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Stem Anatomy and Nodal Vasculature of Some Egyptian *Vicia* Species (Faboideae-Fabaceae)

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Abstract: Stem anatomy and nodal vasculature are studied in seven *Vicia* species. The stem anatomy shows the presence of 30 characters proved to be taxonomically useful, of these: outline of cross section, number of its sides, presence of brown tanniniferous cells in the epidermis, presence of cortical fibre strands, number of vascular bundles in the ring and pith cell shape. Only the epidermis of *V. sativa* is characterized by the presence of some brown tanniniferous cells and the cortex of *V. faba* is characterized by the presence of four cortical fibre strands, two above the two polar bundles and two above the two cortical bundles in the wings. At the beginning of nodal vasculature, the number of the vascular bundles increases from ten to fourteen in *V. sativa*, *V. ervilia*, *V. monantha* and *V. villosa*, from nine to thirteen in *V. peregrina*, from nine to twelve in *V. narbonensis*, or from 18-20 in *V. faba*. The studied *Vicia* species are characterized by an unusual type of stele in which the two cortical bundles divide each into two bundles; one supplies the stipules and the other enter the leaf base. The median petiole trace divides into three leaving the main stele and passes out to the petiole. Each lateral branch of this median trace divides again and the outermost bundle fuses with the incoming lateral petiole trace from cortical bundles supply. The five bundles in the petiole compose of the lower most three are entirely median in derivation, while the two uppermost bundles are composite. Thus the lateral leaf traces are derived from the two cortical bundles of the lower internode below the insertion of their leaf, beside to the two lateral bundles deriving from the median leaf trace and the stipules are supplied by the two cortical vascular bundles only and not by the main vascular ring. A complete replacement occur for the two cortical bundles, they substitute at each node by two new bundles originating from the two lateral bundles of the main ring opposite to them supplying the stipules of the next node. Axillary bud is supplied from the two lateral bundles on either side of the median leaf gap.

Key words: Fabaceae, Faboideae, nodal vasculature, stem anatomy, *Vicia*

INTRODUCTION

The genus *Vicia* L., belongs to tribe Vicieae (Faboideae-Fabaceae) and comprises 140 species of erect, climbing or scrambling herbs. It is mainly distributed in N temperate Europe, extending to S America, Hawaii and tropical E Africa (Kupicha, 1981; Mabberley, 1997). Täckholm (1974) recognized 12 *Vicia* species and Boulos (1999) enumerated 14 species of *Vicia* occurring in Egypt.

Metcalfe and Chalk (1950) mentioned that the stems of *Lathyrus* and *Vicia* have special features, stems of *Lathyrus* species are flattened on one side providing two wings each containing a vascular bundle. Rudall (2000) found that the vasculature of the internodal regions of the stem is arranged in a cylinder of separate or fused collateral bundles in *Vicia faba* and some stems have cortical or medullary bundles, which may be associated with the leaf vasculature.

The nodes distinguished from the internodes by the departure of their traces (Pant and Mehra, 1964), which is usually marked in the main vascular cylinder by the formation of one or more gaps. They may be unilacunar, trilacunar or multilacunar depending on the number of gaps (Metcalfe and Chalk, 1979). Hegde and Tilak (1982) described the anatomy of the seedling of fourteen genera of Leguminosae and all the genera showed a three-trace, trilacunar pattern at the third node, while the seedling anatomy of *Vicia faba* cv. T-44 at the third node presented a single gap in the vascular ring of each cotyledon by Gupta and Murty (1986). Fotedar (1987) examined the vasculature of the leaf in *Juglans regia* and found that the node is multitrace trilacunar. Furthermore, Minu and Murty (1989) studied the nodal vasculature in

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Leucaena leucocephala and the first foliar node was three traced, trilacunar with two intrapetiolar stipules and the axillary bud is supplied by the vascular tissue on either side of the median leaf gap.

Kupicha (1975) recorded observations on the vascular anatomy of the tribe Vicieae and mentioned that the tribe Vicieae is characterized by an unusual type of stele in which the lateral leaf-traces are present as cortical bundles in the internode. The nodal anatomy is often characteristic of families and therefore taxonomically significant, particularly the number and arrangement of the leaf traces and gaps (Metcalfe and Chalk, 1979; Rudall, 2000). Gupta and Murty (1986) recorded that there are two cortical bundles originating from the cotyledonary traces and transverse up to the third node and the cortical bundles formed from the vascular ring in the nodes above the third node traversing and supplying the stipules of the leaves.

