



Root tuberization patterns in Commelinaceae: a new anatomical approach

Ricardo Silva Batista Vita¹ · Nanuza Luiza de Menezes¹ · Marco Octávio de Oliveira Pellegrini² ·
Delmira da Costa Silva³ · Gladys Flávia de Albuquerque Melo-de-Pinna¹

Received: 15 July 2025 / Revised: 1 March 2026 / Accepted: 12 March 2026
© The Author(s) 2026

Abstract

Tuberous roots in monocots exhibit considerable variation in shape and size, yet their development and anatomical diversity remain poorly understood in Commelinaceae. This study investigates the morphological and anatomical diversity of tuberous roots in seven species representing three of the four major lineages in the family—*Palisota*, Commelineae, and Tradescantieae s.s.—with emphasis on the tuberization patterns. Tuberous root samples from *Palisota barteri*, *Commelina platyphylla*, *Dichorisandra leucophthalma*, *D. marantoides*, *D. radicalis*, *D. thyrsiflora*, and *Tradescantia pallida* were collected and fixed in FAA50 and embedded in both Historesin and Paraplast. We carried out morphometric analyses, as root diameter, cortical cells number (CCN), vascular parenchyma cell number (VPCN), and phloem area (PA), from transverse sections of tuberous roots, and we revealed two distinct morphological types of tuberous roots: the stalked shaped characterized by having two non-thickened extremities and a thickened median region and the sessile shaped, with thickening uniform along all the root. In general, the dermal system and cortical region on the thickened region of both stalked and sessile shaped roots, do not differ from what was observed in the non-thickened region. However, in the vascular system our results show four anatomical patterns: (1) In the first pattern the parenchyma of the centre of the vascular cylinder in the non-thickened region is not lignified, whereas, in the thickened region, it is composed of lignified cells; (2) In the second pattern, all parenchyma cells of the centre of the vascular cylinder are lignified, both in the non-thickened and in the thickened region; (3) In the third pattern the parenchyma of the centre of the vascular cylinder is composed of non-lignified cells in both regions (thickened and non-thickened); and (4) The last pattern reveals a fragmentation of the vascular system observed only in *Dichorisandra*, and the parenchyma of the centre of the vascular cylinder in the non-thickened region is lignified, whereas, in the thickened region, it is composed of expanded storage parenchyma. In the cortical region, a protective tissue was observed only in *Tradescantia pallida*, from subepidermal or outer cortical cells. Bifurcated root hairs were observed in *Commelina platyphylla*, *P. barteri* and *T. pallida*. With the new contributions to the understanding of the structural processes involved in root tuberization in Commelinaceae, we encourage studies using this approach in other Commelinoideae, and especially in other subfamilies as Cartonematoideae.

Keywords Monocot · Phellogen-like · Root anatomy · Vascular system

1 Introduction

Commelinaceae comprises ca. 41 genera and 731 species, being the largest and most widespread family of Commelinales (Christenhusz and Byng 2016), currently divided into two subfamilies, Cartonematoideae and Commelinoideae, which are supported by morphological and molecular data (Faden and Hunt 1991; Evans et al. 2003; Burns et al. 2011). Commelinoideae shows several delimitation problems, especially in tribe Tradescantieae, which was recovered as paraphyletic due to the uncertain positioning of the subtribe

✉ Gladys Flávia de Albuquerque Melo-de-Pinna
gfmppinna@usp.br

¹ Laboratório de Anatomia Vegetal, Departamento de Botânica, Instituto de Biociências, Universidade de São Paulo, São Paulo, Brazil

² University of the Witwatersrand, Johannesburg, South Africa

³ Laboratório de Anatomia Vegetal, Departamento de Ciências Biológicas, Universidade Estadual de Santa Cruz, Rodovia Jorge Amado, Km 16, Ilhéus, Bahia CEP 45662-900, Brazil

Palisotinae. *Palisota* Rchb. ex Endl., the single genus of Palisotinae, is either recovered as the sister-group of the remaining members of the subfamily Commelinoideae (Evans et al. 2003; Zuiderveen et al. 2011), as the sister-group of the tribe Commelineae (Burns et al. 2011), or in a polytomy along with Commelineae and Tradescantieae s.s. (Pellegrini et al. 2017, unpublished data). Thus, Commelinaceae is currently organized in four main lineages: Cartonematoideae, *Palisota*, Commelineae, and Tradescantieae s.s.

There is great diversity of underground storage structures in Commelinaceae that may be of cauline (e.g., corms, rhizomes) or radicular origin, like tuberous roots (Faden 1985, 1998; Hunt 2001). Tuberous roots are modified storage organs that usually accumulate water, carbohydrates, or proteins (Stevens 1911; Bresinsky et al. 2013; Appezzato-da-Glória 2015), and may originate from the axial root, distal expansion of lateral roots, proximal expansion of the taproot (Appezzato-da-Glória 2015), or from underground adventitious roots (Hagiladi et al. 1996). In Commelinaceae, tuberous roots are recorded in three of the four main lineages: *Palisota*, Commelineae (i.e., *Aneilema* R.Br. s.l., *Anthericopsis* Engl., *Commelina* L., *Dictyospermum* Wight, *Murdannia* Royle, and *Pseudoparis* H.Perrier), and Tradescantieae s.s. (i.e., *Callisia* Loeffl. s.l., *Cyanotis* D.Don s.l., *Dichorisandra* J.C.Mikan, *Geogenanthus* Ule, *Gibasis* Raf., *Gibasoides* D.R.Hunt, *Matudanthus* D.R.Hunt, *Siderasis* Raf. emend. M.Pell. & Faden, *Thyrsanthemum* Pichon, *Tradescantia* L. emend. M.Pell., *Tripogandra* Raf., and *Weldenia* Schult.f.) (Faden 1985, 1998; Hunt 2001; Pellegrini et al. 2017). Despite the occurrence of tuberous roots in Commelinaceae, its anatomical development is still little explored in the literature. An important study regarding root anatomy in 52 species, representing 19 genera of Commelinaceae was performed by Hofreiter and Tillich (2002), including tubers and no storage roots. According to the authors, a great diversity of character reveals a wide potential to systematic and phylogenetic approach in Commelinaceae. However, there are no studies that demonstrate the anatomical differences found in thickened and non-thickened regions in the two types of tuberous roots, sessile tuber and stalked tuber.

The development of tuberous roots is usually the result of the increase of parenchyma cells of the secondary vascular tissue from activity of the cambium in eudicotyledons (Appezzato-da-Glória 2015). On the other hand, in monocots, there is no vascular cambium and the tuberization process occurs mainly by the expansion of the parenchyma tissue in the cortex, and, in some cases, of the vascular system (Arber 1925; Hofreiter and Tillich 2002). The hypertrophy of the cortex may occur mainly by the activity of the endodermis, originating from a radiated region. This meristematic activity of the endodermis is reported by many authors in the roots of gymnosperms, monocots and eudicots (Williams 1947; Van Fleet 1961; Melo-de-Pinna

and Menezes 2002, 2003; Menezes et al. 2005). When the tuberous roots show increase of the vascular tissue, there is an expansion of the central parenchyma, with or without a change in the number of the protoxylem groups, as in *Triglochin procera* (Juncaginaceae) and *Hemerocallis fulva* (Xanthorrhoeaceae), respectively (Arber 1925).

In Commelinaceae, despite the extensive descriptive work on root anatomy carried out by Hofreiter and Tillich (2002), there are no studies on the tuberization process comparing different morphologies. Here we present a first anatomical study about the tuberisation process, including thickened and non-thickened regions, in two different types of tuberous roots (sessile tuber and stalked tuber), according to Faden and Hunt (1991).

2 Material and methods

We collected the underground system of three specimens, in the adult phase, from each one of the seven species. The samples were obtained at the campus of the Universidade Estadual de Santa Cruz (UESC), at the living collection of the Universidade de São Paulo (USP), at the Rio de Janeiro Botanical Garden (RJBG). Vouchers were deposited at the HUESC, SPF and RB herbaria (Table 1).

Tuberous roots, including the root apex, were fixed in FAA 50 (formaldehyde, acetic acid and ethanol 50%–Johansen 1940) for 48 h and then stored in 70%. With use of a digital caliper, we defined the thickened and non-thickened regions. Hand-free sections were stained with safranin 1% and Astra blue 1% (1:9) in ethanol 50% (Bukatsch 1972). Samples of root apex and some mature regions were dehydrated in an ethanol/tert-butanol series (50–100%), embedded in Paraplast following the methodology described by Ruzin (1999), and then, stained with Astra blue 1% in ethanol 50% and safranin 1% in ethanol 50% (Kraus et al. 1998, modified). Samples of the thickened

Table 1 List of the studied species and their vouchers

Taxon	Voucher
Subtribe Palisotinae	
<i>Palisota barteri</i> Hook.f	Pellegrini 499 (RB)
Tribe Commelineae	
<i>Commelina platyphylla</i> Klotzsch ex Seub	Pellegrini 479 (RB)
Tribe Tradescantieae, subtribe Dichorisandrinae	
<i>Dichorisandra leucophthalmus</i> Hook	HUESC 15732
<i>Dichorisandra marantoides</i> Aona & Faden	Pellegrini 492 (RB)
<i>Dichorisandra radicalis</i> Nees & Mart	HUESC 15733
<i>Dichorisandra thyrsoiflora</i> J.C.Mikan	RSBV 98 (SPF)
Tribe Tradescantieae, subtribe Tradescantiinae	
<i>Tradescantia pallida</i> (Rose) D.R.Hunt	HUESC 15737

region of the roots were embedded in Historresin (LeicaH) and stained in 0.05% Toluidine Blue O in 0.1 M of phosphate buffer at pH 6.8 (O'Brien et al. 1965). To detect callose in the sieve plates of the sieve elements in thickened region, samples were stained with aniline-blue and analyzed in the fluorescence microscope (UV), according to the methodology described by Smith and McCully (1978a, b). Transverse and longitudinal sections (5–10 µm) of the all embedded material were made using a rotary microtome Reichert-Jung Autocut 2040 and mounted in Entellan.

Analysis of occurrence and distribution of amyloplasts were under polarized light with the aid of the image digitization system IM50 attached to a Leica DMLB microscope.

