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Study of Anatomical Characters in Relation to Resistance Against Brinjal Shoot and Fruit Borer

¹M. Mofazzel Hossain, ²M. Shahjahan, ³A.K.M. Azad-ud-doula Prodhan,

¹M. Sirajul Islam and ¹M. A. Begum

¹Bangladesh Rice Research Institute, Gazipur-1701, Bangladesh

²Department of Entomology, ³Department of Crop Botany, Bangladesh Agricultural University, Mymensingh, Bangladesh

Abstract: Resistance or susceptibility of brinjal varieties/lines to shoot and fruit borer seems to be related with some anatomical characters. Varieties/lines having thick cuticle, broad and thick collenchymatous area (hypodermis), compact parenchyma cells in the cortical tissue, small area in the cortical tissues, more vascular bundles with narrower spaces in the interfascicular region, and compact arrangement of vascular tissue with lignified cells and small pith were the main characters of resistant/tolerant varieties. On the other hand, thinner cuticle and collenchymatous area (hypodermis), loose parenchyma cells in the cortical region, larger spaces between vascular bundles i.e., interfascicular region and large pith, less number of trichomes, soft parenchymatous cells in the interfascicular region, might be responsible for the susceptibility to brinjal shoot and fruit borer.

Key words: Brinjal, shoot borer, fruit borer, *Leucinodes orbonalis*, resistance mechanism, plant anatomy

Introduction

Host plant characters including morphological or structural qualities interfere with insect behaviour such as mating, ovipositing, feeding and food ingestion. Pubescence and tissue hardness limit insect mobility acting as structural barriers (Webster, 1975). Various mechanical resistance factors in plants such as solidness of stem, thickness of tissues, anatomical adaptations, and protective structures affect the use of a plant as a host by phytophagous insects like brinjal shoot and fruit borer (*Leucinodes orbonalis* Guenee, Pyralidae: Lepidoptera). Identification of a morphological or biochemical factor governing resistance helps in the development of a rapid screening technique. Resistance may be due to antixenosis or non-preference and appears to have a biochemical basis, although non-preference of some cultivars has been attributed to anatomical characters. The yield loss caused by brinjal shoot and fruit borer pest has been estimated up to 67% in Bangladesh (Islam & Karim, 1991) and up to 63% in Haryana, India (Dhankar *et al.*, 1977). The damage by *L. orbonalis* starts soon after transplanting of seedling and continues till the last harvest of the fruits. In the early stage of crop growth, the newly hatched larvae bore into the petioles, midribs of large leaves and young tender shoots. They close the entry holes with their excreta and feed inside (Butani and Jotwani, 1984). Due to the larval activity within the shoot the transmission mechanism of the plant sap is affected, causing dropping and the withering of the shoots (Alam and Sana, 1962). At the later stage of plant growth, the larvae bore into the flower buds and also enter into the tender fruits, generally through the calyx without leaving any visible sign of infestation (Butani and Jotwani, 1984). At this stage of plant growth, the insect damages both shoots and fruits. Secondary infections by certain bacteria may cause further deterioration of the fruits (Islam and Karim, 1994). As a result, the brinjal becomes rotten which is unfit for human consumption. Compact vascular bundles in a thick layer, with lignified cells and less area of pith in the shoot provided some resistance to *L. orbonalis* (Panda *et al.*, 1971; Chelliah and Srinivasan, 1983). Similarly, hardness of fruit skin and flesh due to compact seed arrangement and tight calyx affects larval entry into fruits which provided some degree of resistance to brinjal shoot and fruit borer (Lall & Ahmad, 1965). Ali *et al.* (1994) examined the anatomical characters of 28 brinjal varieties/lines, which indicated that fruits with compact mesocarp and pulp suffer lower infestation than those with loose mesocarp and pulp. Mishra *et al.* (1988) tested 24 brinjal varieties for resistant to *L. orbonalis* and found that leaf color and phyllotaxy are not related to pest resistance, but tightly arranged seeds in the mesocarp, thick fruit skin and closely packed

vascular bundles in pulp are possible causes of resistance in some varieties.

Panda *et al.* (1971) evaluated 19 brinjal varieties for resistant to *L. orbonalis* and ascertained that the resistant varieties (Thorn Pendy, Black Pendy, H-165 and H-407) possess heavily lignified sclerenchymatous layers and closely packed vascular bundles and seeds are packed tightly in the mesocarp. They also noticed that the thick pithy stem and loose calyx of the susceptible varieties facilitated larval entry. Therefore, the present work has been undertaken to study the anatomical characters in relation to resistance against brinjal shoot and fruit borer.

Materials and Methods

Selected tolerant, moderately tolerant, susceptible and highly susceptible varieties/ lines of brinjal (Hossain, 1997) were grown in Bangladesh Agricultural University farm in Kharif season, 1997 (Table 1).

