LIPIDS OF DEFECTIVE COTTON SEEDS AND THE INFLUENCE OF MICROFLORA ON THEIR COMPOSITION

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A comparative study has been made of the lipid composition of defective and firstgrade cotton seeds after their prolonged storage under industrial conditions. On the basis of the change in the lipids of the seeds subjected to thermal spoilage it is proposed to consider the process of spontaneous heating as a group of enzymatic processes taking place mainly in not fully ripe seeds when they are damaged by pathogenic microflora.

The cotton seeds that are delivered to oil factories, must be stored for a long time before they are processed. During this period, in addition to post-harvesting ripening, a number of biochemical processes takes place in them the nature and intensity of which are determined by the quality of the seeds stored and by the conditions of their storage. With an unfavorable combination of these factors a process of spontaneous heating takes place in the pile of seeds leading to their spoilage, and consequently, to a deterioration in the composition of the raw material sent for processing. It has also been observed that oil seeds undergo spontaneous heating considerably faster than grain seeds [1].

The external characteristics of defective seeds are a rust color of the fuzz of the seed coat and a changed color of the kernel of the seeds — from light brown to black [2]. These characteristics show considerable changes in the protein and carbohydrate components, since the main cause of the darkening of the seeds of cereal and oil crops is considered to be a reaction between amino acids and reducing sugars (melanoidin formation) [3]. In actual fact, in defective cotton seeds the level of protein has fallen somewhat and the fractional composition of the protein and carbohydrates has changed, but there are no hydrophilic melanoidins [4].

It has been shown that in the lipids of defective seeds of oil crops (sunflower, caster oil plant) the levels of free fatty acids, of peroxide, epoxide, and keto compounds of unde-termined structure, and of waxes and pigments have increased [1, 5].

In the present paper we give the results of a study of the lipids of defective cotton seeds in comparison with those of first-grade seeds, and the results obtained are used to elucidate the mechanism of spontaneous heating.

A batch of industrial seeds with intact coats after their mechanical opening up were separated according to the color of the kernel into two groups: sample I — defective seeds with brown and dark brown kernels (30.5% of the weight of the batch); and (II) — first-grade with respect to the color of the kernel (68.2%); the remaining 1.3% were empty seeds.

The free lipids and then the bound lipids were exhaustively extracted from the comminuted samples (kernels with coats). The lipids of batches I and II scarcely differed in color, the free lipids being light orange and the bound lipids dark brown. The protein residue I after complete defatting and drying was dark brown.

Table 1 gives some indices of the seeds and lipids from which it can be seen that the defective seeds differed little from the first-grade seeds with respect to such basic industrial indices as moisture content, acid No. of the lipids, and the amount of their bound form. But the defective seeds contained 1.4 times less gossypol, and half of it was present in the bound form, while in (II) the bulk of it was in the free form.

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TABLE 1. Characteristics of Stored Cotton Seeds

Index	Sample I	Sample II
Weight of 1000 seeds, g	93.6	101.2
Moisture content on the absolutely dry		
matter, %	6.5	5.7
Amount of free lipids (oil content), %	15.9	17.4
Acid No. of the free lipids, mg KOH	1.2	0.9
Amount of gossypol in the free lipids, %	0.015	0.1
Amount in the hexane-defatted seeds, % of	1	
total gossypol	0.95	1.36
free gossypol	0.48	1.27
Amount of bound lipids, %	2.9	2.7
Amount of gossypol in the bound lipids, %	2.15	9.43

TABLE 2. Compositions of the Fatty Acids of the Free and Bound Lipids of Stored Cotton Seeds (%, GLC)

	Lipi	ds I	Lipids II		
Acid	free	bound	free	bound	
14:0 16:0 16:1 18:0 18:1 18:2 Σ sat Σ unsat	0.5 18,1 1,2 1,7 18,4 60,1 20.3 79,7	0.2 19.8 Tr. 1,9 17,2 60.9 21,9 78,1	0,3 18,9 0,9 1,1 17,1 60,7 21,3 78,7	0.2 22.6 Tr. 3,5 24,5 49.2 26.3 73,7	