The morphometrical parameters, as root diameter, cortical cells number (CCN), vascular parenchyma cell number (VPCN), and phloem area (PA), were obtained from transverse sections of tuberous roots by using the software ImageJ (Rasband 2013) with sampling value of $n=3$ for each analysis. Measurements of the phloem area were obtained by tracing the vascular system using image vectorization of serial transversal sections, including thickened and non-thickened regions. The amplitude between the non-thickened region (smaller diameter) and thickened region (bigger diameter) was defined as a percentage from the equation $D = (d_i - d_f) / d_i \cdot 100$, where D is the percentage of the diameter, d_i is the initial diameter of the non-tuberous region and d_f is the final diameter of the tuberous region. The results of the mean and standard deviation of each parameter were generated using the R-3.4.2 (<https://cran.r-project.org/>) and the illustrations were done in the Corel Draw software.

3 Results

3.1 Morphology of tuberous roots

Two morphological types were observed in the tuberous roots of the studied species: sessile and stalked tuber (Table 2). The stalked type is characterized by having two non-thickened extremities and a thickened median region (Fig. 1a–d). In the sessile type, the thickening is uniform along all the roots (Fig. 1e). The four studied species of *Dichorisandra* show stalked tuberous roots (Fig. 1a–c). Although the tuberous roots of *P. barteri* (Fig. 1d) also have been classified in this study as stalked, they present less prominent thickening when compared to tuberous roots in *Dichorisandra*. The other two studied species, *T. pallida* and *C. platyphylla* (Fig. 1e), present sessile tuber.

3.2 Root apical meristem of the tuberous roots

Transverse and longitudinal sections of the root apical meristem (RAM, Fig. 2) in both stalked and sessile tuberous

roots show a protostelic organization, and the endodermis as the inner boundary of the cortex. At a certain distance from the apical initials, the endodermis, in the meristematic phase, gives rise to the inner region of the cortex (Fig. 2b). The phloem and xylem strands are organized in separate units on the periphery of the vascular cylinder and the central region may be differentiated in vascular parenchyma cells, as in *Palisota barteri* (Fig. 2a–b) and *Dichorisandra radicalis* (Fig. 2c, d).

3.3 Dermal and cortical region of the mature roots

The dermal system in non-thickened region of both sessile and stalked tuberous roots, is composed by the uniseriate epidermis, followed by a hypodermis that may show one to two layers of cells (Fig. 3a–c). Root hairs were observed (Fig. 3c), which may be bifurcated in *Commelina platyphylla*, *P. barteri* and *Tradescantia pallida* (Fig. 3d). Sub-epidermal periclinal divisions were observed in *T. pallida* (Fig. 3c) resulting in three to four layers of cells. The cortex is differentiated into three regions: (1) external zone, composed by smaller isodiametric cells and reduced intercellular spaces; (2) median region, with bigger isodiametric cells and with intercellular spaces; and (3) internal region, with a radiated arrangement of cells from the endodermis (Fig. 3e–f). Some canals were observed with raphides (Fig. 4a). Idioblast containing starch (Fig. 4b) were observed in the cortex in all species. The endodermis cells deposit suberin lamellae in the transverse and inner tangential walls of the endodermis cell resulting in a U-shaped in all analyzed species (Fig. 4c–f), except in *C. platyphylla* and *T. pallida* (Fig. 4g–h).

In the thickened region of both sessile and stalked tuberous roots, the organization of the dermal system and cortex do not differ from what was observed in the non-thickened region, i.e., they have a uniseriate epidermis, a distinct uniseriate or biseriate hypodermis and a cortical organization in three zones (Fig. 5). However, a difference was observed in the endodermis cells of the thickened region that presents U-thickening only in *C. platyphylla* (Fig. 5a).

3.4 Vascular system of the mature roots

In thickened and no-thickened regions of the sessile and stalked tuberous roots, the pericycle is uniseriate (Fig. 4c–h), protostele polyarch (Figs. 5; 6) with 5 to 27 protoxylem strands and idioblasts containing starch in the parenchyma of the central cylinder in *Dichorisandra* spp. and *Tradescantia pallida*. In the other species, there is no starch in the vascular cylinder.

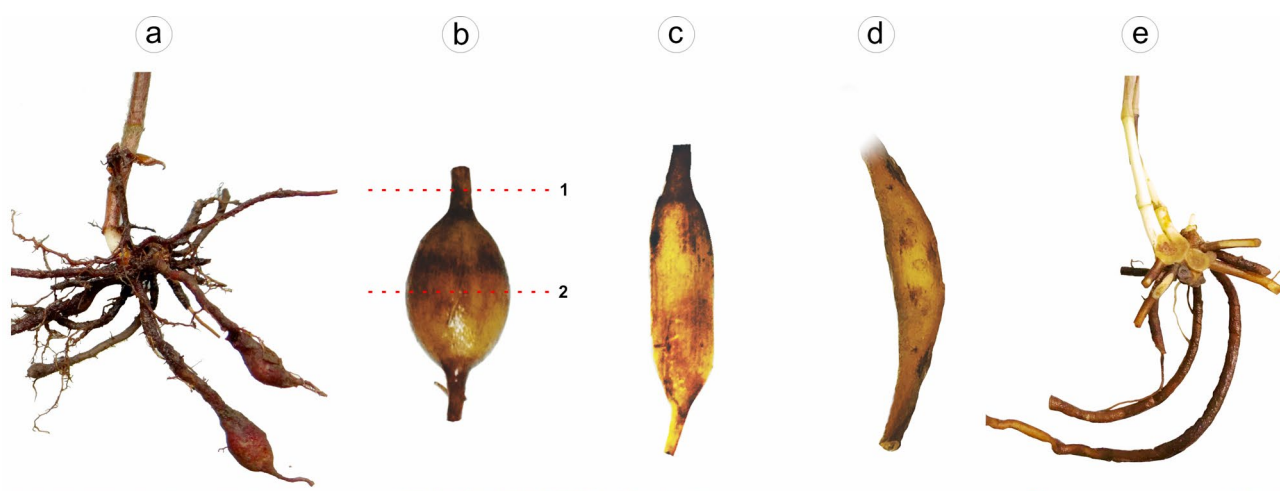
The centre of the vascular system, in both thickened and non-thickened regions, may be lignified as in *P. barteri* (stalked tuber, Figs. 5b; 6b) or parenchymatic as in *Tradescantia pallida* (sessile tuber, Figs. 5c; 6c). In *Commelina*

Table 2 Morphology and morphometrical parameters of the tuberous roots in the analyzed species. CC = Number of cortical cells; VPC = Number of vascular parenchyma cells; PA = Phloem area (μm^2)

	<i>Commelina platyphylla</i> (m $\pm$ sd)	<i>Palisota barteri</i> (m $\pm$ sd)	<i>Tradescantia pallida</i> (m $\pm$ sd)	<i>Dichorisandra leucophthalmos</i> (m $\pm$ sd)	<i>D. marantoides</i> (m $\pm$ sd)	<i>D. radialis</i> (m $\pm$ sd)	<i>D. thyrsiflora</i> (m $\pm$ sd)
Morphological types	Sessile	Stalked	Sessile	Stalked	Stalked	Stalked	Stalked
Vascular Pattern	Vascular Pattern 1	Vascular Pattern 2	Vascular Pattern 3	Vascular Pattern 4	Vascular Pattern 4	Vascular Pattern 4	Vascular Pattern 4
Diameter (mm) of the thickened region	3.46 $\pm$ 0.6	7.25 $\pm$ 0.61	5.13 $\pm$ 0.45	6.63 $\pm$ 3.06	12.68 $\pm$ 2.21	5.69 $\pm$ 3.13	9.95 $\pm$ 2.62
Diameter (mm) of the non-thickened region	2.01 $\pm$ 0.24	2.79 $\pm$ 0.61	2.66 $\pm$ 0.39	1.27 $\pm$ 0.49	1.46 $\pm$ 0.51	1.12 $\pm$ 0.23	1.75 $\pm$ 0.58
Amplitude (%)	72.13 $\pm$ 48.48	159.85 $\pm$ 55.02	92.85 $\pm$ 33.66	422.04 $\pm$ 577.9	768.5 $\pm$ 162.56	408.03 $\pm$ 419.59	468.6 $\pm$ 115.67
Diameter of the vascular cylinder (mm) of the thickened region	0.52 $\pm$ 0.10	0.91 $\pm$ 0.02	0.96 $\pm$ 0.10	3.70 $\pm$ 2.65	7.61 $\pm$ 1.73	3.18 $\pm$ 2.06	6.39 $\pm$ 2.18
Diameter of the vascular cylinder (mm) of the non-thickened region	0.39 $\pm$ 0.01	0.80 $\pm$ 0.06	0.51 $\pm$ 0.03	0.63 $\pm$ 0.28	0.45 $\pm$ 0.12	0.27 $\pm$ 0.03	0.55 $\pm$ 0.34
Amplitude (%)	33.34 $\pm$ 24.48	13.75 $\pm$ 5.58	88.24 $\pm$ 30.52	487.30 $\pm$ 170.73	1591.1 $\pm$ 119.73	1077.8 $\pm$ 801.82	1061.8 $\pm$ 801.27
CC of the thickened region	4253.67 $\pm$ 298.96	3025.67 $\pm$ 175.05	3293 $\pm$ 25.24	3968.33 $\pm$ 2844.08	8570.67 $\pm$ 3603.14	2728.33 $\pm$ 2598.88	4276.33 $\pm$ 1319.31
CC of the non-thickened region	3079 $\pm$ 695.55	2234 $\pm$ 418.76	1205.33 $\pm$ 193.56	1071.33 $\pm$ 358.7	1472.33 $\pm$ 705.66	563.33 $\pm$ 61.17	1548 $\pm$ 1214.8
VPC of the thickened region	267 $\pm$ 123.16	713.33 $\pm$ 193.54	425.67 $\pm$ 209.5	1416.33 $\pm$ 1689.34	2717.67 $\pm$ 2606.13	524.33 $\pm$ 576.52	1618.67 $\pm$ 923.76

Table 2 (continued)

	<i>Commelina platyphylla</i> (m ± sd)	<i>Palisota barteri</i> (m ± sd)	<i>Tradescantia pallida</i> (m ± sd)	<i>Dichorisandra leucophthalmos</i> (m ± sd)	<i>D. marantoides</i> (m ± sd)	<i>D. radicalis</i> (m ± sd)	<i>D. thyrsiflora</i> (m ± sd)
VPC of the non-thickened region	214.33 ± 124.36	910.67 ± 173.08	165 ± 88.39	212.33 ± 85.42	317.33 ± 150.38	115.67 ± 35.73	421 ± 356.58
PA of the thickened region	18,215.16 ± 11,205.34	43,773.99 ± 4900.17	47,321.37 ± 3997.4	47,060.36 ± 41,693.91	70,253.78 ± 30,661.85	35,295.76 ± 20,220.95	60,173.88 ± 27,253.12
PA of the non-thickened region	8473.29 ± 3468.47	33,388.6 ± 7704.95	22,357.7 ± 3354.50	4761.17 ± 1330.65	9659.39 ± 3203.05	5185.29 ± 1155.18	18,942.1 ± 12,478.46

**Fig. 1** Morphology of tuberous roots of the stalked (**a**, **b**, **c**, **d**) and of the sessile (**e**) types. **a** *Dichorisandra radicalis*. **b** *Dichorisandra thyrsiflora*. Non-thickened region (1) and thickened region (2). **c** *Dichorisandra leucophthalmos*. **d** *Palisota barteri*. **e** *Commelina platyphylla*

platyphylla (sessile tuber), lignification occurs only in the thickened region (Fig. 6a), when compared with the non-thickened region, which is parenchymatic (Fig. 5a). In the four species of *Dichorisandra*, the parenchymatic central region is wide (Fig. 6d–f), different from the observed in the non-thickened region where the parenchyma is lignified (Fig. 5d–f).

