

enable the citrus fruit to withstand loss of water, especially during a relatively long hot period.

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## Nutritional Composition of Okra Seed Meal

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The nutrient content of okra seed (*Abelmoschus esculenta* Moench) was investigated. Okra seed contained 21% protein, 14% lipids, and 5% ash. Removal of the seed hulls by grinding and sifting produced a meal with 33% protein, 26% lipids, and 6% ash. The protein of okra seed has a chemical score of 55, with isoleucine the first limiting amino acid. A saturated/unsaturated fatty acid ratio of 1:1.55 was found in the oil, with principal fatty acids as follows: 42% linoleic, 34% palmitic, and 18% oleic. Minerals of whole seeds included 135 mg of Ca/100 g and 335 mg of Mg/100 g with much lesser amounts (<5 mg/100 g) of copper, iron, manganese, and zinc; sifting out hull material resulted in an increase in iron (11 mg/100 g), zinc (14 mg/100 g), and magnesium (518 mg/100 g) concentrations.

The potential of the seed of okra, *Abelmoschus esculenta* Moench, as a high-oil, high-protein crop for the temperate zone and the tropics has now been amply demonstrated. Probably Woodruff (1927), who pointed out that after oil is extracted from okra seeds a high-protein meal remains similar to that of cottonseed meal, was the first to recognize this. Nevertheless, in spite of consid-

erable investigation of the oil and its qualities during the period from 1930 to the present, the potential of okra seed has never been realized. In 1975, Karakoltsidis and Constantinides reviewed part of the literature and studied the protein, oil, vitamin, and mineral content of the whole seed. Their studies have stimulated other investigators who see okra seed as an important crop of high potential.

The problem of removal of the okra seed hull from the kernel has not been sufficiently studied. Nevertheless, in an attempt to devise a system for small-scale use, Martin and Ruberté (1979) ground the seed by a hand mill and separated two fractions, chiefly hulls and chiefly kernels, by sieving. The okra meal so produced contained 33% protein and 32% oil. This meal was used in partial substitute for wheat flour and cooking oil to prepare a wide variety of baked products, including bread. The high acceptability of such products further stimulated interest

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in the possibility of okra seed as a new crop.

In these studies the nutrient composition of a simply prepared okra seed meal suitable as a household product is reported.

#### MATERIALS AND METHODS

Okra seeds were obtained from the third generation of an interbreeding population established from 238 normal okra varieties, grown in Mayaguez, Puerto Rico. Seeds were relatively fresh (less than 1 year of age) and had been refrigerated until sent to Brigham Young University (Provo, UT) for analysis. Their germinability was high (more than 75%). At Brigham Young University the seeds were maintained at room temperature at relatively low humidity until ground as meal. Pieces of seeds and deformed or immature seeds were removed before grinding.

Ground okra seed meal (GM) was produced by grinding whole seeds to a fine meal in an electric grinder. The meal was composed of ground hulls and kernels. Sieved okra seed meal (SM) was produced by sieving ground meal once using a no. 16 screen. The sieving process separated much of the hull material from the ground kernels. SM, however, did contain some residual hull material together with the ground kernels.

Proximate chemical analyses were performed on GM and SM. These analyses determined the moisture, total lipids, and total ash according to standard methods (AOAC, 1975); protein was determined by the Kjeldahl method in which percent nitrogen (N) was multiplied by 6.25.

Lipids were extracted by using the Goldfish extraction procedure. Petroleum ether (bp 30–60 °C) was refluxed through the sample for 4 h to extract lipids; the fatty acid composition of SM was determined by using gas chromatography analysis of methyl ester derivatives of extracted lipids. Even-numbered fatty acid methyl esters were identified and quantitated by using a Varian series 2400 gas chromatograph equipped with a hydrogen flame ionization detector under the following conditions: detector temperature, 230 °C; injector temperature, 215 °C; column temperature, linear program from 140 to 200 °C at a rate of 2 °C/min. Carrier gas (N<sub>2</sub>) flow rate was 15 mL/min; compressed air flow rate was 300 mL/min; hydrogen flow rate was 15 mL/min. A 6 ft long × 1/8 in. i.d. stainless steel column packed with 10% SP-2330 on 100/120 Supelcoport was utilized. Areas under the chromatograph peaks were integrated by using an Autolab Model 6300 integrator.

Odd-numbered fatty acid methyl esters were identified and quantitated by using a Packard series 7300 gas chromatograph under the following conditions: detector temperature, 230 °C; injector temperature, 210 °C; column temperature, linear program from 120 to 220 °C at a rate of 1 °C/min. Carrier gas (N<sub>2</sub>) flow rate was 38 mL/min; compressed air flow rate was 425 mL/min; hydrogen flow rate was 35 mL/min. An 8 ft long × 2 mm i.d. U-shaped glass column packed with 3% OV-1 on Chromosorb W (80/100 mesh) was utilized. Areas under the chromatograph peaks were integrated by using an Infotronics Model CRS-104 integrator.

