

ALKALOIDS, SOME CONSTITUENTS AND ANTI-MICROBIAL ACTIVITY OF *CONVOLVULUS ARVENSIS* L.

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The three organs of *Convolvulus arvensis* L. (flower, green parts and root) were phytochemically screened. The results revealed the presence of sterols and/or triterpenes, carbohydrates and/or glycosides, proteins and/or amino acids, coumarins, flavonoids, alkaloids and /or nitrogenous base and traces of tannins. Some pharmacopoeial constants were determined. Free sugars and pectin substances were studied after hydrolysis by using Paper Chromatography (PC) and Thin Layer Chromatography (TLC) techniques. Percentage of crude protein, were found to be 25.00%, 43.65% and 18.75 % for flower, green parts and root, respectively. Compound, free and total amino acids were identified qualitatively using PC and TLC, and quantitatively by using amino acid analyzer. Aspartic acid showed the highest concentration (18.8%) in green parts, followed by Valine (5.45%) in flower and Phenyl alanine (5.71%) in root. While Alanine has the lowest concentration in both roots and flowers (0.01% and 0.32%, respectively) but in green parts it was Cystine (0.50%). Total alkaloids estimated by three methods; acid-base titration, non aqueous titration and gravimetric results were found to be 0.091, 0.10 and 0.12 % for flower, green parts and root respectively calculated as tropine. Tropine, choline hydrochloride and betain hydrochloride were isolated and identified by using IR, ^{13}C -NMR and ^1H NMR. Anti-microbial screening were carried out for the different plant organ extracts and showed broad activity against some bacterial and fungus strains.

Keywords: Tropine, choline hydrochloride, *Convolvulus arvensis*, Anti-microbial activity, alkaloids compounds.

On reviewing the literatures, it was found that the family Convolvulaceae contains a vast number of species, which vary in their chemical constituents and uses. These constituents were alkaloids, carbohydrates, lipids, phenolic

and resins. Family Convolvulaceae is one of the plant kingdom, which includes a number of very important medicinal plants.

Convolvulus scammonia L. is one of the most important plant of this family as it is the source of scammony (Tschirch and Stock, 1933) which is one of the oldest remedies known for treatment of jaundice, headache, purgative, rheumatic and skin disease (Ibn Sina, 1968 and Egyptian Pharmacopoeia, 1972).

Another important plant in this family is *Ipomoea batatas* cultivated as a vegetable crop for production of sweet potato tubers, it can be considered as a main source of human food because it is very rich in vitamins B, C, D and G, while its leaves contain insulin like compound so it is antidiabetic (Abdallah *et al.*, 1986).

In folk medicine there are many other medicinal uses of Convolvulaceae such as tonic (*Ipomoea digitata* and *Cressa cretica*), toothache (*Convolvulus bidentatus*) (Walter and Memory, 1977), purgative (*Merremia alata*, *Argyrea capitata* and *Ipomoea pedicellaris*), laxative (*Ipomoea indet*) for headache (*Ipomoea gracilis*) (Sirivon, 1973) for rheumatoid and as skin lesion to treat dermatitis caused by the stink of jelly fish (*Ipomoea pes caprae*) (Hostettmann *et al.*, 1905 and Perry and Metzger, 1980).

Reviewing the previous work *Convolvulus arvensis* L. was found to have very little chemical and biological studies. It was mentioned in folk medicine that the leaves of this plant have purgative activity (Roberto, 1982), it also used in asthma (Bulus, 1983), jaundice (Thomas, 2000) and as antihemorrhagic (Shahina, 1994).

Some phytochemical studies were carried out on *Convolvulus arvensis* L. and showed that it contains alkaloids (Hilal *et al.*, 1983 and Aripova and Yunusov, 1983) phenolic compounds (Coltte, 1960), sterols (Shahina, 1994) and resin (Jertzky and Risse, 1940). So it was found that it is of interest to study this plant and check out scientifically its uses in folk medicine and traced any new biological activities

MATERIALS AND METHODS

Plant Materials

The aerial parts (leaf, stem and flower) and root of *Convolvulus arvensis* L. were collected from El-Arish (North Sinai) during 1999 and were identified by Prof. N. El-Hadidi, Professor of Botany, Botany Department, Faculty of Science, Cairo University and by comparison with plant description in flora of Egypt (Täckholm, 1974) as well as herbarium specimens at Desert Research Center

Authentic Material

Reference material for sugars and amino acids, were purchased from E. Merck, Darmstadt, Germany.

