

POSSIBLE CHEMOTAXONOMIC SIGNIFICANCE OF THE OCCURRENCE OF *CIS*-4-HYDROXY-L-PROLINE IN SANTALACEAE

RAMADASAN KUTTAN, KALATHOOR S. V. PATTABHIRAMAN and
AMURTUR N. RADHAKRISHNAN

Wellcome Research Unit, Christian Medical College Hospital, Vellore-632004, Tamil Nadu, India

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Key Word Index—*Santalum*; *Osyris*; *Scleropyrum*; *Thesium*; Santalaceae; *cis*-4-hydroxy-L-proline; chemotaxonomy.

Abstract—*cis*-4-Hydroxy-L-proline was detected in three genera (four species) of Santalaceae.

THE UBIQUITOUS presence of *trans*-4-hydroxy-L-proline (*trans*-Hyp) in the bound form as a constituent of plant cell-wall material has been well documented.^{1,2} *cis*-4-Hydroxy-L-proline (*cis*-Hyp), however, has a restricted distribution in the plant kingdom, being first reported in the bound state in *Amanita phalloides*³ and in the free state in the leaves of *Santalum album* L.⁴ *cis*-Hyp was reported in a few other species of *Santalum*,^{5,6} and a study of its distribution⁵ and on its biosynthesis⁷ in the plant is available. This communication is an extension and consolidation of the data available on the occurrence of *cis*-Hyp as a free amino acid in members of Santalaceae available in India and in a few that have been procured from elsewhere. The data strongly suggests that the presence of this imino acid may be useful as a chemotaxonomic index for Santalaceae.

From the data summarized in Table 1, it is seen that *cis*-Hyp is present in all specimens examined, although there is a wide variation in its concentration. Quite obviously *Santalum* is very rich in *cis*-Hyp and the amount may be as much as 10% of the dry weight of the seed coat of the fruits.⁵ It is of interest to note that *cis*-Hyp was detectable in a specimen of *Scleropyrum* leaves preserved in the dry state for over 10 yr.

Though the present study is somewhat limited in scope, the presence of *cis*-4-hydroxy-L-proline in the Santalaceae may serve as a useful index in chemotaxonomy. It is hoped that this report will generate further interest in studying other members of the large Santalaceae family not seen in India.

EXPERIMENTAL

The plant specimens were collected from different parts of India and the rarer ones by experts of the Botanical Survey of India. Three specimens were earlier obtained from Dr. H. S. McKee.⁵ The following methods were used during this work: powdering, extraction and determination of Hyp by a modification of the Neuman and

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⁶ MCKEE, H. S. and URBACH, G. (1955) *Nature* **175**, 470.

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TABLE I. DISTRIBUTION OF *cis*-4-HYDROXY-L-PROLINE IN SANTALACEAE

Name of plant	Part analyzed	Conc. (g/100 g dry powder)	Ref.
<i>Santalum album</i> L.	Leaf	0.3–2.4	4, 5
	Pericarp	10.6	
	Flowers	0.3	
<i>S. yassi</i> , <i>S. austrocaledonicum</i> , <i>S. obtusifolium</i> , <i>S. murrayanum</i>	Leaf	0.3–2	5, 6
<i>Osyris arborea</i> Wall.	Leaf	0.34	
<i>Scleropyrum wallichianum</i> Arn	Seed	0.12	
<i>Thesium himalense</i> Royle	Leaf	Traces	This study
<i>Thesium wightianum</i> Wall.	Leaf	0.2	
	Leaf and stem	Traces	

Logan⁹ method (Radhakrishnan *et al.*⁵); 2-D PC system;⁸ specific test for HP;¹⁰ separation of *cis*- and *trans*-HP as Cu-salts¹¹ and as free amino acids by the amino acid analyzer.¹² The optical configuration of Hyp was confirmed by treatment with sheep kidney D-amino acid oxidase¹³ both *cis*- and *trans*-D-Hyp being efficient substrates.¹⁴ Failure of formation of pyrrole-2-carboxylic acid was proof for the L-configuration. Hyp used for some of the above work was isolated by a combination of ion-exchange and paper chromatography. The isomer in Santalaceae is *cis*-4-hydroxy-L-proline. When extracts were highly colored as in the case of *Scleropyrum* specimens, they were decolorized with charcoal⁴ and passed through a small Dowex-50 resin column and subsequently eluted with 1 N NH₄OH, which was removed before analysis.

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