African Journal of Biotechnology Vol. 8 (6), pp. 1116-1120, 20 March, 2009
Available online at http://www.academicjournals.org/AJB
ISSN 1684–5315 © 2009 Academic Journals

Full Length Research Paper

Influence of dietary supplementation on biotransformation of locust beans (*Parkia biglobosa*) to condiment

B. Daramola*, O. A. Fasominu, O.J. Oje and O.O. Makanju

Department of Food Technology, Federal Polytechnic P. M. B 5351, Ado Ekiti, Ekiti State, Nigeria.

Accepted 24 December, 2008

Influence of four types of dietary supplements namely; cooked onion extract (COE), Iru water extract (IWE), glucose solution (GLS) and asparagine solution (ASS) on biotransformation of locust beans to condiment was studied. Assessment of biotransformation markers: free amino acids, free total sugar and total titratable acidity showed that supplementation enhanced biotransformation comparatively to sample without supplement (COS). Biotransformation was most enhanced at Day 2 in COE and IWE in comparison to GLS, ASS and COS. Assessment of selected dietary elements revealed that some elements (P, Fe and Mg) decreased during biotraformation. The change may be associated with the necessity of the dietary elements for accomplishment of biotransformation. Analysis of sensory scores (P=0.05) indicated that supplementation (COE, IWE) conferred no adverse influence on organoleptic characteristics of the condiment.

Key words: Locust beans, dietary supplementation, biotransformation markers, dietary elements, natural condiment.

INTRODUCTION

Fermented locust beans (*Parkia biglobosa*) called “Iru” among the people of Western Nigeria is the most important condiment of natural origin in West Africa and Central Savannah region (Odunfa , 1986). The proximate composition indicates that Iru is rich in protein, lipid and minerals. Therefore, the preponderance of application of the condiment in culinary preparations suggests that the flavourant could contribute some nutrients notably minerals and vitamins to nutritional requirements.

Beaumont (2002) reviewed that active microbial metabolism is required to facilitate the culinary changes observed in fermented locust beans. Summarily, conditions for optimum growth of microorganisms are: availability and balance nutrients, supply of gaseous atmosphere such as carbon dioxide, oxygen, and nitrogen, as well as hydrogen ion concentration, temperature and osmotic pressure Olutiola et al., 2000).

Therefore, nutrient enrichment, the most important microbial growth factor in natural fermentation process could accentuate proliferation of microorganisms responsible for conversion of locust beans to condiment. The objective of this study was to evaluate using selected fermentation markers, the influence of dietary fermentation on biotransformation of locust bean to condiment.

MATERIALS AND METHODS

Materials

The dominant raw materials, locust beans (Iru) and Onion (*Allium cepa*) were obtained from a local market. Glucose and asparagine used were of analytical grade.

Preparation of locust bean condiment

Traditional method of production of locust bean condiment (Iru) as reported by Omafuvbe et al. (2004) was adopted with slight modification. African locust beans were manually shelled to obtain the cotyledon prior to water immersion boiling. Shelling was carried out in order to obviate the labourious heating and washing necessary for cooking and removal of cotyledon of beans with seed coat prior to fermentation. The shelled beans (cotyledons) were boiled in

*Corresponding author. E-mail: daramola_bode@yahoo.co.uk.

Table 1. Proportion for preparation and coding of samples for supplementation.

Supplement	Weight (g)/volume (ml) of supplement extraction medium	Level (%) of supplementation of sample and corresponding code		
		1	5	10
Iru	1/2	IWE-1	IWE –2	IWE – 3
Cooked onion	3/4	COE-1	COE-2	COE-3`
Glucose	1/20	GLS-1	GLS-2	NED
Asparagine	1/20	ASS-1	ASS-2	NED

NED = Not included in experimental design.

water for 3½ h. The cooked cotyledon mass was subsequently wrapped hot in black polythene bag and incubated in jute bag and allowed for biotransformation (fermentation).