TABLE 3. Composition of the Free Lipids of Stored Cotton Seeds

	Amount, % by weight of			
Free lipids	Sample I	Sample II		
Neutral lipids	95.7	97.4		
Carbohydrates	Tr.	0.3		
Triacylglycerols (TAGs)	87.1	92.6		
Epoxyacyldiacylglycerols (EAGs)*	0.4	0.3		
Free fatty acids (FFAs)	1.2	1.0		
Monohydroxyacyldiacylglycerols (HAGs)*	1.8	2.4		
Diacylglycerols (DAGs)	5.8	1.8		
Sterols	1.7	1.4		
Hydroxyepoxyacyldiacylglycerols (HEAGs)*	Tr.			
Dihydroxyacyldiacylglycerols (DHAGs)*	1.6			
Trihydroxyacyldiacylglycerols (THAGs)*	Tr.			
Monoacylglycerols (MAGs)	0.4	0.2		
Polar lipids Gosspyol pigments	} 4.3	1.6		

\*Triacylglycerols with two residues of ordinary fatty acids and one residue of epoxy, monohydroxy, hydroxyepoxy, dihydroxy, or trihydroxy acid.

The compositions of the fatty acids (FAs) of the free and bound lipids I and II were determined by the GLC method (Table 2).

It follows from the results of the analysis (Table 2) that the FA compositions of lipids I and II were also practically identical. Some differences were observed in the ratios of the individual FAs of the bound lipids I and II. We have found previously that the bound lipids of a first-grade kernel are more saturated than the free lipids [6]. This difference was also observed in the compositions of the FAs of lipids II, the bound form having not only more of the 18:0 and 16:0 acids but also of the 18:1 acid. In sample I the free and the bound lipids were identical with respect to the composition of their FAs and were characterized by a high level of the 18:2 acid. On comparing the compositions of the bound lipids I and II, a high relative amount of the 18:2 acid can be seen in I because of a fall in the level of the 18:1, 18:0, and 16:0 acids in it.

To evaluate the degree of change of the individual lipid components, the free lipids were analyzed in more detail, being separated by a combination of column chromatography (CC) and thin-layer chromatography (TLC). For this analysis, the free lipids were isolated without the action of high temperatures. Polyamide and silica gel were used for CC. On the polyamide, the neutral lipids were separated from contaminating polar lipids and gossypol pigments. Then the neutral lipids were separated on a silica gel column into individual classes and narrow fractions. The sterols were eliminated from the fractions by precipitation from methanol. The amounts of HAGs, DAGs, and more oxidized forms of TAGs were determined gravimetrically from the ratio of normal and oxidized FAs obtained after alkaline hydrolysis of the corresponding fractions.

The polar lipids and the pigments eluted from the polyamide were rechromatographed by TLC in systems 1-4, and qualitative reactions were carried out for glycolipids, phospholipids, and gossypol. The results obtained are shown in Table 3.

It can be seen from Table 3 that in the hexane-extractable lipids I the proportion of polar lipids had increased, and there was a small amount of TAGs in the neutral lipids, but a high level of products of their hydrolytic cleavage and oxidation (mainly DAGs and DHAGs). The amount of FFAs in the defective seeds had hardly changed, though there were no peroxide compounds, and the amount of natural HAGs had not increased. Furthermore, it can be seen from the ratio of the products of the lipolysis and oxidation of the TAGs that on spontaneous heating hydrolytic processes take place more intensively than oxidative processes. Some indices of Tables 1-3 — the considerably smaller weight of seeds I, the lower amount of total gossypol and of HAGs in them — indirectly confirm the incomplete ripeness of seeds [7] undergoing spontaneous heating.

The FAs were isolated by alkaline hydrolysis from individual classes of lipids. The composition of the ordinary FAs was determined by the GLC method (Table 4).

In the defective seeds the FA composition of the TAGs had not changed but in the FFAs I there was a somewhat smaller amount of the 16:0 and 18:1 acids. The composition of the FAs of the polar lipids reflected a high degree of saturation of the species of glyco- and phospholipids that passed into the hexane-extractable fraction.

The acids isolated from the oxidized TAGs were separated by preparative TLC in system 3 into ordinary and oxidized species. The presence of an epoxy group in the latter was confirmed by a qualitative reaction. For further study, in view of the complexity of their composition, the oxidized acids were separated according to their polarities by TLC in system 4 into three groups: epoxy acids ( $R_f$  0.85); monohydroxy and hydroxyepoxy acids ( $R_f$  0.52); and polyhydroxy acids ( $R_f$  0.32).

The epoxy acids in the form of their MEs were investigated by GLC, and it was found that they contained 95% of an epoxy-18:1 and 5% of an epoxy-18:0 acid. Their structures were not determined.