Table 1: Selected tolerant, moderately tolerant, susceptible and highly susceptible varieties/lines of brinjal

Types of resistance	Variety(s)/ Lines
Tolerant	Jhumki, Baromashi
Moderately tolerant	Uttara, Singnath, Sada ball, BLO96, BLO117 (Khotkhotia-1), BLO81 (Dhohazari-1), BLO101, BLO72 (Dhohazari-2), BLO45, Islampuri, Laffa
Susceptible	BLO95, BLO85, BLO98, BLO114, BLO118 (Khotkhotia-2), Borka
Highly susceptible	Nayankajal

The experimental plots were prepared with proper ploughing and laddering using recommended fertilizer doses (Rashid, 1993). Cowdung was applied as basal dose at the rate of 15 tons/ha while urea, TSP and MP were applied at the rate of 115kg N₂, 72kg P₂O₅ and 75kg K₂O per hectare, respectively. Half of urea, TSP and MP were applied as basal dose and remaining half of urea and MP were applied in two equal top dressings at 21 days after transplanting and at the flowering. Intercultural operations (viz., weeding, roughing, irrigation) were performed as and when necessary throughout the growth period of the crop. Chemical control measures were not taken against insect pests. Secondary shoots were collected on 100 days after transplantation from the infested plants. These shoots were fixed and preserved in formalin-aceto-alcohol (FAA) solution (Johansen, 1940). Transverse sections of the stem were made from fresh and fixed materials following free hand sectioning, using ordinary razor blades. Sections were made through the region between 3rd and 4th leaf from the top. The uniform thin sections were selected,

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stained in saffranin and mounted in 50% glycerine temporarily following the general principle of Johansen (1940). The slides were examined under microscope to study the variation of anatomical characteristics of resistant, tolerant and susceptible brinjal varieties.

Results

Anatomical characteristics of different level of resistant varieties/lines are discussed below:

Anatomical characters of highly susceptible varieties/lines: Stem of highly susceptible variety, Nayankajal, has single layered epidermis with small and large cells (Fig.1b). The cells are oval, round or slightly rectangular in shape and more or less uniform in size. Epidermal cells are thin walled and bears thin cuticle. Trichomes are very few in number, simple and branched. The hypodermis consists of 6-7 layers of moderately thick collenchymatous cells beneath the epidermis (Fig.1a, b). The parenchymatous cells (8-9 layers) of cortical region are thin walled with prominent intercellular spaces.

Vascular bundles are poorly developed. Vascular tissues form a ring of various thickness showing a beaded structure. The cambial activity is higher in the vascular bundles compared to that of the interfascicular region. The interfascicular region, which is more in number compared to other varieties, is composed of less lignified tissues (less number of vessels and fibers, and more number of parenchyma cells). The pith of this variety is large compared to that of the other varieties/lines. Pith contains parenchymatous

cells, which are more or less similar in size as compared to the cortical tissue.

Anatomical characters of susceptible varieties/lines: Epidermis of the stem of susceptible varieties, BLO 95 and BLO 98, is single layered with thin to moderately thick cuticle (Fig. 2a, b and 3a, b). Epidermal cells are oval or round in shape. The variety BLO 95 contains trichomes that are single and branched with or without a distinct stalk (Fig. 2a). There are 6-7 layers of thick collenchymatous cells beneath the epidermis. Parenchyma cells (7-8 layers) in the cortical region are larger in size with prominent intercellular spaces. Vascular tissues form a ring with various thicknesses. The radial thickness of the interfascicular region is less compared to the fascicular (vascular bundle) region. Cambium is more active in the fascicular region compared to the interfascicular region. Interfascicular region consists of less lignified cells (less number of fibers and vessels and more number of parenchymatous cells). Radial thickness of interfascicular region of variety BLO 95 is higher compared to that of the variety Nayankajal. The pith area is less compared to that of Nayankajal. Abaxial side of the pith is composed of small sized cells compared to that of Nayankajal. The epidermis of variety BLO 98 bears less number trichomes compared to variety BLO 95.

Anatomical characters of moderately tolerant varieties/lines: Stem of moderately tolerant varieties, Islampuri, Singnath and Uttara,

Table 2: Anatomical features of different varieties/lines for their degree of resistance.

