Detarium microcarpium Bread Meal: It's Physiological Effects on the Postprandial Blood Glucose and Insulin Levels of Healthy Non Diabetics Subjects

Onyechi, U. A.
Department of Home Science Nutrition and Dietetics, University of Nigeria Nsukka, Nigeria.
Email: uche.onyechi@yahoo.com. **Phone:** +23408066794814

Abstract

This work studied the effect of Detarium bread meal on the postprandial plasma glucose and insulin levels of healthy participants. The subjects of the study were ten healthy non diabetic male subjects who were fed two meals, an experimental bread meal containing detarium flour and a control bread meal made from wheat flour only. The test meals consisted of two small bread rolls, 38g of apricot jam (Robinson's) and water to make a total meal weight of 400g. The available carbohydrate portion of the meal was 75g. The bread rolls contain 50g carbohydrate mostly in the form of starch. The jam provided 25g of available carbohydrate in the form of sucrose. The experimental breads rolls provided 5g of s-NSP as calculated from the nutrient analysis plus s-NSP from the brown flour. The subjects visited the metabolic kitchen twice a week after an overnight fast. All the subjects ate the two types of meals detarium and control bread meal in random order. The subjects were weighed and their heights were taken. A three days food record was kept to ensure adequate carbohydrate intake. Fasting blood samples were taken. Postprandial blood samples were taken from the subjects at 30, 60, 120, and 150 minutes from the commencement of the meal. The blood samples were analyzed for glucose and insulin levels. Data obtained were analyzed using ANOVA. The results of the incremental plasma glucose level showed a significant bread meal effect (Wilks' Lambda 11.1 df 6 and 18; p=0.0049) and a significant time effect at (p=0.0129). The result showed a significant difference between the effect of the detarium bread compared to the control bread meal at (p=0.0008). The result also showed there was a significant difference for Detarium bread meal at 90, 120 and 150 minutes when compared to the control bread meal. ANOVA also showed a significant meal effect on the incremental insulin levels (Wilks' Lambda 16.0; df 2, 18; p=0.0016) and a significant time effect (p=0.0230). There was a significant difference on the plasma insulin levels between the control and the Detarium bread meals (p=0.0022). Detarium bread showed a significant difference on the plasma insulin levels at 30, 60 and 150 minutes.

Keyword: Detarium, Bread meal, Plasma glucose, Plasma insulin, Healthy subjects

Introduction

Incorporation of Detarium flour into traditional Nigerian soup which was fed to healthy subjects resulted in significant lower post-prandial plasma glucose levels compared to the control meal (Onyechi, 2009). Lower plasma insulin levels were also seen after consumption Detarium soup meal compared to the control meal, though the result was not significant (Onyechi, 2009). However, the consumption of this type of traditional soup using thickeners like Detarium has been on the decline in most urban areas. These soup meals are consumed more in the rural areas while urban meals are getting very westernized (Onyechi, 1995). An attempt was therefore made to find an acceptable, palatable food vehicle for Detarium flour.

Studies have been done to introduce soluble non starch polysaccharide (s-NSP) into a variety of different foods (Fuessl *et al*., 1986; Peterson *et al*., 1987; Fairchild *et al.*, 1990). A wide variety of foods have been developed like crisp bread (Jenkins *et al*., 1978a); biscuits (Ellis *et al*., 1988); pasta (Tognarelli *et al*., 1986); wheat bread (Apling and Ellis 1982; Peterson *et al*., 1987) and breakfast cereals (Fairchild *et al.*, 1996). The most commonly used s-NSP has been guar gum, a galactomannan extract of the Indian cluster bean cyamopsis tetragonoloba which is normally produced as flour for commercial use. Guar containing wheat bread was one of the early food products to be investigated in acute and long term studies (Apling and Ellis, 1982). Most clinical studies in the literature have indicated that wheat bread containing guar gum improves carbohydrate tolerance and causes a reduction of plasma cholesterol levels in healthy and diabetic subjects (Ellis *et al*., 1991; Peterson *et al.*, 1987). Wheat bread was considered to be a suitable medium for incorporating guar gum because it could be used as a part of any meal. Bread has been classified as a high glycaemic index food (BNF, 1990). However adding polysaccharide gum to wheat bread transforms the bread into a food with low or medium glycaemic index food similar to legumes or pasta (Ellis, 1994).