The hydroxy acids of the other two groups were analyzed by UV spectroscopy and by the mass spectrometry of their trimethylsilyloxy (TMS) derivatives. The assignment of the fragments was made by using our own [8] and literature [9] results.

The acids with R<sub>f</sub> 0.52 did not differ with respect to the quantitative set of hydroxy acids of the C-18 and C-17 series (mass spectrum) and with respect to the amount of conjugated  $\alpha$ -OH-18:2 acids ( $\lambda_{max}^{hexane}$  232) from the corresponding lipids of first-grade ripe seeds [8]. As additional components in them, we identified 18:1 and 17:1 hydroxyepoxy acids.

The acids with  $R_f$  0.32 were transparent in UV light and, according to their mass spectrum, consisted of saturated and monoenic dihydroxy and trihydroxy acids of the C-17 and C-18 series.

Portions of the acids of the two groups were combined, and the mixture was hydrogenated with Pd/Al. The hydrogenated product was converted into TMS derivatives and analyzed by GLC on nonpolar and polar phases, and also by mass spectrometry (Table 5).

In view of the minor amount of the OH-14:0, OH-15:0, and OH-16:0 acids and the complexity of their mass spectra, the positions of the OH groups in them were not established.

Acid	ТА	Gs	EAGs	FFA	S	HAGs, DAGs	HEAGS, DHAGS, THAGS	MAGs	polar lipids
-	I	II	I	1	11	1	1	I	1
$12:0 14:0 16:1 18:0 18:1 18:2 \Sigma_{sat}\Sigma_{unsat}$	0,6 18,8 1,1 1,7 17,7 60,1 21,1 78,9	0,5 20,3 0,8 1,1 16,3 61.0 21.9 78,1	0.2 0.6 24.3 0,5 1.5 20,4 39.5 26,6 73,4		0 6 30,9 1,3 1,3 25,7 40,2 32,8 67,2	Tr . 0,4 18,7 1.0 1.6 28,3 50.0 20.7 <b>7</b> 9,3	Tr. 0,9 25.8 1,3 20,7 49.0 29,0 71,0	Tr 1.1 24.1 Tr. 6,1 24.6 44,1 31,3 68,7	1.8 64,7 Tr 4.9 14,6 14.0 71.4 28,6

TABLE 4. Composition of the Fatty Acids of Individual Classes of the Free Lipids of Stored Cotton Seeds (%, GLC)

Class

The quantitative composition of the hydroxy acids was evaluated by comparing the results of GLC on two phases.

It follows from Table 5 and the mass spectra of unhydrogenated samples that among the monoenic hydroxy acids of the C-17 and C-18 series isomers with hydroxy groups at C-12 predominated, and among the dienic acids with conjugated systems of ethylenic bonds the 9-hydroxy and 12,13-dihydroxy isomers. The composition of the oxidized TAGs and the structures and set of oxidized FAs show that under the conditions of spontaneous heating the enzymatic oxidation of the lipids is not only intensified but takes place specifically with the predominant formation of dihydroxy compounds.

We then investigated the change in the bound lipids, which for this purpose were isolated by cold extraction. The compositions of the bound lipids I and II were determined with the aid of TLC in several systems by comparing the mobilities of the individual classes and of model compounds and by using specific reagents. In bound lipids I and II we detected qualitatively the same set of components of the glyco- and phospholipids [6], of the gossypol pigments, and of all classes of neutral lipids. Visually, lipids I were distinguished by a small amount of pigments, esters of steryl glycosides, phosphatidylcholines, and TAGs The qualitative compositions of the gossypol pigments I and II also differed. In system 6 the bound lipids I revealed a single gossypol spot ( $R_f$  0.52, G-1), while in II there were another two spots (G-2,  $R_f$  0.33; G-3,  $R_f$  0.26).

It is known from the literature that ripe cotton seeds contain not only gossypol but also its methoxy and dimethoxy derivatives [10]. There are indications that when cotton seeds are stored the amount of gossypurpurin — a product of the interaction of gossypol with amino acids [4, p. 106] — increases, and under the action of moisture at high temperatures, reactions of gossypol with phospholipids and with sugars are possible [11].

