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POLYPRÉNOLS OF NEW VARIETIES OF COTTON

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Abstract

This study presents the results of research conducted on the homologous composition of polyprénols found in the leaves of new cotton (*Gossypium*, *Malvaceae*) varieties. The analysis revealed that the leaves of early-maturing, salt- and drought-tolerant, as well as disease-resistant cotton varieties – Narpay, Kelajak, UzFA-703, and UzFA-705-can serve as promising sources of polyprénols. The extraction of polyprénols was carried out using standard chromatographic techniques. The study contributes to the identification of plant-based sources of biologically active compounds and supports the utilization of cotton plant by-products in biotechnology and pharmacology. These results suggest the potential for further development and application of polyprénol-containing preparations derived from cotton leaf biomass, particularly from stress-tolerant cultivars bred under the conditions of Uzbekistan.

Keywords: leaves, *Gossypium*, *Malvaceae*, UzFA-703, UzFA-705, Kelajak, Narpay, extraction, HPTLC, polyprénols

Introduction

Cotton is a versatile plant capable of meeting many human needs (Sadykov A. S., 1985). It belongs to the genus *Gossypium* of the family *Malvaceae*, with numerous regionally adapted varieties differing in earliness, wilt resistance, growth type (dwarf or tall), and fiber type (fine-fibered, medium-fibered, etc.) (Khidirova N. K., Mamatkulova N. M., Kikteev M. M., Shakhidoyatov Kh. M. 2000;

Mamatkulova N. M., Eshmirzaeva N., Khidirova N. K., Shakhidoyatov Kh. M., 2001). All parts of the cotton plant are valuable raw materials for industry, and around 1,200 different products essential to human life can be obtained from them. For example, hydrolyzed alcohol and protein-rich fodder yeast can be derived from the stems; gossypol and medicinal preparations from the roots; citric and malic acids, the growth stimulant

Uchkun, and a poultry feed concentrate provisionally named *Gossipren* from the leaves (Khidirova N. K., Zakirova R. P., 2019; Orpov A. O., Khidirova N. K., Akhmedov B. N., Ismatova R. A., Sheralieva S., 2019; Khidyrova N. K., Asatova S., Mamatkulova N. M., Yuldashev S. U., Umarov A. A., Shakhidoyatov Kh. M., 2008; Zakirova R. P., Kurbanova E. R., Khidirova N. K., 2020).

The chemical composition of cotton leaves has been studied under the guidance of Academician A. S. Sadykov, later under Professors Ya. V. Rashkes and Kh. M. Shakhidoyatov. At the end of the last millennium, Abu Ali Ibn Sina (Avicenna) wrote that cotton seeds help with cough, chest, and stomach diseases, and that cotton leaf juice stops childhood diarrhea (Abdullaeva D. A., 2017).

In modern traditional medicine, cottonseed oil is used to treat skin diseases. Cotton ash is applied externally as a hemostatic agent (Sadykov A. S., Turulov A. V., 1967). In official medicine, the plant is still under investigation. It has been determined that the total flavonoids from cotton flowers have capillary-strengthening, hypotensive, anti-inflammatory, hepatoprotective, and gastroprotective properties. Gossypol exhibits antiviral, immunostatic, and antitumor properties (Rashkes A. M., Khidyrova N. K., Shakhidoyatov Kh. M., 1997; Ismailov A. I., Karimdzhannov A. K., Islambekov Sh. Yu., Rakhimkhanov Z. B., 1994; Khidyrova N. K., Shakhidoyatov Kh. M., 2002).

Based on this, the use of cotton leaves as a valuable and accessible raw material for producing agricultural and medicinal preparations can be considered.

Previously, we studied polyphenol concentrates isolated from the leaves of cotton varieties T-1, 108-F, S-6524, and others (Rashkes Ya. V., Rashkes A. M., Nadzhimov U. K., Khidyrova N. K., Shakhidoyatov Kh. M., Musaev Zh. A., 1989). There is great interest in polyphenols as potential agents for wound healing due to their high regenerative activity (Syrov V. N., Vais U. V., Egamova F. R., Khushbaktova Z. A., Mamatkulova N. M., Shakhidoyatov R. Kh., 2012). When applied to flat full-thickness skin wounds in rats, the polyphenol concentrate from cotton leaves had a pronounced effect on proliferative and synthetic processes, thereby accelerating wound

healing. The effect of the investigated polyphenol concentrate was similar to that of sea buckthorn oil, but manifested to a greater extent (Vais E. V., Yusupova S. M., Zokirova U. T., Khushbaktova Z. A., Syrov V. N., 2019).