On the basis of these results detarium flour was incorporated into wheat flour to produce bread rolls. The physiological effect of Detarium bread meal was examined to determine the effect on post-prandial blood glucose and insulin levels in healthy subjects. This novel approach could break the monotony of using detarium flour soups only.

The aim of the study was to provide variety in the use of locally available foods that is high in s-NSP in Nigeria. *Detarium microcarpium* locally known as "offor" is leguminous crop seed belonging to the Subdivision Caesalpinoideae (Balogun and Fetuga, 1986). Preliminary studies (Onyechi *et al*., 2007a) on the composition and nutrient analysis of Detarium showed that detarium was high in s-NSP

(55.9g/100g). *Detarium* has been shown to have similar rheological properties as medium grade guar gum (M90) (Onyechi, 1995). Both detarium and guar gum had the same physiological effect on rats as both plants significantly reduced the fasting plasma cholesterol level of rats (Onyechi *et al.*, 2007b). Detarium soup meal also had significant reduction on the incremental plasma glucose level in healthy non-diabetic subjects (Onyechi, 2009). These results indicated a positive physiological effect of detarium.

Detarium flour was therefore incorporated into wheat flour to produce bread rolls. These were fed to healthy non-diabetic subjects. The physiological effects on postprandial incremental blood glucose and insulin levels were determined and compared to control bread meal effect.

Materials and Methods

Subjects: Ten healthy non-diabetic male subjects from King's College, London, participated in the study. Written information was given to each subject and consent forms were signed. The General practitioners (GP) of the participants were written to ascertain their health status with respect to the study. The protocol was approved by the King's College Research and ethical committee.

Preparation of detarium bread: The composition and quantity of the ingredients used in the preparation of the bread is shown in Table 1. Chorleywood bread process (Apling and Ellis, 1982) was used in the preparation of the bread rolls. Ploughman's brown flour (Sovereign, Allied Mills, London) was the type of flour and flora brand (Unilever, UK) was the hydrogenated vegetable fat used. Each batch of the bread rolls contained variable amounts of water depending on the viscosity of the flour. Detarium flour was incorporated into the bread as a replacement for wheat flour. The weight of the dough was calculated such that a total of 50g carbohydrate was contained in the two bread rolls. Each detarium bread roll contained 2.5g s-NSP. Two hours after baking the bread rolls were frozen in self sealed freezer bags at -20^{o}C until required for experimental use.

Table 1: Food ingredients used in the preparation of control bread and detarium bread rolls.

Ingredients	Quantity of ingredients (g/1000g flour)	
	Control bread	Detarium bread
Brown flour	1000	850
Salt	18	18
Detarium flour	0	150*
Fat (hydrogenated)	7	7
Improver	100	100
Fresh yeast	25	25
Water	675	900

** Equivalent to 63g soluble fibre (Onyechi, 1995)*

Glucose analysis: The glucose and insulin increments (changes relative to fasting values) were determined at 30, 60, 90, 120, and 150 minutes after the subjects had consumed detarium and control bread rolls. The plasma glucose was measured by standard glucose oxidase method (Werner *et al.*, 1970) using a Bushrangers Mannhein kit (Boehringer Mannheim House, Bell Lane, Lewes BN7 1LG). The frozen deproteinized plasma was allowed to thaw and mixed in a rotamixer for 2 minutes. A 100 ul of the supernatant was mixed with 5 ml of the reagent which contains buffer, enzymes and chromogen. The sample was mixed in a rotamixer and incubated in a water bath at 20-25^{o}C for 40 minutes avoiding direct exposure to sunlight. The absorbance of the sample and the standard were measured against a blank in a spectrophotometer at 610 nm.

