

Effects of Jicama- (*Pachyehizus erous*) Based Oral Rehydration Solution on Intestinal Alkaline Phosphatase in Diarrheagenic Rats

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Abstract

Diarrhea results in the disruption of intestinal architecture and marked decrease in the activity of intestinal brush border enzymes, including Alkaline phosphatase (ALP). This study was done to determine how well jicama (*Pachyrhizus erosus*) based oral rehydration solution (ORS) can improve intestinal enzyme activity in diarrheagenic rats. Animals were divided into 4 groups; the control group (animal received distilled water and rat chow), Group A (animals were induced with diarrhea and left untreated), Group B (animal were induced with diarrhea and treated with World Health Organization Glucose Electrolyte Solution (WHO GES), while Group C (animal were induced diarrhea with jicama-based ORS (JB-ORS). Diarrhea was induced by feeding the animal with 5ml/100g body weight of 20% solution of D- mannitol daily. After treatment, the rats were sacrificed under ketamine anesthesia, dissected and their small intestines were removed, homogenized, and assayed for alkaline phosphatase enzyme. Jicama ORS increased alkaline phosphatase (ALP) levels (higher than WHO-ORS) and resulted in the normalization of body weights in rats following diarrhea induction. The results suggest that jicama could be used as food-based ORS to restore to normal metabolic activities in diarrheic condition.

Keywords: Diarrhea, Alkaline phosphatase (ALP), Jicama (*Pachyrhizus erosus*), Oral Rehydration Solution (ORS), Intestinal enzyme activity.

Introduction

Approximately ten children under the age of five died from diarrheal disease every minute in 1980, killing a little over five million children under the age of five [1]. These fatalities happened as a result of the 338 million young ones in these years of life who experienced around 1 billion bouts in developing nations (with the exception of China), and they were probably more common in households with lower incomes [2]. Being dehydrated is credited with 60-70% of diarrhea-related fatalities. Majority of these diarrhea-related fatalities can be avoided by using Oral Rehydration Therapy or Solution (ORT/ORS), which may avoid and manage this dehydration [3]. Due to its widespread usage in the medical industry and the fact that friends and family are capable of using it at one's residence, this technical advancement opens up significant new opportunities for lowering the incidence of child fatalities [4]. These two home-prepared solutions and readymade salts are acceptable forms of ORT that can be given to treat diarrhea [4]. Just 10 percent in all paediatric bouts of diarrhea can now be treated with the two hundred million oral substitute's packs that are annually produced. Accelerating the manufacture of ORS and spreading more knowledge about the at-home prompt intervention of diarrhea are urgently needed [5]. Researchers learned in the beginning of the seventies that a straightforward solution of glucose and electrolyte salts could be used to replenish people's body water-especially newborns and kids who were dehydrated as a result of diarrhea. The oral rehydration solution (ORS) packs established the focal point of international initiatives to lower diarrheal diseases and mortality. The awareness and adoption of ORT, which in alongside ORS involves a higher fluid

intake via accessible sources like as nursing milk, rice syrup infused with salt, stews, and grain meals, has been a part of the effort to reduce dehydration throughout time [5]. By the late 1990s, ORS/ORT is thought to have played a significant role in avoiding over a million cases of diarrhea-related fatalities yearly. But at the present, understanding and use of ORS/ORT have stalled in some nations while it has declined in certain others [5]. Salts of phosphorus play a significant role in daily living. They are employed in a variety of technological applications in addition to the manufacturing process. Each form of life contains phosphorus, meaning that without it, humans would perish. For animals, humans, and plants to be properly nourished, at least daily requirement consumption is necessary. By utilizing adenosine triphosphate (ATP), a natural energy carrier, they play a role in transmitting energy within cells [6]. On the contrary, if several companies were to remake without employing phosphate salts, they would face substantial difficulties and a significant struggle, and it is certain that some of the now-existing items would completely fade off circulation or just remain occasional [6]. A hydrolase enzyme called alkaline phosphatase (ALP, ALKP) (EC 3.1.3.1) is in charge of liberating phosphate residues off a variety of compounds, such as protein chains, nucleotides, and alkaloids [7]. Dephosphorylation refer to the term used to describe the removal of the phosphate group. Alkaline phosphate enzymes work most efficiently in an environment with a high pH, as implied by their name. This particular enzyme has an extensive variety of uses since it plays numerous essential functions in both humans and other uses [7].