Characters	Highly susceptible	Susceptible	Moderately tolerant	Tolerant
Epidermis:				
A. Layer	Single	Single	Single with wavy circumference	Single
B. Cell:				
a. Shape	Oval, round or slightly rectangular	Oval or round	Oval, round, slightly rectangular or square	Oval or tangentially elongated
b. Size	Small and large, thin walled	Uniform, thin walled	Smaller, thick walled, slightly wavy in nature	Uniform and smaller thick walled
C. Cuticle	Very thin to thin	Thin to moderately thick	Moderately thick to thick	Thick
D. Trichome	Very few, simple and branched	Few, simple and branched with or without a distinct stalk	Numerous, compact in Islampuri, simple, branched with or without a distinct stalk.	Numerous, compact in Baromashi, simple, branched with or without a distinct stalk.
Cortex:				
A. Collenchyma (Hypodermis)				
a. Layer	6-7 layers	6-7 layers	7-8 layers	8-9 layer
b. Cell wall corner	Moderately thick	Thick	Very thick	Very thick
B. Parenchyma cell				
a. Layer	8-10 layers	7-9 layers	6-8 layer	5-7 layer
b. Shape	Round, oval or polygonal	Round, oval or polygonal	Round, oval or polygonal	Round, oval or tangentially flattened
c. Size	Large with prominent intercellular spaces in middle region but smaller in abaxial and adaxial region	Larger with prominent intercellular space	Smaller, intercellular space smaller and less in number	Abaxial and adaxial cells are smaller
C. Wall	Thin walled with prominent intercellular spaces	Thin walled with prominent intercellular spaces	Thick walled	Thick walled apparently less Number of intercellular spaces
Vascular bundle				
a. Development	Poorly developed	Poorly developed	Moderately developed	Well developed
b. Number	Many	Many	Many	Many
c. Size	Smaller, vascular tissues form a ring with various thickness showing a beaded structure	Smaller, vascular tissues form a ring with various thickness, thick in fascicular region and thin in interfascicular region, interfascicular region is wider in some places. Lignified tissues are comparatively more in interfascicular region compared to highly susceptible variety.	Large, vascular tissues form a ring with uniform radial thickness, wavy circumference. Interfascicular region is composed of lignified tissues.	Large, vascular tissues form a well-developed ring with uniform radial thickness. Fibers are more in numbers, pericycle discontinuous, thick walled and highly lignified.
d. Cambial activity	Higher in fascicular compared to interfascicular region.	Higher in vascular bundle and less in interfascicular region.	More or less uniform throughout the vascular ring.	Uniform throughout the ring
e. Ray regions	Many, less lignified tissues i.e., less number of vessels and fibers and large number of parenchyma.	Thick walled, very few fibers in the interfascicular region.	Contains more thick walled fiber cells and thin walled ray cells compared to susceptible variety.	Well developed and more number of thick walled fibre cells, compact and highly lignified.
Pith	Large and connected with the cortex through thin walled ray parenchyma.	Smaller compared to highly susceptible variety and connected with the cortex through thick walled ray parenchyma.	Slightly smaller than the susceptible variety	Smaller pith with exception in Jhunki

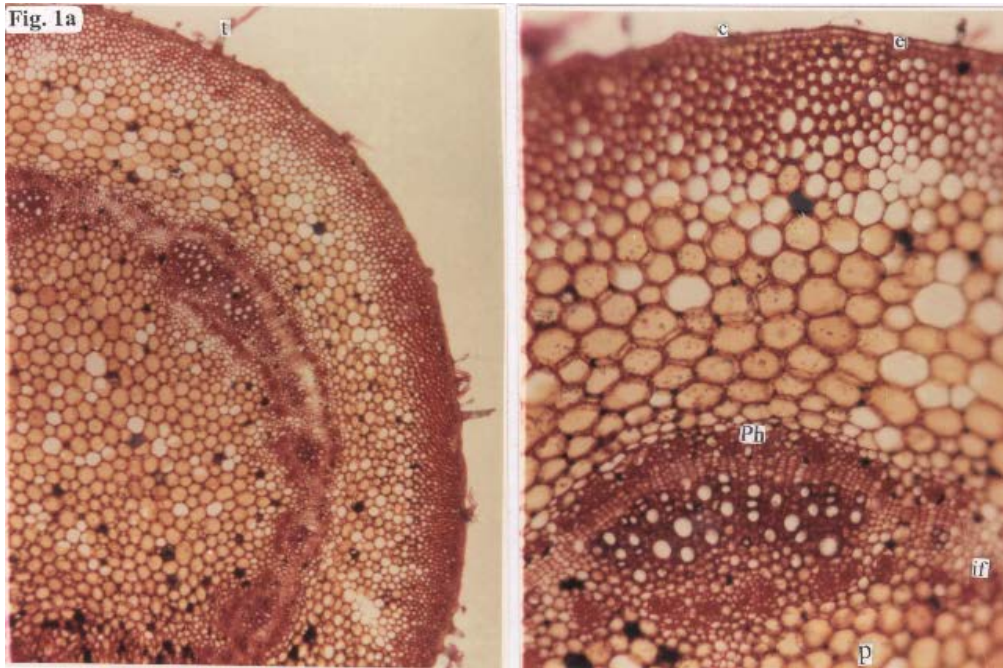


Fig. 1: a: T.S. of the stem of Nayankajal showing thin cuticle (c), few trichomes (t), cortical (cor) region with collenchyma (col) and parenchyma (par) cells, beaded like vascular bundle (Vb), interfascicular (if) region, and pith (p). X 37.
 b: T.S. of the stem of Nayankajal showing epidermis (e) with thin cuticle (c), parenchyma (par) cells with prominent intercellular spaces in the cortical (cor) region, beaded like vascular bundle (Vb), phloem (ph), cambium (Ca) and xylem (x), interfascicular (if) region, and pith (p). X90.