In order to identify the pigments, we isolated them from the bound lipids I and II by preparative TLC in system 6, in which the pigments are not separated from the accompanying polar lipids. The amounts of aldehyde groups of gossypol were determined spectrophotometrically in the fractions isolated, and the lipid classes were revealed by the supplementary chromatography of the fractions in systems 1, 2, and 5. To characterize the pigments we used UV spectroscopy and qualitative reactions.

The results showed that G-1 was separated in admixture with FFAs, phytosterols, esters of steryl glycosides, and monogalactosyldiacylglycerols (MGDGs). The amount of aldehyde groups in this fraction of lipids I was 27.1%, and in II 6.5%. The UV spectrum —  $\lambda_{max}^{C_2H_5OH}$  237, 258, 294, and 376 nm — and qualitative reactions with solutions of phloroglucinol and SbCl<sub>3</sub> were characteristic for gossypol.

Fraction G-2 contained steryl glycosides, digalactosyldiacylglycerols (DGDGs) and 19.1%, of aldehyde groups it had a UV spectrum with  $\lambda_{\max}^{C_2H_5OH}$  238, 260, 295, and 378 nm, and gave positive qualitative reactions with the reagents mentioned.

Fraction G-3 proved to be a mixture of DGDGs, phospholipids, and pigments containing 4.6% of aldehyde groups, and it had  $\lambda_{\max}^{C_2H_5OH}$  377 nm and gave positive qualitative reactions for gossypol.

All the pigment fractions had a yellow-brown coloration.

According to these results, the pigments of fractions G-2 and G-3 had the binaphthyl skeleton of gossypol and also aromatic aldehyde groups but were not methoxy derivatives of gossypol. No other products of the transformation of gossypol or its interaction with the organic substances of the seeds were detected.

Thus, in spite of the dark color of the kernels of the defective seeds, their lipids were less colored than those of the first-grade seeds.

Such changes in the lipid composition of the defective seeds as those in the ratio of TAGs and the products of their decomposition, DAGs and FFAs, the fall in the level of the 16:0, 18:0, and 18:1 acids, the passage of the bound form of sterols into the free form, and the specific nature of the set of oxidized TAGs are difficult to explain from the point of view of thermal damage alone.

It is known that when oil and grain seeds are stored for long periods the main factors affecting the development and intensity of spontaneous heating are a high moisture content, a high temperature of the seeds, a high relative atmospheric humidity, the presence of dirt, especially oil impurities, the degree of ripeness of the seeds, and the presence of a micro-flora [1, 3, 5, 12].

In the opinion of M. K. Kristensen [12] the microflora plays an important role in the spontaneous heating of moist seeds, since pathogenic fungi and, in particular, those of the genera *Candida* and *Aspergillus*, in the process of intensive growth can raise the temperature of the material by 20°C in a day.

On the basis of the results that we have obtained and those given in the literature, the sequence of biochemical processes in the spontaneous heating of oil seeds can be represented in the following way. Pathogenic fungi, or else "storage molds," can develop under aerobic or anaerobic conditions if there is a nutrient substrate, a high moisture content, and a certain initial temperature [12]. Plant residues present in a batch of seeds are moister and are copiously populated by field fungi, while oil impurities contain a fairly large amount of FFAs [13], which, as is well known [14], are a good substrate for microflora. Furthermore, pathogenic fungi possess a complex group of enzymes, including extracellular enzymes [15]. On parasitizing tissues enriched with lipids they are capable of splitting complex lipids.

If oil seeds of different degrees of ripeness with a high moisture content and a high degree of contamination are stored at moderate temperatures and atmospheric humidities, the optimum conditions for the intensive multiplication of pathogenic fungi are created in the heap at the points of accumulation of plant and oil residues. Because of exothermic biodegradation and the absence of gas exchange, in a certain volume — a focus of spontaneous heating — the temperature rises and the medium in the space between the seeds becomes more humid and anaerobic, since in their development the fungi consume  $O_2$  and liberate  $CO_2$ .

In their turn, in freshly harvested unripe seeds a weak activity of enzymatic, particularly respiratory, systems is still retained. A rise in temperature and an increase in humidity activate these systems [16], which is also accompanied by the liberation of energy (through respiration) and consequently, leads to a still higher rise in temperature.

Under these conditions changes take place primarily in the properties of the seed coat [17], impairing its protective functions. The possibility appears of the penetration of pathogenic fungi into the seeds, where they develop mainly in the germ, killing it and changing its color [12]. In this process they produce metabolites apparently capable of changing the biochemistry of the seeds. It has been found that in spite of a rise in the acid No. of the lipids, the lipase activity of the seeds themselves is weak under the conditions of heat damage [16]. Consequently, the change in the lipids is largely the result of the vital activity of the microflora in them.

