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PHYTOCHEMICAL STUDIES ON EGYPTIAN ARACEAE SPECIES

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The family *Araceae* is represented in Egypt by four genera viz. *Pistia, Arisarum, Biarum* and *Eminium*. *Arisarum* and *Eminium* are of the most common terrestrial genera and each is represented in Egypt by one species viz. *Arisarum vulgare* Torg-Tozz var. *veslingii* (Schott) Engl. and *Eminium spiculatum* (Blume) Ktze (= *Helicophyllum crassipes* Schott).

This paper deals with the chemsitry of these two wild Egyptian species.

Experimental and Results

Material[1]

The corms of the wild species were collected from the Western Mediterranean Coastal region extending from the west of Alexandria to Sallum. The collection of *Arisarum vulgare* was earlier than that of *Eminium spiculatum*, being in January and February, while the latter in March and April, 1963–1965. The freshly collected plant material was sliced, air dried at 50° C, and reduced to No. 60 powder.

General Analysis

The phytochemical screening of the corms of both species revealed mainly the presence of mucilages, tannins, unsaturated volatile amines and alkaloids, but did not establish the presence of essential oils, flavonoids and saponins.

Carbohydrates

Identification of the Free Sugars by Paper Chromatography

The carbohydrates were extracted from the defatted powdered corms by the pyridine method (Malpress and Morrison, 1949). Investigation of the sugars was carried out by ascending and descending paper chromatographic technique using n-butanol-acetic acid-water (4 : 1 : 5). Development of the spots was performed by spraying with p-anisidine-phosphoric acid reagent (Hough et al., 1950). The chromatograms revealed the presence of fructose, glucose, sucrose and raffinose in both species. After hydrolysis with 1 N sulphuric acid, the paper chromatography, using ethyl acetate-pyridine-water (2 : 1 : 2) (Jermyn and Isherwood, 1944), revealed the presence of fructose, glucose and galactose.

Investigation of the Mucilage

The mucilages in the corms of both species were prepared according to the method of Ahmed et al. (1965). The white mucilage powder, in each case, gave a negative test for nitrogen

[1] The systematic identification of the plants was carried out by Dr. K. H. Batanouny, Botany Department, Faculty of Science, Cairo University.

and had no reducing power. The yield, the ash, the viscosities (dynamic and kinematic) and the insoluble residue (after hydrolysis) of the mucilages of *Eminium spiculatum* and *Arisarum vulgare* are shown in Table 1.

Table 1

The yield, the ash, the viscosities (dynamic and kinematic) and the insoluble-residue (after hydrolysis) of the mucilages of *Eminium spiculatum* and *Arisarum vulgare*

Species	Mucilage %	Ash %	Viscosities		Insoluble Residue %
			Dynamic	Kinematic	
Eminium spiculatum	2.836	6.56	0.9887	0.986	2.465
Arisarum vulgare	3.967	3.86	2.8564	2.837	1.666

The hydrolysis of the mucilages was effected by heating with 4% sulphuric acid for 20 hours, on a boiling water bath. Investigation of the hydrolysates was carried out by paper chromatographic analysis (Smith, 1961).

The hydrolysate of *Eminium* mucilage was found to be composed of D-galacturonic acid, D-galactose, D-glucose, D-mannose and L-rhamnose, while that of *Arisarum* mucilage composed of D-glucuronic acid, D-galactose, D-mannose, L-rhamnose and D-glucuronolactone.

Organic Acids

Following the procedure described by Paech and Tracey (1955), and using the fresh juices of both *Eminium* and *Arisarum* corms as starting materials, a purified concentrate of the polybasic organic acids was prepared in each case. The systematic identification of the organic acids was carried out by paper as well as thin-layer chromatographic technique (Neher, 1964), using butanol-acetic acid-water (12 : 3 : 5) as solvent system and bromocresol green (Kurt, 1964) and aniline-xylose reagents (Smith, 1961) as spray reagents.

