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CHEMICAL CHARACTERISTICS AND BIOLOGICAL ACTIVITY OF LIPIDS FROM *Chenopodium album* SEEDS

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The content and composition of neutral lipids (NL), glycolipids (GL), phospholipids (PhL), and fatty acids (FA) from Chenopodium album *seeds are established for the first time. NL contained 15 identified FA; PhL, 16; and GL, 19 constituents including trans-18:1n9. The stress-protective effects of NL, FA, methyl esters of FA, and unsaponifiable substances (US) on the growth and development of wheat and cotton sprouts were studied under salt stress conditions. NL at a concentration of 0.001% and US and FA at a concentration of 0.01% had the greatest effects on the growth of wheat roots. US at a concentration of 0.001% stimulated growth of roots and stems of cotton sprouts; FA, only growth of roots at a concentration of 0.0001%.*

Keywords: *Chenopodium album*, neutral lipids, glycolipids, phospholipids, fatty acids, unsaponifiable substances, stress-protective activity.

Chenopodium album L. (white goosefoot, Amaranthaceae, formerly Chenopodiaceae) is an annual herbaceous plant that is widely distributed throughout the world, including Uzbekistan [1]. In some countries, *C. album* is considered a weed in field and garden crops; in others, it is cultivated for grain and is treated as an alternative source of edible food and drugs [2–4].

The aerial part of *C. album* is used in arid regions as fuel and raw material for producing dyes and feed. Young leaves of the plant are used as a vegetable in many Asiatic countries because of their high food value [2]. Extracts of various parts of *C. album* are traditionally used in folk medicine of several countries as a diuretic, laxative, sedative, hepatoprotective, hypotensive, anti-inflammatory, anthelminthic, and antitumor agent [2, 5, 6]. Seeds and their extracts have been prescribed for diseases of the liver and spleen and stomachache [7]. The aqueous decoction of *C. album* seeds was reported to possess contraceptive activity [5].

Furthermore, the plant exhibits allelopathic properties by inhibiting the growth of other wild species and controls the development of viruses, fungi, and soil nematodes [8]. *C. album* possess high salt resistance over a broad range of salt concentrations [9].

Lipids from *C. album* seeds are poorly characterized. Their contents in the seeds have been reported and vary in the range 5.8–8.9% [10–12]. Carotenoids (up to 6.61 mg/100 g) [10] and fatty acids (FAs) including oleic (37.9%), linoleic (26.1%), palmitic (17.4%), eicosenoic (20:1n9, 3.9%), and lignoceric (24:0, 1.1%) acids were also reported [13].

The goal of the present work was to study the chemistry of the lipid complex from *C. album* seeds growing in Uzbekistan and to find the stress-protective activity of separate lipid constituents on wheat and cotton crops under saline conditions.

Air-dried and ground seeds in a Soxhlet apparatus afforded neutral lipids (NL). The pulp was dried in air. Polar lipids (PL) were extracted from it by the Folch method. The NL contained free FAs. The NL were hydrolyzed by alkaline base to afford unsaponifiable substances (US). The PL were fractionated by column chromatography into glycolipids (GL) and phospholipids (PhL). The yields of lipid groups were found gravimetrically. Table 1 presents the results.

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TABLE 1. Parameters of Lipids from *C. album* Seeds

Parameter	Content
Moisture and volatiles, mass% of seeds	4.20
Neutral lipids with actual moisture, mass% of seeds	8.42
Neutral lipids in abs. dry substance, mass% of seeds	8.78
Free fatty acids, mass% of NL	1.29
Unsaponifiable substances, mass% of NL	7.90
Polar lipids, mass% of seeds, including:	0.75
glycolipids	0.28
phospholipids	0.47

TABLE 2. Composition and Content of Fatty Acids in Lipids from *C. album* Seeds, GC, mass% of Acids

Acid	NL	GL	PhL	Acid	NL	GL	PhL
10:0	–	0.21	–	18:2n6	53.71	39.01	27.69
12:0	–	0.29	–	20:0	0.46	0.78	0.28
14:0	0.17	0.92	0.35	20:1n9	1.20	0.74	0.45
15:0	–	0.44	0.24	22:0	0.54	1.51	0.91
16:0	9.07	19.72	33.70	22:1n9	1.94	1.18	0.47
16:1	0.44	0.53	1.82	24:0	0.17	1.21	0.62
17:0	–	0.41	0.26	24:1n9	0.20	1.36	0.10
18:0	1.34	5.30	2.71	26:0	0.18	0.80	–
cis-18:1n9 + 18:3	29.27	23.78	28.02	Σsat.	11.93	31.59	39.07
trans-18:1n9	1.31	1.81	2.38	Σunsat.	88.07	68.41	60.93

Table 1 shows that the NL content in *C. album* seeds growing in Uzbekistan agreed with the literature. The content of total lipids (NL and PL) was 9.53%. The NL included high contents (almost 8%) of US.

