



ISSN: 0975-833X

## RESEARCH ARTICLE

### SOYA FORTIFIED YORGHURT – WAY FORWARD TO INDIGENOUS AND PROBIOTIC PRODUCT DEVELOPMENT FOR SUSTAINABLE FOOD SECURITY

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#### ARTICLE INFO

##### Article History:

Received 29<sup>th</sup> October, 2011

Received in revised form

7<sup>th</sup> November, 2011

Accepted 17<sup>th</sup> December, 2011

Published online 31<sup>st</sup> January, 2012

##### Key words:

Biotechnology,

Probiotic,

Fortification,

Indigenous Foods.

#### ABSTRACT

Kenya is endowed with a wide variety of foods. However, there is need to fortify the locally available foods, especially for the vulnerable groups. Designing, creating and monitoring second generation products are prominent opportunities for biotechnology research which increasingly offer new and larger market sectors. This experimental research sought to enhance the deficient iron content in plain yoghurt by fortifying it with locally available ingredients (soya flour) and making use of fermenting action of live bacteria. The activity of live bacteria converts the lactose in the milk into lactic acid, which, because of its acidity, reacts causing the proteins in the milk to solidify. Soy is widely used for fortification of other foods, due to its high protein and iron content. This property was employed in the fortification of yoghurt. The findings of this research show how the nutritional and health value of home made plain yoghurt, by fortifying it with soya flour and culturing with starter containing probiotic bacteria, can be improved to increase specifically the protein and iron amounts in the yoghurt. Production of functional foods like probiotics should thus be encouraged, especially for food aid to deal with consequences of food insecurity like malnutrition.

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#### INTRODUCTION

Over one billion people continue to experience the hardship of hunger, a figure which is on the rise even amidst the riches of the 21<sup>st</sup> century. Engulfed within the vortex of population growth, economic instability and climate change, food security has become the most intractable challenge for national and global governance. Food security is defined by access to sufficient and affordable food; it can relate to a single household or to the global population (FAO, 2009). The first Millennium Development Goal (MDG) falls short of food security aspirations in seeking only to reduce by half the proportion of the world's population experiencing hunger. The first of two benchmarks of measuring progress is the "minimum dietary energy requirement" for each person as stipulated by the UN Food and Agriculture Organization (FAO). This naturally varies by age and sex so that a weighted average is calculated for each country based on its population profile; typically this average is just below 2,000 kilocalories per day. Despite the political commitment to reduce world hunger, the number of people lacking access to this minimum diet has risen from 824 million in the baseline year 1990 to 1,020 million in 2009. These figures for 2009 are collated by FAO from national household surveys conducted in the period 2004-2006, provisionally updated with analysis by the Economic Research Service of the US "when all people at all times have access to sufficient, safe, nutritious food to

maintain a healthy and active life" (WFS, 1996). Commonly, the concept of food security is defined as including both physical and economic access to food that meets people's dietary needs as well as their food preferences.

#### Causes of Food Security Related to Poor Governance

There are three negative direct influences on food security which have been allowed to flourish in the absence of firm governance. The first is a long decline in the scale of investment in agriculture in the developing world; the second is the exercise of inappropriate rules for trade and investment between rich and poor countries. The third is our global tolerance of extreme inequality which in this context permits the diversion of valuable food resources (Food Policy Research Institute, 2009).

#### Neglect of Agriculture

Despite the World Bank estimate that growth of rural economies accelerates poverty reduction four times faster than other sectors, the proportion of foreign aid allocated to agriculture fell from 18% in 1979 to less than 5% in 2007. African governments have likewise neglected their commitment to Maputo Declaration which called for 10% of national budgets to be dedicated to agriculture by 2008. The consequence of this prolonged lack of investment is an inadequate infrastructure for rural economies. Poor roads, irrigation and storage facilities impede efficiencies. Insecure

