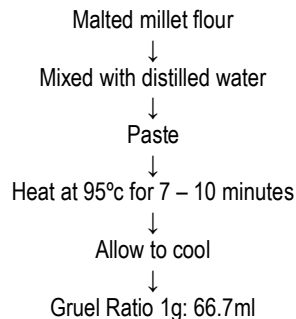


Maconkey Agar (MCA) – Acumedia, Maryland
Sabour and Agar (SBA) – Oxoid, England.

Gruel Production

Flour and distilled water were mixed in a glass beaker and heated in a boiling water bath to reach a cooking temperature of 95°C

within 7 to 10 min. The gruel was left at room temperature to cool. Gruels were prepared from either ungerminated flours or germinated flours or ungerminated flours mixed with 5 to 10% germinated flour added before or after cooking (Mousa et al., 1992).



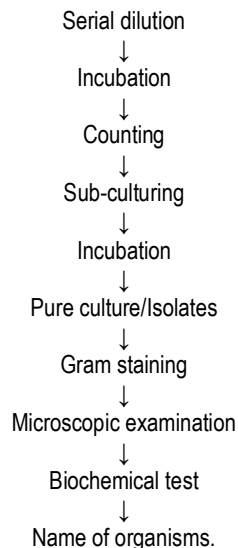
Flow chart for the preparation of gruel (Mousa et al., 1992).

Microbial Analysis

Viable bacteria, yeast and mould count were determined on preparation of gruel. First, a pilot test was carried out to determine how highly contaminated the sample were, so as to be able to determine the suitable dilution

factors. At the end of the test, 10⁻⁶ and 10⁻⁷ and 10⁻⁸ dilution powers were chosen. Eight-fold serial dilution were inoculated on dry nutrient agar plates, using the spread plates method as described by Mackie and McCartney (1978) to determine the surface viable count.

Preparation of sample for serial dilution.



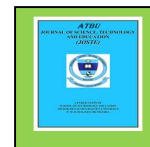
Flow-chart for microbial analysis Mackie and McCartney (1978).

Corresponding author: Adams, S. D.

✉ regence2000@yahoo.com

Home Economics Department, Federal College of Education, Zaria.

© 2023. Faculty of Tech. Education, ATBU Bauchi. All rights reserved



PROCEDURE FOR ENUMERATION

1.0g of the sample each was transferred into dilution tubes containing 9ml of sterilized physiological saline and allowed to stand for 15 min. It was then shaken vigorously to suspend the microorganism in the diluents. Eight-fold serial dilution was made from the suspension by withdrawing 1ml from the suspension and discharging into dilution tube containing 9ml of sterile physiological saline by means of sterile bacteriological pipette. This was labeled 10^{-1} dilution. Another 1ml was withdrawn from the 10^{-1} dilution and transferred into another 9ml diluents, this becomes 10^{-2} dilution. The serial dilution process continued until 10^{-8} dilution was reached for each millet sample (Mackie and McCartney, 1978).

0.1ml of each of the 10^{-6} , 10^{-7} and 10^{-8} dilution for each sample were pipette and discharged on the surface of the already prepared dried nutrient agar plates labeled according to the sample plate and dilution powers. Sterile fine wire loop was used to spread the suspension on the surface of the agar medium. The plates were incubated at 37°C for 24 h to 48 h to allow maximum growth before cooling. The plates were removed after incubation and colonies were counted using colony counter. The viable count was calculated by multiplying the colonies counted by the dilution powers and taking the average of the three plates per sample (i.e. 10^{-6} , 10^{-7} and 10^{-8}) (Ogundana, 1989). The results were explained as log of Colony Forming Unit (CFU) per gram of the sample. From the viable count plates, isolation and identification of these microorganisms were carried out using methods described by Harrigan and McCance (1976).

Sub-culturing was done by picking individual colonies by differing in cultural or colonial characteristics like the colour of the colonies, shape and size of the colonies, using sterile wire loop. The colonies are streaked on freshly prepared, solidified and dried McConkey agar plates separately. The plates were incubated at 37°C for 24 h. The sub culturing step was repeated over and over until pure discrete colonies were obtained as pure isolates. The pure isolates obtained were

subjected to grams staining; this marked the beginning of the identification of the unknown microorganisms present. This step was followed by the observations of the slides under the microscope. The gram reaction exhibited by each microorganism indicates the type of selective media used, which is part of the identification process. This process continued with biochemical test like the sugar fermentation, methyl red, coagulase, catalase, motility test (Harrigan and McCance, 1976).