Despite the fact that the general fatalities due to sickness caused by diarrhea has progressively decreased

through the past few decades, acute diarrhea remains an important concern in many parts of the globe [8].

Introducing oral rehydration therapy (ORT) proved crucial in lowering the death rate from diarrhoea. World Health Organisation oral rehydration solution (WHO-ORS) was first evaluated for the treatment of cholera, and it was later demonstrated to be effective regardless of the source of diarrhea or the patient's age [9]. However, neither the volume of stools nor the length of diarrheal episodes are significantly shortened by the WHO-ORS. One of the possible agricultural crops has been widely grown for centuries in practically every country in the globe, including Indonesia, the jicama (*Pachyrhizus erosus*; Fabaceae) [10]. According to recent researches, an edible jicama tuber contains a variety of bioactive substances, such as vitamin C, flavonoids, vitamin B1, vitamin B2 as well as inulin. The consumption of jicama tuber, whether it is provided fresh or transformed, has been shown to have significant positive impacts on health [11]. Jicama extract was used in a study on mice that served as a model for type 2 diabetes, and the results showed that after six weeks of therapy, both blood glucose levels and HbA1c, an objective indicator of glycemic control, had substantially reduced [12].

Furthermore, the receptivity to both insulin and glucose was much increased, and activity of liver function was entirely restored, demonstrating that jicama extract prevents the evolution of diabetes by improving the responsiveness to insulin [13]. A substantial inhibition impact of jicama extract regarding the activities of glucose synthesis enzyme (-glucosidase) and -amylase activity was shown in different research using mice injected with streptozotocin (these mice are used as a model for type 1 DM), were studied to understand how to lower the rise in blood glucose levels after eating

[14-15]. This suggests that ingesting fresh jicama tuber has a positive impact on heart function [16].

As a result, better ORSs for treating acute diarrhea demonstrating that jicama extract prevents the evolution of diabetes by improving the responsiveness to insulin [17]. A substantial inhibition impact of jicama extract regarding the activities of glucose synthesis enzyme (-glucosidase) and -amylase activity was shown in different research using mice injected with streptozotocin as subjects to reduce the postprandial blood glucose increase. Jicama juice has been shown to stimulate circulatory nitrate and nitrite levels, which prevents platelet aggregation brought on by collagen [18]. This suggests that consuming fresh jicama tuber has a positive impact on cardiovascular health. As a result, improved ORSs for the treatment of acute diarrhea are still being looked for and tested.

Research Aim

This study aim to examine the impacts of an oral rehydration solution made from Jicama (*Pachyrhizus erosus*) (JB-ORS) on intestinal Alkaline Phosphatase (ALP) activity in diarrheagenic rats. By comparing the effects of JB-ORS with the standard World Health Organization Glucose Electrolyte Solution (WHO GES) and untreated conditions, the study seeks to determine the potential of jicama-based ORS in improving intestinal enzyme activity and restoring normal metabolic functions following induced diarrhea.

Filling Knowledge Gap

1. Enhancement of Intestinal Enzyme Activity: The study addresses a gap in knowledge by investigating the impact of jicama-based ORS on Alkaline Phosphatase (ALP) activity. While

dehydration and diarrhea disrupt intestinal enzyme activity, particularly ALP, the study explores the potential of jicama-based ORS to mitigate this decrease and enhance ALP levels.

