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Harnessing the Underutilized Value of Pineapple Pulp in South Western Uganda; Application in Fortifying Wheat Flour for Healthier Consumption

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Abstract: *In Uganda, pineapple processing into juice and wine is the fastest growing pineapple processing venture, during which this study observed that large amounts of pineapple pulp left behind, which contains abundant dietary fiber, is discarded as industrial waste, resulting in the loss of valuable food nutrients. Proximate analysis and processing into dry pineapple pulp powder, which is later used for fortifying commercial wheat flour, provides opportunity for production of a highly nutritious and affordable product with enormous health benefits. This paper therefore explores the feasibility of harnessing the untapped value of abundant dietary fiber content in fresh pineapple pulp, for use in improving dietary fiber content in commercial wheat flour. Fortifying commercial wheat flour using dry pineapple powder is a reliable way of enriching wheat flour with dietary fiber and ascorbic acid. This process transforms commercial wheat flour, widely consumed cereal flour in Uganda, into a much healthier food option for the fast-growing use of fast foods most of which utilize wheat flour especially in fried and baked products.*

Keywords: pineapple pulp, dietary fiber, fortification, health benefits

1. Introduction

Among many developing countries where horticultural value chains are being promoted for livelihood improvement, increase in fruit production has, unfortunately, not been matched by vertical integration of production with processing (Rolle, 2006). Little attention has been given to promoting investment in produce processing, despite this being a critical means of expanding and diversifying fruit products to increase market opportunities for fresh fruits, and add value to fruit products, while minimizing postharvest losses.

More specifically, in the Uganda food processing industry, harnessing the high dietary fiber content in pineapples for enrichment of widely consumed cereal flours, is one of the feasible processing ventures, considering the high dietary fiber content in pineapple fruit, as it has been reported by several researchers (Lund and Smoot, 1982; Bartolome and Ruperez, 1995; Gorinstein *et al*, 1999).

Many of the above studies have however focused more on utilizing waste from other pineapple parts; leaves and shell, and not the pulp left behind after juice extraction. Larrauri *et al.*, (1997), for example, reported that dietary fiber powder prepared from pineapple shell has 70.6% total dietary fiber, with better sensory properties than commercial dietary fibers from apple and citrus fruits. Krueger *et al* (1992) further highlights that dietary fibers and phenolic antioxidants, available in fresh pineapple, could be used as impending nutraceutical resource, capable of offering significant low-cost nutritional dietary fiber supplement for low-income settings.

Much as the booming market of functional food has created a great demand for utilization of natural resources to obtain

particular nutrients, Makinde and Sonaiya (2010) point out that, the greatest challenge regarding harnessing pineapple pulp as a dietary fiber source, is the difficulty incurred in effectively drying this high-moisture by-product, the high cost of drying equipment needed, and the evident lack of affordable and appropriate alternative processing methodologies. The problem is more severe in developing countries because of undeveloped or non-existent processing technical capacity needed for conversion of such by-products into useful products (Odeyinka *et al* 2003)

Several recent studies in fresh fruit processing indicate that large amounts of pineapple pulp containing dietary fiber, are discarded as wastes in the processing of fruits into fruit juices, resulting in the loss of food nutrients and the increased production of organic waste (Akcom, 2012). Unfortunately, though, a growing number of people living a sedentary lifestyle and struggling with lifestyle diseases in developing countries, are missing out on these discarded sources of dietary fiber. This paper therefore focuses on affordable methods of harnessing the untapped value in form of dietary fiber, which happens to be very abundant in fresh pineapple pulp, and its application in improving dietary fibers content in commercial wheat flour.

2. Literature Survey

According to Muyanja and Turyagenda (2006), Uganda is endowed with fertile soils that support production of various fruits and vegetables. Most fruits produced in Uganda are of significant economic importance, and if well harnessed, have great export potential. The fruit chains that have been developed to the level of processing include; pineapples, passion fruits, apple bananas (Ndiizi), Gros Michel bananas (Bogoya), avocado, citrus, mangoes, papaya and jackfruit (UIA, 2001).

MAAIF (2004) highlights that the fruit and vegetable sector in Uganda contributes significantly to the non-traditional exports. In addition, the country's current fruit production lies at 384, 000 tones, of which 90% is marketed locally without any processing. Much as the recent global trends are such that fruit and fruit products trade is increasing, fruit processing in Uganda is very under exploited. More specifically focusing on the pineapple value chain, only a limited number of processors and exporters are currently involved in pineapple processing (Brett et al 1996).

With growing interest and investment in fruit and vegetable production to alleviate household poverty in developing countries (Dixon *et. al.*, 2001), it is evident that in Uganda, fresh fruit processing for juice and wine production is evidently growing steadily (UIA, 2009), being driven by urbanization, growth in incomes and middle class, as well as among the more nutrition and health conscious population. There is more consensus now among fruit supply chain actors about the fact that agricultural processing improves the viability, profitability and sustainability of fruit production systems, by accruing farm incomes, generating rural employment, and earning foreign exchange. This calls for designing context specific and innovative strategies for proper postharvest management of fresh produce.

Ackom (2012) observed that vast amounts of dietary fiber-containing fruit components are usually discarded as pulp, left behind as "wastes" in the processing of fruits into fruit juices, results in the loss of valuable food nutrients. This scenario is also seen to be common along the pineapple value chain in Uganda, whereby large amounts of pineapple pulp are simply discarded as waste after either domestic or even industrial extraction of juice, as this is never considered to be lost.

According to Eskicioglu (2015) however, the number of dietary fiber enriched food products introduced to the food market recently has been increasing, due to several beneficial effects of dietary fibers, mainly on the digestive system. Because of this trend, there are deliberate efforts geared towards finding new sources of dietary fiber, such as agronomic by-products, that can be used as ingredients in the food industry (Rodríguez *et. al.*, 2006; Redondo-Cuenca *et. al.*, 2008). This paper thus describes findings from a study that investigated the feasibility of using dry pineapple pulp to enrich commercial wheat flour with dietary fiber. Tyroller and Zwickenpflug (2002) further emphasize that the most commonly consumed dietary fiber products have been those derived from cereals, however, over the past decade, high dietary fiber materials from fruits are increasingly being sought after.

A major observation in Uganda is that to many actors along the pineapple value chain, the pineapple pulp left after extraction of pineapple juice and wine is never put to any beneficial use, yet this pulp is rich in digestive fiber, which is essential for human health (Dhingra. *et.al.*, 2012). If well harnessed, pineapple pulp could be used to boost dietary fiber content of many commonly and widely consumed flours, say maize, cassava or even wheat flour. The World's Healthiest Foods-USA (2017), during their campaign of popularizing health-promoting foods "that can change your

life" indicated that the health benefits of wheat depend entirely on the form in which it is eaten. These benefits will be few if one selects wheat that has been processed into 60% extraction, bleached white flour- which is unfortunately the commonest on the Ugandan local market. This realization has prompted many food scientists to advocate for "enrichment" of processed wheat flour with the nutrients that make it more nutritious.

