Journal of Research in Biology

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Anatomical investigations in *Silybum marianum* (L.) Gaertn.

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ABSTRACT:

Anatomical studies have been carried out for various parts of species *Silybum marianum* (L.) Gaertn. According to the available literature very little work has been done on this aspect. Keeping this in view presently, the root, stem and leaf of this plant has been studied in detail for its anatomical variations. Stem anatomy is almost similar to other members of the family. The Transverse Section of the leaf shows the presence of accessory vascular bundles. Similarly Transverse Section of the root has revealed the occurrence of secondary growth. In normal conditions, secondary growth if occur, has to be present in other plant parts also e.g., stem. But it has not been observed in stem during present investigation. Further, stomata have not been found on any of the two epidermal layers. In addition, to best of our knowledge, no anatomical details has been worked out for this species in the past except one study made on root anatomy as discussed in the results. So present findings related to the anatomy of this particular species may be treated as pioneer.

Keywords:
Asteraceae, anatomy, leaf, root, stem, microscopy, *Silybum marianum*.

Web Address:
http://jresearchbiology.com/Documents/RA0149.pdf.

Article Citation:
Sidhu MC and Saini P.
Anatomical Investigations in *Silybum marianum* (L.) Gaertn.
journal of research in Biology (2011) 8: 603-608

Dates:
Received: 21 Nov 2011 /**Accepted:** 23 Nov 2011 /**Published:** 12 Dec 2011

Journal of Research in Biology

INTRODUCTION:

Silybum marianum (L.) Gaertn. was previously known as *Carduus marianus*. It is a member of family Asteraceae (Compositae) commonly known as Daisy family. Its synonyms are *Carduum marianum* L., *Carduus marianum* L. and *Cnicus marianum* L. Its botanical name is derived from a Greek word "Silybon"or "Silybos" which means tassel or tuft. It is an annual or biennial species occurring throughout the world. But Eeastwood (1901) designated it as a stout annual species having white patched leaves with prickly margins. *Silybum marianum* is an annual herb and many of its aerial parts bear thorns. Plant height may vary from place to place. It also grows as biennial species in certain areas. The stem is tall, erect and branched towards the apex. The leaves are alternate, large and glabrous. The seeds bear very fine, long silky and creamy white pappus. The fruits are referred to as achenes. Anatomical studies illustrate the internal organisation of the species and it has been placed between morphology and cell biology. According to Dilcher (1991), anatomical details are important for Palaeobotanists for the reconstruction of complete plant. Anatomical details can also be helpful in studying the systematic position and evolutionary relationship between the species (Dengler, 2002). Dhyani et al. (2009) while analysing the anatomical features of *Lilium polyphyllum* suggested that these types of studies are important for botanists, medical researchers and taxonomists. In case of a species like *Silybum marianum* which is of high medicinal importance, anatomical characterisation has great significance to avoid adulteration (Eltahir and AbuEReish, 2010). *Silybum marianum* has anticancer, antidiabetic, antidote to mushroom poisoning, anti-fibrotic, antihepatotoxic, antiinflammatory, antioxidant, hepatoprotective, hypocholesterolaemic, immunomodulatory, kidney protective, neuroactive and neuroprotective etc. properties (Corchete, 2008). Besides this it has also been used in various veterinary medicines. In addition to its benefits for human health, it also has some negative aspects. In several parts of the world, it is regarded as a toxic weed. To the best of our knowledge, till today the internal organisation of *Silybum marianum* has not been worked out in detail. Hence this area needs a thorough and extensive exploitation. Attempts have been made to study the internal structure of root, stem and leaves.

MATERIAL AND METHODS:

The plant material was collected from its natural habitats in Chandigarh, India. The root, stem and leaf were fixed in the fixative for their proper preservations. For longer duration these samples were shifted to 70% alcohol. The Transverse Sections of root, stem and leaf were obtained manually. Different sections were stained using safranin and fast green, then observed under the microscope. Photographs of the well stained sections showing cellular differentiation have been taken.