The organic acids present in the corms of *Eminium spiculatum* were found to be succinic, citric and tartaric acids, while those of *Arisarum vulgare* were found to be citric and tartaric. No free oxalic acid was detected in both species.

Lipids

The dried powdered corms of both species, were separately exhausted with petroleum ether (b. p. 50–75° C) in a continuous extraction apparatus. The last traces of the solvent were removed completely in a vacuum oven at 50° C. The comparative analyses (A.V., S.V., I.V., U.M. and T.F.A.) of both oils were carried out and the results obtained are shown in Table 2.

The fatty acids of both oils were prepared by saponification and the soap solution, after removal of the unsaponifiable matter, was rendered acidic with dilute sulphuric acid and the fatty acids were extracted with ether. The saturated fatty acids, recovered after decomposing the unsaturated acids by the permanganate oxidation method (Kuemmel, 1958), were subjected to paper chromatographic analysis according to a modified procedure (Gad et al., 1961), in which the Spireri method (1954) was used.

The chromatograms obtained revealed the presence of myristic, palmitic, stearic, arachidic, behenic and lignoceric acid in *Arisarum vulgare*, and only pal-

mitic, behenic and lignoceric acid in *Eminium spiculatum*. The unsaturated acids were determined quantitatively by the ultraviolet spectrophotometric analysis (A.O.A.C., 1957) and the data obtained are shown in Table 3.

Table 2

The percentages of oils of *Eminium spiculatum* and *Arisarum vulgare* and their physical and chemical analyses

Species	Oil %	Sp. gr.	$(n)_{30}^{D}$	F.A. %	A.V.	S.V.	I.V.	Total Fatty acids		Unsap. %
								Satd.	Unsatd.	
Eminium spiculatum	0.657	0.9506	1.480	71.267	59.714	133.55	114.50	5.69	94.31	11.656
Arisarum vulgare	0.550	0.9290	1.474	74.051	51.100	126.44	78.50	10.72	89.28	9.802

Table 3

Component fatty acids of the oils of *Eminium spiculatum* and *Arisarum vulgare*

Species	Component Fatty Acids %				
	Oleic	Linoleic	Linolenic	Diene	Satd.
Eminium spiculatum	70.1	20.2	2.00	2.01	5.69
Arisarum vulgare	65.2	19.5	1.42	3.16	10.72

Preparation and Fractionation of the Unsaponifiable Matter of Eminium spiculatum

75 mg of the purified fixed oil were saponified with 0.5 N alcoholic potassium hydroxide for 3 hours under reflux condenser in the usual manner. The alcoholic solution was concentrated by evaporation, diluted with water and shaked several times with fresh portions of ether until complete extraction was effected. The combined ethereal extracts were washed with water till free from alkalinity, then dried over anhydrous sodium sulphate. Distillation of the ether yielded 8.7 gm.

The unsaponifiable matter (7 gm) was fractionated on alumina (250 gm), and the course of the chromatographic fractionation was followed on plates of silica gel G. The results obtained are shown in Table 4.

The two non-steroidal crystalline compounds viz. $(C_7H_{16})_n$ m. p. 46–48° C, and $(C_{15}H_{30}O)_n$ m. p. 76–78° C, were shown to be a saturated aliphatic hydrocarbon and a saturated aliphatic alcohol respectively, as proved by I.R. and elementary analysis. Both showed C-H absorption maximum at 3.4 μ, CH_2 and CH_3 grouping at 6.8 and 7.2 μ. The second compound showed, in addition, OH absorption maximum at 2.85 μ.

Analysis: calculated for $(C_7H_{16})_n$ C, 84.00; H, 16.00;
found C, 84.32; H, 15.68.
calculated for $(C_{15}H_{30}O)_n$ C, 79.24; H, 13.20;
found C, 80.30; H, 12.90.

