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Fruit anatomy of species of *Solanum* sect. *Acanthophora* (Solanaceae)

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Received 11 October 2007; accepted 7 January 2008

Abstract

The fruits of 10 species of *Solanum* sect. *Acanthophora* were studied. Cross and/or longitudinal and/or tangential microtome sections, stained mostly with astra blue/basic fuchsin, were made for microscopic examination. Three different kinds of cells were found in the epidermis, immediately below which a hypodermis, consisting in any of four types of structures, was always found. The mesocarp exhibits two histologically differentiated zones, an external one (formed by normal or spongy parenchyma, according to the species), and an internal one, commonly juicy, and with proliferations among the seeds. The diagnostic value of all these structures is assessed. Morpho-anatomical information is used to define fruit types beyond the berry, traditionally described for *Solanum*, and the probable dispersal syndrome related to them is discussed. Fruit similarities are slightly noticeable in several cases, while differences may be the result of their sexual system – which affects specially the size – and their histology, which is related to the dispersal syndrome. The comparison of our data with previous molecular phylogeny of the section suggests that a significant morphological variation is not associated with significant DNA sequence changes.

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Keywords: Anatomy; Epidermis; Fruit; Phylogeny; *Solanum*; subgen. *Leptostemonum*

Introduction

Solanum subgen. *Leptostemonum* (Dunal) Bitter is a large group (ca. 450 species) within *Solanum*, comprising almost one-third of the genus (Bohs, 2005; Levin et al., 2005; Weese and Bohs, 2007). The species of this subgenus have a worldwide distribution, with the greatest species richness in South America, Africa, and Australia. Several of them are economically important, for example *S. melongena* L. (aubergine or brinjal eggplant), *S. aethiopicum* L. (scarlet eggplant), *S. quitoense* Lam. (naranjilla or lulo) or *S. sessiliflorum* Dunal (cocona). One of the main characteristics defining

the subgen. *Leptostemonum* is the presence of sharp epidermal prickles on stems and leaves in all, except for a few, taxa. Most members of the subgenus have stellate hairs and attenuate anthers with small terminal pores.

Within subgen. *Leptostemonum*, sect. *Acanthophora* Dunal is distinguishable and easily grouped due to the existence of simple hairs on the upper leaf surface (Levin et al., 2005; Nee, 1991; Whalen, 1984). This section is monophyletic (Levin et al., 2005), and its diversity center is located in eastern Brazil. It includes about 20 herbs and small shrubs mainly, adapted to disturbed areas and secondary open forests. The importance of sect. *Acanthophora* lies in the fact that some of its species are considered invasive weeds. For instance, *S. viarum* Dunal (“tropical soda apple”), is naturalized in USA, Africa, and Asia (Bryson and Byrd, 1994; Welman,

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2003), while *S. palinacanthum* Dunal (“pocote”, “joá bagudo”); invades roadsides and crop fields in Argentina and Brazil (Auler Mentz and Oliveira, 2004; Nee, 1991). Conversely, other species of the section are cultivated for their ornamental value (e.g. *S. mammosum* L., “apple of Sodom”, “cow’s udder” or “nipple fruit”; and *S. capsicoides* All., “cockroach berry”).

Species of sect. *Acanthophora* exhibit great variation in their fruit morphology. Some of them (*S. mammosum*, *S. palinacanthum*) produce large fruits, reaching up to 5.5 cm in diameter and with a spongy mesocarp. Other species produce smaller fruits with a thinner spongy layer (*S. aculeatissimum* Jacq., *S. viarum*) while others are said to be irregularly dehiscent (*S. capsicoides*, *S. platense* Dieckmann) (Levin et al., 2005). Finally, some species (e.g. *S. atropurpureum* Schrank) develop small, red, juicy fruits. Despite this variability, no anatomical studies have been performed in fruits of *Acanthophora* species, except for *S. mammosum* (Miller, 1969), so all the discussion about evolution of fruit traits has been superficial. Moreover, the study of fruit anatomy in the entire subgenus has been broadly neglected. Thus, aiming to achieve a comprehensive knowledge of the fruit anatomy within the species of *Leptostemonum*, and to evaluate probable taxonomical and evolutionary incidences, we started a carpological analysis in members of different sections of the subgenus. As a result, some groups, such as the Argentine representatives of sect. *Melongena* (sensu Nee, 1999), revealed their vast heterogeneity concerning fruit traits (Chiarini and Barboza, 2007), supporting the paraphyly of this section (Levin et al., 2006). We are now presenting results in species of sect. *Acanthophora*, as a contribution to clarify the fruit evolutionary pattern of this group, and to understand the relationship between structure and function.

Materials and methods

Ten wild species of *Solanum* sect. *Acanthophora* were analysed. The following are the voucher data of the studied material:

Solanum aculeatissimum Jacq. BRAZIL. **Santa Catarina**. Mun. Monte Castelo, 28°38'12"S, 50°15'06"W, 23-II-2006, Barboza et al. 1617 (CORD).

Solanum aenictum C. V. Morton. ARGENTINA. **Corrientes**. Santo Tomé, 7-I-2002, Chiarini 536 (CORD).

