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DIABOLINE FROM STRYCHNOS POTATORUM

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Plant. Strychnos potatorum L.f., a muchbranched small- to medium-sized tree of Central and South India, Sri Lanka and Burma, and also East and northern South Africa [1]. Sources. See ref. 2 and Table 1. Uses. In India the various parts of the plant have been widely applied in both the Ayurvedic and Yunani systems of medicine and the seeds (Sanskrit kataka) are particularly well known on account of their use in clearing muddy water—a use which has been in vogue since the time of Charaka and Sushruta [3], but which is not known from Africa [4].

Previous work. On the basis of microchemical studies Rosoll [5] considered strychnine to be present in the seeds. Other investigators criticized his findings [6,7] and interpreted their own results as indicating the absence of alkaloids [6,8]. In contrast, Dymock et al. [9] obtained partially purified extracts which gave positive tests for brucine, while Mathis and Duquénois [10] extracted 0.068–0.075% total alkaloid, comprising three components, which afforded negative tests for strychnine.

Present work. The seeds yielded 0.3% Et₂O-CHCl₃(2:1)-soluble alkaloids. TLC and GLC indicated that diaboline was the major component and that it was accompanied by an acetyldiaboline and by traces of other bases. Diaboline was isolated via its hydrochloride and its identity established by

chromatographic and spectroscopic studies. Preparative TLC of the total bases gave a mixed fraction whose MS confirmed the presence of diaboline and the acetyldiaboline. The leaves gave 0.096% and the trunk bark 0.091% total alkaloids. In both these mixtures, according to TLC and GLC, diaboline was again the major base; the occurrence of the acetyldiaboline and traces of other alkaloids was also noted.

For comparison, the alkaloid extracts from a number of herbarium samples of both Indian and African origin were also examined by TLC and GLC (see Table 1). In all cases, diaboline could be identified as the major alkaloid.

Diaboline has been obtained from American, African, and Asian species of *Strychnos* [11] and although recent long-term feeding experiments with *S. nux-vomica* L. have shown that it is not itself on the biosynthetic pathway to strychnine and related alkaloids, the corresponding deacetyl compound—Wieland-Gumlich aldehyde—is efficiently incorporated into strychnine [12].

EXPERIMENTAL

Methods. The systems and sprays for TLC and preparative TLC [13] and the system for GLC [14] have been previously described.

Extraction of the alkaloids. Air-dried material was shaken with 4 vol. Et₂O-CHCl₃(2:1) and left for 10 min; 0.4 vol. dil. NH₄OH was added and the whole shaken mechanically for 6 hr, after which it was extracted (Soxhlet) for 2 hr. The organic phase was removed and shaken with 5 × 0.8 vol. N H₂SO₄. After basification (conc. NH₄OH) the combined extracts were extracted exhaustively with CHCl₃. The combined CHCl₃ extracts were washed (H₂O), dried (Na₂SO₄),

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Collector's no.	Plant part	Locality and date	Crude alkaloid (%) [14]	Alkaloid test [14]
Leippert 5968	Leaves	Tanzania, Northern	0.14	+
		Prov.; 22 June 1965		
Newman 22	Leaves	Tanzania, Central	0.63	++++
		Prov.; 1965		
Greenway, Savidge and Kanuri 14084	Leaves	Tanzania, Southern	0.22	+ +
		Highlands Prov.;		
		11 March 1970		
Greenway, Savidge and Kanuri 14084	Fruits	Tanzania, Southern	0.11	++++
		Highlands Prov.;		
		11 March 1970		
ST 10/29/K1	Seeds	India: Balfour,	0.06	++++
		19th C		
ST 10/29/K2	Seeds	India; commercial,	0.08	++++
		20th C		

Table 1. Herbarium samples of Strychnos potatorum L.f.

and taken to dryness. TLC and GLC indicated that seed, leaves and bark extracts all had diaboline as the major base; small amounts of an acetyldiaboline and other alkaloids also appeared to be present.

Isolation and identification of diaboline. A soln of the crude seed bases in EtOH was acidified (EtOH–HCl) and fractionally pptd with EtOAc giving small pellets of diaboline HCl-ide (IR). The identity of the free base as diaboline was established by TLC, GLC, IR, NMR, and MS in comparison with the authentic alkaloid. Preparative TLC of the seed bases gave a main band and the residue from it proved to be still a mixture, which TLC and MS showed to consist of diaboline (M⁺ 352) and the acetyldiaboline (M⁺ 394).

Extraction of the alkaloids from the herbarium samples. The samples extracted are listed in Table 1. The method of extraction used has been described previously [14]. TLC and GLC showed that diaboline was the principal alkaloid in all the extracts examined.

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