

THE CHROMATOGRAPHIC STUDIES FOR DIFFERENTIAL IDENTIFICATION OF HOMOEOPATHIC DRUGS OF ASCLEPIAS SPECIES

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INTRODUCTION

The genus *Asclepias* belongs to the family *Asclepiadaceae*. They are perennial herbs generally 0.6m. to 1.5m. high. They are mostly found in America, and in some parts of Africa. A few species are also found in India. Four species of *Asclepias*, viz. *A. incarnata*, *A. syriaca*, *A. tuberosa* and *A. vincetoxicum* are used in Homoeopathy. *A. incarnata* is used in chronic gastric diseases of a mucus character, catarrhal discharges in leucorrhoea, dropsy and diabetes. *A. syriaca* is used in bronchitis, dropsy, catarrhal fever, hay fever, headache, indigestion, influenza, uterine pain etc. *A. tuberosa* is used in asthma, bilious fever, bronchitis, catarrhal fever, colic, cough, diarrhoea, dysentery, ophthalmia etc. and *A. vincetoxicum* is used in dropsy, diabetes, great thirst, and profuse urination.

Literature survey revealed the presence of asclepiadin (an emetic principle) an alkaloid, two acrid resins, volatile oil, fixed oil, albumin, starch, pectin and glucose in *A. incarnata*; nicotine in *A. syriaca*; sterol, an oil and three unidentified crystalline fractions in *A. tuberosa*. No chemical work have been reported on *A. vincetoxicum*. *A. curassavica* a drug found in India but not used was reported to contain asclepagonin $C_{23}H_{32}O_6$, clepogonin $C_{23}H_{32}O_6$, curassavogenin $C_{23}H_{32}O_7$, asaurogenin $C_{23}H_{32}O_7$ and calotropagonin.

MATERIALS AND METHODS

For percolation the powdered plant material was initially soaked in 70% alcohol for *A. incarnata*, 75% for *A. syriaca* and 60% for *A. tuberosa* and *A. vincetoxicum*. The choice of percentage of alcohol was made in consonance with the prescribed approximate requirements for, preparation of the homoeopathic mother tinctures. After 24 hrs. of percolation, 100 ml extract of each plant material was evaporated on waterbath to remove alcohol. The remaining aqueous portion was divided into two equal parts. One part (in each case) was extracted thrice with 25 ml (10+10+5) of chloroform and aqueous layers (A, A₁, A₂, A₃ & A₄) and chloroform layers (B, B₁, B₂, B₃ & B₄) were separated. Second aqueous parts were made alkaline with Ammonium hydroxide solution and then extracted with 25ml of chloroform thrice (10+10+5) and the chloroform layers (C₁, C₂, C₃ &

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C.) were separated and examined for the presence of alkaloids. The T.L.C. plates of aqueous layers (A series) and of chloroform layers (B & C series) were chromatographed over silica gel 'G' plates 200 μ 20 \times 5cm, BDH, using ethyl-acetate: butanone: formic acid: water (5:2:2:1 v/v) for aqueous layers and benzene: methanol (9:1 v/v) for chloroform layers respectively. T.L.C. plates of aqueous layers were sprayed with aniline-phosphoric acid and developed plates of chloroform layers (B series) sprayed with antimony trichloride and the plates of C series were sprayed with Dragendroff's reagent. The plates were also examined under U.V. light before spraying.

RESULTS & DISCUSSION

The chromatographic study of aqueous layers (A series) showed the presence of one spot at Rf. 0.71 in *A. incarnata* and at Rf. 0.75 in *A. syriaca* but no spot appeared in *A. tuberosa* and *A. vincetoxicum*.

The chloroform layer (B series) developed in benzene-methanol (9:1 v/v) showed seven spots in *A. incarnata* at Rf. 0.40, 0.47, 0.55, 0.74, 0.86, 0.94 & 0.99; three spots in *A. tuberosa* at Rf. 0.33, 0.94 & 0.99; eight spots in *A. vincetoxicum* at 0.33, 0.43, 0.49, 0.55, 0.66, 0.74, 0.80 & 0.94 and four spots in *A. syriaca* at Rf. 0.28, 0.40 & 0.99 under ultra violet light.