Solanum atropurpureum Schrank. ARGENTINA. **Corrientes**. Santo Tomé, 7-I-2002, Chiarini 531 (CORD); Goya, Arroyo Guazú, 29°50'26"S, 59°24'24"W, 3-XII-2002, Barboza et al. 355 (CORD).

Solanum capsicoides All. ARGENTINA. **Corrientes**. Ituzaingó, Isla Apipé Grande, 4-XII-2002, Barboza

et al. 394 (CORD); BRAZIL. **Santa Catarina**. Trombudo Central, 24-XI-2003, Mentz et al. 274 (CORD, ICN); Garuva, 24-II-2006, Barboza et al. 1623 (CORD); **São Paulo**. From Salesópolis to Paraibuna, SP 077, km 108.5, 25-II-2006, Barboza et al. 1641 (CORD); **Rio de Janeiro**. Floresta de Tijuca, 15-VII-2003, Barboza et al. s.n. (GUA 48406). *Solanum incarceratum* Ruiz et Pav. BRAZIL. **São Paulo**. Estrada Itú-Jundiá, 25°15'36"S, 47°15'34"W, 29-VI-03, Marcondes et al. s.n. (CORD 1028).

Solanum mammosum L. ECUADOR. **Napo**. XII-1995, Hunziker s.n. (CORD 1024).

Solanum palinacanthum Dunal. ARGENTINA. **Córdoba**. Capital, 1-VI-2001, Chiarini 465 (CORD); **Tucumán**. Famaillá, 1-IV-1977, Hunziker et al. 23081 (CORD); PARAGUAY. **Caaguazú**. 25°22'43"S, 56°00'42"W, 12-XII-2002, Barboza et al. 495 (CORD).

Solanum platense Dieckmann. ARGENTINA. **Misiones**. San Ignacio, ayo. Macaco, 7-XII-2002, Barboza et al. 441 (CORD).

Solanum tenuispinum Rusby. ARGENTINA. **Salta**. Santa Victoria, Baritú, 2-X-2001, Barboza et al. 292 (CORD); Rosario de Lerma, 24°58'15"S, 65°35'37"W, 6-III-2002, Negritto et al. 293 (CORD); **Catamarca**. Andalgalá, Río Chacras, 27°23'01"S, 65°59'29"W, 23-II-2003, Barboza et al. 629 (CORD). *Solanum viarum* Dunal. ARGENTINA. **Corrientes**. Santo Tomé, 6-I-2002, Chiarini 533, 537 and 538 (CORD); Ituzaingó, 14-V-2004, Barboza et al. 1006 (CORD); **Misiones**. Gral. Manuel Belgrano, 25°42'24"S, 54°5'48"W, 30-XI-2003 Barboza et al. 819 (CORD).

For microscopic examination, whole or cut up ripe fruits were preserved in a formaldehyde–acetic acid–ethanol mixture, then dehydrated in a 50–100% ethanol series, and embedded in Paramat[®] resin. Cross and/or longitudinal and/or tangential microtome sections 10–12 µm thick were stained mostly with a 1% astra blue solution in a 1% water/basic fuchsin solution in 50° ethanol. Astra blue stains cell wall polysaccharides such as cellulose and pectins, while basic fuchsin shows affinity for lignified, suberized or cutinized walls, i.e., structures embedded in phenolic substances (Kraus et al., 1998). Basic fuchsin also stains chloroplasts and nucleic acids. In some cases, additional sections were stained with a 0.05% cresyl blue solution in water (Pérez and Tomassi, 2002).

The specimens were visualized using a Zeiss Axiophot microscope. The images were captured with a digital camera assembled to the microscope.

In order to enrich the discussion, data about the sexual system of the species studied were drawn from the literature, and missing information was found via our observations in the field.

Results

Most of the taxa have spherical fruits, with the exception of *S. tenuispinum*, which has ovoid fruits, and *S. mammosum*, which has an anomalous fruit, 3–5-lobed at base.

According to Knapp (2002) the fruits of *Solanum* sect. *Acanthophora* studied here are berries in a conventional sense. In the Spjut's (1994) system, and according to our observations, fruits are either berries, i.e. simple fruits with an indehiscent pericarp – containing many seeds embedded in a solid fleshy mass, supported by an epicarp that is less than 2 mm thick – or a carcerulus, which is a fruit resembling the true berries, but with an aerial space between the seeds and the pericarp when the fruit is completely ripe, as is the case with peppers (*Capsicum* spp.) (Fig. 1A–D, Table 1).

In general, fruits of the investigated species are small to medium-sized, varying from 1.35 cm (*S. incarceratum*) to 4 cm (*S. mammosum*) in diameter. When mature, fruits are of a single colour: red or orange-red (*S. capsicoides* and *S. atropurpureum*), yellow (most of the species), or greenish yellow (*S. incarceratum* and *S. platense*).

Seeds belong to two types: winged, i.e. a strongly flattened seed, with the seed coat forming a prominent wing, 0.8–2 mm wide (e.g. *S. platense*; *S. capsicoides*, Fig. 2A; *S. atropurpureum*, Fig. 2B), or bulky, i.e. a slightly flattened or non-flattened lenticular seed, whose seed coat margin is not thickened at all (e.g. *S. viarum*, Fig. 2C; *S. mammosum*, Fig. 2D).