Materials, Solvent Systems and Reagents for Chromatography

Adsorbents: pre-coated silica gel 60 F254 plates (E-Merck) for TLC and silica gel 60, (70-230 mesh, Merck) for column chromatography, sulphonated polystyrene resin, type Ultra Pac 8 for ion-exchange chromatography (IEC) were used.

Solvent systems: (a) ethyl acetate-methanol-acetic acid water (65:15:10:10), (b) butanol- acetic acid – water (4:1:5), (c) chloroform: ether (1: 1 v/ v), (d) ethyl acetate – formic acid (75:25) were used for developing the chromatoplates. The following chromatographic reagents were prepared (Stahl, 1969).

- 1- Naphthoresorcinol-sulphuric acid, for carbohydrate.
- 2- Aniline phthalate, for carbohydrates.
- 2- Ninhydrin (0.2% w/v in acetone) for amino acids.
- 3- Dragendorff's reagent, for alkaloids (Balbaa *et al.*, 1981).

For spectrophotometric estimation of amino acids in IEC fractions, ninhydrin was used as 0.3% (w/v) solution in dimethyl sulfoxide/lithium hydroxide containing 0.4% hydrantine at pH 5.2.

Apparatus

- 1- Kjel-for automatic nitrogen analyser Model 16210 (Foss America Inc.) for determination of crude proteins.
- 2- LKB 4151, Alpha plus amino acid analyzer (attached with photometric and automatic integrator) for amino acid analysis.
- 3- GLC pye unicom gas chromatographic equipped with dual flame ionization detector and dual chamber for gas chromatographic analysis.
- 4- Mass spectra were recorded using EI mass (Chro N29 MY 5526) Ver. Ion Uie
- 5- Spectra were measured on a Perkin Elmer 783 Infrared spectrometer (IR).
- 6- Varian 500 MHz spectrometer for Proton Nucleus Magnetic Resonance (¹HNMR) using TMS as internal standard.

Procedures**Phytochemical study****Phytochemical screening**

Powdered samples of the three organs of *Convolvulus arvensis* L (green part, flower and root) were subjected to preliminary phytochemical screening.

1. Preparation of carbohydrate extracts

Low molecular weight sugar components were extracted with boiling ethanol (Karawya *et al.*, 1984) (90%) from the plant powder (100 g). The residue left after evaporation of ethanol was dissolved in hot pyridine, filtered and evaporated to dryness at room temperature. The pyridine extract was dissolved in 2 ml aqueous isopropanol (10%) and saved for chromatographic study.

Water-soluble polysaccharides of the plant were obtained from the residual marc (after extraction with ethanol) by successive extraction with water extract, ammonium oxalate and dilute hydrochloric acid, proteins were removed from prepared aqueous extracts by coagulation with acetic acid (Plimmer, 1926; Kertese, 1951; Pigman, 1957 and Whistler *et al.*, 1965). Polysaccharides were precipitated from each prepared extract by the addition of ethanol 95% (4 volumes). Purification of the precipitate was carried out by solubilisation in water re-precipitation and thorough washing with ethanol. Purified precipitates were kept dry (vacuum desiccators), their yield and physico-chemical characters were recorded.

Preparation of polysaccharide hydrolysates

Samples of 100 mg of purified extracts (hot and cold) were hydrolysed by heating with 2 ml of 0.5M H₂SO₄ (Balbaa *et al.*, 1981) in sealed ampoules for 20 hours at 100°C. The hydrolysates were freed from SO₄²⁻ by treatment with BaCO₃. The resulting solutions were extracted with hot pyridine and treated as before in ethanolic extracts and saved for chromatographic study.

2. Chromatographic investigation of carbohydrate extracts

a) The two carbohydrate extracts viz. ethanolic (90%) and polysaccharide hydrolysates of the plant were examined for simple sugars by TLC (Thin Layer Chromatography) using silica gel G and solvent system a, and PC (Paper chromatography) using solvent system b and Spray reagent No. (1).
b) GLC examination: The dried polysaccharide hydrolysate, free sugars, and authentic samples (5 mg each) were separately silylated and treated as follows:

Five micro litre of trimethylsilane derivatives of the sample or authentic was injected into the gas chromatography by means of a micro-syringe applying the following condition:

Column, SE 30% packed column; initial temperature 150 °C; temperature program, 60 °C/min; final temperature of 300 °C; flame ionisation detector temperature of 220 °C; injection temperature of 270 °C; time of run, 25 min; chart speed, 1 cm/2 min; volume of sample injected, 5 micro litre; nitrogen flow rate, 30 ml/min; hydrogen flow rate, 33 ml/min and air flow rate, 330 ml/min.