Insulin analysis: The Boehringer Mannhein diagnostic kit based on enzyme immunological reactions was used for the quantitative determination of human insulin *in-vitro*. The ES 22 combi step analyzer program B auto machine was used. Precipath IM was the quality control serum used to run each analysis and values were within the stipulated range. Five standards were used which ran in duplicates along with duplicate samples of the control and test sera. The machine automatically dispensed and washed out the tubes with reaction solutions. The tubes were automatically read after the incubation period by passing along a conveyer into the spectrophotometer, where the solution is aspirated out and absorbance plotted. A computer program was used to read the blank, standard, control serum and the test sample for each run. The absorbance readings were calculated in the calculation mode and the standard curve plotted. The concentration of plasma insulin in the test sample was calculated from the standard curve.

Statistical analysis: The difference between the effects of the control and detarium bread meals on the blood glucose and insulin were analysed by repeated measure of analysis of variance, ANOVA, SAS Statistical package, (SAS Institute Inc., 1985). Significance difference between the control and the detarium bread meals were accepted at $p<0.05$.

Results

Subjects: Ten subjects between the ages of 21 and 39 years participated in the study. The body weight varied between 57 and 94 kg and calculation of the BMI showed that all but three were within desirable range of weight for height. Each subject consumed two small bread roll of control and detaruim that weighed 98g and 125g each respectively.

Nutrient composition of the bread rolls by calculation: The nutrient composition of the bread rolls indicated that the detarium bread had more moisture and fat than the control bread, while the control bread had more protein and carbohydrate than the detarium bread (Table 2). Weight of cooked dough to provide 50g carbohydrate was lower than the raw dough, 98g and 125g respectively for control and detarium bread rolls. The proximate analysis indicated that the control

bread rolls had higher protein (7.8g) than the detarium bread (6.8g). With regards to energy, the control bread had higher energy profile than the detarium bread (Table 2).

Table 2: Composition (g/100) of raw bread dough by calculation

Nutrients	Quantity of nutrients (g)	
	Control bread	Detarium bread
Moisture	39.1	46.1
Protein	7.8	6.8
Fat	0.4	0.8
Fibre	4.2	4.2
Available carbohydrate	68.6	60.9
Total energy Kcal/100)	309.6	276.3

Weight of the raw dough used to provide 50g carbohydrate was 126g (Control bread) and 160g (detarium bread).

Effect of the bread meals on the plasma glucose levels of the subjects: The result showed that fasting blood glucose levels were found to be within the normal range for non-diabetic subjects. The pooled mean of the fasting blood glucose level for the subjects was 4.30mmol/L. The post-prandial rise in blood glucose levels was expressed as incremental blood glucose levels which were calculated relative to the fasting values. The mean incremental blood glucose levels were shown in Table 3.

Table 3: Plasma glucose levels subjects fed Control bread and Detarium bread meals.

Time (min)	Plasma glucose level (mmol/L)	
	Control bread meal	Detarium bread meal
30	1.30 ±0.18	0.64 ±0.11
60	1.62 ±0.16	0.94 ±0.12
90	1.61 ±0.15 [a]	0.58 ±0.08[b]
120	0.31 ±0.25 [a]	0.13 ±0.07[b]
150	0.94±0.16 [a]	0.12 ±0.06[b]

Superscripts are significantly different means.

Analysis of the data using ANOVA showed a significant bread meal effect (Wilks' Lambda 11.1; df 6 and 18; (p = 0.0049) and a significant time effect at (p = 0.0129). Comparison of the mean incremental blood glucose rise after the consumption of the control bread and the detarium bread showed a significant difference between the detaruim and control meals at (p = 0.0008). When the difference between the bread meals was analyzed at each time interval, the result showed that there was a significant difference for detarium bread at 90, 120 and 150 minutes when compared with the control.

Effect of the bread meal on the plasma insulin levels: The result showed that the pooled mean fasting insulin levels was 15.07Uu/ml and within the normal range for non diabetics. Incremental plasma insulin levels were calculated relative to the fasting levels at all postprandial times and shown in Table 4. ANOVA showed that there was significant bread meal effect (Wilks' lambda 16.0; df 2,18 p = 0.0016).

