EVALUATION OF PROBIOTIC CONTENT OF COMMON COMPLEMENTARY FOODS USED IN MUBI METROPOLIS, ADAMAWA STATE, NIGERIA

***Nwankwo, R. N.[1], T a'awu, K.G.[2], Moshood, M .[1] and Madu, G.[1]**

1 Department of Nutrition and Dietetics, Federal Polytechnic Mubi.

2 Department of Food Science and Technology, Federal Polytechnic Mubi.

*Corresponding author [1]: ritanwanko82@gmail.com

ABSTRACT

Background: Probiotic bacteria are becoming increasingly important in the context of human nutrition based on the role they play in immunological, digestive and respiratory functions.

Objective: This study investigated the probiotic content and strengths of some complementary foods commonly used in Mubi Adamawa state, Nigeria.

Materials and Methods: Locally made cereal pastes (*kamu)* made from cereal grains sorghum, millet, and maize were purchased from the Mubi general market and coded as LSG, LMT and LMZ, respectively. Three most commonly used commercial complementary foods in Mubi metropolis were also purchased from Mubi market packed in cans of 450g each and coded as CC1, CC2, and CC3 respectively. The basic ingredients in each commercial product were recorded from the labels on the packages. De ManRogasa Agar was used to isolate the probiotic bacteria in all the samples using standard methods of AOAC (2000). Colony count and fungi identification were carried out.All analyses were done in triplicates.Data was analysed for means and standard deviation using Statistix 9, version 9.1(2012).

RESULT: Commercial complementary foods CC1 and CC2 had Lactobacillus species isolated with bacteria count of 7.5×10^2 and 8.7×10^2 Cfu / g respectively while CC3 had no bacterial specie isolated. Local complementary food LSG had no probiotic bacteria isolated while LMT and LMZ had Lactobacillus species isolated with bacteria count of 5.4×10^2 and 6.5×10^2 (Cfu/g) respectively. Commercial complementary foods CC1, CC2 and CC3 had no yeast isolated. Local complementary food LSG had the least yeast count of 1.01×10^3 (Cfu / g) of *Saccharomycescerevasiae*. LMT had 6.06×10^2Cfu /g and LMZ had the highest yeast count of 9.26×10^2 (Cfu / g) of *Saccharomyces Cerevasiae*.

CONCLUSION: Local complementary foods used in this study contained both probiotic bacteria (*Lactobacillus* species) and yeast (*Saccharomyces Cerevasiae*).

KEY WORDS: Evaluation, Probiotics, Complementary Foods.

INTRODUCTION

Probiotics are live microorganisms (in most cases bacteria) found in the human gut mainly in the intestine or in some foods (1). They are also called friendly bacteria or good bacteria. Probiotics influence health in general or specifically by improving the overall health when ingested as bacterial or yeast supplement (2). Probiotic bacteria are becoming increasingly important in the contest of human nutrition because of the role they play in immunological, digestive and respiratory functions as well as their significant effect in alleviating infectious disease in children(3).

Probiotic delivery system are usually neutriceutical products, commercial food based products and fermented dairy products (4). Other foods that may provide probiotics are fermented cereal gruel commonly called '*ogi'* made from maize, sorghum and millet. These cereal products are generally used in preparing complimentary foods in Nigeria (5).Momka (5) reported that fermented cereal products have been claimed to contain probiotics but may not be in the amount or in the form that is necessary to achieve the health benefits required. It is necessary to assess the probiotic content of the common complimentary foods in Mubi in other to determine their effectiveness as probiotic delivery system. Global markets of probiotics are expanding, it is necessary to harmonize national and international regulations and guidelines to evaluate the efficacy and safety of probiotic bacteria in most foods (6). To achieve this aim, evaluation of probiotic content of common complementary foods will be very useful.

