International Journal of Food and Nutritional Science

e-ISSN 2320 -7876 www.ijfans.com
Vol. 7, No. 1, January 2018

Research Paper ***Open Access***

FOOD POISONING BY BACILLUS CEREUS

H Lalawmpuii[1]*, A Lalruatdiki[1], Losa Rose[1], Krishnakanth Reddy[1] and Lalrengpuii Sailo[1]

*Corresponding Author: H Lalawmpuii, ✉ opihnialum@gmail.com

Received on: 1st May, 2017 Accepted on: 11th October, 2017

Bacillus cereus is a Gram positive bacteria, found in a wide variety of food of plant and animal origin. They are normally non-pathogenic but may cause illness when food improperly cooked or stored are consumed. The illness is usually manifested in two forms: emetic syndrome and diarrheal syndrome. The food poisoning caused by *Bacillus cereus* are usually underreported as the disease is self-limiting and usually last for less than 24 hours. Adopting necessary precautions and proper heating of food is imperative for the prevention of the illness caused by *Bacillus cereus*.

Keywords: *Bacillus cereus*, Emetic toxins, Enterotoxins, Food poisoning

INTRODUCTION

Bacillus cereus is a Gram-positive aerobic or facultatively anaerobic, motile, spore-forming, rod-shaped bacterium known to cause illness due to consumption of improperly cooked food. They are commonly found in soil and are naturally present in a wide range of food products both of plant and animal origin.

They belong to the genus *Bacillus*, first described and classified by Ferdinand Cohn in 1982. The genus Bacillus has more than 60 species most of which are considered non-pathogenic. *Bacillus cereus* have close phenotypic and genetic (16S rRNA) relationships to several other *Bacillus* species, especially *anthracis*. The spores of *Bacillus cereus* are resistant to heat, cold, radiation, desiccation and disinfectants.

They can grow in food within a wide range of temperature and pH with the optimum being 30-40 °C and 6 to 7 pH, respectively. As mentioned earlier they are normal component of microflora of foods such as meat, milk, rice, vegetables, etc. Although their presence is seen in a wide range of food, they normally do not impose health risk but cause illness when food improperly cooked or stored are consumed.

TOXINS PRODUCED BY BACILLUS CEREUS AND THEIR ASSOCIATED SYMPTOMS

The food poisoning due to *Bacillus cereus* are underreported as the illness is mild and self-limiting and usually last for less than 24 hours (Granum and Lund, 1997). Two type of food poisoning are reported: Diarrhoel type and emetic type. A special surface structure of *B. cereus* cells, the S-layer, has a significant role in the adhesion to host cells, in phagocytosis and in increased radiation resistance (Kotiranta, 2000). Other potential virulence factors include secreted phospholipases, haemolysins, proteases, and other degradative enzymes. *Bacillus cereus* produces one emetic toxin (ETE) and three different enterotoxins: HBL, Nhe and CytK.

Emetic toxin (ETE) is a ring-shaped structure of three repeats of four amino acids with a molecular weight of 1.2 kDa. It is highly resistant to pH between 2 and 11, to heat, and to proteolytic cleavage. It is not antigenic but stimulate the vagus afferent by binding to 5-HT3. Cereulide dose of

[1] Department of Livestock Products Technology, COVS, GADAVASU, Ludhiana 141004.

approximately 9.5 µg/kg BW is required to cause onset of emetic syndrome (Finlay *et al.*, 1999). The emetic toxin is pre formed in the food such as rice, pasta, pastry and noodles and is characterized by relatively short incubation period of 0.5 to 5 hours. Symptoms of the illness include nausea, vomiting and malaise and last for less than 24 hours.

Two of the three enterotoxins are involved in food poisoning which are produced within the host body. They both consist of three different protein subunits that act together. HBL the primary virulence factor is also a hemolysin. Nhe is not a hemolysin but is the most common while CytK is a single component protein, not involved in food poisoning. All three enterotoxins are cytotoxic and cell membrane active toxins that will make holes or channels in membranes. The infective dose to cause diarrhoel symptoms is about 10^5 to 10^7 cells. The incubation period is comparatively longer than the emetic type, i.e., about 8-16 hours. Consumption of improperly cooked meat products, milk, fish, vegetables, soup, etc., cause the illness characterized by abdominal pain, watery diarrhea and occasional nausea.

