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ACIDS AND THEIR FUNCTIONALLY SUBSTITUTED ESTERS FROM *Amaranthus cruentus* SEED OIL

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The acid composition of seed oil of Amaranthus cruentus *and the synthesis of their glycidyl and pyridine-containing esters are studied. It is demonstrated that 67% of the total acids are C_{18}-polyunsaturated linoleic and linolenic. A new method for preparing glycidyl esters of C_{18}-unsaturated carboxylic acids is developed by reacting their salts with ECG in an aprotic medium to produce the corresponding glycidyl esters. The reaction of the glycidyl esters and pyridine salts with carboxylic and phosphonic acids produces the propanolpyridine esters of the acids that combine the properties of the acids and pyridinium salts and are promising in the search for biologically active compounds.*

Key words: *Amaranthus cruentus*, seeds, fatty acids, glycidyl and propanolpyridine esters.

Extensive research in the last two decades has produced new information on the biological role of unsaturated fatty acids. Several studies have established that they are very important bioregulators of intracellular biochemical reactions and various physiological processes that occur in humans [1]. Both free fatty acids and their esters, in particular, glycerine esters [2] and acid amides [3] possess such activity.

The unsaturated fatty acids oleic, linoleic, and linolenic, which are found in plant oils, are unsubstituted fatty acids and the principal components of vitamin F [4]. They affect the aggregation of blood thrombocytes and cholesterol level. They exhibit antisclerotic and immunostimulatory activity. Free acids, especially linoleic and linolenic, and their esters can suppress the growth of malignant tumors [5-7]. The presence of these acids in seed oil of amaranth [8] and the antitumor activity of amaranth oil [9, 10] have been reported.

In the present article, results are presented from a study of the composition of acids from seed oil of amaranth *Amaranthus cruenthus*, which is indigenous in Tatariya. The oil was obtained by extraction and was saponified in aqueous alcohol. The mixture of acids was analyzed by chromatography-mass spectrometry and high-resolution mass spectrometry. The qualitative and quantitative composition of the acids was determined by chemical-ionization (CI) mass spectrometry because the peaks of the acid molecular ions can be too weak or even absent in electron-impact mass spectra. The base peaks in the CI mass spectra are those of the quasimolecular ions $[MH]^+$ (100%). The quantitative ratio of the acids was estimated using the ratio of intensities of the quasimolecular ions because the volatility of the acid fractions was quite similar. The mass of the $[MH]^+$ ion for both the acid mixtures and their derivatives was determined precisely by peak coincidence at 10,000 resolution and was compared with that calculated. These results and data for the acid composition and content are listed in Table 1. The analysis showed that seed oil of *A. cruentus* contains five acids. Table 1 shows that the principal acid is linoleic (53% of the total). The total contents of polyunsaturated acids (linoleic and linolenic), 67%, in *A. cruentus* oil is close to that of linseed.

The important role of unsaturated fatty acids and the pharmacological properties that they exhibit make it attractive to prepare various derivatives of them, e.g., salts, esters, and amides. One of the most promising types of esters of unsaturated carboxylic acids is glycidyl esters, which can act as key compounds in the synthesis of new derivatives of the aforementioned acids. A method for preparing glycidyl esters of linoleic and oleic acids in 65-90% yield has been reported. It consists of the

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TABLE 1. Acids of *A. cruentus* Seed Oil

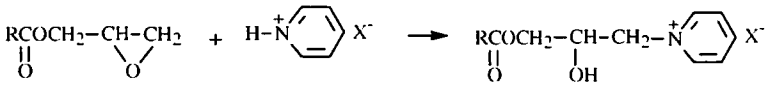
Acid	Mass $[MH]^+$		Content, %
	found	calc.	
Palmitic	257.247	257.2481	7
Linolenic	279.231	279.2324	14
Linoleic	281.249	281.2481	53
Oleic	283.265	283.2637	21
Stearic	285.280	285.2794	4

TABLE 2. Glycidyl Esters of Acids of *A. cruentus* Seed Oil

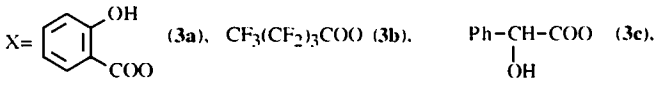
Glycidyl ester of acid	Mass $[MH]^+$		Content, %
	found	calc.	
Palmitic	313.272	313.2743	10
Linolenic	335.257	335.2586	16
Linoleic	337.275	337.2743	49.5
Oleic	339.288	339.2899	18.5
Stearic	341.305	341.3056	3

reaction of acid salts with epoxychloroglycerine (ECG) in the presence of quaternary ammonium catalysts [11, 12]. We proposed a new method for preparing glycidyl esters of these acids in addition to linolenic acid that produces the corresponding esters in quantitative yield. The method is based on the use of the polar aprotic solvent DMF, which enables the process to be carried out without a catalyst. A mixture of acids is used that includes amaranth oil. The acids are not separated in order to preserve the uniqueness of the oil. The composition and structure of the resulting esters were determined by CI mass spectrometry analogous to that for determining the acid mixture (Table 2).