University of California health services (2004) describes dietary fiber as an important part of a healthy diet, important for moving food and waste efficiently through the digestive system. Several studies have also revealed that consumption of food with higher fiber content gives profitable physiological effects and health benefits, (Buttriss & Stokes 2008; Borchani et al. 2011). According to Kucerova *et.al.*, (2013), soluble dietary fiber can be partially split by digestive enzymes in the upper gastrointestinal tract. It increases the viscosity of the stomach and intestinal content, thus slowing down the mixing of their content. It also limits the access of pancreatic amylases and lipases to substrates, which in turn limits the absorption of nutrients by the intestinal wall. It may also slow down the passage of the intestinal content and decrease the diffusion of nutrients. This emphasizes the relevance of digestive fiber in controlling weight gain.

The many benefits of dietary fiber intake have prompted attention to be directed towards development of products enriched with fiber. According to the Food and Drug Administration (FDA) (2013), to have a product with a "high source of fiber" and "good source of fiber", it must contain 20% or more fiber. With the demand for healthily oriented products, products with a low content of sugar, low energy and a high content of fiber increasing daily in wheat products, fortifying wheat using pineapple pulp powder offers a viable way forward to meet this demand. With respect to this, Martinez et al., (2012) observed that, 75% of carbohydrates in a cup of raw pineapple chunks consist of sugars, 11% of these carbohydrates, is made up of dietary fiber. This implies that a cup of raw pineapple chunks can provide up to 11 % of the recommended daily fiber intake. For most Ugandans, wheat is not a traditional staple food, but its consumption is rapidly growing in prominence, particularly among urban and high-income households, due to its ease and variety of preparation (Haggblade and Dewing, 2010). The national annual wheat consumption currently lies in the range 26, 000-37, 000 tones and the demand has been growing steadily over the last 8 years (Sekitooleko and Vogel, 2011). Much as wheat has a fair amount of dietary fiber, the content can be enhanced if an appropriate source of dietary fiber is identified and appropriately utilized.

If pineapples are to be used as a source of fiber for enriching wheat flour in Uganda, drawing from their own study findings, Serena & Knudsen (2007) state that fruits and vegetables are usually inexpensive, abundantly available and are a good source of dietary fiber. We are therefore certain of a sustainable source of the dietary fiber, owing to the increasing acreage being devoted to pineapple production and the growing processing of pineapple into juice and wine, in Uganda. In addition, it is important to remember that

dietary fibers from fruits have a high proportion of soluble dietary fiber and bioactive associated compounds (Spiller, 1986). Also, the dried fruit pulp waste powder of pineapple is highly nutritious and a good source of dietary fiber – 8.5g/100g, Vitamin C- 8.8mg/100g.

3. Problem Definition

Muyanja and Turyagenda (2006) highlighted that in Uganda, pineapple ranks high among fresh fruits with remarkable potential for processing and diversification into various products. Only a limited number of processors and exporters however, are currently involved in any form of pineapple processing (Brett et al 1996). Such under exploited processing and value addition opportunities represent a huge untapped potential for pineapple value chain actors to more profitably benefit from their engagement in this chain. This is also a strong indication of the available investment opportunities in pineapple processing and value addition, with special focus on meeting the growing demand in both the in country and export market (MAAIF, 2004).

Based upon such evident unharnessed potential, there was an evident need for considering full scale and innovative fresh fruit processing attempts and development of new market driven products, to make the pineapple value chain more productive. Drawing from the observation that the number of dietary fiber enriched food products introduced to the food market recently has been increasing, due to several beneficial effects of dietary fibers, mainly on the digestive system (Eskicioglu, 2015), this paper explains findings from an experimental study which was carried out to determine the feasibility of utilizing pineapple pulp, to improve digestive fiber content in wheat flour, as a means of reducing postharvest losses along the pineapple value chain in SW Uganda, and accrue better income to the value chain actors involved. This was in response to the observation that among most pineapple juice and wine processors, the pineapple pulp left after extraction of pineapple juice and wine was never put to any beneficial use, yet this pulp is rich in digestive fiber, which is essential for human health (Dhingra *et al.*, 2012).

Most of these processors do not regard the discarding of this “left-behind” pulp as a loss in any kind, partly because most of them are unaware of the nutritional value of this pulp and how best it can be harnessed to utilize it for better use. The pineapple pulp is a dependable natural source of digestive fibre, and this fibre, as indicated by several epidemiological studies, is important for reducing the risk of cardiovascular diseases, various types of cancer, and type 2 diabetes by enhancing the digestive process, stimulating bowel movements, lowering cholesterol, and exerting a positive influence on blood sugar levels (Higgins, 2004; (Venn, 2004; Thewissen, 2008).

4. Methodology/Approach- Preparation of pulp powder

Pineapples for the experiment were collected in equal amounts from the 3 study sites (52 per site), washed with soap, peeled, sliced into smaller pieces and blended. The

juice was then filtered out of the pulp using a muciline cloth. The pineapple pulp was then cleaned of pineapple peels and other extraneous matter. 1000 g of the pulp was soaked in 2000 ml of the Ca (OH)₂ solution to neutralize its PH. Sub-samples of the cleaned pulp were then soaked in varying concentrations of food – grade calcium hydroxide for 15 minutes, followed by 15 minutes soaking in water, to modify the pH of the pulp. The concentration of the solution was varied from 0.01 M - 0.1 M to obtain a desired close to neutral PH.

The pulp samples were then air dried by spreading the pulp out on a tray and exposing it to a constant air current generated by 2 electric fans, at 25 degrees Celsius for over 5 hours after which the dry pineapple pulp was placed in an electric moisture analyzer to determine the effectiveness of the drying method used. The dry pulp was then hammer-milled into the final product (dry pineapple pulppowder). The pulp powder was then sieved through a sieve of hole diameter of 30 microns to produce a powder with uniform particle size.

Fortification of the wheat flour was done at 5%, 10% and 15%, in order to compare and determine the best % at which fortification will produce an acceptable product. The effect of fortification of wheat flour with pineapple pulp powder, on the physical properties of wheat was also investigated;

Proximate analysis was carried out on the unfortified wheat flour, the pineapple pulp powder and the fortified wheat flour sample using the standard analytical methods:

Crude protein was determined using the Kjeldahl, nitrogen combustion that follows three successive stages: Digestion, Distillation and Titration according to (A.O.A.C. method 977.02) – (1990) (A.O.A.C. method 977.02) – (1990). Two Kjeldahl digestion tubes were collected and label with a permanent marker close to the top. 0.1 g of dry sample were weighed out on to a filter paper (W1). The filter paper containing the dry sample was folded and carefully transferred to a labelled digestion tube. Another filter paper was placed in a second digestion tube to act as a blank. Two catalyst tablets (copper sulphate + potassium sulphate) were added to each tube, followed by 10 cm³ concentrated H₂SO₄ and mixed by gentle swirling. 10 cm³ H₂O₂ was then added in a fume cupboard while watching carefully for frothing. The peroxide was then added slowly at first. The tubes were placed in the digestion block and digested until clear (30 minutes), carefully checking to ensure that there were no black carbon particles visible. The tubes were allowed to cool until barely warm but still liquid, after which 70 cm³ distilled, Water was added.

