## LIPID COMPOSITION OF PAULOWNIA SEEDS GROWN IN BULGARIA

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### ABSTRACT

The chemical composition of seeds from paulownia (*Paulownia tomentosa*) was investigated. The main components in the triacylglycerol fraction of the oil were linoleic (64.1%), oleic (21.2%) and palmitic acids (7.3%).  $\gamma$ -Tocopherol (approx. 100.0%) predominated in the tocopherol fraction, and in the sterol fraction –  $\beta$ -sitosterol (79.2%), campesterol (10.3%) and stigmasterol (7.7%). In the seeds were established 10.6 % protein, 9.5 % cellulose and 38.2 % hydrolysable carbohydrates.

**Key Words:** Paulownia tomentosa seeds, chemical composition, seed oil, fatty acids, sterols, tocopherols.

## INTRODUCTION

Paulownia (fam. *Scrophulariaceae*) is a subtropical tree, which grows in many countries all over the world. It is a comparatively new plant for Bulgaria, where the main species grown is *Paulownia tomentosa* is grown in the country. The tree is cultivated for its valuable wood, which is used to produce musical instruments, furniture wooden planes, etc. It is used for building griddles for wind protection, for recovering burnt down forests and for antierosion plantations. The plant is also used for production of high quality honey, because its blossom has a pleasant aroma (Hu, 1961; Bonner and Burton, 1974).

The leaves of the plant act as a dust catcher in polluted areas. It was found that they could be used for animal fodder because of their biochemical composition (Zima et al., 2010).

Extracts, rich in flavanoids were obtained from the fruit of *Paulownia tomentosa*. They are widely used in medicine due to their antiradical activity (Smejkal et al., 2007a, b).

The fruits contain from 800 to 1000 seeds (Bonner and Burton, 1974). There is no written data concerning the content and composition of the biological active substances of seed oil from paulownia, such as fatty acids, sterols and tocopherols. The above issue is the main subject of the present research.

### **MATERIALS AND METHODS**

*Samples.* The seeds from mature fruit of *Paulownia tomentosa*, harvest 2010, in the region of Plovdiv, Bulgaria, were used for investigation. The plant material was milled (0.5 mm) and its moisture content was determined by drying it up to constant weight, at 105°C (Russian Pharmacopoeia, 1990). The samples were analyzed for the content of: seed oil, proteins (Vinogradova et al., 1991), cellulose (Updegraff, 1969), hydrolysable carbohydrates (Vinogradova et al., 1991). The values were represented on the base of absolute dry weight.

*Isolation of seed oil and determination of oil content:* The seeds (100 g sample) were extracted with n-hexane in Soxhlet for 8 h. The solvent was partly removed in a rotary vacuum evaporator, the residue was transferred to pre-weighed glass vessel and the rest of the solvent was removed under stream of nitrogen to a constant weight, in order to determine the oil content (ISO 659, 1998).

Sterols. Unsaponifiables were determined by weight after saponification of the glycerides oil and extraction with hexane (ISO 18609, 2000). The unsaponifiable matters (100 mg, precisely measured) was applied on 20 cm x 20 cm glass plates (ca. 1 mm thick Silica gel G layer) and developed with n-hexane : acetone, 100 : 8 (by volume). Free sterols ( $R_f = 0.4$ ) were detected under UV light by spraying the edges of each plate with 2',7'-dichlorofluorescein, they were then scraped, transferred to small glass columns and eluted with diethyl ether. The solvent was evaporated under a stream of nitrogen and the residue was weighed in small glass containers to a constant weight. Sterol composition was determined by GC using HP 5890 gas chromatograph (Hewlett Packard GmbH, Vienna, Austria) equipped with a 25 m x 0.25 mm DB – 5 capillary column (Agilent Technologies, Santa Clara CA, USA) and a flame ionization detector. Temperature gradient was from 90°C (hold 2 min) up to 290°C at a rate 15°C/min and then up to 310°C at a rate of 4°C/min (hold 10 min); the injector temperature was 300°C and the detector temperature was 320°C. Hydrogen was used as carrier gas at a flow rate 0.8 ml/min; split 50:1. Identification was confirmed by comparison of retention times with those of a standard mixture of sterols ( ISO 12228, 1999).