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tenure and exclusion from affordable credit limit the aspirations of small farmers (Countries in Crisis, Food and Agriculture Organization). There is also neglect in the area of agricultural research, especially in developing countries. This has led to poor food production and minimal innovations in this field. Biotechnology is a major aspect in agricultural research which is at times termed as agribiotechnology. It involves the use of microbes majorly used as vectors and the use of plant and animal cells. Biotechnology is important in increasing crop yields and improving the production frequency in a given season. Production of food crops with superior qualities or characteristics is made possible with biotechnology. Drought and pest resistant crops have been produced through biotechnology together with the production of nutritionally dense food products. With all of the above factors, this paper will focus on improving security, especially with the objective of coming up with a product that will help alleviate malnutrition, more so, in children. All this is done using locally available ingredients to come up with an indigenous product employing biotechnology in innovation. This paper is based on a research done at small scale and the details highlighted therein. The rationale behind this research is to actually improve the nutritional and health value or benefits of home made plain yoghurt. This was done by fortifying it with soy flour and culturing it with a starter containing probiotic bacteria; this with the aim of increasing the protein and iron amounts specifically in the yoghurt. Yoghurt is low in iron content (0.1 mg/100g) compared to soya flour (12.0 mg/100g) (Aileen & Hans, 2007). Yoghurt is low in proteins compared to soya; yoghurt has 21% protein per 100g while soya has 33% per 100g.

### Probiotics

The Greek translation for Probiotic is "for life." A probiotic is a microorganism in a food or supplement containing live microorganisms that are present in sufficient numbers to actively enhance consumers' health by improving the balance of microflora or microorganisms in the gastrointestinal tract. Today, probiotic bacteria, such as *Lactobacillus* and *Bifidobacterium*, are added to fermented foods and other foodstuffs (Aubertin, 2001). Probiotics must be able to survive the aerobic condition of the product in which they are contained, as well as the acidic condition of the stomach. They must be able to survive the bile levels and pancreatic secretions into the small intestine (Shah, 2001). Yoghurt is the most familiar product containing beneficial microorganisms. Yoghurt is the most familiar product containing beneficial microorganisms. There are, however, other foods that may contain added probiotics, such as sour cream, fruit juices and buttermilk. Fermented foods, such as cheese and sauerkraut, which contain beneficial bacteria, have been a part of the human diet for centuries. Probiotics are also available in tablet, spray, capsule, or powder forms. Probiotics must be able to survive the aerobic condition of the product in which they are contained, as well as the acidic condition of the stomach. They must also be able to survive the bile levels and pancreatic secretions into the small intestine (Shah, 2001). Probiotics may have a number of benefits. Certain probiotics have lactase activity, which is needed to properly digest lactose or milk sugar. For example, people that are lactose-intolerant may be able to eat yogurt that contains organisms that produce lactase enzymes (Guo, 2001). Probiotics may

also help keep the intestinal tract more acidic, making it difficult for disease causing organisms or pathogens to persist. Scientists agree that bacteria compete for nutrients in the intestinal tract so if probiotics are present in abundance the more virulent organisms may be crowded out. The immune system may also be stimulated by probiotics. Studies in children suggest that probiotics can help repress infections and allergic responses (Guo, 2001). Probiotics may reduce antibiotic-associated infections and diarrhea. Diarrhea is a common side effect of antibiotic treatment. Antibiotics can destroy both good and bad microorganisms in the intestinal tract. A number of probiotics show promise in alleviating antibiotic related diarrhea. Other health benefits of probiotics, which have been documented or indicated, include suppressing colon cancer, preventing vaginitis, reducing serum cholesterol, improving bowel regularity and maintaining remission in inflammatory bowel disease.

### Benefits of Yoghurt

Not only is yogurt a wonderful quick, easy and nutritious snack that is available year-round, but researchers are finding evidence that milk and yogurt may actually add years to your life as is found in some countries where yogurt and other fermented dairy products (like kefir) are a dietary staple. Yogurt is a fermented dairy product made by adding bacterial cultures to milk, which causes the transformation of the milk's sugar, lactose, into lactic acid. This process gives yogurt its refreshingly tart flavour and unique pudding-like texture, a quality that is reflected in its original Turkish name, Yoghurtmak, which means "to thicken." As a general rule, we favour low-fat dairy products rather than products made from whole milk although we understand that there are individuals for whom whole milk dairy products may be appropriate. It has nutritional benefits beyond those of milk. People who are moderately lactose-intolerant can enjoy yoghurt without ill-effects, because much of the lactose in the milk precursor is converted to lactic acid by bacterial culture (Yale-New Haven Hospital Nutrition Advisor, 2004). Yoghurt also has medical uses, in particular for a variety of gastrointestinal conditions (Kolars *et al.*, 1999; Adolfsson *et al.*, 2004). Yoghurt is also useful in preventing antibiotic-associated diarrhoea (Ripudaman *et al.*, 2003). One study suggests that eating yoghurt containing *L. Acidophilus* helps prevent vulvovaginal candidiasis, though the evidence is not conclusive (Ringdahl, 2000). Yoghurt is believed to promote good gum health, possibly because of the probiotic effect of lactic acids present in it (dentalblogs.com, 2008). A study published in the International Journal of Obesity (2005) also found that the consumption of low fat yoghurt can promote weight loss. In the trial, obese individuals who ate 3 servings of low fat yoghurt a day as part of low calorie diet lost 22% more weight than the control group who only cut back on calories and did not have extra calcium. They also lost 81% more abdominal fat (Dairy Augmentation of Total and Central Fat Loss in Obese Subjects, 2004). Yoghurt is excellent for preventing yeast infections, strengthening the immune system and relieving ulcers.