The present study deals with the stem anatomy and nodal vasculature in seven *Vicia* species for showing the characters of taxonomic values of the stems and understanding the behaviour of the bundles of the wings and main ring in the stipules and petiole vasculature.

MATERIALS AND METHODS

Seven *Vicia* species were collected from their natural habitats in Egypt as weeds of cultivation, especially *Triticum* species, taxa classification, names and places of collection are given in Table 1. For anatomical studies, fresh stems and fifth node and internode were fixed in FAA solution, then transferred to 70% ethanol and dehydrated through a graded ethanol series. Samples were then embedded in paraffin wax M.P. at 56-58°C (Johansen, 1940). Using a rotary microtome, serial sections (12 μ) were obtained and double stained by safranin and light green (1% solution in 50% ethanol) according to Willey (1971). They were then dehydrated through an alcohol series and mounted in Canada balsam. These sections were done and photographed using a Zeiss photomicroscope at Agricultural Botany Department, Faculty of Agriculture, Suez Canal University. Where the same anatomical characters are shared by more than one species, only one photograph was presented.

RESULTS AND DISCUSSION

Stem anatomy (Table 2 and Fig. 1 and 2a): The outline of cross section of stem is ±rhombic in *V. narbonensis*, *V. sativa* and *V. ervilia*, ovate to rhombic in *V. peregrina* and *V. monantha*, or ± quadrate to rhombic in *V. faba* and *V. villosa*. The stem has four sides with two wings in *V. faba* and *V. narbonensis* (Fig. 1a and b), six sides (two sides right and 4 sides left direction) in *V. sativa* and *V. ervilia* (Fig. 1d and e), or six sides (two sides left and 4 sides right direction) *V. peregrina*, *V. monantha* and *V. villosa* (Fig. 1c, f and 2a). The maximum value of average thickness of stem at wings and polar bundles are (3200 and 4000 μ) in *V. faba*, while the minimum ones (1507 and 1524 μ) are found in *V. ervilia* and *V. villosa*, respectively. In addition, the highest wing length and width are observed in *V. faba* (760×520 μ) and the lowest ones are present in *V. ervilia* and *V. villosa* (302 and 222 μ, respectively).

The epidermal cells are semi-rectangular to semicircular with thin cuticle, the maximum value of their average thickness is (60 μ) in *V. faba* (Fig. 1a) and their lowest value (24 μ) is noticed in *V. ervilia* (Fig. 1e). All the epidermal cells are homogenous (all their cells with equal sizes) in some of the studied species, or heterogenous (some cells larger than the other cells) in *V. narbonensis*, *V. sativa* and *V. villosa*. Only the epidermis of *V. sativa* is characterized by the presence of some brown tanniniferous cells (Fig. 1d). This is in agreement with Metcalfe and Chalk (1950) who mentioned the presence of these cells in the epidermis of *Lathyrus pratense*.

The outer cortex is formed of angular collenchyma cells in 4-7 layers, while 5-10 layers at the four poles and inner cortex comprises large isodiametric cells. The maximum values of average thickness of collenchyma and parenchyma tissues of cortex region (75 and 280 μ) were found in *V. peregrina* and *V. faba*, respectively, while, the minimum ones (40 and 79 μ)are observed in *V. faba* and *V. villosa*, respectively. Moreover, The highest value of average thickness of wing collenchyma tissue is (400 μ) in *V. faba* and the lowest ones is (75 μ) in *V. sativa*. These cortical characters are similar with the data which obtained by Hassan (1992) in some *Trigonella* species.