The digestive system contains a variety of different types of bacteria. Some of these bacteria are beneficial (e.g. *Enterobacterium* and *Lactobacillus*) and some are detrimental (e.g. *Enterobacteriaceae* and *clostridiumspp*.) (7).These bacteria live naturally in the body and assist in breaking down the food eaten. Probiotics in foods when ingested help the bacteria that already live in the intestine to breakdown food for healthy digestion. For the body to be healthy, the good bacteria (probiotics) must maintain control over the bad bacteria to establish a positive balance between the two (8). In a healthy gut, these friendly bacteria compete with the disease causing variety to outnumber them. The goal of probiotics is to reinforce the health bacteria to avoid dysbiosis. Once the bad bacteria or disease causing bacteria outnumbers the friendly bacteria, they precipitate problem. Probiotics when ingested either in common foods or as supplements reinforce the health promoting bacteria. It is necessary that the complementary foods should contain the type of probiotics that exist in the gut of the child at the onset of complementary feeding to maintain homeostasis.

Complementary feeding is the provision of food to infants in addition to breast milk from 6 months of age when breast milk is no longer enough to meet the nutritional need of the infants (9). The transition from exclusive breastfeeding to family food is very vulnerable. During weaning there are significant

changes on the gastrointestinal microbiota (9). Deletarious alterations of the gastrointestinal microbiota can result in pathogenical process while measures that stimulate its development and stability such as the use of probiotics are beneficial (10). Hiembach*et al.* (11) reported that some digestive disorders result when the balance of friendly bacteria in the intestine becomes disturbed. It is very imperative that the initial complementary foods, commercial or locally made should contain reasonable amount of probiotics to maintain a positive balance in the child's gastrointestinal tract. Some commercial complementary foods are claimed to have probiotic content necessary for proper upbringing of the infants (12). Most home made complementary foods are made of fermented cereal products (9).Experts demand that complementary foods must contain the necessary probiotic bactaria for proper infant gastrointestinal microbiota (13, 14). Several researches have been conducted to evaluate the suitability of different cereal grains to enhance probiotic bacteria growth and maintain their viability (13, 14, and 16). Some researches has also been conducted on the probiotic effects of complementary foods (9, 19) but there is paucity of data on the probiotic content and probiotic strength of common complimentary foods used in Mubi metropolis. It is necessary to identify the probiotic content and strength of the various commercial and locally made complementary foods used in Mubi metropolis. This will provide evidence based recommendations to the mothers. This calls for the trust of this study to determine the probiotic content and strength of some commercial and locally made complementary foods used in Mubi metropolis in Adamawa state Nigeria.

Materials and Methods

Sourcing of materials

Cereal pastes as is used in preparing complementary foods in Mubi metropolis commonly called '*ogi'* or '*kamu'* made with cereal grains sorghum, millet, and maize were purchased from the Mubi general market in sterile containers and coded as LSG, LMT and LMZ respectively. Three most commonly used commercial complementary foods in Mubi metropolis were also purchased from Mubi market package in cans of 450g each and coded as CC1, CC2, and CC3 respectively. The basic ingredients in each commercial product were recorded from the labeling on the tins. The manufacturing companies were recorded. The material for isolating probiotics (DemanRagosa Agar) was bought from chemical material stores in Mubi. The analysis was carried out in the Food microbiology laboratory of the Department of Food Science and Technology, Federal Polytechnic Mubi. All analyses were done in triplicates.

Table 1 shows the composition of commercial and locally made complementary foods used in this study as shown on the labels.

Table 1: Commercial and local complementary food used in this study.

complementaryfoods	composition
CC1	rice and milk powder
CC2	maize and milk powder
CC3	maize grits
LSG	Sorghum paste ('ogi')
LMT	Millet paste ('ogi')
LMZ	Maize paste('ogi')

KEY:
CC1 = Commercial Complementary Food 1
CC2 = Commercial Complementary Food 2
CC3 = Commercial Complementary Food 3
LSG = Local complementary food sorghum
LMT = Local complementary food millet
LMZ = Local complementary food maize

METHODS

Preparation of Samples

30gm each of 'Ogi' or 'akamu'- cereal paste of sorghum, millet, and maize was kept in sterile containers and labeled, kept for probiotic isolation. Each commercial complementary food was measured out (30gm) in sterile containers,labeled and kept in samples sterile containers.

Preparation of Media for probiotic isolation.

Preparation of media for probiotic isolation was done according to method of De Man (16).
The solution was distributed into sterile petri dishes kept in insulated cupboard.

