Developing methods for anatomical identification of the genus Zygophyllum L. (Zygophyllaceae) in Saudi Arabia. Original research

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Abstract: Zygophyllum L., the largest genus of Zygophyllaceae comprises about 100 species known from the Mediterranean to Central Asia, more than eleven species growing in Saudi Arabia specially in desert and saline habitats. This study deals with taxonomy of Zygophyllum species growing in Saudi Arabia depending on anatomical characters of leaves and petioles to eleven investigated species. Anatomical features of leaves and petiole of the eleven investigated Zygophyllum species show characters of major importance such as stem outline, leaf outline, the arrangement of leaf vascular tissue, in addition to the number of vascular bundles in the inner leaf whorl, and characters of minor importance such as leaf mesophyll, and the branching of the main vascular bundle in the petiole Those characters enable us to separate Z. simplex from the other ten species by its cup shape transverse section in stem. Leaf vascular tissue is arranged either in straight line, or in two whorls . Petiole vascular bundles tissue arranged in two whorls the outer with number of vascular bundles, the inner whorl with two type: one main central vascular bundle and one main central vascular bundle associated with two peripheral vascular bundles. According to all those anatomical characters an artificial key explain the difference between the eleven investigated Zygophyllum species. Life Science Journal. 2011;8(3):451-459] (ISSN: 1097-8135). http://www.lifesciencesite.com.

Keywords: Zygophyllum species, anatomical features, leaf vascular tissue

Introduction

The genus Zygophyllum L., the largest genus of *Zygophyllaceae*, comprises about 100 species known from the Mediterranean to central Asia, South Africa and Australia. It includes perennial shrubs or under-shrubs, with succulent cylindrical rarely flattened leaves, simple or 1-2 foliate (Boulos 2000).El Hadidi (1977) described *Zygophyllum migahidii* and *Z.mandavillei* as new species to Saudi Arabia. Migahid (1978) recognized 7 species of *Zygophyllum* grow in Saudi Arabia: *Zygophyllum album, Z. coccineum, Z. decumbens, Z. gaetulum, Z. mandavillei, Z. migahidii* and *Z. simplex* . In her revision of the genus, Hosny (1978) added another 5 species of *Zygophyllum* viz. Z. *aegyptium, Z. berenicense, Z. propinquum. Z. qatarense* and *Z. hamiense*. Collonette (1999) added the following 2 species: *Z. boulosii* and *Z. fabago*. Ahmed (1989) carried out an anatomical study on *Zygophyllum album* in Egypt and reported the effect of the different ecological factors on the stem growth. These anatomical characters mentioned in her study highlighted the importance of investigated these features of the *Zygophyllum* species growing in Saudi Arabia which can't be done before.

Therefore the aim of this works: 1- Investigate the anatomical characters of each of the eleven *Zygophyllum* species growing in Saudi Arabia; 2-Based on these anatomical features recognized, an artificial key utilizing major and minor characters for the diagnosis of these species will be developed.

1. Materials and Methods:

2.1. Sample collection:

Fresh materials of 9 *Zygophyllum* species growing wild in sand plain region and salt marches were collected. Attention was paid for studying specimens who were collected from localities representing the geographical range of each species (Table 1). Two herbarium specimens were studied where the fresh materials was not available during this seasons. The collected materials were identified according to Migahid (1978) and Chaudhary (2001). Samples of the identified materials were kept at Botany Department Faculty of Science King Abdul-Aziz University (Girls section).

2.2 Sample preparation:

For anatomical investigations specimens were fixed in formalin- glacial acetic acid- ethyl alcohol .Petioles and leaves blade were microtome serially at 10-15 **µ** after being embedded in paraffin wax. Sections were stained in saffranin and light green dehydrated in alcohol-xylol series, cleared in clove oil and mounted in Canada balsam, photographed by Nikon Microscope.

3. Results:

3.1. Anatomical result:

In table (2&3) the 11 investigated *Zygophyllum* species were arranged according to systematic treatment followed by Van Huyssteen (1937). Two species viz. *Z. decumbens* and *Z. simplex* belong to section Bipartita of subgenus *Agrophyllum*, while *Z.album ; Z. boulosii ; Z. coccineum; Z. hamiense; Z. mandavillei; Z. migahidii; Z. propinquum. Z. qatarense* belong to section Mediterranea of the same subgenus. The last row comprise *Z. fabago* belong to section Fabago of subgenus *Zygophyllotypus*, the anatomical characters are arranged horizontally.