In the species with sessile tuberous roots and in *P. barteri* (stalked tuberous root), the increase in the diameter of the vascular cylinder did not alter the organization of the vascular system, maintaining the alternate pattern of phloem and xylem strands (Fig. 6a–c). On the other hand, in the four species of *Dichorisandra* with stalked roots, there is a fragmentation of the vascular tissues with the increase of the vascular cylinder (Fig. 6d–f). Only in *D. marantoides*, it was observed phloem invagination in the direction of the center of the vascular cylinder (Fig. 7a–d). In the remaining species of *Dichorisandra*, the phloem strands are fragmented

and scattered around the vascular system, adjacent to the pericycle, as observed in *D. radicalis* (Fig. 8) and *D. leucophthalmos* (Fig. 9).

From our results it is possible to identify four anatomical patterns of the vascular cylinder (Figs. 10, Table 2; S1). In the first pattern observed only in *C. platyphylla* (Figs. 10a; 11a–b), the vascular cylinder parenchyma of the non-thickened region is not lignified, whereas, in the thickened region, it is composed of lignified cells. In the second pattern, found only in *P. barteri* (Figs. 10b; S1i–j), all parenchyma cells of the vascular cylinder are lignified, both in the non-thickened region and in the thickened region. In the third pattern observed only in *T. pallida* (Figs. 10c; S1c–d), the vascular cylinder parenchyma is composed of non-lignified cells in both regions (thickened and non-thickened). Finally, in the last pattern observed only in the four species of *Dichorisandra* (Figs. 10d; S1e–f; S1k–n), the center of the vascular cylinder is occupied for lignified parenchyma cells in the

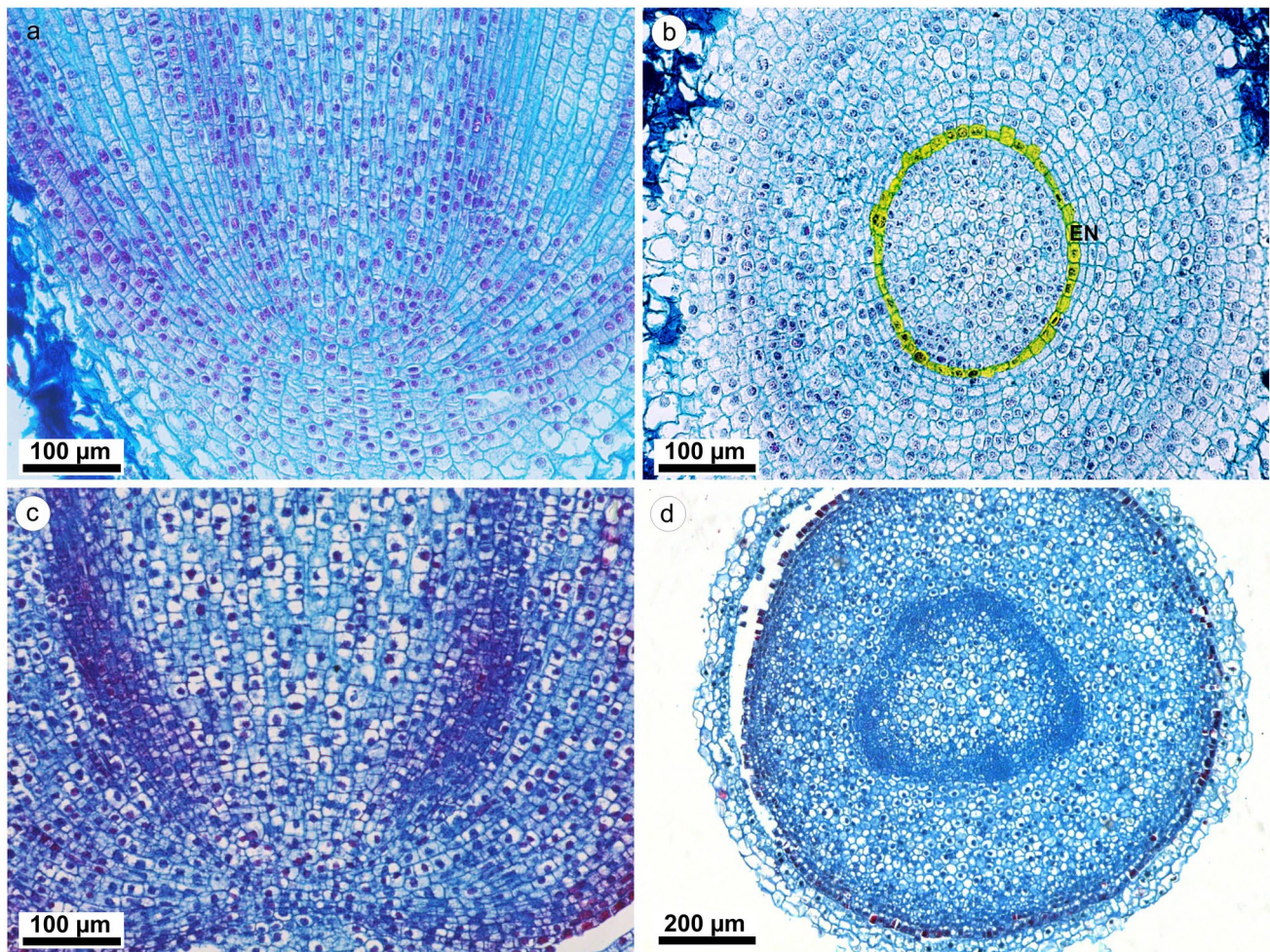


Fig. 2 Root apex of *Palisota barteri* (a, b) and *Dichorisandra radicalis* (c, d). **A.** Transverse section showing the root apical meristem with procambial cells in the center of the vascular system. In figure b, it is possible to observe an endodermis (En) in the meristematic phase (highlighted in yellow) originating the innermost region of the cortex. **c.** Longitudinal section of the root apical meristem. In figure d, note differentiation of the parenchyma vascular cells in the central region

non-thickened region and parenchyma not lignified in the thickened region. Only in this pattern, occurs a peculiar vascular architecture due to the fragmentation of the vascular system.

3.5 Morphometrical parameters of the thickened and non-thickened regions

The morphometric parameters observed in the seven analyzed species were summarized in Table 2. Comparing the diameter of the non-thickened regions among the seven species, a small variation was verified in the total diameter, like in *D. radicalis* (1.12 ± 0.23 mm) and *P. barteri* (2.79 ± 0.61 mm), and in the vascular cylinder diameter (*D. radicalis* 0.27 ± 0.03 mm/*P. barteri* 0.80 ± 0.06 mm). In the thickened regions, there is more variation, the largest

diameter recorded for *D. marantoides* (12.68 ± 2.21 mm) and the smaller diameter in *C. platyphylla* (3.46 ± 0.6 mm).

Dichorisandra species show a larger variation between the diameters of thickened and non-thickened roots, as *D. marantoides* (1.46 ± 0.51 mm/ 12.68 ± 2.21 mm), with around 768% of thickening (“swelling”), while *C. platyphylla* has the smaller increase (2.01 ± 0.24 mm/ 3.46 ± 0.6 mm) around 75% of thickening. The increase in the number of cortical parenchyma cells (CC) and of the vascular cylinder (VPC) is more expressive in stalked tuberous roots. *Dichorisandra marantoides* presents the bigger variation of CC ($1472.33 \pm 705.66/8570.67 \pm 3603.14$), an increase of around 482%, and the bigger variation of diameter of the vascular cylinder (0.45 ± 0.12 mm/ 7.61 ± 1.73 mm), of about 1591%.

The increase in the number of parenchyma cells is not directly proportional to the increase of the root diameter. While *D. marantoides* had an increase of 768% in the total