The extracellular lipases of fungi are specific to the chain lengths of the fatty acids and to their positions in acylglycerols, and substrates for them are not only TAGs but also lipoproteins [15]. In fungi of the genus *Candida* phospholipases A<sub>1</sub> and C have been detected, the products of the vital activity of which are FAs from the sn-l positions of the phospholipids, DAGs, glycerophosphoryl bases, and 2-acylphospholipids. In unripe cotton seeds the level of polar lipids is higher than in ripe ones [7]. In the sn-l positions of the main cottonseed phospholipids the 16:0, 18:0, and 18:1 acids predominate [18], these being liberated on

$\frac{1}{1} \frac{1}{1} \frac{1}$	B**
OH - 14:0 1 0 48 0 3 215 200 (0 1)	
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	015
$\begin{array}{c c} 9 - OH - 17 : 0 \\ 12 - OH - 17 : 0 \\ 13 - OH - 17 : 0 \end{array} \right\} 1.61 15,8 357 (1,0) 341 (0,6) 301 315 $	215 173 159 <b>(</b> 12 <b>,3</b> )
9, 10(12, 13)-ep 13(9)-OH - 17:0 2,35 0.6 259	229, 159
$\begin{array}{c c} 9\text{-}OH-18:0\\ 12\text{-}OH-18:0\\ 13\text{-}OH-18:0 \end{array} \right\} 2.9  65.1  371(1,7)  355(1,3)  259(26,3)\\ 301(24,8)\\ 301(24,8)\\ 315(46,6) \end{array}$	229 (11,6) 187 (41,16) 173 (41,6)
9,10- di -OH-17:0 9,13-di -OH-17:0 9,10(12,13)-ep- 3,87 5,6 445 (0,8) 429 (0,6) 259 259	201 (6.2) 159
$\begin{array}{c ccccccccccccccccccccccccccccccccccc$	243 ( <b>4,5</b> ) 173 159, <b>26</b> 1
9.10,13 (9,12,13)- tri -OH-17:0 9,10-di -OH-18:0 5,19 5,4 5,33 (0,1) 517 (0,1) 259 070	159
9,13-di -OH-18:0 ) 12,13-di -OH-18:0 ) 5.51 4.0 459 (0,6) 443 (0,4) 259 301	173 173, 275 (2,8)
9,10,13 (9,12,13)- tri-OH-18:0 6,82 0,8 547 (0,1) 531 (0,1) 259	173

TABLE 5. Composition of the MEs of the TMS Derivatives of Hydrogenated Hydroxy Acids from Defective Cotton Seeds

**\***m/z 73−100%.

\*\*A - fragment containing the -COOCH3 end of the chain; B - fragment containing the CH3 end of the chain [8].

phospholipolysis. The FFAs, the level of which in the bound lipids and also in the DAGs and lysophospholipids will be the first to rise at moderate temperatures or on spontaneous heating, are enriched with these acids. Part of the FFAs - possibly only certain ones (16:0, 18:0, 18:1) - are utilized by the microflora, and this is also accompanied by the liberation of heat.

The lipoxygenases of pathogenic fungi have scarcely been studied, but mono- and dihydroxy and epoxy derivatives of fatty acids have been detected among their metabolites [19]. An increase in the amount of epoxide and carbonyl compounds has been detected in the spontaneous heating of sunflower seeds in the presence of a microflora [4].

The low degree of oxidation of the lipids of cotton seeds can be explained by the antioxidant action of gossypol which, in exerting this action, passes into an oxidized form capable of binding irreversibly with components of the seeds, and this makes its own contribution to the darkening of the gel fraction.

The combined action of the enzyme systems of pathogens and of the seeds causes hydrolytic and oxidative breakdown not only of the lipids but also of the sugars (the main substrates of respiratory enzymes) and proteins. The products of glycolysis are used for respiration, and proteolysis leads to a rapid change in the fractional composition of the proteins [3].

In their process of development, pathogenic fungi liberate metabolic water which raises the moisture content of the seeds and of the space between the seeds to a level promoting the multiplication of thermophilic bacteria [12]. The latter may raise the temperature to 75°C, when the pathogenic fungi die. A further rise in the temperature takes place because of the absence of gas exchange.