In turn, the tubes were placed into the distillation unit and twisted to seal.. The door on the apparatus was closed and the cycle was started. The apparatus automatically added alkali to the tube and then bubbled steam to distil off the Ammonia. This was collected in boric acid. The titration went on automatically. The volume of acid used was displayed on the front of the apparatus. When the titration was finished the tubes were emptied automatically, and the volume of acid used was recorded. The next tube was inserted, and the process continued.

Total Nitrogen and Protein in the food, measured in dry sample according to the following formula:

$$\text{Total Nitrogen} = \{(VI - BL)\} \times 0.1 \times 0.0014 \times \{\text{Total Dry Matter of Food}\} / W1$$

Where:

Weight of food sample = W1

Volume of acid in sample titration = V1

Volume of acid in blank titration = BL

Concentration of acid, M = 0.1 M;

Where 0.1 is molar concentration of the HCl and 0.0014 is factor for equivalence of HCl to NH₃

Percentage Nitrogen and Protein In original, wet sample was calculated as follows:

$$\text{Nitrogen, \%} = \{(VI - BL)\} \times 0.1 \times 0.0014 \times 100 \times \% \text{Dry Matter} \times 100$$

To convert to protein, the value was multiplied by the nitrogen-to-protein conversion factor 6.25, which is a standard.

For Crude fat (A.O.A.C. method 920.39), the Soxhlet extraction thimble was dried at 105°C to constant weight, removed, cooled to room temperature in a desiccator, and weighed to the nearest 0.1 mg. The sample was carefully added to the extraction thimble, taking care not to overfill the thimble, leaving at least a 1 cm gap between the sample and the top of the thimble. The filled thimble was weighed to the nearest 0.1 mg. A plug of glass wool was placed on top of the sample to prevent sample loss during the extraction. Several boiling chips were placed into a clean, dry receiving flask or beaker, the container weighed with chips, to the nearest 0.1 mg and recorded as the tare weight of the container. The Soxhlet apparatus was assembled using at least 160 mL of 95% ethanol. The thimble was inserted and heated at reflux for 24 hours. Periodically the reflux rate was checked, and the heating rate adjusted to give four to five solvent exchanges per hour in the Soxhlet thimble. When the extraction time was complete, the thimble was removed, and the sample transferred to a Buchner funnel. All residual solvent was removed by vacuum filtration and the sample washed thoroughly with 95% ethanol, collecting all of the filtrate. The biomass was allowed to air dry in the Buchner funnel while it was still attached to the vacuum system. The filtrate was combined from the previous step and any solvent from the upper section of the Soxhlet apparatus with the solvent in the 250 mL flask. The flask was placed on the rotary evaporator and the solvent removed under vacuum using a water bath temperature of 45 ± 5°C to heat the flask during evaporation. After all of the visible solvent had been removed by the rotary evaporator, the flask was placed in a vacuum oven (75-100 torr) at 40 ± 1°C for 24 ± 1 hour. The flask was removed at this time and allowed to cool to room temperature in a desiccator. The flask was weighed and recorded as the total weight to the nearest 0.1 mg.

Calculation

The oven dry weight of the sample was calculated using the average total solids content.

$$\% \text{ Extractives} = \{\text{Weight of container} + \text{Residue}\} - \{\text{Tare weight of container}\} * 100 / \text{ODW}$$

The numbers of extractives in the sample were calculated on a percent dry weight basis.

$$\text{ODW} = \{\text{Weight of thimble} + \text{sample}\} - \{\text{Weight of thimble}\} * \% \text{ Total solids} / 100$$

Where ODW = Oven Dry Weight

Crude fiber (A.O.A.C. method 930.10), 2g of ground material were extracted with petroleum ether to remove fat (Initial boiling temperature. 35 -38°C and final temperature 52°C), followed by boiling 2g of dried material with 200mL of sulphuric acid for 30min with bumping chips. The mixture was filtered through muslin and washed with boiling water until washing were no longer acidic. Mixture was boiled with 200mL of sodium hydroxide solution for 30min, followed by filtering through muslin cloth again and washing with 25mL of boiling 1.25% H₂SO₄, three 50mL portions of water and 25mL alcohol. The residue was removed and transferred to ashing dish (pre-weighed dish W1). The residue was dried for 2h at 130 ± 2°C, and the dish cooled in a desiccator, then weighed to obtain (W2). The mixture was ignited for 30min at 600 ± 15°C, followed by cooling in a desiccator and reweighed to obtain (W3).

Calculation

% crude fiber in ground sample =

$$\{\text{Loss in weight on ignition (W2 - W1) - (W3 - W1)}\} / \{\text{Weight of the sample}\} * 100$$

Total ash content (A.O.A.C. method 930.05) was obtained by Organic matter in the sample was burnt off at as low temperature as possible in stages; first to char the product thoroughly and finally to ash at 550° C in a muffle furnace. The inorganic matter left after burning organic matter was cooled and weighed, as follows;

Crucibles were placed in muffle furnace to heat at 550° C for 15 minutes. Crucibles were removed, then cooled in a desiccator for one hour and weighed obtaining the value (W). 2 g of sample were weighed into the crucible (W1). Sample was kept on a hot plate till smoking ceased and sample became thoroughly charred. The crucibles were placed inside the muffle furnace and heated to 550° C for 5 hours. The furnace was let to cool after which crucibles containing ash were taken out, cleaned to a white residue in. Traces of carbon still evident were removed by cooling the crucible, adding 2 ml of water and stirred with a glass rod to break up the ash, followed by drying on steam bath and placed in muffle furnace and again heating at 550° C.

Total Carbohydrate 100mg of the sample was weighed into a boiling tube. The sample was hydrolyzed by keeping it in a boiling water bath for 3 hours with 5mL of 2.5 N-HCl and cooling to room temperature. The mixture was neutralized with solid sodium carbonate until the effervescence ceased. The volume was made up to 100mL and the mixture centrifuged. The supernatant was collected and 0.5 and 1mL aliquots were taken for analysis. The standards were prepared by taking 0, 0.2, 0.4, 0.6, 0.8 and 1mL of the working standard. '0' serving as blank. The volume was made up to 1mL in all the tubes including the sample tubes by adding distilled water followed by adding 4mL of anthrone reagent. The mixture was heated for eight minutes in a boiling water bath and cooled rapidly and read, the

green to dark green color, at 630nm. A standard graph was drawn by plotting concentration of the standard on the X-axis versus absorbance on the Y-axis. From the graph the amount of carbohydrate present in the sample tube was calculated.

Calculation

Amount of carbohydrate present in 100mg of the sample = {mg of Glucose} / {Volume of test sample} * 100

The crucible was cooled in a desiccator and reweighed to obtain (W2), weight of the crucible containing ash.