*Tocopherols.* Tocopherols were determined directly in the oil by high performance liquid chromatography (HPLC) by a Merck-Hitachi (Merck, Darmstadt, Germany) unit equipped with a 250 mm x 4 mm Nucleosil Si 50-5 column (Merck, Darmstadt, Germany) and a fluorescent detector Merck-Hitachi F 1000. The operating conditions were as follows: mobile phase n-hexane:dioxan, 96:4 (by volume), flow rate 1.0 ml/min, excitation 295 nm, emission 330 nm. 20 µkl 1% solution of crude oil were injected. Tocopherols were identified by comparing the retention times to those of authentic individual pure tocopherols. The tocopherol content was calculated on the base of tocopherol peak areas in the sample vs. tocopherol peak area of the standard tocopherol solution (ISO 9936, 1997).

*Fatty acids*. The total fatty acid composition of the oil was determined by GC after transmethylation of the respective sample with 2N methanolic KOH at 50°C according to Christie, 2003. Fatty acid methyl esters (FAME) were purified by TLC on 20 cm x 20 cm plates covered with 0.2 mm Silica gel 60 G layer (Merck, Darmstadt, Germany) with mobile phase n-hexane : acetone, 100 : 8 (by volume). Determination was performed on a gas chromatograph equipped with a 30 m x 0.25 mm x 25  $\mu$ m (I.D.) capillary EC 30-Wax column (Hewlett Packard GmbH, Vienna, Austria) and a flame ionization detector. The column temperature was programmed from 130°C (hold 4 min), at 15°C/min to 240°C (hold 5 min); injector and detector temperatures were 250°C. Hydrogen was the carrier gas at a flow rate 0.8 ml/min; split was 50:1. Identification was performed by comparison of retention times with those of a standard mixture of FAME subjected to GC under identical experimental conditions (ISO 5508, 2000).

## RESULTS

The general chemical composition of the seeds was presented in Table 1, the fatty acid composition of the oil - in Table 2, and the individual sterol composition - in Table 3.

Characteristics	Content, % (wt/wt)
Moisture	15.5
Seed oil	20.3
Protein	10.6
Cellulose	9.5
Hydrolysable carbohydrates	38.2

Table 1:	Chemical Composition of Seeds

	Fatty acids	Content, % (wt/wt)		
Saturated				
C <sub>12:0</sub>	Lauric	0.4		
C <sub>14:0</sub>	Myristic	0.1		
C <sub>15:0</sub>	Pentadecanic	0.1		
C <sub>16:0</sub>	Palmitic	7.3		
C <sub>17:0</sub>	Margarinic	0.1		
C <sub>18:0</sub>	Stearic	3.6		
C <sub>20:0</sub>	Arahinic	0.4		
C <sub>22:0</sub>	Behenic	1.0		
	Monou	nsatured		
C <sub>14:1</sub>	Myristoleic	0.5		
C <sub>16:1</sub>	Palmitoleic	0.1		
C <sub>18:1</sub>	Oleic	21.2		
C <sub>20:1</sub>	Gadoleic	0.5		

# Table 2. Fatty Acid Composition of Seed Oil

Polyunsaturated				
C <sub>18:2</sub>	Linoleic	64.1		
C <sub>18:3</sub>	Linolenic	0.3		
C <sub>20:2</sub>	Eicosadienoic	0.3		

# Table 3: Sterol Composition of Seed Oil

Sterols	Content, % (wt/wt)	
Cholesterol	tr.	
β-Sitosterol	79.2	
Campesterol	10.3	
Stigmasterol	7.7	
$\Delta^5$ - Avenasterol	1.2	
$\Delta^7$ - Stigmasterol	1.2	
$\Delta^7$ - Avenasterol	0.4	

### DISCUSSION

The contents of protein, cellulose and hydrolysable carbohydrates were lower than those in the above mentioned reports ((Hu, 1961; Bonner and Burton, 1974). The amount of substances examined in our study and forming the chemical composition of the seeds was approximately 94.1%. Non-nitrogenous extractible substances which formed the rest up to 100.0% are not part of our study.

