



***Celtis ehrenbergiana* (Celtidaceae): meristematic tissue in the thorns**

[*Celtis ehrenbergiana* (Celtidaceae): tejido meristemático en las espinas]

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Abstract

The goal of this research is to establish the presence of meristematic tissue (buds) in the thorns of "tala" (*Celtis ehrenbergiana*). This species has been mentioned as having medicinal properties, and this tree has high ecological value since it protects the environment. Thorn samples have been processed by standard light microscope techniques. Only one of the two geminate thorns located at the apex of actively growing branches has two buds: one in the apical and the other in a lateral subapical position. It would be interesting to assay these portions which have numerous meristematic cells with division capacity using "in vitro" micropagation technology.

Keywords: anatomy; *Celtis ehrenbergiana*; Celtidaceae; conservation; medicinal plants; thorns.

Resumen

El tala (*Celtis ehrenbergiana*) es un árbol con espinas que posee propiedades medicinales, brinda alimento y refugio a otras especies y protege el ambiente. Se realizó el estudio anatómico de las espinas de tala a fin de establecer la presencia de tejido meristemático (yemas). Las muestras de espinas fueron procesadas empleando técnicas estándares para microscopía óptica. Solamente una de las dos espinas geminadas localizadas en la proximidad del ápice de ramas en activo crecimiento presenta dos yemas: una apical y otra lateral en posición sub-apical. Sería interesante ensayar estas porciones con células meristemáticas en activa división con técnicas de micropagación *in vitro*.

Palabras Clave: *Celtis ehrenbergiana*; Celtidaceae; conservación, espinas; plantas medicinales

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INTRODUCTION

The "tala" is also known by the common names: "churqui tala", "desert hackberry", "huasteco", "ibirá yuazú", "shiny hackberry", "spiny hackberry", "tala amarillo", "tala árbol", "tala blanco", "yoasí", "yoasí-guazú". It has a widespread distribution in the New World, the South of the United States, Mexico, Haiti, Puerto Rico, Brazil, Bolivia, Paraguay, Uruguay and Argentina. This tree has many applications, for example in Argentina, where native people use it for shade and firewood due to its high calorific power; its hard and heavy wood is used to make farm tools. In medicine, leaf infusion with honey is used to cure coughs, colds, headaches, chest pain, and acts as an expectorant, a hepatic protector, a digestive aid and may be used to control cholera (Hieronymus, 1882; Toursarkissian, 1980; Lahitte *et al.*, 1995, 1998; Demaio *et al.*, 2002; Barboza *et al.*, 2006; Peña-Chocarro *et al.*, 2006; Hernández *et al.*, 2009). It has calcifying properties (Carrizo *et al.*, 2005). The decoction of its leaves and branches is drunk in case of poisoning, and the decoction of the bark may be used for appetite stimulation, wound washes and disinfection (Martínez Crovetto, 1981). The root has tinctorial properties, giving a brown color resistant to water and sunshine (Del Valle Perea *et al.*, 2007). The fruits are edible (Lahitte *et al.*, 1998; Arenas, 2003). This native tree has educational, cultural, and ecological importance, contributing to the conservation of the environment. In Argentina, Buenos Aires province, with the objective to protect the "monte of tala", also known as "talar", the Gestión Talares Bonaerenses was created. These "talares" are very important, e.g., as a part of birds and several insect species diets, soil erosion control, and ecological balance preservation (Mérida and Athor, 2006; Aramburú *et al.*, 2009). The "talar" is reducing its distribution area due to the agricultural frontier expansion; its seeds have lower germination ability, and the shoots present difficulty growing (Abedini and Ruscitti, 1996; Abedini *et al.*, 2000).

Cell culture technology is one way to contribute to species conservation. Some assays of *in vitro* micropropagation had been carried out (Abedini *et al.*, 2000; Adema *et al.*, 2007), however when stems and thorns were used, difficulties were found in promoting new shoot development (Adema *et al.*, 2007). According to Romanczuk (1976), the thorns of genus *Celtis* may have one node or a small number of nodes. Hunziker and Dottori (1976) distinguished between leafless thorns and leafy thorns. Romanczuk

and del Pero de Martínez (1978) on the basis of thorn characteristics recognized two groups of species: one having thorns with invisible nodes, and the other comprising species with visible nodes as they produce small branches with leaves on them. On the basis of these observations we believe that thorns have areas (buds) with meristematic cells, and locating these areas with meristematic activity might be useful for the multiplication of this species. Our objective was to analyze the anatomy of thorns and find one or more specific areas with meristematic tissue (buds).

MATERIALS AND METHODS

Family *Celtidaceae* (= *Ulmaceae*), *Celtis ehrenbergiana* (Klotzsch) Liebm. Synonyms: *C. tala* Gillies ex Planch., *Momisia ehrenbergiana* Klotzsch, *M. integrifolia* Wedd., *M. lancifolia* Wedd., *Celtis tala* Gillies ex Planch. var. *weddelliana* Planch., *C. sellowiana* Miq., *C. bonplandiana* Planch., *C. tala* Gillies ex Planch. var. *gaudichaudiana* Planch., *C. tala* var. *tala* (= var. *gilliesiana* Planch.) f. *obtusata* Chodat & Hassl., *C. pallida* Torr., *C. tala* Planch. var. *pallida* (Torr.) Planch., *C. spinosa* Spreng. var. *pallida* (Torr.) M. C. Johnst., *C. flexuosa* Wedd. var. *glabrifolia* Griseb., *C. tala* Planch. var. *chichape* (Wedd.) Planch. f. *subpilosa* Kuntze, *C. tala* Planch. var. *chichape* (Wedd.) Planch. f. *subtomentosa* Kuntze, *Sarcomphalus punctatus* Urb. & Ekman, *Celtis punctata* (Urb. & Ekman) Urb. & Ekman (Berg and Dahlberg, 2001; Zuloaga *et al.*, 2008).