Calculation:

Ash % = {(W2 - W) / (W1 - W)} x 100

Where,

W = Weight of empty crucible

W1 = Weight of empty crucible + Sample

W2 = Weight of empty crucible + Ashed Sample

Ascorbic acid (A.O.A.C. method 967.21a)

The amount of ascorbic acid was determined using an oxidation reduction reaction whereby a solution containing ascorbic acid was used to standardize the iodine solution. Three sub-samples of 0.1g of the sample were weighed and each was placed in a numbered 125 ml Erlenmeyer flask. 30 ml distilled water was added followed by 5 drops of starch solution to each flask. A 25 ml burette was rinsed and filled with the iodine solution and titrated against the solution containing ascorbic acid. The procedure was repeated for the other two ascorbic acid solutions

Effect of fortification using pineapple pulp powder on the physical properties of wheat flour

The physical properties investigated in this section included water absorption capacity, Fat absorption capacity, Foaming capacity and stability, and Gelation capacity.

To determine the water absorption capacity, 15 ml of distilled water was added to 1 g of the flour in a weighed 25 ml centrifuge tube. The tube was agitated on a vortex mixer for 2 minutes. It was centrifuged at 4000 rpm for 20 min. The clear supernatant was decanted and discarded. The adhering drops of water was removed and the reweighed. Water absorption capacity is expressed as the weight of water bound by 100 g dried flour.

For the Fat absorption capacity, 10 ml refined corn oil was added to 1 g of the flour in a weighed 25 or 80 ml centrifuge tube. The tube was agitated on a vortex mixer for 2 min. It was centrifuged at 4000 rpm for 20 min. The volume of free oil was recorded and decanted. Fat absorption capacity is expressed as ml of oil bound by 100 g dried flour. 2 g flour sample and 50 ml distilled water was mixed in a blended at room temperature. The suspension was stirred for 5 min at 1000 rpm. The total volume after 30 sec was recorded. It was allowed to stand at room temperature for 30 min and the volume of foam recorded. The percentage increase in volume after 30 sec is expressed as foaming capacity. 2-20% suspension was prepared capacity of the blends increased as more SPF was with 5 ml distilled water in test tube. The tube containing the suspension was heated for 1 h in a boiling water bath. It was cooled rapidly under running cold water. The test-tube was cooled for 2 h at 4C. The o test was

inverted to see if content will fall or slip off. The least gelation concentration is that concentration when the sample from the inverted test tube does not fall or slip.

Microbiological suitability of the fortified wheat for human consumption

To ascertain suitability of the fiber fortified wheat flour for human consumption, microbiological analyses of total viable counts and coliform counts of bacterial colonies; and the yeast and mould counts were conducted on the pineapple pulp product using standard microbiological procedures;

Total viable count Using separate sterile pipets, decimal dilutions of 10^{-2} , 10^{-3} , 10^{-4} of the homogenate samples were prepared by transferring 10 ml of previous dilution to 90 ml of diluent. While avoiding all sampling foam, all dilutions were shaken 25 times in 30 cm (1 ft) arc within 7 s. 1 ml of each dilution was pipetted into separate, duplicate, appropriately marked petri dishes. Dilution bottle were reshaken 25 times in 30 cm arc within 7 s and allowed to stand more than 3 min before it is pipetted into petri dish. 15 ml of plate count agar (cooled to $45 \pm 1^\circ\text{C}$) was added to each plate within 15 min of original dilution. Agar was added to the latter two for each series of samples. Agar was then immediately added to petri dishes, since the sample diluent contained hygroscopic materials. Agar and dilution water were poured onto control plates for each series of samples. Sample dilutions and agar medium were immediately mixed thoroughly and uniformly by alternate rotation and back-and-forth motion of plates on flat level surface. Agar was left to solidify. The solidified petri dishes were inverted and incubated promptly for 48 ± 2 h at 35°C , taking care not to stack plates when pouring agar or when agar is solidifying

Total viable count was calculated as follows;
Plates with 25-250 CFU.

$$N = \frac{\sum C}{[(1 \times n_1) + (0.1 \times n_2)] \times (d)}$$

a. Calculate the APC as follows:

$$\frac{(31 + 31) \text{ colonies}}{0.0015 \text{ ml}} = 4.1 \times 10^4$$

Where:

N = Number of colonies per ml or g of product

$\sum C$ = Sum of all colonies on all plates counted

n_1 = Number of plates in first dilution counted

n_2 = Number of plates in second dilution counted

d = Dilution from which the first counts were obtained

Coli form count 50 g of the sample transferred into a sterile high-speed blender jar. 450 ml of Butterfield's phosphate-buffered water were added to the sample and blended for 2 minutes. Decimal dilutions were prepared with sterile Butterfield's phosphate diluent. All suspensions were shaken 25 times in 30 vortex mix for 7 s. Using at least 3 consecutive dilutions, 1 mL aliquots were inoculated from each dilution into 3 LST tubes for a 3 tube MPN analysis (1 mL or 5 mL pipet used for inoculation). Pipet was held at angle so that its lower edge rests against the tube and not more than 15 min were allowed to elapse from time the sample was blended until all dilutions were inoculated in

appropriate media. LST tubes were incubated at $35^{\circ}\text{C} \pm 0.5^{\circ}\text{C}$, examined and reactions recorded at 24 ± 2 h for gas, i.e., displacement of medium in fermentation vial or effervescence when tubes were gently agitated. Gas-negative tubes were re-incubated for an additional 24 h and examined and reactions recorded against at 48 ± 3 h. Confirmed test was performed on all presumptive positive (gas) tubes. From each gassing LST tube, a loopful of suspension was transferred to a tube of BGLB broth, avoiding pellicle if present. BGLB tubes were incubated at $35^{\circ}\text{C} \pm 0.5^{\circ}\text{C}$ and examined for gas production at 48 ± 3 h. The most probable number (MPN) of coliforms was calculated based on proportion of confirmed gassing LST tubes for 3 consecutive dilutions

Using 25 g from each subsample was analyzed. An appropriate amount of 0.1% peptone water was added to the weighed sample to achieve 10^{-1} dilution, then homogenize in a stomacher for 2 min so as to obtain a dilution of 10^{-6} . Spread-plate method was used for Plating and incubation of sample, whereby 0.1 ml of each dilution was aseptically pipetted and pre-poured on solidified DRBC agar plates and spread inoculum with a sterile, bent glass rod. Plates were incubated in the dark at 25°C , taking care not to stack plates higher than 3 and not to invert. Plates were let to remain undisturbed until counting. Plated were counted after 5 days of incubation. If there is no growth at 5 days, plates were re-incubated for another 48 h to prevent counting colonies before the end of the incubation period because handling of plates can result in secondary growth from dislodged spores, making final counts invalid. Plates containing 10-150 colonies were counted bearing in mind that if mainly yeasts are present, plates with 150 colonies are usually countable, and if substantial amounts of mold are present, depending on the type of mold, the upper countable limit would have to be lowered. Results were reported in colony forming units CFU/ml based on average count of triplicate set. Counts were rounded off to two significant figures.