Table (1): The collected specimens of Saudi Arabia *Zygophyllum* species

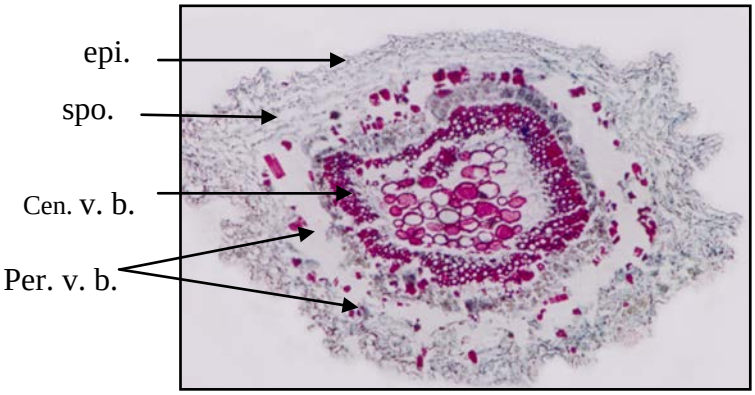
Species	***Locality***	***Date of collection***
1-Z. album	El-Sheaba 90 Km south Jeddah South-Obher 30Km south Jeddah	15/2/2003 20/2/2003
2-Z. boulosii	North –Obher 20 Km north Jeddah	12/7/2003
3-Z. coccinum	North–Obher 40Km north Jeddah South-Obher 30Km south Jeddah Jeddah-Makka road	7/3/2003 20/2/2003 25/3/2003
4-Z. mandavillei	East part Abou-Hedrea road	12/2/2003
5-Z. migahidii	EL-Raid 20Km south El-Mattar road	15/4/2003
6-Z. qatarense	East part Abou-Hedrea road	12/2/2003
Z. simplex 7-	El-Sheaba 90 Km south Jeddah Jeddah-Makka road	15/2/2003 25/3/2003
8-Z. propinquum	Al-Shaiba, 80 Km south of Jedda	9/4/1999 Herbarium specimens
9- Z. fabago	Al-Asawia,10Km before Tabarjal, S.Abedin,M.Al-Tahya, M.Al-Said,5-iv-88(19038-RIY)	Herbarium specimens
10-Z. hamiense	10 km north Dammam road	22/4/2003
11-Z. decumbens	20Km North west Jeddah	17/3/2003

Table (2): Petiole anatomical characters of the Zygophyllum species in Saudi Arabia

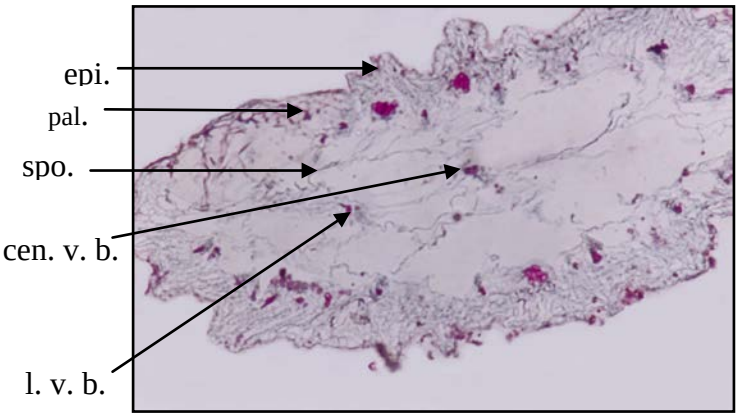
Characters	*Agrophyllum*										*Zygophllotypus*
	Bipartita		*Mediterranea*								*. fabago*
	Z. decumbens	*Z. simplex*	*Z. album*	*Z . boulosii*	*Z. coccineum*	*Z. hamiense*	*Z . mandavillei*	*Z . migahidii*	*Z. propinquum*	*Z. qatarense*	*Z. fabago*
Out line	Cylindrical wavy margin	--------	Cylindrical	linear	kidney	kidney	linear	Cylindrical	Cylindrical	Cylindrical	winged irregular
Mesophyll	unilateral	--------	bilateral	bilateral	bilateral	bilateral	bilateral	bilateral	bilateral	bilateral	unilateral
Main vascular tissue	central asymmetric vascular cylinder	--------	central cress shape V.B. accompanied by 2 lateral V.B.	central tribranched V.B. accompanied by 2 lateral V.B.	central cress shape V.B. accompanied by 2 lateral V.B.	only central V.B.	central cress shape V.B. accompanied by 2 lateral V.B.	only central cress shape V.B.	central cress shape V.B. accompanied by 2 lateral V.B	central cress shape V.B. accompanied by 2 lateral V.B	complete vascular cylinder accompanied by 2 lateral V.B

Table (3): Leaf blade anatomical characters of the *Zygophyllum* species in Saudi Arabia

