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CHEMICAL INVESTIGATION OF *Callisia fragrans*

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Lipids of leaves, runners, and stems of Callisia fragrans *were investigated. Classes of neutral, glyco-, and phospholipids and their fatty-acid compositions were determined. The contents of ascorbic acid and organic acids and chlorophyll and carotinoid pigments in various plant organs were established.*

Key words: *Callisia fragrans*, neutral lipids, glyco- and phospholipids, fatty acids, pigments.

Callisia fragrans (basketplant) belongs to the Commelinaceae family. Its leaves and runners contain biologically active flavanoids and phytosteroids. Vitamins C, PP, and B_2 and the microelements Fe, Cr, Ni, and Cu are found in the juice [1, 2]. Basketplant is used to treat burns, tuberculosis, arthritis, bronchial asthma, infertility, and skin, oncological and cardiovascular diseases. Preparations prepared from *Callisia* leaves and runners suppress pathogenic microflora in the intestine, relieve pain, reduce the activity of the pancreas and spleen, and fight inflammation [3].

We carried out a physical chemical investigation of the leaves, runners, and stems of *C. fragrans* grown indoors and determined the contents of total lipids, organic acids, chlorophyll and carotinoid pigments, and vitamin C (Table 1).

The contents of these compounds in this plant have not been reported.

Table 1 shows that the plant mass was mainly leaves. The largest fraction of lipids was observed in runners. The total contents of carotinoid and chlorophyll pigments and ascorbic acid were dominated by those in the leaves.

Carotinoids and xanthophylls were isolated as before [4].

TLC of the acetone extract of fresh leaves on Silufol using system 1, on silica gel + $CaCO_3$ (1:1) using system 2, and on cellulose using system 3 established that chlorophylls "a" and "b" and carotenes "α" and "β" were present. Neoxanthine and anteraxanthine were identified from xanthophylls. The ratio of chlorophylls "a" and "b" in leaves was 67.0 and 23.0%, respectively, of their total mass [5].

The extract of runners and stems contained anthocyans, which had R_f 0.67 for TLC on silica gel using system 4.

Lipids of leaves and runners were separated by column chromatography (CC) over silica gel into neutral (NL), glyco- (GL), and phospholipids (PL) by elution successively with $CHCl_3$, $(CH_3)_2CO$, and CH_3OH. The contents of lipid groups were established gravimetrically and gave the following values:

Lipid class	Leaves	Runners
NL + pigments	58.6	56.4
GL + pigments	28.8	24.2
PL	12.6	19.4

It can be seen that the prevalent fractions in all plant parts were the NL and GL. TLC of NL using system 5 detected the following lipid classes: paraffinic, olefinic, and aromatic hydrocarbons; carotinoids; sterol and triterpene acetates; triacylglycerides; free fatty acids; sterols; triterpenols; triterpenic acids; and chlorophylls.

The GL composition was established by TLC using systems 7 and 8 and standard GL samples. The following GL classes were detected: sulfolipids, digalactosyldiglycerides, sterolglycosides, cerebrosides, and monogalactosyldiglycerides.

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TABLE 1. Properties of Fresh *Callisia fragrans*

Property	Leaves	Runners	Stems
Plant mass, %	53.5	12.7	33.8
Moisture, %	94.4	92.2	93.8
Content:			
total lipids per abs. dry wt., %	2.3	3.9	2.0
organic acids, %	0.19	0.18	0.14
ascorbic acid, mg%	25.6	14.1	18.3
Total carotinoid pigments, mg%	7.40	0.74	0.14
Carotenes, mg%	4.10	0.43	0.05
Xanthophylls, mg%	3.30	0.31	0.09
Chlorophylls, mg%	67.8	17.9	14.7

TABLE 2. Fatty-Acid Composition of Lipid Classes of *Callisia fragrans* (GC, mass %)

Acid	Leaves				Runners				Stems
	total lipids	NL	GL	PL	total lipids	NL	GL	PL	total lipids
10:0	1.0	1.4	0.9	1.2	0.2	Tr.	Tr.	Tr.	0.5
12:0	2.1	2.3	1.6	1.5	4.3	0.2	1.7	3.5	2.4
13:0	1.3	0.4	0.9	1.1	1.9	1.1	1.0	1.5	2.7
14:0	1.9	3.2	1.2	1.3	2.1	2.3	2.7	2.3	2.1
15:0	0.3	0.8	0.9	0.6	0.4	0.6	4.9	5.0	1.9
16:0	22.2	24.3	21.7	20.5	33.8	30.2	20.9	39.1	34.1
16:1	3.2	4.8	6.8	7.5	3.4	2.9	9.3	9.9	3.5
17:0	1.0	1.2	1.3	1.2	0.9	0.9	1.1	1.0	1.1
18:0	9.2	9.1	7.8	9.1	12.1	17.8	9.9	13.8	11.5
18:1	19.6	22.4	17.7	13.2	19.3	18.8	28.5	10.4	16.9
18:2	23.1	22.9	18.5	20.7	16.7	22.9	15.3	10.1	18.8
18:3	15.0	7.2	20.7	22.1	4.9	2.3	4.7	4.4	4.1
$\Sigma_{sat.}$	39.1	42.7	36.3	36.5	55.7	53.1	42.2	55.2	56.3
$\Sigma_{unsat.}$	60.9	57.3	63.7	63.5	44.3	46.9	57.8	34.8	43.7