2. Comparative Evaluation: By comparing the effects of JB-ORS with both untreated conditions and the standard WHO GES, the study adds a crucial dimension to understanding the efficacy of jicama-based rehydration solutions. This comparison offers insights into whether jicama-based ORS has superior effects on ALP activity and normalization of body weights compared to existing treatments.

3. Natural Food-Based Solution: The research introduces a novel approach by utilizing jicama, a naturally occurring edible tuber, as the base for ORS. This fills a gap in research related to alternative and potentially more accessible rehydration solutions, contributing to the field of diarrheal disease treatment and management.

4. Functional Benefits of Jicama: The study builds on existing evidence of jicama's health-promoting effects, such

as its potential to improve glycemic control and cardiovascular health. By exploring its impact on ALP activity, the research extends the understanding of jicama's potential benefits beyond its traditional established roles.

5. Application in Diarrheal Management: The study bridges the gap between traditional oral rehydration therapy (ORT) and novel food-based approaches. By demonstrating the potential of jicama-based ORS to enhance ALP activity, the research contributes to the broader knowledge of effective strategies for managing acute diarrhea and associated complications.

To sum up, this study aims to investigate the effects of jicama-based oral rehydration solution on intestinal Alkaline Phosphatase activity in diarrheagenic rats. By addressing gaps in knowledge related to enzyme activity restoration, comparative efficacy, natural food-based solutions, and functional benefits of jicama, the study contributes valuable insights to the field of diarrheal disease treatment and management.

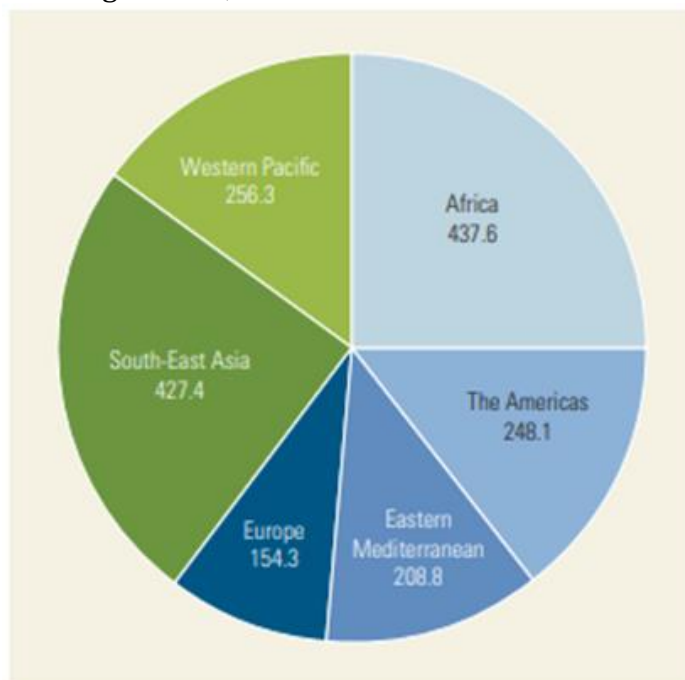


Figure 1 Regional d, ages 0-4 years, 2010 [19]