Supplementation of samples

Supplements were prepared into solution, and added at 1, 5 and 10% volume of extract per weight of cooked locust beans. Both cooked onion and Iru were separately blended using domestic blender and sieved using 0.5 mm size sieve to obtained the extract of each supplements. Also, glucose and asparagine were dissolved in distilled water. Details of proportion for preparation, and coding of samples for supplementation are shown in Table 1. The extracts and solutions were added at the stipulated levels (Table 1) of volume of extract per weight of cooked locust beans and thoroughly mixed prior to wrapping in polythene bag and incubated in jute bag for biotransformation (fermentation) process. Each supplemented sample was wrapped and coded for the day (1 - 3) to be examined such that once a sample was opened, it was discarded.

Determination of free amino acids and soluble sugars

Samples at various processing stage were dried in hot air oven at 70℃, grounded and defatted. The free amino acids in the defatted samples were extracted with 80% ethanol (v/v) in accordance with the method of Odibo et al. (1990). The free amino acids in the ethanolic extract were estimated using the ninhydrin colorimetric method (Rosen, 1957) using glycine as standard.

The soluble sugars were evaluated according to the method of Dubois et al. (1956) with slight modification (samples were not digested using heat). The ethanolic defatted aliquot was filtered prior to addition of phenol and concentrated sulphuric acid and absorbance was measured at 490 nm after 10 min. Sugar content was calculated using a standard curve prepared using 0 – 100 µg glucose.

Determination of titrable acidity

Hot water extracts of milled samples were titrated against standard alkaline in accordance with AOAC (1990) method. Results were expressed in mg lactic acid equivalent/g sample.

Mineral analysis

Analysis of the selected minerals in this study was accomplished using the method of AOAC (1990). Samples were analysed using Alpha-4-(chemtech-analytical)-atomic-absorption pectrophotometer.

Sensory evaluation

Using multiple comparison test, sensory evaluation of the supplemented biotransformed locust beans and non-supplemented samples was carried out by 10 panelists comprised of students of the Dept. of Food technology, Federal Polytechnic, Ado-Ekiti. The panelists were selected based on the history of their familiarity with processing of locust beans condiment. Sensory attributes evaluated were, colour, odour, taste, texture and mucillageneous characteristics using a score scale of 1 to 9 where 1 indicates extremely inferior and 9 indicates extremely superior (Larmond, 1979).

Statistical analysis

One way analysis of variance (ANOVA) using repeated measure was conducted. When significant (P=0.05) difference was observed, means were separated using Tukey's test (Snedecor, 1956).

RESULTS AND DISCUSSION

Influence of dietary supplementation namely glucose (GLS), asparagine (ASS) solutions, cooked onion (COE) and Iru (IWE) extract on biotransformation of locust beans to condiment was investigated. Selection of the supplements was informed essentially by microbial nutritional requirement as follows: glucose as carbon source, asparagine as nitrogen source, cooked onion as nutrient composite source for carbon, nitrogen, vitamins, minerals and others (Sharma and Cavalli, 2002) and Iru water extract as source of condiment inherent biotransformation stimulant or inoculum starter. Progress of biotransformation (fermentation) was evaluated using free amino acids (FAA), hydrolyzed soluble sugars (HSS) and titratable acidity (TTA) as biotransformation markers. Results of HSS and ASS shall were not interpreted for samples in which glucose and asparagine were the supplements.

Supplemented and control samples in this study showed increment in the result of evaluated biotransformation markers at variable amounts (Table 2) on daily basis. This is in agreement with an earlier report of Odunfa (1986), Omafuvbe et al. (2004) on progress of fermentation of *P. biglobosa* without supplement (COS).

The free amino acids (mg glycine/g sample) hydrolysedat day 1 was highest (0.1225 - 0.1313) for COE and

Table 2. Profile of selected markers during biotransformation of locust beans to condiment