Table 4

The results, obtained from the chromatographic fractionation of the unsaponifiable matter of *Eminium spiculatum*

Fractions (10 ml)	Solvent	Sterol test	R_F I	R_F II	Empirical formula	m. p. ° C
1– 64	Petroleum ether (50–70° C)	–	0.77	–	$(C_7H_{16})_n$	46–48
65–264	Benzene-Pet. ether (30 : 70)	–	–	–	Yellow non-crystalline sub.	
265–318	Benzene-Pet. ether (50 : 50)	–	–	–	Yellow resinous liquid	
319–361	Benzene-Pet. ether (50 : 50)	–	–	0.19	$(C_{15}H_{30}O)_n$	76–78
362–491	Benzene-Pet. ether (50 : 50)	–	–	0.29 0.24 0.14		
492–515	Benzene	–	–		Resinous oil	
516–611 (EM_1)	Benzene	+	–	0.08	$C_{29}H_{50}O$	138
612–659 (EM_2)	Benzene	+	–	0.09		138–139
660–707 (EM_3)	Benzene	+	–	0.09		149–151

I: Solvent Benzene-Acetone (90 : 10).
II: Solvent Benzene.

The steroidal fractions EM_1, EM_2 and EM_3 were purified repeatedly by crystallisation from chloroform-methanol until constant m. p. They were shown to be chromatographically pure having one and the same R_F value (0.08), using silica gel G and benzene as solvent system. On the other hand when the acetyl derivative of the steroidal fractions were examined by the reactive layer technique (Peereboom, 1964), using silica gel G impregnated with silver nitrate solution, and the system of petroleum ether-chloroform-acetic acid (75 : 25 : 0.5), four spots were revealed in each of the steroidal fractions, upon spraying with antimony trichloride reagent.

Preparation and Fractionation of the Unsaponifiable Matter of Arisarum vulgare

The unsaponifiable matter of the fixed oil of *Arisarum* was prepared in exactly the same way of *Eminium* and the fractionation followed the same line of treatment. The results obtained are shown in Table 5. A saturated aliphatic hydrocarbon $(C_7H_{16})_n$, m. p. 54–56° and a saturated aliphatic alcohol $(C_{25}H_{50}O)_n$, m. p. 70–73° C, as proved by I.R. and elementary analysis, were isolated.

Analysis:	calculated	for $(C_7H_{16})_n$	C, 84.00; H, 16.00;
	found		C, 83.97; H, 16.03.
	calculated	for $(C_{25}H_{50}O)_n$	C, 81.96; H, 13.66;
	found		C, 81.46; H, 14.03.

Table 5

The results, obtained from the chromatographic fractionation of the unsaponifiable matter of *Arisarum vulgare*

Fractions (50 ml)	Solvent	Sterol test	R_F I	R_F II	Empirical formula	m. p. ° C
1– 12	Petroleum ether (50–70° C)	–	0.9	–	$(C_7H_{10})_n$	54–56
13– 32	Benzene-Pet. ether (30 : 70)	–	–	–	Yellow oily residue	
33– 50	Benzene-Pet. ether (50 : 50)	–	–	–		
51–100	Benzene	–	–	0.14	$(C_{25}H_{50}O)_n$	70–73
101–175 (AR_1)	Methanol-Benzene (1 : 99)	+	–	0.09	$C_{29}H_{50}O$	136–138

I: Solvent Benzene-acetone (90 : 10).
II: Solvent Benzene.

The steroidal fraction (AR_1) was purified by repeated crystallisation from chloroform-methanol until constant m. p. The analytical data of this fraction and its derivatives viz. the acetate and 3,5 dinitrobenzoate were identical with those of the steroidal fraction (EM_1) isolated from *Eminium*. When the acetyl derivative of the steroidal fraction (AR_1) was examined by the reactive thin-layer technique, four spots (the same as those of *Eminium*), were revealed.