RESULTS AND DISCUSSION:

Attempts have been made to study the internal organization of root, stem and leaf of this species. The results obtained from the present investigations are as follows:

Stem:

The outline of Transverse Section of *Silybum marianum* stem is nearly circular. The outer most layer of the stem is epidermis which is single layered and cuticularised (Figure 1-a). The cortex is made up of collenchymatous and parenchymatous tissues. There is present a continuous layer of Chlorenchyma cells and Collenchymatous cells above the cortical cell layers. Endodermis is quite distinct. Its cells contain large starch grains and hence referred as the starch sheath. Multilayered pericycle is irregular and made up of large distinct sclerenchymatous cells (**Figure 1-c**). The pericycle encloses the vascular bundles. Vascular bundles are arranged in a ring (**Figures 1-a & b**). They are conjoint (xylem and phloem are lying on the same radius), collateral (xylem lies inwards and phloem outwards), closed (absence of cambial strip between the xylem and the phloem) and exarch (metaxylem towards centre and protoxylem faces the periphery) type. Xylem is further differentiated into protoxylem and metaxylem (**Figure 1-d**). The parenchymatous tissues constitute the conjuctive tissue between the two vascular bundles. The well developed parenchymatous pith is located in the centre (Figure1-a).

Root:

In Transverse Section of the root, the outer most layer is epidermis which is single layered and consists of thin walled parenchymatous cells. Some of these cells get elongated to form the new roots which are later designated as the lateral roots. Cortex is being depressed and reduced due to the occurrence of secondary growth in roots. There is

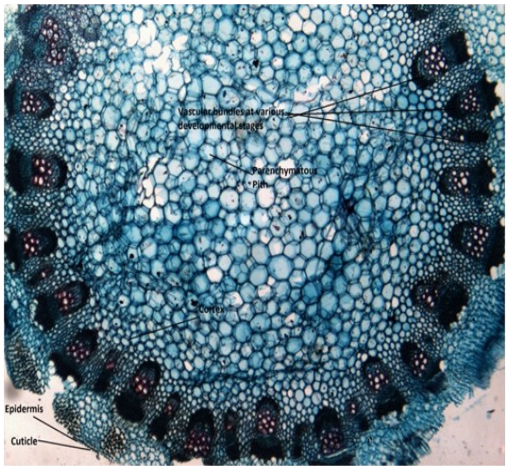

Fig. 1-a (T. S. Stem; Magnification - 4x)

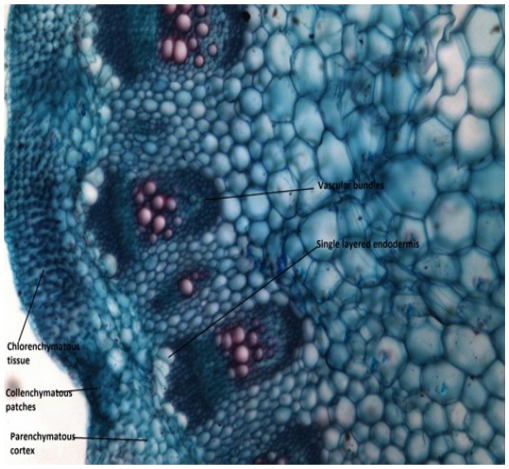

Fig. 1-b (T. S. Stem; Magnification - 10x)

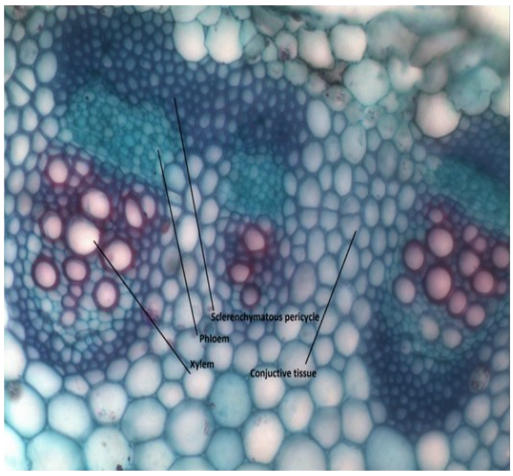

Fig. 1-c (T. S. Stem; Magnification - 20x)

