

# THE USE OF FIVE TECHNIQUES TO EXTRACT PROTEIN, OIL AND GOSSYPOL CONTENT OF OKRA SEEDS (*Abelmoschus esculentus* (L) moench)

W. UBI, H. IGWE, S. EKPE and O. IBOR

(Received 29 November, 2004; Revision accepted 21 July, 2005)

## ABSTRACT

Okra (*Abelmoschus esculentus* (L) moench) seeds of two cultivars, Emerald and Clemson spineless were taken from the harvest of previous study and used for this investigation. The seeds were ground by five techniques, extracted with water and filtered to yield a milk-like emulsion. The five techniques are: the use of blender with dry seeds, hand grinding with dry seeds; use of blender with soaked seeds for 20 hours, hand grinding with soaked seeds for 20 hours, thereafter, soaked for 24 hours after grinding. Proteins, oils and gossypol were analyzed using standard laboratory techniques. (Technique 3). Higher percent of protein and oil were recovered from the seeds soaked 20 hours in water and then ground with household blender. Results values of (technique 3) were significantly ( $P < 0.05$ ) higher than all other treatments. The values of emerald cultivar in terms of protein, oil and gossypol contents were significantly ( $P < 0.05$ ) higher than values obtained from Clemson spineless cultivar, given the same treatment. Okra seeds curd is an attractive and nutritious food that can be prepared with household-scale techniques of greater economic value. The implications of the results are discussed in light of okra seeds nutrition.

**KEYWORDS:** Five techniques, Protein, Oil and gossypol content of Okra seeds

## INTRODUCTION

Okra, (*Abelmoschus esculentus* (L) moench) appears to have potentials as high protein crop of the tropics when grown for its seeds, (Karakoltsides and Constantivides, 1975). Okra is presently used as a warm season vegetable, young pods of which are used boiled, in soups or fried. It is produced throughout the tropics and warmer parts of the temperate zone; and is especially popular in India and West Africa.

The seeds of Okra have often been used for various purposes. They have been roasted and ground, as a coffee substitute. Franklin et al. (1979). Edible oil has been extracted from the seeds. Goldblatt (1969). The seeds have been ground into meal and use in cooking. These under documented uses have seldom been the object of scientific attention. Martin and Ruberte, (1979) and Powell, (2002) have received such uses and still add another, the preparation of vegetable curd from the seeds.

Vegetable curd prepared from okra seeds has many advantages as a food. It is easy to prepare, even with household appliances and without the use of electricity. It is attractive in taste and appearances. It is versatile in its uses, and can be substituted for cheese, in recipes.

Further, it is rich in protein and oil. The production of okra seed and its use in vegetable curd could enrich diets in many parts of the tropics. Seeds of malvacea, such as cotton seeds often contain gossypol, a toxic polyphenole compound. Gossypol, irritates the gastrointestinal tract and leads to pulmonary edema of the lungs paralysis, and in extreme cases, death, Goldblatt, (1969).

However, many gossypol deleterious effects were not considered in this study, but it is good to avoid cultivars with high gossypol. In developing a new food product from okra seeds it is desirable to measure protein, oil and gossypol content and in this study we report levels of this substances as influenced by treatments.

## MATERIALS AND METHODS

Okra seeds of Emerald and Clemson spineless were obtained from a multiple source population based on collection from previous experiment, Ubi, (2004). The seeds were sieved to remove small seeds and impurities, washed to remove floating seeds and dried at 40°C for twenty-four hours in Gallen kamp moisture dried oven. All test lost weighed 100g. All test mentioned were performed in three replicated samples. Five different techniques were used to grind or extract seeds as follows.

1. Dried seeds were ground for 1 minute in a housed hold Blender. About 5 volumes of water were added and stirred to mix thoroughly.
2. Dried seeds were ground as finely as possible with a hand operated seeds grinder. About 5 volumes of water were added and the mixture thoroughly stirred.
3. Seeds were soaked in 5 volumes of water for 20 hours rinsed and ground for 1 minute in a household blender in the same volume of water.
4. Seeds were soaked in 5 volumes of water for 20 hours, rinsed and ground without water with a hand operated grain seed grinder. The same volume of water was added and the mixture was thoroughly stirred.
5. Seeds were prepared as described in 4 but the ground mixture in water was soaked for an additional 24 hours.