PL were analyzed by two-dimensional TLC on silica gel using systems 9 and 10. This revealed seven PL classes: phosphatidylinosites (PI), phosphatidylcholines (PC), phosphatidylethanolamines (PE), phosphatidylglycerines (PG), phosphatidic acid (PA), and two unidentified PL.

Carbohydrates precipitated from the lipid extract from leaves after solvents were removed. These were analyzed by paper and thin-layer chromatography. TLC on silica gel using system 11 detected a spot with R_f 0.32 that corresponded to a glucose standard.

The fatty acid compositions of NL, GL, and PL and total lipids were determined by GC (Table 2).

The set of fatty-acid components was the same in all lipid classes. Their contents did differ. The total fraction of unsaturated acids was prevalent in lipids of leaves and GL of runners. The mass of saturated acids was elevated in other classes of runner lipids and lipids of stems. The saturated acid with the highest content in all lipid classes of the studied plant organs was 16:0. Lipids of runners and stems were especially rich in it. The fraction of 18:0 acid was significant (from ~10 to 18%) in these same plant parts. The distribution of C_{18} unsaturated acids was different for both the separate lipid classes and the plant organs. The content of 18:3 acid in PL and GL of leaves was >20%; in NL, only 7.2%; whereas it was <5% in lipids of runners and stems. The fraction of 18:1 acid was greatest in GL of runners, 28.5%; in PL of runners, only 10.4%. The content of 18:2 acid in separate lipid classes of leaves varied little, from 18.5 to 22.9%, this acid was prevalent in NL (22.9%) of runners whereas its mass was the same as 18:1 acid in PL, 10.1%.

EXPERIMENTAL

GC was performed on a Chrom-5 instrument using a column packed with 5% Reoplex-400 on N-AW at 196°C and N_2 flow rate 30 mL/min.

TLC was performed using the following solvent systems: hydrocarbons:C_6H_6:CH_3OH (60:15:4, 1), hydrocarbons:$(CH_3)_2CO$:C_6H_6:*i*-PrOH (69.5:25:4:1.5, 2), hydrocarbons:C_6H_6:$CHCl_3$:$(CH_3)_2CO$ (50:35:10:5, 3), *n*-BuOH:CH_3CO_2H:H_2O (4:1:2, 4), Et_2O:hydrocarbons (3:7, 8:2, 5:5, 5), $CHCl_3$:CH_3OH (25:1, 6), $CHCl_3$:$(CH_3)_2CO$:CH_3OH:CH_3CO_2H:H_2O (65:20:10:10:3, 7), $(CH_3)_2CO$:C_6H_6:H_2O (91:30:8, 8), $CHCl_3$:CH_3OH:NH_4OH (25%) (65:35:5, 9), $CHCl_3$:CH_3OH:CH_3CO_2H:H_2O (14:5:1:1, 10), $CHCl_3$:CH_3OH:NH_4OH (25%) (100:40:7, 11).

Total lipids were extracted three times by $CHCl_3$:CH_3OH [6]. The combined extracts were washed with $CaCl_2$ solution (0.04%) to remove nonlipid components. Carotenes were isolated by hydrocarbons; xanthophylls and chlorophylls, by $(CH_3)_2CO$:EtOH (3:1, v/v). Their quantitative contents were determined by photocolorimetric method.

Sterol and triterpenol acetates were hydrolyzed by KOH in CH_3OH (10%). The unsaponified fraction was extracted by Et_2O and analyzed by TLC using system 5. Triterpene acids were isolated as before [7] and identified by Lieberman—Burchard and Sal′kovskii reactions and by mobilities in TLC on silica gel using oleanolic acid (R_f 0.60 using system 6) as a standard.

Components of lipid groups were identified by specific qualitative reactions and chromatographic mobilities in an adsorbent layer and by comparison with standards. NL were developed by iodine vapor and aqueous H_2SO_4 (50%); GL, by α-naphthol; PL, by Vas′kovsky and Dragendorff's reagent and ninhydrin.

Hydrolysis of lipids and isolation of fatty acids and their methylation were performed as before [8].

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