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ANATOMICAL AND BIOCHEMICAL STUDY OF *LACTUCA SERRIOLE* L. FROM THE ASTERACEAE SPECIES GROWN IN THE WEST OF IRAQ

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ABSTRACT : The present study was aimed to explore the important of *Lactuca serriole* L. (Synonym : *Lactuca scarioala* L.), were growth wildly in west of Iraq and used un traditional medicine. When we studied anatomical and Biochemically, the anatomically study description different internal tissues of the plant organs (leaf blades, petioles, stems and roots) and measured the details. These characteristics have include epidermis studies are considered important characters like cell forms which were square and compact with non- divisible bristles. The vascular system of the root, stem, petioles and leaves was of good structure with wide ribs and thick fibers. The chemical characteristics of the plant include a qualitative and quantitative identification of some active components such as alkaloids, glycosides, saponins and tannins has been carried out, Phenols with the highest ratio (7.88%) have been pathogenic tested for some pathogenical bacteria using the minimum inhibitory concentration method. A good inhibition activity was found on *Pseudomonas aeruginosa* with a rate of 603 mg/ml. This has been compared with some antibiotics, including ciprofloxin and erythromycin.

Key words : *Lactuca serriole* L., traditional medicines, Asteraceae species.

INTRODUCTION

Several types of plants are common in Iraq due to environmental and typographical variation extending from the north to the south passing through the middle regions of the country (Al-Rawi, 1964). In this regard, Rechinger (1968) states that anatomical characteristics have been used for classification purposes tens of years ago some modern classification for classification ranking have been adopted. The accurate active chemical content has a relationship with plant classification taxa. Moreover, it helps in differentiation between plants depending on the smell, tats, or both together. (Reasume, 2010). *Lactuca serriole* Linn. (Synonym: *Lactuca scarioala* L.) is a member of the Asteraceae family is one of such plants used in traditional medicine widely grown in Iraq and world (Uniyal *et al*, 2006). It is commonly called prickly lettuce, wild lettuce or milk thistle and has various local names among Nigerians. It is native to Europe, Africa, and Asia including Iraq. Mohammad (2013) and Al- Rajab (2015), furthermore the leaves are oblong or lanceolate, pinnated with fine spines along the veins and edges. They get progressively smaller as they reach the top of the plant and measure 3.5 – 25cm long by 1 – 20cm wide 3,4. The young leaves are eaten raw as salad or cooked, although it has somewhat bitter taste (Blamey, 2003). Prickly Lettuce is one of the medical wild herbs an antibiotic against inflammation, bacterial diseases,allergy, muscle activation,immunity, enhancer and antioxidant (Kim, 2001). This significance can be attributed to the active chemical content of *Lactuca serriole* particularly lactucine, lactucone, saponin, phenols, vitamins, beta carotene, iron and triterpenoid (Marco *et al*, 1992; Abiach and Marco, 2008).

MATERIALS AND METHODS

The plant aerial part of Prickly Lettuce collected from desert of western Iraq among flowering period in June 2017, after war the samples were dried under and then homogenized into fine powder using a mortar and pestle then stored in airtight (Sofowore, 2002).

Anatomical study: the fresh plant part is used in anatomical process. Studying the cross sections of species has depended on taking samples from roots, leaves and petioles from specific unified regions during field visits. These samples have been kept in FAA solution by adding

5 ml formalin, 5 ml of snow acetic acid and 95 ml of 70% ethyl alcohol for 24 hr. Then, the sample underwent a series of processes to obtain permanent slides according to the modified method of Nassur Allah (2007). Venation was studied by drying leaves using an alkaline solution (Al-Lami, 2002).

Phytochemical analysis : The presence of some chemical compounds in the pulverized samples of plants were determined using standard methods such as (tannins, flavonoids, saponin, carbohydrate, resin, alkaloids and phenolic compounds) (Evans, 2002; Omoregie *et al*, 2010).

Biological study : Phenolic compounds were extracted with a weight of 86 g of Lactuca leave powder of. These were put in a class flask of 1 lit capacity. Then, 600 ml of distilled water was added with 10 % v/ v of Hcl. Reflex extraction was carried out for 8 hrs. Then the extraction was filtrate and cooled and 50 ml of ethyl acetate was added to each 50 ml of the extract. This process was repeated for three times, then the extracts were centrifuged and dried using a rotary evaporator at 45°C. The extract was kept to carry out the inhibitions test with concentrations, 25mg for each 100ml, 300 mg for each 100 ml distil water and 400 mg for each 100 ml of distilled water. The concentrations were as follows : 25%, 300% and 400% for three concentrations according to the random design and LSD (Harporne, 1984). The inhibition activity was tested using the lowest concentration of phenolic compounds which observation a visible colony of bacterial growth after incubation at 37°C for 24 h. (Abu-Darwish *et al*, 2015). Antibacterial activities of phenolic compounds of prickly lettuce was used against *Staphylococcus aureus*, *Proteus vulgaris*, *Peudomonas aeruginosa* and *Escherichia coli were* obtained from Hospital of wamen Lab. The technique used in our study is the whole assay as described in Ayoola *et al* (2008). Which is believed to test the sensitivity of microbial strains to phenols on a nutrient broth media then it was compared with antibiotic (Amoxicillin, Erthromycin and Ciprofloxin) had standard active concentration (2.5 mg ml^{-1}). Moreover, the highest concentration affecting the bacteria was tested using nutrient agar (Rooppashree *et al*, 2008).