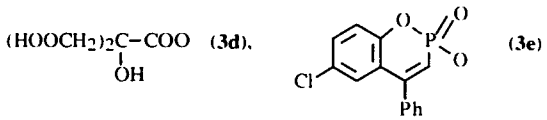
As already noted, glycidyl esters of unsaturated fatty acids can act as key compounds for the preparation of various derivatives. We used glycidyl esters of amaranth oil (**1**) to prepare new functionally substituted esters of the acids, propanolpyridine salts **3**.



1 **2** **3**



(3a), $CF_3(CF_2)_3COO$ **(3b)**, Ph–CH(OH)–COO **(3c)**,



(3d), **(3e)**

Salts **3** are prepared by the reaction of glycidyl esters **1** with pyridine salts **2** of organic and phosphorus-containing acids under conditions analogous to those given in the literature [13]. The synthesized salts **3** are thick viscous oils of yellow to brown color. The epoxide ring is opened under mild conditions to give the corresponding esters **3a-e** in quantitative yields. This was shown by IR spectroscopy using the disappearance of the characteristic epoxide bands (860 and 908 cm^{-1}) and the appearance of bands corresponding to hydroxyl stretching vibrations (3200-3300 cm^{-1}).

EXPERIMENTAL

Mass spectra were obtained by chemical ionization on a Finnigan MAT-212 instrument. Samples were directly introduced into the ion source. The reacting gas was pentane. IR spectra were recorded on a UR-20 instrument as films between KBr plates. $^{31}P\{^1H\}$ NMR spectra were recorded on a Bruker MSL-400 (162 MHz) instrument.

Preparation of *A. cruentus* Seed Oil. Amaranth seeds (377 g) were ground, placed in a Soxhlet extractor, and extracted with boiling CH_2Cl_2 (1 l) for 16 h. The solvent was removed at atmospheric pressure. The solid was held under vacuum until CH_2Cl_2 was completely removed. Yield of oil, 118 g (5.9%) from amaranth seeds (1622 g).

Preparation of *A. cruentus* Seed-oil Acids. Amaranth oil (118 g) was dissolved in ethanol (300 ml, 75%) and treated with hexane (50 ml) and KOH (10 g). The mixture was heated with stirring at 50-55 C for 2 h. Then, the reaction mixture was treated with water (200 ml) and extracted with hexane. Yield of neutral products from hexane, 8.92 g (7.6%). The lower layer, which contained acid salts, was neutralized by adding dilute H_2SO_4 (until the pH was 3). The organic acids were extracted with benzene (3×200 ml). The benzene extracts were combined and washed with water until the washings were neutral. The solvent was removed under vacuum. Yield of acids, 94.38 g (88.5%).

Preparation of Glycidyl Esters (1) of *A. cruentus* Seed Oil Acids. Acids (94.38 g) were treated with KOH (18.84 g) and dissolved in water (150 ml). The water was removed under reduced pressure. The solid (104.44 g) was treated with dry DMF (400 ml) and ECG (27 g) and heated with stirring for 5 h (at 100-110 C). The DMF was removed. The solid was filtered off in a Buchner funnel. Yield of glycidyl esters (**1**), 94.9 g. IR spectrum (cm^{-1}): 860 and 908 (epoxide ring), 1730 (COOR).

Preparation of Esters (3) of *A. cruentus* Seed-oil Acids. A solution of glycidyl esters (**1**, 2.0 g) and pyridinium 2-hydroxyphenylbenzoate (1.28 g) in pyridine (10 ml) was held for three days at 20 C. The pyridine was removed under vacuum. Yield of esters (**3a**), 3.08 g, thick oil). IR spectrum (cm^{-1}): 1180 (OH), 1260 (OH-phenol), 1590-1640 ($C_6H_5COO^-$), 1740 (C=O), 3020-3140 (C=C–H), 3200 (OH). Compounds **3b-e** were obtained analogously.

Esters **3b** (3.34 g) were obtained from pyridinium perfluoropentanoate (2.0 g) and glycidyl derivatives (**1**, 2.0 g). IR spectrum (cm^{-1}): 1180 (OH), 1670-1690 (COO^-), 1735 ($COOCH_2$), 3020-3140 (C=C–H), 3200 (OH).

Esters **3c** (2.8 g) were obtained from pyridinium 2-hydroxy-2-phenyl acetate (1.18 g) and glycidyl derivatives (**1**, 1.73 g). IR spectrum (cm^{-1}): 1180 (OH), 1590-1640 ($C_6H_5COO^-$), 1740 (C=O), 3020-3140 (C=C–H), 3200 (OH).

Esters **3d** (10.1 g) were obtained from pyridinium citrate (3.15 g) and glycidyl derivatives (**1**, 7.47 g). IR spectrum (cm^{-1}): 1130, 1180 (OH), 1580-1610 (COO^-), 1720-1740 (C=O), 3020-3140 (C=C–H), 3200 (OH).

Esters **3e** (2.3 g) were obtained from pyridinium 2-oxo-6-chloro-4-phenylbenzo[e]-1,2-oxaphosphorin-3-en-2-oate (1.27 g) and glycidyl derivatives (**1**, 1.15 g). IR spectrum (cm^{-1}): 1100 (OH), 1210-1250 (POC), 1280 (P=O), 1740 (C=O), 3020-3140 (C=C–H), 3200 (OH). ^{31}P NMR spectrum (DMF): δ_P 4.2 ppm (br. s).

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