Sample	Day 1			Day 2			Day 3		
	FAA	HSS	TTA	FAA	HSS	TTA	FAA	HSS	TTA
IWE –1	5.96	217.00	0.52	13.13	364.00	0.71	15.75	43.00	2.29
IWE-2	6.13	322.00	0.62	16.63	400.70	0.79	12.25	43.00	2.35
IWE-3	6.13	129.00	0.62	6.13	440.30	0.78	14.88	257.00	2.36
COE-1	3.50	214.00	0.53	27.13	429.00	0.65	32.38	450.00	1.87
COE-2	7.00	407.00	0.52	26.25	514.00	0.72	33.25	493.00	1.88
COE-3	12.25	407.00	0.63	28.00	560.00	0.93	21.88	493.00	2.12
GLS-1	1.75	557.00	0.54	8.75	472.00	0.58	7.00	605.20	1.68
GLS-2	13.13	600.00	0.51	2.63	386.00	0.50	1.75	771.5o	1.47
ASS-1	12.25	235.70	0.50	41.13	215.00	0.84	42.00	192.90	1.89
ASS-2	13.25	218.00	0.50	42.07	226.00	0.84	42.10	200.00	1.89
COS	2.63	278.60	0.60	5.25	257.00	0.78	15.25	320.00	1.88

Results are average of two determinations.COE = cooked onion extract, IWE = Iru water extract, GLS = glucose solution, ASS = asparagine solution, HSS = soluble sugars (µg glucose equivalent/g), FAA = free amino acids (mg glycine equivalent/g), TTA = titratable acidity (mg lactic acid equivalent/g). Numerical value of sample code implies level of supplementation.

GLS supplemented samples and 0.0613 was observed for IWE in comparison to low (0.0263) free amino acids of the COS.

As stated earlier, data (free amino acids) obtained for ASS-1 deserve no interpretation, because the observed value was not due to biotransformation but due to asparagine of the solution added to the sample prior to fermentation/biotransformation. Evaluation of samples at day 2 showed that COE-2 and IWE-2 samples had about 3 times the amounts (Table 2) of FAA hydrolysed in GLS-2, which implies that addition of cooked onion and Iru extracts enhanced biotrans-formation of locust beans to condiment (Iru) than addition of glucose. However, all the supplements resulted to improvement in hydrolysis of amino acids compared to COS. Similar trend was observed for Day 3. Thus addition of COE and IWE improved hydrolysis of amino acids comparatively to glucose supplementation.

Considering the result of FAA across the days of biotransformation, while FAA increased in day 2 comparatively to day 1 in all the samples, same was not observed in data for day 3 when compared with data for day 2, because the increase was marginal when observed. However, appreciable increase in FAA was obtained in the COS at days 5.

Moreover, supplementation at level 3 yielded no better results than corresponding level 2 supplementation.

Assessment of HSS (mg glucose/g sample) at day 1 showed that addition of cooked onion extract produced the highest amount (0.4407) of soluble sugars. The amounts of hydrolysed soluble sugars in IWE-1 (0.315) and ASS-1 (0.218) apparently were not very different from the value (0.2786) observed for COS.

However, increase in soluble sugars evaluated at Day 2 doubled for IWE-2 and even greater value for COE–2 samples. The soluble sugars determined for ASS-2 sample was low when compared with the value for the COS at day 2. The results observed for Day 3 appeared slightly different from the stated trend, in that HSS content evaluated amounted to no noticeable difference in the hydrolysed soluble sugars when compared with Day 2 results. But it was observed that HSS proportionally increased during the days in the control sample. This result showed that biotransformation was actively in progress during days 1 and 2 of the process. Generally, there was increase in titratable acidity (TTA) of samples during biotransformation. Similar result was earlier reported by Ikenebomeh et al. (1986). TTA (Table 2) of the samples on day 1 appeared similar with minuscule difference in amounts of TTA values. However, at Day 2 progressive increment in TTA was observed in all the samples, with the exception of GLS supplemented samples. A slight increase in favour of IWE–2, COE–2 and ASS-2 samples in comparison to COS-2 at day 2 was observed. At day 3, TTA of Iru and cooked onion extracts supplemented samples increased comparatively to TTA of COS-3 sample with no difference in TTA values for asparagine and glucose supplemented samples.

The high TTA of IWE-3 samples may be due to onset of undesirable biotransformation. This is because the TTA of other samples were similar to TTA of the COS sample. The TTA determined for asparagine-supplemented samples deserves no interpretation because of the amino acids inherent to the added supplement.