3. Determination of nitrogen content (crude protein)

This was carried out according to the method of the Association of the Official Analytical Chemistry (A.O.A.C) (British Pharmacopoeia, 1980) using Kjeldahl's automatic nitrogen analyser. Determination was carried out (3 times) on 1 g air-dried powdered samples of the plant under investigation.

Extraction of free amino acids

A defatted powdered plant sample (10 g) of each organ was percolated with 50% ethanol. The concentrated residue was dissolved in absolute ethanol, left overnight in a refrigerator, and filtered to dispose the precipitated extraneous matter. The concentrated residue was finally dissolved in 2 ml 10% aqueous isopropanol and kept for both chromatographic study and quantitative determination using the amino acid analyser.

Isolation and hydrolysis of protein

A defatted powdered plant sample (40 g) of each plant organ was stirred in 10% sodium chloride solution for one hour (Lederer and Lederer, 1957) and filtered. The filtrate was treated with an equal volume of trichloroacetic acid (10%). The precipitated protein was separately collected by centrifugation, successively washed with trichloroacetic acid (5%), ethanol and ether followed by drying in vacuum desiccators.

Acid hydrolysis was carried out by refluxing 10 mg of the isolated protein of each sample with 10 ml 6N hydrochloric acid for 20 hours (Lederer and Lederer, 1957). The reaction mixture was evaporated to dryness under reduced pressure at 30 °C. A part of the residue was dissolved in 10 ml water to which 10 mg activated charcoal was added, stirred and filtered. The concentrated filtrate was dissolved in 1 ml 10% aqueous isopropanol and saved for chromatography. The other part of the residue was used for the quantitative determination of the combined amino acids.

Identification of free, combined and total amino acids

PC and TLC were performed applying ascending double development technique with solvent systems a and b. The air-dried chromatograms were revealed with ninhydrin spray reagent No. 3.

Free and total amino acids were qualitatively determined using LKB 4151 plus amino acid analyser. A powdered sample (50 mg) of the plant was hydrolysed with 6N hydrochloric acid; the evaporated hydrolysates were suspended in citrate buffer (pH 2.2) and filtered. The filtrate was diluted with buffer to 6 ml (equivalent to 50 mg of original powder) and 0.5 ml volume of the obtained solution was injected in the amino acid analyser (Steven *et al.*, 1989).

4. Extraction and isolation of the alkaloids

Fresh flower (500 gm) and two kg powder of green parts and root were separately extracted with 70% ethanol until exhaustion the alcoholic extract of each plant organ was concentrated under vacuum at a temperature not exceeding 44 °C, acidified with HCl (3%), and filtered. The filtrate obtained from each plant organ was separately extracted with chloroform to remove undesirable matters. The acidic aqueous layer was adjusted to alkaline pH with ammonia and the liberated alkaloid bases were extracted by chloroform till exhaustion

The combined chloroform extracts of each plant organs were separately filtered over anhydrous sodium sulphate and evaporated under vacuum to yield fraction I (chloroform soluble alkaloids) the water – soluble bases (chloroform – insoluble alkaloids) were precipitated as reinecates after adjustment of the pH 1-2

(50% HCL), their hydrochloride prepared and represented fraction II.

Examination of the different alkaloidal extracts of the different plant organs were carried out on silica gel G plates. Solvent systems c and d were used for samples of fractions I and II, respectively. Spots were located using Dragendorff's reagent.

Preparative TLC technique was adopted for the separation and isolation of alkaloidal bases from both fractions I and II, using silica gel G (1.0 mm thick) and pilot location of bands with Dragendorff's reagent. Bands separated from fraction I (system g) and corresponding to main spot in each plant organ. All bands were scraped off and eluted with methanol, elutes (methanol and 50 % methanol) were freed from solvents under reduced pressure, checked for purity by TLC examination using different solvent systems and kept dry for further phytochemical investigation.

Characterization and identification of the separated alkaloidal bases were done using TLC and interpretation of their $^1\text{H-NMR}$, $^{13}\text{C-NMR}$ and IR spectral data.