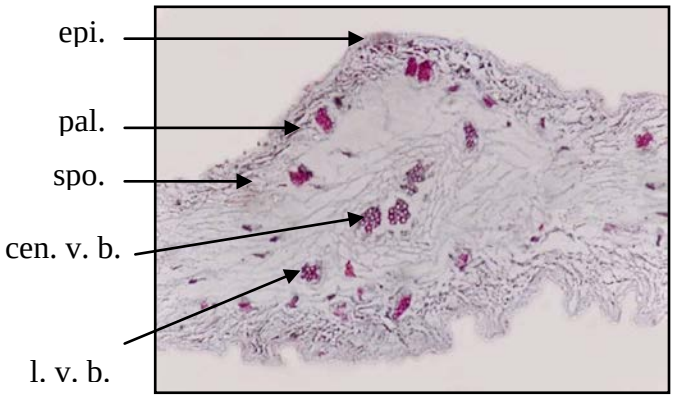
Characters	Agrophyllum										Zygophllotypus
	Bipartita		*Mediterranea*								*. fabago*
	Z. decumbens	*Z. simplex*	*Z. album*	*Z . boulosii*	*Z. coccineum*	*Z. hamiense*	*Z . mandavillei*	*Z . migahidii*	*Z. propinquum*	*Z. qatarense*	*Z. fabago*
Out line	winged linear	elliptical	elliptical	linear	kidney	kidney	linear	linear	elliptical	linear	winged linear
Mesophyll	unilateral	bilateral	bilateral	bilateral	bilateral	bilateral	bilateral	bilateral	bilateral	bilateral	unilateral
Main vascular tissue	central V.B. accompanied by 6 lateral V.B. at same level	only central V.B.	central dichotomous V.B. accompanied by 2 lateral V.B.	central V.B. accompanied by 2 lateral V.B.	5-7 central separated V.B accompanied by 2 lateral V.B.	5-7 central separated V.B.	central V.B. accompanied by 2 lateral V.B.	central V.B. accompanied by 1 lateral V.B.	only central dichotomous V.B.	only central cress shape V.B.	complete vascular cylinder accompanied by 5 lateral V.B .in each wing
Crystals	----------	druses	----------	----------	druses	druses	----------	----------	druses	druses	----------



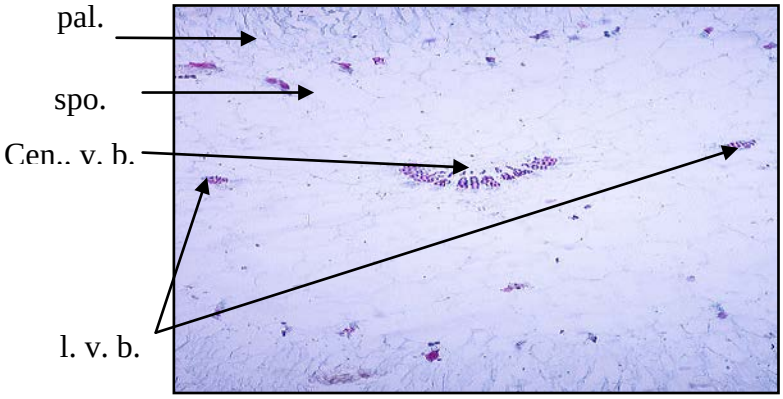
T.S. in *Z. decumbens petiole* x 50
Figure.2b



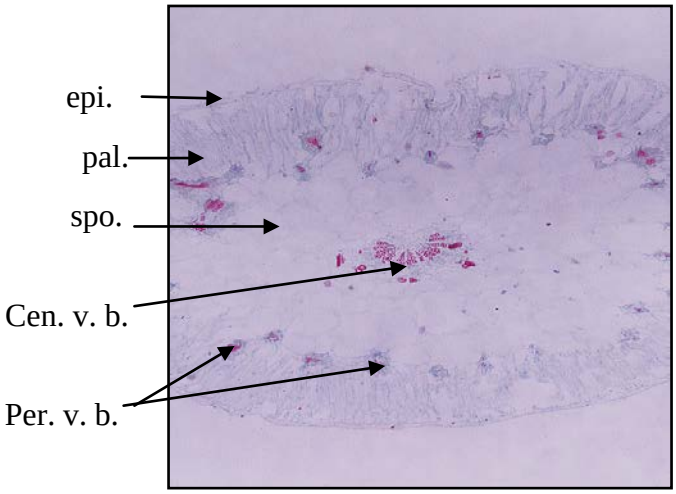
T.S. in *Z. mandavillei petiole* x 50
Figure.3b