Polyphenol fractions were extracted from cotton leaves with high yields of 1–3.5% of the plant's air-dry mass (ADM). Their chemical structure and homologous composition were determined, and standardization was performed (Rashkes A. M., Khidyrova N. K., Rashkes Ya. V., Mirkhodjaev U. Z., Nadzhimov U. K., Shakhidoyatov Kh. M., 1995). It was established that polyphenols from cotton leaves exhibit gastroprotective activity in models of "stress-induced," acetylsalicylic acid-induced, and ethanol-induced ulcers, and also have therapeutic effects in a model of chronic ulcers caused by acetic acid (Vais E. V., Yusupova S. M., Zokirova U. T., Khushbaktova Z. A., Syrov V. N., 2019).

Materials and Methods

A study was conducted on the polyphenols of cotton leaves from newly developed promising varieties: UzFA-703, UzFA-705, Kelajak, and Narpay, created by researchers from the Institute of Genetics and Experimental Plant Biology (Kakhkharov I. T., 2010).

The *Kelajak* variety was developed at the Institute of Genetics and Experimental Plant Biology of the Academy of Sciences of the Republic of Uzbekistan (Kakhkharov I. T., 2010). It was obtained by crossing the cotton species *Gossypium hirsutum* L. and *Gossypium barbadense* L., resulting in the L-6161 line. Through hybridization between L-6161 and the Bulgarian variety 146 (*Gossypium hirsutum* L.), the early-maturing *Kelajak* bush variety was created. The plant has a large bush type with a main stem height of 80–90 cm and 3–5-lobed leaves. Its average yield is 37.2 centners per hectare, and the vegetation period is 110–115 days. *Kelajak* differs from other varieties in fiber yield and quality, adaptability to different climatic conditions, early ripening, high yield, drought and salt tolerance, and its characteristic of self-pruning.

The *UzFA-703* variety was developed by hybridizing lines L-6161 and L-45. The main stem height is 100–120 cm, stems are pale green, and leaves are 3–5-lobed. The average

yield is 36.3–40.0 centners per hectare, with a vegetation period of 110–115 days. It differs from standard varieties by its earliness, high yield, and tolerance to drought and salinity.

The *UzFA-705* variety was created through multiple hybridizations between line L-6161 and line L-417. The main stem height is 90–100 cm, stems are green, and leaves are 3–5-lobed. Its average yield is 35.0–41.0 centners per hectare, with a vegetation period of 115 days.

The *Narpay* variety was developed from line L-470. It stands out for its fiber quality, early ripening, wilt resistance, drought and salt tolerance, and high yield under diverse climatic conditions. The main stem height is 100–120 cm, stems are pale green, and leaves are 3–5-lobed. The average yield is 37.0 centners per hectare, and the vegetation period is 115 days.

The *Namangan-77* industrial variety was developed by crossing variety 159-F with the semi-wild species *Gossypium punctatum*. Through subsequent selection from this cross, the industrial early-ripening *Namangan-77* variety was developed (authors: V. A. Avtanomov, M. Saidakhmedov). The plant has a large bush, the main stem height is 100–110 cm, and it is wilt-resistant. The average yield is 29–52 centners per hectare, with a vegetation period of 100–115 days.

Plant Material

Cotton leaves were collected from the lower tiers of plants grown on the same plot in September 2024.

The total extractive substances and neutral compounds were isolated according to described methods. A standard sample was obtained from the leaves of *Rhus coriaria* (Mamatkulova N. M., Khidirova N. K., Mamadrakimov A. A., Shakhidoyatov Kh. M., 2014).

Qualitative and quantitative analysis of polyprenols was performed using high-performance thin-layer chromatography (HPTLC) with a device from Camag (Switzerland) (Khidyrova N. K., Mamatkulova N. M., Mukarramov N. I., Shakhidoyatov Kh. M., 2012).