### A Summary of the Health Benefits of Probiotics

This yoghurt drink contains probiotic cultures and omega-3 fatty acids. Probiotics are live micro-organisms which may help maintain a healthy intestinal flora. Probiotic products

usually contain *Lactobacillus* and *Bifidobacterium* bacteria, both of which are normal inhabitants of the human digestive system. Prebiotics are non-digestible food ingredients which may be beneficial to the colon by selectively stimulating the growth and/or activity of probiotic bacteria in the colon. They are principally oligosaccharides. Omega 3 fatty acids are essential for fundamental human biochemical processes, such as growth and repair.

In addition to promoting healthy digestion and boosting immunity, studies have shown that probiotics may:

- Lower cholesterol
- Regulate hormones
- Reduce symptoms of Irritable Bowel Syndrome (IBS)
- Relieve symptoms of thrush
- Prevent eczema in children
- Shorten the duration of diarrhoea
- Prevent and treat vaginal yeast infections
- Decrease risk of certain cancers e.g. prostate and cervical.

## MATERIALS AND METHODS

### Research Design

An experimental research design was used in this study. The design was important in facilitating product formulation using the various ingredients and their subsequent mixing in the laboratory. The laboratory setting or environment allowed the testing of different parameters of this product. This type of research design made it easy to obtain information, and was appropriate and effective. The experimental design is also flexible, economical and minimizes bias and maximises the reliability of the data collected and analyzed.

### Study Area and Target Population

The Soya fortified yoghurt was subjected to a sensory testing to ascertain its potential consumer acceptance in the market. Sensory testing was conducted at Chepkoilel campus among the students' population. Chepkoilel campus is located 9 kilometres from Eldoret town (the CBD) along the Eldoret-Ziwa-Kitale road. The campus was formerly Moi Teachers College but is now a constituent college of Moi University and a science-based campus that has grown steadily from initial one Department of Forestry to 5 Schools and 16 Academic Departments with a student population of over 6,000 students. It has a well stocked library (another is under construction), laboratories, hostels and lecture halls. The major activities in the campus is lecturing and learning, although there are other activities such as farming, that is, plant cultivation, animal husbandry and bee keeping. The research was conducted in the Food Laboratory under the School of Agriculture and Biotechnology, Department of Family and Consumer Sciences. A target population of 90 informants (sensory panellist) was determined to provide data for subjective analysis of the product.

### Sampling Design and Procedures

Out of the target population of 90 students, two groups were formed. One group consisted of 36 students and the other had 54 students. The group of 36 students was selected using

purposive/judgemental sampling design, a non-probability sampling design. This group consisted of those students pursuing BSc. Food Science and Nutrition and BSc. Food Service Technology academic programmes who were fourth-years. Simple random sampling together with snow-ball sampling technique was then used to get the 2<sup>nd</sup> sample of 54 students from the general student community of Chepkoilel.

### Data Collection Method

The soya fortified yoghurt was produced and subjected to an acceptability test, the data of which was collected through the use of questionnaires. The questionnaires were made up of close-ended items to ease answering and data analysis. In general, data was collected using consumer panels (a transitory consumer panel).

### Equipments and Ingredients Used

#### Equipment

- Plastic jugs (2)
- Weighing scale
- Oven
- Thermometer
- Sieve
- Measuring cylinder
- Metal spoon
- Pots (2) or medium sized sufurias
- Double boiler
- Containers
- Stirrer
- Refrigerator
- Lid or plastic wrap

#### Ingredients

- One quart milk
- ¼ to ½ cup non-fat milk e.g. skim milk powder. (optional)
- 2 tablespoon existing plain yoghurt with live cultures or freeze dried bacteria.
- Sugar (30g)
- Soy flour (40g)
- Clean boiled water.