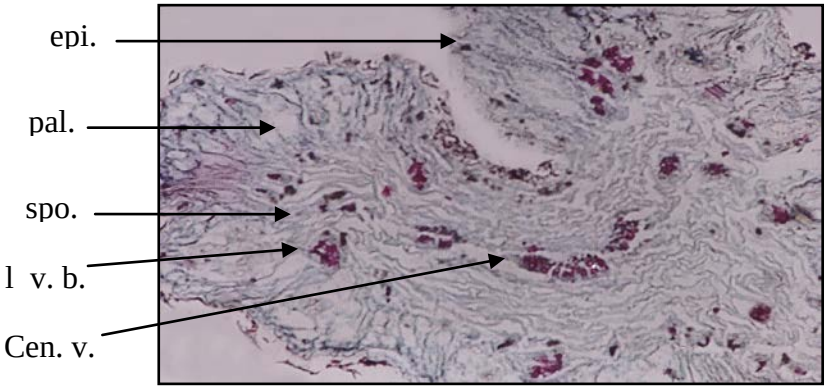
T.S. in *Z. boulosii petiole* X50
Figure.4b



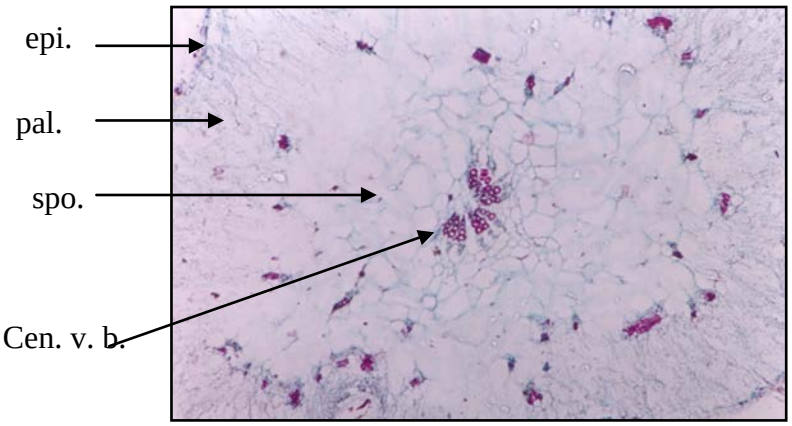
T.S. in *Z. album petiole* X 50 Figure.5b



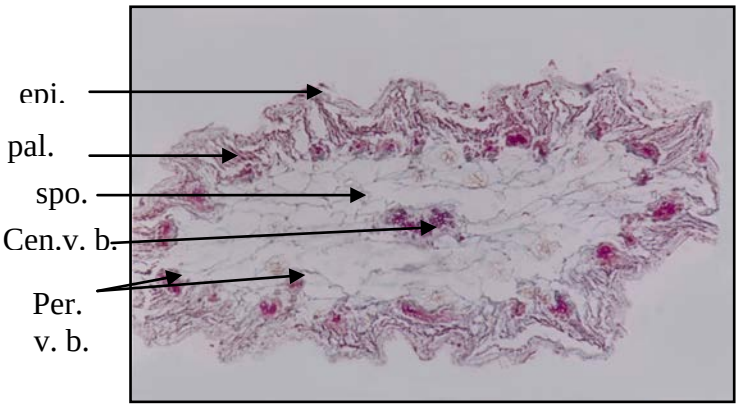
T.S. in *Z. migahidii petiole* x 50 Figure.6b



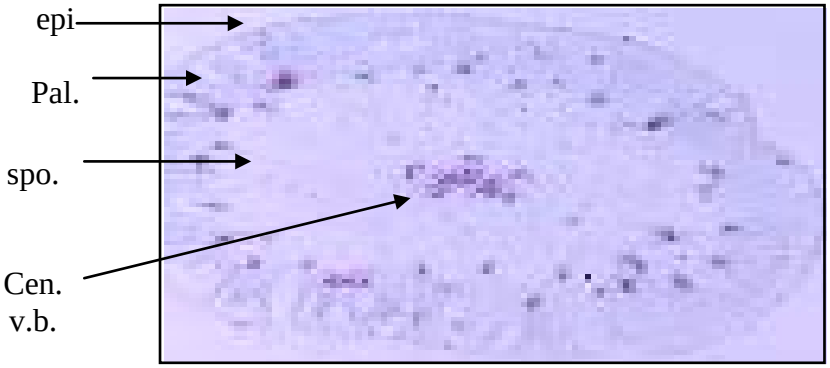
T.S.in *Z. propinquum petiole* x 50 Figure.7b



