

# Compositional Differences Among Crambe Samples and Between Seed Components<sup>1</sup>

F. R. EARLE, J. E. PETERS and I. A. WOLFF, Northern Regional Research Laboratory,<sup>2</sup> Peoria, Illinois; G. A. WHITE, New Crops Research Branch,<sup>3</sup> Beltsville, Maryland

## Abstract

Data germane to the processing and utilization of crambe as a new oilseed include information on variations to be expected in gross composition of the seed (fruit) and its component parts.

Seventy-five samples of *Crambe abyssinica* Hochst. ex R. E. Fries from experimental plantings in 17 states have been analyzed. Samples as received contained 16 to 62% pod material (pericarp), the extremes representing samples with many seeds removed from the pod or with many pods containing no seed. The amount of pericarp was most often between 25 and 40%. Oil content of seed (without pericarp) ranged from 36 to 54%, with most samples between 40 and 48%; crude protein from 22 to 37%, usually 25 to 30%; and erucic acid in the oil from 39 to 60%, usually 53 to 59%. Total thioglucoside content in 30 samples ranged from 8 to 10% calculated as *epi-progoitrin* in oil-free meal, although 2 samples were between 4 and 5%.

One sample was hand-separated into pericarp (40%) and seed, and the latter was further separated into seed coat (8%), cotyledon (82%) and hypocotyl (10%). The pericarp contained only 0.4% lipid and the respective seed fractions 17, 55 and 38%. Their corresponding protein contents were 4, 23, 23 and 34%; and their thioglucoside contents (oil-free meal), 0.1, 2.1, 10.9 and 13.0%.

Cotyledon and hypocotyl were quite similar in amino acid composition. Neither contained hydroxyproline, which is in both pericarp and seed coat.

## Introduction

AMONG THE MANY PLANT SPECIES under examination in the U.S. Department of Agriculture as potential new oilseeds, crambe (*Crambe abyssinica* Hochst. ex R. E. Fries) attracted attention because its oil contains approximately 55–60% erucic acid, more than any oil now available in world commerce. Rapeseed oil, usually containing 35–50% erucic acid, is one of the leading vegetable oils of the world, being exceeded in tonnage produced only by soybean, peanut, cotton, coconut, sunflower and perhaps olive and palm oils (17). Comparatively little of the oil is traded outside the country of production. To meet United States needs for erucic acid oil, about 4 million lb of rapeseed oil (17) is imported annually. Discussions with industrial chemists suggested that a domestic source of an oil rich in erucic acid could find immediate use and that utilization might be increased markedly by research on applications of the oil and of products chemically derived from it (1).

*Crambe abyssinica* is related to the rapes and mustards (family Cruciferae) and is native to the Mediterranean area. It grows about 3 ft tall and produces

seed about 1/16 in. in diameter, in spherical single-seeded pods (silicles) about 1/8–3/16 in. in diameter (Fig. 1). It was grown experimentally in Russia as early as 1932 (14,18), in Sweden (10), and in Canada soon after 1950 (10). There has been no continuing commercial production.

Following analyses of crambe oil in 1957 in our search for new industrial oils, plantings were made at various Agricultural Experiment Stations to provide both additional seed for study and preliminary information regarding climatic adaptability and response to agronomic practices. The favorable outcome of these and later plantings created increasing interest and resulted in plantings of about 100 acres for industrial-scale processing (12) in 1963 and plantings of several-fold this acreage by a commercial company in 1965.

Although optimum areas and conditions for production are not yet fully established, crambe seems to be rather widely adapted to areas where it can make its major growth during a relatively cool season. Present conventional farming equipment can be used for planting and harvesting. Crambe seems resistant to insects and diseases and, even in early experimental plantings, has given excellent seed yields.

Since these favorable agronomic characteristics suggest that production can be easily increased to meet demand, crambe may become an established crop and be subjected to numerous scientific investigations. Accordingly, results of compositional studies are presented here to indicate the variation to be expected from the strains of crambe now available when grown under different climatic and cultural conditions. The data give only a general view. Since the samples are not from statistically designed experiments, they do not provide precise correlation between compositional variation and causative factors. Such detailed studies remain for the future. Also reported are results on analyses of morphological components of the crambe



FIG. 1. *Crambe abyssinica*, 73 days after planting. Close-up shows flowers and developing pods.