### Experimental Set-up for Product Formulation

- Heat milk to 185°F (85°C) using two pots that fit inside one another to create a double boiler or water jacket effect. This will prevent the milk from burning. Stirring should be done occasionally. If this cannot be done and you must heat the milk directly, be sure to monitor it constantly, stirring all the while. If you do this, have a thermometer; 185°F (85°C) is the temperature at which milk starts to froth.
- Cool the milk to 110°F (43°C). The best way to achieve this is by a cold water bath. This will quickly and evenly lower the temperature, and requires only occasional stirring. If cooling at room temperature or in the refrigerator, you must stir more frequently. Do not proceed until milk is below 120°F (49°C), and do

not allow it to go below 90°F (32°C). 110°F (43°C) is optimal.

- Warm the starter and then let the starter yoghurt sit at room temperature while you are waiting for milk to cool. This will prevent from being too cold when added.
- Add non-fat (skimmed milk powder) dried milk, if desired. About ¼ to ½ cup non-fat dry milk at this time will increase the nutritional content of the yoghurt. The yoghurt will also thicken more easily. This is especially helpful if you are using non-fat milk.
- Add the starter; add 2 table spoons of the existing yoghurt, or add the freeze dried bacteria.
- Put the mixture in containers. Pour your milk into a clean container(s). Cover each one tightly with a lid or plastic wrap. Allow the yoghurt bacteria to incubate, keep the yoghurt warm and still to encourage bacterial growth, while keeping the temperature as close to 100°F (38°C) as possible. An oven with a pilot light is one option. After 7 hours, you'll have a custard-like texture, a cheesy odour, and possibly some greenish liquid at the top. The longer it sits beyond 7 hours, the thicker and tangier it will become.
- Filter the soy flour (40g), i.e. make it to a paste then heat to boiling point after adding some one litre of water. This is to remove any anti-nutritional factors from the soy.
- Add the 30g of sugar to improve the taste.
- Add the soy flour "liquid" to the cooled yoghurt and agitate or mix thoroughly by stirring.
- Refrigerate the yoghurt. Place it in the fridge for several hours before serving.
- Additional flavours can be added, e.g. vanilla essence (25g) and jams and syrups among others.
- Package in containers or bottles (500ml).

### Microbial Count Determination

- A sample of soy yoghurt was used as the neat solution to be serially diluted. 1 ml of the yoghurt was pipette into 9 ml of distilled water in a test tube and shaken to mix.
- 1 ml of this dilution was taken and pipette into another test tube containing 9 ml of distilled water to make 10<sup>-2</sup> dilution. This process continued up to 10<sup>-4</sup> dilution.
- 0.1 ml of 10<sup>-3</sup> and 10<sup>-4</sup> dilutions was pipette on a solidified agar in Petri dishes and incubated at 25°C for 24 hours.
- The number of colony forming units (CFUs) was determined. Each colony was assumed to have resulted from growth of an individual bacterium. Therefore, three numbers of bacteria.
- The colonies were counted using the manual colony counter.
- The number of bacteria in the original samples was determined by multiplying by 1000 for 10<sup>-3</sup> dilution and by 10000 for 10<sup>-4</sup> dilution to obtain the number of bacteria in 0.1 ml of the original sample.

- It then multiplied by 10 to obtain the number of bacteria in 1 ml. An average of the two dilutions was obtained to come up with the microbial count.

### Shelf Life Determination

Static test was used to determine the shelf life of the product. It was defined or done by storing the product under a given set of environmental conditions selected as representative. The product was placed in refrigerated conditions and assessed daily. Foul smell started or was detected first after three-and-a-half weeks. But three weeks (3) was taken to be on the safe side.

### pH determination

The product was taken to a chemistry laboratory and by use of pH metres; the pH was ascertained to be 5.0.

## RESULTS

### Number of Participants

The number of participants in the study were determined in relation to their age groups and the courses they studied. Among those who were aged 15-20 years, 11 studied agriculture, 13 studied science, 5 studied business related courses, and 8 studied education related courses. Among those who were aged between 23 and 25 years, 29, 4, 3 and 2 participants studied courses related to agriculture, science, business and education respectively. For those who were above 25 years of age, 13 of them studied agriculture, 2 studied science, none studied business and 2 others not studied and education related courses.

Figure 3: TASTE RESULTS.