Mineral assays were performed on acid-digested samples of GM and SM. The method of additions was utilized for each mineral assay. All determinations were made by using a Perkin-Elmer Model 306 atomic absorption spectrophotometer. Specific minerals analyzed included calcium, cadmium, chromium, copper, iron, magnesium, manganese, and zinc. Bovine liver extract (National Bureau of Standards Reference Material No. 1577) was used as a standard throughout the analyses to determine the recovery of minerals. The digestion of GM was carried out

Table I. Proximate Composition of Ground Okra and of Ground and Sifted Okra Seed Meal (%)

component	ground okra meal (GM) <sup>c</sup>	sifted okra seed meal (SM) <sup>c</sup>
moisture	7.80 ± 0.24	7.44 ± 0.57
protein <sup>a</sup> (N × 6.25)	21.10 ± 0.38	32.50 ± 0.31
total lipids <sup>a</sup>	13.58 ± 0.44	25.57 ± 0.29
total ash <sup>a</sup>	5.28 ± 0.02	6.37 ± 0.04
total carbohydrates <sup>b</sup>	60.04 ± 0.84	35.56 ± 0.64

<sup>a</sup> Expressed on dry weight basis. <sup>b</sup> Calculated by difference. <sup>c</sup> Mean ± standard error (triplicate determinations).

by using a nitric acid–perchloric acid procedure (AOAC, 1975). SM was digested by using a sulfuric acid–hydrogen peroxide (30%) treatment (Culver et al., 1975).

Amino acid analysis was performed on dried and defatted SM. Acid hydrolysis of the sample was carried out for 22 h by using 6 N HCl under nitrogen. The hydrolysate was filtered to remove insoluble humin and dried on a rotary evaporator, and the dried residue was taken up with 4 mL of 0.2 M citrate buffer (pH 2.2). Cystine and cysteine were determined as cysteic acid following performic oxidation of SM according to Moore (1963). The tryptophan content of SM was determined following basic hydrolysis of the sample in barium hydroxide monohydrate (Tkachuk and Irvine, 1969; Miller, 1967). All amino acid analyses were performed on a Beckman Model 121 amino acid analyzer. Dual-column separation of the amino acids provided for qualitative and quantitative analysis of basic and acidic + neutral amino acids. Quantitation of sample amino acids was determined by comparison of peak areas of a commercial amino acid standard to peak areas of the SM hydrolysate samples.

#### RESULTS

In producing GM the complete seed was included in the sample material. SM in these tests included 50% of the original seed weight.

The proximate compositions of GM and SM are summarized in Table I. It is evident that the removal of much of the hull material from GM is beneficial in increasing the percentages of protein, total lipids, and total ash in SM. The sifting process produced a meal that is approximately 54% higher in protein content, 88% higher in lipid content, and 20% higher in ash content. The meal was 41% lower in carbohydrate content.

The fatty acid composition of SM extracted lipids is given in Table II. Previously published results of okra seed fatty acid content are given for comparison. A large percentage (61%) of the fatty acids of okra seed are unsaturated. The high percentage of linoleic acid (42%), an essential fatty acid in human nutrition, makes okra seed oil a desirable food oil. The correspondingly low percentage of linolenic acid (<1%), also an essential fatty acid in human nutrition, does not seriously affect the nutritional aspects of the seed oil as the linoleic and linolenic acids are interconvertible in the human system.

The mineral compositions of GM and SM are summarized in Table III. Contents of calcium, iron, magnesium, and zinc were higher in SM than in GM, indicating that these minerals are located principally in the kernel of the seed. However, SM contained less copper and manganese than GM, indicating that these minerals are present at a higher concentration in the hull of the seed. From a nutritional standpoint, the magnesium, zinc, and iron contents of SM are outstanding.

The amino acid composition of SM is given in Table IV as compared to the provisional amino acid scoring pattern of the Food and Agriculture Organization of the United

Table II. Percent Fatty Acid Composition of Okra Seed Lipids

fatty acid <sup>a</sup>	sifted okra seed (present study) <sup>b</sup>	whole seed (Karakoltsidis and Constantinides, 1975)	whole seed (Sengupta et al., 1974)
14:0	0.24	0.12	0.2
15:0	trace	trace	
16:0	33.72	33.53	30.2
16:1	0.56	0.14	0.4
17:0	0.64	0.24	
17:0	trace	trace	
18:0	3.28	3.81	4.0
18:1	17.88	29.29	24.4
18:2	42.15	31.48	40.8
18:3	0.24	1.42	
19:0	1.12	trace	
19:0		0.09	
21:4		trace	
22:0	0.16		
24:4		0.12	

<sup>a</sup> Fatty acids are expressed as number of carbon atoms in chain to number of double bonds. <sup>b</sup> Based on two replicate analyses.