The pericarp comprises three clearly distinguishable zones: the exocarp, the mesocarp, and the endocarp.

Exocarp

The cuticle is highly variable and usually thick (especially in *S. incarceratum*, Fig. 4F), and it can be smooth (e.g. *S. atropurpureum* Fig. 3D), undulate (e.g. *S. capsicoides* Fig. 4A), or grooved (e.g. *S. aculeatissimum*, Fig. 4D). In all species, cuticular wedges are present among the epidermal cells, these wedges being notably deep in *S. atropurpureum* (Fig. 3D) and in

S. aenictum (Fig. 3B). Ventilation clefts and stomata are lacking in all cases.

The epidermis consists of different kinds of cells according to the species, as follows:

- (1) A unistrate layer of small, isodiametric cells, with dense content and cellulosic walls, e.g. *S. platense* (Fig. 4E) or *S. aculeatissimum* (Fig. 4D).
- (2) A unistrate layer of relatively large, bottle-shaped cells, here designed as lageniform cells, with a noticeable nucleus, dense content and simple pits. These cells can be weakly lignified, but not enough so as to be considered true sclereids, e.g. *S. aenictum* (Fig. 3B).
- (3) A layer of brachysclereids (*S. capsicoides*, Fig. 4A and B).

Immediately below the epidermis, a hypodermis is differentiated, consisting in any of the four following kinds of structures:

- (a) Compressed parenchymatous cells, with dense content, arranged in 1 or 2 layers (e.g. *S. incarceratum*, Fig. 4F).
- (b) Rounded parenchymatous cells containing rhombohedral crystals of calcium oxalate, in a single continuous layer or alternating with non-crystalliferous cells, below which some collenchymatous layers are found (e.g. *S. viarum*, Fig. 3A).
- (c) Sclerenchymatous cells, i.e., true brachysclereids, naturally orange coloured, followed by 1–2, or several true collenchymatous layers (e.g. *S. capsicoides*, Fig. 4A and B). Transitional forms between sclereids and parenchyma cells develop along the boundary between the exocarp and the mesocarp.
- (d) Several layers of very thickened and lignified cell walls, taller than wide, which could be considered a sclerified collenchyma. In *S. mammosum* and *S. palinacanthum* (Fig. 3E), the cells of these layers adopt a strange “amoeboid” appearance due to their irregular outline; a normal collenchyma is developed underneath.

The epidermis and the hypodermis constitute a unit, the exocarp, which generally has layers that gradually decrease their degree of lignification from the outside to the inside of the fruit.

Usually, when the fruit is immature, the cell layers located below the epidermis (or below the crystalliferous layer or layer of fibres, when present) have chloroplasts and chromoplasts. In mature fruits, the chloroplasts disappear and the cells become compressed. A collenchyma is always present, within which the number of layers and the degree of lignification vary according to the species.



Fig. 1. Senescent fruits of the carcerulus type in *Solanum* sect. *Acanthophora* species. A longitudinal cut shows the disposition of the seeds: (A) *S. aenictum*, (B) *S. viarum*, (C) *S. capsicoides*, and (D) *S. platense*. The bar represents 1 cm.

Table 1. Macroscopic and anatomical fruit features of the 10 species of *Solanum* sect. *Acanthophora* studied

Species	Fruit type	Sexual system	Seeds type	Fruit colour	Exocarp			Mesocarp	
					Cuticle	Epidermis	Hypodermis	External zone	Internal zone
<i>S. aculeatissimum</i>	C	A	Bulky	Yellow	Grooved	Unistrate, isodiametric cells with dense content (Fig. 4D)	1–2 layers of normal to some sclerified collenchyma (Fig. 4D)	20 layers of spongy tissue over the veins and 20 underneath (Fig. 4C)	Tiny proliferations among the seeds (Fig. 4C)
<i>S. aenictum</i>	C (Fig. 1A)	A	Bulky	Yellow	Grooved with deep cuticular wedges	Unistrate, lageniform cells (Fig. 3B)	1–2 layers of normal collenchyma	60–65 layers of spongy tissue. Starch (in unripe fruits)	Absent
<i>S. atropurpureum</i>	B	H	Winged (Fig. 2B)	Red	Smooth, with deep cuticular wedges	Unistrate, short lageniform and trapezoidal cells (Fig. 3D)	1–2 layer of normal collenchyma (Fig. 3D)	A few layers of normal to somewhat spongy tissue	Tiny proliferations
<i>S. capsicoides</i>	C (Fig. 1C)	A	Winged (Fig. 2A)	Red	Undulate	1–2 layers of brachysclereids, naturally orange coloured (Fig. 4A and B)	Sclerenchyma or sclerified collenchyma, followed by normal collenchyma (Fig. 4A)	20 layers of spongy tissue over the veins and 20 underneath	Absent
<i>S. incarceratum</i>	B	H	Winged	Yellow greenish	Undulate	Unistrate, isodiametric cells with dense content (Fig. 3D)	1–2 layers of compressed cells with dense content (Fig. 3D)	5 layers of spongy tissue over the veins and 5 underneath	Some tiny proliferations
<i>S. mammosum</i>	C	A	Bulky (Fig. 2D)	Yellow	Smooth, with wedges	Unistrate, tall lageniform cells	Special sclerified collenchyma (amoeboid appearance)	40 layers of spongy tissue over the veins and 60 underneath, with the first layers dense, and the following loose	Absent
<i>S. palinacanthum</i>	C	A	Bulky	Yellow	Grooved	Unistrate, tall lageniform cells (Fig. 3E)	2–3 layers of special sclerified collenchyma (amoeboid appearance) (Fig. 3E), followed by normal collenchyma	Spongy tissue, 20–30 layers over the veins and 20 underneath (Fig. 3F)	Absent
<i>S. platense</i>	C (Fig. 1D)	A	Winged	Yellow greenish	Grooved	Unistrate, isodiametric cells with dense content (Fig. 4E)	1–2 collenchymatous layers	10 layers of spongy over the veins and 20 underneath (Fig. 4E)	Absent
<i>S. tenuispinum</i>	B	H	Winged	Yellow	Smooth	Unistrate, lageniform cells (Fig. 3C)	1–2 collenchymatous layers (Fig. 3C)	6–7 layers of spongy tissue over the veins and 5–6 underneath	Tiny proliferations (Fig. 3C)
<i>S. viarum</i>	C (Fig. 1B)	A	Bulky (Fig. 2C)	Yellow	Grooved	Unistrate, isodiametric cells with dense content (Fig. 3A)	1 layer of living cells containing a crystal, followed by 2–3 layers of laminar collenchyma (Fig. 3A)	18–20 layers of spongy tissue over the veins and 20 underneath	Absent