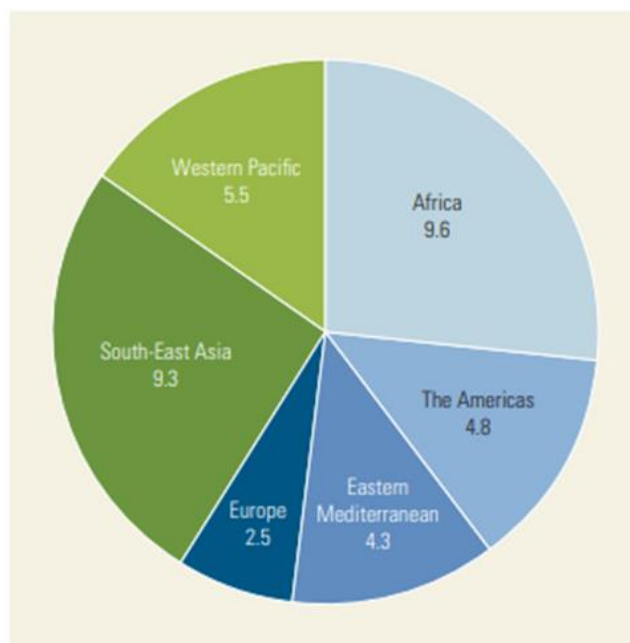


Figure 2 Regional burden of severe diarrhea episodes, ages 0-4 years, 2010 [19]

Materials and Methods

Preparation of Jicama-ORS

A 200 g portion of jicama was properly washed and boiled in 1000 ml of distilled water for 30 min, after which it was dried for several days to remove all moisture present, when confirmed dried, it was grinded to powdery form. A 100 g of the cooked jicama was used in place of glucose in standard Glucose Electrolyte Solution (GES) formulae [20]. The jicama powder was boiled in 1000 ml of water for 10 min and stirred continuously. The solution was brought to room temperature, and then 300 ml of orange juice, 2.5 g of baking powder and 1.75 g of cooking salt respectively were added to provide the ions: chloride bicarbonate, sodium and potassium. The resulting solution was allowed to cool and stored in refrigerator [20].

Experimental Animals

The animals used in this research were fifty (50) weaning albino rats of the Wister strain were obtained from Lagos

State College of Medicine, Nigeria. They were kept in an appropriate environment with temperature of 26 °C and relative humidity of 65% with unrestricted exposure to food and water throughout the experiment.

Chemicals and reagents: Each used chemical was of laboratory quality.

Induction and Treatment of Diarrhea

At the end of two weeks acclimatization period, 5 distinct groups of 8 rats each were formed from the entirety of the rats that weighs from 100 g to 200 g. The other 10 weighed below 100 g and therefore were excluded. One group served as the positive control, while the other groups were induced with osmotic diarrhea. Diarrhea was induced by force feeding the animals with 5 ml/100 g body weight of 20% solution of D-mannitol daily between 9 am and 10 am using an oral canular [21]. Diarrhea manifestation was checked by monitoring parameters such as, rate of food consumption, faecal output, color and texture of faeces, body weight, ruffled fur, hunched back, hair loss and

other signs. For the treatment of diarrhea, the five groups were labeled as follows:

- i. Group A: (Positive Control): In this group all animals were administered with distilled water only.
- ii. Group B: (Negative Control): In this group, all the animals had diarrhea but received no treatment.
- iii. Group C: In this group, the diarrheic rats were treated with the original WHO formulate.
- iv. Group D: In this group, the diarrheic rats were treated with jicama oral rehydration solution.

Preparation of Homogenates

The test subjects were properly weighed and sacrificed after chloroform anaesthesia administration at the end of the experiment. The animals were cut open and their intestines were immediately removed and chilled on ice. The intestines were homogenized using porcelain mortar and pestle. Four parts (w/v) cold distilled water were used in the homogenization. The nuclei and large cell fragments were removed by centrifugation at low speed, while the supernatant was stored in a refrigerator in labelled eppendorf tubes.

Biochemical Analysis

Determination of the Serum Total Proteins

This was done according to the biuret method [22] using a commercially available kit from Randox laboratories (N/Ireland).

Alkaline Phosphatase Activity

With slight adjustments, the Dahlqvist [23] method was used to conduct the intestinal disaccharidase activity assay. The amount of D- glucose liberated from the disaccharides; lactose, sucrose, and maltose were measured using a glucose oxidase/peroxidase system. Enzyme activity was expressed as the amount of glucose liberated by lactase in $\mu\text{mol}/\text{mg}$ protein at 37 degrees. Alkaline phosphatase activity was measured using the Naftalin *et al.* [24] technique using 4-methyl-p- nitrophenol phosphate as the substrate. Enzyme activity was expressed as the amount of P-nitrophenylphosphate in units hydrolyzed per milligram protein and measured spectrophotometrically at 405 nm and 37 °C.

