

## Anatomical and Chemical Analysis in *Solidago chilensis* var. *chilensis* (Asteraceae)

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**SUMMARY.** *Solidago chilensis* Meyen var. *chilensis* (Asteraceae) is a weed with an ample distribution in Argentina. The objective of this paper was to deepen the organs anatomy knowledge and to perform a preliminary chemical evaluation to detect constituents present in them. Root, rhizome, aerial stem, leaf and inflorescence structures were studied using conventional methods. Histochemical methods were used to identify plant constituents, inulin, lipophilic substances, and starch. Plant extracts of many species are used in biological control of pathogen microorganisms. *Solidago chilensis* var. *chilensis* can be seen as a potential source of extracts production to control pathogens.

### INTRODUCTION

*Solidago chilensis* Meyen var. *chilensis* “vara de oro” (Asteraceae), is a perennial herb widely distributed in Buenos Aires province and North of Argentina growing from 0-2500 m a.s.l. <sup>1-5</sup>. It is a rhizomatous and invasive weed with gregarious habit living along the roadside and uncultivated farmlands <sup>6,7</sup>. The saponins of this species resulted toxic for livestock <sup>8</sup>. However, this species is mentioned in the literature as medicinal, the decoction of leaves and flowers is drunk as a diuretic remedy, and the root has anti-cephalalgic and anti-litic properties <sup>9</sup>. It is also used as anti-helminthic, anti-rheumatic, anti-septic, anti-inflammatory, anti-oxidant, and it has gastroprotective activity; in external application is used to treat wounds, trauma, contusions and sciatic <sup>10-17</sup>. It is a honey plant and recently was cultivated observing its utility as ornamental for xeric regions <sup>18</sup>. The widely distribution, easy reproduction, abundance, and numerous potential applications of this species justify the objective to deepen the organs anatomy knowledge and to perform a preliminary histochemical evaluation to detect the constituents present in them.

### MATERIALS AND METHODS

#### *Plant materials*

Complete and fresh plants were collected roadside Río de La Plata avenue, between La Plata and Berisso cities, at 34°50' 49.40" Lat. S, and 57°50' 48.49" Long. W; also were collected at the Facultad de Ciencias Agrarias y Forestales (FCAyF), Jardín Botánico y Arboretum “C. Spigazzini”, Universidad Nacional de La Plata (UNLP) [34°55'S, 57°56'W], Buenos Aires province. The voucher specimens were deposited in the herbarium, Hernández # 99 and 100 (LPAG, acronym according to Thiers, 2011) <sup>19</sup>. The botanical nomenclature follows Zuloaga *et al.* <sup>5</sup>, and the site www.darwin.edu.ar.

#### *Anatomical analysis*

Fresh organs (root, rhizome, stem, leaf, and inflorescence) were fractioned. To avoid alterations each one was fixed with FAA 70% (formalin: glacial acetic acid: ethanol) <sup>20</sup>. Longitudinal and cross-sections were obtained in the middle part of each organ. To distinguish different structures the sections were bleached in 50% sodium hypochlorite (NaOCl), washed three times with distilled water, and stained with 80%

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alcoholic solution of safranin or 1% aqueous solution of cresyl violet. Slides were mounted on glycerin jelly<sup>20</sup>. Structures were examined by means of a light microscope (LM), Leitz SM lux equipped with a lucid camera used to prepare the schematic drawings. LM images were obtained by means of a color PAL CCD camera attached to a LM Gemalux. The images were captured and digitalized by means of Hyper Media Center software. Symbols used in schematic drawings and terminology used in organs description are according to Metcalfe & Chalk<sup>21,22</sup>. We used the term reservoirs to refer secretory structures found in every organ as was suggested by Lersten & Curtis<sup>23</sup>. Trichomes descriptions are in agreement with Ramayya<sup>24</sup>.

**Chemical analysis**

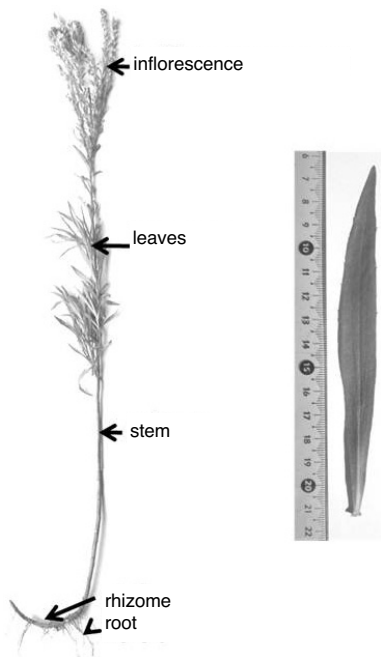
Inulin was developed by a natural alcohol precipitate test produced by FAA solution. Histochemical tests were performed for lipophilic substances with a saturated alcoholic solution of Sudan IV<sup>20</sup>, and for starch with iodine-potassium iodide (IKI)<sup>25</sup>.