Isolation of Polyprenols (General Method)

Air-dried leaves of cotton varieties *Ke-lajak*, *Narpay*, *UzFA-703*, and *UzFA-705*

(100 g each) were sequentially extracted with 3000 ml of 96% ethanol (1000 ml × 3), followed by 300 ml of benzene. To each extract, 90 ml of a 50% aqueous potassium hydroxide (KOH) solution was added, and the mixture was shaken at room temperature for one hour. The mixture was then diluted with water, and the benzene layer was separated, washed with water until neutral, and dried over anhydrous Na₂SO₄.

The organic layer was concentrated using a rotary evaporator to obtain the total extractives. Ten grams of the extractive mixture were weighed and subjected to column chromatography for fractionation. Silica gel of the KSK brand (100/250 mesh) was used as the adsorbent, with an adsorbent-to-substance ratio of 40:1. Elution was carried out first with hexane, then with a hexane–chloroform system, gradually increasing polarity.

As a result, polyprenol fractions were obtained with purity ranging from 94% to 98%, and yields of 2.82%, 2.72%, 2.49%, and 2.52% of the air-dry plant mass, respectively. The purity of polyprenols was monitored using thin-layer chromatography (TLC) on Silufol UV-254 plates in solvent systems: benzene–ethyl acetate (24:1) or hexane–chloroform (1:2). The detection reagent was a 1% solution of vanillin in concentrated sulfuric acid. The content of polyprenols in the fractions was determined using TLC and high-performance TLC (HPTLC) methods (Mamatkulova N. M., Eshmirzaeva N., Khidirova N. K., Shakhidoyatov Kh. M. 2001; Khidyrova N. K., Mamatkulova N. M., Mukarramov N. I., Shakhidoyatov Kh. M. 2012).

Discussion of Results

To determine the polyprenol content, the total amount of neutral compounds (NC) was isolated using the method described in (Mamatkulova N. M., Khidirova N. K., Mamadrakimov A. A., Shakhidoyatov Kh. M. 2014), and the content of polyprenols was determined by high-performance thin-layer chromatography (HPTLC) (Khidyrova N. K., Mamatkulova N. M., Mukarramov N. I., Shakhidoyatov Kh. M. 2012). The results are summarized in Table 1.

Table 1. Yields of total extractives (TE), neutral compounds (NC), polyprenols (PP) in % of air-dry mass and polyprenol homologue content in % of sample weight

№	Cotton Variety	TE, %	NC%	PP%	PP Homologue Content		
					n=10	n=11	n=12
1.	Namangan-77	11.6	5.2	2.53	Trace	23.10	8.5
2.	Kelajak	11.5	4.1	2.82	14.90	75.71	9.39
3.	Narpay	12.5	4.4	2.60	8.40	80.58	11.02
4.	UzFA-703	10.1	3.8	2.49	Trace.	85.27	14.72
5.	UzFA-705	11.1	4.0	2.57	Trace	82.6	17.29

* Standard polyprenols isolated from sumac (*Rhus coriaria*) leaves contained polyisoprenoid units ranging from C10 to C13, with the following composition: decaprenol (5.37%), undecaprenol (60.85%), dodecaprenol (20.47%), and tridecaprenol (13.31%)

Discussion of Results

The data presented in Table 1 and HPTLC analysis of polyprenols indicate that the isolated compounds consist of three polymer homologues containing 10–12 isoprenoid units, with undecaprenol (n=11) being the dominant component.

Among the studied varieties, Kelajak showed the highest polyprenol content. This variety is characterized by self-topping, which is economically beneficial as it eliminates the need for mechanical topping. The quantitative composition of polyprenol homologues in Kelajak was as follows: decaprenol – 14.90%, undecaprenol – 75.71%, and dodecaprenol – 9.39% (Figure 1).

In other cotton varieties such as UzFA-703 and UzFA-705, the homologue profile is slightly different: decaprenol is present in trace amounts, while undecaprenol content reaches 85.27% and 82.6%, respectively.