The qualitative compositions of the NL, GL, PhL, and US constituents were found using analytical TLC on silica gel and Silufol plates using various solvent systems, as reported before [14].

The NL from *C. album* according to the analyses consisted of hydrocarbons, FA esters with phytosterols and triterpenols, triacylglycerides (main constituent), free FAs, triterpenols, and phytosterols. Hydrocarbons, aliphatic alcohols, triterpenols, and phytosterols (main constituent) were identified in the US on Silufol plates using hexane–Et_2O (4:1).

The predominant constituents of GL were steryl glycosides; minor constituents, monogalactosyl- and digalactosyldiacylglycerides. The PhL contained phosphatidylcholines (main constituent), phosphatidylethanolamines, phosphatidylinositols, and minor amounts of phosphatidic acid.

NL, GL, and PhL were hydrolyzed by alcoholic base to establish the FA composition. The isolated FAs were methylated by diazomethane. The FA methyl esters were analyzed by GC. Table 2 presents the analytical results.

The presence of elaidic acid (*trans*-18:1n9) in the FA methyl esters was confirmed by IR spectral data and Ag^+-TLC. The IR spectrum exhibited a weak vibrational band for an isolated *trans*-olefinic group at 980 cm^{-1} [15]. The Ag^+-TLC chromatogram of the FA methyl esters had a spot with R_f 0.65 that corresponded to that of a model sample of elaidic acid methyl ester (R_f 0.67).

Acid 18:3 was also identified by Ag^+-TLC by comparing the chromatographic mobility of a spot with R_f 0.5 to that of linseed oil methyl esters (18:3, R_f 0.52) because the methyl esters of *cis*-18:1n9 and 18:3 were not separated under the used GC conditions.

Table 2 shows that the five acids reported in the literature [13] were identified in the FA compositions of all lipid groups from *C. album* with an additional 10 in NL, 11 in PhL, and 14 in GL.

Lipids from *C. album* were dominated by unsaturated FAs. The total content of 18:1 + 18:3 and 18:2 in NL was 82.98; in GL, 62.79; in PhL, 55.71%. The total contents of high-molecular-mass monoenoic FAs eicosenoic (20:1n9), erucic (22:1n9), and nervonic (24:1n9) were in the range 3.34% (NL) to 1.02% (PhL). The highest level of palmitic acid occurred in PhL (33.7%).

TABLE 3. Effect of Lipids from *C. album* Seeds on Growth of Wheat and Cotton Sprouts

Substance	Concentration, %	Wheat, cm		Cotton, cm	
		root length	stem height	root length	stem height
Control (H_2O)	–	3.24 ± 0.5	2.21 ± 0.8	2.71 ± 0.3	1.8 ± 0.3
Floraxan	0.00001	4.33 ± 0.5	3.14 ± 0.6	3.61 ± 1.0	2.56 ± 0.6
NL	0.01	2.47 ± 1.3	2.03 ± 1.0	2.65 ± 0.4	1.3 ± 0.3
	0.001	4.28 ± 0.8	2.49 ± 1.2	1.73 ± 1.5	1.31 ± 0.4
	0.0001	3.42 ± 1.3	2.45 ± 0.6	2.61 ± 0.8	0.91 ± 0.8
US	0.01	4.35 ± 0.7	2.68 ± 0.8	1.71 ± 0.9	1.2 ± 0.5
	0.001	3.13 ± 1.0	2.52 ± 1.1	3.31 ± 0.4	2.63 ± 0.6
	0.0001	3.05 ± 0.7	2.86 ± 0.5	2.86 ± 0.3	1.81 ± 0.5
FA	0.01	4.02 ± 0.6	2.83 ± 0.6	2.46 ± 1.0	2.01 ± 1.0
	0.001	3.5 ± 0.7	2.89 ± 0.6	2.85 ± 0.9	2.05 ± 0.6
	0.0001	3.62 ± 0.9	2.53 ± 0.7	3.46 ± 0.5	2.06 ± 0.8
MEFA	0.01	2.7 ± 0.9	1.52 ± 1.0	2.65 ± 0.4	1.53 ± 0.8
	0.001	3.81 ± 0.8	2.83 ± 0.5	2.1 ± 0.3	1.6 ± 0.4
	0.0001	3.28 ± 0.5	2.18 ± 0.5	2.23 ± 0.8	1.7 ± 1.0