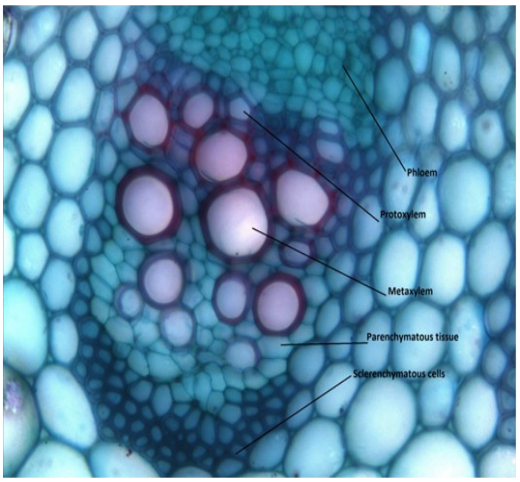

Fig. 1-d (T. S. Stem; Magnification - 40x)

present a discontinuous peridermal layer which later on differentiated into phellem and the phellogen (**Figure 2-a**). Only two to three layers of cortical cells are visible in reduced form. The formation of distinct layers of secondary xylem and phloem suggests the occurrence of secondary growth (**Figure 2-b**). Secondary phloem is red in colour while the primary phloem has been compressed and only some of its remnants can be seen. There are present distinct layers of cambial cells (2 layers) which are responsible for the secondary growth. The major portion of root is occupied by the secondary xylem which is distinguished by its vessels or xylem tracheids. Xylem tracheids are usually two to three celled. There are present distinct layers of thin walled ray parenchyma cells. Both of these components (vessels and ray parenchyma) form the secondary xylem while the primary xylem is present towards the centre in a crushed miniature form. In the centre there is present a small sized parenchymatous pith. It is surrounded by certain sclerenchymatous cells which forms a portion of the xylem tissue (**Figures 2-c & d**). The occurrence of secondary growth in the root seems to be an abnormal feature since *Silybum* growing in the present investigation area occurs as an annual species and formation of growth rings in an annual plant is an exception. Fritz and Saukel (2011) while studying the anatomy of subterranean organs of some medicinally used species observed the presence of secondary phloem. They were of the opinion that root anatomy of *Silybum marianum* is similar to that of *Onopordum acanthium* and *Cnicus benedictus* but preceding secondary growth was the distinctive feature of *Silybum marianum* root. Their findings substantiated the present observations regarding the secondary growth in the root portion only.

Leaf:

The leaves are alternate, large, white veined,

Fig. 2-a (T. S. Root; Magnification - 4x)

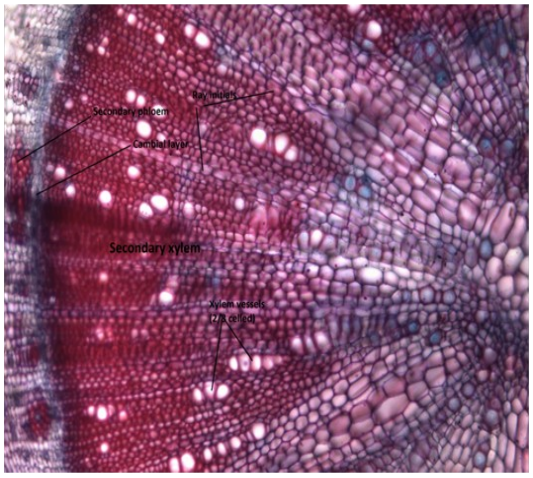

Fig. 2-b (T. S. Root; Magnification - 10x)

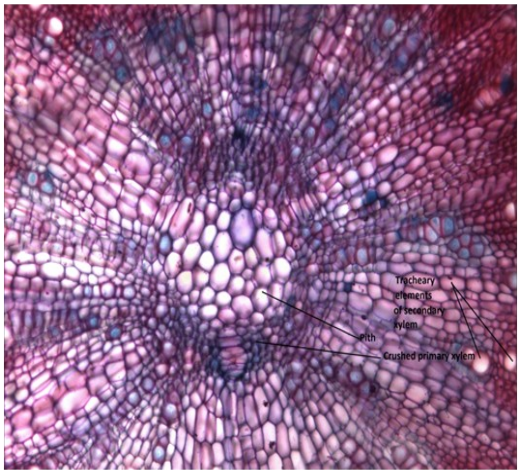

Fig. 2-c (T. S. Root; Magnification - 20x)