Abbreviations: A: andromonoecious species; B: berry; C: carcerulus; H: hermaphrodite species.

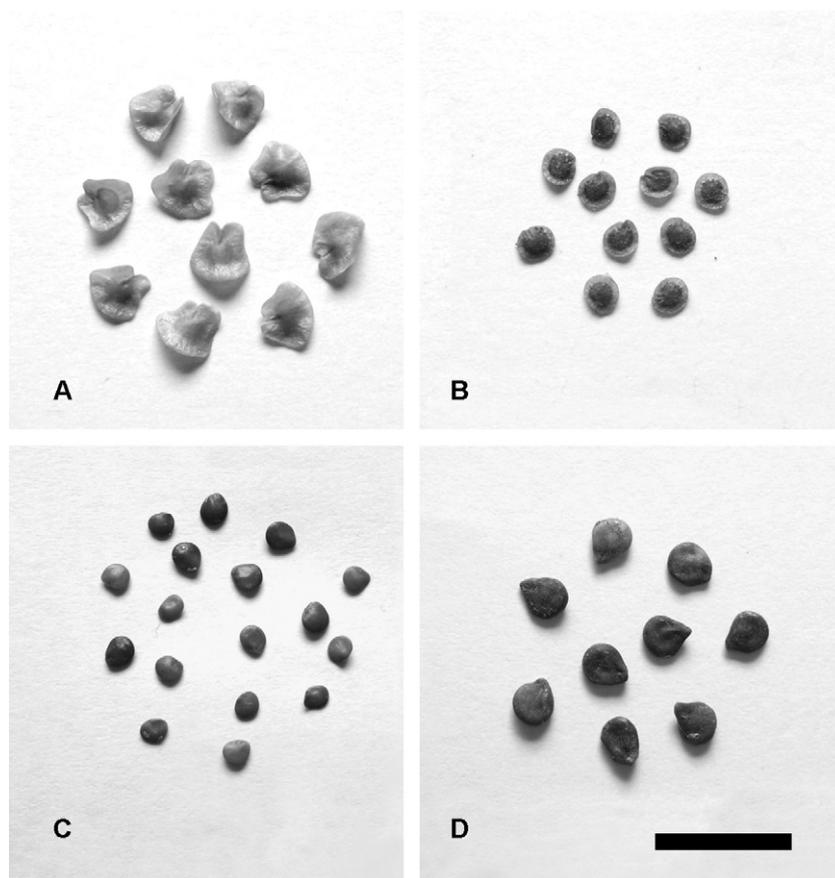


Fig. 2. Seed types of *Solanum* sect. *Acanthophora* species: (A and B) winged seeds and (C and D) bulky seeds. (A) *S. capsicoides*, (B) *S. atropurpureum*, (C) *S. viarum*, and (D) *S. mammosum*. The bar represents 1 cm. All pictures at the same scale.

Mesocarp

The number of layers of this tissue gives the pericarp its thickness. The higher the number of mesocarp layers, the thicker the pericarp. Fruits with a thick pericarp contain usually more than 10 layers. The mesocarp consists in two histologically differentiated zones: an external one (immediately below the hypodermis, evidenced with astra blue), and an internal one, identified with basic fuchsin. The external zone may adopt two forms, according to the kind of cell arrangement:

(1) A spongy parenchymatic tissue, resembling the albedo of the hesperidium, formed by big, very vacuolated or almost empty, loosely connected cells, with large intercellular spaces (e.g. *S. palinacanthum*, Fig. 3F). The cells increase their size towards the endocarp, and the cell walls get loose and undulated. At maturity, the mesocarp is not in direct contact with the seeds, therefore the fruit belongs to the carcerulus type (Fig. 1A–D). The number of parenchymatous layers varies according to the species; for example, 40 layers over the veins and

60 beneath them are found in *S. mammosum*, while only 10 layers over the veins and 8 underneath are found in *S. tenuispinum* (Fig. 3C).