Inoculation of Samples

Serial dilution of the samples were performed from 10^{-1} – 10^{-5} according to theproceedure of De Man (16). 1ml of the appropriate sample dilution was transferred to the dry Petri dish containing the rogosa Agar. The plate was incubated at 37^{0}c for one week after which the result of the growth was counted.

Sample Staining Technique and Colony count.

Sample staining technique using oil immersion lens as described by De Manrogosa(16) was used. Colony count was done using the illuminated colony counter and tally counter as described by Adams (17). Fungi identification was carried out by macroscopic and microscopic examination according to Glover (28).

Statistical Analysis

All the analyses were conducted in triplicate. Data was analyzed using Statistix 9, version 9.1(2012) statistical package for mean and standard deviation.

RESULTS

Table 2: shows the isolated bacteria species and bacterial count of colony forming unit per gramme (cfu/g) of commercial and local complementary foods. Commercial complementary foods CC1 and CC2 had *Lactobacillus* species with bacteria count of 7.5 x 10^2 and 8.7 x 10^2 Cfu / g respectively while CC3 had no bacterial specie isolated. Local complementary food LSG had no probiotic bacteria isolated while LMT and LMZ had *Lactobacillus* species isolated with bacteria count of 5.4 x 10^2 and 6.5 x 10^2 (Cfu/g) respectively.

Table 2: Isolated bacteria species and bacterial count (cfu/g) of the samples.

Complementary foods	Probioticbacteriaisolated	Bacteriacount (Cfu/g)
CC1	Lactobacillus spp	7.5 x 10^2 ±10.00
CC2	Lactobacillus spp	8.7 x 10^2 ±10.00
CC3	Nill	Nill± 0.00
LSG	Nill	Nill± 0.00
LMT	Lactobacillus spp	5.4 x 10^2 ±9.00
LMZ	Lactobacillus spp	6.5 x 10^2 ±10.00

Mean SD, n= 3

KEY:

CC1 = Commercial Complementary Food 1
CC2 = Commercial Complementary Food 2
CC3 = Commercial Complementary Food 3
LSG = Local complementary food sorghum
LMT = Local complementary food millet
LMZ = Local complementary food maize
Nill = Not Identified
Cfu/g = colony forming unit per gramme

1.01 x 10^3 (Cfu / g) of *Saccharomycescerevasiae*. LMT had 6.06 x 10^2Cfu /gandLMZhad the highest yeast count of9.26 x 10^2 (Cfu / g) of *Saccharomycescerevasiae.*

Table 3 shows the isolated yeast and yeast count (Cfu/g) of commercial and local complementary foods used in the study. Commercial complementary foods CC1, CC2 and CC3 had no yeast isolated. Local complementary food LSG had the least yeast count of

Table 3: Isolated yeast and yeast count (Cfu / g) of the samples

Complementary food	Yeast isolated	Yeast count Cfu/g
CC1	Nil	Nil± 0.00
CC2	Nil	Nil± 0.00
CC3	Nil	Nil± 0.00
LSG	Saccharomyces*cerevasiae*	1.01 x 10^3Cfu/g± 6.00
LMT	*Saccharomycescerevasiae*	6.06 x 10^2Cfu/g± 1.00
LMZ	*Sacrcharomycesc erevasiae*	9.26 x 10^2Cfu/g±1.00

Mean SD, n= 3

KEY:

CC1 = Commercial Complementary Food 1
CC2 = Commercial Complementary Food 2
CC3 = Commercial Complementary Food 3
LSG = Local complementary food sorghum
LMT = Local complementary food millet
LMZ = Local complementary food maize
Nill = Not Identified
Cfu/g = colony forming unit per gramme