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<sup>2</sup> No. Utiliz. Res. Dev. Div. ARS, USDA.

<sup>3</sup> Crops Research Division, ARS, USDA.

fruit: pericarp, seed coat, cotyledon and hypocotyl. These data may be pertinent to future processing operations.

### Materials and Methods

Seed for analysis was supplied by USDA's New Crops Research Branch (NCRB) mainly from experimental plots grown by workers at various State Agricultural Experiment Stations. The first sample analyzed in our program came from the seed collection of the NCRB's Plant Introduction Section. It had been accessioned in 1950 as Plant Introduction (PI) No. 189139 without information as to its foreign origin. Most of the results in this paper derive from experimental plantings of PI 189139 and PI 247310 (accessioned May 1958 from Sweden), but a few results derive from introductions from Russia in 1962 (PI 281728-281737) or lines obtained from Canada by individual investigators. Samples from larger, multiacre plantings on 12 farms in 1963 are also included. Most of the samples came from the northern half of the United States, but some came from Texas where crambe can be planted early enough to mature before hot weather or, in some instances, planted to grow through the winter.

Many of the early samples were analyzed with the seed in the pod (pericarp) as received except that stems and foreign matter were removed. Most of these samples were also milled to remove the pericarp and analyzed as cleaned seed. Sample preparation varied with the purpose of the analysis; the usual procedure was to remove and weigh stems and foreign matter, remove and weigh pericarp, and analyze clean seed.

The sample to be separated into structural components was milled in a burr mill carefully adjusted to crack the pericarp without damaging the seed. The pericarp was then removed by aspiration. The seed was broken in the readjusted mill and separated into fractions consisting of seedcoat, cotyledon and hypocotyl by a combination of aspiration, sieving and handpicking (Fig. 2).

Methods of analysis have been reported previously for oil and protein content (7), oil composition and properties (11), amino acids (21) and thioglucosides (19).

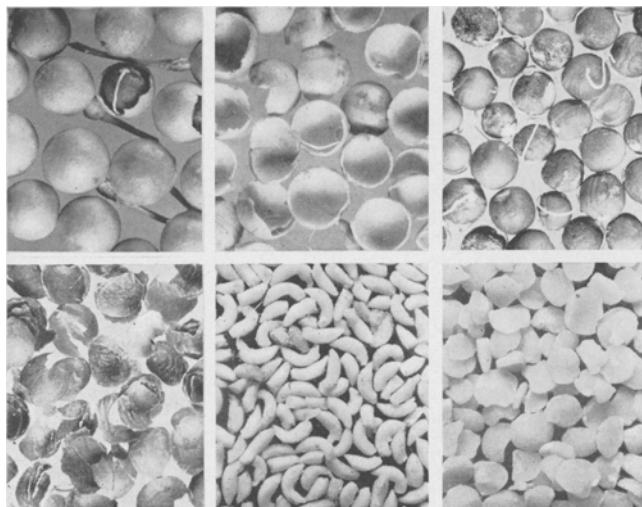


FIG. 2. Crambe pods, seeds and structural components. From top left: pod, pericarp and seed. From bottom left: seedcoat, cotyledon and hypocotyl. Magnification  $2\frac{1}{2} \times$ .

## Results and Discussion

### Seed Composition

Analyses of "seed plus pericarp," or seed in the pod, have practical value because present methods of harvest collect the seed primarily in this form. Such analyses provide information regarding the quantity of oil in a given lot of seed as purchased, in storage, or being processed. However, the presence of unfilled pods and of seed from which the pod has been threshed in harvesting prevents effective use of such analyses for comparing compositions of crambe samples with one another. The wide variation in oil content of crambe seed (Fig. 3) has not yet been correlated with specific genetic or environmental factors. The occurrence of numerous samples having 45-50% oil in the seed is favorable to commercial processing for oil, and the presence of three samples containing more than 50% oil suggests that future crops might have even more desirable composition. Presumably, cultural and climatic factors are involved in this variation in composition rather than genetic strains alone, because the three samples richest in oil came from three States in two years and at least two of the three are the same stock as most of the other samples.