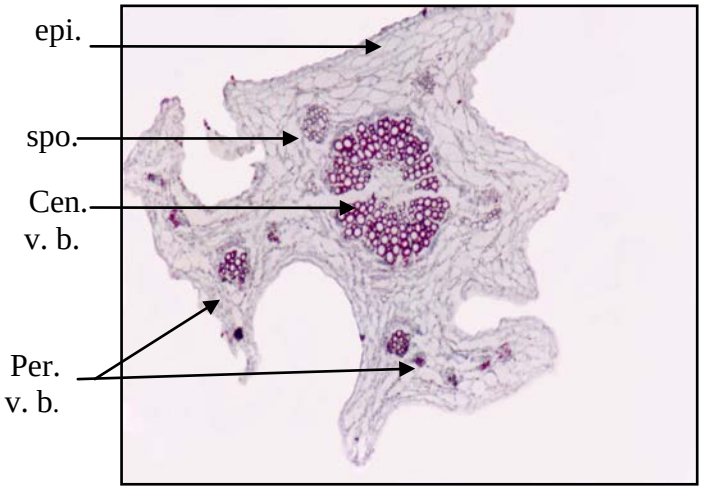
T.S. in *Z. coccineum petiole* X50
Figure.8b



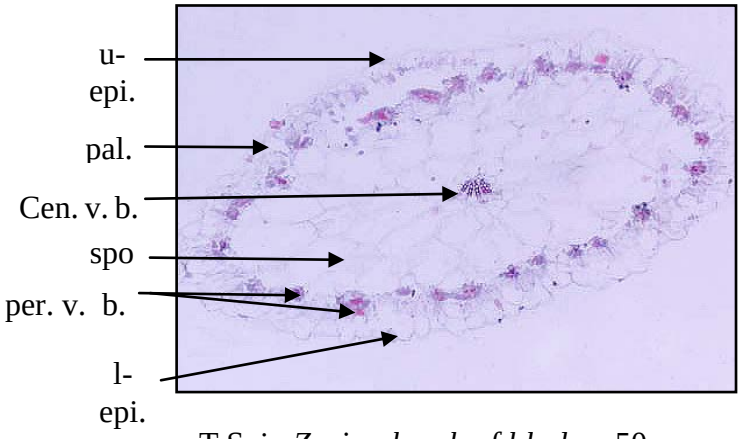
T.S. in *Z. hamiense petiole* x 50 Figure.9b



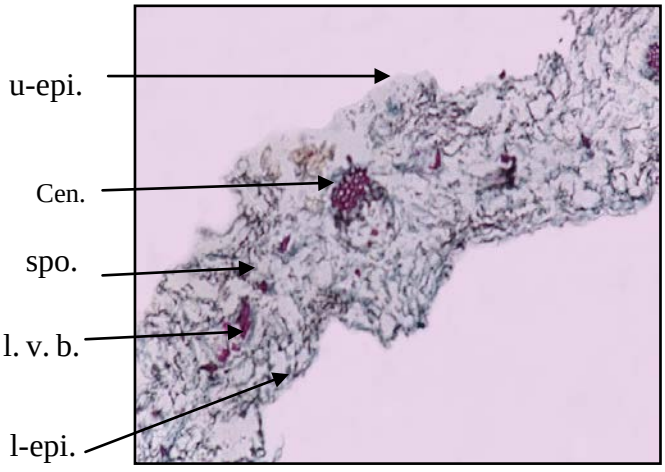
T.S. in *Z. qatarenes petiole* x 50 Figure.10b



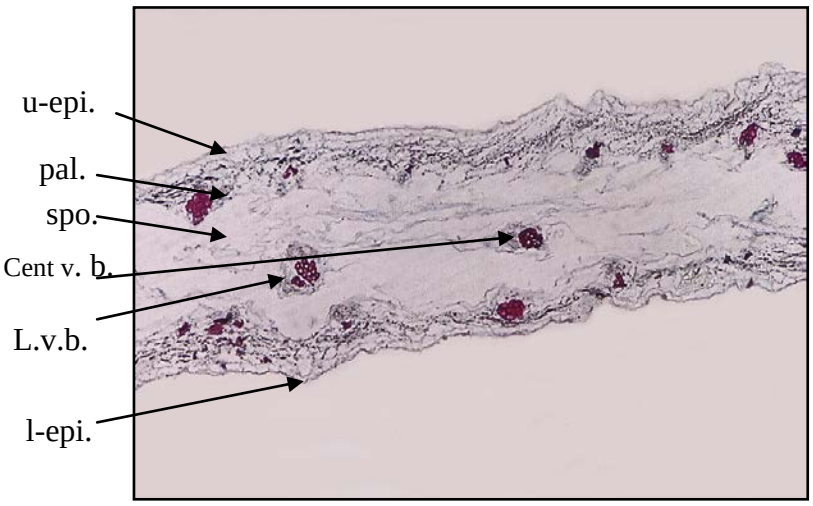
T.S. in *Z. fabago petiole* x 50 Figure.*11b*