Table 4: Plasma insulin levels of subjects fed control bread meal and Detarium bread meal

Time (min)	Plasma insulin level (Mu/L)	
	Control bread meal	Detarium bread meal
30	22.16 ±3.55	10.84±2.47
60	24.16 ±2.69	17.46 ±3.97
90	36.71[a] ±5.76	24.92[a] ±4.44
120	14.99 [a]±4.25	5.28[a] ±1.41
150	4.34 ±2.01	1.47 ±0.47

There was also a significant time effect (p = 0.0230), when the control bread was compared to the detarium bread.

Discussion

The result of this study showed that when healthy subjects were fed bread meals containing 75g carbohydrate and 6g s-NSP from detarium flour, there was a significant main meal effect (p=0.0049) compared to the low fibre control bread. When the incremental plasma glucose level of detarium was compared to the control bread, a significant lowering effect was found after the consumption of detarium bread (p=0.0008) at 90,120 and 150 minutes post-prandially. Comparison of the effect of the control bread and the detarium bread on insulin level also showed a significant meal effect between the detarium bread and the control bread (p=0.0016). The detarium bread showed a significant lowering effect (p=0.0016) at 90 and 120 minutes. The detarium bread meal proved to be significantly effective in lowering plasma glucose and insulin levels. Jenkins *et al.*, (1980) listed the mechanisms by which guar gum modulates postprandial hyperglycaemia and hormone responses. These mechanisms may be similar to the mechanism by which detarium lowered postprandial glucose and insulin levels. Detarium and guar gum are strikingly similar. Both are legumes and contain s-NSP. Both have similar rheological properties and detarium has a molecular weight similar to medium grade guar gum (Onyechi, 1995). These mechanisms of action include viscosity effect. Jenkins *et al.*, (1978a) showed a positive correlation between the peak rise of blood glucose and 2-hour post-prandial glucose level with the viscosity of four viscous NSP, guar, tragacanth, pectin and methyl cellulose in healthy subjects. Reduction of insulin response – Jenkins *et al.*, 1976 showed that fibre enriched meal produced a marked flattening of the postprandial glycaemia and insulin response. Other studies in the literature have observed similar effects with high NSP meals (Jenkins *et al.*, 1977; Wolever *et al*., 1979; Morgan *et al.*, 1979). Jenkins *et al.*, 1980 indicated that there was overall flattening of the endocrine response induced by viscous NSP as shown by Morgan *et al*., (1979) in which GIP response was flattened after guar supplementation. Slow absorption – Jenkins *et al*., (1978b) indicated that the depressed postprandial glycaemic response with s-NSP was due to slow absorption rather than malabsorption. These results were later confirmed by Leeds *et al*., (1978) who showed that guar did not cause carbohydrate malabsorption due to the

absence of hydrogen breath in the subjects. Gastric emptying small intestine absorption, Jenkins *et al*., (1980) pointed out that delayed mouth to caecum transit times are associated with ingestion of the viscous NSP and showed a significant relationship between mouth-to-caecum transit with viscosity (Jenkins *et al*., 1978). Holt *et al*., (1979) suggested that delayed gastric emptying may be of great importance in flattening post-prandial glycaemia. Jenkins *et al*., (1978b) explained that viscous NSP prevents gastrointestinal hurry and slows the rate of nutrient absorption resulting in flatter post-prandial levels of glucose, metabolites and hormones.

It is possible that the positive physiological effect of detarium in modulating postprandial glucose and insulin profile could be attributed to the above mentioned mechanisms listed by Jenkins *et al.*, (1980). Detarium in addition to having the same physiological properties as guar gum both s-NSP had the same effect on the rats. Both detarium and guar gum covariates such as weight gain, food intake, faecal output and they both lowered the plasma cholesterol levels of rats Onyechi *et al*., (2007b). The similarity of effects of detarium to guar gum suggests this indigenous food could be a useful adjunct to the treatment of diabetes mellitus in Nigeria and detarium bread rolls may provide a variety to the diet of the diabetics that resides in the urban areas.

