

Phytochemical study of South American *Ilex*

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ABSTRACT The structure of five triterpenoid saponins (matesaponins 1-5) isolated from *Ilex paraguariensis* is reported. Structural elucidation was performed using mass spectrometry and NMR techniques.

ABSTRAK Struktur 5 saponina triterpenoid dari *Ilex paraguariensis* dilaporkan. Elusidasi struktur dijayakan dengan menggunakan kaedah RMN dan kaedah spektrometri jisim.

(*Ilex paraguariensis*, Aquifoliaceae, matesaponin, maté)

INTRODUCTION

The genus *Ilex* (Aquifoliaceae) consists of over 400 species, which grow as trees or shrubs. Major centres of distribution are Central and South America and Asia.

In Brazil and Paraguay, *Ilex paraguariensis* is cultivated for the preparation of a mild stimulant beverage (called maté) made by infusion of the dried leaves. However, several indigenous *Ilex* species have been reported as adulteration or substitutes for the genuine maté product. More particularly, *I. argentina*, a species from the subtropical subandean rainforest of Northwestern Argentina and Eastern Bolivia that shows several morphological similarities with *I. Paraguariensis* has been recommended for the maté production [1]. The geographic gap between these two plants, incited us to start our South American *ilex* phytochemical program by the study of the saponin content of these two species. As a preliminary result, we herein present the structure of five saponins isolated from *I. paraguariensis*.

EXPERIMENTAL

The dried leaves of *I. paraguariensis* (200g) were extracted with EtOH-H₂O (4:6). The gum obtained

after evaporation of the solvent was dissolved in H₂O and successively extracted with CHCl₃, EtOAc and *n*-BuOH. The *n*-BuOH fraction was washed with 1% NaOH solution and the residue obtained after evaporation on the *n*-BuOH was repeatedly chromatographed over Si gel (CHCl₃-EtOH-H₂O, 8: 4: 0.5) to give matesaponins (1 - 5).

RESULTS AND DISCUSSION

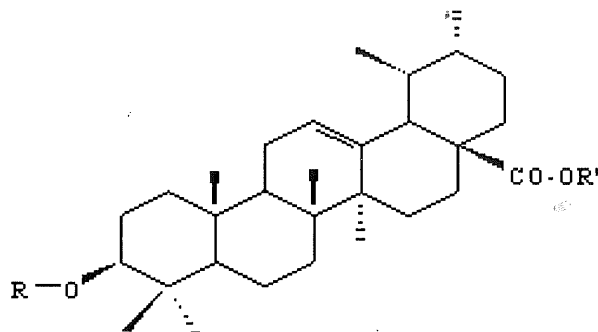
The structure of the five saponins was established from the native derivatives or from their peracetylated counterparts using a combination of mass and NMR spectroscopy techniques [2-4]. The aglycone of the five saponins was shown to be ursolic acid from ¹³C NMR data. The structure of the sugar side chain was deduced using COSY, HOHAHA and ¹³C-¹H correlation spectra (¹J and ³J). Complete ¹³C NMR data of matesaponins 1 -5 are listed in Table 1.

Other minor saponins have also been isolated from *I. paraguariensis*, their aglycone has already been identified as ursolic and oleanolic acid. Their complete structure elucidation will be reported in due time.

Table 1 ¹³C NMR spectral data (δ, ppm) of 1-5₁ (75 MHz)