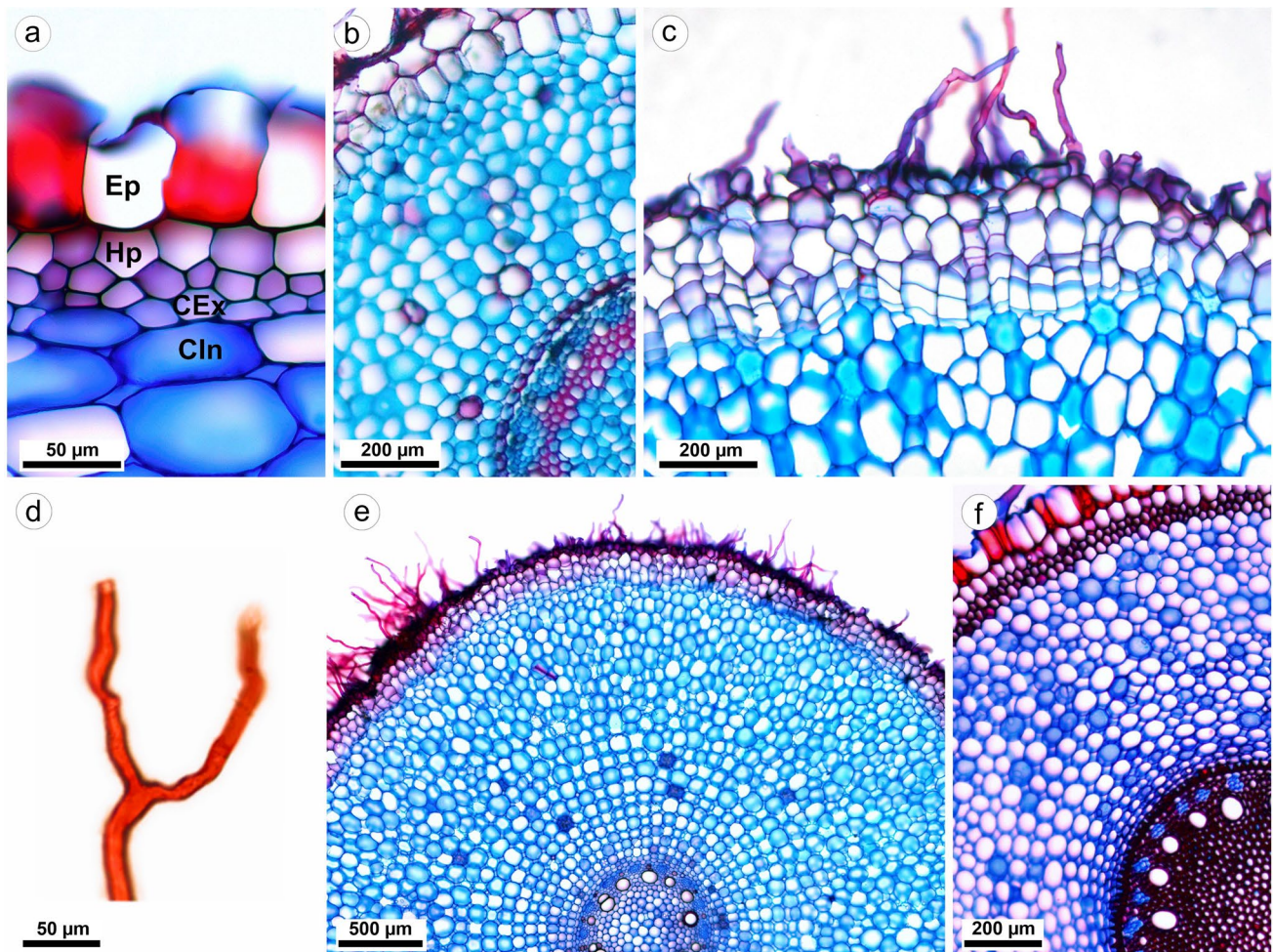


Fig. 3 Transverse sections of the non-thickened region and thickened region in tuberous roots. **a** *Palisota barteri* with uniseriate layers of the epidermis (Ep) and of the hypodermis (Hp). **b** *Dichorisandra marantoides*. **c** *Tradescantia pallida* showing a region of dividing cells (Storied Cork). **d** Details of bifurcated root hair in *Palisota barteri*. **e** *Tradescantia pallida* showing a radiated inner cortex. **f** *Palisota barteri* showing a lignified central cylinder. CEx, external cortex; CIn, internal cortex

diameter of the root, the number of CC increased 482,10%. This relation was also verified between the increase of vascular cylinder diameter (1591.1%), and the number of parenchyma cells of the vascular cylinder (756.41%) and the phloem area (627.31%), indicating that the variation of the cells number is not the main factor of thickening in the stalked tuberous root.

An increase of the phloem area was observed in all species, although sessile tuberous roots presented smaller variation (e.g., 111.65% in *T. pallida* and 114.97% in *C. platyphylla*), while stalked tuberous roots presented a bigger increase (e.g., 888.42% in *D. leucophthalmus*). *Palisota barteri*, although is considered stalked, presents a smaller variation in the vascular cylinder diameter (13.75%) and in the phloem area (31.1%). This same species presents smaller variations of CC (35.43%) and negative values for VPC (−31.1%).

In sessile tuberous roots, was observed small variations of morphometrical parameters. In *C. platyphylla*, variations of total diameter (72.13% of increase) and diameter of the vascular cylinder (33.3% of increase) were close of the found values for CC (38.15% of increase), VPC (24.57% of increase) and phloem area (114.97% of increase). When compared with *C. platyphylla*, small variations were found for *T. pallida*, except for CC (173.2%) and VPCN (157.98%).

In general, all species present an increase of the vascular cylinder, where the smaller variation was recorded for *P. barteri* (0.80 ± 0.06 mm to 0.91 ± 0.02 mm), an increase of 13.75%, followed by sessile species. The largest variations were found in the remaining species with stalked tuberous roots, as in *D. marantoides* (0.45 ± 0.12 mm/ 7.61 ± 1.73 m), which had an increase of 1591.1% (Table 2).

Despite *T. pallida* show the largest increase of vascular cylinder diameter (88.24%) and VPC (157.98%) when

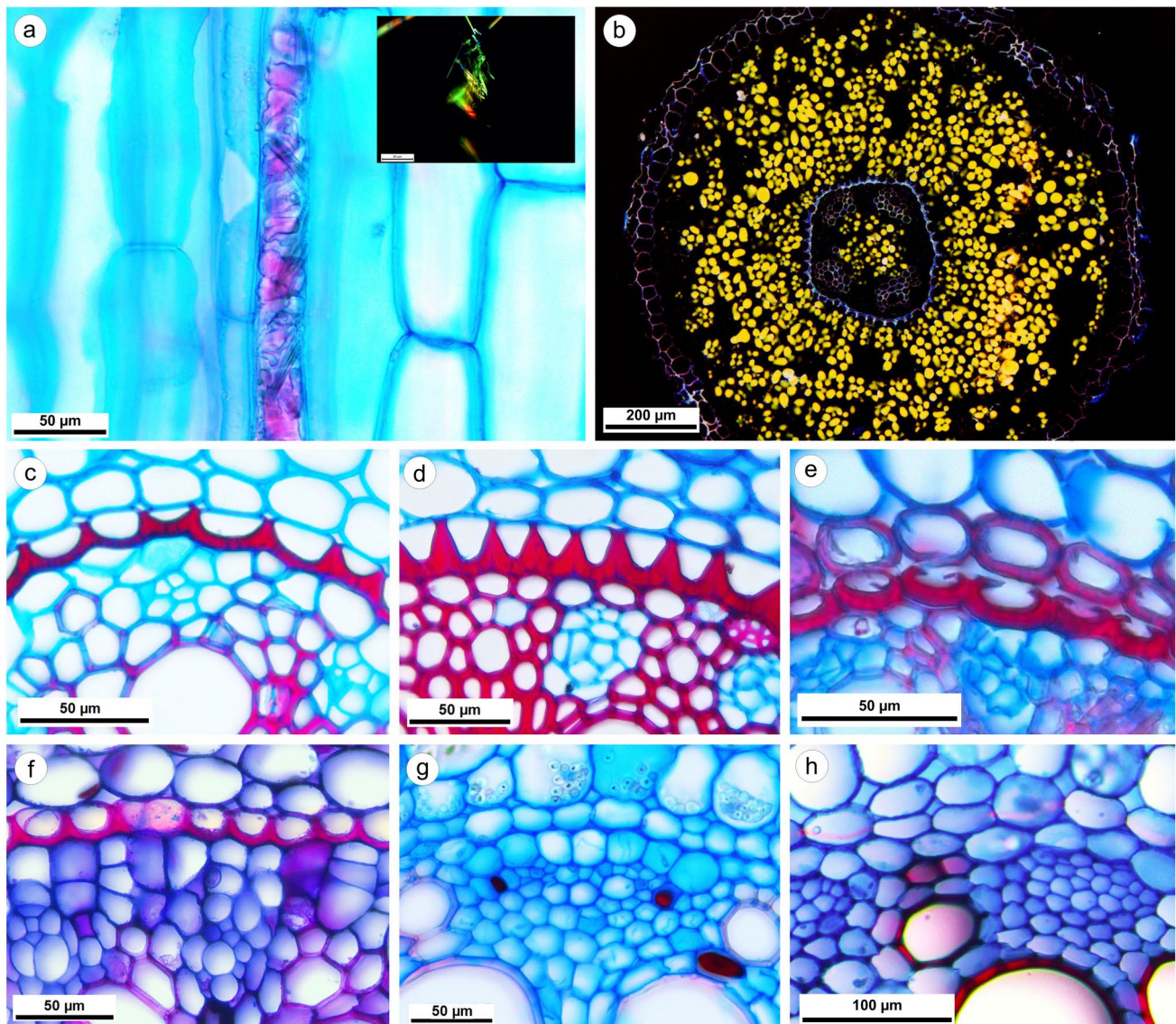


Fig. 4 Anatomical features of the cortical region. **a** Longitudinal section of *Commelina platyphylla* in the non-thickened region of the tuberous root indicating a canal containing raphides. Detail presenting raphides under polarized light. **b** *Dichorisandra radicalis* showing the distribution of starch in the non-thickened region of the tuberous root under polarized light. **c–f** Details of the inner region of the cortex showing the endodermis and pericycle in the non-thickened region of the tuberous root of *Dichorisandra radicalis* (**c**), *Palisota barteri* (**d**), *Dichorisandra marantoides* (**e**), *Dichorisandra thyrsiflora* (**f**), *Commelina platyphylla* (**g**), and *Tradescantia pallida* (**h**)

compared with *C. platyphylla*, that presents smaller increase of vascular cylinder diameter (33.3%) and VPC (24.57%), there is no expressive variation between phloem areas of both species, that are 111.65% and 114.97%, respectively.

4 Discussion

In addition to previous evidence that root anatomy in Commelinaceae provides characters of potential systematic value (Hofreiter and Tillich 2002), the detailed study of tuberous roots—based on comparative analyses of thickened and

non-thickened regions across distinct morphological types—enabled the identification of anatomical features not previously reported for the family.