Preference Sensorial Analysis of a fried product made with wheat fortified with pineapple pulp

To determine which sample was most acceptable for the consumers, chapatti was made out of plain unfortified wheat as a control, as well as the 5%, 10% and 15% pineapple-fortified wheat samples. Twelve (12) panelists were each presented with two different coded samples of chapattis made out of each of the fortification levels of wheat and the control which was chapatti made out of unfortified wheat flour and were instructed to indicate on questionnaires which product they preferred in a simple paired preference test.

This test was found to be the most appropriate for gathering consumer information, to determine if consumers like the product if one level of fortification is preferred over another and to ascertain if the consumers intend to take up the product. The method was preferred for use in this study because it accorded the researcher an opportunity to put one product up against another, and instruct the respondent to indicate which one the respondent preferred hence paired. The method however does not determine the 'preference' measure (how much one product is liked or how much more one product is liked versus another).

5. Results and Discussion

Pineapple pulp for fortification originated from the researcher's observation during documentation of post-harvest handling practices, mainly among pineapple juice processors and wine producers, that usually after extraction of the juice, the pineapple pulp left behind is never put to any significant use but is rather simply discarded as waste.

Interviews with these processors highlighted the fact that they do not consider discarding the pulp as loss, since they have already obtained the juice/wine. It was also realized that these chain actors' failure to recognize discarding the remaining pulp as loss stems from their lack of information regarding the nutritional value of the pulp and its usefulness in enriching human diet for better health. The experimental investigations revealed that it is indeed feasible to produce dry pineapple pulp powder from the pulp.

A comparison of the mean amount of dry pulp produced per 1000g of fresh fruit obtained from the three different pineapple producing locations using a paired sample T-test revealed that pineapple produced in Isingiro gave a significantly greater amount of pulp (mean: 510.4g/1000g of fresh fruit), than those from Bushenyi (mean: 408.84 g/1000g of fresh fruit) and Ntungamo ($p < .005$) (ref: table...). Pineapple produced from Ntungamo gave the least amount of dry pulp (mean: 226.15g/1000g of fresh fruit, as illustrated in fig 24.below.

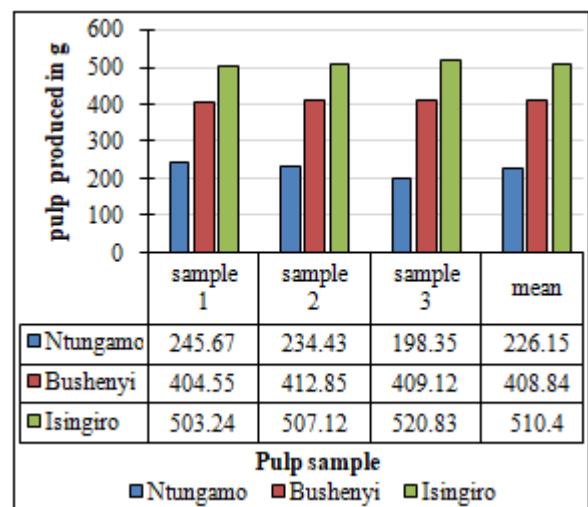


Figure 1: Dry pulp production per 1000g of fresh pineapple fruit

Comparing mean amounts of pulp produced per 100g of fresh pineapple revealed that amount of pulp produced was significantly different across the study site, [$F = (2, 18) = 1131.89, p = 0$].

A posthoc comparison test further indicated that the mean quantity of pulp produced per 100g of fresh pineapple was significantly different across the sites as shown below.

Moisture content of pulp

Analysis of the moisture content of pineapple pulp obtained from fresh pineapple fruits from the three different production sites showed that pineapples from Ntungamo had

the highest moisture content in their pulp (mean: 84.17%), which was not significantly higher than that of pulp from pineapple produced in Bushenyi (mean: 83.44%)-[F(21) = 1221.3, (p=.0033, but fairly greater than that of pineapples produced in Isingiro (mean: 76.12%). (See figure 25 below)

revealed that, generally, the moisture content in fresh pineapple pulp reduced in the same order, regardless of the site the pineapples were obtained (fig 26.). After air drying for 8.5 hours, there was no significant difference [F(19) = 213.4, (p= 0.061 in the moisture content of the final dry pineapple pulp from the different production sites. The pulp from pineapples produced in Ntungamo dried faster than that from all the other two sites

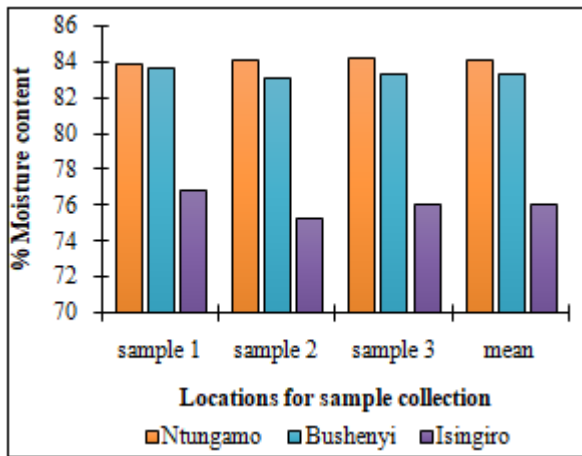


Figure 2: Moisture content of pineapple pulp

Drying behavior of pineapple pulp

During the preparation of the dry pineapple pulp, a comparison of drying behavior of fresh pineapple pulp obtained from the three different pineapple production sites

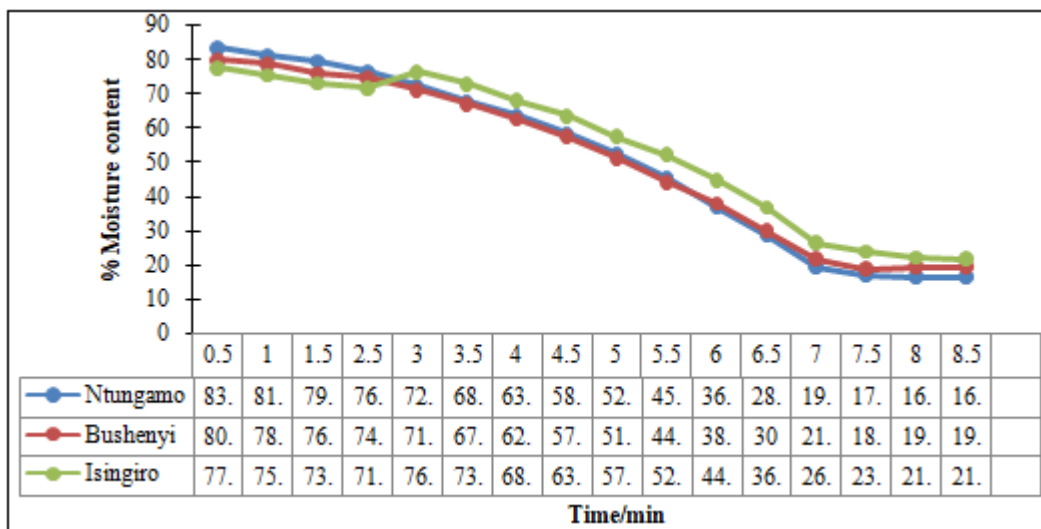


Figure 3: Drying behavior of pineapple pulp from different production sites

Proximate composition of dry pineapple pulp

After partitioning into and analyzing for the composition of six components; moisture, ash, crude protein (or Kjeldahl protein), crude lipid, crude fiber and nitrogen-free extracts (digestible carbohydrates) in the dry pineapple pulp powder (proximate analysis), this study revealed that crude fiber contributed the greatest percentage composition in dry pineapple pulp (mean: 29.6%) followed by Ascorbic acid (mean: 20.4%), see fig 4 below. These findings proved dry pineapple pulp to be a reliable source for both crude fiber and ascorbic acid.