All of the mixtures were filtered, through cotton cloth. The filter was squeezed by hand to remove excess liquid. The residue was dried at 40°C until weight was stable and then weighed. The milky liquid was heated to 80°C. Ten grams of Commercial Epsom salt (magnesium sulphate 7H<sub>2</sub>O) was then added and the liquid was removed from the heat. After 10 minutes the liquid was filtered through three layers of cheese cloth. After filtering the water of approximately two volumes of the wet precipitate was poured over the filter twice to remove salts and non precipitate substances was poured over the filter twice to remove salts and non precipitated substances. The

W. UBI, Ministry of Agriculture, No. 3 Barracks Road, Calabar, Nigeria.

H. IGWE, Cross River State Agriculture Development Project, Calabar, Nigeria,

S. EKPE, Permanent Secretary, Ministry of Agriculture, 3 Barracks Road, Calabar, Nigeria.

O. IBOR, Forestry Commission, Calabar, Nigeria.

filtered curd in the cheese cloth was pressed between two blocks of wood on which a 2kg – weight was placed for 1 hour.

The extracted oils however were considered part of the dry weight of the sample. The original seeds milk residues and Wheys were tested for protein, oil and gossypol content. Protein was determined on defatted samples. Nitrogen was determined by the micro-kjeldahl technique (AOAC, 2000). Protein content was calculated as  $6.25 \times N$ .

Fatty substances were extracted with N-heptane in a soxhlet apparatus. The solvent was evaporated and the residue was dried to a constant weight to determine oil content. The oil containing gossypol and determined by the official method of the American Oil Chemists Society (2000). The entire chemical analysis was done at the Science Laboratories of Alabama Agricultural and Mechanical University Normal, United States of America.

**Statistical Analysis:** Data were subjected to analysis of variance (ANOVA), and means compared by the use of Duncan's Multiple range test at (0.05) probability level.

## RESULTS

The results of effect of grinding and extraction techniques on the amount of residue are presented in Table 1.

Grinding dry seeds with a household blender significantly ( $P < 0.05$ ) produced a finer and more uniform product in the two cultivars than grinding by hand. Soaked seeds were more easily ground with the household blender than by hand, because the wet meal was resilient, and particles appeared to be larger. The meal soaked for an extra 20 hours fermented, as evidence by a foul odor and turbid appearance.

The effect of different techniques on extraction of dry matter from the samples can be shown in two ways:

By dry weight of non extractable residue, and by dry weight of vegetable curd. Grinding of wet seeds with a household blender (technique 3) significantly ( $P < 0.05$ ) resulted in a lighter residue and heavier vegetable curd (Table 1) than did the other techniques.

The calculations of recoveries made in the two cultivars during the study showed that technique 5, soaking of seeds after grinding resulted in a much reduced recovery of oil and a somewhat reduced recovery of protein compared with other treatments (Table 2).

**Table 1:** Effect of grinding and extracting techniques on amount of residue, vegetable curd and sample lost in Whey (g/original 100g).

Techniques	Milk Seed condition		Residue not extracted (g)	Wet (W) (g)	Dry Weight (g)	Amount unaccounted for (g)
	Emerald Cultivar					
1.	Blender	Dry	55.2C	36.4b	18.1b	17.0c
2.	Hand	Dry	54.5C	37.2b	16.7bc	18.6ab
3.	Blender	Soaked 20 hours	62.6a	48.6a	21.5a	19.0ab
4.	Hand	Soaked 20 hours	58.3b	45.5a b	21.2a	13.1d
5.	Hand	Soaked 20 hours, before 24 hours after grinding	54.6c	32.1c	13.6c	12.2a
<b>Clemson spineless cultivar.</b>						
1.	Blender	Dry	53.5c	38.6c	19.6b	18.5c
2.	Hand	Dry	57.2b	39.4c	17.2c	19.2c
3.	Hand	Soaked 20 hours	60.5ab	48.6a b	21.6a	20.3b
4.	Hand	Soaked 20 hours	54.1c	50.7a	22.0a	15.3d
5.	Hand	Soaked 20 hours before 24 hours after grinding	53.7c	36.8 cd	14.8d	22.5a

Differences followed by the same letters are not statistically different ( $P < 0.05$ ).

**Table 2:** Content of protein oil and gossypol in vegetable curds made from different grinding techniques and percent of original oil and protein recovered.