Statistical Analysis

One-way ANOVA was used to analyze the variables using Sidak's test. Utilizing version 8.5.2 of graph pad prism for graphs.

Results

Mean Body Weights

Effect of diarrhea on body weight of rats is displayed in Figure 1.

Serum Total Protein

Serum protein determined is shown in Figure 2 below.

Alkaline Phosphatase Activity

Alkaline phosphatase activity in tissue homogenates of rats is demonstrated in Figure 3.

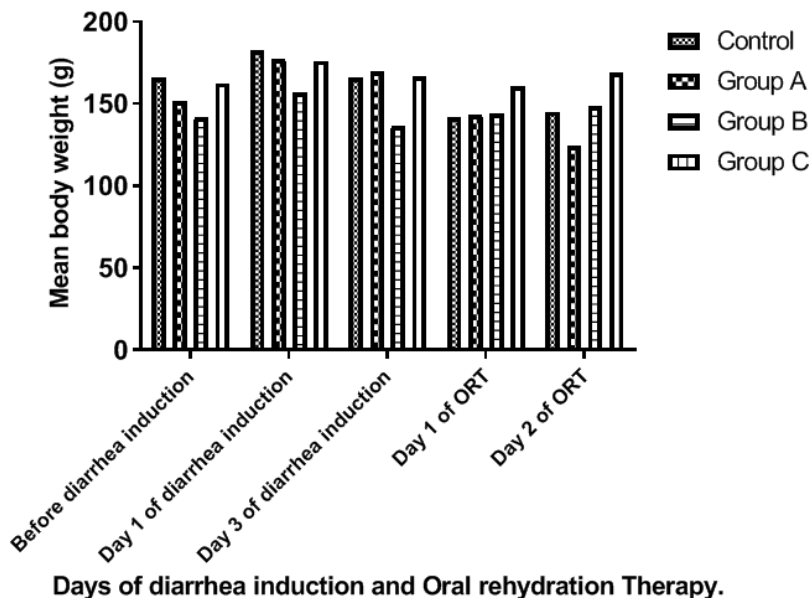


Figure 1 Effect of diarrhea on body weight of rats. Control (received distilled water and rat chow); Group A (diarrhea induced without treatment); Group B (diarrhea induced with WHO-ORS treatment); and Group C (diarrhea induced with Jicama based ORS). One-way ANOVA was used to analyze the variables using Sidak's test

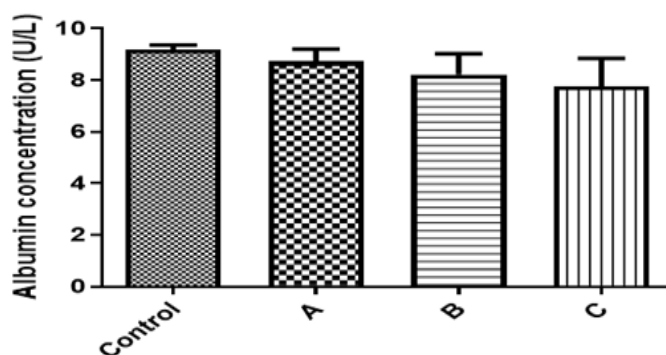


Figure 2 Serum protein determination. Control (received distilled water and rat chow); Group A (diarrhea induced without treatment); Group B (diarrhea induced with WHO-ORS treatment); and Group C (diarrhea induced with Jicama based ORS). One-way ANOVA was used to analyze the variables using Sidak's test

