RESEARCH ARTICLE

AN ANALYSIS OF THE PROBIOTIC BACTERIA IN THE COMMONLY FERMENTED MAIZE, OIL BEAN, AND CASTOR OIL IN NIGERIA

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Abstract

Active probiotic organisms are essential bacteria considered live microorganisms obtained from fermented foods. There is increasing evidence that probiotics are necessary for human health. This study aimed to isolate and characterize the active probiotic microorganisms in Nigeria's commonly fermented food samples. Maize, castor oil, and African oil beans were the primary fermentable food samples used in the study. The result revealed the presence of active probiotic organisms such as Pediococcus, Micrococcus, Lactobacillus, and Bacillus species. The study concluded that these organisms are responsible for the fermentation of carbohydrates and protein-rich seeds.

Introduction:

The term Probiotics has been widely used to describe specific biological agents, generally consumed as food supplements, which can positively impact the microbial ecology of the host. Although the exact definition of probiotics continues to evolve, however, they are commonly referred to as live microorganisms with favorable health effects on the host when consumed in adequate amounts (Barbara et al., 2018; Das et al., 2020; Gasbarrini et al., 2016; Gasilina & Belmer, 2015; Haghshenas et al., 2017; Hussein et al., 2021; Jackson & Lovegrove, 2012; Orel, 2013; Salisu, 2014). Nonetheless, the description of probiotics is centered on live microbes, which contrasts to prebiotics that are nondigestible food ingredients. The probiotic bacteria generally belong to the Lactobacillus and Bifidobacterium genera (Rossi et al., 2011; Saarela et al., 2002). Evidence abounds that suggest that the consumption of probiotics favorably influences numerous parts of the innate nonspecific immune system, such as the promotion of mucin production, inhibition of pathogenic bacteria, decrease in gut permeability, macrophage activation, and phagocytic capacity.

Active probiotic organisms are essential microorganisms generally regarded as live microbes found in fermented foods, which enhance the digestive tract and promote the immune system. Probiotics also influence carbohydrate and protein fermentation, enriching microbiota with high saccharolytic activity and low proteolytic activity. Probiotics have attracted huge research attention in the recent decade (Anukam & Reid, 2007; Barba-Vidal et al., 2019; Bronn et al., 2013; Coman et al., 2019; de Melo Perreira et al., 2018; Dowarah et al., 2018; Guemonde & Salminen, 2006; Holvoet et al., 2013; Shokryazdan et al., 2017; Shinde, 2012). Similarly, substantial developments have been recorded relating to the classification of probiotics and the authentication of health-related implications linked with adequate intake of probiotics (Akter et al., 2020; Amin et al., 2020; Beenaat et al., 2012; Bharti et al., 2020; Fernandez & Marette, 2017; Hewadmal & Jangra, 2019; Holvoet et al., 2013; Klerebezem et al., 2019; Park et al., 2014; Shah, 2000). Research has underscored numerous mechanisms of action of probiotics.
relating to prevention and treatment of several diseases, such as antimicrobial activity and suppression of bacterial growth, enhancement of barrier activity, immunomodulation, suppression of human T-cell proliferation, and initiation of an immune response (Peluso et al., 2007; Resta-Lenert & Barrett, 2003; Rioux & Fedorak, 2006).

Maize, African oil bean, and castor oil are among the commonly fermented food crops in Nigeria that has been extensively emphasized in the literature (Abdoulaye et al., 2018; Njoh et al., 2017; Adekoya et al., 2017; Adiaha, 2017; Akande et al., 2012; Olasupo et al., 2016). The fermentation process describes the microbial breakdown of carbohydrates and other substances to release alcohol, carbon dioxide, and energy. The fermentation processes play an essential role in food technology industries. In the traditional fermentation process, natural microorganisms are employed to prepare and preserve different types of food. The methods add to the nutritional value of foods and enhance flavor and other desirable qualities associated with digestibility and edibility. Fermented foods reflect foods that have been subjected to the action of microorganisms or enzymes to achieve desirable biochemical changes cause significant modification to the food (Singh et al., 2012).