Conclusion: The result of the study showed a significant lowering effect on the post-prandial glucose and insulin levels of healthy non diabetic subjects. On the basis of this result it was decided that detarium could be studied further with non-insulin diabetic subjects. This to ascertain if there would be any improvement in the postprandial glucose and insulin profiles of the diabetics.

Acknowledgements

The author gratefully acknowledges Professor P.A. Judd for her supervision, Dr. P.R. Ellis for his contributions and help, Professor H.N. Ene-obong for her help in the purchase of the foods; my colleagues at King's College, London, Mrs Rose Colokasia and David Lincoin for their expert technical assistance; Professor Simon Ross-Murphy for his helpful discussion on the physico-chemical properties of the polysaccharides; Mr Peter Milligan for advice on statistics. The author also is grateful to Dr. Hans Englyst (MRC Dunn Clinical Nutrition Centre, Cambridge) for his help with the analysis on the NSP and starch of the foods. This work could not have been possible without the support of Association of Commonwealth Universities that granted a scholarship to the author.

References

Apling, E.C. and Ellis, P.R. (1982). Guar bread: Concept to application*. Chemistry and Industry*, 23: 950-954.

Balogun, A.M. & Fetuga B.L. (1986) Chemical composition of some under-exploited leguminous crop seeds in Nigeria. *Journal of Agricultural Food Chemistry*, 34:189-192.

British Nutrition Foundation (BNF) (1990) *Complex carbohydrate in foods.* The report of the British Nutrition Foundation's Task Force Chapman &Hill, UK.

Ellis P.R. (1994) Polysacchride gums: their modulation of carbohydrate and lipid metabolism and role in the treatment of diabetes mellitus. *In: Gums and stabilizers for food industry*. No 17, pp207-216 (J.O.Phllips. PA Williams and JD Wedlock). Oxford University Press, Oxford.

Ellis, P.R. Dawoud,F.M. and Morris, E.R. (1991) Blood glucose, plasma insulin and sensory responses of guar containing wheat breads: effects of molecular weight and particle size of guargum*. British Journal of Nutrition*, 66: 363-379.

Ellis, P.R., Kamalanathan, T. Dawoud. F.M., Strange, R.N. and Coultate, T.P. (1988) Evaluation of guar biscults for use in the management of diabetes: tests of physiological effect and palatability in non-diabetic volunteers. *European Journal of Nutrition*, 42: 425-435.

Fairchild, R.M., Ellis, P.R., Luzio, S., Byrne, A., Mir, M.A (1996) A new breakfast cereal containing guar gum reduces postprandial plasma glucose and insulin concentrations in normal weight human subjects. *British Journal of Nutrition*, 76: 63-73.

Fairchild, R.M., Daniels, C.E.J., Ellis , P.R., Naqui, S.H.M., Kwan, R.M.F and Mir, M.A. (1990) Effcet of two types of guar gum in solid and liquid foods on postprandial blood glucose, plasma insulin and c-peptides in healthy subjects. *Proceedings of Nutrition Society*, 49: 54A.

Fuessl, H.S., Williams, G., Adrian, T.E., Bacarese-Hamilton A.J. and Bloom, S.R. (1986). Guar in NIDD: effects of different modes of administration on plasma glucose and insulin responses to a starch meal. *Practical Diabetes*, 3: 258-260.

Holt, S., Heading, R.C., Carter, D.C., Prescott, L.F., and Tothill, P., (1979). Effect of gel fibre on gastric emptying and absorption of glucose and paracetamol*. Lancet* , I: 636-639.

Jenkins, D.J.A., Wolever, T.M.S., Tayor, R.H., Ghafari, H., Jenkins, A.L., Baker, H. & Jenkins, M.J.A. (1980). Rate of digestion of foods and postprandial glycaemia in normal and diabetic subjects*. Br Med J.* 281: 14-17.