Grinding techniques	Protein (%)	Oil (%)	Gossypol (%)	Percent recovered	
				Protein	Oil
<b>Emerald Cultivar</b>					
1	35.2b	47.8ab	0.013c	26bc	54b
2	35.1b	40.7b	0.017b	28b	41c
3	45.4a	51.2a	0.019b	31a	65a
4	34.2b	48.7ab	0.016b	27b	64a
5	31.5c	36.1c	0.037a	23c	38d
<b>Clemson spineless Cultivar</b>					
1	33.1b	44.2ab	0.012b	24.2b	51.2b
2	34.2b	38.1b	0.011b	26.0ab	37.1d
3	41.2a	45.6a	0.015b	29.3a	56.0a
4	34.3b	44.7ab	0.014b	22.1c	54.3 ab
5	34.3b	38.2b	0.028a	22.1c	52.4c

Difference followed by the same letters are not statistically different ( $P < 0.05$ )

The highest gossypol content (0.037) was obtained from the fermented sample (technique 5) and was significantly ( $P < 0.05$ ) higher than those of other treatments. High gossypols content in the case of the first 4 techniques were related to efficiency of extraction. Highest protein content (45.1%) and oil (51.2) in emerald cultivar and protein (41.9%) and oil (46.5%) in Clemson spineless, were obtained from (technique 3) and in the case of soaked seeds ground by blender and these were significantly ( $P < 0.05$ ) higher than those of other treatments. The percent of original protein and oil recovered was highest in the case of soaked seeds ground by blender (technique 3) and these were significantly ( $P < 0.05$ ) higher than those of other treatments in two cultivars and the amount of residue accounted for, followed closely the same trend throughout the study period.

The species differed significantly in their response to these treatments. Emerald cultivar, on the average, had the highest residue not extracted (62.6g) and had greater wet weight and dry weight and these were significantly ( $P < 0.05$ ) higher than those of Clemson spineless in the case of soaked seeds ground by household blender (technique 3). Equally, the protein contains 45.5%, oil (51.2%), and gossypols, (0.019%) of emerald were significantly higher than values obtained from Clemson spineless (Table 2).

#### DISCUSSIONS

The investigation has revealed that high protein and high vegetable oil product can be made at the household level from okra seeds, using simple techniques. Since okra seeds can be produced conveniently on the small farm and the seeds can be stored in a container for a reasonable length of time, okra seed curd could be a new nutritious food for the individual household or can be easily adapted to large-scale production in the tropics.

The gossypol content of okra seed curd appears to be potentially, a serious problem associated with toxicology. Practically all gossypol research seems to concentrate on cotton seeds meal, used both as food and as a feed. The U.S. Food and Drug administration has established a tolerance level of free gossypol in cotton seed meal of not more than 0.045%. Food and Agricultural Organisation of the United Nations recommended not more than 0.06% free or 1.2% total gossypol in meals for human use, Pons (1976).

In this study, measurements of gossypol content of okra seeds curd are well within this indicated tolerance. The high gossypol content reported in the case of fermented sample, (Technique 5) is suggested to be due to more efficient

extraction by fermentation and increased concentration caused by loss of oil and protein.

#### CONCLUSION

The third technique in this study that involved soaked seeds ground by household blender has been suggested for adoption as this method gave more protein content and oil and is generally affordable. Gossypol can be avoided by selection of cultivars with low gossypol content. However, a more thorough study of the possible long-term toxicological implications of gossypol should be done with test animals as to come up with a better understanding of the dangers of high gossypol content in okra feed meals.

#### REFERENCES

- AOAC, Association of Official Analytical Chemists, 1990. Official methods of Analysis 15<sup>th</sup> edition Association of official Analytical Chemist Washington D.C. United States of America.
- Franklin, W. M., Felek, L. Reberte, R. and Satiago, A., 1979. Protein oil and gossypol content of vegetable curd made from okra seeds. *Journal of Food Sciences*, 44: 1517 – 1529.
- Goldblatt, B., 1969. Gossypol in "Toxic" Constituents of plants food stuffs. Ed. Liend, I. E. P. 211. Academy Press New York.
- Karakoltsides, P. A. and Constantinides, 1975. Okra seeds a new protein Source. *Journal agric. Food Chem.* 23(6): 1204.
- Martin, F. W. and Reberte, R., 1979. Milling and use of okra seeds meal at the household level. *Journal of Agric. Univ. Puerto Rico.* 63:1
- Pons, W. A., Jr, 1976. Gossypol analysis past and present *AOAC.* 60: 252-259.
- Powell, Smith, 2004. Nutrition facts and food composition. Analysis of okra raw. Home and Garden Information carter, Lxington Extension Agent pp. 1-4.
- Ubi, W., 2004. Response of Okra (*Abelmoschus esculentus* (L) *moench*) to some mulch materials. *Global Journal of Agricultural Science.* 3(1): 53-57