RESULTS AND DISCUSSION

Cross section of stem

The epidermis layer in the cross section of the stem (Fig. 1), cell form and cuticle layer covering it. The cuticle in the cross section appeared in a thin and plain form without bumps. The layer appeared with three to five rows of parenchyma cells in small to medium size. They took a circular and asymmetrical shape. As for the region of protuberances that appeared with a hight of five to eight rows in the cross section of the stem to form a collenchyma platelet texture, cells there were characterized by small size and oval form. Beneath this texture, there was a layer consisting of two rows of parenchyma cells of larger cells compared with the previous cells. In the bundle close to the vascular cylinder, lanticiferous ducts appeared surrounded with circular cells. The epidermis thickness was 104 micrometer. The cortex in the vascular cylinder was wide composed of thick cells. It formed a semicircular shape with the compune pack. Elements and transforming units of wood texture were of the radial order with close circular vein, there form changes to become asymmetrical towards the pith.

Cross section of leaf : Data of vertical section of the leave can be observed in Fig. 2. The vertical section of the blade leave were covered by waxy protective nature. The cuticle covering the epidermis, which were oval to circular and extended with 12 micrometers thickness. The palisade layer had no compact cells with small distainc separating then it was rectangular to oval extended in shape ordered in one to two rows. Thickness of the vertical layer was 125 micrometer. Cells of the sponge parenchyma layer were in extended ordered with five to six rows with a thickness of 78 micrometers. The middle vein was conical with a thickness of 440 micrometers. The number of transferring units in each pack was 12 and the number of vascular in the middle vein was three, one big vascular bundle in the middle of the cross section and two smaller on its sides.

Venations

Venation was of the brochdiodromous type as in Fig. 3, in which secondary veins don't end at the age. Rather, they are associated with series of prominent arches with one mid rib linearly extend along with the base of the leaf. Moreover, the two sides using extended leaning

Table 1 : Anatomical measurements of leaf *L. serriole*.

Middle vain			Mesophyll						Cuticle thickness
Number of phloem elements	Number of xylem elements	Thickness of vascular bundle	Sponge Thickness	Sponge layers	Palisade thickness	Palisade layers	Lower epidermis thickness	Epidermis thickness	
5	12	367	78	5	124	2	12	15	2.2

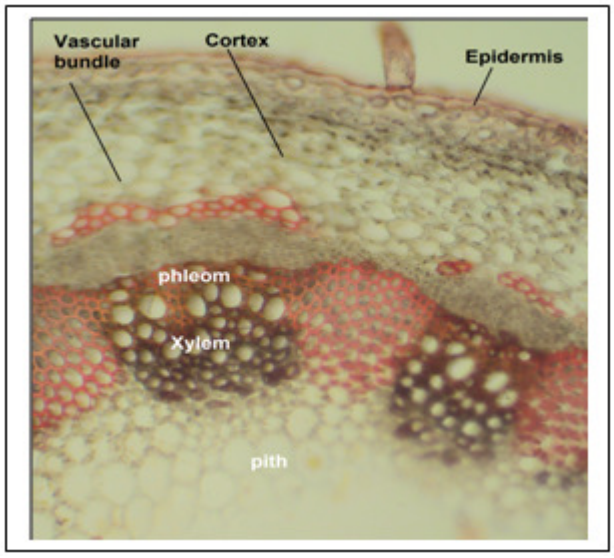

Fig. 1 : Transction of the *L. serriole* stem, 40x.

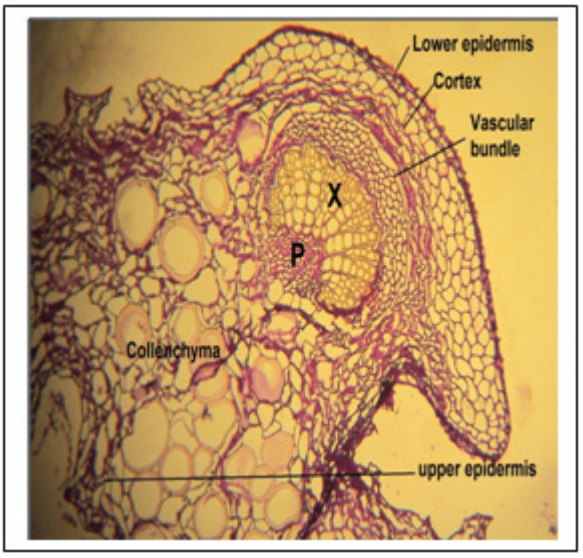

Fig. 2 : Transction of the *L. serriole* leaf, 40x.