Jenkins, D.J.A., Wolever, T.M.S., Nineham, R., Taylor, R., Metz, G.L., Bacon, S. and Hockaday, T.D.R. (1978a). Guar crispbread in the diabetic diet. *British Medical Journa*l, 279: 1744-1746.

Jenkins, D.J. A., Wolever, T.M.S., Leeds, A.R., Gassull, A.M., Haisman, P., Dilawari, J., Goff, D.V., Metz, G.L. & Alberti, K.G.M.M. (1978b) Dietary fibre, fibre analogues and glucose tolerance: importance of viscosity. *British Medical Journal*, 279: 1392-1394.

Jenkins, D.J.A., Leeds, A.R., Gassull, M.A., Cochet, G., and Alberti, G.M.M. (1977) Decrease in postprandial insulin and glucose concentration by guar and pectin. *Ann Inter Med*, 86: 20-23.

Jenkins, D.J.A., Leeds, A.R., Gassull, M.A., Wolever, T.M.S., Goff,D.V. Alberti, G.M.M. and Hockaday, T.D.R. (1976). Unabsorbable carbohydrate and diabetes: decreased postprandial hyperglycaemia*. Lancet,* ii: 172-174.

Leeds, A.R., Bolster, N., and Truswell, A.S. (1978) Guar gum anf glucose absorption: Absence of evidence for malabsorption. *Proceeding of Nutrition Society*, 37: 89A.

Morgan, L.N., Goulder, T.J., Tsiolakis, D, Marks, V. and Alberti, K.G.M.M.(1979) The effect of unabsorbable carbohydrate on gut hormones: modification of postprandial GIP secretion by guar. *Diabetologia* 17: 85-89.

Onyechi, U.A. (2009) The effect of Detarium microcarpium soup meal on the plasma glucose and insulin levels of healthy non-diabetic adults. *Nigerian Journal of Nutritional Science.* 30: (1) 10-18.

Onyechi, U.A. Judd, P. A. and Ellis, P.R. (2007a) The processing and nutrient analysis of some unexploited Nigerian plant foods. *West African Journal of foods and Nutrition*, 8: (1) 35-44.

Onyechi, U.A., Bell, S. Judd, P.A. and Ellis, P.R. (2007b) A comparative study of the effectiveness of two unexploited Nigerian plant foods (Detarium and Cissus) to Guar gum as a positive control on cholesterol levels of rats. *West African Journal of foods and Nutrition*. 7: (1) 45-53

Onyechi, U. A. (1995). *Potential role of indigenous Nigerian foods in the treatment of non-insulin dependent diabetes mellitus (NIDDM)*. PhD Thesis. Kings College, University of London.

Peterson, D.B., Ellis, P.R., Baylis, J.M., Fielden, P., Ajodhia, J., Leeds, A.R. and Jepson, E.M. (1987) Low dose guar in a novel food product: Improved metabolic control in non-insulin dependent diabetes. *Diabetic Medicine*, 4: 111-115.

Peterson, D.B, Ellis, P.R., Baylis, J.M., Frost, P.G., Leeds, A.R., J Epson, E.M., (1984) Effects of guar on diabetes and lipids – food and pharmacology compared. *Diabetology*, 27: 319A

Tognarelli, M., Miccoli, R., Giampietro, O., Cerri, M., and Navalesi, R., (1986) Guar pasta: A new diet for obese subjects? *Acta Diabetology*, 23: 77-80.

SAS Institute Inc. (1985) SASUser's Guide: Statistics, 5th ed. SAS Institute Inc. North Carolina USA.

Werner, W., Rey, H.G and Wellinger, H. (1970) On the properties of a new chromagen for the determination of glucose in blood according to th GOD/POD method*. Zetschrift fur Analytische Chemie,* 252:224-228.

Wolever, T.M.S., Jenkins, D.J.A., Nineham, R. and Alberti, K.J.M (1979). Guar gum and reduction of postprandial glycaemia: effects of incorporation into solid foods, liquid foods or both. *British J. Nutrition*.41: 505-510.