Table III. Mineral Composition of Ground Okra Seed and Sifted Okra Seed Meal<sup>a</sup>

mineral	ground okra seed (GM), mg/100 g of dry seed	sifted okra seed meal (SM), mg/100 g of dry meal
Ca <sup>a</sup>	135.0 ± 9.0	169.0 ± 6.0
Cr	<0.32	<0.32
Cu	1.74 ± 0.06	1.28 ± 0.12
Cd	<0.08 ±	<0.08
Fe	4.18 ± 0.86	11.2 ± 2.3
Mg <sup>b</sup>	335.0 ± 15.0	518.0 ± 76.0
Mn	2.14 ± 0.16	1.89 ± 0.11
Zn	5.78 ± 0.13	13.7 ± 1.2

<sup>a</sup> Based on three replicate analyses. <sup>b</sup> Dilutions prepared in 1% lanthanum solution.

Nations (FAO/WHO, 1973). The protein analyzed provides a sufficient amount of tryptophan, threonine, total sulfur-containing amino acids, leucine, and tyrosine + phenylalanine. The limiting amino acids and their corresponding amino acid or chemical scores are isoleucine 55, valine 69, and lysine 87. The value of tryptophan indicates that this amino acid is in excess of the FAO/WHO (1973) suggested level.

## DISCUSSION

The fatty acid composition of okra seed oil in the present study correlates closely with previously published data. The major deviations among the studies occur in the percentages of oleic (18:1) and linoleic (18:2) acids. The present study indicates a lower percentage of oleic acid and a higher percentage of linoleic acid than in previous reports (Karakoltsidis and Constantinides, 1975; Sengupta et al., 1974). However, the sum percentage of oleic + linoleic acids in all three studies is consistent: 60.0, 60.8, and 65.2%.

The ratio of saturated to unsaturated fatty acids, 1:1.55, in the present study is close to the 1:1.66 ratio reported by Karakoltsidis and Constantinides (1975). The study reported by Sengupta et al. (1974) indicated a saturated/unsaturated fatty acid ratio of 1:1.90.

The present investigation of minerals in ground okra seed indicates approximately one-half the previously reported amounts of calcium and iron (Karakoltsidis and Constantinides, 1975). The discrepancy may be explained by the different methodologies used in the respective

Table IV. Amino Acid Composition of Okra Seed (Based on Two Replicate Analyses)<sup>f</sup>

amino acid	sifted okra seed meal	ref pattern (FAO/WHO, 1973)	amino acid score <sup>b</sup>	whole okra seed <sup>e</sup>
Trp	94	60	100	-
Lys	297	340	87	502
His	146			186
ammonia	117			-
Arg	583			778
Asp	824			966
Thr	257	250	100	273
Ser	397			419
Glu	1270			1280
Pro	284			379
Gly	376			361
Ala	329			368
Cys + cystine	189	220	100	96 <sup>f</sup>
Val	214	310	69	400
Met	141	c		81 <sup>f</sup>
Ile	138	250	55	290
Leu	428	440	97	529
Tyr	203	380	100	225
Phe	275	d		294

<sup>a</sup> Values are expressed in milligrams of amino acid per gram of sample N. <sup>b</sup> Amino acid score = (mg of amino acid in test protein)/(mg of amino acid in reference pattern) × 100. <sup>c</sup> Included in cysteine + cystine value. <sup>d</sup> Included in tyrosine value. <sup>e</sup> Data calculated from Karakoltsidis and Constantinides (1975). <sup>f</sup> Values believed to be low due to partial loss during acid hydrolysis.

mineral assays or the different seed samples used. The present study utilized atomic absorption spectrophotometric techniques to quantitate the minerals whereas the previous investigators used AOAC (1975) chemical methods. Bovine liver extract (Reference Material No. 1577), used as a standard in the present investigation, confirmed the recovery of trace minerals. The present methodology, therefore, suggests confidence in the calcium and iron values found. Sieving can be concluded to be a useful process for increasing iron, manganese, and zinc contents.

In the only previously published amino acid analysis of okra seed, Karakoltsidis and Constantinides (1975) reported that the amino acids isoleucine, valine, and lysine were not limiting. Table IV indicates the amino acid composition of the sifted okra seed meal and comparative data reported by Karakoltsidis and Constantinides (1975). Variations between the respective amino acid profiles can be partly explained by variety differences and/or the degree of seed maturation at time of analysis, or possibly these are associated with the sieving process. However, with such close correlation between the two studies, it was concluded that the values obtained in the present study for valine, isoleucine, and lysine indicate these as the limiting amino acids in sifted meal.