Discussion

Lactobacillus spp and *saccharomyces cerevisiae* are the organisms Table 2 and 3) isolated from the local complementary foods. This connotes that *Lactobacillusspp* and *saccharomycescerevisiae* might be responsible majorly for the sour taste of 'ogi' that resembles that of yoghurt. Odunmfa*et al.,* (18) identified *lactobacillus* isolates as an important micro flora of African fermented foods and reported *ogi* as a good source of probiotics and bacteriocin. Also Odunmfa*et al.* (18) reported that a special type of *ogi* was able to stop the growth of diarrhoea causing bacteria. Commercial products CC1 and CC2 had higher number of bacterial count than CC3 (Table 2) and all locally made complementary foods used, probably because of the milk content in CC1 and CC2 (Table 1). Milk serves as a good substrate for probiotic bacteria growth (19), this is shown in the study as the complementary foods containing milk had more probiotic bacteria count than all others both local and commercial. According to Momka (5) not all micro organisms of *ogi* fermentation are always found in all fermentations as shown in this work, Sorghum had the least yeast count and no bacteria count while maize had the highest bacteria count (6.5 x 10^2Cfu/g) and yeast count (9.26 x 10^2Cfu / g) (Tables 1 and 2). There is significant difference between probiotic content of Sorghum and the other cereal pastes at $p \leq 0.05$. Sorghum is known to have poor nutritional quality which may have affected the probiotic bacteria growth. All the locally made complementary foods contain yeast probiotics (Table 2), while the commercial foods had no yeast growth (Table 1), probably because of the fermentation process used in the local production. Fermentation has been reported as the main process of probiotic delivery in cereal foods (20). Most cereals are good sources of all nutrients beside their prebiotic content. These constituents make cereals a suitable medium for probiotic bacteria growth (20). However the commercial complementary foods had higher bacteria strength probably because of their milk component which serves as a good substrate for growth of probiotic bacteria (21). The buffering capacity of milk helps to improve the survival of probiotic flora in the GI tract also (21), yet many researches reports that fermented foods remain the main vehicle to deliver probiotic bacteria (9, 22). Even though there is no specified level of probiotic bacteria in foods that would guarantee biological activity, it is increasingly recommended to ingest 10^8 CFU / day (23). All the complementary foods used in this study contain low values of probiotic bacteria when compared with the 10^8 recommendation; however they can still help to maintain homeostasis of the infants (24). Nestle (25) reported that some complementary foods contain probiotics (lactic acid bacteria) similar to those found in the digestive system of breast fed babies; result of this research supports this assertion. All the samples except LSM contain lactic acid bacteria (*Lactobacillus* spp.).

Although the commercial complementary food products contain milk and therefore had more probiotic bacteria content (CC1, CC2), they had no yeast growth to make a balanced gastrointestinal microbiota. Fuller (10) reported that food scontaining mixed culture of probiotic microbes improves the health of the host by improving intestinal microbial balance. The local complementary foods made of whole cereals are known good sources of non-digestible carbohydrates that besides promoting several beneficial photochemical effects can act as prebiotics that selectively promotes the growth of lactobacilli and bifidobacteria in the colon of the babies (26).Whole grains are known to be good sources of many beneficial phytochemicals and therefore beneficial to infants nutrition (27). Fermented cereals, as a result of the

fermentation process used in producing local complementary foods in this study might have made more available the nutrients for probiotic bacteria and yeast growth as shown in Tables 1 and 2. Glover (28) also reported isolating *saccharomyces cerevisiae* frompito a fermented cereal product in various parts of Ghana.

CONCLUSION

The local complementary foods used in this study contained mixed probiotics more than the commercial foods($p < 0.05$). More studies are needed for other types of complementary foods containing cereals and legumes used in complementary feeding.

REFERENCES

1 Macance, J.I. (2011). National Center for Compementary and Alternative Medicine. Available at nccam.nih.gov/health / probiotics accessed, July 12, 2011.

2 Gallant,R. (1989). Probiotics in man and animals. *J.APPL.Bacteriol*. 66(5):365-378.

3 Didari, T. and Mozaffari S. (2014). A systematic review of the safety of probiotics. Expert opinion on drug safety, 13 (2): 227-239.

4 FAO, (2012). Food Agriculture Organization. Working group on drafting guidelines for the evaluation of probiotics in foods, London, Ontario, Canada.

5 Momka, K. (2014). Complementary feeding of young children in developing countries; age of introduction of complementary foods and the weaning dilemma: Geneva, World Health Organization.