For identification, polyprenols were isolated using column chromatography (CC) on silica gel with yields ranging from 2.49% to 2.82% of the air-dry plant material. Their structure was confirmed using IR spectroscopy, ^1H and ^{13}C NMR spectroscopy, and mass spectrometry.

IR spectral data of polyprenols included:

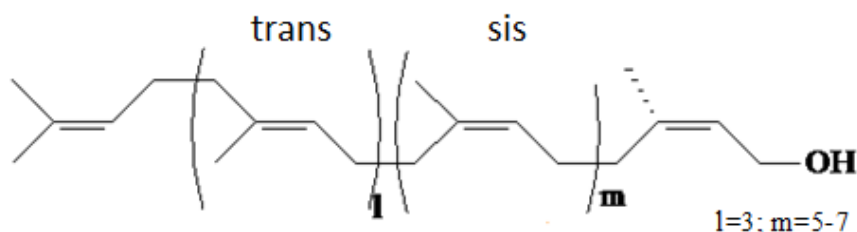
Absorption at 837 cm^{-1} : characteristic of C–H deformation in tri-substituted olefins ($\text{CH}_2\text{--C}(\text{CH}_3)=\text{CH--CH}_2$), 1001 cm^{-1} : C–O stretching in allylic primary alcohols ($\text{CH=CH--CH}_2\text{OH}$), 1376 cm^{-1} : C–H deformation in CH_3 groups, 1448 cm^{-1} : CH_2

and CH_3 group vibrations, 1668 cm^{-1} : C=C stretching in the isoprenoid chain, 2856 , 2927 , 2962 cm^{-1} : C–H stretching of CH_2 and CH_3 groups, 3024 cm^{-1} : =CH group vibrations, 3561 cm^{-1} : free hydroxyl group, 3332 – 40 cm^{-1} : associated hydroxyl groups from polymer interactions.

Mass spectrometry revealed weak molecular ion peaks at: m/z 698 ($\text{C}_{50}\text{H}_{82}\text{O}$) – dodecaprenol, m/z 766 ($\text{C}_{55}\text{H}_{90}\text{O}$) – undecaprenol, m/z 834 ($\text{C}_{60}\text{H}_{98}\text{O}$) – tridecaprenol, along with fragment ions at m/z 680, 748, 816, and smaller fragments 153, 135; 85, 67; 69, consistent with known fragmentation patterns for polyprenols from other sources (Khidyrova N. K., Rakhmatova M. Zh., Kukina T. P., Shakhidoyatov Kh. M.).

^1H NMR spectrum showed: Singlets at δ 1.62 and 1.56 ppm – cis- and trans-methyl groups ($\text{CH}_2\text{--C}(\text{CH}_3)\text{--CH}$), Multiplet at δ 1.90–2.05 ppm (centered at 1.96 ppm) – methylene protons of the isoprenoid chain, Doublet at δ 3.97 ppm ($J = 7.5\text{ Hz}$) – methylene protons of the terminal $\text{--CH--CH}_2\text{OH}$ group, Broad singlet at δ 5.05 ppm – olefinic protons in the middle of the chain, Triplet at δ 5.31 ppm ($J = 7.5\text{ Hz}$) – terminal olefinic proton.

^{13}C NMR spectrum showed: δ 32.21 ppm – methylene carbons in trans-cis (E-Z) arrangements, δ 32.41 ppm – cis-cis (Z-Z), δ 39.91–39.93 ppm – trans-trans (E-E) and terminal trans (ω -E) configurations. These chemical shift values confirm the structure of cotton-derived polyprenols.



Conclusion

The conducted research demonstrated that the polyprenol content in the leaves of early-maturing, drought- and salt-tolerant cotton varieties – Kelajak, Narpay, UzFA-703, and UzFA-705-was 2.82%, 2.72%, 2.49%, and 2.52% of dry matter, respectively. Among these, the Kelajak variety showed the highest polyprenol yield. It is es-

pecially noteworthy for its self-topping trait, which is advantageous from an agricultural and economic standpoint, reducing the need for mechanical topping. These findings indicate that the Kelajak variety is a promising source of biologically active polyprenols and can be used for further biochemical and pharmacological studies in natural product research.

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