NON GASTRO-INTESTINAL INFECTIONS

Bacillus cereus is also known to cause non gastrointestinal infections like endopthlamitis, respiratory tract infections, CNS infection, gas gangrene like infection, cutaneous infection, endocarditis, osteomyelitis and urinary tract infection. *B. cereus* produces a potent β-lactamase conferring marked resistance to β-lactam antibiotics (Bottone, 2010).

DETECTION AND ISOLATION

Isolation of *Bacillus cereus* in selective agar can be done from suspected food, faeces and vomitus of the patients. Advance techniques like multiplex PCR, MLST, AFLP and LAMP can also be used for its detection. Detection of the toxins can be done using cell culture (vero cells) where it produces vacoulation activity, ELISA, RPLA and bioassay such as Boar Spermatozoa Test. Further, test kits for their detection are also available commercially (Bacillus Diarrhoeal Enterotoxin Visual Immunoassay (TECRA International Pty Ltd., Australia and BCET-RPLA Toxin Detection Kit (Oxoid).

PREVENTION AND CONTROL

Since the organism is commonly found in food of plant and animal origin, it is imperative that they are destroyed by proper heating. Food if not consumed immediately, should be chilled or hold at appropriate temperature to prevent the spore germination and multiplication of the vegetative cells. In any case, holding the food at room temperature for extended period should be avoided.

REFERENCES

- Bottone E J (2010), "*Bacillus cereus*, a Volatile Human Pathogen", *Clin Microbiol Rev.*, Vol. 23, No. 2, pp. 382-398, doi: 10.1128/CMR.00073-09.
- Finlay W J J, Logan N A and Sutherland A D (2000), "*Bacillus cereus* Produces Most Emetic Toxin at Lower Temperatures", *Letters in Applied Microbiology*, Vol. 31, pp. 385-389, doi:10.1046/j.1472-765x.2000.00835.x.
- Granum P E and Lund T (1997), "*Bacillus cereus* and its Food Poisoning Toxins", *FEMS Microbiol Lett.*, Vol. 157, No. 2, pp. 223-228.
- https://www.fda.gov/food/foodborneillness contaminants/causesofillnessbadbugbook/ ucm070492.htm
- Kotiranta A, Lounatmaa K and Haapasalo M (2000), "Epidemiology and Pathogenesis of *Bacillus cereus* Infections", *Microbes Infect.*, Vol. 2, No. 2, pp. 189-198.

IJFANS
International Journal of Food
And Nutritional Sciences

IJFANS
International Journal of Food And Nutritional Sciences

Volume 7, Issue 1, January 2018, www.ijfans.com e-ISSN: 2320-7876

INTERNATIONAL JOURNAL OF FOOD AND NUTRITIONAL SCIENCES

IMPACT FACTOR ~ 1.021

Official Journal of IIFANS

IJFANS International Journal of Food and Nutritional Science

e-ISSN 2320 -7876 www.ijfans.com
Vol. 7, No. 1, January 2018

Research Paper *Open Access*

NUTRITIONAL COMPOSITIONS OF MILLET-BASED WEANING FOOD SUITABLE FOR INFANTS UNDER THE AGE GROUP OF 6-12 MONTHS

P Nazni[1]*

*Corresponding Author: P Nazni, ✉ naznip@gmail.com

Received on: 12th November, 2017 Accepted on: 15th December, 2017

Weaning is the process of gradually introducing an infant mammal to what will be its adult diet and withdrawing the supply of its mother's milk. Objective of the study is to find the proximate, mineral, amino acid and anti-nutritional composition of formulated weaning mixes. In this study, four types of weaning food based on barnyard millet flour (21-30%), defatted soy flour (18%), sugar (52.5-58.55) and skim milk powder (49.5-52.5%) was prepared based its standard procedure. The developed complementary mixes were analysed for its proximate, mineral, amino acid and anti-nutrient compositions based on its standard procedure. The results of the study revealed that, mix 1 and mix 4 had lower (1.02 and 1.2) moisture content than mix 2 and mix 3 but which were significantly higher moisture content. Highest protein content was observed in mix 3 (18.90 g/100 g) and mix 4 showed high carbohydrate of 77.25 g/100 g than mix 1 and mix 2 combinations. Highest sodium, potassium, calcium, magnesium and phosphorus was observed in mix 4. Mix 4 showed high manganese content 16.49 mg/100 g, high iron 21.20 mg/100 g, zinc with high level of 6.75 mg/100 g and copper 15.51 mg/100 g. Regarding amino acid compositions, mix 4 (30% Barnyard millet flour, 18% defatted soya flour, 52.5% of sugar and 49.5% of skim milk) showed highest value of isoleucine, leucine, lysine, cystine, methionine, tyrosine, phenylalanine, theonine, valine, histidine, arginine, glutamic acid, serine, proline, glysine and alanine. Conclusion of the study revealed that, nutritionally the formulated samples were better than a commercial complementary food in terms of proximate, minerals and amino acid compositions.