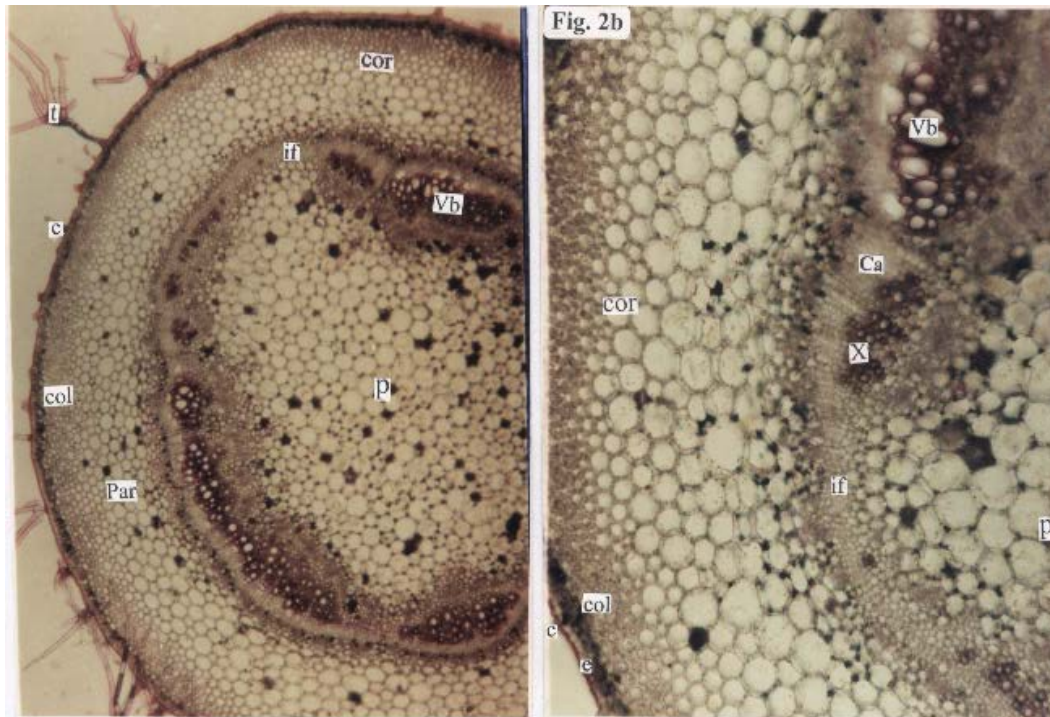


Fig. 2: a: T.S. of the stem of BLO 95 showing thin cuticle (c), branched trichomes (t), cortical (cor) region with collenchyma (col) and parenchyma (par) cells, vascular bundle (Vb), interfascicular (if) region and pith (p). X37.
 b: T.S. of the stem of BLO 95 showing single layered epidermis (e) with thin cuticle (c), Vascular tissues form a ring with various thicknesses, parenchymatous (par) cells with intercellular spaces in cortical (cor) region, phloem (ph), cambium (Ca), xylem (x), less lignified fibers and vessels in interfascicular (if) region and pith (p). X90.