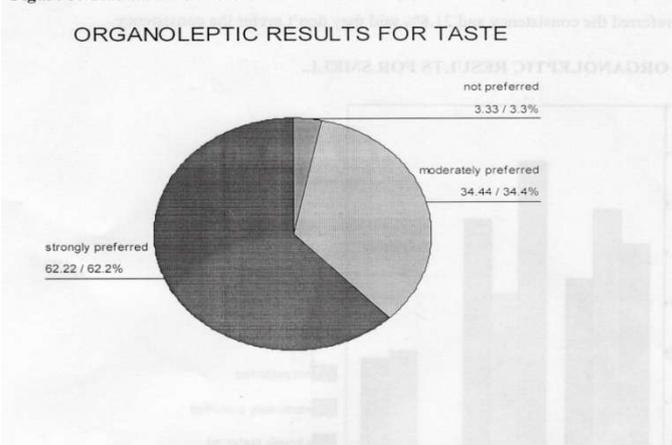
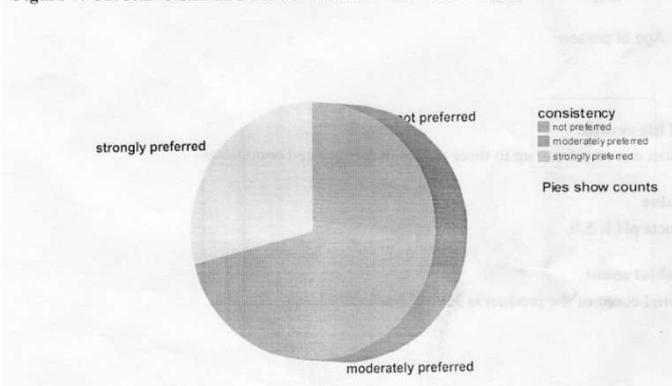


Figure 4: ORGANOLEPTIC RESULTS FOR CONSISTENCY



## Organoleptic Results for Colour

The participants of different age groups showed varied degrees of colour preferences, and the results were presented in form of percentages. Among those aged between 15 and 22 years, 40% did not prefer colour, another 40% moderately preferred colour and 43% strongly preferred. Among those aged between 23 and 25 years, 20% showed no colour preference, 47% and 40% showed moderate and strong preference respectively. Among those above 25 years of age, 40% not showed no preference, 13% not moderately preferred and 18% strongly preferred. 50% of the respondents said they moderately preferred the consistency, while 28.2% said they strongly preferred the consistency and 21.8% said they don't prefer the consistency.

Figure 5: ORGANOLEPTIC RESULTS FOR SMELL.

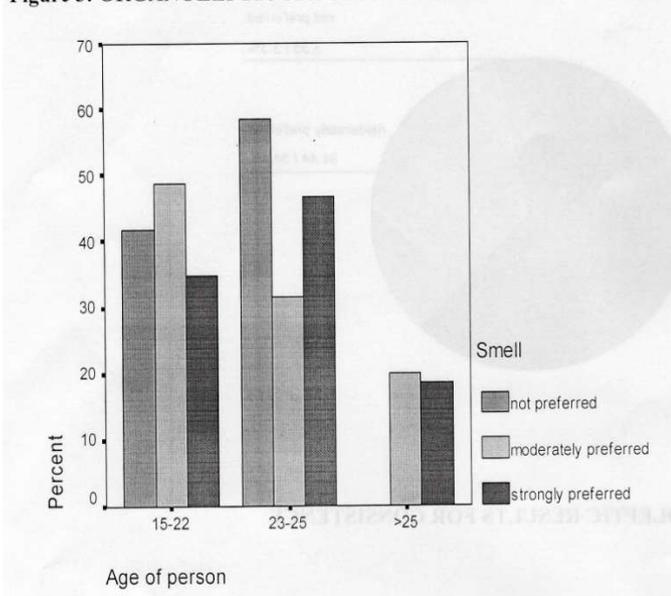


Table 1: Analysis of the Food Record

Food	Amount	energy	Carbohydrate.
Cow's certified milk, min. 45% fat (dry matter) 1000g	1000 g	671.6 kcal	48.0 g
Sugar	80 g	324.5 kcal	79.8 g
Drinking water	250 g	0.0 kcal	0.0 g
Soya bean flour (excess oil removed) bitter principle	120 g	236.0 kcal	0.7 g
Skimmed-milk powder	125 g	460.4 kcal	64.4 g
Yoghurt partially skimmed	20 g	9.2 kcal	0.8 g

Meal analysis: energy 1701.7 kcal (100%), carbohydrate 193.8 g (100%)

## Shelf Life Results

The product can stay fresh up to three weeks in refrigerated conditions.