Keywords: Barnyard millet, Soy flour, Amino acid, Anti-nutrients

INTRODUCTION

Nutritional wellbeing is a sustainable force for health and development and maximization of human genetic potential. The nutritional status of a community has therefore been recognized as an important indicator of national development. In other words, malnutrition is an impediment in national development and hence assumes the status of national problem. For solving the problem of deep-rooted food insecurity and malnutrition, dietary quality should be taken into consideration. Diversification of food production must be encouraged both at national and household level in tandem with increasing yields. Growing of traditional food crops suitable for the area is one of the possible potential successful approaches for improving household food security.

The importance of balanced weaning supplements for promoting children growth and wellbeing is well recognized. Growth and development of infants and children towards mature adult stage is the result of number of competing, complementary, and interacting influences. The most important influence is the supply of sufficient nutrients of adequate quantity and quality. The economically affluent and elite generally meet growing infants needs through commercially standardized foods. These foods are excellent

[1] Department of Clinical Nutrition and Dietetics, Periyar University, Salem-11.

and meet the nutritional requirements of young children in both developed and developing countries. However, the products are too expensive for the target groups in developing countries.

In India, around 63% of the children under five years of age are malnourished. This accounts for 75 million malnourished children and is one of the worst levels in the world (Rhode, 1994). Nutritionally inferior diets and improper feeding practices are major contributing factors to the development of childhood malnutrition (Huffman and Martin, 1994). Many attempts have been made by government, international organizations and commercial enterprises to manufacture and market a formula that provides a balanced weaning food for the child (WHO, 1998; and SPCFA, 2000). However, these formulae have proved too expensive to be used by those within the range of very poor (Chandrasekhar *et al.*, 1988).

Millets also referred to as coarse cereals are a variety of small edible grasses belonging to the grass family (*Gramineae/Paniceae*). These are distributed in about 10 genera and 20 species in all (Lupien, 1990). Millet is a collective term referring to a number of small seeded annual grasses that are cultivated as grain crops, primarily on marginal lands in dry areas in temperate, subtropical and tropical regions (Baker, 1996). The millets include five genera of the *Panaceae* family (*Panicum, Setaria, Echinochloa, Pennesetum and Eleusine*). The most important cultivated species are: Proso millet (*Panicum miliaceum*), Foxtail millet (*Setaria italica*), Japanese barnyard millet (*Echinochloa frumentacea*), Finger millet (*Eleusine coracana*) and Koda millet (*Paspalum scrobiculatum*).

Low-cost, high protein food supplement development for weaning infants is a constant challenge (Schmidt, 1983). This is particularly important in developing countries where malnutrition problems are still common particularly during weaning. In African countries weaning can be a period of problems and vulnerability to the survival of a child, where traditional foods are characterized by the low nutrient density and high bulk. The baby is either weaned directly onto the family diet early in life or continues to be breast-fed, with sub optimal nutrient intake. In either case, the nutrition of a child suffers and optimum growth cannot be ensured. So, the introduction of traditional semi-solid foods to infants in developing countries is unsatisfactory in timing, and the quality and quantity of the food is insufficient. The effect on children is growth flattering and the result is malnutrition disease later. Moreover, the high prices of proprietary weaning food and of vegetable and animal protein and the non-availability of nutritious foods, combined with faulty feeding practices and late introduction of supplementary food, are all aggravating the disorder.

Protein-energy-malnutrition generally occurs during the crucial transitional phase when children are weaned from liquid to semi-solid foods. During this period, because of their rapid growth, children need nutritionally balanced calorie-dense supplementary foods in addition to the mother milk (Cameroon and Hofvander, 1971; and Berggren, 1982). Severe protein energy malnutrition results in kwashiorkor and marasmus, while inadequate growth or stunting produced as a result of poor supplementation is described as hidden malnutrition. This is because the child may appear healthy while being severely malnourished. The effects of protein-calorie malnutrition on morbidity and mortality among infants in under-privileged socio-economic groups have been well recognized. Impaired physical growth and mental development have been scientifically verified (Davidson and Passmore, 1986).