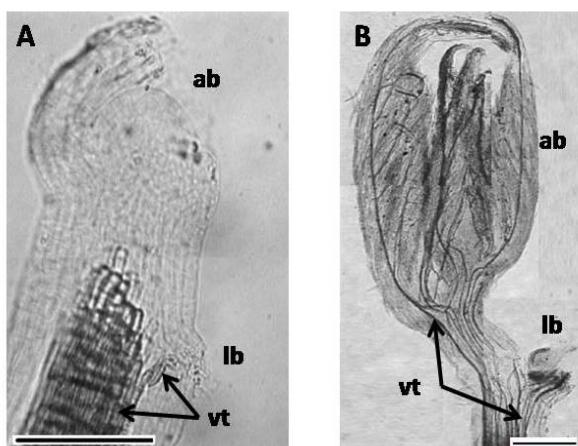
It is a shrub or a tree up to 4-12(-15) m tall, with arching branches, and straight thorns, solitary or geminate, up to 2-8(-20) mm long. Leaves are simple and positioned one per node in an alternate arrangement. Petiole 2-8 mm in length, and ovate, elliptic or oblong leaf blade shapes up to 1-5(-10) x 0.5-3(-7) cm, with asymmetrical base, acute or obtuse apex and irregularly dentate margin; chartaceous to subcoriaceous consistency. Glandular (pluricellular) and non-glandular trichomes (cystoliths) are present all over the surfaces. Pocket domatia are found on the abaxial surface in the main vein axils (Dottori, 1976). Stipules are deciduous. Inflorescences have imperfect and perfect flowers. Fruit is a yellow to orange rounded drupe in fall; 0.4-1 cm long and 6 mm diam. (Romanczuk, 1987; Biloni, 1990; Berg and Dahlberg, 2001).

The study was performed using living thorn samples collected from trees growing in the Arboretum and Botanical Garden "C. Spegazzini", Facultad de Ciencias Agrarias y Forestales,

Universidad Nacional de La Plata, Argentina, where the LPAG herbarium and the laboratory are located and in which the study was carried out. Other specimens from the LP herbarium were also surveyed (acronym according to Holmgren *et al.* 1990). ARGENTINA. Buenos Aires province, Pdo. La Plata: City of La Plata, Oct-2008, A. M. Arambarri 301 (LPAG). San Luis province. La Capital: Alto Pencoso, Feb 1914, *Rodrigo s.n.* (LP 027957). Santa Fe province, Dpto. 9 de Julio: Tostado, Feb 1936, *M.M. Job s.n.* (LP 30395). Santiago del Estero province, Dpto. Guasayán: Cerrillos, Jan 1950, *M.M. Job 2818* (LP 905690).

To analyze the thorn, freehand longitudinal semithin sections were obtained. Some thorn samples were cleared to complement the study using the method of Dizeo de Strittmatter (1973). Slides have been made with the sections and cleared thorn material stained with Safranin O in 80% ethanol and mounted in gelatin-glycerine. The microscopic slides were observed under light microscope (LM) Gemalux. LM images were captured by means of a color PAL CCD camera attached to an LM, and they were digitalized by means of Hyper Media Center software.

Figure 1. *Celtis ehrenbergiana*.



Thorn longitudinal view. A, longitudinal section: vt, vascular tissue of lateral bud; ab, apical bud; lb, lateral bud. B, thorn view at a more developed stage: vt, vascular tissue irrigating both buds; ab, dominant apical bud, with conspicuous leaves primordium; lb, lateral bud. Scales: A, 100 µm, B, 500 µm.

RESULTS AND DISCUSSION

The thorn longitudinal section (Fig. 1A) and the longitudinal cleared thorn (Fig. 1B) show two buds: one in the apical and the other in a lateral subapical position. From an exomorphological point of view, the

thorns of *Celtis ehrenbergiana* are straight and without nodes.

However, buds were found in the anatomy study, and node presence can be deduced. The presence of these thorns with conspicuous buds is in agreement with the description of Romanczuk and del Pero de Martínez (1978) about the existence of thorns giving branches which grow in zig-zag pattern. In the light of our results, only some thorns have active meristematic tissue (buds) and develop shoots. Perhaps this was one reason for new undeveloped shoots in the material assayed by Adema *et al.* (2007). It means that the selection of thorns is important when they are used in assays of *in vitro* micropagation.

CONCLUSIONS

Only some thorns develop shoots. One of the two geminate thorns located at the apex of actively growing branches has two buds. Therefore, these portions which have numerous meristematic cells with division capacity might be assayed with cell culture technology.

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