Examination of the cortical region revealed tangential cell divisions in the subepidermal or outer cortex of *T. pallida*. Similar divisions have been described in Dioscoreaceae, Araceae, and Asparagaceae (Bucherer 1889; Priestley and Woffenden 1922; Jura-Morawiec et al. 2015). In eudicots, periderm may originate from epidermal, subepidermal, or pericyclic tissues, or from secondary phloem, forming phellogen, phelloderm, and phellem (Bary 1877; Höhnelt 1878; Beeckman and Eshel 2024). Although often considered

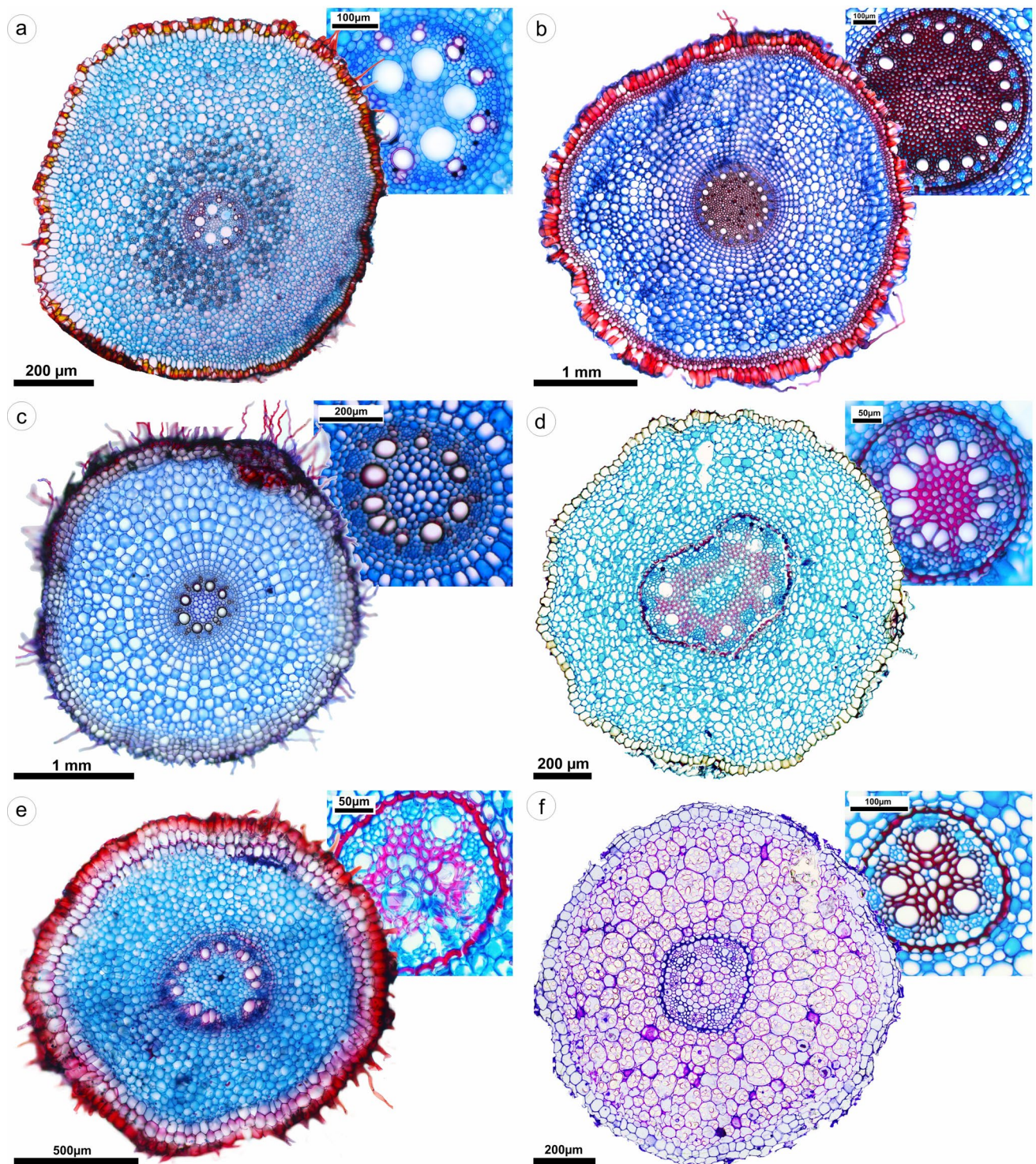


Fig. 5 Transverse sections of the non-thickened region in tuberous roots. *Commelina platyphylla* (a) and *Palisota barteri* (b). Details of the vascular cylinder with lignified parenchyma and endodermis with suberin lamellae. c *Tradescantia pallida*. The detail is a bigger magnification of the vascular cylinder indicating the absence of cell wall secondary thickening in the endodermis and in the parenchyma. d *Dichorisandra marantoides*, showing the vascular cylinder in a non-thickened region before the beginning of the thickening process. e *Dichorisandra leucophthalmus*, showing the vascular cylinder with lignified parenchyma. f *Dichorisandra radicalis*, showing the vascular cylinder with lignified parenchyma

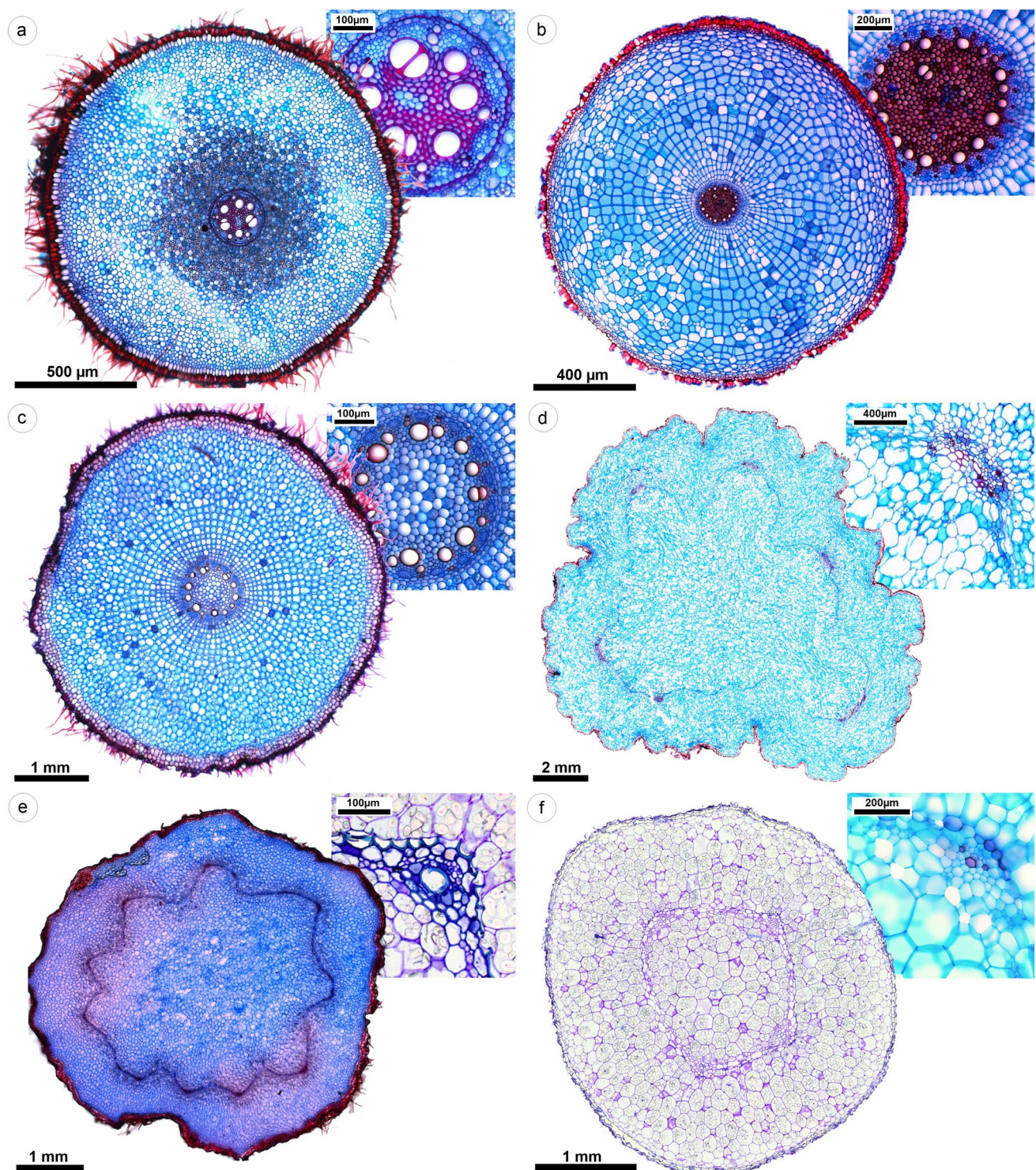


Fig. 6 Transverse sections in the thickened region of the tuberous roots. **a** *Commelina platyphylla*, showing in detail the lignified vascular cylinder in the central region. **b** *Palisota barteri*, with lignified parenchyma in the central cylinder. **c** *Tradescantia pallida* indicating the absence of lignification in the central region of the vascular cylinder. **d–f**. *Dichorisandra* species, showing an increase of the central region of the vascular cylinder and a detail of one of their six poles of xylem in *Dichorisandra marantoides* (**d**), *D. leucophthalmus* (**e**) and *D. radicalis* (**f**)