Verification of the Nature of the Isolated Sterol Fractions by Gas-Liquid Chromatography and Mass Spectrometry

The sterols eluted from the alumina column, and purified by repeated crystallisation from chloroform-methanol, were further purified by precipitation as sterol digitonides (Baughton and Wheatly, 1959), and subjected to gas-liquid chromatographic analysis, using the following conditions:

Apparatus:	Pye chromatograph "104".
Column dimension:	Glass column, 90 cm length and 0.25 cm diameter, U tube
Solid support:	Celite gas chromosorb P 80/100 mesh.
Stationary phase:	Methyl substituted gum rubber (General Electric Co.) SE-30 (2.7%).
Detector:	Flame ionization.
Carrier gas:	Nitrogen, flow rate of 60 ml/minute.
Temperature:	260° C, injection pre-heater, 280° C.
Recorder:	Honeywell with chart speed of 1 inch/3 minutes.
Sensitivity:	50×10^3.
Sample size:	2–5 µl (containing 20–50 µg) of 1% solution of the sample in spectroscopic chloroform.

n-Octacosane was added to all the samples, before injection as internal standard.

The results obtained, showed that the isolated sterols (EM_1, EM_2 and EM_3) from *Eminium*, as well as AR_1 from *Arisarum*, are a mixture of three sterols viz. β-sitosterol, stigmasterol and campesterol.

The most decisive information about the exact composition of the steroid fractions was obtained by the molecular mass spectrographic analysis. The parent peak (m/e 413) in the mass spectrum corresponded to the unsaturated sterol C_{29} (β-sitosterol). Additional peaks (m/e 411, 399 and 397) corresponding to stigmasterol, campesterol and dehydrocampesterol were also detected. The percentages of the different sterols in the steroidal fractions, as obtained from mass spectrum analysis, were shown in Table 6.

Table 6

The percentages of the component sterols of the steroid fractions of *Eminium spiculatum* and *Arisarum vulgare*

Steroid Fraction	Sterols %			
	β-sitosterol	Stigmasterol	Campesterol	Dehydro-campesterol
EM_1	45.2	25.0	23.1	6.7
EM_2	25.2	23.0	44.7	7.1
EM_3	14.1	11.9	66.3	7.7
AR_1	47.7	19.8	25.8	6.7

Proteins

The amino acids in the proteins's hydrolysate of the defatted powdered corms of both *Eminium* and *Arisarum* were studied chromatographically. Acid hydrolysis was carried out by 6 N hydrochloric acid in a sealed tube according to the method of Cramer (1954). The alkaline hydrolysis was carried out with barium hydroxide (Block et al., 1958).

A two-dimensional paper chromatographic technique was used to determine the composition of the acid hydrolysis using n-butanol-acetic acid-water (4 : 1 : 5) in one direction and phenol-water in the other direction. The identification of the amino acids was accomplished by comparing the chromatogram with that shown by Zahn (1951), who used sec. butanol-formic acid-water in one direction and phenol-water in the second. According to Cramer (1954), the butanol-acetic acid-water system had very similar properties to sec. butanol-formic acid-water system.

Equivalent chromatographic patterns were obtained by the analysis of the corms of both *Eminium spiculatum* and *Arisarum vulgare*. The following amino acids were identified: aspartic, glutamic, cystine, lysine, histidine, arginine, glycine, threonine, proline, tyrosine, valine, methionine, phenylalanine, isoleucine, leucine and tryptophan.

Alkaloids

Alkaloids of Eminium spiculatum

The alkaloids were extracted from the defatted powdered corms (13 kg) with ethanol. The ethanol-free extract was treated with aqueous hydrochloric acid (pH 2) and extracted with

chloroform (Fraction I). The acidic aqueous solution was rendered alkaline with ammonium hydroxide (pH 8) and extracted with chloroform till exhaustion (Fraction II). The mother liquor, after the chloroform extraction, still responding to alkaloidal tests was treated with ammonium reineckate to precipitate the alkaloids present (Fraction III).

Fraction I

The brown viscous residue (2.6 gm) was subjected to column chromatography, using aluminium oxide (Brockmann). Elution with benzene-chloroform (50 : 50) gave non-alkaloidal substance which melted at 137–140° C. On the other hand chloroform-methanol (95 : 5) eluted 2 bases (R_F 0.56 and 0.61) as shown by TLC using chloroform-methanol (85 : 15) as developing solvent and sprayed with modified Dragendorff's reagent. Trials to crystallise out any of these components were unsuccessful.