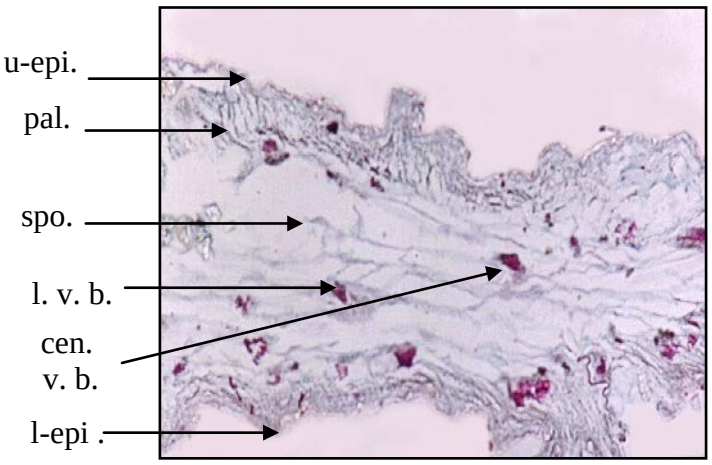
T.S. in *Z. simplex leaf blade* x 50
Figure.1



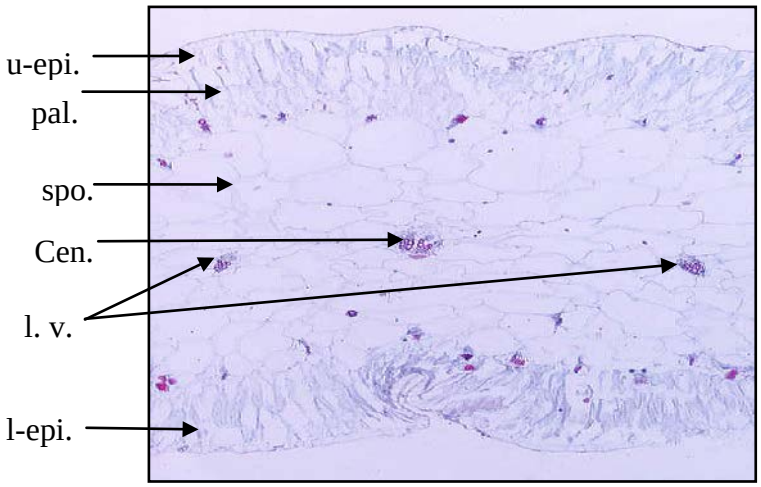
T.S. in *Z. decumbens leaf blade* x 50
Figure.2a



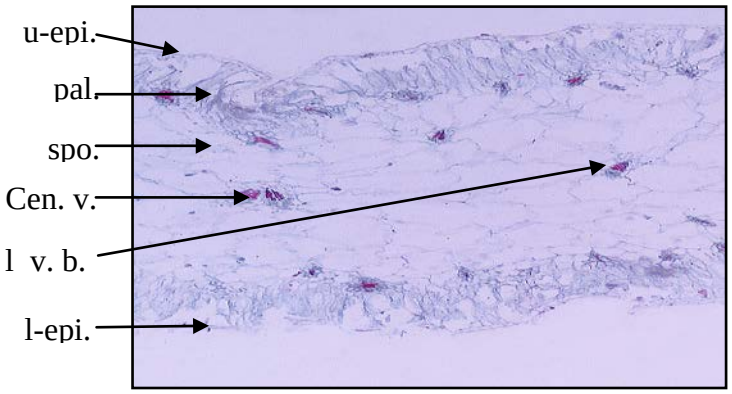
T.S. in Z. mandavillei leaf blade x 50 Figure.3a



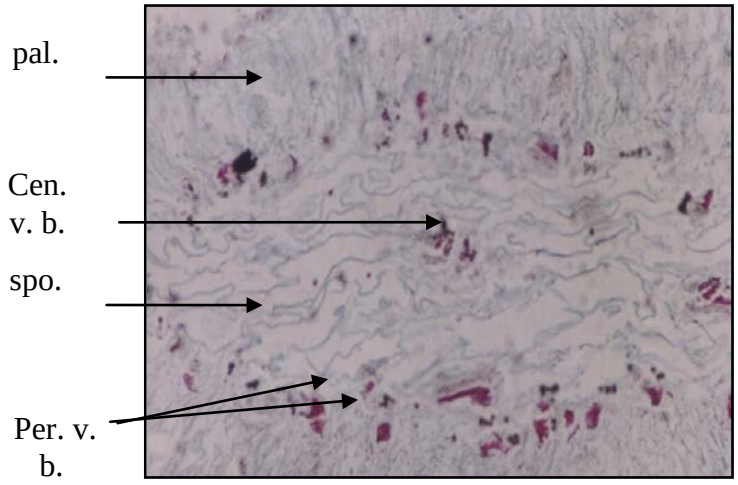
T.S.in *Z. boulosii leaf blade* X50 Figure.4a