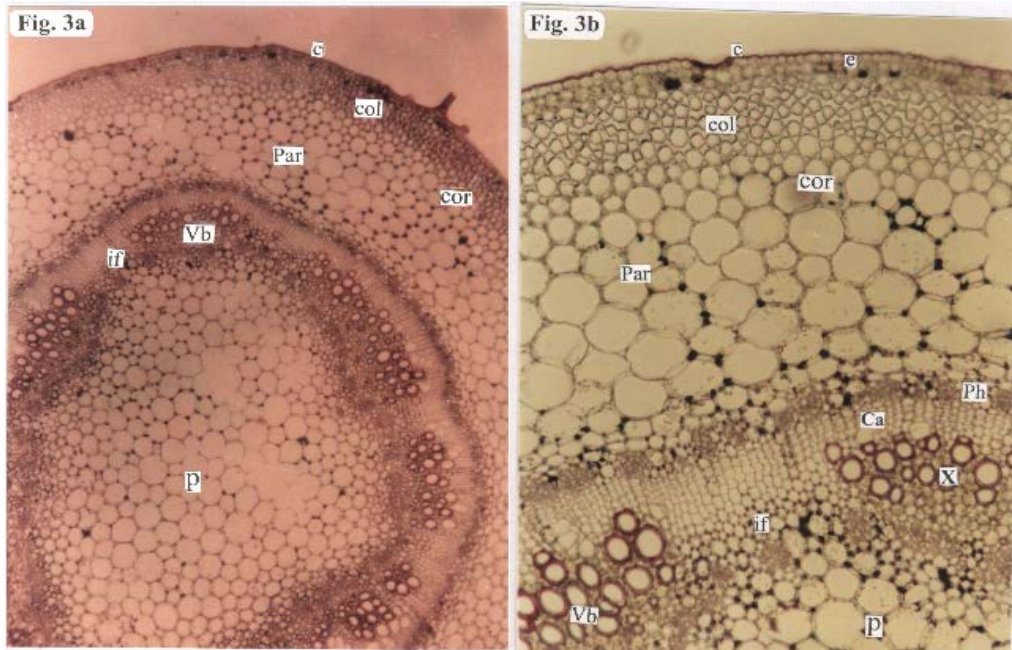


Fig. 3: a: T.S. of the stem of BLO 98 showing few trichomes (t) with moderately thick cuticle (c), thick collenchymatous (col) cells beneath the epidermis (e), vascular tissues form a ring with various thickness, large sized parenchyma (par) cells in cortex (cor) and pith (p). X37.
 b: T.S. of the stem of BLO 98 showing moderately thick cuticle (c), 6-7 layers of thick collenchymatous (col) cells beneath the epidermis (e), epidermis consists of oval or rectangular parenchyma cells, vascular bundle with phloem (ph), cambium (Ca), and xylem (x). Vascular tissues form a ring with various thickness, large sized parenchyma (par) cells in cortex (cor) and pith (p). X90.

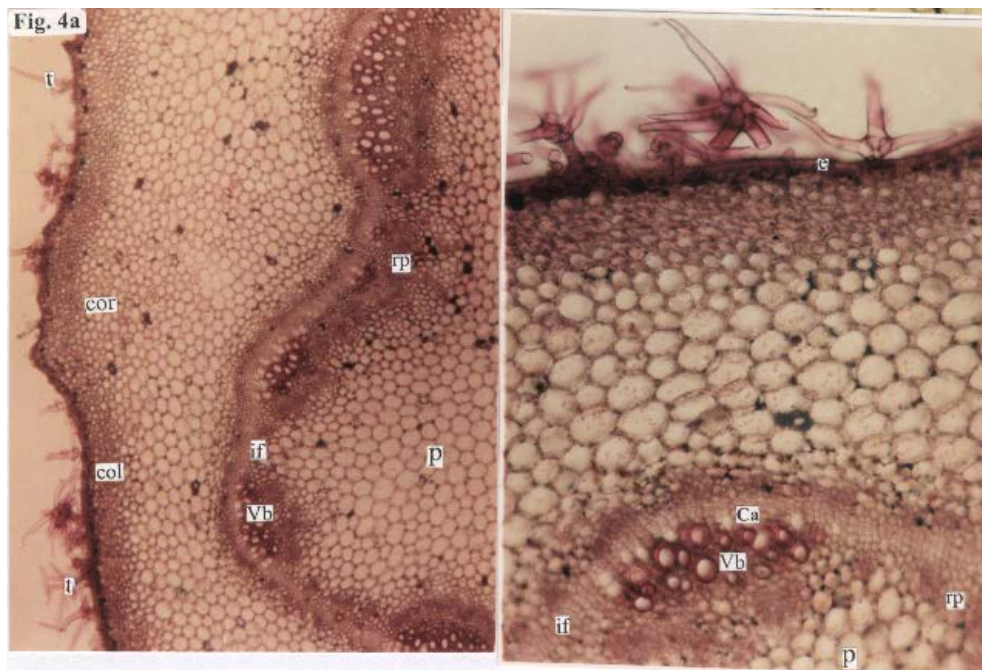


Fig. 4: a: T.S. of the stem of Islampuri showing wavy single layered epidermis (e), large number and branched trichomes (t) with or without a distinct stalk, vascular bundles (Vb) are irregular in radial thickness, cortical (cor) region is small in ridged portion and large in furrow region, interfascicular (if) region with ray parenchyma (rp). X37.
 b: T.S. of the stem of Islampuri showing wavy single layered epidermis (e), large number and branched trichomes (t) with or without a distinct stalk, cortical (cor) region with collenchyma (col) and parenchyma (par) cells, vascular bundles (Vb) are irregular with phloem (Ph), cambium (ca) and xylem (x), large interfascicular (if) region with ray parenchyma (rp). X90.