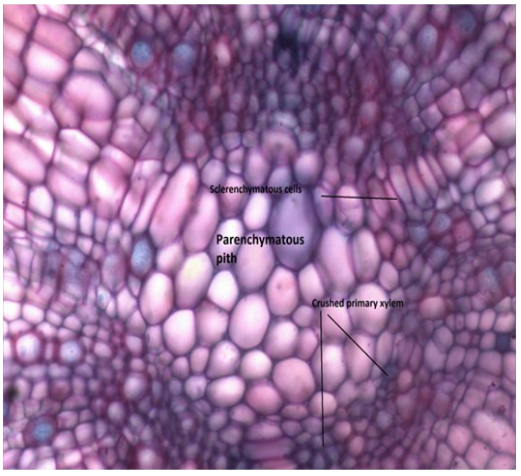

Fig. 2-d (T. S. Root; Magnification - 40x)

glabrous and have strong spiny margins. The main and an important feature is the presence of white streaks on the leaf surface. The leaf of *Silybum* is of dorsiventral type. The Transverse Section shows the presence of single layered upper and lower epidermis which is made up of compactly arranged barrel shaped cells. Both the surfaces are covered up with a thick and wavy cuticle. It is more on the upper epidermis and lesser on the lower epidermis (**Figure 3-a**). The stomata are usually absent in the epidermal layers which is a distinct feature. Inner to the epidermis present mesophyll cells which is differentiated into two to three layered parenchymatous palisade cells. It consists of green coloured columnar cells which are arranged in compact rows. Each palisade cell contains several chloroplasts positioned around its walls. A prominent and large vascular bundle (collateral and closed type) is present in the region of midrib. A layer of parenchymatous cells separates the vascular bundles from the epidermis. The vascular bundles are encircled by a parenchymatous bundle sheath cells (**Figure 3-c**). The upper and lower sides of the vascular bundle are covered by bundle sheath extensions and sclerenchymatous cells. In addition, there are present two to four layers of collenchyma cells near the upper and lower epidermis. Each vascular bundle consists of xylem present towards the upper epidermis and phloem close to the lower epidermis. Xylem is differentiated into metaxylem and protoxylem. Protoxylem vessels are smaller in size and facing towards the upper epidermis (**Figure 3-d**). Phloem has various components like sieve tubes, companion cells and phloem parenchyma cells. Along with the largest vascular bundle present in midrib, there are present certain accessory bundles which are smaller in size and they are present towards the upper epidermis near the wing of the leaf (**Figure 3-b**). These accessory vascular bundles may be developed to meet the requirements in greater amount through translocation system. For example to provide

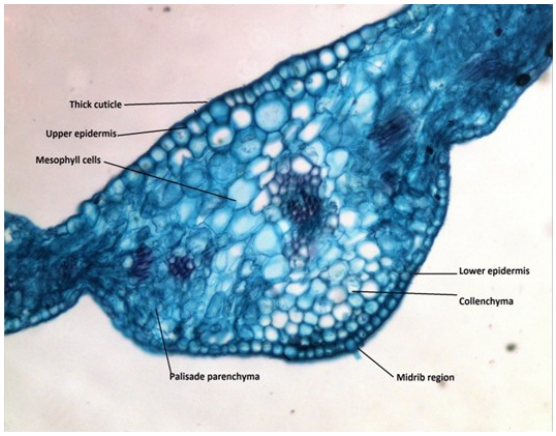

Fig. 3-a (T. S. Leaf; Magnification - 4x)