Analysis of seed and seed plus pericarp for protein content shows distribution patterns (Fig. 4) comparable to those for oil in the same samples.

As factors favoring high-oil crambe seeds are learned and utilized, the protein content of the seed will probably decrease in conformity with the inverse relationship normally existing between two constituents that make up such a large proportion of the seed. In the future it may be possible to increase the total oil and protein in the seed, now about 75%, and, perhaps, to increase the oil content preferentially.

### Oil Composition

With the exception of two samples, all crambe oils in this survey contained from 51 to 60% erucic ester (GLC area %) (Fig. 5). In contrast, rapeseed oil, the present commercial source of erucic acid, may contain as much as 55% (8), but is more likely to have less than 50% and even as little as 30% (4,5). The two exceptions among the crambe samples were grown in Alaska in successive years from the same seed source used for many of the other samples. The low erucic content, 39-40%, may result from a climatic effect peculiar to the extreme northern latitude. Omitting the Alaskan samples, other principal constituents in crambe oil are oleic ester, 15-20%; linoleic, 7-11%; linolenic, 4-8%; eicosenic, 2-6%;  $C_{24}$ , up to 4%; and saturated acids, 4-8%. Typical composition of crambe seed oil is given in Table I. No

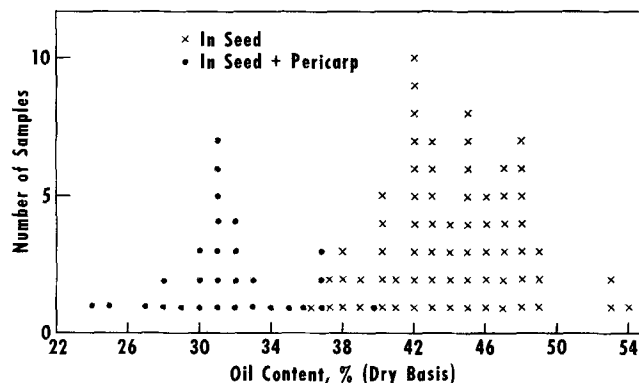


FIG. 3. Oil content of crambe seed and seed plus pericarp.

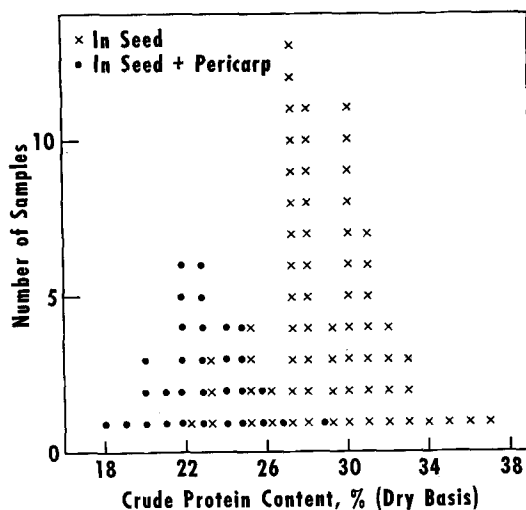


FIG. 4. Protein content of crambe seed and seed plus pericarp.

definite relationship was evident between content of crucic acid and the percentage of oleic, linoleic or linolenic acids.

#### Oil-Free Meal

Meal available after removal of oil from crambe seed (without pericarp) contains 46–58% crude protein, a proportion desirable for feed supplements. The amino acid composition of the meal indicates good nutritional quality and potential value for feed use. The amino acid data (Table II) on the seed used for study of morphological components are in good agreement with those of samples reported previously (21).