Quantitative estimation of total alkaloids

The following methods (Egyptian Pharmacopoeia, 1953) were used for estimation of total alkaloids content. The results were compared gravimetric, acid – base titration and non aqueous titration (BP and EP), the alkaloidal content was calculated as atropine.

Powdered samples, 100 g each were used for gravimetric estimation and extracted as previously described to yield chloroform soluble alkaloids (fraction I) Samples (0.5 g) of the residues, representing fraction I of each plant extract, were subjected to acid –base and non – aqueous titration

Tropine: (24mg) white needle crystals, mp. 232-233 °C, $R_f=0.8$ (system f) $^1\text{H-NMR}(\text{CDCl}_3)$: δ 4.1(1H, H-3), 3.2 (2H, s, H1,5), 2.2 (3H, NCH_3) and 1.8-2 (4H, complex pattern, H2,4,6,7). $^{13}\text{C-NMR}(\text{CDCl}_3)$: δ 62.5 (for C-3), 60.0 (for C-1 and C-5), 39.1 (for N-CH_3), 38.5 (for C-2 and C-4) and 26.4 (for C-6 and C-7).

Choline hydrochloride: 16 mg, white crystals, mp.303-305 °C, $R_f=0.3$ (system g). IR (Kbr.) cm^{-1} : 3000 (OH), 1400(CH), 800-1600 finger print area. It is identical with authentic material

Betaien hydrochloride: white crystals 15mg, mp.290-293 °C. $R_f=0.40$ (system g).IR (Kbr.) cm^{-1} : 2500 (CH_3), 1750 (C=O), 1400(CH), 800-1600 finger print area. It is identical with authentic material

Biological studies

Anti microbial activity

Preparation of extract

Total and successive extracts of the plant organs (fresh flower, green part and root) were dissolved in each solvent in a concentration of 10% and tested for antimicrobial activity.

Microorganisms used

Bacillus subtilis, *Micrococcus kristina*, *Sarcina maxima*, *Staphylococcus aureus*, *Actin baum*, *Salmonilla sp.*, *Moraxella lacunata*, *Echerichia coli*, *Candida albicans*, *Aspergillus niger*, *Aspergillus flavus*, *Pseudomonas auregenosa*, *Saccharonmyces cereviseae*, *Mycobacter pheli*, *Penicillium chrysogemum* and *Klebsiella pneumonia*.

Method

The antimicrobial activity was carried out using the paper disc technique (Duguid *et al.*, 1978). Sterilized paper discs (Whatmann No.3) of 0.5 cm diameter impregnated with the specific concentration of the total and successive extracts of different plant parts were placed upon the surface of the tested organism (after dryness from the solvent) inoculated plates.

After incubation at 30-32 °C for 24 hours (for bacterial organisms) and 7 days (for fungi), the plates were examined for any zone of inhibition around the disc which indicate that the organisms were affected by the tested extracts. Each treatment was replicated three times. Plates containing solvent only and free of compounds served as control. The diameters of the inhibition zone were determined in mm.

RESULTS AND DISCUSSION

Phytochemical study

1) Preliminary photochemical screening of the plant revealed the presence of: carbohydrates and/or glycosides, sterols and/or triterpenes, proteins and/or amino acids, coumarins, tannins, flavonoids, alkaloids and /or nitrogenous bases and saponins. No volatile oils or cardenolides were detected.

2) *Carbohydrate constituents*. The polysaccharides precipitated from the different extracts (hot and cold) were obtained as amorphous, greyish-white, odourless and tasteless powders. They dissolved readily in 20 parts of water at 25 °C forming viscous opalescent, colloidal solutions. They gave negative tests for mucilage (Whistler *et al.*, 1965) and positive tests for pectin's (Kertese, 1951).

The total extractable pectins amounted were found to be 0.67, 1.31 and 8.2 % for flower, green parts and root respectively (calculated on the dry weight basis).

PC and TLC examination (solvent systems A and C) and GLC of sugar components of 90% ethanol extract revealed the presence of xylose,

glucose, fructose, galactose, sucrose, and unidentified peaks. Pectin hydrolysate showed the same sugars as the free, in addition to, galacturonic acid and arabinose, absence of sucrose and the unidentified peaks.

3) Protein and amino acids content. Percentage of crude protein, as determined by the A.O.A.C method, was found to be 25.00, 43.65 and 18.75% for flower, green parts and root, respectively.

The free amino acids and amino acid composition of protein hydrolysates were qualitatively studied by PC, TLC and amino acid analyser (Table 1). They gave data for at least 17 components.