Motility test

The motility test used is the hanging drop method. The spirit lamp was lit and the surface area was cleaned with a disinfectant. A wire loop was used to take a colony from the growth and put inside the bottle containing the peptone water. The inoculated sample were incubated into a slide cover and observed with microscope. This was done with an inflamed wire loop.

RESULT AND DISCUSSION

The result as shown in Table 3 and 4 indicated that the count of unmalted gruel obtained at the production stage (i.e. before cooling and after cooling), has no microbial count present before cooling. This is due to the heating/cooking of the gruel, since heat tends to inhibit the growth of microorganisms. However, after cooling the gruel the aerobic count were present in all the samples. In table 3, the gruel (WA-27) had the highest number of bacterial colonies observed followed by WA-21. The gruel having the least bacterial count is WA-14.

In table 4, it could be seen that the gruel having the highest mould count is WA-25, then followed by WA-27 and the least are WA-14, WA-18 and WA-32 having 0.47 each.

Table 5 shows the distribution of microorganisms in unmalted millet based gruel. It could be seen that *Escherichia coli* appear in all the gruel, second to *Escherichia coli* in appearance was *Aspergillus niger*, *Enterococcus faecalis*; *Aspergillus flavus* and *Schizosaccaromyses pombe* have the least.

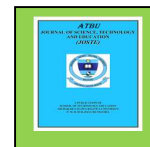


Table 3: Total bacterial plate count (\log_{10} CFU/ml of unmalted millet based gruel

Millet cultivars	Sample before cooling	Sample after cooling
WA-14	ND	1.17
WA-18	ND	1.49
WA-21	ND	1.64
WA-25	ND	1.46
WA-26	ND	1.44
WA-27	ND	1.88
WA-29	ND	1.50
WA-32	ND	1.47

ND = not detected
 Mean of triplicate determination

Table 4: Total mould count (\log_{10} CFU/ml) of unmalted millet based gruel.

Millet cultivars	Sample before cooling	Sample after cooling
WA-14	ND	0.47
WA-18	ND	0.47
WA-21	ND	0.69
WA-25	ND	1.98
WA-26	ND	0.69
WA-27	ND	0.95
WA-29	ND	0.60
WA-32	ND	0.47

ND = not detected
 Mean of triplicate determination

Table 5: Distribution of Microorganisms and their Frequency of Occurrence in Unmalted Millet Based Gruel

Gruel Cultivars	From Millet	Microorganisms
		<i>Enterococcus faecalis</i>
		<i>E. coli</i>
		<i>Aspergillus niger</i>
		<i>Aspergillus flavus</i>
		<i>Schizosaccharomyces pombe</i>
WA-14		8
WA-18		-
WA-21		18
WA-25		-
WA-26		-
WA-27		-
WA-29		4
WA-32		-

- = absent

Frequency calculation was based on percentage of occurrence in plated material

In Table 6 and 7 it indicated the microbial count of malted millet based gruel obtained before cooling and after cooling of gruel. It could be seen that before cooling the gruel, there was no microbial count due to heating or cooking this is because

heating/cooking inhibits the growth of microorganisms.

In table 8, the gruel that had the highest bacterial count are WA-25 and WA-26. WA-14 had the least. Whereas the gruel that had the highest mould count is WA-26, followed by WA-27 and the least is WA-21 (Table 7).

Corresponding author: Adams, S. D.

✉ regence2000@yahoo.com

Home Economics Department, Federal College of Education, Zaria.

© 2023. Faculty of Tech. Education, ATBU Bauchi. All rights reserved



Table 8 shows the distribution of microorganisms in malted millet based gruel. *Aspergillus niger* and *Aspergillus flavus* appear to be the most organism that had the highest frequency of occurrence, almost 70%. This is because they are seed borne fungus.

The occurrence of *Escherichia coli* might be due to fecal contamination through water. The organism isolated seems to appear to be similar organism isolated by Livingstone et al, (1992). Generally *Schizosaccaromyses pombe* causes the fermentation of farm produce.