	1*	2§	3¶	4*	5#
C-1	38.8	40.2	38.8	39.2	39.5
C-2	26.5	27.1	26.1	26.3	27.1
C-3	88.6	90.5	90.0	87.8	89.2
C-4	39.4	40.2	39.1	39.5	39.9
C-5	55.7	57.0	55.8	55.7	56.8
C-6	18.3	19.3	18.4	18.2	19.1
C-7	32.9	33.5	33.2	32.7	34.0
C-8	39.9	40.2	39.7	39.7	39.9
C-9	47.9	48.4	48.3	47.7	48.7
C-10	36.8	37.8	36.9	36.7	37.6
C-11	16.8	24.1	24.3	23.0	24.2
C-12	125.9	127.1	126.1	125.7	128.5
C-13	138.3	139.0	137.0	137.7	139.0
C-14	42.3	43.0	42.3	42.1	43.0
C-15	28.5	29.3	26.1	28.4	29.1
C-16	24.5	24.1	23.5	24.3	25.0
C-17	48.2	48.4	47.8	48.0	48.8
C-18	53.1	54.0	52.8	52.8	53.8
C-19	39.1	40.1	39.3	38.7	40.0
C-20	38.9	40.1	39.1	38.9	39.9
C-21	30.5	31.6	30.8	30.4	31.4
C-22	36.7	37.6	36.2	38.9	37.3
C-23	28.0	28.6	28.0	27.7	28.5
C-24	15.6	16.3	16.6	17.1	16.3
C-25	15.6	14.2	15.7	15.5	17.3
C-26	17.5	17.7	17.2	16.6	17.8
C-27	23.5	24.1	23.4	23.3	23.8
C-28	176.1	177.8	175.2	175.9	177.2
C-29	23.6	17.7	17.4	16.7	17.7
C-30	21.1	21.6	21.3	20.9	21.6
glcI-1	95.5	95.6	91.5	95.3	95.4
glcI-2	73.9	75.2	70.3	74.6	74.1
glcI-3	78.7	77.8	72.1	77.6	77.6
glcI-4	71.0	71.0	68.5	70.6	71.2 ^c
glcI-5	78.9	77.8	71.3	78.3	77.6
glcI-6	62.1	62.3	67.9	69.1	69.4
glcII-1		100.9	104.2	104.4 ^b	
glcII-2		71.2	74.8	74.1	
glcII-3		72.9	77.7	76.1	
glcII-4		69.1	71.0	76.2	
glcII-5		72.9	77.8	77.6	
glcII-6		62.1	62.2	61.5 ^a	
glcIII-1				104.2 ^b	
glcIII-2				74.1	
glcIII-3				77.6	
glcIII-4				70.6 ^c	
glcIII-5				77.6	
glcIII-6				62.0 ^a	
glcIV-1	106.0	101.8	100.6	104.3	104.2 ^b
glcIV-2	75.5	75.2	70.1	74.3	74.6
glcIV-3	78.1	77.8	72.1	78.1	77.6
glcIV-4	71.4	71.4	68.4	71.1	71.2 ^c
glcIV-5	78.4	77.8	71.6	77.8	77.6
glcIV-6	62.5	62.6	61.7	62.1	62.1 ^a
ara-1		107.2	105.2	103.7	104.8
ara-2	71.7	77.8	73.1	78.1	73.5
ara-3	83.9	83.8	76.9	81.7	80.8
ara-4	69.1	69.4	73.1	67.8	68.2
ara-5	66.7	66.6	64.3	64.4	64.9
rha-1		104.6		101.5	101.6
rha-2		72.0		72.0	71.4
rha-3		72.0		72.1	73.5
rha-4		73.7		73.5	74.6
rha-5		70.5		69.6	69.8
rha-6		17.1		18.2	18.2

a, b, c attributions with the same index are interchangeable.
 * Spectrum recorded in C₅D₅N., § Spectrum recorded in CD₃OD, ¶ Peracetylated matesaponin 3, spectrum recorded in CDCl₃,
 # Spectrum recorded in CD₃OD/C₂D₆SO.



- 1: R = β -D-glucose-(1 Φ 3)- α -L-arabinose; R' = β -D-glucose. [R = glcIV-(1 Φ 3)-ara; R' = glcI].
 2: R = β -D-glucose-(1 Φ 3)-[α -L-rhamnose-(1 Φ 2)]- α -L-arabinose; R' = β -D-glucose. [R = GlcIV-(1 Φ 3)-[rha-(1 Φ 2)]-ara; R' = glcI].
 3: R = β -D-glucose-(1 Φ 3)- α -L-arabinose; R' = β -D-glucose-(1 Φ 6)- β -D-glucose. [R = glcIV-(1 Φ 3)-ara; R' = glcII-(1 Φ 6)-glcI].
 4: R = β -D-glucose-(1 Φ 3)-[α -L-rhamnose-(1 Φ 2)]- α -L-arabinose; R' = β -D-glucose-(1 Φ 6)- β -D-glucose. [R = glcIV-(1 Φ 3)-[rha-(1 Φ 2)]-ara; R' = glcII-(1 Φ 6)-glcI].
 5: R = β -D-glucose-(1 Φ 3)-[α -L-rhamnose-(1 Φ 2)]- α -L-arabinose; R' = β -D-glucose-(1 Φ 4)- β -D-glucose-(1 Φ 6)- β -D-glucose. [R = glcIV-(1 Φ 3)-[rha-(1 Φ 2)]-ara; R' = glcIII-(1 Φ 4)-glcII-(1 Φ 6)-glcI]

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