Aspartic acid was found to be the major component in green parts (18.8 mg/g), followed by cystine in flower (5.82 mg/g) and alanine in root (5.71 mg/g). Arginine, cystine and proline showed the lowest concentration (0.19, 0.50 and 0.0123 mg/g, respectively) in flower, green parts and root respectively.

4). Isolated compounds. Three alkaloids and nitrogenous bases were isolated and identified by their melting point, ¹HNMR, ¹³CNMR spectral data and super imposed IR (for the two quaternary bases only) in addition to comparison with published data (Manske *et al.*, 1950 and Geoferey, 1962) as tropine, betaine hydrochloride and choline hydrochloride

Total alkaloids estimated by three methods: Acid-base titration, non aqueous titration and gravimetric results were found to be 0.091, 0.10 and 0.12 % for flower, green parts and root respectively calculated as tropine.

Biological study

Antimicrobial activity

The flower is the most active organ of *Convolvulus arvensis* as anti microbial against most of the studied bacteria and fungi in its ether and alcoholic extracts, followed by green part ethyl acetate and alcoholic extracts and finally ether and ethyl acetate extracts of the root. *Sarcina maxima* was the most affected bacterium (inhibition zone ranged from 6 to 16 mm), affected by most extracts of different organs followed by *Staphylococcus aureus* and *Escherichia coli* (Tables 2 and 3).

TABLE (1). Free protein hydrolysate and total amino acids for different organs of *Convolvulus arvensis*.

Amino acids	Flower			Green parts			Root		
	T.	H.	F.	T.	H.	F.	T.	H.	F.
Aspartic	3.69	2.29	0.77	18.18	6.48	11.7	4.09	1.84	2.25
Glutamic	2.57	2.25	0.32	6.48	3.81	2.67	2.46	1.44	1.02
Serine	4.08	3.48	0.60	8.54	3.99	4.55	2.31	-	2.31
Glycine	1.58	1.33	0.25	6.20	4.55	1.66	0.99	0.18	0.81
Histidine	2.09	1.69	0.40	4.41	1.40	3.01	2.50	0.86	1.64
Arginine	0.19	-	0.19	0.91	0.50	0.41	1.34	1.08	0.26
Threonine	0.99	0.76	0.23	2.01	1.12	0.89	1.11	0.68	0.43
Alanine	0.32	0.32	-	0.62	0.32	0.30	0.01	-	0.01
Proline	1.39	0.91	0.48	2.58	1.40	1.18	2.35	1.35	1.00
Tyrosine	4.94	4.03	0.91	8.90	6.01	2.96	0.50	-	0.50
Valine	5.45	5.01	0.44	10.50	7.13	3.45	5.03	3.20	1.83
Methionine	4.66	0.83	3.83	3.27	1.73	1.54	2.63	0.72	1.91
Cystine	5.82	-	5.82	0.50	-	0.50	0.55	-	0.55
Isolucine	1.84	0.31	1.53	1.71	0.76	0.95	0.37	0.32	0.05
Lucine	4.21	0.63	3.58	2.93	2.03	0.90	2.62	1.22	1.40
Phenyl Alanine	3.02	0.54	2.48	2.81	1.04	1.77	5.71	5.18	0.53
Lysine	3.02	-	3.02	7.59	2.80	4.79	1.85	0.95	0.90

T= total, H=hydrolysate, F= free.

TABLE (2). Antibacterial activity of plant extracts.

Organism	Flower					Green parts			Root		
	E.	Ch	E.A.	95% Alc.	50% Alc.	E.A	95% Alc.	50% Alc	E.	E.A	50% Alc.
<i>B. sub.</i>	6	-	6	8	6	8	8	7	-	6	6
<i>Mic. k.</i>	10	-	9	12	7	8	7	6	8	9	7
<i>Sar. max.</i>	12	6	8	14	16	14	12	6	15	12	8
<i>St. aur.</i>	12	6	11	9	7	11	7	7	9	9	8
<i>Salm. sp.</i>	8	-	9	8	-	7	-	6	8	8	6
<i>Mor. lac.</i>	7	-	6	8	7	7	10	7	7	9	6
<i>E. coli</i>	6	-	8	8	7	8	8	7	6	8	9

B. sub. = *Bacillus subtilis*, *Mic. k.* = *Micrococcus kristinae*, *Sar. max.* = *Sarccina maxima*, *St. aur.* = *Staphylococcus aureus*, *Salm. sp.* = *Salmonella sp.*, *Mor. lac.* = *Moraxella lacunata*, *E. coli* = *Escherichia coli*, E.= ether, Ch= chloroform, EA.= ethyl acetate and Alc= alcohol

TABLE (3). Antifungal activity of plant extracts on fungi.