Fraction II

The solvent-free alkaloidal residue (1.6 gm) was subjected to ether fractionation. The ether-soluble (IIa) and the ether-insoluble (IIb) fractions were about 1.3 and 0.3 gm respectively.

Ether-Soluble Fraction (IIa)

Both paper (Whatmann No. 1, butanol-acetic acid-water 4 : 1 : 5) and TLC (silica gel G and alumina G, butanol/acetic acid/water 5 : 1 : 2, chloroform-methanol 50 : 1; chloroform-methanol 50 : 2.5) techniques revealed the presence of one alkaloid spot (modified Dragendorff, Block et al., 1958). The crude alkaloidal residue was purified by passing through a column of activitated silica gel. Elution with chloroform-methanol (98 : 2) gave "Base A".

Base "A" (ethanol), m. p. 90–94° C exhibited λmax (ethanol) at 270 mμ. The infra-red absorption spectrum showed an NH_2 absorption band at 2.8–3.15 μ, C-H at 3.35 μ and a ketonic band at 6.37 μ. The tetraphenylboron derivative (Sinsheimer and Smith, 1963), melted at 110–115° C. Trials to prepare other crystalline derivatives were unsuccessfull.

Ether-Insoluble Fraction (IIb)

TLC using silica gel G or alumina G and developing with the above solvent mixtures showed the presence of at least two alkaloids (R_F were 0.74 and 0.94, using alumina G and developing with chloroform-methanol 5 : 1). The alkaloidal residue was chromatographed on a column of neutral alumina. Benzene eluted a non-alkaloidal substance, which melted at 239–241° C, while chloroform eluted two alkaloids in traces. Trials to separate them were unsuccessful. Elution with chloroform-methanol (95 : 5) gave "Base B".

Base "B" (chloroform-methanol), m. p. 210–213° C, exhibited λmax (ethanol) at 205 and 275 mμ. The infra red spectrum showed a broad absorption band at 2.93–3.20 μ indicative of N-H grouping; at 3.35 indicative of C-H; at 6.2 indicative of C = C grouping, in addition to CH_2 bands at 6.9 and 7.2 μ.

Analysis calculated	for $C_{20}H_{35}O_2N$	C, 74.76; H, 10.90; N, 4.35;
	found	C, 74.48; H, 10.56; N, 4.19.

Fraction III

The alkaline mother liquor was acidified to pH 4 with 10% HCl, and the remaining bases were precipitated with ammonium reineckate following the modified procedure of Panous (1949). Purification of the crude free base by passing through a column of aluminium oxide and elution with acetone gave "Base C".

Base "C"; picrate, m. p. 271–274° C (decomp.)

Analysis calculated for $C_6H_{12}NO.C_6H_3N_3O_7$, C, 41.98; H, 4.36; N, 16.32;
found C, 42.08; H, 4.53; N, 17.00.

Base "C" reineckate, m. p. 275–278° C.

Analysis calculated for $C_6H_{12}NO.C_4H_{10}N_7S_4Cr$,
C, 26.6; H, 4.88; N, 24.88; S, 28.44;
found C, 25.82; H, 4.64; N, 23.05; S, 30.39.

The hydrochloride derivative was prepared according to the method used by Kapfhamer (1930) and Hogg et al. (1961), and further more purified by preparative TLC using silica gel G. It exhibited maximal absorption at 204 and 285 mμ.

Alkaloids of Arisarum vulgare

The fractionation of the total alcoholic basic extract, was carried out according to the procedure adopted in *Eminium spiculatum*. All the fractions were chromatographed on alumina. About 6 alkaloids could be detected by TLC, but non could be isolated in a pure form. However, the major alkaloid was retained in the aqueous solution and was obtained by precipitation as reineckate. The reineckate melted at 272–274° C.

Analysis calculated for $C_6H_{12}NO.C_4H_{10}N_7S_4Cr$,
C, 26.60; H, 4.80; N, 24.90; S, 28.44;
found C, 25.85; H, 4.50; N, 23.10; S, 30.30.