6 Sanders, M.E and Huis,V.J.(1999). Bringing a Probiotic- containing food to the Market ; Microbiological, Product, Regulatory and Labelling Issues. Antoine Vanleehook, 76, 293-315.

7 Gilliland, S.E.(1990). Health and Nutritional benefits from lactic acid bacteria. FEMS Micro.Rev.*:* 87, 175-188.

8 Oswari, H., Bisgaard, H.,Bomehykke, K., Chaves, B.L., Stor T. and Poludan-Muller, G.(2013). Reduced diversity of the intestinal microbiota during infancy is associated with increased risk of allergic diseases and malnutrition: J. Nutr; 13: 236 – 356.

9 Guarno, F. andSchoafsmaG.J.(1998). Probiotics in complementary food. Inter. J. of FdMicrob.; 39, 237-238.

10 Fuller, R.(1996). Probiotics in man and animals .J.APPL.Bacteriol. 66 (5): 365-78.

11 Hamilton-Miller, J.M. (2003). The role of probiotics in the treatment and prevention of Helicobacterpylori infection. Inter.J.of Antimicrobial Agents, 22(4), 360 -366.

12 Okida, F.(2015). Infant and young child feeding . Availablehttp://friso.com.mx/en/product-friso-detail/15/friso-gold-3?p=1&id-fag=14(date assessed 25/8/2017).

13 Maya, P. and Stanton, C. (2007). Probiotic bacteria. Legistrative Framework-Requirement, Evidence Basis; Ameri.J.Nutr., 137, 3850-853.

14 Odunfa, H. Helland M.H. Nardu J. (2007). Growth and metabolism of probiotics in fermented non-alcoholic cereal based foods. Int. J. of food Microbiol: 87, 17-27.

15 Ihekoronye, A.I.andNgoddy, P.O. (1985). Integrated Food Science and Technology for Tropical Schools, Ltd. London, Macmillan Publisher.

16 DeMan,J.C., Rogosa, M and Share, M.E (1960). A medium for cultivation of Lactobacilli. *J.APPL.Bacteriol*. 45(3):163-243.

17 Adams, M.R. and Moss, M.O.(1999). Food Microbiology. Royal Society of Chemistry, Cambridge, 257-260.

18 Odunfa, H. Helland M.H. Nardu J. (1997). Growth and metabolism of probiotics in fermented non-alcoholic cereal based foods. Int. J. of food microbiol: 87, 17-27.

19 Laurens-Hattingh A. Viligoen B.C. (2001). Yoghurt as probiotic characteristic and starter organisms. J. of clinical nutr. (suppl) 3745-3795.

20 Blandino, A.-Aeeri M.E., Pandilla, S.S., Cantero, D., Webb,C.(2003). Cereal based fermented foods and beverages: Rev. Food Research Inter: 36, 527-543.

21 Farnworth, E.R. (2007). Growth of probiotic bacteria and Bifidobacteria in a Soy Yoghurt Formulation. *Int. J. food Microbiol*. (116), 174-181

22 Parvez, S.(2006). Probiotics and their fermented Food products are beneficial for Health.-Review. *J.of APPL. Microbio.* 100 1171-1185.

23 FAO/WHO (2002). Working Group On Drafting Guidelines For The Evaluation Of Probiotics In Food , London, Ontario, Canada.

24 Jean,P.(2015). Probiotic channels. Available frisoGold3enAmazon.Com.mx/Ahorraenfr isogold3date assessed 25/8/2017).

25 Deabo,P.(2016). Nestle cereals.Available atwww.nestle-cwa./en/brands / baby foods / cerelac 24/8/2017).

26 Probiotics and prebiotics- Progress and challenges. *Int. Dairy Journal* 18, 969-975.

27 FAO / WHO (2001). Health and nutritional properties of probiotics in food including powder milk with live lactic acid bacteria. Argentina; FAO of the United Nations and world Health Organization,Expert Consultation Report.

28 Glover, R.K. and Abaido,R.C.(2005). Biodiversity of Sacchromycescerevisiae isolated from a surver of 'pito' production sites invarious parts of Ghana. Systm.andAppl.Microbiol. 28(8): 755-76