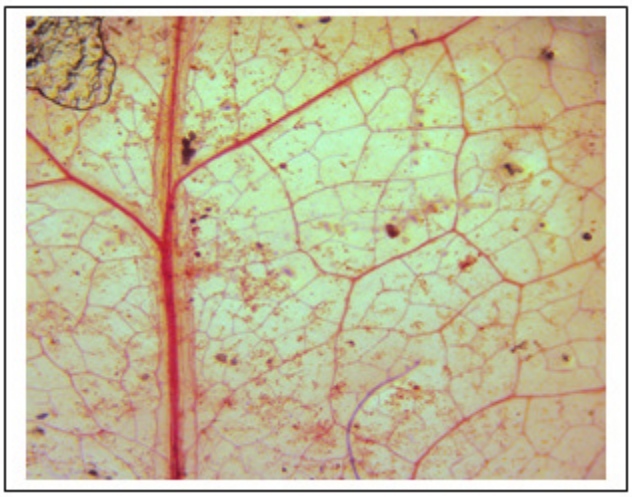

Fig. 3 : Surface section of the *L. serriole* leaf venation.

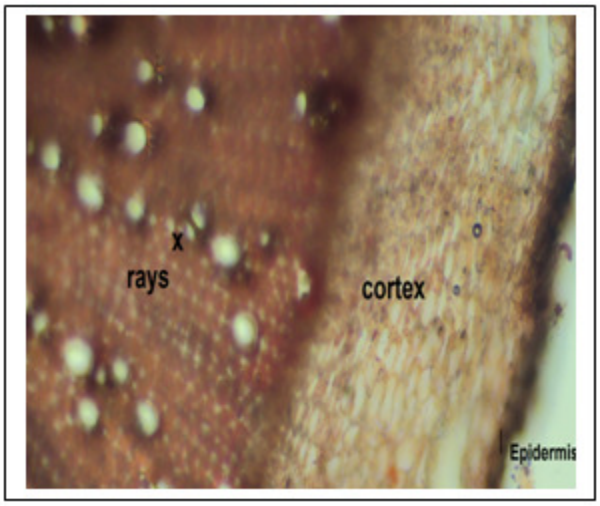

Fig. 4 : Transaction of the *L.serriole* root, x: xylem 40x.

toward the leaf edge. In its final third to unite with a series of branches at the age. The distance between the secondary and tertiary veins is almost non homogenous with the types. The areoles composed as a result of the meeting between tertiary veins are multifaceted. The areoles is of the complete and perfect type, whereby all veins constituting the areoles are perfect and constitute a vaccum. Ends of veins are either simple linear or simple curved.

Cross sections of root : Root was characterized as tap roots (Fig. 4), Table 2, centered in the vascular tissue including the xylem. The epidermis area was wide with several forms. It was extending outside and from within, it was square to oval. Lanticiferous ducts were spread between epidermis cells. The cortex took the form of undulate composed of square to extend and crossed cells with the epidermal tissue with oval cells. The cambium, on the other hand was narrow with crossed cells extending to those of the cortex. The xylem tissue next to the cambium was with square cells, which represent xylem parenchyma intersected with some transferring units which appeared in the form of pairs or groups. The center of the root was occupied with parenchyma cells which were ribbed with a thin wall. The narrow area which represents the center of the roots was occupied by the xylem texture in all types. Remains of primary wood, which were softer than the secondary wood can be observed. The primary xylem is pressed whereby only some compact veins can be seen.

Cross sections of petiole : The epidermis in the petiole appeared with circular cells followed by parenchyma tissue in the form of epidermis with circular to oval cells. The number of packs was 3 and 3-5 layers end as in Table 2. The numbers of transferring units were 4 to 7. Xylem in the transferring packs was with good texture, whereby transferring units were clearly visible