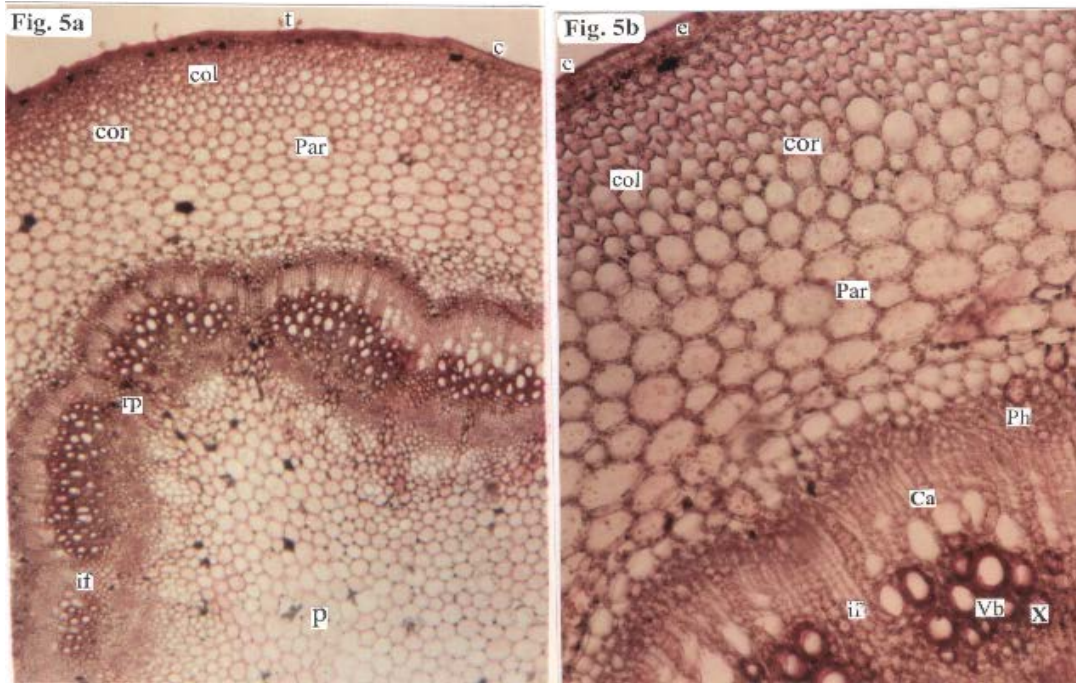


Fig. 5: a: T.S. of the stem of Singnath showing very few and small trichomes (t) with moderately thick cuticle (c), The outer circumference of vascular ring is wavy in nature, very short interfascicular (if) region with ray parenchyma (rp). Pith (p) and cortical (cor) regions are composed of parenchymatous (par) cells with prominent intercellular spaces. X37.
 b: T.S. of the stem of Singnath showing single layered epidermis (e) with moderately thick cuticle (c), very short interfascicular (if) region with ray parenchyma (rp), vascular bundle (Vb), cambium (Ca). Pith (p) and cortical (cor) regions are composed of parenchymatous (par) cells with prominent intercellular spaces. X90.

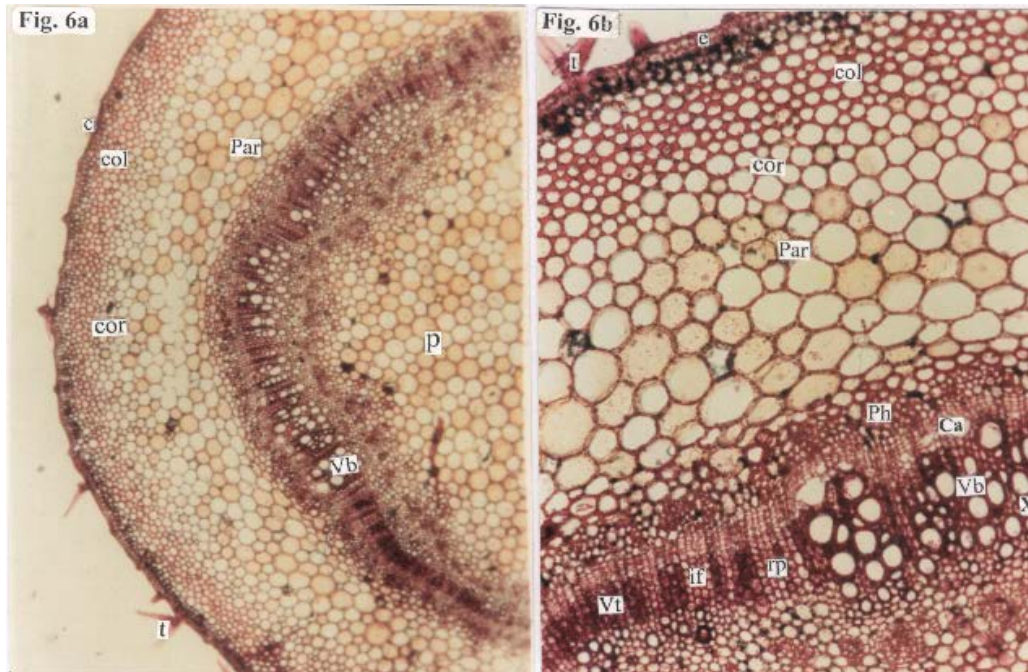


Fig. 6: a: T.S. of the stem of Uttara showing pointed trichomes (t) on single layered epidermis (e), very thick collenchymatous (col) cells. Pith (p) and cortical (cor) regions are composed of parenchymatous (par) cells with prominent intercellular spaces. Vascular bundles (Vb) form a ring with uniform thickness. X37.
 b: T.S. of the stem of Uttara showing trichomes (t) on single layered epidermis (e), 7-8 layers of very thick collenchymatous (col) cells. Pith (p) and cortical (cor) regions are composed of parenchymatous (par) cells with prominent intercellular spaces. Vascular bundles (Vb) form a ring with uniform thickness and consist of phloem (ph), cambium (Ca) and xylem (x). X90.