Discussion

The study of the free carbohydrates, by paper chromatographic analysis, gave similar qualitative picture in both *Eminium spiculatum* and *Arisarum vulgare*, revealing the presence of raffinose, sucrose, glucose and fructose. However, the sugar components of the mucilage hydrolysates, showed certain differences. The mucilage of *Eminium* was found to be a mixture of D-galacturonic acid, D-galactose, D-glucose, D-mannose and L-rhamnose, while that of *Arisarum* composed of D-glucuronic acid, D-galactose, D-mannose, L-rhamnose and D-glucuronolactone.

The organic acids of the corms, prepared by the lead method, as tested by TLC and paper chromatographic techniques revealed the presence of citric, tartaric and succinic in *Eminium* and citric and tartaric in *Arisarum*. The absence of oxalic acid was proved in both species.

The unsaturated fatty acids were oleic, linoleic, linolenic, and conjugated dienes in oils of both *Eminium* and *Arisarum*. The saturated acids were myristic, palmitic, stearic, arachidic, behenic and probably lignoceric in *Arisarum* and only palmitic, behenic and probably lignoceric in *Eminium*.

The methodology involved in the fractionation of the unsaponifiable matter of the studied steroids disclosed interesting findings about the validity of the different physicochemical techniques in the separation and identification of the closly related

sterol mixtures, a problem considered to be one of the most intericate in phytochemical investigations. The physical fingerprints of the isolated sterols viz. TLC, U.V. and I.R. analysis, as well as the constant melting ranges of the sterols themselves or of their prepared derivatives, were unable to uncover their complexity. The application of gas-liquid chromatography threw more light on the nature as well as the relative amounts of the individual components of the sterol mixtures. The concrete desicive conclusion about the qualitative and quantitative determination of the individual components of the sterol mixtures was achieved by the application of molecular mass spectrometry. The qualitative composition of the sterol fractions in both *Eminium* and *Arisarum* were the same viz. β-sitosterol, stigmasterol, campesterol and dehydrocampesterol.

An interesting example about the deceptive intricate complex mixture of steroid fractions isolated from plants was encountered in the isolation of an apparently pure substance, m. p. 235–240° C (Liebermann-Burchardt positive) during the hunt for triterpene acid in *Eminium spiculatum*. The substance, after tedious solvent fractionation procedure for its isolation and purification was proved to be composed of at least 7 different components of which 3 are known sterols, applying the molecular mass spectroscopy.

The amino acids of the two species were alike as proved by paper chromatography.

The review of literature uncovers the relatively little attention given to the alkaloids of the *Araceae* plants. The majority of the isolated alkaloids, so far, are of unknown structure, while the few reported are of simple nature viz. coniine and 5-hydroxytryptamine. The family, however, is characterised by the ample presence of a variety of volatile amines e.g. ethylamine, isobutylamine, tertiary methylamine and isoamylamine.

The alkaline steam distillate, tested by paper chromatography, (Table 7), of both species revealed 8 spots in *Eminium*, of which one was a tertiary base (Dragendorff) and 7 spots in *Arisarum*, of which one was of tertiary nature of the same R_F (0.1) as that of *Eminium*.

The study of the alkaloids of *Eminium spiculatum* resulted in the isolation of 3 main fractions. The first was extractable by chloroform from acid medium and presumably contained the neutral bases. This fraction revealed 2 minor components which could not be obtained in any crystallisable form. The second fraction was extractable by chloroform from alkaline medium and was further separated into ether-soluble and ether-insoluble fractions. Base "A" and Base "B" were isolated from the ether-soluble and the ether-insoluble fractions respectively. The third fraction, which contained the major alkaloid component of the plant was the water soluble. This revealed one alkaloid "Base C", which could be obtained in a preparative scale by the reineckate precipitation. Certain characters of the bases viz., m.p., U.V., I.R., and preparation of derivatives were reported.