Data in Table 2 show that 15 fatty acids were determined, constituting 100 % of the total oil content. The correlation saturated : unsaturated fatty acids was 13.0 : 87.0, as monounsaturated acids were 22.3% and polyunsaturated – 64.7%. Palmitic acid (7.3%) predominated in the fraction of saturated fatty acids, representing 56.2% of their total content. Oleic acid (21.2%) was predominant among the monounsaturated acids, representing 24.4% of their total content, and linoleic acid (64.1%) – among the polyunsaturated, with a share of 73.7%. The results showed that regarding their content of the oil, paulownia seeds were similar to other nontraditional oil-bearing materials such as grape seeds, water melon, melon, tobacco, as well as to fruits from the *Lamiacea* family (Shterbakov, 1963; Zlatanov and Antova, 2004).

Such information concerning fatty acid composition is not found in the specialized literature. This is the reason why a comparison cannot be done.

The fatty acid composition of paulownia seeds compared to those of other trees, such as almond, apricot, plum and cherry showed that the correlation oleic-linoleic acid was reversed. For the examined seeds of paulownia it was 1:3, while for the above mentioned materials it was 3:1, as in the case of almond (respectively 77.0 - 84.0% and 20.0%), apricot (respectively 60.0 - 79.0% and 18.0 - 32.0%), and plum (respectively 70.0 - 72.0% and 20.0 - 24.0%); for cherry fruit it was 2:1 (respectively 47.0 - 50.0% and 28.0%) (Popov and Ilinov, 1986; Shterbakov, 1963). The higher content of linoleic acid in paulownia seeds could be attributed to the special features of the species, related to its pattern of cultivation.

Regarding the individual presence of oleic and linoleic acid, the oil from paulownia seeds was similar to the oils from other nontraditional materials such as grape seeds (respectively 12.0 - 33.0% and 45.0 - 72.0%), watermelon (respectively 13.0 - 20.0% and 60.0 - 70.0%), tobacco (respectively 15.0 - 24.0% and 60.0 - 78.0%) and poppy seeds (respectively 28.0 - 30.0% and 60.0%) (Popov and Ilinov, 1986; Shterbakov, 1963). The general fatty acid profile, like that of the edible sunflower oil, was found to contain very low amounts of the undesirable saturated palmitic acid, in contrast to the widely used olive oil (7.5 - 20.0%) and peanut oil (6.0 - 16.0%) (Gunstone et al., 2007).

Sterols are present in the so called non-saponificated part (3.9%) and they are an important component of the seed oil. Their total content in the investigated oil was found to be 0.6 %. The individual composition is presented in Table 3. The most significant contribution to the total content of sterols was by  $\beta$ -sitosterol (79.2%), followed by campesterol (10.3%) and stigmasterol (7.7%). It is obvious from the data, that regarding its sterol content and composition, paulownia seed oil was similar to the findings by Zlatanov and Ivanov (1995) for fruits from the *Apiaceae* family and by Gunstone et al., 2007 for sunflower oil.

The total content of tocopherols in the oil was comparatively low – 33.7 mg/kg, and was dominated by  $\gamma$ -tocopherol (nearly 100.0%), while the rest of tocopherol types were identified only in trace amounts. It is well-known that  $\gamma$ -tocopherol has antioxidant properties, which are extremely important for increasing the antioxidant stability of the oil, as well as for widening its application in different industries – food, cosmetics, pharmacy, technology. Regarding its content of  $\gamma$ -tocopherol the examined oil proved superior to a number of common food oils, for example – corn oil (50.0 – 62.0%) and soya oil (60.0 – 85.0%), thus showing similarity to some non-traditional oils, such as pyrene oils of morello (93.1%) and apricot (96.0%) (Popov and Ilinov, 1986).

## CONCLUSION

The total content and the composition of the glyceride oil isolated from paulownia (*Paulownia tomentosa*) seeds are similar to the characteristics of the oils from other oilbearing plant seeds. Paulownia seeds can be used as a non-traditional material for producing oil rich in biologically active substances as sterols and tocopherols for nutritive purposes, as well as for an additive in fodder mixtures in order to enrich them with valuable nutrients.

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