**RESULTS**

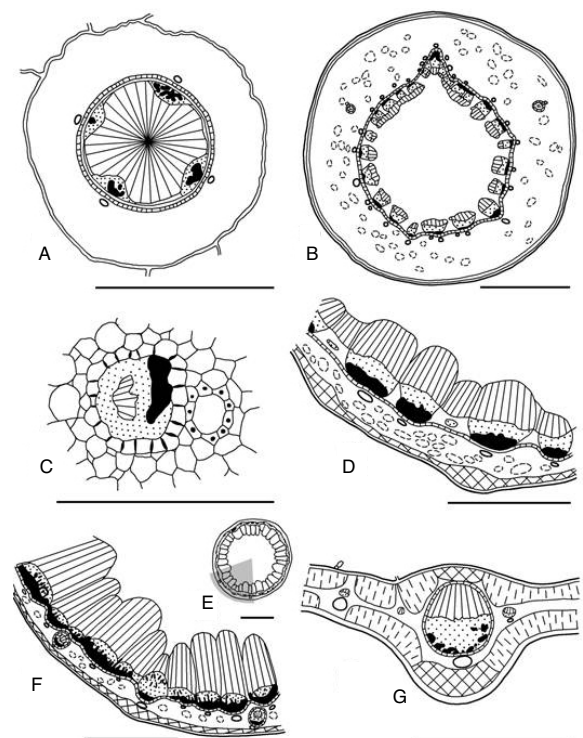
**Anatomical analysis**

Roots are thin (Fig. 1). The cross-section is rounded in outline (Fig. 2A; Fig. 3A). The rhizodermis is one layer of quadrangular cells. The periderm is formed by a phellogen installed su-

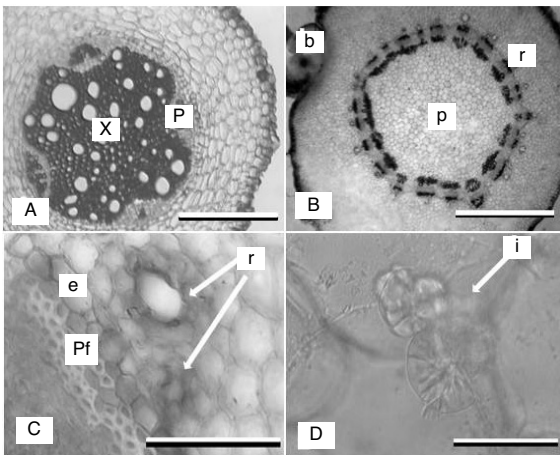
perificially (1-2 layered, initial periderm). In the primary cortex there are 10-12 layers of parenchyma. The endodermis has Casparian band. In the vascular cylinder, the pericycle surrounds the secondary vascular structure. The central part of the root is occupied by a mass of secondary xylem and may be see four clusters of phloem with fibers (Fig. 3A). In the cortex and in front of each cluster of phloem there is one reservoir. The rhizome is soft, yellowish with smooth surface (Fig. 1). The cross-section is circular in outline (Fig. 2B), having buds (Fig. 3B). The epidermis is one layer of small quadrangular cells. The periderm is formed by a phellogen installed superficially with 1-2 layers



