The Significance of Some Pharmacognostical Studies and the Anthraquinone Content in the Identification of Egyptian Cassia Species.

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Abstract : The presence or absence of the different anthraquinone derivatives in Cassia species are recorded. The derivatives are studied by paper and thin-layer chromatography before and after hydrolysis and oxidative hydrolysis. Chromatographic and spectral data are recorded as well as their quantities. Some morphological aspects and numerical values are mentioned.

Introduction

Anthraquinones occur widely in Cassia plants, not only in the pods but also in the leaves and roots and they are characteristic of the genus (1). From Cassia acutifolia Del. and C. angustifolia Vahl, sennoside A, B, C and D were isolated (2-6), in addition to rhein, aloe-emodin, chrysophanol and other derivatives (7-11,23). Previous work on C. fistula L. showed that the pulp contain rhein and fistulic acid (12,13), the leaf sennosides in the flowering season (14) and the flowers rhein and fistulin (12). C. obovata Collad leaves were found to contain rhein , aloe-emodin, sennidins, chrysophanol, and sennosides A and B (15). Dianthraquinones were isolated from C. siamea Lam. bark (16 - 18) .

The genus Cassia contains about 400 species and there is no recent report about their systematic study, although Hegnauer (19) referred to the distribution of anthraquinones in plant kingdon . From these species seventeen are grown and cultivated in Egypt and are collected for the performance of this study. The leaves were chosen as a representative organ and the components were derived to the free aglycones. However, the original form whether C- or O-glycoside is mentioned. The identification of the components was performed chromatographically and by UV measurements .

Experimental

Material: Samples of the leaves of Cassia speies were collected from non-flowering, flowering and early fruiting plants cultivated in the experimental stations of Horticulture and Ornamental plants at Alexandria, Cairo, Aswan and the experimental station of medicinal plants, Assiut University. The leaves were dried at $60°_C$ and powdered.

Method: 5 g powdered leaves of each species were extracted with hot methanol(70%) and the obtained extracts were employed for TLC examination. An aliquot was extracted, after concentration with ether and the mother liquor was hydrolysed with 2N sulphuric acid and extracted with ether again. Both of the etherial extract, in each case, was fractionared into carboxylic and non-carboxylic components by the use of aqueous sodium bicarbonate. Oxidative hydrolysis was carried out adopting the method described by Gyanchandani et al (I969)) (2I). The anthraquinone derivatives were estimated according to the method of Rowson et al (I959)(22). For identification of the presence or absence of anthraquinones, Borntrager reaction was employed.

TLC: silicagel plates and the solvent systems: Ethylacetate-methanol- water (I00:I6.5:I3.5); n-propanol-ethylacetae-water(4:4:3) benzene-ethylacetate(4:I); and alkaline silicagel(N/I0 NaOH) using as solvent system: benzene-ethylacetate-acetic acid(75:20:5).

PC: Whatman no. I and n-butanol-acetic acid-water(4:I:5); petroleum - ether saturated with 97% methanol. After development the obtained spots were scraped or cut off, eluted and used for UV measurements.

Results

From the seventeen species examined, six were found to contain anthraquinones (table 1). The derivatives occur in both glycosidic and free forms except in C. simea Lam. which contain only free forms. With the exception of C. javanica , chrysophanol was present in all species. C. fistula leaves showed the presence of traces of glycosides based on rhein at the beginning of the flowering stage and samples collected at other seasons failed to give a positive test.

The closely similar morphologically C. siamea and C. renigera could be distinguished by the presence of anthraquinones only in the former. C, javanica is distinguished from C. siebriana by the presence of C-glycoside based on rhein in the leaves of C. javanica. The absence of anthraquinones is obseved in C. nodosa however, Rizvi et al (20) isolated a glycoside nodoside from the flowers.

Sennosides and sennidins are detected in the leaves of C. acutifolia and C. angustifolia and traces in C. fistula leaves. Table 2 summerises the distribution of anthraquinones whether free or combined in the species examined, omitting C. fistula as the leaves contain traces of these constituents.

The chemical analysis of the leaves showed that those of C.acutifolia contain the highest percentage of anthraquinones:2.1 to 2.88% w/w followed by C. angustifolia 2.09 2.73 %w/w. C. obovata was found to contain 0.84 to 0.89% w/w and C.siamea 0.14 to 0.25 % w/w. The lowest contents were those of C. javanica 0.05 to 0.057 % w/w and C. didymobotrya 0.013 to 0.014 % w/w.

Table 3 presents the measurements of some morphological aspects and numerical values of the examined leaves which give an additve charecter for each species beside the chemical screening.

Table 1. The presence or absence of anthraquinones in the leaves of the different Cassia species.