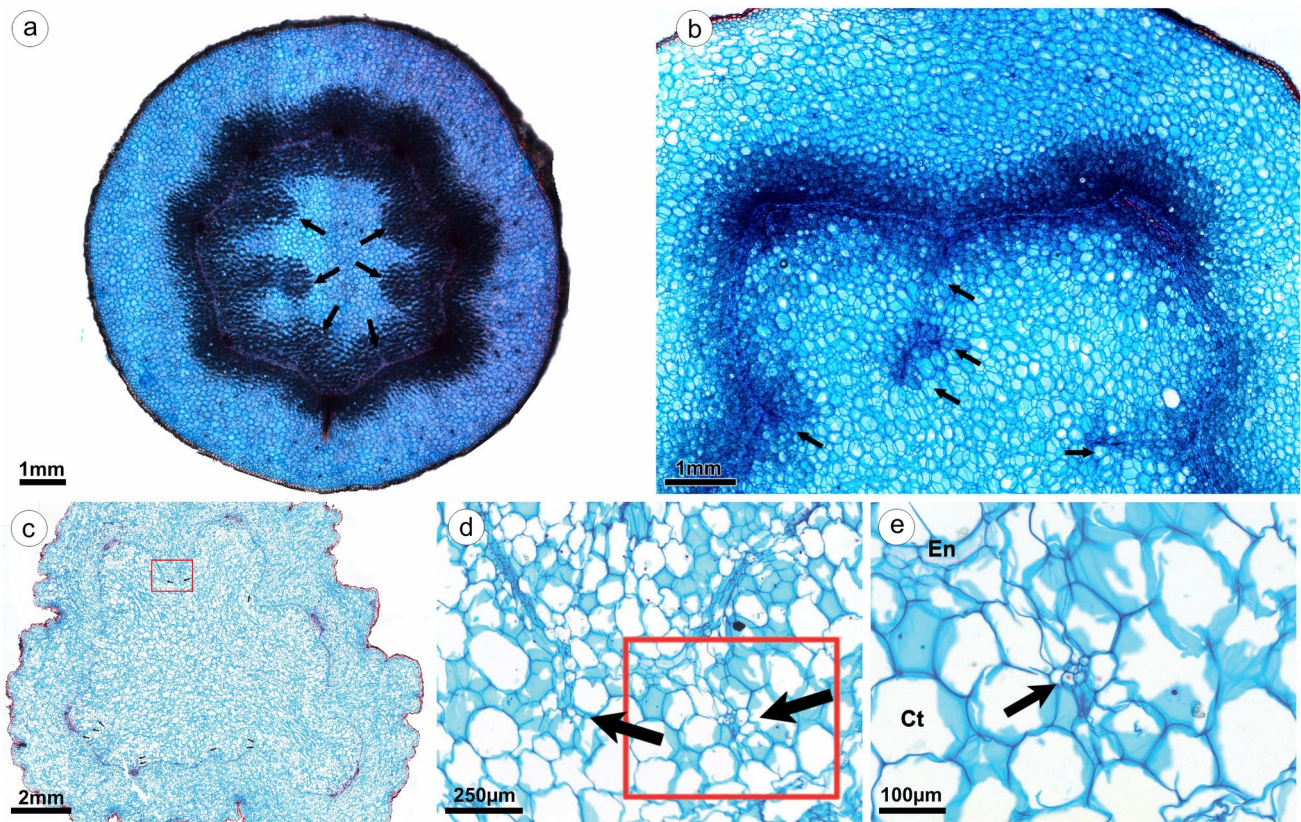


Fig. 7 Transverse sections of the thickened region of *Dichorisandra marantoides* indicating phloem invaginations (arrows). **d–e** The detail is a bigger magnification of the selected region in the figure **C**. En, endodermis; Ct, cortex

absent in monocots, periderm or periderm-like tissues have long been reported (Bary 1877; Scott and Brebner 1893; Schoute 1902; Arber 1925), especially in roots and rhizomes. Secondary protective tissues occur in several families, including Araceae, Arecaceae, Asparagaceae, Bromeliaceae, Dioscoreaceae, Gramineae, Juncaceae, Liliaceae, Musaceae, Pandanaceae, Strelitziaceae, Zingiberaceae, and Typhaceae (Engler and Krause 1908; Eames and MacDaniels 1947; Esau 1960, 1965; Fahn 1982; Evert and Eichhorn 2006). Terminology remains inconsistent among authors, encompassing *phellogenetic meristem*, *etagenkambium*, *storied cork*, and *cork cambium* (Bucherer 1889; Schoute 1902; Spratt 1920; Trockenbrodt 1990).

According to Eames and MacDaniels (1947), most monocots do not develop a true periderm, though “primary initials” may give rise to storied cortical layers, as originally termed *Etagen kork* (Beijer 1927). Other authors adopted equivalent terms (Bary 1877; Krauss 1949; Esau 1960; Tenório et al. 2012). While some consider these divisions non-phellogenetic (Evert and Eichhorn 2006; Steeves and Sawhney 2017), others describe cortical mother-cell zones (*Rinden Mutterzellen*) (Roseler 1889) or relate them to endodermal modifications (Schoute 1902; Priestley and Woffenden 1922). In *T. pallida*, the presence of subepidermal

division zones supports the interpretation of a storied cork sensu Schoute (1902). The tissue lacks radial continuity, indicating a temporary meristematic zone typical of monocots. Whether *T. pallida* forms a persistent periderm derived from an active phellogen, as reported for Dioscoreaceae (Lawton and Lawton 1969), remains to be determined.

Regarding tuberous root morphology, Hofreiter and Tillich (2002) reported two storage-root types—spindle-shaped and tuber-shaped—though without explicit definitions. Based on their illustrations, the spindle-shaped type likely corresponds to the sessile type of Faden and Hunt (1991), whereas the tuber-shaped type corresponds to the stalked type. This interpretation aligns the present findings with those of Hofreiter and Tillich (2002) for *Dichorisandra*. However, for *Palisota barteri*, we classify the species as stalked, whereas Hofreiter and Tillich (2002) described it as sessile. The discrepancy may reflect differences in the degree of cortical expansion, which constitutes the main storage parenchyma and may drive the stalked morphology observed here.

In monocots, storage parenchyma generally arises from hypertrophy of cortical or vascular tissues, as documented for *Roscoeia cautleoides* (Zingiberaceae), *Triglochin procerum* (Juncaginaceae), and *Dioscorea discolor* (Dioscoreaceae)

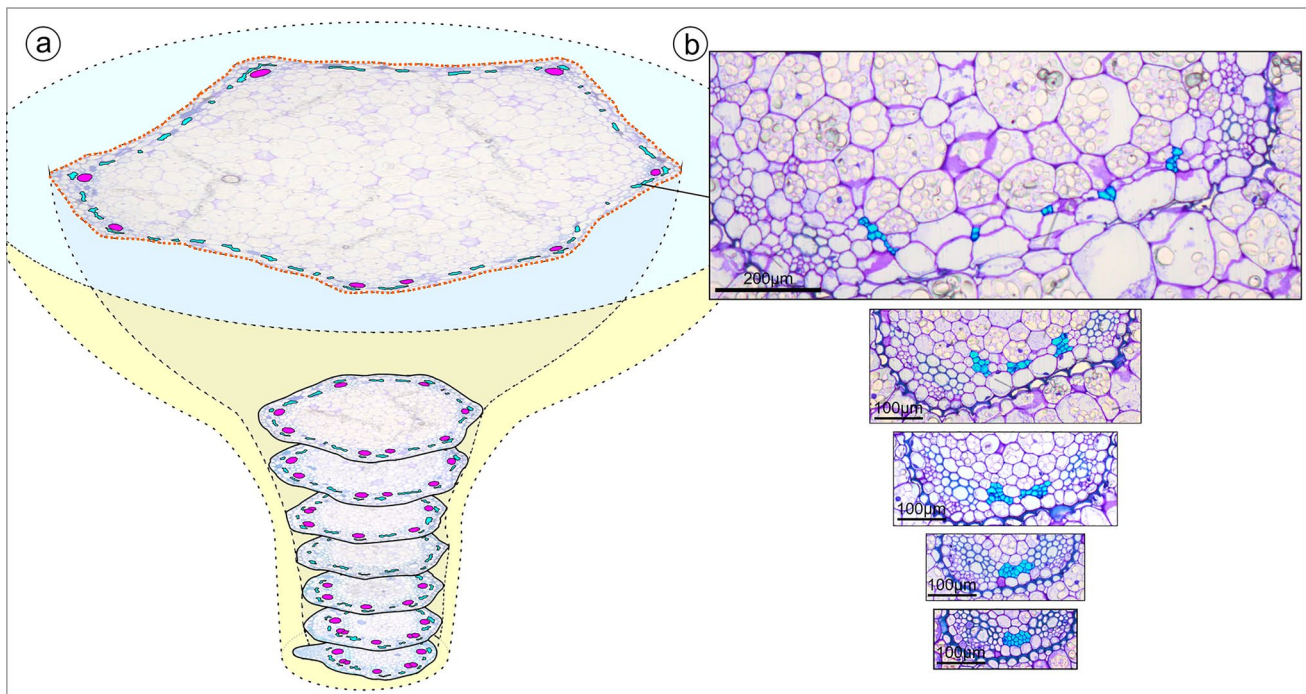
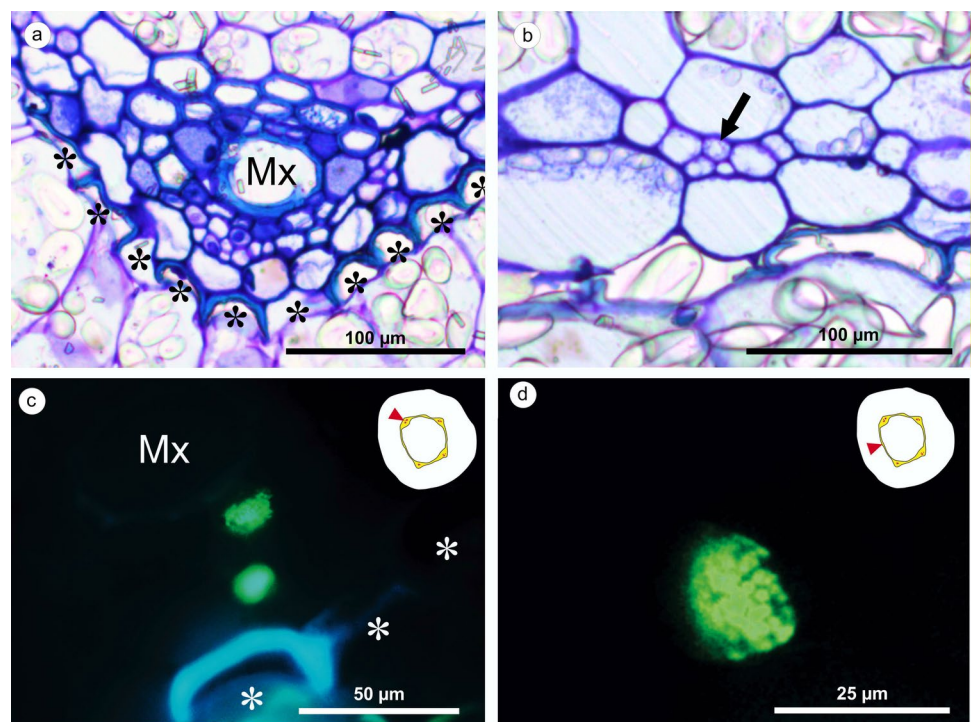


Fig. 8 Tracing of the vascular system in the tuberous root of *Dichorisandra radicalis* by using image vectorization, indicating the localization of xylem and phloem elements after the fragmentation of the vascular system (a). b Cell tracking method combined with serial transverse sections showing the localization of phloem elements (blue) during fragmentation (or scattering) of primary phloem strands as expansion (tuberization) of the vascular system occurs