**Figure 1.** Organs of *Solidago chilensis* var. *chilensis* plant. Scale: 30 cm. On the right is a detail of one leaf.



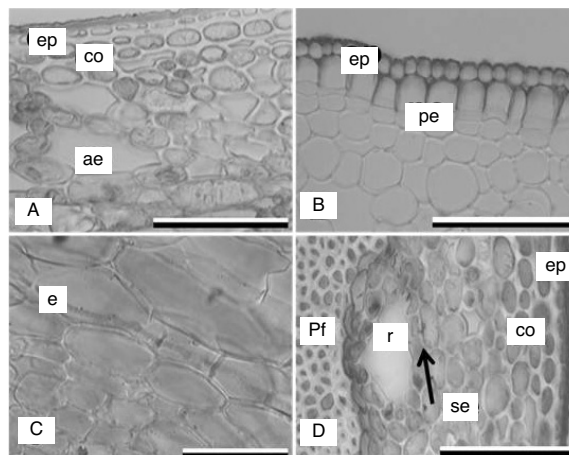
**Figure 2.** Schematic drawings of each vegetative organ of *Solidago chilensis* var. *chilensis* in cross-sections. **A**, root: a general view. **B**, rhizome: shows an ample cortical aerenchyma, and eustele with an ample parenchyma in the pith. Also may be see two concentric vascular bundles in the cortex. **C**, Detail of one cortical concentric vascular bundle. **D**, shows primary structure in a stem of 3 mm diameter. **E**, shows stem outline rounded-waved. **F**, details of secondary structure in a stem of 5 mm diameter. See concentric vascular bundles in a narrow cortex. **G**, the cross-section of leaf showing biconvex middle vein with collateral vascular bundle surrounded by endodermis and a reservoir situated in front of phloem. See the collenchyma to adaxial and abaxial surfaces, and isobilateral mesophyll. The reservoirs were indicated with circle of full line. Scales: A, G: 500 µm; C: 200 µm; B, D-F: 1mm.



**Figure 3.** Root and rhizome traits in cross-section. **A**, root, see the vascular cylinder showing secondary xylem (X) and four clusters of phloem (P). **B**, rhizome, in the general view, may be seen ample pith (p), eustele with 1-3 reservoirs (r) in front of each collateral vascular bundles, and a bud (b). **C**, rhizome, detailed view showing two reservoirs (see arrow) in front of phloem fibers (Pf), and separated by endodermis (e). **D**, Inulin crystals (i) in parenchymatic pith of the rhizome. Scales: A, 200  $\mu$ m; B, 1mm; C, 100  $\mu$ m; D, 50  $\mu$ m.

of large quadrangular cells. In the primary cortex there are several layers of aerenchyma (Fig. 2B). In the vascular cylinder, the vascular bundles are collateral, forming a typical eustele around the pith which consists of isodiametric cells (Figs. 2B and 3B). In the cortex appear concentric vascular bundles, each enclosed by endodermis and external to it, to the rhizome peripheral side one reservoir (Figs. 2B and C). In the cortex, external to endodermis and in front of each vascular bundle appear 1-3 reservoirs (Figs. 3B and C). Between cortical parenchyma and vascular cylinder appears the endodermis with conspicuous Casparian band (Fig. 3C). The cortex and pith tissue is rich in inulin crystals (Fig. 3D). The aerial stem is cylindrical and erect, little branched, 1.80 m tall (Fig. 1). The most important differences with the rhizome are: the presence of collenchyma; presence of only one reservoir in front of each vascular bundle, and a narrow cortex. The cross-section outline is rounded-waved (Figs. 2D-F).

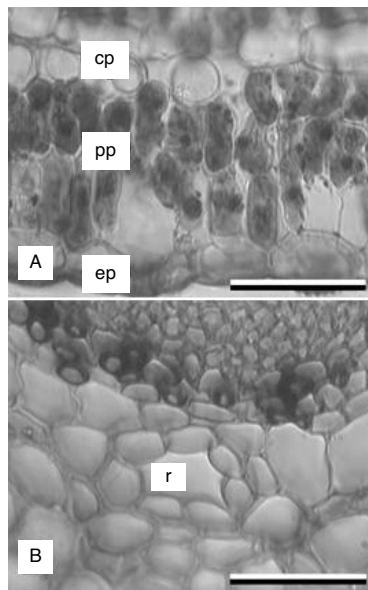
The epidermis one layered is coated with a thick cuticle (Fig. 4A). The periderm is formed by a phellogen installed superficially (Fig. 4B). In the cortex there are 2-6(-8) layers of laminar collenchyma, followed by 6-10 layers of aerenchyma (Fig. 4A), in which the most inner layer is an endodermis with Casparian band



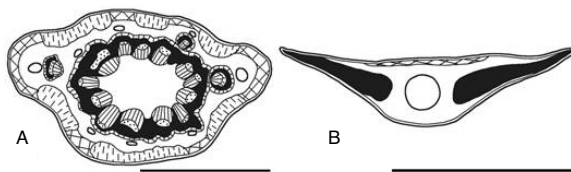
**Figure 4.** Stem traits in cross-section. **A**, epidermis with cuticle (ep), laminar collenchyma (co), and aerenchyma (ae). **B**, epidermis (ep), and superficial periderm (pe). **C**, details of endodermis Casparian band (e). **D**, stem periphery showing epidermis (ep) with cuticle, collenchyma (co), reservoir (r) situated in front of phloem fibers (Pf) exhibiting the uniseriate secretory epithelium (se). Scales: A, B, D: 100  $\mu$ m; C, 50  $\mu$ m.