Sensory scores of the samples are shown in Table 3. Iru, cooked onion extracts and glucose solution supplemented samples were adjudged better than control sample in terms of colour. Asparagine supplemented sample was rated least. The colour changed to green at day 3. This was probably an indication of growth of wild bacteria.

Table 3. Sensory score of biotransformed locust beans with and without dietary supplements.

Quality attribute	IWE-1	IWE-2	IWE-3	COE-1	COE-2	COE-3	ASS-1	GLS-1	GLS-2	COS
Day 2										
Colour	8.00a	7.57a	8.00a	3.71bc	3.00bc	2.29c	2.29c	8.00a	8.00a	6.00ab
Odour	6.00abc	5.86abc	8.71a	8.00ab	7.43abc	6.86abc	6.86abc	4.29c	5.00bc	4.42c
Taste	7.53ab	7.50ab	7.80a	8.43a	7.46b	5.00c	5.00c	5.00c	5.21c	6.00c
Texture	5.86a	4.71a	6.14a	6.43a	7.00a	5.71a	5.71a	6.29a	6.00a	7.57a
Mucillage	5.86a	5.29a	6.86a	7.43a	7.43a	4.71a	4.71a	4.71a	5.29a	6.43a
Day 3										
Colour	8.43abc	7.86abc	7.00bcd	7.85abc	6.14cde	5.29dc	1.14f	9.57a	9.29ab	4.43e
Odour	5.14ab	5.43ab	6.29ab	7.29a	5.71ab	7.14a	4.71ab	4.43ab	7.14a	6.29ba
Taste	8.24a	8.00a	8.00a	6.58b	6.43b	8.00a	4.6c	4.12c	5.00bc	6.00b
Texture	8.00a	7.57a	7.86a	7.29a	7.14a	7.43a	7.57a	6.71a	7.14a	3.86a
Mucillage	6.00bcd	5.00d	5.29cd	6.71ab	7.71a	5.57bcd	6.00bc	5.86bcd	6.43bc	7.71a

See Table 2 for interpretation of sample codes.
Any two means in a row not followed by same letter(s) are significantly (p=0.05) different.

Table 4. Dietary element composition (mg/100g) of selected locust beans samples.

Dietary element	Samples		
	$^{1}COS^{0}$	$^{2}COS^{3}$	^{3}COE–3
Ca	211.20	272.25	232.65
Fe	11.62	7.69	9.50
Zn	8.91	12.54	7.92
P	1822.26	1492.26	1542.75
Na	70.95	84.15	81.84
Mg	130.35	123.75	121.11
Mn	34.32	33.66	33.99
K	120.45	128.04	124.41

Results are average of 2 determinations. $^{1}COS^{0}$ = ^{1}Cooked sample day O, $^{2}COS^{3}$ = ^{2}Cooked sample day 3, ^{3}COE–3 = ^{3}cooked onion extract supplemented (level 3, day 3) sample.

Odour, the most important quality identification factor in locust beans condiment (Iru) also could be taken as measure of extent of biotransformation. COE–1 and COE–2 were significantly rated better than other samples of day 2. At day 3, the odour of COE–1, COE–2 and GLS samples was significantly preferred to other samples. IWE samples were rated least in terms of odour. This result has a linear relationship with TTA of the samples (Table 2). Therefore, high biotransformation TTA could connote excessive biotransformation. Excessive biotransformation is not desirable due to associated repulsive odour.

In terms of taste, IWE and COE samples were preferred to COS both at day 2 and 3. The texture of COS was only significantly different at the p=0.01 and not at p=0.05 from the other samples.

Summarily, sensory score showed that IWE and COE supplemented samples were preferred in colour to ASS and COE samples, but comparable to GLS, and possessed comparable odour to COS. Thus COE and IWE extracts enhanced biotransformation of locust beans to condiment without adversely affecting the sensory attributes of the condiment.

Changes in nine dietary elements (minerals) were evaluated in selected three sample namely: COS day 0, COS day 3, COE–3 day 3 in order to examine change in dietary elements during biotransformation with view to identity dietary elements that may be important for biotransformation of locust beans to condiment (Iru). Table 4 shows the result of the analysis.