Table 1: Classification and collection data of the studied *Vicia* species (Classification based on Kupicha, 1976; Maxted, 1993)

Subgenus	Section	Species	Place of collection	Date of collection
Vicia	Faba	*V. faba* L. cv. Giza 2	Ismailia, Fanara	15.3.2004
		V. narbonensis L.	Dessuk, Aboe Ghoneina	22.3.2004
	Peregrinae	*V. peregrina* L.	El Sharkia, Menia El Kameh, El Saadiin	26.3.2004
Vicilla	Vicia	*V. sativa* L.	El Arish, Airport protected area	20.3.2004
	Ervilia	*V. ervilia* (L.) Willd.	East El Arish, Wadi Hareidin	03.4.2004
	Cracca	*V. monantha* Retz.	El Arish, Airport protected area	20.3.2004
		V. villosa Roth	Agricultural museum garden, Dokki	05.4.2004

Table 2: Stem anatomical characters of the seven examined *Vicia* species

Species recording data	*V. faba*	*V. narbonensis*	*V. peregrina*	*V. sativa*	*V. ervilia*	*V. monantha*	*V. villosa*
Outline of cross section	Quadrate to rhombic	Rhombic	Ovate to rhombic	±Rhombic	±Rhombic	Ovate to rhombic	±Quadrate to rhombic
No. of cross section sides	4	4	6	6	6	6	6
Average thickness of cross section at wings (μ)	3200	2400	1725	2100	1507	1700	1555
Average thickness of cross section at polar bundles (μ)	4000	2675	1725	2400	1555	1800	1524
Average wing length and width (μ)	760×520	500×375	375×325	375×375	302×286	400×375	365×222
Average thickness of epidermis (μ)	60	38	25	38	24	50	32
Size of epidermal cells	Homogenous	Heterogenous	Homogenous	Heterogenous	Homogenous	Homogenous	Heterogenous
Presence brown tanniniferous cells in epidermis	-	-	-	+	-	-	-
Average thickness of collenchyma (μ)	40	50	75	63	48	63	48
Average thickness of wing collenchyma (μ)	400	175	125	75	111	150	95
Average thickness of parenchyma (μ)	280	150	125	150	111	150	79
Presence of two winged cortical fibre strands	+	-	-	-	-	-	-
Presence of two polar cortical fibre strands	+	-	-	-	-	-	-
No. of vascular bundles in the ring	18	9	9	10	10	10	8
Average length and width of cortical (wing) bundles (μ)	240×160	225×125	125×100	150×125	127×127	150×125	95×79
Average length and width of polar bundles (μ)	400×200	350×150	200×150	375×300	254×254	275×200	206×175
Average length and width of lateral bundles (μ)	240×80	150×125	200×150	250×200	111×79	150×100	143×95
No. of xylem vessels of cortical (winged) bundles	9	14	8	8	9	9	7
No. of xylem vessels of polar bundles	8-14	10-16	10-13	15-32	20-25	7-15	17-26
No. of xylem vessels of lateral bundles	1-3	2-5	3-8	4-16	2-4	1-4	3-6
Average thickness of pith at wings (μ)	1400	575	500	625	508	550	318
Average thickness of pith at polar bundles (μ)	2040	1300	675	1050	555	675	508
Pith cell shape *	1	2	1	2	1	2	1

(*) Pith cell shape: (1) Polygonal to isodiametric; (2) Polygonal to isodiametric with undulated wall

Only the cortex of *V. faba* is characterized by the presence of four small cortical fibre strands, two above the two polar bundles and two above the two cortical bundles in the wings (Fig. 1a). The presence of these cortical fibre strands is recorded by Gourley (1931) two in *Pisum sativum*, Gupta and Murty (1986) only two above the two polar bundles in *Vicia faba* cv. T- 44 and Govila (1982) four in *Lens esculenta*. According to Gourley (1931) these fibre strands give mechanical support to the stem.

The central vascular cylinder consists of discrete collateral primary bundles which are usually individually distinct arranging in an elliptical ring around the pith. These bundles are two large polar bundles, the other small lateral bundles separating by wide medullary rays and additional minor traces derived from the neighbouring major bundles. They vary in number from 8 bundles in *V. villosa* to 18 bundles in *V. faba*, 9 in *V. narbonensis* and *V. peregrina*, or 10 in the other examined species. Gupta and Murty (1986) recorded the presence of eight vascular bundles in the main ring at the third internode in *Vicia faba* cv. T- 44, while the stem of the same species (*Vicia faba* cv. Giza 2) have 18 bundles at the fifth internode in the present study due to its large size. The maximum values of average length and width of polar bundles are (400 and 300 μ) in *V. faba* and *V. sativa*, respectively and the minimum ones are (200 and 150 μ) in *V. narbonensis* and *V. peregrina*. While, the highest values for average length and width of lateral bundles (250 and 200 μ) are observed in *V. sativa* and the lowest ones (111 and 79 μ) are noticed in *V. ervilia*. Also, the dense number of xylem vessels per polar and lateral bundle (15-32 and 4-16) are recorded in *V. sativa* and the little ones (10-13 and 1-3) are observed in *V. peregrina* and *V. faba*, respectively.