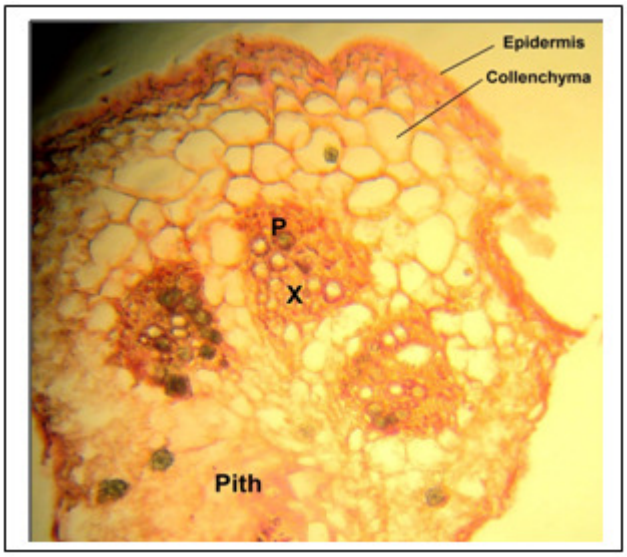

Fig. 5 : Transaction of the *L.serriole* leaf petiole, x: xylem, p: phloem 40x.

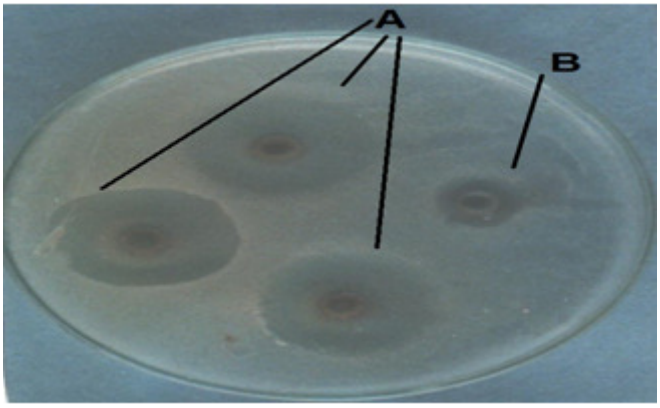

Fig. 6 : Inhibition diameter cons. 6.3 mg ml^{-1} from lettuce phenols in nutrient agar on bacteria *Pseudomonas aeruginosa* (A = Phenols extract, B = control ciprofloxin).

Table 2 : Anatomical measurements of stem, root and petiole of *L. serriole*.

Number of element per vessels	Number of xylem layers	Phloem thickness	Xylem thickness	Number of vascular bundles	Number of parenchyma layers	Number of collenchyma layers	Cortex thickness	Section format	Cuticle thickness
11	7	65	91	4	2	5	98	Circular with bumps	Stem
6	4	36	54	3	3	4	44	Circular irregular	Petiole
8	12	77	68	5	4	2	252	circular	root

Table 3 : Phytochemical screening of *L. serriole* leaves.

Phytochemical constituents	Amount %
Flavonoids	15.64
Carbohydrates	10.5
Saponins	5.22
Resin	2.29
Phenolic compounds	7.88
Alkaloids	13.6
Tannins	10.33

in arc – shape. Moreover, the cortex was with good texture whereby the fibers were clearly visible with bundle cap form as in Fig. 5.

Chemical testes results of *L. serriole* leaves showed that they contain active substances for flavonoids, tannins, clayoxides, alkaloids, saboons and flavodat as in Table 3. The presence of these active substances in the plan gives it great biological activity and ability in inhibiting pathogens as well as being antioxidant. This fact has been confirming it by Baloun *et al* (2017). The availability of flavonoids

Table 4 : Antibactrial activity *of L. serriole* phenolic [Minimum Inhibitory Concentration (MIC) measured (cfu ml^{-1})].

Tested bacteria	MIC(mg ml^{-1})			Antibiotic		
	25	30	40	Am	Er	Ci
Staphylococcus aureus	2.4	2.8	5.6	3.9	2.7	0.2
Proteus vulgaris	2.4	3.7	4.7	2.9	3.2	0
Escherichia coli	3.8	3,9	4.6	2.2	3.6	1.4
Pseudomonas aeruginosa	2.3	4.7	6.3	4.6	4.1	3.3

Am= Amoxicillin, ,Er= Erhtromycin, Ci= Ciprofloxin, LSD = 1.42

with this high percentage enables the plant to gain inhibitory significance against pathogenic organisms inflammations caused by bacteria and fungi (Margariteli, 2016; Al-rajab *et al*, 2018). Also, it enhanced the activity of the immunity system in the living organism (Kim, 2001).

Biological test

"Flavonoids are the most important type of active compounds and the most predominant in prickly lettuce. It is active against inflammations. Also, it inhibits the activity of cancer cells and many mutant material substances by removing free radicals and enhancing immunity (Haleem *et al*, 2013). The results as in Table 4 and Fig. 6 of MIC test and zones of inhibition showed significant effect on all pathogenic bacteria we were testing. Several studies have confirned the positive relationship between plants content's of flavonoids and its activity against pathogenic bacteria (Ulloa-Urizar *et al*, 2013; Janbaz *et al*, 2001).

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