Fig. 9 Transverse sections of the thickened region of the tuberous root of *Dichorisandra leucophthalmus* indicating sieve plate in the phloem cells. a Detail of one xylem pole showing the endodermis (asterisks) as boundary layer between cortex and vascular system. b Small region of phloem between two xylem poles. c–d Detection of sieve plates stained with Aniline Blue. Details indicate the position of callose plates. c Phloem in front of the metaxylem (Mx). d Phloem adjacent to the pericycle in an area away from the metaxylem. The sieve plate indicated in this figure is the same cell indicated (black arrow) in Fig. 9b. Details indicate the relative position of the phloem in the transverse section (red arrowhead). Mx, metaxylem; Asterisk, endodermis



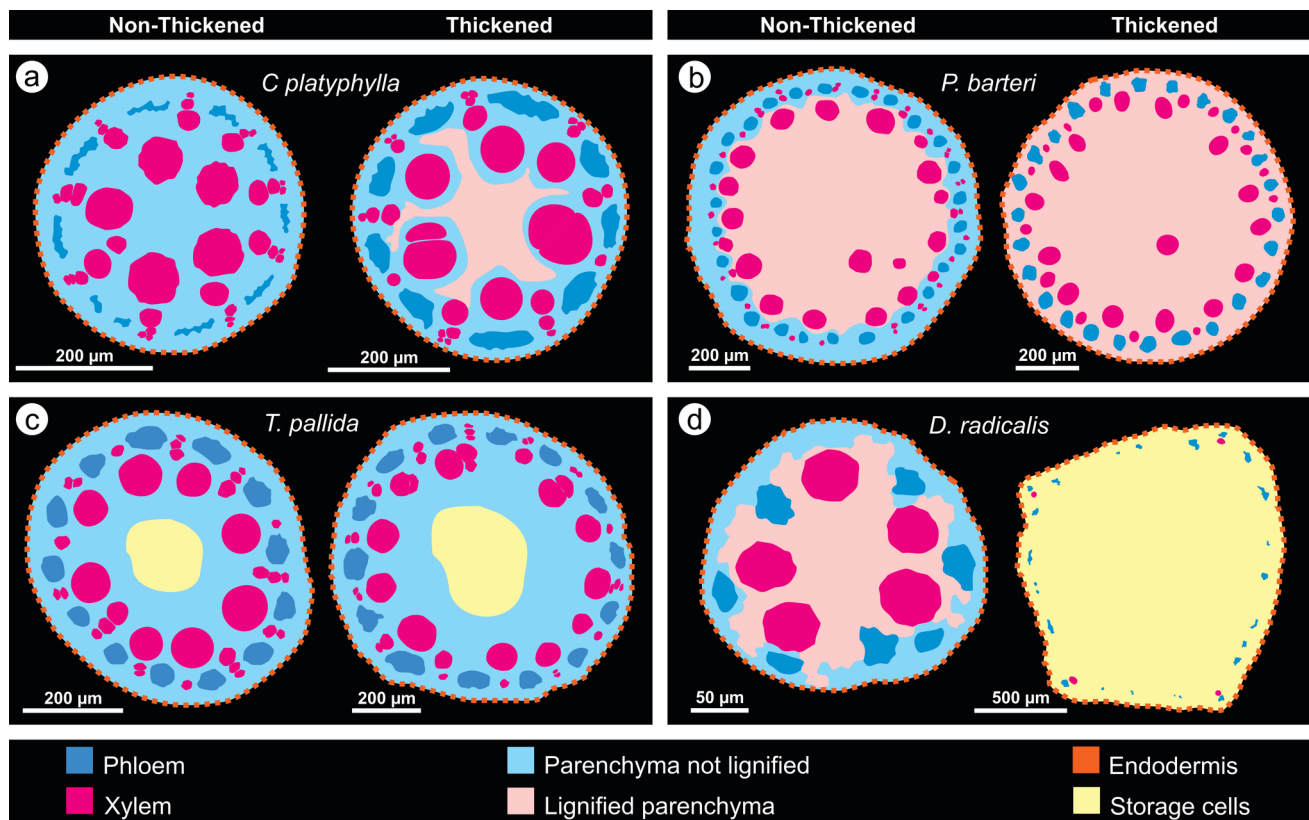


Fig. 10 Schematic representation of the four patterns of vascular architecture of *Commelina platyphylla* (a), *Palisota barteri* (b), *Tradescantia pallida* (c), and *Dichorisandra radicalis* (d)

(Arber 1925). Among the species analyzed, expansion of both cortical and vascular parenchyma occurred only in *Dichorisandra*. In *D. marantoides*, the vascular cylinder diameter increased by 1591.1%, but the rise in vascular parenchymatic cell number (756.41%) and phloem area (627.31%) alone does not account for this increase, indicating that both cortical and vascular expansion contribute to thickening of stalked tuberous roots.

Lignified cells were present in the central vascular region of both thickened and non-thickened root portions, except in *T. pallida*. In *Dichorisandra*, lignification predominated in non-thickened regions, whereas in *C. platyphylla* and *P. barteri* it predominated in thickened regions. Extending these observations to the study of Hofreiter and Tillich (2002) suggests that vascular-center lignification occurs in both tuberous and non-tuberous roots in Commelinaceae. Although some authors associate reduced lignification with enhanced lateral-root production and starch accumulation (Koshimizu and Nishida 1949; Khan et al. 2016), this pattern does not fully explain the variability observed here, since tuberization

and lateral-root formation also occur in species with lignified central parenchyma, such as *P. barteri*.

By contrast, fragmentation of vascular architecture—specifically xylem–phloem arrangement—was observed only in *Dichorisandra*, a pattern also documented by Hofreiter and Tillich (2002) for *D. acaulis* and *D. reginae*. Although preliminary, this vascular configuration may hold taxonomic value and should be investigated further in additional species of the genus.

In conclusion, tuberous roots in Commelinaceae exhibit distinct anatomical patterns related to starch storage, lignification of the vascular center, and expansion of cortical and vascular parenchyma. These features may vary along a single root, but the overall vascular architecture remains stable despite parenchymal expansion. The structural insights provided here contribute to the understanding of root tuberization in Commelinaceae and highlight the need for similar studies in other Commelinoideae and in additional subfamilies, particularly Cartonematoideae.

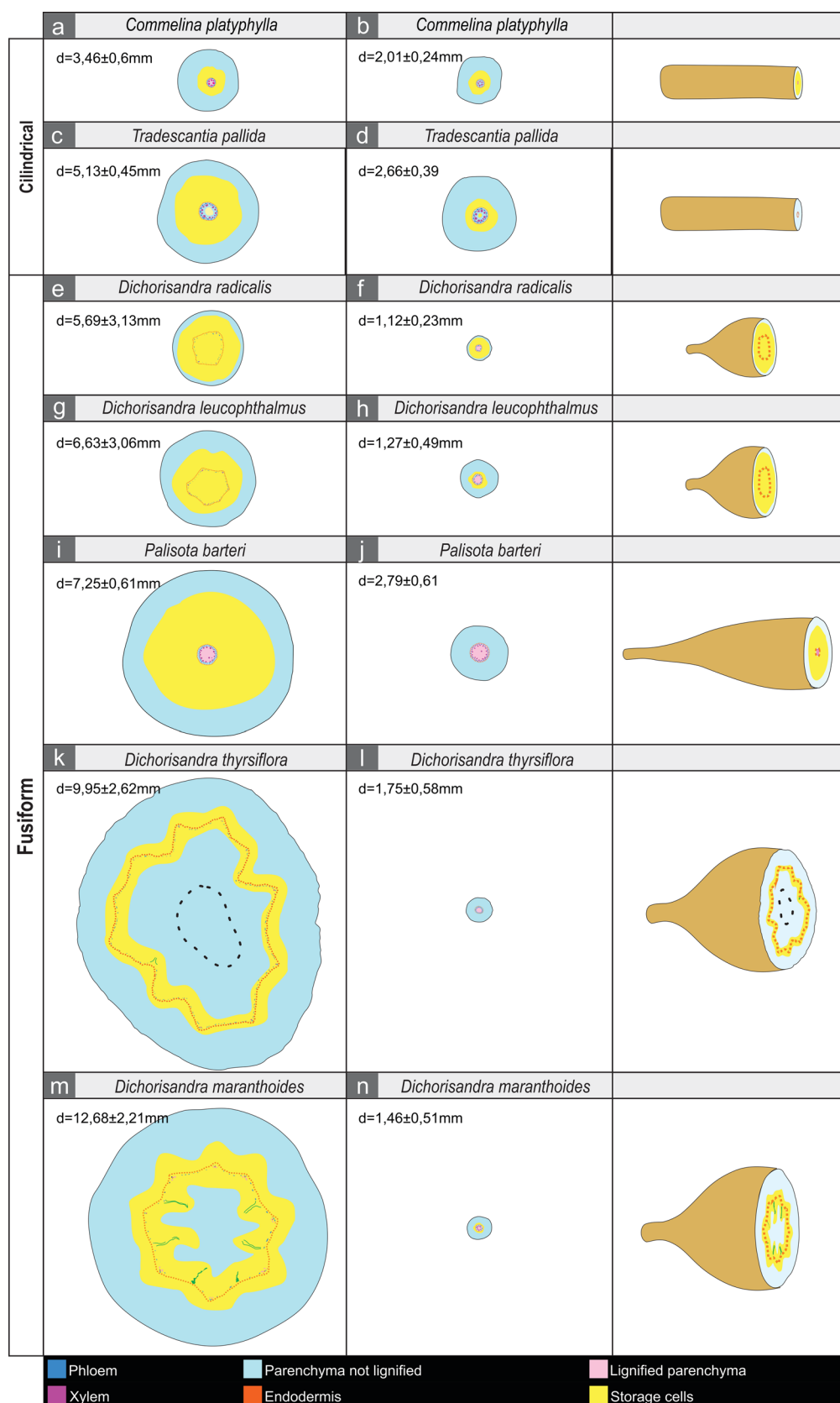


Fig. 11 Schematic representation of the relationship between regions of bigger and smaller tissues distribution, and their root morphology

Acknowledgements The authors thank the editors of BJB and anonymous reviewers for suggestions.

Author contributions RSBV and GFMP conducted anatomical analysis. All authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Funding The Article Processing Charge (APC) for the publication of this research was funded by the Coordenação de Aperfeiçoamento de Pessoal de Nível Superior - Brasil (CAPES) (ROR identifier: 00x0ma614). This work was supported by Fundação de Amparo à Pesquisa do Estado de São Paulo (Grants 2015/15158-4); Conselho Nacional de Desenvolvimento Científico e Tecnológico (CNPq) for productivity Grants to GFAMP and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior (CAPES) for Grants to MOOP and RSBV.