On spraying the same T.L.C. plates with antimony trichloride solution, ten spots appeared in case of *A. incarnata* at Rf. 0.30, 0.40, 0.43, 0.49, 0.63, 0.68, 0.76, 0.85, 0.90 & 0.95; six spots in *A. syriaca* at Rf. 0.41, 0.63, 0.68, 0.72, 0.90 & 0.95; six spots in *A. tuberosa* at Rf. 0.41, 0.49, 0.55, 0.63, 0.70 and 0.85 and seven spots in *A. vincetoxicum* at Rf. 0.18, 0.23, 0.30, 0.36, 0.41, 0.49 & 0.53. All spots were violet blue in colour.

The T.L.C. plate of chloroform layers of C series when developed with benzene-methanol (90:10 v/v) gave three-blue fluorescence spots in *A. incarnata* at Rf. 0.18, 0.22 & 0.94; four spots in *A. syriaca* at Rf. 0.18, 0.22, 0.69 & 0.94; and five spots in *A. tuberosa* at Rf. 0.14, 0.18, 0.22, 0.31 & 0.94; and seven spots in *A. vincetoxicum* at Rf. 0.18, 0.22, 0.26, 0.29, 0.59, 0.90 and 0.94. All spots were giving blue fluorescence except spot at 0.90 of *A. vincetoxicum*, which was pink in colour.

The above plate on spraying with Dragendroff's reagent four spots in *A. incarnata* at 0.10, 0.18, 0.31 & 0.94; four spots in *A. syriaca* at 0.03, 0.22, 0.131 & 0.94; three spots in *A. tuberosa* at Rf. 0.03, 0.10 & 0.94; four spots in *A. vincetoxicum* at Rf. 0.07, 0.10, 0.31 & 0.94.

The alcoholic extracts were diluted (100 times with water) and screened for ultra violet spectrum. All the four species of *Asclepias* gave a peak between 245-280 nm but the chromatographic results show that the four species of *Asclepias* give different results and it is very convenient to differentiate them from one another chromatographically.

CHROMATOGRAPHIC RESULTS OF ASCLEPIAS SPECIES

	Detection	<i>A. incarnata</i>	<i>A. tuberosa</i>	<i>A. vincetoxicum</i>	<i>A. syriaca</i>
Aqueous layer (A)					
Solvent System—ethyl acetate: butanone: formic acid: water 5 : 2 : 2 : 1 (v/v)	Sprayed with aniline phosphoric acid reagent	0.71 (brown spot)	Nil	Nil	0.75 (brown spot)
Chloroform layer (B)					
Solvent System—Benzene methanol 90 : 10 (v/v)	U. V. light all gives blue fluorescence	0.40, 0.47, 0.55, 0.74, 0.86, 0.94, 0.99.	0.33, 0.94, 0.99.	0.33, 0.43, 0.49, 0.55, 0.66, 0.74, 0.80, 0.95.	0.28, 0.40, 0.924, 0.99.
	Sprayed with SbCl ₅ reagent. All gives violet blue colour	0.30, 0.40, 0.43, 0.49, 0.63, 0.68, 0.76, 0.85, 0.90, 0.95.	0.41, 0.49, 0.55, 0.63, 0.70, 0.85.	0.18, 0.23, 0.30, 0.36, 0.41, 0.49, 0.53.	0.41, 0.63, 0.68, 0.72, 0.90, 0.95.
Chloroform layer (C)					
Solvent System—Benzene methanol (90 : 10 v/v)	U. V. light all blue	0.18, 0.22, 0.94.	0.14, 0.18, 0.22, 0.31, 0.94.	0.18, 0.22, 0.26, 0.29, 0.59, 0.90, (pink) 0.94.	0.18, 0.22, 0.69, 0.94.
	Sprayed with dragendroff's reagent (brown spot)	0.10, 0.18, 0.31, 0.94.	0.03, 0.10, 0.94.	0.07, 0.10, 0.31, 0.94.	0.03, 0.22, 0.31, 0.94.

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