Defatted crambe meal also contains some 8–10% thioglucosides, primarily *epi*-progoitrin, which place restrictions on use of the meal in animal feeds (6,19). The occurrence of two samples with only 4 and 5% of thioglucosides suggests that the content of thioglucoside may be reduced by cultural methods, but the effective factors are not yet known. When the low-thioglucoside samples were planted in another location, the progeny contained the usual large amount of thioglucosides. The meal also contains sinapine

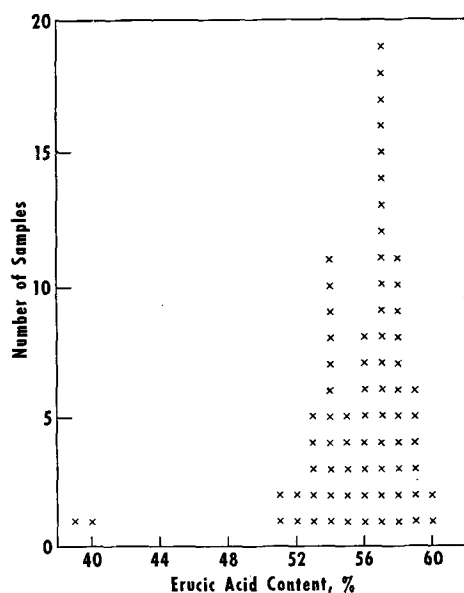


FIG. 5. Erucic acid in crambe oil, as methyl ester in mixed methyl esters, GLC area %.

TABLE I  
Composition and Characteristics of Oil from Crambe Seed and Seed Parts

Characteristic	Pericarp	Seed	Seed-coat	Hypocotyl	Cotyledon
Iodine value	....	92	95	100	93
Refractive index, $n_D^{40}$	....	1.4650	1.4666	1.4660	1.4648
Unsaponifiables, %	34	....	5	3	1.4
Esters in mixed esters, GLC area %					
12:0	0.1	0	Tr	....	0
14:0	0.8	0.1	0.1	0.1	0.1
16:0	11.8	1.6	2.8	4.5	1.5
16:1	1.6	0.3	4.1	0.5	0.3
18:0	2.3	0.8	0.3	0.9	0.5
18:1	8.7	17.9	12.5	13.4	15.1
18:2	12.0	6.9	12.7	13.2	6.7
18:3	7.3	6.7	4.0	7.2	6.1
20:0	1.9	0.7	0.5	0.5	0.2
20:1	8.9	2.5	15.2	4.7	2.0
20:2	....	0.3	....	0.5	....
22:0	1.4	1.4	0.8	0.5	0.7
22:1	39.6	58.6	45.7	46.0	59.1
22:2	....	0.3	0.7	....	0.1
22:3	....	0.3	....	....	....
24:0	1.2	0.3	....	4.7	2.4
24:1	1.1	1.2	....	3.4	4.5
Other	1.3	0.1	0.6	....	0.7

(2) and related substances that may influence its utilization as feed.

Feeding tests with cattle on a finishing ration containing 10% protein supplement suggest that crambe meal can replace one-third of the soybean meal without effect on weight gains (3). When more than a third of the soybean meal was replaced, feed consumption and weight gains were reduced. Feed efficiency, however, was not altered and no toxic effects were noted in gross examination of the animals and carcasses (3). In contrast, the untreated meal is toxic to rats and chicks. Essentially all the toxic components can be removed by extraction of the meal with 80% aqueous acetone, but the loss of meal solids amounts to 20% (19). After hydrolysis of the thioglucoside by endogenous enzymes, the toxic materials can be removed by extraction with 90–98% aqueous acetone, and the loss of meal is reduced to about 10% (16). The search for improved methods of seed processing and meal treatment is continuing. Pretreatment of the meal with ammonia (9) or other chemicals may have promise.