(2) A non-specialized or normal parenchyma, consisting of regular, vacuolated, medium-sized cells with small intercellular spaces (*S. atropurpureum*, Fig. 3D).

Regardless of the cellular arrangement of the mesocarp external zone, it is always followed by an internal zone, which is commonly juicy, and develops proliferations among the seeds. The cells are large, with dense content filled with grana, which disorganize and release their own content to the locules and produce a mucilage-like substance that surrounds the seeds in the ripe fruit. In several species this mucilaginous content turns black at air contact, perhaps due to its phenolic or saponinic nature.

The thickness of each zone varies notably according to the species. For instance, in species with a thick spongy external zone, the internal zone has few layers or is altogether absent (*S. palinacanthum*, *S. platense*, Fig. 4E) while in other species it is well developed (*S. atropurpureum*, *S. aculeatissimum*, Fig. 4C).

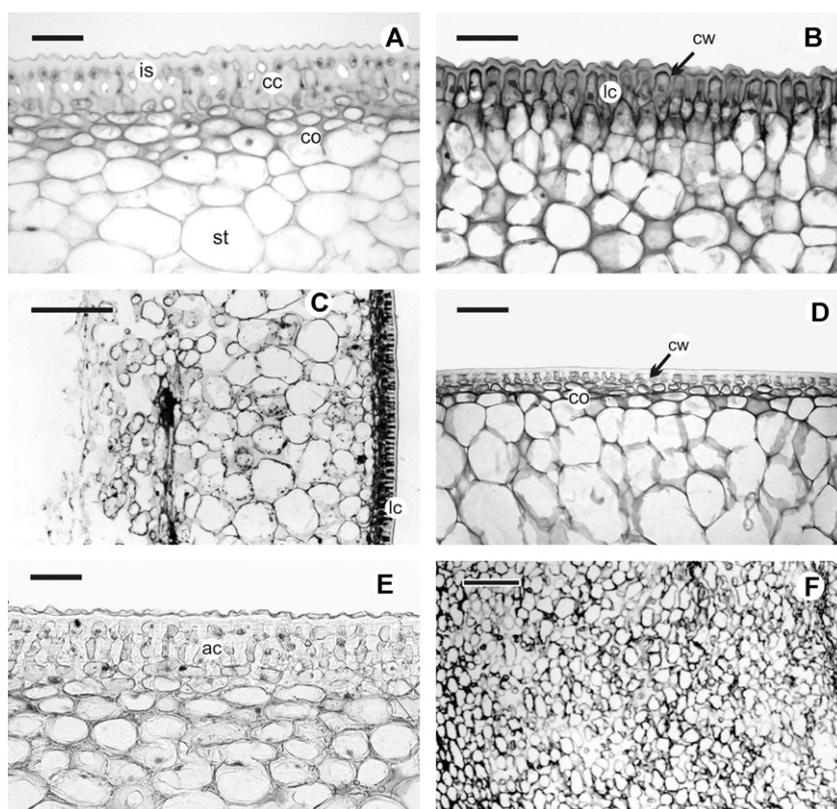


Fig. 3. Photomicrographs of fruit anatomy in *Solanum* sect. *Acanthophora* species, showing different structures: (A) *S. viarum*, (B) *S. aenictum*, (C) *S. tenuispinum*, (D) *S. atropurpureum*, (E) *S. palinacanthum*, and (F) detail of the spongy mesocarp in *S. palinacanthum*. The bars represent 50 μm in A–E, and 100 μm in F. Abbreviations: ac = amoeboid cells; bs = brachysclereids; cc = cells containing a crystal; co = collenchyma; cw = cuticular wedges; is = isodiametric cells; lc = lageniform cells; sc = sclerified collenchyma.

Stone cells or sclerosomes, widely present in many sections of *Solanum* and in related genera, are absent in sect. *Acanthophora*, at least in the species analysed here.

Endocarp

Finally, no specific particularities were observed in the endocarp. This layer, which is very difficult to observe due to its delicate structure, is uniseriate and lacks stomata in all cases.

Discussion

Trait assessment

Regarding the fruit itself, there is a slight similarity among the different species, but several constraints beyond phylogeny may be at play to result in this manner. The external resemblance concerns size and colour, while histological characters define each taxon. For instance, the lageniform cells are present in some

species with large yellow fruits (e.g. *S. aenictum* and *S. palinacanthum*), but not in other yellow-coloured ones (e.g. *S. aculeatissimum*).