MATERIALS AND METHODS

Procurement and Preparation of Raw Materials

Barnyard millet was procured from Tamilnadu Agricultural University, Coimbatore. The millets were cleaned properly, dried and stored cleanly for further use. Major ingredients such as defatted soya flour, sugar and skim milk powder were procured from local market.

Processing of Barnyard Millet

The selected barnyard Millet was soaked for 24 hours, steamed for 20 minutes and dried. By using the roller, the millets were polished and its outer layer was removed. The cleaned grains were pulverized using a plate mill to obtain whole flour, which was be further sieved through a 44-mesh sieve (BSS). The obtained flour was stored in an airtight container until further use.

Malting

Defatted Soya flour was steamed for 20 minutes, dried and powdered and packed in air tight container for further use. Sugar and Skim milk powder was purchased from the local market checked for its purity and stored until further use.

Blend Formulations

The blends were prepared or mixed from the individual flour ingredients in the percentages or proportions as shown in

Table 1: Composition of Blends

Mixes	Processed Barnyard Millet Flour	Defatted Soy Flour (20 min Steamed)	Sugar	Skim Milk Powder
Mix 1	21	18	58.5	52.5
Mix 2	24	18	55.5	52.5
Mix 3	27	18	54	51
Mix 4	30	18	52.5	49.5

Table 1. The developed composite mixes nutrient, amino acid, fatty acid, minerals and anti-nutrient properties were analysed using standard procedure.

Proximate Analysis

Proximate Analysis Standard procedures of AOAC were used to determine the moisture content, crude fat, crude protein, ash and the total carbohydrates have been obtained by difference (AOAC, 1990). Energy value was calculated using the Atwater's conversion factors (Spackman *et al.*, 1958; and Harper, 2003).

Minerals Analysis

Minerals such as potassium and sodium content, calcium, magnesium, manganese, iron, zinc and copper were determined by Atomic Absorption Spectrophotometer, Hitachi Model 180-80, and Ion Chromatographic Analyzer ICA model IC 100 (AOAC, 2000).

Amino Acid Composition

The developed samples amino acid was measured on hydrolysates using amino acid analyzer Sykam-S7130 based on high performance liquid chromatography technique.

Anti-Nutritional Parameters

The anti-nutrients such as saponin, tannin, alkaloid, flavonoid, cyanogenic glycoside. Oxalate, phytate, trypsin inhibitor and haemagglutinin were determined by the modified Vanillin assay (Butler *et al.*, 1982).

Statistical Analysis

Data obtained were analysed for its mean ± standard deviation. Duncan multiple range test was used to compare the differences between the means. Significance was accepted at $p \leq 0.05$.

RESULTS AND DISCUSSION

The developed complementary feeding mixes nutritional parameters were discussed with the following headings.

Proximate Analysis of the Developed Complementary Mixes

The proximate analysis of the developed complementary mixes was shown in Table 2.

Table 2 shows result of proximate composition of the developed complementary foods. The moisture (dry basis), carbohydrates, protein, fat and ash g/100g ranged from 1.02-1.2%, 74.56 -77.25 g/100 g, 16.78-18.90 g/100 g, 1.59-2.83 g/100 g and 3.18-3.42 g/100 g respectively. Mix 1 and mix 4 had lower (1.02 and 1.2) moisture content than mix 2 and mix 3 but which were significantly higher moisture content. Moisture content of food is an important index of their susceptibility to microbial spoilage. When the moisture content is on the high side, it encourages the growth of microorganisms. Moisture content would therefore indicate low growth of bacteria and fungi (Temple *et al.*, 1996). This thus predisposes such food to degradation and enhances its perishability.

The recommended protein content (grams of protein per 100 kcal of food) for complementary foods is of 0.7 g/ 100 kcal, from 5 to 24 months. In most countries, the protein requirements of infants are met when the energy intake is appropriate, except if there is a predominant intake of low-protein foods (WHO/UNICEF, 1998). In this present study, highest protein content was observed in mix 3 (18.90 g/100 g) than mix 1 (18.58 g/100 g) and mix 2 18.11 g/100 g. According to the protein Advisory Group, guidelines for weaning foods should be 20% of proteins, fat levels of up to 10%, moisture content 5% to 10% and total ash content not more than 5% (FAO/WHO/UNICEF/Protein Advisory Group (PAG), 2007). Diets with this high content of protein can be helpful not only for weaning children but also for children already suffering from PEM.

Among the four mixes, mix 4 showed high carbohydrate of 77.25 g/100 g than mix 1 and mix 2 combinations. For infants with an average breast milk intake and who eat at least three meals a day containing