#### Composition of Seed Parts

By present methods, most crambe is harvested without disruption of the pod or silicle. The outer part of the pod (pericarp) can be rather easily removed from the seed by milling followed by aspiration. The

TABLE II  
Amino Acid Content of Oil-Free Meal from Crambe Seed and Seed Parts (g/16 g N)

Amino acid	Pericarp	Seed	Seed-coat	Hypocotyl	Cotyledon
Lysine	1.5	5.1	7.4	5.6	5.5
Methionine	0.3	1.6	1.2	1.8	1.5
Arginine	1.3	5.7	4.4	6.5	6.4
Glycine	1.8	5.2	5.0	5.8	5.1
Histidine	0.4	2.4	2.0	2.5	2.9
Isoleucine	1.4	3.7	3.7	3.9	3.4
Leucine	2.4	5.9	5.1	6.5	5.4
Phenylalanine	1.5	3.4	3.2	4.5	3.4
Tyrosine	0.7	3.0	3.2	2.7	2.5
Threonine	1.7	4.2	5.0	4.0	4.9
Valine	1.8	4.5	5.0	4.7	3.8
Alanine	1.7	4.0	4.0	4.4	3.6
Aspartic acid	3.1	6.0	5.9	6.2	6.8
Glutamic acid	3.6	14.2	11.2	16.7	14.8
Hydroxyproline	1.1	0.9	5.9	0.0	0.0
Proline	1.7	5.5	6.7	5.7	5.7
Serine	1.9	3.5	4.2	3.5	3.5
Nitrogen as amino acids, % of N	23	74	72	76	70
Nitrogen as ammonia, % of N	26	13	14	15	15

seed can then be further milled, but quantitative separation of the seed fractions by the procedures used here required hand separation as a final step. The pods, seeds and seed parts are shown in Figure 2.

The proportion of pericarp (Table III) in the sample used for the separation of structural components is near the upper limit expected for good quality seed. For most samples the amount of pericarp is between 25 and 40%, but in a few, perhaps grown under sub-optimum conditions and containing many unfilled pods, the pericarp constituted more than 50% of the sample. Major constituents of the pericarp are lignin, 30%; pentosans, 23%; and  $\alpha$ -cellulose, 19%. The low levels of oil and crude protein in the pericarp, the small proportion of the total nitrogen accounted for as amino acids and the high level of lignin suggest that the pericarp will have little nutritive value for nonruminant animals. The level of thioglucoside in the pericarp is extremely low. Tests with rats confirm the absence of toxicity from the pericarp; reduced mean gain in weight is probably attributable to the increased amount of fiber (19). In a preliminary experiment, crambe pericarp incorporated into ruminant feed appeared to be an acceptable roughage that caused no decrease in weight gain or feed efficiency (3).

The seedcoat is rich enough in oil and protein to have significant nutritional value and its thioglucoside content is low enough so that the amount present contributes little to the total in the meal. The

lysine content of the protein is unusually high, and the methionine content somewhat low, but neither deviation will have much effect on the use of seedcoat in a protein feed supplement. The high content of hydroxyproline, while perhaps of no nutritional significance, is in contrast to the levels in the other seed components, and is in agreement with Van Etten's findings on the occurrence of hydroxyproline (20).

The cotyledon is higher in oil content and lower in protein than the hypocotyl but, otherwise, the gross composition of the two fractions is very similar. As with urease activity in the soybean (13), myrosinase activity in crambe is about twice as high (0.24 vs. 0.14  $\mu$  eq sulfate from sinigrin/min/mg defatted material) in the hypocotyl as in the cotyledon (15). Differences noted here suggest no reason for trying to separate these components in future commercial milling. Together they constitute 92% of the seed in the sample studied here, and their analyses are in good agreement with the analysis for the whole seed.

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TABLE III  
Analyses of Crambe Seed and Seed Parts

Constituent	Pericarp	Seed	Seed-coat	Hypocotyl	Cotyledon
Proportion of sample as harvested, %	40	60	5	6	49
Proportion of seed, %	....	100	8	10	82
Component analyses, % dry basis					
Oil	0.4	47	17	38	55
Crude protein	4	25	23	34	23
Ash	8	....	4	5	4
Oil-free meal analyses, % dry basis					
Crude protein	4	51	29	56	52
Phosphorus	0.2	....	0.3	1.8	2.1
Sulfur	0.0	2.4	0.7	3.1	2.6
Thioglucoside, as <i>epi</i> -progoitrin (6)	0.1	8.8	2.1	13.0	10.9
Calcium	2.2	....	1.0	0.7	1.0
Magnesium	0.2	....	0.1	0.6	0.7