Although colour is a variable feature, yellow fruits predominate. Colour has been attributed to the dispersal syndrome (Van der Pijl, 1982), and brightly coloured fruits would be more attractive to birds (Edmonds and Chweya, 1997). This should be the case in red-fruited species, but *S. capsicoides* or *S. atropurpureum* do not fit this hypothesis very well. In fact, *Solanum capsicoides* fruits have other features, such as their large size and their spongy mesocarp, which make them unsuitable for birds. In addition, both *S. capsicoides* and *S. atropurpureum* have winged seeds, presumably for wind dispersion.

Cipollini et al. (2002) found a significant connection among yellow colour, seed number, and mass. They also described a pattern in which large, yellow fruits with a low nutrient, toxic and highly dry matter content, differ from small, red, and black fruits with a high nutrient, non-toxic, and watery content. Nevertheless, colour cannot be easily linked with any morpho-anatomical structure in the species studied here, since fruits with the same colour show different tissue types.

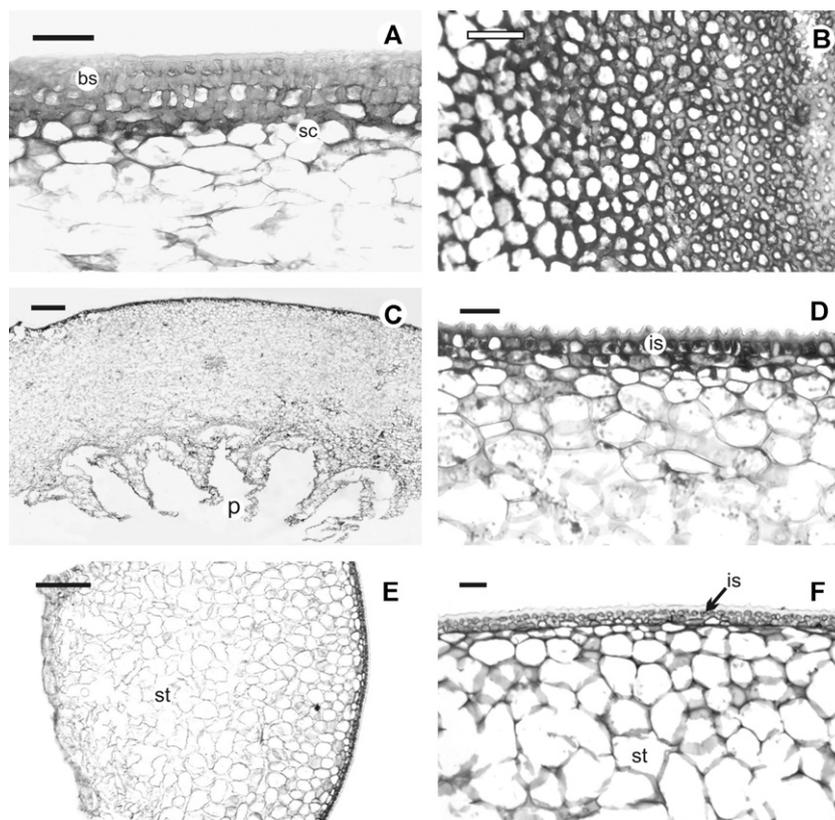


Fig. 4. Photomicrographs of fruit anatomy in *Solanum* sect. *Acanthophora* species, showing different structures: (A) *S. capsicoides* in cross-section, bar 50 μ m; (B) *S. capsicoides* in tangential section, showing the sclereids, bar = 50 μ m; (C) pericarp of *S. aculeatissimum*, bar = 0.5 mm; (D) exocarp of *S. aculeatissimum*, bar = 100 μ m; (E) *S. platense*, bar = 100 μ m; and (F) *S. incarceratum*, bar = 100 μ m. Abbreviations: bs = brachysclereids; is = isodiametric cells; p = proliferations among the seeds; sc = sclerified collenchyma; st = spongy tissue.

The cuticle has no diagnostic value, since it varies from one species to another. This situation has already been noticed in other “spiny *Solanum*” (Chiarini and Barboza, 2007; Dottori and Cosa, 1999, 2003).

The epidermis has shown a significant diversification. For instance, some species present isodiametric cells with dense content, and others present lageniform cells (which is apparently a distinctive feature of the section) while sclereids are unique in *S. capsicoides*. These different types of epidermal cells could be related to fruit size and dehiscence type. Lageniform cells of large fruits would be the result of a tendency for the epidermal cells to become lignified and tall in response to external constraints, while epidermal sclereids in *S. capsicoides* could be related to the cracking of the fruit at the senescent stage.

The presence of a hypodermis, mainly constituted by collenchyma, is constant in the majority of the analysed species. However, there is a specific variation in the number of layers and the degree of lignification. The collenchymatous hypodermis is common in fruits with a thick outer skin, which is the case of many berries and drupes, such as some species of *Ribes*, *Berberis*, and

Paris (Roth, 1977) and even berries of some members of Solanaceae (Filippa and Bernardello, 1992; Valencia, 1985), and more precisely in *Solanum* (Dottori and Cosa, 1999; Klemt, 2007; Roth, 1977). The function of the hypodermal cells possibly comprises providing mechanical support, or, in some cases, participating in the dehiscence mechanism (Dyki et al., 1997; Klemt, 1997). The collenchyma is followed by a thick-walled parenchyma, but it is difficult to draw a line between the two tissue types, which is a well-known fact (Roth, 1977).