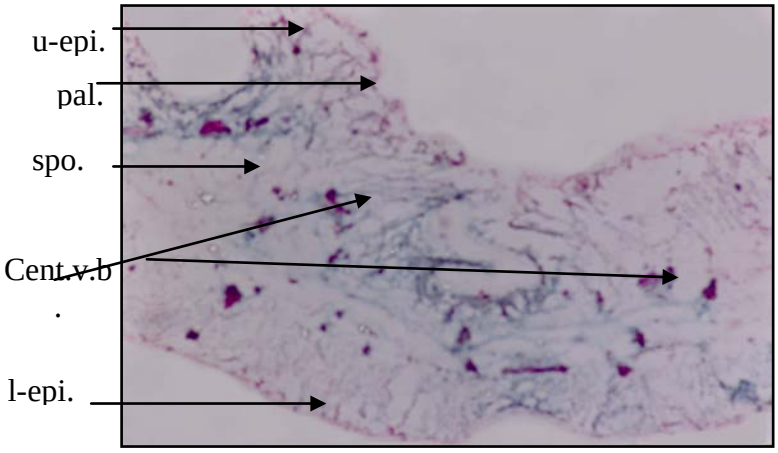
T.S. in *Z. album leaf blade* X 50 Figure.5a



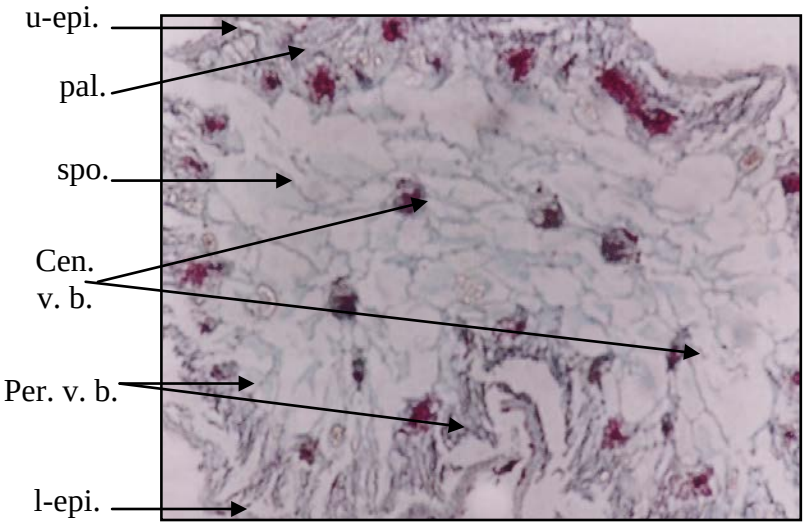
T.S. in *Z. migahidii leaf blade* x 50 Figure.6a



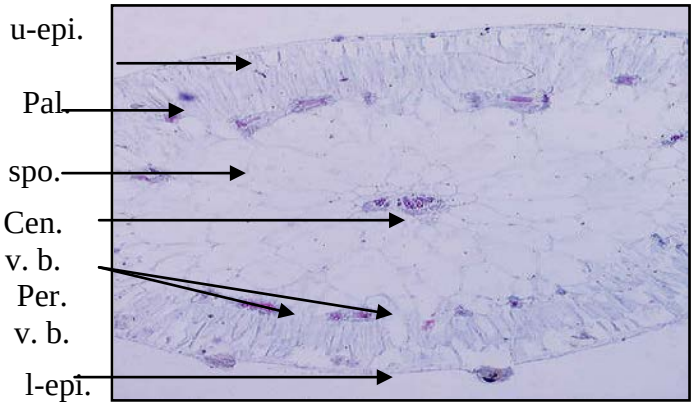
T.S. in *Z. propinquum leaf blade* x 50 Figure.7a



T.S.in *Z. coccineum leaf blade* X50 Figure.8a



Z. hamiense leaf blade x 50 T.S. in Figure.*9a*



T.S. in *Z. qatarenes leaf blade* x 50 Figure.10a

3.2. Statistical results:

In table (4 &5) and figure (12&13) the distribution of lateral vascular bundles and their position relative to the central vascular bundle varied from one species to another in leaf blade except in Z. *boulosii and Z. mandavillei* .were similar. Also the distribution of lateral vascular bundles and their position relative to the central vascular bundle varied from one species to another in leaf petiole except in *Z. album, Z.coccineum, Z. mandavillei, Z propinquum and Z.quatarense* which are similar