C. acutifolia Del.	+	C. limensis Lam.	_
C. angustifolia Vahl	+	C. nodosa Buch-Ham.	_
C. annuora L.	_	C. obovata Collad.	+
C. bicapsularis L.	_	C. obtusifolia L.	_
C. didymobotrya Fres.	+	C. pendula Willd.	_
C. glauca Lam.	_	C. renigera Bois.	_
C. glomifera L.	_	C. siebriana Dc.	+
C. javanica L.	+		

+ = present , _ = absent.

Table 2. The distribution of the different anthraquinones in the examined leaves.

	C. acutif.	C. angust.	C. didym.	C. fruv.	C. obov.	C. [illegible]
A. Before hydrolysis:						
Chrysophanol	+	+	+	–	+	+
Emodin	+	+	–	–	–	–
Aloe-emodin	+	+	+	–	+	–
Rhein	+	+	–	traces	+	–
Physcion	+	+	–	–	–	+
Sennidins	+	+	–	–	+	–
Sennosides	+	+	–	–	+	–
Others	non-rhein glycosides		O-glycoside of rhein-like subst.	C-glycosides of rhein	–	cassiamin A
B. After acid hydrolysis:						
Chrysophanol	±	±	±	–	±	±
Emodin	±	±	–	–	±	–
Aloe-emodin	+	+	+	–	+	–
Rhein	+	+	–	–	+	–
Sennidins	+	+	–	–	+	–
Others	–	–	rhein-like subst.	–	–	traces of cassiamin A
C. After oxidative hydrolysis:						
Chrysophanol	–	–	–	–	–	traces
Emodin	–	–	–	–	–	–
Aloe-emodin	+	+	–	–	±	–
Rhein	+	+	–	+	+	–
sennidins	traces	traces	–	–	–	–
Others	–	–	rhein-like subst.	–	–	–

+ = present , – = absent , ± = detected in traces.

Table 3. Some morphological charecters and numerical values of the examined leaves.

	C.acutif.	C.angust.	C.didym.	C.javan.	C.obov.	C.siam.
No.of pairs of leaflets	4-7	7-9	16-22	9-19	5-6	8-10
Size of leaflets:						
length,mm	230-580	250-550	300-400	200-240	80-300	350-800
Width,mm	5-15	5-13	10-20	9-15	7-13	15-25
Stipule	present 1-2 mm	present 1-2 mm	present ovate	-	present ovate	-
Apex	acute spiny	acute	acute spiny	mucronate	round spiny	round spiny
Stomatal-index	11.5-12.1 -13.0	17.2-18.5 -20.0	20.2-21.1 -22.2	8.8-9.1 -10.2	11-14.5- -16.3	13.0-14.1 -15.2
Palisade ratio	4.5-9.5- 18 and 3.5-7.1- 14.5	4.1-7.5- 12 and 2.5-5.2- 12.0	2.1-3.1- 4.2	2.8-4.1- 5.2	10.5-10.7 -11.1 and 6.3-6.5- 7.1	4.9-5.8- 7.0
Vein islet number	25.2-29.6	19.5-22.4	30-32	3.0-4.0	12.0-13.2	4.1-6.3

Table 4. Physical constants of the anthraquinones present in Cassia species.

Compound	m.p.	$R_f \times 100$	UV λ_{max} in methanol
Aloe-emodin	224	45[a]	225,256,278,287,425nm.
Chrysophanol	197-8	81[a]	225,257,279,288,433nm.
Emodin	256-7	62[a]	223,253,266,289,438nm.
Physcion	206	80[a]	225,255,266,288,436nm.
Rhein	330	43[a]	229,259,431nm.
Sennidin A	280-7	46[b]	261,335,366nm.
Sennidin B	230-5	33[b]	262,310,374nm.
Sennoside A	220-30	26[b]	269,341,370nm.
Sennoside B	180-6	13[b]	266,297,360nm.

[a] alkaline silicagel and benzene – ethylacetate – acetic acid (75 : 20 : 5)

[b] silicagel and n-propanol – ethylacetate – water (4 : 4 : 3)

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اهمية بعض الدراسات العقاقيرية والمحتوى الانثراكينونى فى تعريف انواع نبات الكاسيا المصرية

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تم فى هذا البحث دراسة اوراق ١٧ نبات من جنس الكاسيا المنتشرة فى جمهورية مصر العربية ولقد اثبت ان ٦ فقط من هذا النبات تحتوى على الانثراكينون سواء فى حالة حره او كجلوكوسيد وقد تم تحويل الجلوكوسيدات المتعرف عليها الى انثراكينون حر عن طريق التحليل المائى او التحليل فى وجود عوامل مؤكسده ٠

وقد درست كروماتوجرافيا بواسطة الطبقة الرقيقة والورق ٠

ولقد تم فصل واستخلاص المواد الانثراكينونية الممثلة الموجودة فى اوراق هذه النباتات بواسطة الكروماتوجرافيا وتم التعيين الكمى لها بواسطة الطرق الفيزيائية والاسبكتروسكوبية ٠