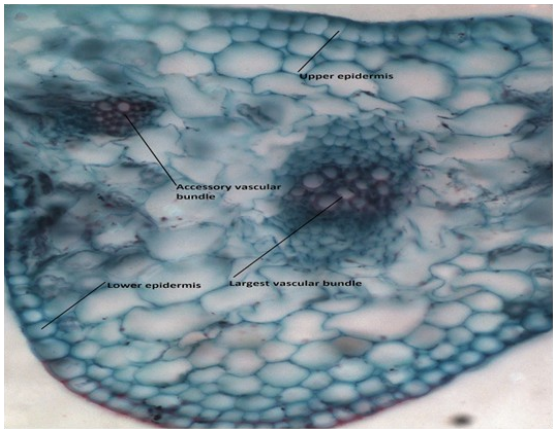

Fig. 3-b (T. S. Leaf; Magnification - 10x)

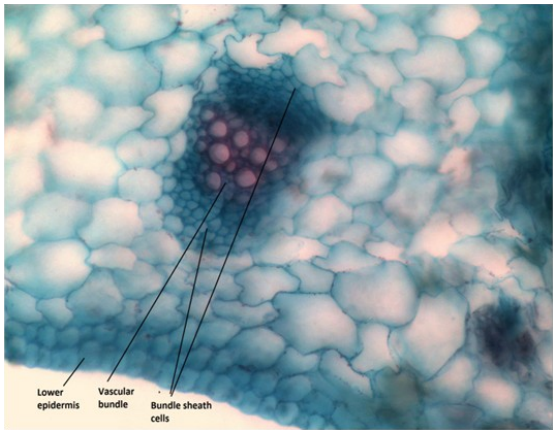

Fig. 3-c (T. S. Leaf; Magnification – 10x)

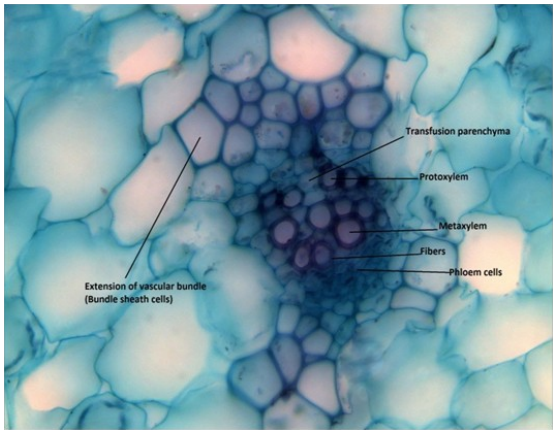

Fig. 3-d (T. S. Leaf; Magnification - 20x)

nourishment to the densely crowded flowers (Holroyd, 1928). The presently investigated species show profuse growth during winter season and flower nearly at the end of the winter season in this study area. Accessory vascular bundles must have been developed to overcome winters on reserve food material.

In the available literature, there is hardly any report on the complete anatomical studies of this species. We have come across only one publication (Fritz and Saukel, 2011) discussing the occurrence of secondary phloem in the root portion of this particular species. Since *Silybum marianum* is a plant of high medicinal importance both in traditional systems of medicines and in modern medical treatments, its anatomical characterisation is of great significance for its precise identification. So, the main contribution of *Silybum marianum* is its quite astonishing and vital medicinal properties due to which it is gaining huge importance in medical science. Due to large number of medicinal applications, it proved to be highly beneficial for human beings.

CONCLUSION:

Since in the present investigation area, plant usually occurs as an annual weed, preferably in the undisturbed regions, the exact reason behind the preceding secondary growth in the root portion is not clear. It can only be assumed that plant is shifting from its annual habit to the perennial ones as it has been reported (its biennial nature) in some parts of the world. The initiation of secondary growth in the root portion further confirm its changing habit towards perennial species nature since once the roots have retained viability, then it can give rise to new plants in the next season. This character (secondary growth) has not appeared in the stem portion of the species. So this is a case where only one plant part has shown the occurrence of secondary growth. Further investigations are in process to understand the reason behind this characteristic feature of the root. However, anatomical characterisation of this species is almost

similar to many other species. Leaf anatomy has shown the occurrence of some accessory vascular bundles. This character has already been reported by many researchers and it is there to meet the translocation requirements during the unfavourable conditions.

ACKNOWLEDGEMENT:

Authors are thankful to the Chairperson, Department of Botany, Panjab University, Chandigarh for providing necessary facilities during this work.

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