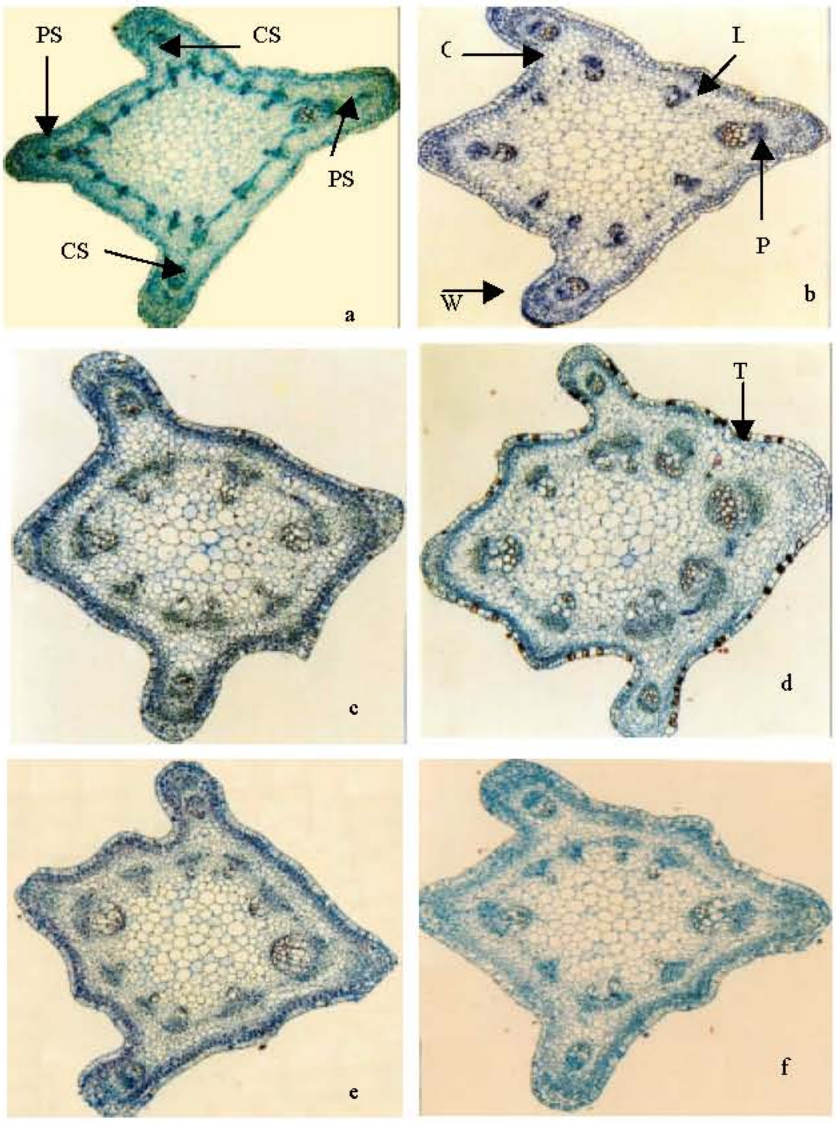

Fig. 1(a-f): Photographs of cross sections of the stem of the studied species. (a), *V. faba* (x 25);(b), *V. narbonensis* (x40); (c), *V. peregrina* (x40); (d), *V. sativa* (x40); (e), *V. ervilia* (x63); (f), *V. monantha* (x40). C = Cortical bundles; CS = Cortical fibre strand; L = Lateral bundle; P = Polar bundle; PS = Polar fibre strand; T = Tanniniferous cell, W = Wing

In all studied species, two cortical vascular bundles are observed, one in each wing. The presence of these cortical bundles in the stem has been observed in a number of species with winged or grooved stem (Metcalfe and Chalk 1950). The maximum values of average length and width of the cortical bundles are (240 and 160 μ) in *V. faba* and the minimum ones are (95 and 79 μ) in *V. villosa*. While, the dense number of xylem vessels per cortical bundle (14) are recorded in *V. narbonensis* and the little ones (7) are observed in *V. villosa*.