Fermentation (biotransformation) of samples (COS, COE-3) at day 3 resulted to decrease in evaluated total amount of dietary elements content compared to COS at day 0. This result appeared to be at variance with previous study (Ikenebomeh et al., 1986). However, the authors gave no report on amounts of individual dietary elements. But, inspection of the result of each of the assessed dietary elements in the present study showed that while the amounts of Ca, Zn, Na and K increased the amounts of P, Fe, Mg and Mn decreased. It is arguable that during fermentation, increase in dietary element could be due microbial release or synthesis because one of the advantages of fermentation (biotransformation) during food processing is to release or unlock nutrients (Potter and Hotchkiss, 1996) and decrease in mineral content could be due to microbial utilization. Potter and Hotchkiss (1996) stated that one of the possibilities of microbial effect during fermentation (biotransformation) is that when microorganisms ferment (biotransform) food constituents, they derive energy in the process. In addition, Odunfa (1986) asserted that biotransformation of locust beans to Iru is associated with temperature up to 45°C. Moreover, microorganisms source for energy by releasing enzymes mostly requiring dietary elements in nature as co-factor for activation.

Conclusion

Addition of cooked onion extract (COE) and Iru water extract (IWE) enhanced biotransformation of locust beans to condiment (Iru) comparatively to glucose and asparagine under the conditions employed in this study. The result of this investigation suggests that composite nutrient such as present in COE and IWE rather than absolute carbon source (glucose) or nitrogen-source (asparagine) could enhance conversion of locust beans to condiment. In addition, dietary element such as phosphorus, magnesium and iron were identified to be important for biotransformation of locust beans to condiment. The simple but effective process could be adapted for improvement of the technology of traditional fermentation of locust beans to condiment.

REFERENCES

AOAC (1990). Official Methods of Analysis. 14th Edition. Association of Official Analytical Chemists, Washington DC.Beaumont M (2002). Flavouring composition prepared by fermentation with *Bacillus spp*. International J. Food Microbiol. 75:189-196.

Dubois M, Gilles KA, Hamilton JJ, Reber PA, Smith F (1956). Colorimetric method for determination of sugars and related substances. Anal Chem 28: 350-354.

Ikenebomeh MJ, KOK R, Ingram JM (1986). Processing and fermentation of the African locust bean. (*Parkia filicoidea* Welw.) to produce dawadawa. J. Sci. Food Agric. 37: 272-282.

Larmond E (1979). Laboratory Method of Sensory Evaluation of Food (Publ. No. 1637). Dept of Agriculture, Ottawa, Canada.

Odibo FC, Nwabunnia F, Osuigwe DI (1990). Biochemical changes during fermentation of *Telfaria* seeds for Ogiri production. World J. Microbiol. Biotechnol. 6: 425-427.

Odunfa SA (1986). Dawadawa. In: Reddy NR, Pierson MD, Salunkhe DK (Eds). Legume-Based Fermented Foods. ERC Press, Boca Raton pp. 173-189.

Olutiola PO, Famurewa O, Sonntag HG (2000). An introduction to general microbiology – a practical approach. Bolabay publications Obafemi Awolowo Way, Lagos Nigeria, p. 46.

Omafuvbe BO, Falade OS, Osuntogun BA, Adewusi SRA (2004). Chemical and biochemical Changes in Africa. Locust bean (*Parkia biglobosa*) and melon (*Citrullusvulgaris*) seeds during fermentation to condiments. Pak. J. Nutr. 3(3): 140-145.

Potter NN, Hotchkiss JH (1996). Food Science CBS publishers and distributors (1st Indian Edn) New Delhi, India, pp. 264-275.

Rosen H (1957). A modified ninhydrin Colorimetric analysis for amino acids. Arch. Biochem. Biophys. 67: 10-15.

Sharma JL, Caralli S (2002). A dictionary of food and nutrition CBS. Publishers and Distributors. New delhi – 2, India pp. 518-521.

Snedecor GW (1956). Statistical Methods 5th edn Iowa State college Press.