Proximate composition of pure commercial wheat flour

For comparison, when proximate analysis was conducted on pure commercial wheat flour, crude fiber was found to be in very limited quantities (mean: 0.51%). The wheat flour was however found to be rich in crude protein (mean: 10.23%),

as illustrated in fig 31. below. These results indicate that pineapple pulp is a reliable source for crude fiber, and if used to fortify pure wheat flour, would go a long way in improving its nutritional composition. Since dry pineapple pulp had already been identified as a rich and reliable source for crude fiber, this study found it suitable for fortifying pure wheat flour (fig 4).

Proximate composition of pure commercial wheat flour fortified with dry pineapple pulp powder

Since in this experiment, fortification of pure wheat flour was done by addition of variable amounts of dry pineapple pulp powder to the wheat; 5%, 10% and 15%, proximate analysis of the various samples of fortified wheat flour revealed that that generally, composition of crude fiber increased with increasing amounts of dry pineapple pulp powder added to the wheat flour during fortification, though

not so for crude protein and crude fat as shown in figure 4 below.

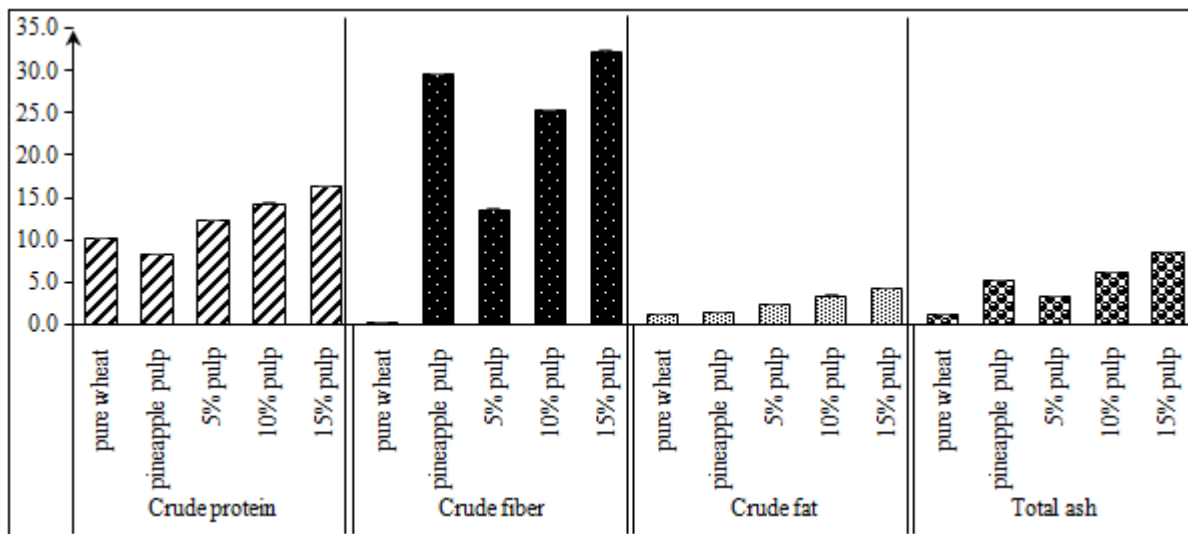


Figure 4: Proximate composition of fortified wheat samples

One-way ANOVA comparison of means also revealed that there was a significant increase crude fat content as a result of fortifying pure wheat with pineapple pulp [F (45) = 327.92, p = 0]. A posthoc comparison of the means revealed that the mean crude fat content was significantly different at different amounts of pulp added to pure wheat.

A comparison of means through one-way ANOVA also showed that fortification of pure wheat with pineapple pulp significantly increased the crude fiber content in pure wheat at 0.05% level of significance: [F (45) = 16249.35, p = 0]. A posthoc test further showed that the mean crude fiber content was significantly different for samples with different amounts of pineapple pulp added.

Unlike all other parameters investigated by proximate analysis, one-way ANOVA test revealed that there was no significant increase in crude protein content as a result of fortifying pure wheat with pineapple pulp, at 0.05% level of significance, [F (36) = 0.922, p = 0.44].

Microbiological safety of commercial wheat flour fortified with dry pineapple pulp powder

In order to ascertain the microbiological safety of the fortified wheat flour for human consumption, an analysis on pure wheat, pineapple pulp powder and fortified wheat was conducted in a certified food analysis laboratory at coca cola industries, which revealed that for all the three samples tested, total coliform counts and moulds were found to be well in range with respect to the acceptable standard, while yeast levels were slightly high (fig 5), but still in acceptable range for human consumption. The fortified wheat flour was thus certified by the food laboratory as fit for human consumption and a certificate of analysis was issued.

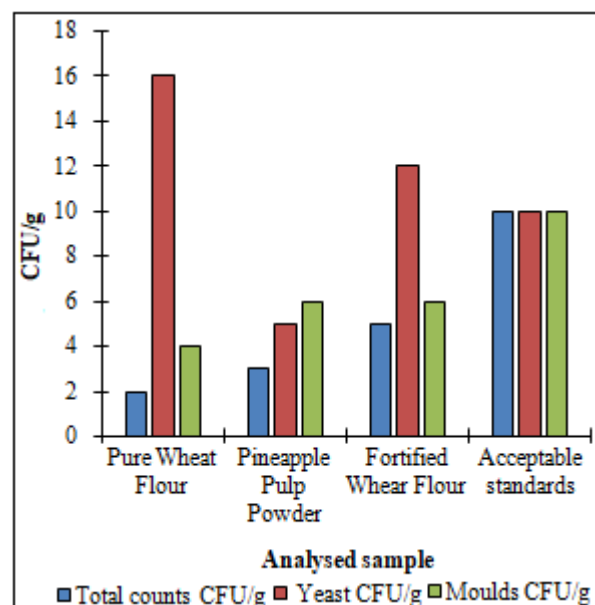


Figure 5: Comparison of Microbiological safety of pure and fortified wheat

Effect of fortification with pineapple pulp powder on the physical properties of wheat flour

Based on the fact that fortification of flours may affect functional properties of the flour, this study investigated the effect of fortification with varying percentages of pineapple pulp powder on four selected functional properties of wheat flour; oil absorption capacity, water absorption capacity, gelation capacity and foaming capacity, expected to influence the application and use of the fortified product.