The frequently fermented food in Nigeria includes cereals (Maize), beverages (palm wine), legumes (oil bean, castor oil seed), and tubers (cassava). This investigation is aimed to isolate and characterize the active probiotic organisms existing in fermented maize (Zea mays), oil bean (Ricinus communis), and African oil bean (Pentadethra macrophylla Benth).

Materials and Method:
Glassware and other materials were sterilized correctly and dried. The traditional fermented maize, oil bean, and castor oil seed were collected from different sources and subjected to laboratory analysis. According to Da Silva et al. (2013), all media were prepared according to the standard procedures. Furthermore, each fermented food sample’s grams were mashed with laboratory pestle and mortar and mixed with purified water as a diluent in a sterile sample bottle. Nevertheless, characterization and identification of isolates, Gram staining of the isolates, biochemical test for identification of bacteria, catalase test, citrate test, indole test, motility test, methyl red test, plaiting, and sugar fermentation were appropriately employed.

Results:

Table 1: Viable colony count of bacteria isolated from the fermented food samples.

| Plates | Samples            | Nutrient Agar | De Man Rogosa Sharp (MRS) |
|--------|--------------------|---------------|---------------------------|
| 1      | Zea mays           | 1.6x10^6      | 2.2x10^6                  |
| 2      | Zea mays           | 4.0x10^5      | 1.12x10^6                 |
| 3      | Pentadethra macrophylla benth | 1.6x10^5 | 1.04x10^6                  |
| 4      | Pentadethra macrophylla benth | 1.8x10^6 | 2.2x10^6                  |
| 5      | Ricinus communis   | 1.4x10^6      | 2.68x10^6                 |
| 6      | Ricinus communis   | 1.08x10^6     | 2.40x10^6                 |

Table 2: Morphology Characteristics of Isolates.

| Plate | Sample name | M e d i a | M o r p h o l o g y |
|-------|-------------|----------|-----------------|
| 1     | Z. m a y s 1 | Nutrient | Cream, smooth, circular, small, flat, entire, transparent |
|       | Z. m a y s 2 | Nutrient Mrs. | p u n c t i f o r | r m |
|       | Z. m a y s 3 | M r s . | Cream, smooth, circular, moderate, flat, unulate, transparent filamentous small. |
|       | Z. m a y s 2 | Nutrient | Cream, smooth, circular, big, flat, entire transparent white spindle, moderate amber small punctiform |
|       | P. macrophylla benth 1 | Nutrient | Cream, smooth, circular, big, flat, entire transparent white spindle, moderate amber small punctiform |
|       | P. macrophylla benth 2 | M r s | Cream, smooth, circular, big, flat, entire transparent irregular small punctiform |
|       | R. communis 1 | Nutrient | Cream, smooth, circular, small, flat entire transparent irregular small. |
|       | R. communis 2 | M r s | Cream, smooth, circular, small, flat, entire transparent irregular small. |
|       | R. communis 1 | M r s | Cream, smooth, circular, small, flat, entire transparent irregular small. |
Table 3:- Biochemical Test on the Isolate.

| Sample name | Glu | Lau | Suc | Fru | Mann | Indole | Catalase | Citrate | Motility | Oxidase | Methyl red | Gram reaction | Presumptive isolated organisms |
|-------------|-----|-----|-----|-----|------|--------|----------|---------|----------|---------|------------|---------------|-------------------------------|
| *Z. m., R. c* | AG  | A   | A G | AG  | AG   | -      | -        | +       | -        | -       | -          | +ve, rods      | Lactobacillus sp            |
| *R. communis* | AG  | A   | A G | AG  | AG   | -      | +        | -       | -        | -       | -          | +ve, cocci     | Micrococcus sp           |
| *P. macrophylla bent, R.c* | A   | A   | A   | A   | A    | -      | +        | +       | -        | -       | -          | +ve, long rod  | Bacillus sp             |
| *Z. m. mays*   | A   | A G | AG  | AG  | AG   | -      | -        | +       | -        | -       | -          | +ve, cocci short chains | Pediococcus sp         |
| *Z. m. mays*   | AG  | A   | AG  | AG  | AG   | -      | +        | -       | -        | -       | -          | +ve, rod        | Lactobacillus sp         |
| *P. macrophylla bent* | AG  | A   | A G | AG  | AG   | -      | +        | -       | +        | +       | -          | +ve, cocci single | Micrococcus sp          |
| *Z. m. mays*   | A   | A G | AG  | AG  | AG   | -      | +        | -       | -        | -       | -          | +ve, rod single and cluster | Lactobacillus sp        |