The pith is more or less squared and formed of ±polygonal to large isodiametric parenchyma cells with conspicuous intercellular spaces. These isodiametric cells have undulating walls only in *V. narbonensis*, *V. sativa* and *V. monantha*. The highest value of average thickness

of pith at wings and polar bundles are (1400 and 2040 μ) in *V. faba*, while the lowest ones (318 and 508 μ) are found in *V. villosa*. These results are similar with the data which obtained by Metcalfe and Chalk (1950 and 1979).

Nodal vasculature

Nodal vasculature in *V. villosa* Fig. 2(a-h): The observation of the nodal anatomy revealed close similarity between all the studied *Vicia* species, therefore, *V. villosa* will be discussed in detail. The ring of the vascular tissue is elliptical shape consisting of eight collateral conjoint bundles, two large bundles designated as polar bundles (A and E) and the other six lateral bundles are arranged in between them (B, C, D, F, G and H), three on each side (Fig. 2a), in addition to the two cortical bundles (I and J).

In the base of the node, the number of the vascular bundles increases from eight to twelve due to trifurcating the two lateral bundles (B and H) adjacent to the polar bundle (A) into six bundles, three each (B_1, B and B_2) and (H_1, H and H_2), one from each side (Fig. 2b). The two small bundles (B_2 and H_2) are the two cortical bundles of the next internode.

The polar bundle (A) moves into the cortex leaving a single gap in the main vascular ring and begins to trifurcate (Fig. 2c). This completely separate into three bundles immediately (A_1, A and A_2) within the cortex of the node, which later enters the leaf base as leaf traces. The middle bundle (A) is comparatively larger than the other two lateral bundles acting as median leaf trace (Fig. 2d).

Slightly above it, the axillary bud supply (Bud) extends from the two bundles (B_1 and H_1) adjacent to the leaf gap into the cortex which join laterally (Fig. 2c-f). After formation the leaf and axillary bud supply, the leaf gap is filled up immediately by the fusion of the two lateral bundles (B_1 and H_1) adjacent to this gap and the central cylinder is again completed (Fig. 2d-g).

The two wings gradually expand covering nearly the node except for the portion opposite the leaf base (Fig. 2d). The two cortical vascular bundles (I and J) present in the two wings and the two lateral leaf bundles (A_1 and A_2), all bifurcate, each into two bundles (I_1, I_2, J_1, J_2, A_{1a}, A_{1b}, A_{2a} and A_{2b}) (Fig. 2e and f). This step appear clearly in the cross section of nodal vasculature of *V. sativa* (Fig. 2b). The resulting two cortical bundles (I_2 and J_2) split each into three bundles extending into the expanded portion of the wing. This is in agreement with Kupicha (1975) and Gupta and Murty (1986) who reported that the stipules of the foliage leaves in most of the *Vicia* species are supplied by the cortical vascular bundles only and not by the central cylinder.

While, the two bundles (J_1 and A_{1b}) converge from each other, then fuse to give the adaxial petiole trace as well as the two bundles (I_1 and A_{2b}) and the adaxial side of the petiole supply by two traces (Fig. 2f). This is in accordance to Kupicha (1975, 1977) who mentioned that all the genera of tribe Vicieae except *Cicer* are characterized by an unusual type of cauline vasculature in which the lateral leaf traces are present in the internode below the insertion of each leaf as a pair of cortical bundles.

The two bundles (A_{1a} and A_{2a}) are the median petiole traces, while the bundle between them (A) is the abaxial petiole trace. Therefore, the leaf base has five vascular bundles in the middle supplying the leaf petiole and three to nine vascular bundles in the lateral portions supplying the two adnate stipules on the leaf side. The leaf base gradually separates from the node along with the two winged portion as a single structure which appears as a sheathing leaf base surrounding most of the node (Fig. 2f and g) and the stem have the original number of vascular bundles.