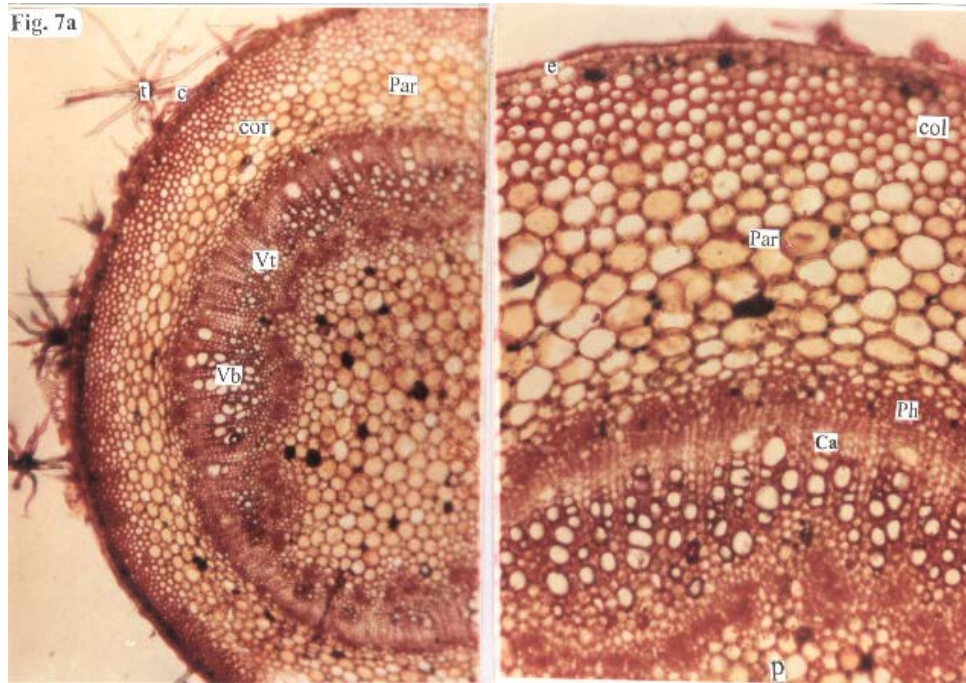


Fig. 7: a: T.S. of the stem of Baromashi showing more branched trichomes (t) with thick cuticle (c), very thick collenchyma (col) and parenchyma (par) cells in cortex (cor), well-developed vascular bundle (Vb) and small pith (p). X 37.
 b: T.S. of the stem of Baromashi showing single layered epidermis (e) with thick cuticle (c), 8-10 layers of very thick collenchymatous cell in cortex (cor), well-developed vascular bundle (Vb), phloem (Ph), cambium (Ca), xylem (x) and small pith (p). Phloem fibers are well developed. X90.