Organism	Flower				Green parts			Root		
	E	E.a	95% alc	50% alc.	E.a.	95% alc	50% alc.	E.	E.a.	50% alc.
<i>Cand. alb.</i>	7	-	-	-	-	9	-	-	7	-
<i>Asp. nig.</i>	-	-	-	-	-	-	-	-	-	8
<i>Asp. flav.</i>	-	-	-	-	-	-	-	-	6	7
<i>Ps. aur.</i>	-	-	7	8	7	-	-	-	6	6
<i>Sac. cer.</i>	8	-	-	-	-	-	-	-	7	-
<i>Myc. ph.</i>	9	-	12	10	8	8	6	-	8	8
<i>Pn. chr.</i>	-	-	6	6	-	-	-	-	-	6

Cand. alb. = *Candida albicans*, *Asp. nig.* = *Aspergillus niger*, *Asp. flav.* = *Aspergillus flavus*, *Ps. aur.* = *Pseudomonas auregenosa*, *Sac. cer.* = *Saccharomyces cerevisiae*, *Myc. ph.* = *Mycobacter phely*, *Pn. chr.* = *Penicillium chrysogenum*, E.= ether, E.a.= ethyl acetate and Alc.= alcohol.

RECOMMENDATIONS

From the present study, it was found that, *Convolvulus arvensis* can be used as a treatment to many micro organisms by preparing the crude ethanolic extract in a proper pharmaceutical form. It also can be used as good source of protein due to its high contents especially in green parts (43%).

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قلويدات وبعض المحتويات والتأثير المضاد للبكتريا لنبات العليق

امانى شفيق عواد ، نوال هداية محمد وخديجة فهمى عامر
قسم النباتات الطبية والعطرية - مركز بحوث الصحراء - المطرية - القاهرة - مصر

أظهر المسح الفيتوكيميائى لنبات العليق احتوائه على المواد الكيميائية التالية: ستروليدات و/أو ترائى تربينات، كربوهيدرات و/أو جليكوزيدات، بروتينات و/أو أحماض أمينية، كومارينات، فلافونيدات، قلويدات و/أو قواعد نيتروجينية و قليل من التانينات. تم تحديد بعض الثوابت الدستورية (نسبة الرماد الكلى والذائب فى الحمض والذائب فى الماء ونسبة الرطوبة). درست السكريات الحرة والبكتين بعد حملاته، باستخدام كروماتوجرافيا الطبقة الرقيقة والورقة والغاز السائل. تم تقدير نسبة البروتينات فى جميع أجزاء النبات (الأوراق، الأجزاء فوق أرضية والجذور) وبلغت ٢٥%، ٦٥؛ ٤٣% و ٧٥؛ ١٨% على التوالى، وقد درست الأحماض الأمينية الحرة والمركبة والكلية دراسة نوعية باستخدام كروماتوجرافيا الورقة والطبقة الرقيقة، وكميا باستخدام جهاز تحليل الأحماض الأمينية وقد اظهر التحليل أن حامض الأسبرتك هو الأعلى تركيز (١٨؛ ٨%) فى الأجزاء الخضراء ويليه حمض الفالين (٥؛ ٤٥%) فى الأزهار ثم حمض الغينيل النين (٥؛ ٧١%) فى الجذور، أما الأحماض الأقل تركيزا كان الألسين والسيستين.

تم فصل قلويد تروبين وقاعدتين نيتروجينيتين هما بيتاين هيدروكلوريد وكولين هيدروكلوريد والتعرف عليهما باستخدام أجهزة الأشعة فوق الحمراء وجهاز الرنين المغناطيسى للبروتون والكربون كما تم تحديد نسبة القلويدات تبعا لدستور الأدوية المصرى والبريطانى. تم دراسته تأثير المستخلصات المختلفة للأجزاء الثلاثة (الأزهار، الأجزاء الخضراء والجذور) على الميكروبات وقد أعطت نتائج ايجابية على بعض أنواع البكتريا والفطريات. ومن النتائج يمكن استخدام هذا النبات كمصدر للبروتين وكمضاد للبكتريا.