The two small bundles (B_2 and H_2) gradually move outwards into the cortex forming the two new cortical bundles and tissues where they present projects slightly outside forming the two wings (Fig. 2f and g). This means that complete replacement occur for the two original cortical bundles in the wings at each node in agreement with Muller (1937), Kupicha (1975) and Gupta and Murty (1986).

At the level of separation of the leaf base, the axillary bud also separates from the node and its supply repeatedly divides (Fig. 2g and h).

Nodal Vasculature in the remaining studied *Vicia* species Fig. 3(a-f): The remaining studied species have the same way as *V. villosa*, but differ in the number of the bundles. At the beginning of nodal vasculature, the number of the vascular bundles increases from ten to fourteen in three species (*V. sativa*, *V. ervilia* and *V. monantha*) (Fig. 3a, K and L is the excess two bundles), while, it increases from nine to thirteen in *V. peregrina*. Only, two species have small deviations from the other examined species, the number of the vascular bundles increases from nine to twelve in *V. narbonensis*, or from 18-20 in *V. faba*. In *V. narbonensis*, the lateral bundle (H) adjacent to the polar bundle (A) behave as usual, but the other lateral bundle (B) bifurcate to two bundles only (B_1 and B) and the cortical bundle (B_2) originate from the neighbour lateral bundle (C) as (C_1) which split into two bundles (C and C_1) (Fig. 3e). In *V. faba*, the two lateral bundles (B and H) adjacent to the polar bundle (A) behave as the lateral bundle (B) in *V. narbonensis*, while the two cortical bundles of the next internode originate from the third or the fourth lateral bundles for the polar bundle (A) (Fig. 3d).