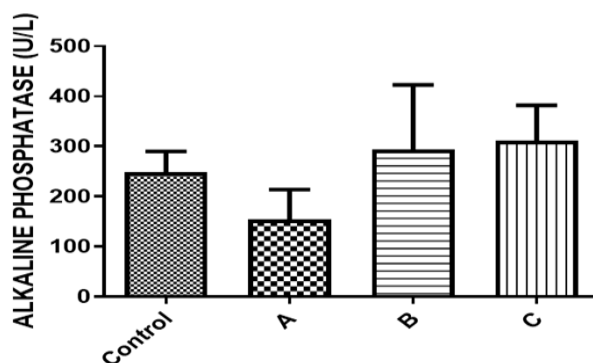


Figure 3 Alkaline phosphatase activity in tissue homogenates of rats. Control (received distilled water and rat chow); Group A (diarrhea induced without treatment); Group B (diarrhea induced with WHO-ORS treatment); and Group C (diarrhea induced with Jicama based ORS). One-way ANOVA was used to analyze the variables using Sidak's test

Discussion

Poorly digestible food can lead to diarrhea by disrupting the digestion process, causing decreased nutrient absorption, higher water and salt output, and faster digestion [25]. When rats were given D-mannitol for three days, it caused a type of diarrhea called osmotic diarrhea, there was no symptom of diarrhea. Which prompt the use of immunocompressor followed by double dosage of D- mannitol on the rats which makes most of the rats dead leaving 5 rats in each group (A-D). The following features: dehydration, changes in faeces and loss of appetite were observed. The effect of diarrheal induction and oral rehydration therapy on respective body weights is displayed in Figure 1. The average body weight of the rats induced with diarrhea (Groups A and C) gradually increased on commencement of oral rehydration solution (ORT) while there is a decrease in the body weight Group B.

There was a marked decrease in body weights of the rats at the peak of diarrhea induction (day 1-3) suggesting secondary malnutrition associated with malabsorption. Treatment with oral based rehydration solution result in the restoration of body weights and weight gain as a result of jicama-based ORS.

There was no significant different of osmotic diarrhea and ORT on the activity of Alkaline phosphatase between Group B and C (Figure 3). Though the group treated with the jicama-based ORS "Group C" and the group treated with World Health Organization Oral Rehydration Solution (WHO-ORS) "Group B" has the ability to repair damage intestinal mucosa, as depicted in Figure 2. The serum total protein determination from this study shows that jicama-based ORS does not significantly decrease albumin concentration (Figure 3). There was a slight increase in the control group when compared to Groups A, B, and C.

The result of alkaline phosphatase on serum shows no significant differences between the Groups (A-D), but there is an increase in the untreated group "Group B" and the WHO-ORS group "Group C". However, the jicama-based ORS group shows a decrease in the serum alkaline.

Conclusion

One of the possible ways to reduce the high osmotic value is to prepare Oral Rehydration Solution (ORS) from common food sources of carbohydrate polymers. Food based oral rehydration solutions are from common food sources of carbohydrate compounds. Most food-based ORS are formed with rice, though other cereals and tubers and legumes have been used. They are cheap, effective, available, and practical and of high nutritional benefits.

To sum up, the results suggest that jicama could be used as a food-based ORS to restore normal metabolic activities by placement of lost fluids, and essential salts, prevention of dehydration and improvement of diarrhea outcomes in diarrheic conditions.

Ethics approval and consent to participate

The animals' experimental protocol was in compliance with the Federation of European Laboratory Animal Science Association and the European Community Committee Directive of November 24, 1986 (86/609/EEC). The animals experimental protocol was approved by the Experimental Animal Ethics Committee of the Lagos State College of Medicine, Lagos State University, Lagos.

Consent for Publication

Not applicable.

Availability of Data and Materials

Not applicable.

Conflict of Interests

The authors declare that they have no conflict of interest.

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Author's Contributions

All co-authors were involved in all stages of this study while preparing the final version. All authors read and approved the final manuscript.

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