Note: + = positive, - = Negative, A = Acid, AG = Acid gas, Glu = Glucose, G = Gas, Lac = Lactose, Fru = Fructose, Suc = Sucrose, Mann = Mannitol, Z. m = Z. mays, R. c = Ricinus communis.

Discussion:-

The present study intends to isolate and characterize the probiotic bacteria in some fermented food samples. The research shows the microorganisms isolated from the fermented samples as the bacteria grow on the de Man Rogosa Sharpe (MRS) agar and nutrient agar. The viable colony count of bacteria isolated from the fermented foods, maize, castor oil, and oil bean are shown in Table 1. Indeed, the finding agrees with the standard plate count of colony range of 30-300 cfu on a petri-dish. The bacteria isolated from maize ranges from 1.6x10^5 to 2.28x10^6, oil bean from 1.6x10^5 to 2.2x10^6, and castor oil goes from 1.4x10^5 to 2.68x10^6. The bacteriological characteristics of the bacteria colonies were acknowledged through visual counting from the plates comprising the aliquot dilute samples of each fermented food sample, as shown in Table 2. Table 3 shows the probiotic organisms isolated and characterized from maize, castor oil, and oil bean through the biochemical test. The active probiotic organisms isolated were *Pediococcus*, *Micrococcus*, *Lactobacillus*, and *Bacillus* species. These organisms are accountable for fermentation and can utilize constituents of the fermented foods.

The result is aligned with Aworh (2008), who reported that fermentation improves the texture and flavor of foods imparting a pleasantly sour taste. It also enhances the value of food materials giving it higher quality, detoxification, and better preservation. The Lactobacillus produces acid, which further inhibits the growth of non-desirable organisms. The *Pediococcus* are home fermenters that produce lactic acid. This agrees with the observation of Ogueke et al. (2005), which reported that microorganisms isolated from fermented maize, soil bean, and castor oil seed usually contain proteolytic, lipolytic, and amylolytic ability to breakdown protein, carbohydrates, and lipids. The *Bacillus*, *Lactobacillus*, *Micrococcus*, and *Pediococcus* isolated from the three fermented foods are gram-positive and produce lactic acid and acetic acid, identifying them as probiotics.

Conclusion:-

Lactic acid bacteria are among the most severe microorganisms used in food fermentation. Probiotics are not pathogenic organisms in foods that can positively influence the host's health and modulate the gastrointestinal tract. The study finding revealed that the following organisms were isolated from the fermented foods: Maize: *Lactobacillus* and *Pediococcus* species, Oil bean: *Bacillus* and *Micrococcus* species, castor oil: *Lactobacillus* and *Bacillus* species. *Lactobacillus* and *Bacillus* species are found to be expected. It fermented the foods very well and gave them the desired texture, flavor, and taste. Therefore, it could be concluded that these organisms are responsible for the fermentation of carbohydrates and protein-rich seeds to give them desired fermented products (Maize, Oil bean, and Castor oil). However, certain microorganisms were detected and isolated from the sample. Perhaps, the presence of these microorganisms could be attributed to the poor hygienic condition. Therefore, the study recommends observing proper sanitary conditions while preparing food fermentation. Also, foods that contain active probiotics are recommended to enhance immune system responses, prevent infection and reduce inflammation.
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