Table 6: Total bacterial plate count (\log_{10} CFU/ml) of malted millet based gruel

Millet cultivars	Sample before cooling	Sample after cooling
WA-14	ND	1.17
WA-18	ND	1.49
WA-21	ND	1.44
WA-25	ND	1.96
WA-26	ND	1.96
WA-27	ND	1.85
WA-29	ND	0.90
WA-32	ND	1.38

ND = not detected
 Mean of triplicate determination

Table 7: Total plate mould count (\log_{10} CFU/ml) of malted millet based gruel

Millet cultivars	Sample before cooling	Sample after cooling
WA-14	ND	0.69
WA-18	ND	0.60
WA-21	ND	0.30
WA-25	ND	0.49
WA-26	ND	0.95
WA-27	ND	0.90
WA-29	ND	0.60
WA-32	ND	0.90

ND = not detected
 Mean of triplicate determination

Table 8: Distribution of Microorganisms and their Frequency of Occurrence in Malted Millet Based Gruel

Gruel Cultivars	From Millet	Microorganisms
		<i>Enterococcus faecalis</i>
		<i>E. coli</i>
		<i>Aspergillus niger</i>
		<i>Aspergillus flavus</i>
		<i>Schizosaccaromyse pombe</i>
WA-14	-	4
WA-18	17	6
WA-21	8	-
WA-25	4	4
WA-26	10	-
WA-27	-	14
WA-29	-	4
WA-32	12	14

- = absent
 Frequency calculation was based on percentage of occurrence in plated material



CONCLUSION

The results obtained showed that traditional cooling exposes the gruel, a weaning food to microorganism. These were the organisms identified; *Aspergillus niger*, *Aspergillus flavus*, *Schizosaccaromyces pombe*, *Escherichia coli* and *Enterococcus faecalis*.

Aspergillus flavus produces aflatoxins which are carcinogenic; *Aspergillus niger* causes lung diseases during sporulation stage of the fungus, and is dangerous when consumed. *Escherichia coli* causes diarrhea and this normally is contaminated through faeces.

RECOMMENDATION

To make this product more acceptable, the following recommendations are made;

1. Aseptic technique should be observed in the preparation of the gruel based weaning food
2. Adequately distilled and clean water should be used in the preparation of the gruel
3. Standardized and clean equipment should be used in its preparation.
4. Gruel should be cooled in a closed container to prevent contamination from the surrounding
5. Environment.

REFERENCES

- Agboola, S.A. (1979). *An agricultural atlas of Nigeria*. Oxford University press, 73-88.
- F. A. O. (1991). *Food and Agricultural Organization production year book*, FAO, Rome.
- F. A. O. (2009). *Food and Agriculture Organization of the United Nations, World Development Indicators database; Production estimates and crop assessment division*. USDA; Newyork, The World Bank.
- Harrigan, J. L. and McCance, R. A. (1976). Evaluation of microbiological activity in cereal grains. *Journal of microbiology*. 41: 196- 201.
- Hulse, J. H., Laung, E.M. and Pearson, D.E. (1980). *Sorghum and Millet. Their composition and Nutritive value*. 454-460. London Academy Press.
- Ihekoronye, A. I and Ngoddy, P.O. (1985). *Integrated Food Science and Technology for the Tropics*. (1st Ed.) London; Macmillan Publishers Ltd. 45-59.
- Kent, A. (1983). *Sorghum and Millet. Their composition, uses and value*; 274-282. London; Academy press.
- Livingstone, A. S., Sandhu, J. S. and Maeshi, N. G. (1992). Microbial evaluation of malted wheat chickpea and weaning food based on them. *Journal of Tropical Pediatrics*. (38); 74-77.
- Mackie, A. and McCartney, V. (1978). Bacteria contamination of infant foods. *Indian Journal of Medical Research* 77: 342- 346.
- McCance, R. A. and Widowson, E. M. (1976). *Chemical composition of foods*. Medical Research Council. London, H.M.S.O.
- Mousa, W. A., Tadoo, M. D., Mekhael, K. G. D. M. A. E. H., Shakir, A. H and Abdel-el-Renin, E. A. (1992). Some simple methods of home processing and their implication with weaning food. *Die Nahrung*. 36:1; 26-33.
- Nkama, I. (1993). "Studies on improving the nutritional quality of masa a traditional Nigerian fermented cereal based food". *Report central food technology research institute, Mysore, India*.
- Ogundana, W. (1989). *Microbiology of food. Journal of Medical Microbiology* 13: 57- 61. Churchill Livingstone ELBS.
- Okaisabor, E. K. (1980). The Nigerian Agrarian Revolution and the Nigerian Society for plant protection. NSPP occasional Publication No 3:1-10.
- Onwueme, I. C and Sinha, T. D. (1991). "Field crops in the tropics". CTA wagenengen, Netherlands.
- Singh, T and Susulki, F.W. (1985). Malting of hulls barley cultivars and gluaglea (Lactrivism) Utility wheat. *Journal of food science*, 50; 342-346.

Corresponding author: Adams, S. D.

✉ regence2000@yahoo.com

Home Economics Department, Federal College of Education, Zaria.

© 2023. Faculty of Tech. Education, ATBU Bauchi. All rights reserved