Biological Part. The effects of various stress factors (high salt concentration; low and high temperatures; deficits of water, nutrients, etc.) on the quantitative and qualitative lipid compositions have been reported before [16, 17]. However, few publications on the biological effects of lipids of wild plants on the growth and development of crop species have appeared.

The stress-protective effects of NL, US, FA, and FA methyl esters isolated by us from *C. album* seeds were compared during cultivation of wheat and cotton under saline conditions. For this, seeds were wetted beforehand in aqueous solutions of the compounds and planted in Petri dishes to which NaCl solution (5 mL, 1.0%) was added instead of H_2O. The reference drug was the growth regulator Floraxan. Table 3 presents the results.

Table 3 shows that US at a concentration of 0.01% and NL at a concentration of 0.001% had the greatest effects on root growth in wheat culture. The root length in these versions exceeded the control by 34.2% and 32.1%, respectively, and was practically at the level of the test using Floraxan at a concentration of 0.00001%. FA at a concentration of 0.01% also had a positive effect on root growth. The root length in this instance exceeded that of the control by 24.0%.

The results showed that all lipid samples had various positive effects on wheat sprout stem growth. Samples of US and FA had the greatest stimulatory activity. For example, US at a concentration of 0.0001% increased the stem length by 29.4% as compared to the control; treatment of seeds with FA at a concentration of 0.001%, by 30.8%. FA methyl esters at a dose of 0.001% also exhibited stimulatory activity. The stem length of wheat sprouts was longer than the control by 28.0%.

US at a concentration of 0.001% and FA, 0.0001%, were highly active with respect to cotton roots. The root length exceeded that of the control by 22.1% and 27.6%, respectively. These substances did not have a significant stimulatory effect on cotton stems except for seeds wetted with US solution (0.001%), where the stem length exceeded that of the control by 46.1%.

Thus, the contents and compositions of NL, GL, PhL, and FAs from *C. album* seeds were studied for the first time. US and FAs of NL stimulated growth and development of wheat and cotton sprouts under saline conditions and; therefore, reduced the negative impact of salt stress.

EXPERIMENTAL

IR spectra of FA methyl esters were taken as films on a PerkinElmer Model 2000 FTIR spectrometer. GC of FA methyl esters used an Agilent 6890N instrument with a flame-ionization detector, a capillary column (30 m × 0.32 mm) with HP-5 stationary phase, He carrier gas, and temperature programmed for 150–270°C. FA methyl esters were identified according to the literature [18]. TLC of lipids and CC of polar lipids were performed as before [14]. Ag^+-TLC of FA methyl esters used silica gel L 5/40 plates with added $AgNO_3$ (30%) in benzene.

Fully ripe *C. album* seeds were collected in 2019 in Tashkent Region.

NL were isolated from ground seeds by exhaustive extraction with benzine (bp 72–80°C) in a Soxhlet apparatus. PL were extracted (4 ×) from the dried pulp by infusion with $CHCl_3$–MeOH (2:1) for 16 h. The crude extract of PL was worked up with aqueous $CaCl_2$ (0.04%) to remove nonlipid constituents. The content of free FAs in NL was calculated from the acid number, which was 2.56 mg/KOH/g [19].

Biological tests used seeds of wheat variety "Antonina" and cotton seeds of variety "Sulton". Seeds were wetted with the tested compounds at concentrations of 0.01, 0.001, and 0.0001% for 18 h, and then transplanted into Petri dishes (10 ea.) [20]. The tests were run in triplicate. The controls were seeds wetted with H_2O. The standard was Floraxan [21] at a concentration of 0.00001%. The effects of the compounds on the development of sprouts under salt-stress conditions was studied in Petri dishes with NaCl solution (1%, 5 mL) and cultivation for 5 d in a thermostat at 26°C. Activity was evaluated from the linear growth of the sprouts. Results were mathematically processed by dispersion analysis using Origin Pro software [22].

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