Table 7

R_F of the volatile components, obtained by the alkaline steam distillation of *Eminium spiculatum* and *Arisarum vulgare*

No. of spots	R_F		Colour	
	Eminium spiculatum	*Arisarum vulgare*	Ninhydrin	Dragendorff
1	0.01	0.01	Violet	–
2	0.04	–	Yellow	–
3	0.10	0.10	Yellow	Yellow
4	0.23	0.22	Yellow	–
5	0.27	0.26	Yellow	–
6	–	0.35	Yellow	–
7	0.41	–	Yellow	–
8	0.51	0.50	Yellow	–
9	0.65	0.65	Yellow	–

Solvent: Butanol-acetic acid-water (50 : 1 : 49).

The fractionation of the total alkaloids of *Arisarum vulgare* showed that the chloroform (acidic) fraction contained 6 minor components, while the chloroform (alkaline) fraction contained 7 minor components. The major component was retained in the water soluble fraction, and was also prepared by the reineckate precipitation.

On trying 19 different solvent systems for the separation of alkaloids of both *Eminium* and *Arisarum* on TLC, it was noticed that those systems which afforded good separation in one plant, were unsuccessful with the other plant and vica versa. In those few trials where a fairly good separation was possible for the basic fractions in both plant species, markedly different TLC patterns were obtained in each case. The total alkaloidal spots (revealed by spraying the chromatoplates with modified Dragendorff's reagent) were 7 and 14 for *Eminium spiculatum* and *Arisarum vulgare* respectively.

Coniine, which was previously reported to occur in certain species of Araceae, could not be identified in both species. The possibility that the water-soluble base "C", isolated in the form of reineckate from *Eminium spiculatum* might be choline was chalenged by the mixed m.p. determination of the authentic sample of choline reineckate (m.p. 284–286° C). Admixing choline reineckate with authentic "Base C" reineckate caused marked depression. Moreover, the hydrochloride of Base "C" showed absorption peaks in U.V. region at 204 and 285 mμ. However, the complete exclusion of the presence of choline lacked concrete evidence since its concomitant presence with "Base C" should be expected if the similarity of the R_F, characteristic stain with Dragendorff's reagent and hygroscopicity of its hydrochloride are to be considered.

Summary

A phytochemical investigation of the carbohydrates, lipids, organic acids, proteins and alkaloids of *Eminium spiculatum* and *Arisarum vulgare* was undertaken.

The study of the carbohydrates revealed the presence of glucose, fructose, sucrose and raffinose in both species.

The mucilage, isolated from the corms of *Eminium,* was found to be composed of D-galacturonic acid, D-galactose, D-glucose, D-mannose and L-rhamnose, while that of *Arisarum* was composed of D-glucuronic acid, D-galactose, D-mannose, L-rhamnose and D-glucuronolactone.

The organic acids of *Eminium* were found to be succinic, citric and tartaric, while that of *Arisarum* were citric and tartaric acids.

The saturated and unsaturated fatty acids of the oils were determined. The study of the unsaponifiable matter revealed the presence of β-sitosterol, stigmasterol, campesterol and dehydro-campesterol, in addition to a saturated aliphatic hydrocarbon and a saturated aliphatic alcohol, in both species.

The amino acids components of the two species were identified by paper chromatography.

Using solvent fractionation and chromatographic techniques, Base "A", m.p. 90–94° C, Base "B", $C_{20}H_{35}O_2N$, m.p. 210–213° C, Base "C", $C_6H_{12}ON$, picrate, m.p. 271–274° C, reineckate, m.p. 275–278° C, were isolated from *Eminium spiculatum.* From *Arisarum vulgare,* only one water-soluble base, was obtained on a preparative scale by the reineckate salt. Both species contain few minor constituents and were free from coniine.

The study of the volatile amines of *Eminium spiculatum* and *Arisarum vulgare,* as detected by paper chromatography, revealed 8 and 7 spots respectively. Only one tertiary base was detected in each species.

Zusammenfassung

Es wird über die Ergebnisse einer phytochemischen Untersuchung der Sprosse von *Eminium spiculatum* und *Arisarum vulgare* berichtet.

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