In spontaneous heating, the temperature rises at a high rate and therefore the hydrolytic and oxidative decomposition of the substances is limited. As a result of the denaturation and compaction of the seed protein at high temperatures, lipid-protein interactions are weakened and part of the bound lipids passes into the free form. This transition is also due to some increase in the polarity of the free lipids themselves because of an increase in them of forms more polar than TAGs (DAGs, DHAGs, MAGs). At high temperatures a reaction takes place between the insoluble sugars and the amino acids, leading to a change in the color of the kernel [2]. Colored products are also formed in the reaction of proteins with oxidized lipids [20].

Thus, spontaneous heating is a complex group of biochemical processes taking place more intensively in unripe seeds when these are damaged by storage molds.

The efficiency of the oil-extracting industries is largely determined by the quality of the raw material processed. The increasing proportion of cottonseed raw material harvested by machines is raising the amount of low-grade seeds in the raw material and contributing to their defective state. On evaluating the technological properties of defective seeds on the basis of the results obtained, the following conclusions can be drawn.Defectiveness is not reflected in the index of the yield of crude oil, but because of the low level of TAGs in this oil the index of the yield of neutral oil falls. The oil from defective seeds has low indices of acidity and gossypol content but higher levels of polar lipids and of nonpolar and oxidized acylglycerols, which may be reflected in the efficiency of alkali refining.

## EXPERIMENTAL

UV spectra were taken on an Hitachi instrument and mass spectra of a MKh 1310 (evaporator bulb 60-70°C; ionization chamber 100°C; 50 eV). GLC was performed on a Chrom-4 instrument. The MEs of the ordinary FAs and of the TMS derivatives of the hydrogenated hydroxy acids were analyzed on a  $4 \times 3700$  mm column containing 15% of Reoplex 400 on Chromaton N-AW-DMCS at 200°C; and the MEs of the TMS derivatives of the hydrogenated acids additionally on a  $4 \times 2500$  mm column containing 3% of OV-1 on Chromaton N-AW-DMCS at 210°C. The chart speed was 0.5 cm/min.

CC on silica gel L 100/160 and TLC on silica gel L 5/40 (Chemapol) were performed as described in [21]. CC on polyamide (particle size 1 mm) previously washed with acetone was carried out at a sample—adsorbent ratio of 1:8 with a  $3.4 \times 106$  cm column, the neutral lipids being eluted with hexane—diethyl ether (1:1), and gossypol and polar lipids first with acetone and then with methanol.

The following systems were used for TLC: 1) chloroform methanol-25% NH<sub>4</sub>OH (65:25:4); 2) chloroform methanol-water (65:35:5); 3) hexane-diethyl ether (1:1), 4) hexane-diethyl ether (7:8); 5) heptane-methyl ethyl ketone-CH<sub>3</sub>COOH (43:7:0.5); and 6) benzene-methanol (20:5) [22].

Qualitative reactions for epoxy groups, phosphorus, glycolipids, and gossypol were performed as described in the literature [11, 23].

Samples of seeds were selected in the Tashkent oils and fats combine in June, 1984. The lipids were extracted and the indices of the products were determined as described in [6]. Gossypol was determined in accordance with [11, p. 218]. The cold extraction of free and bound lipids was carried out by steeping with hexane and with a 2:1 mixture of chloroform and methanol, respectively.

The TMC derivatives were obtained as described in [24].

The hydroxy acid MEs were hydrogenated by adding to a solution of 40 mg of the sample in 2 ml of methanol 2 ml of a 5% solution of PdCl<sub>2</sub> in methanol with 50 mg of aluminum powder and heating at  $80^{\circ}$ C for 5 h.

## SUMMARY

Defective cottonseeds differ from first-grade seeds by a smaller amount of gossypol and its free form and by higher levels of the 18:2 acid in the bound lipids and of diacylglycerols, polyhydroxy lipids, and polar lipids in the free lipids.

The oxidized acylglycerols of the defective seeds contain two residues of ordinary and one residue of isologs of monohydroxy, hydroxyepoxy, dihydroxy, or trihydroxy fatty acids with chain lengths of 18 and 17, and also of monohydroxy acids with 16, 15, and 14 carbon atoms.

On the basis of the change in the lipid composition of the seeds subjected to heat spoilage, it is proposed to consider the process of spontaneous heating as a complex group of enzymatic processes taking place mainly in unripe seeds on their damage by a pathogenic microflora.

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