Data availability All data is available within the article or its supplementary material.

Declarations

Conflict of interest The authors declare that they have no conflict of interest.

Open Access This article is licensed under a Creative Commons Attribution 4.0 International License, which permits use, sharing, adaptation, distribution and reproduction in any medium or format, as long as you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons licence, and indicate if changes were made. The images or other third party material in this article are included in the article's Creative Commons licence, unless indicated otherwise in a credit line to the material. If material is not included in the article's Creative Commons licence and your intended use is not permitted by statutory regulation or exceeds the permitted use, you will need to obtain permission directly from the copyright holder. To view a copy of this licence, visit <http://creativecommons.org/licenses/by/4.0/>.

References

- Appezato-da-Glória B (2015) Morfologia de sistemas subterrâneos de plantas. 3i Editora, São Paulo
- Arber A (1925) Monocotyledons: a morphological study. Cambridge University Press, Cambridge
- Bary A (1877) Vergleichende Anatomie der Vegetationsorgane der Phanerogamen und Farne, vol 3. W. Engelmann, Leipzig
- Beeckman T, Eshel A (eds) (2024) Plant roots: the hidden half. CRC Press, Boca Raton
- Beijer JJ (1927) Die Vermehrung der radialen Reihen im Cambium. Rec Trav Bot Neerl 24:631–786
- Bresinsky A, Körner C, Kadereit JW, Neuhaus G, Sonnawal U (2013) Strasburger's plant sciences: including prokaryotes and fungi, vol 1. Springer, Heidelberg
- Bucherer E (1889) Beiträge zur Morphologie und Anatomie der Dioscoreaceen. T. Fischer, Kassel
- Bukatsch F (1972) Bemerkungen zur Doppelfärbung Astrablau-Safranin. Mikrokosmos 61:255
- Burns JH, Faden RB, Stepan SJ (2011) Phylogenetic studies in the Commelinaceae subfamily Commelinoideae inferred from nuclear ribosomal and chloroplast DNA sequences. Syst Bot 36:268–276. <https://doi.org/10.1600/036364411X569471>
- Christenhusz MJ, Byng JW (2016) The number of known plants species in the world and its annual increase. Phytotaxa 261:201–217. <https://doi.org/10.11646/phytotaxa.261.3.1>
- Eames AJ, MacDaniels LH (1947) An introduction to plant anatomy, 2nd edn. McGraw-Hill, New York
- Engler A, Krause K (1908) Araceae–Monsteroideae. Das Pflanzenreich, vol 37. Wilhelm Engelmann, Leipzig
- Esau K (1960) Anatomy of seed plants. John Wiley & Sons, New York
- Esau K (1965) Plant anatomy, 2nd edn. John Wiley & Sons, New York
- Evans TM, Sytsma KJ, Faden RB, Givnish TJ (2003) Phylogenetic relationships in the Commelinaceae: II. A cladistic analysis of *rbcL* sequences and morphology. Syst Bot 28:270–292. <https://doi.org/10.1043/0363-6445-28.2.270>
- Evert RF, Eichhorn SE (2006) Esau's plant anatomy: meristems, cells, and tissues of the plant body: their structure, function, and development, 3rd edn. John Wiley & Sons, Hoboken
- Faden RB (1985) Superorder Commeliniflorae. In: Dahlgren RMT, Clifford HT, Yeo PF (eds) The families of the monocotyledons. Springer, Berlin, pp 374–460
- Faden RB (1998) Commelinaceae. In: Kubitzki K (ed) *The families and genera of vascular plants*. Vol. IV. Flowering plants. Monocotyledons. Alismatanae and Commelinanae (except Gramineae). Springer, Berlin, pp 109–128
- Faden RB, Hunt DR (1991) The classification of the Commelinaceae. Taxon 40:19–31
- Fahn A (1982) Plant anatomy, 3rd edn. Pergamon Press, Oxford
- Hagiladi A, Yang XH, Umiel N, Gilad Z (1996) *Curcuma alismatifolia*. I. Plant morphology and the effect of tuberous root number on flowering date and yield of inflorescences. In: Proceedings of the VII international symposium on flowerbulbs 430, pp 747–754. <https://doi.org/10.17660/ActaHortic.1997.430.119>
- Hofreiter A, Tillich HJ (2002) Root anatomy of the Commelinaceae (Monocotyledoneae). Feddes Repert 113:231–255. [https://doi.org/10.1002/1522-239X\(200208\)113:3/4%3c231::AID-FEDR231%3e3.0.CO;2-2](https://doi.org/10.1002/1522-239X(200208)113:3/4%3c231::AID-FEDR231%3e3.0.CO;2-2)
- Von Höhnelt FR (1878) Über den Kork und verkorkte Gewebe überhaupt. Vienna
- Hunt DR (2001) Commelinaceae. In: Egli U (ed) Illustrated handbook of succulent plants: monocotyledons. Springer, Berlin, pp 247–254
- Johansen DA (1940) Plant microtechnique. McGraw-Hill Book Company, New York
- Jura-Morawiec J, Tulik M, Iqbal M (2015) Lateral meristems responsible for secondary growth of the monocotyledons: a survey of the state of the art. Bot Rev 81:150–161. <https://doi.org/10.1007/s12229-015-9152-8>
- Khan MA, Gemenet DC, Villordon A (2016) Root system architecture and abiotic stress tolerance: current knowledge in root and tuber crops. Front Plant Sci 7:1584. <https://doi.org/10.3389/fpls.2016.01584>
- Koshimizu T, Nishida M (1949) On the relation between the distribution of free auxin in the young sweet potato plant and its root tuber formation. Bot Mag (Tokyo) 62:735–736. <https://doi.org/10.15281/jplantres1887.62.146>
- Krauss BH (1949) Anatomy of the vegetative organs of the pineapple *Ananas comosus* (L.) Merr. III. The root and the cork. Bot Gaz 110:550–587. <https://doi.org/10.1086/335540>
- Kraus J E, Sousa H C, Rezende M H, Castro N M, Vecchi C, Luque R (1998) Astra blue and basic fuchsin double staining of plant materials. Biotech Histochem 73: 235–243
- Lawton JR, Lawton JRS (1969) The development of the tuber in seedlings of five species of *Dioscorea* from Nigeria. Bot J Linn Soc 62:223–232. <https://doi.org/10.1111/j.1095-8339.1969.tb01966.x>
- Melo-de-Pinna GFA, Menezes NL (2002) Vegetative organ anatomy of *Ianthopappus corymbosus* Roque & Hind (Asteraceae–Mutisieae). Braz J Bot 25:505–514. <https://doi.org/10.1590/S0100-84042002012000014>
- Melo-de-Pinna GFA, Menezes NL (2003) Meristematic endodermis and secretory structures in adventitious roots of *Richtera*

- Kuntze (Mutisieae–Asteraceae). *Braz J Bot* 26:1–10. <https://doi.org/10.1590/S0100-84042003000100002>
- Menezes NL et al (2005) Meristematic activity of the endodermis and the pericycle in the primary thickening in monocotyledons: considerations on the “PTM.” *An Acad Bras Cienc* 77:259–274. <https://doi.org/10.1590/S0001-37652005000200006>
- O'Brien T P, Feder N, Mc Cull M E (1965) Polychromatic staining of plant cell walls by toluidine blue O. *Protoplasma* 59: 364–373
- Pellegrini MO, Forzza RC, Sakuragui CM (2017) Novelty in Brazilian *Tradescantia* L. (Commelinaceae). *Phytokeys* 80:1–31. <https://doi.org/10.3897/phytokeys.80.12232>
- Priestley JH, Woffenden LM (1922) Physiological studies in plant anatomy. V. causal factors in cork formation. *New Phytol* 21:252–268. <https://doi.org/10.1111/j.1469-8137.1922.tb07603.x>
- Rasband WS (2013) ImageJ. Bethesda: US national institutes of health. Accessed 27 Nov 2017. Available at: <http://imagej.nih.gov/ij>
- Roseler P (1889) Das Dickenwachstum und die Entwicklungsgeschichte der sekundären Gefäßbündel bei den baumartigen Lilien. *Jahrb Wiss Bot* 20:292–348
- Ruzin SE (1999) Plant microtechnique and microscopy. Oxford University Press, New York
- Schoute JC (1902) Über Zellteilungsvorgänge im Cambium. J. Müller, Jena
- Scott DH, Brebner G (1893) On the secondary tissues in certain monocotyledons. *Ann Bot* 7:21–62. <https://doi.org/10.1093/aob/os-7.1.21>
- Smith MM, McCully ME (1978a) A critical evaluation of the specificity of aniline blue-induced fluorescence. *Protoplasma* 95:229–254. <https://doi.org/10.1007/BF01294453>
- Smith MM, McCully ME (1978b) Enhancing aniline blue fluorescent staining of the cell wall structures. *Stain Technol* 53:79–85. <https://doi.org/10.3109/10520297809111446>
- Spratt AV (1920) Some anomalies in monocotyledonous roots. *Ann Bot* 34:99–105. <https://doi.org/10.1093/aob/os-34.1.99>
- Steeves TA, Sawhney VK (2017) Essentials of developmental plant anatomy. Oxford University Press, Oxford
- Stevens WC (1911) Plant anatomy from the standpoint of the development and functions of the tissues, and handbook of microtechnique. P. Blakiston's Son, Philadelphia
- Tenório V, Sakuragui CM, Vieira RC (2012) Stem anatomy of *Philodendron* Schott (Araceae) and its contribution to the systematics of the genus. *Plant Syst Evol* 298:1337–1347. <https://doi.org/10.1007/s00606-012-0640-6>
- Trockenbrodt M (1990) Survey and discussion of the terminology used in bark anatomy. *IAWA J* 11:141–166. <https://doi.org/10.1163/22941932-90000511>
- Van Fleet DS (1961) Histochemistry and function of the endodermis. *Bot Rev* 27:165–220. <https://doi.org/10.1007/BF02860082>
- Williams BC (1947) The structure of the meristematic root tip and origin of the primary tissues in the roots of vascular plants. *Am J Bot* 34:455–462. <https://doi.org/10.1002/j.1537-2197.1947.tb13017.x>
- Zuidervleen GH, Evans TM, Faden RB (2011) A phylogenetic analysis of the African plant genus *Palisota* (Commelinaceae) based on chloroplast DNA sequences. In: Grand Valley State University Honors Projects 65

Publisher's Note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.