Some hypodermal cells, whose walls are impregnated with lignin, resemble the outline of a true collenchyma. Layers with such features are called here “sclerified collenchyma”. The amoeboid cells in the hypodermis of *S. palinacanthum* and *S. mammosum* are another case of a modified collenchyma. The sclerified collenchyma, as well as the strong thickened walls of the amoeboid cells, may be structures that make the fruits harder and more resistant to deformation, and perhaps are a defence against phytophagous insects. Indeed, fruit features are usually interpreted in relation to vertebrate dispersion and consumption, while insect and microbial

attacks, which are all the more important, have become neglected (Tewksbury, 2002).

Regarding the mesocarp, the presence of a spongy parenchyma is very obvious in some species. This tissue, characterized by large intercellular spaces and cells that change their shape from rounded to elliptical, elongate, and even stellate, was accurately described in *S. mammosum* (Miller, 1969) with the name of “aerenchyma”. Something similar occurs in the albedo of the orange, where parenchyma cells develop arms in different directions (Roth, 1977). We found that, apart from *S. mammosum*, the spongy fruited *Solanum* analysed here also present the so-called “aerenchyma”. The spongy tissue is not a very useful character to distinguish sections, since it is present in several species of sect. *Acanthophora*, but also in a species of section *Melongenina* (Chiarini and Barboza, 2007).

Regarding the pulp of the fleshy or juicy fruited *Solanum*, both the placenta and, especially the pericarp, contribute to form the pulp, a fact already reported for *Physalis peruviana* (Valencia, 1985) and other *Solaneae* (Filippa and Bernardello, 1992; Garcin, 1888; Murray, 1945). Instead, in *Solanum lycopersicum* (sub nom. *Lycopersicon esculentum*) only the placentas are responsible for the formation of the pulp (Murray, 1945; Roth, 1977), which is very peculiar. The disorganization of the inner mesocarp and the endocarp occurs in the same way in the fleshy and juicy fruits analysed, as in *P. peruviana* (Valencia, 1985).

The presence of a layer with a single prismatic crystal in each cell, observed here in *S. viarum*, has been found for several species of sect. *Melongenina* (Chiarini and Barboza, 2007; Dottori and Cosa, 2003). Deposition of calcium oxalate may have evolved as a primary mechanism for controlling the excess of calcium in many plants, providing multiple benefits to different organs (e.g. Franceschi and Horner, 1980; Sakai et al., 1972; Thurston, 1976; Webb, 1999). Nevertheless, the function of crystals in fruits remains unexplained to date.

Nee (1991) has discovered that different kinds of seeds can be found in species of the same section, probably in response to environmental pressures. On their part, Levin et al. (2005) discussed that, within sect. *Acanthophora*, species that share the same type of seed do not form a natural group. In addition, we demonstrate that one type of seed may not be related to any pericarp characteristic, since winged seeds, for instance, are present in species with spongy mesocarp (*S. capsicoides*, *S. platense*) as well as in species with another type of mesocarp (*S. atropurpureum*).

The most remarkable differences among fruits arise from the sexual system (Table 1). In fact, the relationship between andromonoecy and fruit size detected by Whalen (1984) and Whalen and Costich (1986) was also found here, since most of the analysed species have few

hermaphrodite flowers per inflorescence (strong andromonoecy) and grow large fruits. Within these species, size tends to increase through a proliferation of parenchymatic cells in the mesocarp. The increased size could be a way to accommodate more seeds per fruit (Nee, 1986), which is accompanied by changes in placentation patterns. In contrast, the weakly or non-andromonoecious species produce relatively smaller fruits. Outstandingly, large fruits show the lengthened lageniform cells, perhaps as a way to avoid deformation, while small fruits (*S. atropurpureum*, *S. tenuispinum* and *S. incarceratum*) present shorter epidermal cells.

Nevertheless, the relationship between fruit size or sexual system and the different tissue types appears not to be rigorously direct. For example, the spongy tissue is mainly found in many-seeded, large fruits, in species which are usually invasive herbs growing in ruderal environments, in open, sunny areas of the Chaco region, as *S. viarum* or *S. aenictum* (Matesevach, 2002; Whalen, 1984). Thus, the occurrence of these fruits is explainable through ecological and reproductive reasons rather than through phylogenetic relationships. However, the same spongy tissue is also found in *S. tenuispinum*, which has small fruits, and a different habit and habitat (this shrub is typical of rainforests, the Yungas of northern Argentina and southern Bolivia). Although the anatomical pattern of *S. tenuispinum* is similar to that of species of drier regions, the spongy tissue is less thick. The presence of such tissue in a non-andromonoecious species could be a rudimentary feature shared with its andromonoecious relatives.

Dispersion

Usually, fruits are classified into different dispersal syndromes according to their morphological characters. Van der Pijl's (1982) criterion is usually followed, but direct observation of the dispersion is seldom possible. As a consequence, the fruits or seeds are assigned to a dispersal syndrome on the basis of speculations, which leads to puzzling discussions, as Levin et al. (2005) pointed out. As mentioned before, there is an emphasis on the interpretation of fruit traits as adaptations to the interactions of vertebrates (Tewksbury, 2002), but our results indicate that other points of view should be regarded.