Okra seed should prove to be a satisfactory supplement to any diet that may be lacking tryptophan. In addition, okra seed protein contains an amino acid pattern (Table IV) that allows it to serve as an adequate supplement to legume- or cereal-based diets. Methionine is well-known to be the first limiting amino acid in legumes. Okra seed can supply sufficient amounts of methionine in the dietary protein. Threonine is generally the second limiting amino acid in legumes (Jansen, 1977), and okra seed can supplement the legume-based diet with sufficient amounts of threonine as well.

Okra seed can also serve as a supplement to cereal-based diets in which lysine is generally the first limiting amino acid (Jansen, 1977). Although the amino acid score for lysine was 87 (representing the third limiting amino acid

in okra seed), this value is considerably higher than that of most cereal grains. A representative list of cereal grains where lysine is the first limiting amino acid and the respective chemical scores includes barley 64, cornmeal 49, millet 63, oats 68, polished rice 66, ragi 53, rye 62, sorghum 37, teff 51, wheat bulgur 47, and wheat flour 38 (Jansen, 1977).

Thus, the present data suggest that okra seed meal obtained by the simple process of grinding and sieving is an improvement over ground whole seed and should be of high nutritional value.

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## Effects of Temperature and Storage on the Iron and Tin Contents of Commercially Canned Single-Strength Orange Juice

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The effects of storage time and temperature on the corrosion of commercial cans packed with single-strength orange juice showed that accelerated rates of detinning occurred at higher temperatures. Storage of canned juices over a 12-week period showed the following mean weekly rates of tin uptake (mg of Sn/kg juice-week): 0.4 [21.1 °C (70 °F)], 0.8 [26.7 °C (80 °F)], 1.5 [32.2 °C (90 °F)], 2.4 [37.8 °C (100 °F)], 4.0 [43.3 °C (110 °F)], and 12.5 [48.9 °C (120 °F)]. Iron uptake by juice was minimal and indicated that orange juice was not strongly corrosive to the plain carbon steel plate. Storage of juices below 43 °C should not cause the tin contents to exceed 250 ppm (Codex Alimentarius Commission tolerance limit of tin in canned juice) if the juice is stored for 12 weeks or less.

Extensive packaging of liquid foods in tins is due to the many advantages of this type of container, namely, ease of packing and sterilizing, handling and transporting, and minimizing the loss of vitamin potency in a foodstuff because of the anaerobic environment of the sealed can. These containers are made from plain carbon steel plates with a thin coating of tin (American Can Company, 1973).

In an empty tin can, the tinplate is cathodic and the steel base is anodic. The mode of protection is a mechanical one; i.e., the corrosive medium is kept from the corrosion-susceptible steel base by the corrosion-resistant tin layer. However, in the presence of a corrosive acidic liquid, a reversal of polarity takes place. Tin now becomes the local anode and the steel base the local cathode (Lueck and Blair, 1928; Kohman and Sanborn, 1928). Thus, the protective mode of the tin plate is now via an electrochemical process, and this happens in canned acidic fruit juices such as citrus juices.

During the preservation of acid food products, for example, canned citrus juices, an interaction occurs between the components of the canned food and the material of the can. Corrosion in canned acid products is influenced by the chemical composition of the product (Bombara et al., 1970), character of the tinplate (Mahadeviah et al., 1975), and the presence of corrosion accelerators such as sulfites, sulfur dioxide (Saguy et al., 1973), nitrates (Lambeth et

al., 1967), and oxygen (Kefford et al., 1959; Koehler, 1961). Studies (Vaurio, 1950; Koehler and Canonico, 1957) of the corrosion mechanism have shown that the process entails dissolution of the tin coating, dissolution of the steel base, and evolution of hydrogen.

The object of the present work was to evaluate the influence of temperature and storage on the corrosion (dissolution of iron and tin) of tinplate containers packed with single-strength orange juice (SSOJ).

This experiment should also indicate other factors (pH; titratable acidity; Brix/acid), in addition to time and temperature of storage, that are responsible for enhancing the uptake of tin by the juice, thus limiting the shelf life of the product because the tolerance level of tin (250 mg/kg of juice; Codex Alimentarius Commission, 1978) was exceeded.

#### EXPERIMENTAL SECTION

**Materials.** Commercially canned SSOJ was obtained from three processors (A; B; C) during the early (November to January) and mid (January to March) processing seasons and from four processors (A; B; C; D) during the early Valencia (April to May) and late Valencia (May to July) seasons. SSOJ in 46-oz cans was taken directly from the production lines in the processing plants and placed in a laboratory locker at -18 °C. Temperatures of storage lockers were maintained at ±2 °F (about ±1.1 °C).

**Instrumentation.** A Perkin-Elmer Model 503 atomic absorption spectrophotometer was used with a Perkin-Elmer tin electrode-less discharge lamp set at 8 W or a Perkin-Elmer hollow cathode iron lamp. Tin absorbance

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