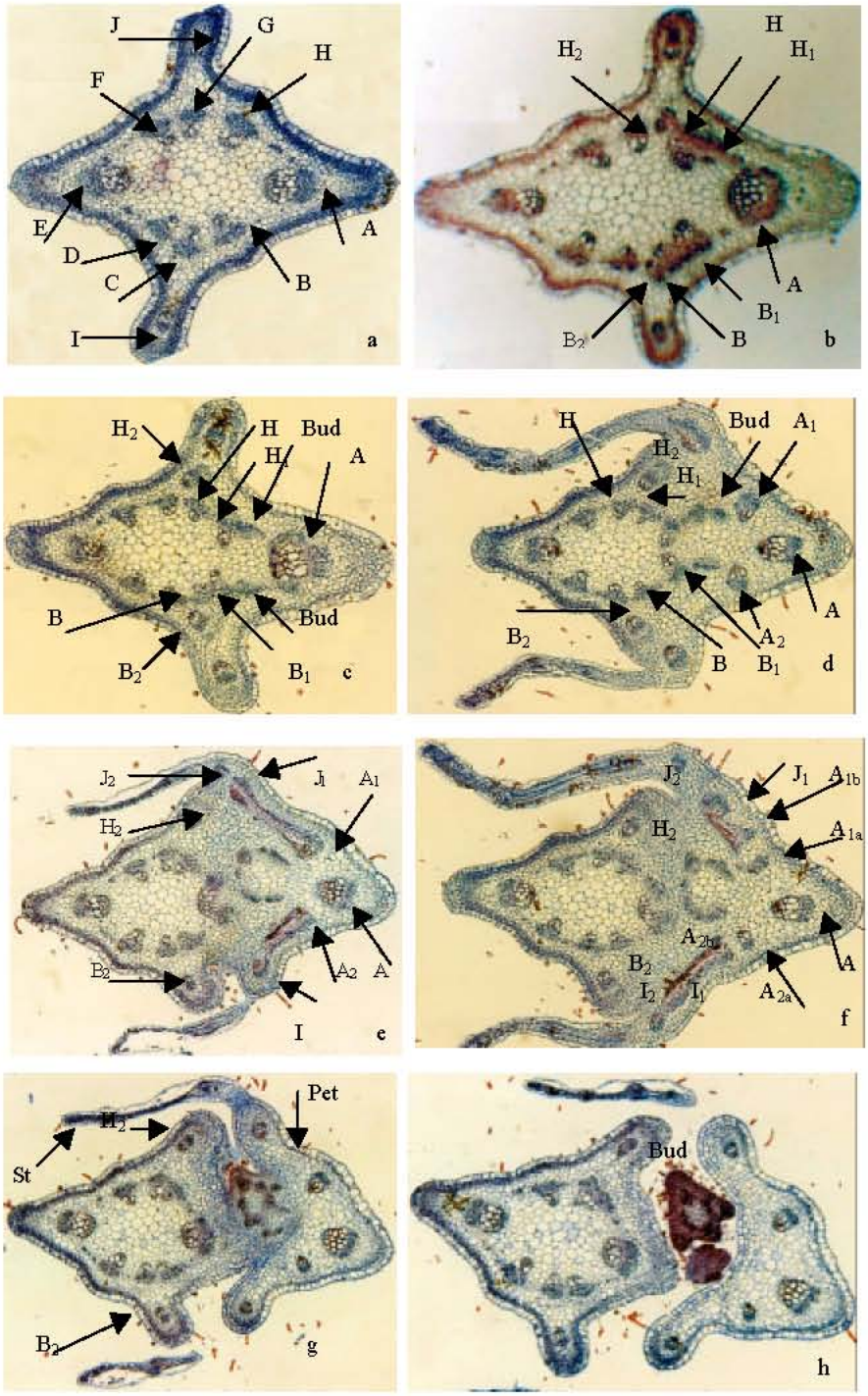

Fig. 2(a-h): (a), Photograph of cross section of the stem of *V. villosa* (x63). (b-h), Photographs of serial cross sections showing the nodal vasculature of the fifth node and internode in *V. villosa* (x40); Bud = Axillary bud supply, Pet = Petiole; St = Stipule

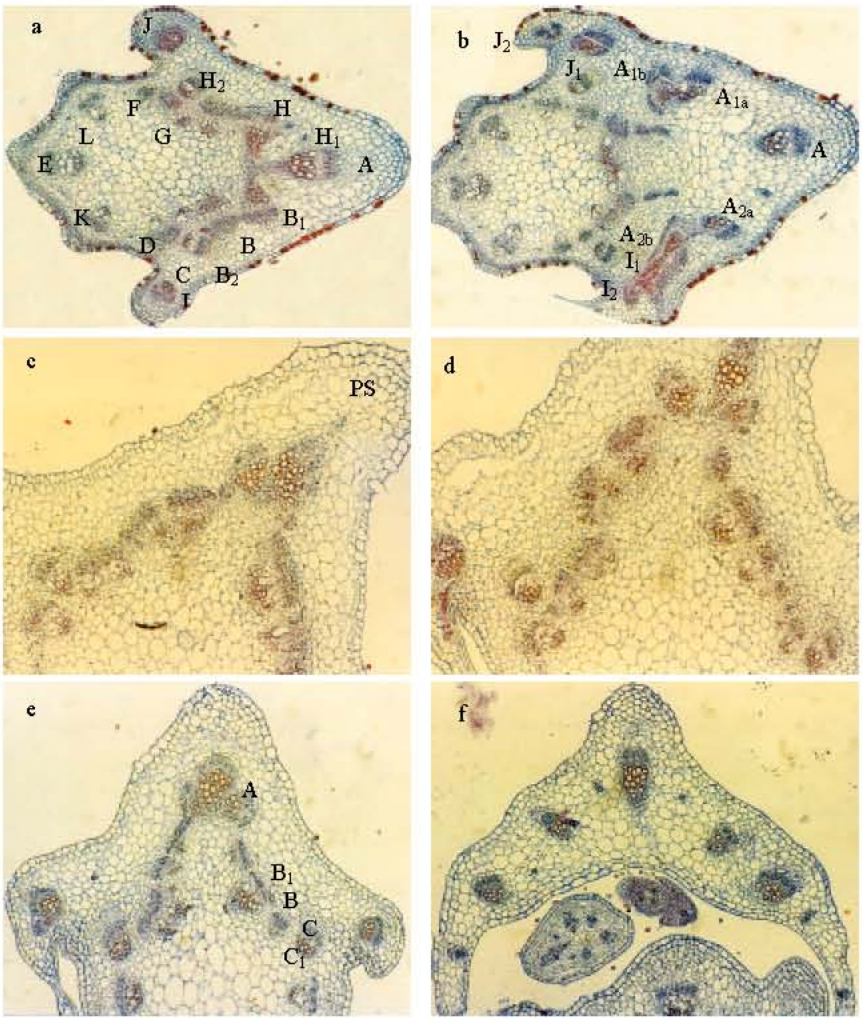

Fig. 3(a-f): Photographs of cross section showing nodal vasculature in these species. (a and b), *V. sativa* (x40); (c and d), *V. faba* (x40); (e and f), *V. narbonensis* (x40)

Only in *Vicia faba*, when the polar bundle (A) moves into the cortex, joins the cortical fibre strand (Fig. 3c) and the newly formed polar bundle also gives a new small fibrous strand which moves outward above it into the cortex forming a cortical fibre strand. The same is happened for the two fibre strands above the two cortical bundles in the same species.