## pH Value

The products pH was 5.0.

## Microbial Count

The bacterial count of the product was  $35 \times 10^4$  bacteria/ml.

Table 2: Results

Nutrient	Analyzed Value	Recommended Value/day	Percentage Fulfilment
Energy	1701.7 kcal	2036.3 kcal	84%
Water	1150.4 g	2700.0 g	43%
Protein	132.3 g (32%)	60.1 g (12%)	220%
Fat	40.9 g (21%)	69.1 g (< 30%)	59%
Carbohydr.	193.8 g (47%)	290.7 g (> 55%)	67%
Dietary fiber	33.0 g	30.0 g	110%
Alcohol	0.0 g	-	-
PUFA	2.3 g	10.0 g	23%
Cholesterol	144.8 g	-	-
Vit. A	456.7 µg	800.0 µg	57%
Carotene	0.2 mg	-	-
Vit. E (eq.)	1.4 mg	12.0 mg	12%
Vit. B1	2.3 mg	1.0 mg	233%
Vit. B2	4.9 mg	1.2 mg	412%
Vit. B6	1.4 mg	1.2 mg	121%
Tot. fol. acid	488.1 µg	400.0 µg	122%
Vit. C	29.7 mg	100.0 mg	30%
Sodium	1242.0 mg	2000.0 mg	62%
Potassium	6173.6 mg	3500.0 mg	176%
Calcium	3134.3 mg	1000.0 mg	313%
Magnesium	634.9 mg	310.0 mg	205%
Phosphorus	3002.0 mg	700.0 mg	429%
Iron	16.2 mg	15.0 mg	108%
Zinc	12.1 mg	7.0 mg	173%

## CONCLUSION

It is possible to fortify yoghurt with soy flour thereby increasing its iron and protein content. This process can be sustainable because the production costs are very low and the required ingredients are available locally. All these will be handy in improving food security in the aspects of food availability and accessibility. The product's shelf life is approximately 3 weeks, but it can be further improved through other means like adding preservatives, this makes it valuable, especially during transportation to regions of food insecurity. It is evident that developing countries are not doing enough in terms of investing in agriculture, and this has led to food insecurity in most of them. Biotechnology, which is one of the many tools of agricultural research and development, could contribute to food security by helping to promote sustainable agriculture centred on smallholder farmers in developing countries. Yet, biotechnology is now a lightning rod for visceral debate, with opposing factions making strong claims of promise. Others think that biotechnology is all about genetically modified organisms and so do not involve themselves in agribiotechnology. Many developing countries have in one time or another been involved in biosafety debate and chosen not to engage in biotechnology. This research will be instrumental in encouraging research in agriculture.

## RECOMMENDATIONS

Food security involves a myriad of other sectors that influence or affect agriculture, and an agricultural revolution that is environmentally sustainable as well as yield-increasing could help future food needs. This revolution will need the political good will to remove policy distortions that discriminate against poor people, investments in rural health and education, as well as rural roads and credit institutions. Governments and other relevant NGOs should be involved in high-quality research, within which biotechnology will have an increasing role in terms of agricultural production. Trade policies, especially by the World Trade Organization (WTO) on agricultural agreements should allow developing countries to re-evaluate and raise tariffs on key products to protect national

food security and employment. There should be rights-based approach to food security which would impose obligations on national governments to implement non-discriminatory and non-political strategies to enable their people to feed themselves. Countries should embed the right to food in their constitutions. African governments have likewise neglected their commitment to the 2003 Maputo Declaration which called for 10% of national budgets to be dedicated to agriculture by 2008. Hence, investment should be increased in African countries by 10% and even more. The 2009 G8 Summit in Italy promised \$20 billion spread over three years for investment in agriculture and developing countries should take up this opportunity. Food should be subject to the same market forces as manufactured goods with minimum state involvement. Small "peasant" farms should be consolidated and alternative livelihoods found for surplus labour. Support and encouragement of small-scale farmers and food processors, in terms of incentives or any other way, should be given priority to improve production of food. Larger farms can then raise capital for the expensive products of modern biotechnology and compete in export markets. Countries should exploit the competitive advantage of crops that grow best in their climate. Use of readily available indigenous foods that are nutritious and promising in alleviating malnutrition should be grown or produced. Production of functional foods should be encouraged especially for food aid to deal with consequences of food insecurity like malnutrition.

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