The study revealed that due to the addition of pineapple pulp powder, oil absorption and water absorption capacities of wheat flour decrease with increasing amount of pulp powder used, implying products from the fortified wheat would be increasingly less soggy with water and oil.

1 below

Table 1: Functional properties of fortified wheat fl Table 1:
Functional properties of fortified wheat flour

Wheat Category	F			
	Oil absorption capacity (gs-1)	Water absorption capacity (gs-1)	Gelation Capacity (%)	Foaming capacity (%)
100% Pure wheat	2.19±0.05	2.43±0.09	10.62±0.10	4.21±0.08
5% pulp 95% wheat	1.94±0.04	2.24±0.06	12.43±0.12	4.28±0.05
10% pulp 90% wheat	1.78±0.05	2.19±0.07	13.71±0.08	4.76±0.10
15% pulp 85% wheat	1.58±0.03	1.73±0.06	4.81±0.07	5.25±0.07
p-value	0	0	0	0

A comparison of means for the effect of fortification on properties of pure wheat indicated that there was no significant effect of fortification on all the properties of wheat investigated, as shown in the table below;

Property	F	df	p value
Gelation	2.37	3, 28	0.09
Foaming	0.91	3, 28	0.05
Water Absorption	1.12	3, 28	0.18
Oil Absorption	1.73	3, 28	0.08

From this study, during air-drying, the moisture content in fresh pineapple pulp generally reduced in the same order, regardless of the site the pineapples were obtained. This implies that the rate at which moisture diffused from the pineapple pulp during air drying did not vary significantly, among pineapples produced from different locations in South Western Uganda. According to Özge *et al.*, (2017), air drying behavior from pineapple is largely influenced by the rate at which moisture diffuses from the pineapple. The rate of diffusion of moisture during drying of pineapples in turn depends on a number of factors; air speed and direction, atmospheric temperature, solute content in pulp as well as the moisture content.

The study further revealed that there was no significant difference in the moisture content of the final dry pineapple pulp from the different production sites. Pulp from pineapples produced in Ntungamo however dried slightly faster than that from all the other two sites. The lower total solute content in pineapples produced from Ntungamo may be the reason for this variation in drying time of the pulp Özge *et al.*, (2017).

During drying of the pulp however, this study opted to air dry the wet samples by spreading the pulp out on a tray and exposing it to a constant air current generated by 2 electric fans, at 25 degrees Celsius for over 5 hours after which the dry pineapple pulp was hammer-milled into the final product (dry pineapple pulp). Unlike Ackom (2012) who used oven drying of pulp, this drying method was considered much more affordable, as it requires less energy, and much more easily adaptable by small scale processors, since it requires no sophisticated equipment. The procedure used in this study could be upgraded in designing and developing a tunnel dryer which employs the same mechanism. In addition, unlike heating the pulp to dry it through the oven, this method maintains the ascorbic acid present in the pulp, since it can easily be destroyed by heating. The method is thus very useful where the pulp is also required for increasing levels of Ascorbic acid in the food that is to be

fortified. This study thus suggests the Sequential stages of dry pulp production as may be used by any upcoming industry interested in production of dietary fiber from pineapples.

Proximate composition of dry pineapple pulp

Findings from this study revealed that crude fiber contributed the greatest percentage composition in dry pineapple pulp (mean: 29.6%) followed by Ascorbic acid (mean: 20.4%). These findings proved dry pineapple pulp to be a reliable source for both crude fiber and ascorbic acid. In line with these findings, Grigelmo (1999) emphasizes the importance of fresh fruits as dependable sources of dietary fiber, in that dietary fiber from fruits is such a significant functional ingredient in food systems as a filler, fat substitute, binder and stabilizer.

He further adds that most fruits have a more balanced dietary fiber profile in terms of soluble and insoluble dietary fiber, as compared to fiber from cereal brans. Several other scholars have also reported that the major natural sources of dietary fiber are fruits and vegetables (Alvarado *et al.*, (2001), Stacewicz *et al.*, (2001), Schauss (2006). According to Schneeman (1987), pineapple contains 0.5 ± 0.03 g/100g soluble dietary fiber and 2.3 ± 0.12 g/100g insoluble fiber, and of the total dietary fiber, $17.8 \% \pm 0.74$ is soluble dietary fiber.

Alvarado *et al.* (2001) has also emphasized that from most studies done on the sources of dietary fiber, the major natural sources of this functional food component have been found to be fruits and vegetables. In the same vein, Venn and Ji (1987) explain that fruits have a more balanced dietary fiber profile in terms of soluble and insoluble dietary fiber as compared to fiber from cereal brans which have been widely used as a fiber supplement in various food applications. Cereal dietary fiber is however, low in soluble dietary fiber.

A study related to this one, by Ackom (2012) also revealed that dried pineapple pulp contains about 23 % and 48 % acid and neutral detergent fiber, respectively. This is closely in line with findings from this study. She thus adds that recovering the fiber for edible uses increases the economic value of pineapple processing and decreases waste. Grigelmo (1999) however warns that one factor which may have negative effects in the food applications of this dietary fiber is the well – known low pH of pineapples, due to the high acid content. He adds that this challenge needs to be addressed for better quality dietary fiber, if pineapple pulp dietary fiber is to have non- limiting applications in the food industry.

The high amounts of ascorbic acid also indicate that air drying which was used is a very appropriate drying method if the dry pulp is required for enriching other foods with ascorbic acid. Unlike other methods, like oven drying, this drying method maintains the nature of ascorbic acid, hence its abundant presence in the dry pulp. According to Hamalatha (2013), pineapple is a rich source of ascorbic acid supplement to our diet. The chemical composition of pineapple flour was in agreement with findings by

Prakongpan *et al.*, 2002 and Ackom and Tano-Debrah (2012).

Proximate composition of pure commercial wheat flour

When proximate analysis was conducted on pure commercial wheat flour, crude fiber was found to be in limited quantities (mean: 0.51%), while the composition of Ascorbic acid in wheat flour was found to be negligible. These results clearly indicate that a reliable source for both crude fiber and ascorbic acid, if used to fortify pure wheat flour, would go a long way in improving its nutritional composition. Since dry pineapple pulp had already been identified as a rich and reliable source for both crude fiber and Ascorbic acid, this study found it suitable for fortifying pure wheat flour with dry pineapple pulp powder. These findings are higher than Oloayee *et al.*, (2006), who found crude fiber composition in pure commercial wheat to be as low as 0.03%. This variation may be due to the difference in level of refinement of the wheat flour. It however still emphasizes the need to fortify wheat flour with dietary fiber, for healthier human consumption. Opoong (2016) however found dietary fiber in wheat to be 0.85%, which is close to 0.51% from this study. He further contends that wheat flour would not be a better source of fiber content since it had significantly lower crude fiber content. This supports the justification from this study, for the need to explore fortifying pure commercial wheat flour with dry pineapple pulp powder. The obtained results of proximal composition however were agreed with Ahmad *et al.*, 2001 who reported that the chemical properties of wheat flours have been studied previously by several researchers and they found that moisture content ranged between 12.5 to 14.6 %, crude protein content 8.23 to 12.71 % and ash content 0.42 to 0.66 (Ahmad 2001).