The petiole includes additional minor traces derived from the neighbouring major ones in *V. faba* and *V. narbonensis* (Fig. 3f).

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REFERENCES

Boulos, L., 1999. Flora of Egypt (*Azollaceae-Oxalidaceae*). Al Hadara Publishing, Cairo, Egypt, pp: 341-348.

Fotedar, R.L., 1987. Nodal anatomy and leaf vasculature in *Juglans regia*. Phytomorphology, 37: 47-52.

Gourley, J.H., 1931. Anatomy of the transition region of *Pisum sativum*. Bot. Gaz., 92: 367-383.

Govila, R., 1982. Studies in reproduction biology of some pulses. Ph.D. Thesis, Meerut University, Meerut, India.

Gupta, M. and Y.S. Murty, 1986. Seedling anatomy of *Vicia faba* cv. T-44. Phytomorphology, 36: 243-250.

Hassan, A.E., 1992. Comparative studies on the Egyptian species of the genus *Trigonella* L. Thesis, M.Sc., Fac. Agriculture, Suez Canal Univ., pp: 46-49.

Hegde, U. and V.D. Tilak, 1982. Anatomy of the seedling of the Leguminosae-I. Proc. Indian Acad. Sci. (Plant Sci.), 91: 281-287.

Johansen, D.A., 1940. Plant Microtechnique, MC. Graw-Hill Book Co. New York.

Kupicha, F.K., 1975. Observations on the vascular anatomy of the tribe Vicieae (Leguminosae). Bot. J. Linn. Soc., 70: 231-242.

Kupicha, F.K., 1976. The infrageneric structure of *Vicia*. Notes from the Royal Botanic Garden, Edinburgh, 34: 287-326.

Kupicha, F.K., 1977. The delimitation of the tribe Vicieae (Leguminosae) and the relationships of *Cicer* L. Bot. J. Linn. Soc., 74: 131-162.

Kupicha, F.K., 1981. Tribe 21. Vicieae (Adans.) DC. (1825), Nom. Conserv. Prop. In: Advances in legume systematics. Polhill, R.M. and P.H. Raven (Eds.), Royal Botanic Gardens, Kew, Part 1: 377-381.

Mabberley, D.J., 1997. The Plant-Book. Cambridge University Press, Cambridge, pp: 59-60.

Maxted, N., 1993. A phenetic investigation of *Vicia* L. subgenus *Vicia* (Leguminosae, Vicieae). Bot. J. Linn. Soc., 111: 155-182.

Metcalfe, C.R. and L. Chalk, 1950. Anatomy of Dicotyledons. Clarendon Press, Oxford, 1: 502-533.

Metcalfe, C.R. and L. Chalk, 1979. Anatomy of the Dicotyledons. Clarendon Press, Oxford, 1: 712-72.

Minu and Y.S. Murty, 1989. Root-shoot transition and nodal vasculature in *Leucaena leucocephala* (Lamk.) De Wit. Phytomorphology, 39: 135-139.

Muller, C., 1937. La tige feuillée et les cotylédons des Viciées á germination hypogée. Cellule, 46: 195-354.

Pant, D.D. and B. Mehra, 1964. Nodal anatomy in retrospect. Phytomorphology, 14: 384-387.

Rudall, P., 2000. Anatomy of Flowering Plants (An introduction to structure and development). Cambridge University Press, Edn., 2: 16-19.

Täckholm, V., 1974. Students' Flora of Egypt. Cairo University, Edn., 2: 272-278.

Willey, R.L. 1971. Microtechnique, A Laboratory Guide. Mcmillan Publishing Co., Inc., N.Y., pp: 99.