(Fig. 4C). In the cortex there are concentric vascular bundles similar to rhizome (Fig. 2C). The vascular cylinder is eustele, which is changing to secondary structure forming a ring of secondary phloem and xylem. In the cortex in front of each cluster of phloem fibers appears one reservoir. The reservoirs are schizogenus in origin having uniseriate epithelium. These secretory structures are the same when are compared the different organs (Fig. 4D). The pith showed some inulin crystals. The leaf is simple, sessile with blade linear-lanceolate attenuated in the base with entire margin, 40-80 (-140) length x 5-10 mm wide (Fig. 1). Epidermal cells in face view, shows straight anticlinal cell walls and anomocytic and anisocytic stomata on both surfaces. Cross-section of the blade shows an epidermis one layered formed by quadrangular-elliptic cells covered by a thick cuticle. Stomata are at level respect to other epidermal cells or slightly elevated on the abaxial surface. Three types of trichomes were found, on the surface they are bulbiferous flagellate and biseriate vesicular, and distributed over the veins and in the foliar margin are simple conical. The middle vein is biconvex in cross-section more prominent to the abaxial surface (Fig. 2G).

The mesophyll is isobilateral having 2-3 layers of short cells of palisade parenchyma to



**Figure 5.** Leaf traits in cross-section. **A**, epidermis (ep), palisade parenchyma (pp), and central colorless parenchyma (cp). **B**, reservoir (r) in the parenchyma of middle vein, situated in front of phloem. Scales: 50  $\mu$ m.



**Figure 6.** Inflorescence. **A**, peduncle traits, in cross-section. **B**, phyllary traits in cross-section. Reservoirs are indicated using full line. Scales: 500  $\mu$ m.

both epidermis, and 1-3 layers of central colorless cells (Fig. 5A). The middle collateral vascular bundle is surrounded by endodermis and on the phloem side the parenchyma exhibit a reservoir (Fig. 5B). Angular collenchyma occurs and it comprehends 2-3 layers next to both surfaces. The inflorescence has from the principal stem a variable number of secondary branches carrying numerous capitula bearing bright golden yellow flowers (Fig. 1).

The cross-sections of the principal stem, secondary branches, capitula peduncle, and phyllaries of the involucre showed secretory reservoirs in front of each vascular bundle, included occupying the vascular bundle place, in the phyllary (Fig. 6A and 6B).

#### Chemical analysis

Inulin crystals were observed in cortical and medullar parenchyma of the rhizome. Small and

few crystals also were seen in pith of aerial stems. When freehand sections of fresh leaves were treated with Sudan IV the secretory reservoirs in every organs showed bright red oil droplets, and when the sections were treated with IKI solution a little quantity of starch was observed only in the endodermis of the stem.

#### DISCUSSION

We found the xeromorphic epidermal characteristics in coincidence with Gil *et al.*<sup>18</sup>, adding the xeromorphic foliar structure amphistomatic equifacial with an isobilateral mesophyll, which has palisade parenchyma to both surfaces. Metcalfe & Chalk<sup>21</sup> indicated that frequently in the family Asteraceae there are cortical vascular bundles. They are leaf trace bundles. It suggested that the cortical vascular bundles in the rhizome are vascular supply to the buds. To respect of secretory structures Evert<sup>26</sup> named cavities when they are short secretory spaces, and duct when they are large. There is not accord among different authors about the secretory structures nomenclature, namely cavities, canals, ducts, secretory inner spaces, tubular cavities and reservoirs<sup>21,23,27-31</sup>. In fact, for the secretory structures of *S. chilensis* var. *chilensis* we adopted the reservoirs term used by Lersten & Curtis<sup>23</sup>, because our longitudinal sections showed coincidence with author's description of these secretory spaces. Every organ exhibited one reservoir situated in front of each vascular bundle, however, in the rhizome one to three were found. This result coincides with the concept that the subterranean condition is favorable to the secretory reservoirs production<sup>29</sup>. The disposition of inulin crystals in clusters adjacent to cell walls have been reported before for other species<sup>29-32</sup>. The result accords well with data on that preferentially inulin is accumulated in subterranean organs<sup>33, 34</sup>, however, in *S. chilensis* var. *chilensis* also was observed a little quantity in pith of aerial stems.

#### CONCLUSIONS

When freehand sections of different organs were treated with Sudan IV the reservoirs showed bright red oil droplets. This coloration suggests they are essential oils. Results are the base for deepen studies to use in biological control of pathogens.

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