Proximate composition of pure commercial wheat flour fortified with dry pineapple pulp powder

Generally, composition of both crude fiber and Ascorbic acid significantly increased with increasing amounts of dry pineapple pulp powder added to the wheat flour during fortification ($p= 0.00$). This implies that fortifying commercial wheat flour using dry pineapple powder is a reliable way of enriching wheat flour with dietary fiber and ascorbic acid. This process transforms commercial wheat flour, widely consumed cereal flour in Uganda, into a much healthier food for human health (Ackom, 2012). According to Venn and Ji (2004), fruits like pineapples, have a more balanced dietary fiber profile in terms of soluble and insoluble dietary fiber, as compared to fiber from cereal brans which have been widely used as a fiber supplement in various food applications. The development of this fortified wheat flour in this study was as well in response to an observation made by Dhingra (2012), who asserts that flour products that are rich in dietary fiber are becoming more widely demanded on the market, hence the need to innovatively fortify widely consumed flours with the readily available sources of dietary fiber. With specific regard to enriching wheat with dietary fiber, several epidemiological studies have reported that the consumption of foods that are rich in dietary fiber, say food products prepared from this wheat flour fortified with pineapple pulp, may reduce the risk of cardiovascular diseases, various types of cancer, and type 2 diabetes and possibly improve body function

regulation. It is also known to enhance digestive process, stimulate bowel movements, lower cholesterol, and exert a positive influence on blood sugar levels (Higgins, 2004). Venn (2004) further indicates that dietary fiber may also negatively influence the occurrence of breast cancer. According to Thewissen (2008), dietary fiber has also been highlighted as important in the prevention of diabetes mellitus, obesity, coronary heart diseases, colon cancer and diverticular diseases among others.

Microbiological safety of commercial wheat flour fortified with dry pineapple pulp powder

This study also revealed that for all the three samples tested total coliform counts and moulds were found to be way below the acceptable standard, while yeast levels were slightly high, but still in acceptable range for human consumption. This confirmed that the fortified wheat produced from this study was safe for human consumption, considering the fact that the microbial load of the dehydrated pulp was low, and coliforms were found less absent in the product. Ackom (2012) however cautions that it may be more appropriate to sterilize the raw pulp after the alkalization before the final dehydration to increase the microbial safety, even though the vitamin C content would be affected.

Effect of fortification with pineapple pulp powder on the physical properties of wheat flour

The study revealed that due to the addition of pineapple pulp powder, oil absorption and water absorption capacities of wheat flour decrease with increasing amount of pulp powder used, implying products from the fortified wheat would be increasingly less soggy with water and oil. This makes the fortified wheat applicable in making less oily deep-fried products. Water absorption capacity has been reported to represent the ability of the products to associate with water under conditions when water is limiting, such as in dough's and pastes. Good water absorption capacity would thus be important in foods such as bakery products which require hydration, to improve handling features.

Gelation capacity and foaming capacity on the other hand was found to increase with increasing amounts of pineapple pulp added, implying increasing applicability of the fortified wheat in products that require making dough. According to Tongpun (2006), foam capacity describes the ability of substance in a solution to produce foam after shaking vigorously, usually because the proteins present in the food product foam when whipped, considering the fact that they are surface active. Appiah *et al.*, (2011) add that foaming properties are also useful as indices of the whipping features of protein isolates in a particular food. This implies that fortification of wheat with dry pineapple pulp powder may have slightly increased the amount of protein present in the mixture.

Sensorial preference of fortified wheat products.

When consumer preference for a fried product (*Chapatti*), made from wheat fortified with dry pineapple pulp powder was conducted, the taste panelist indicated that products from wheat fortified with 5% of dry pineapple pulp powder were considered to be having the most appealing taste, overall. This stems from the fact that the chapatti from this

level of fortification had a great texture, did not get soaked with much oil. These even tasted better than the plain wheat samples, implying that fortification of wheat flour with dry pineapple pulp does not only improve the dietary fibre and Ascorbic acid composition of wheat, but also the taste of deep fried products made there from.

The more appealing taste of deep fried products made from wheat fortified with pineapple pulp powder may be explained by the reasons given for taste preference in a similar study done by Ackom (2012). These include the pineapple flavor in the test samples, the sponginess, softness, better mouth-feel in the chapatti made from the fortified wheat flour.

6. Conclusion

Pineapples produced from Isingiro should be highly prioritized for industrial production of dry pineapple pulp, if industrially viable amounts of the dry pulp are to be obtained. In addition, recommend the phase set up of the pulp production industry as you did it in the lab. How should it be set up?

laboratory developed protocols/manual of producing pineapple pulp as indicated in this study need to be up-scaled to develop an industrial set up of the same, for much bigger production volumes, considering that sufficient pineapple is available an underutilized. This could begin from developing a package product that can be tested on the market, so as to inform the clear direction to be taken in up scaling the procedure to industrial level. There is a need to study the cost effectiveness of industrial set up for pulp production and its utilization in fortifying commonly consumed flours in Uganda.

Proximal composition

Further research studies need to be conducted to ascertain the suitability of fortification using this pulp in other flours which are commonly consumed in Uganda such as cassava flour. With respect to Ascorbic acid, utilization of the pulp “in ready to eat” food supplements that do not require heating would be preferred, as this acid would probably be destroyed with prolonged heating.

Air drying should be promoted as the most appropriate method for drying pineapple pulp for fortification purposes. The high amounts of ascorbic acid retained and protein after air drying indicate that drying method is a very appropriate drying method if the dry pulp is required for enriching other foods with ascorbic a Also, view of the fast-growing incidences of behavior related undesired health conditions; cancer, hypertension, among various others, coupled with the growing popularity of wheat as a major component of household diets for all classes of Ugandans, production of wheat flour fortified with pineapple pulp should be up-scaled to industrial scale, and popularized in the local market. Fortifying commercial wheat flour using dry pineapple powder is a reliable way of enriching wheat flour with dietary fiber and ascorbic acid. This process transforms commercial wheat flour, widely consumed cereal flour in Uganda, into a much healthier food option for the fast-growing use of fast foods most of which utilize wheat flour

especially in fried and baked products. Commercial wheat flour intended for use in fried chapatti should only be fortified with up to 5% dry pineapple flour, if the products are to have an appealing taste, better oil and water absorption capacities. Care must also be taken such that the fortified wheat is safe from microbial contamination for human consumption.

7. Acknowledgement

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