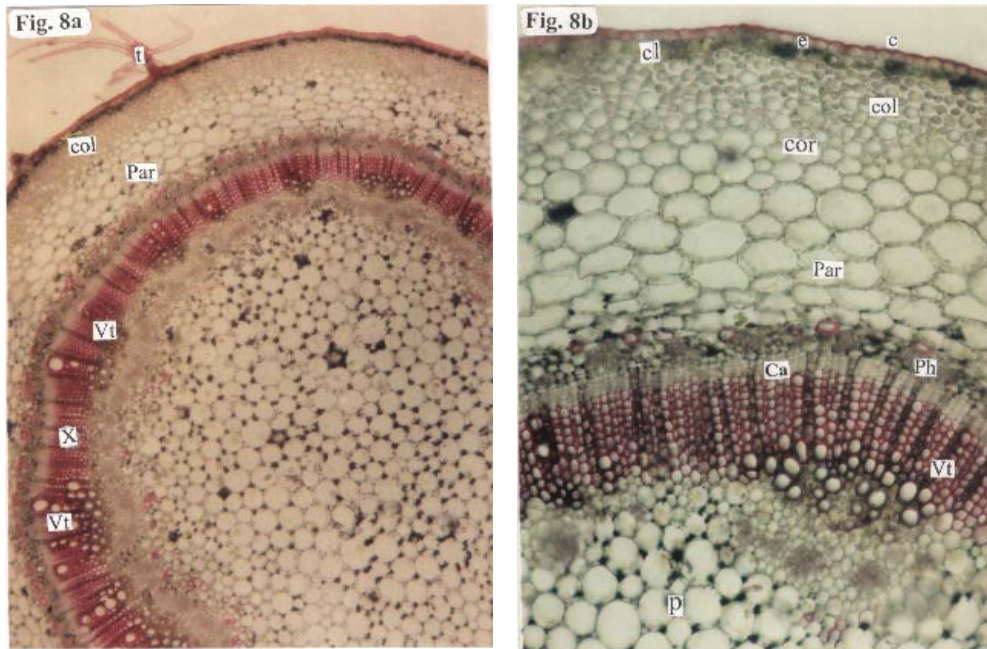


Fig. 8: a: T.S. of the stem of Jhumki showing single layered epidermis (e) with thick cuticle (c), branched trichome (t) with a distinct stalk, collenchyma (col) and parenchyma (par) cells in the cortex (cor), vascular tissue (Vt) forms a ring with uniform radial thickness, well-developed xylem (X) fibers and cambium (Ca). X37.
 b: T.S. of the stem of Jhumki showing single layered epidermis (e) with thick cuticle (c), 1-2 layers of chlorenchyma (cl) cells beneath the epidermis, 7-8 layers of thick walled collenchyma (col), vascular tissue (Vt) forms a ring with uniform radial thickness. Phloem (Ph) and xylem (X) fibers are well developed and lignified. Cambial (Ca) zone is composed of 4-5 layers of tangentially flattened compact cells. X90.

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has single layered epidermis (Fig. 4-6). There are 7-8 layers of very thick collenchymatous cells beneath the epidermis. Epidermal cells are oval, round, slightly rectangular or square in shape. The epidermal circumference is wavy in the variety Islampuri (Fig. 4a). Cuticle is thick to moderately thick. Trichomes are more in number and compact in Islampuri (Fig. 4) compared to those of Singnath (Fig. 5) and Uttara (Fig. 6). Trichomes are branched with or without a distinct stalk in Islampuri (Fig. 4) cortical tissues are larger in size with prominent intercellular spaces. Vascular tissues form moderately to well develop ring with uniform thickness in the variety Uttara (Fig. 6) and irregular in thickness in the variety Islampuri (Fig. 4) and Singnath (Fig. 5). Vascular tissue is moderate to highly develop compared to that of susceptible varieties/lines. The outer circumference of the vascular ring of variety Islampuri and Singnath is wavy (Fig. 4a, 5b). Interfascicular region is composed of lignified tissues (Figs. 4-6). Length of interfascicular region is very short in Singnath (Fig. 5a). There is no prominent interfascicular region in the variety Uttara (Fig. 6b). The ring of the vascular tissue is sometimes interrupted by parenchymatous cells in the interfascicular region thereby the entry of the larvae of brinjal shoot and fruit borer becomes easier through this soft (parenchymatous cell) region.

Anatomical characters of tolerant varieties/lines: Stem of the tolerant variety, Baromashi and Jhumki, bears single layered epidermis with thick cuticle. Epidermal cells are oval or tangentially elongated. The epidermis bears trichomes which are branched with or without a distinct stalk (Fig. 7 and 8). These trichomes frequently impart general antixenosis type of resistance providing an effective barrier that prevents movement and feeding of larvae on the plant surface. Branched trichomes are more in Baromashi (Fig. 7a) compared to that of Jhumki (Fig. 8b). There are 8-10 layers of very thick collenchymatous cell in Boromashi (Fig. 7b) and 7-8 layers in Jhumki (Fig. 8b). The cortical region contains less number (5-7 layers) of parenchymatous tissues compared to that in other varieties/lines. Cells are round, oval or tangentially flattened.

Vascular tissue is composed of lignified cells, which form a well-developed ring with uniform radial thickness (Fig. 7, 8). Fibers are more in number compared to other varieties. Phloem fibers are well developed and lignified. Interfascicular region of the variety Baromashi is less developed compared to fascicular region (Fig. 7a). Pith is small in Baromashi compared with Jhumki.

Discussion

The identification of anatomical features governing resistance can help in the development of a rapid screening technique. We have identified the anatomical features of different types of resistant varieties (Table 2). From the observations it seems that brinjal varieties with thick cuticle, broad and thick collenchymatous area (hypodermis), compact parenchyma cells in the cortical tissue, small area in the cortical tissues, more vascular bundles with narrower spaces in the interfascicular region, compact arrangement of vascular tissue with lignified cells and small pith characters offer some degree of resistance to brinjal shoot and fruit borer (Table 1) whereas susceptible varieties have thinner cuticle and collenchymatous area (hypodermis), loose parenchyma cells in the cortical region, larger spaces between vascular bundles i.e., interfascicular region and large pith, less number of trichomes, soft parenchymatous cells in the interfascicular region are the characters responsible for susceptibility. Brinjal varieties showed resistance to *L. orbonalis* by heavily lignified sclerenchymatous layer and closely packed vascular bundles. Ali *et al.* (1994), and Lall and Ahmed (1965) reported similar phenomenon. We have observed that larval entry is affected by thick cuticle (Baromashi and Jhumki) with pointed multi-cellular trichomes and small pith region. Mishra *et al.* (1988) and Panda *et al.* (1971) observed that

larval entry is also affected by thick cuticle, small pithy stem and pointed unicellular trichomes. The results of the present study are in agreement with those of Lall and Ahmed (1965) and Panda *et al.* (1971).

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