Most of the examined species produce a spongy fruit that never dehisces without the action of external forces, and whose placentas and seeds are not in contact at the senescent stage, i.e. a carcerulus according to Spjut (1994). For some species with such a kind of fruit (*S. viarum*, *S. aculeatissimum*, *S. mammosum*), Cipollini et al. (2002) suggested dispersion by large animals, like mammals. Nee (1991) proposed a “shaker” mechanism as the dispersal syndrome for the spongy fruits of two

other species, *S. capsicoides* and *S. platense*, since he observed a spontaneous rupture of the pericarp, which we could not corroborate. Indeed, no special features were detected in the fruit of *S. platense*, although epidermal sclereids in *S. capsicoides* could be related to the mentioned mechanism. The fact that the five mentioned species have potentially poisonous fruits (Cipollini and Levey, 1997), in addition to their spongy structure, turns dispersion by vertebrates little plausible. We suppose that all of these fruits with spongy tissue are adapted to dispersion by drain water after a rainstorm, the spongy tissue being an adaptation to flotation, as it has been previously suggested (Bryson and Byrd, 1994; Levin et al., 200; Nee, 1979, 1991).

The particularities of the fruit of *S. atropurpureum* should be mentioned. This is a unilocular fruit with a unique combination of red colour, a normal or not spongy and slightly juicy mesocarp (in contrast to its relatives), and winged seeds. Nee (1979, 1991) proposed epizoochory by birds as a dispersal mechanism for this species and for *S. acerifolium* Sendtn., a species with similar fruits, since their seeds do not resist the passage through the digestive system of birds. The intriguing combination of winged seeds and a showy, juicy fruit, can be thus solved.

Section *Acanthophora* could have several dispersal syndromes: hydrochory, epizoochory, endozoochory and the “shaker” mechanism. Thus, it has been proven that, despite the fact that their monophyly has already been confirmed by molecular studies (Levin et al., 2005, 2006), there are diverging pathways in fruit evolution within sect. *Acanthophora*.

Fruit traits and phylogeny

The similarities among fruits may not be synapomorphies, but they could be attributed to convergence in reproductive strategies. Cipollini et al. (2002), in a study that foregrounds phytochemical aspects, states that there is no significant correlation among the fruit types they distinguished and the phylogenetic lineages in *Solanum*. For these authors, fruit typology may be the consequence of physiological constraints, holding an independent evolution of the different dispersal syndromes. Regarding morpho-anatomy, our data lead to similar conclusions.

As mentioned before, an important factor influencing fruit features in *Solanum* subgen. *Leptostemonum* could be the andromonoecy. This sexual system is a convergent phenomenon, a homoplastic character that varies secondarily (Whalen, 1984) and appears and disappears within the clades or natural groups independently (Levin et al., 2006; Whalen and Costich, 1986). The big, many-seeded fruits would then be a collateral effect of the andromonoecy strategy, not a cause, since this sexual system is independent of the fruit anatomy or the seed

type: there are andromonoecious species with big, dry or juicy fruits, with winged or bulky seeds. Andromonoecious species tend to concentrate a high number of ovules in few hermaphrodite flowers (Bertin, 1982; Symon, 1979; Whalen and Costich, 1986); in consequence, they produce large fruits. Thus, the sexual system may have played a very important role in the *Leptostemonum* evolutionary lineages, since it affects not only the reproductive biology of its species (reproductive success, fitness, sexual allocation, etc.), but it also has an influence on the morpho-anatomy of fruits and their dispersal mechanism.

Phytochemical aspects are also very important in the evolution of *Solanum* fruits. For instance, in *Solanum* sect. *Solanum*, the concentration of steroidal glycoalkaloids decreases dramatically as the fruit matures (Carle, 1981; Cipollini et al., 2002), this being perhaps a mechanism to deter seed predators while the fruit is growing without deterring seed dispersers that feed on mature fruits. Nevertheless, in subgenus *Leptostemonum*, the concentration of toxic compounds in ripe fruits remains high (Cipollini and Levey, 1997). These authors suggest that the compounds perform an antifungal function, but that they also deter both predators and dispersers. This may be the case of *S. capsicoides*, *S. viarum*, and *S. mammosum* (Cipollini et al., 2002).

The results of our work suggest that phylogeny is not the main factor influencing the different carpologic characters found in subgen. *Leptostemonum*. We propose that, in many cases, a significant morphological variation is not associated with significant DNA sequence changes. Moreover, fruit traits seem to respond quickly to selection constraints on the dispersal syndromes. Molecular studies showed that some species, like *S. capsicoides* and *S. viarum*, are closely related as regards phylogeny (Levin et al., 2005), but differ notably regarding fruit traits. We tend to think that similarities and differences can be the result of the sexual system and of ecological and physiological conditions.

Acknowledgements

The authors thank Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET, Argentina), Agencia Córdoba Ciencia S. E. (Argentina), SECyT (UNC, Argentina), Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES, Brazil) and Myndel Botanica Foundation (Buenos Aires, Argentina) for financial support.

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