Table (4)Tally for Discrete Variables: VB in Leaf blade

VB type	Count	Percent
5-7centra VB +lateral VB	1	9.09
5-7central VB+2lateral VB	1	9.09
central cress VB	1	9.09
central di VB	1	9.09
central di VB +2 lateral VB	1	9.09
central VB	1	9.09
central VB+1lateral VB	1	9.09
central VB+2lateral VB	2	18.18
central VB+6lateral VB	1	9.09
complete VB + 5 lateral VB	1	9.09
N= 11		

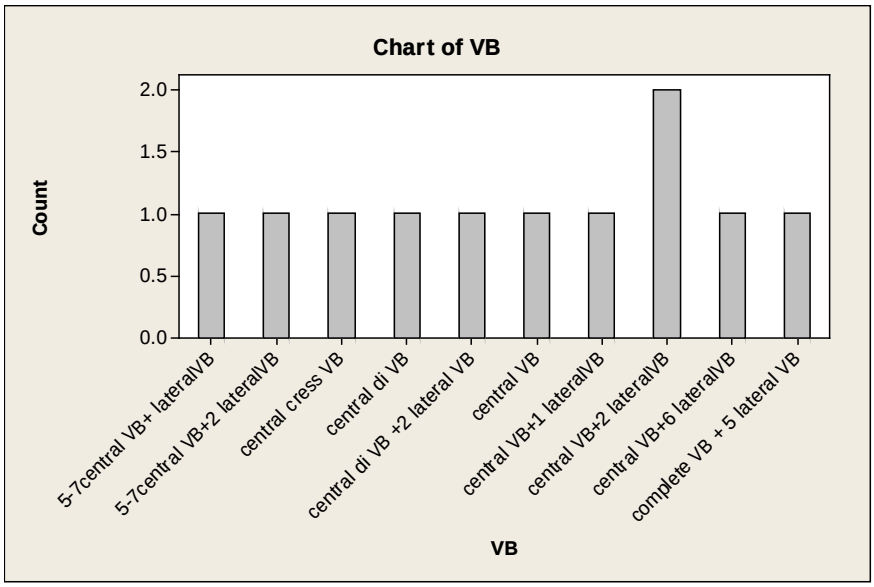
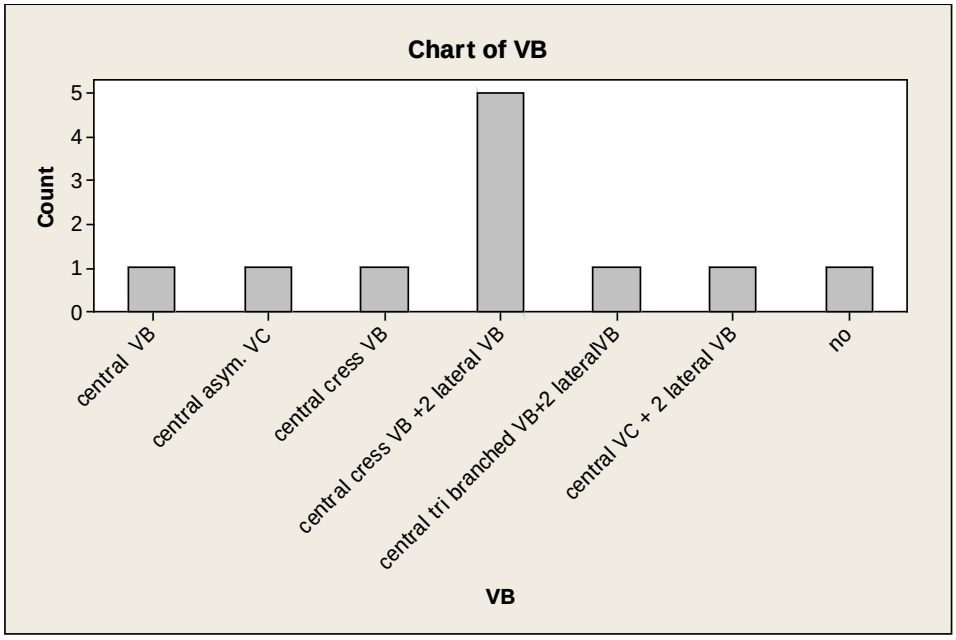


Figure (12) Position of lateral Vascular bundle in relation to central vascular bundle in leaf blade

Table (5) Tally for Discrete Variables: VB in petiole

VB type	Count	Percent
central VB	1	9.09
central asymmetric VB	1	9.09
central cress VB	1	9.09
central cress VB +2 lateral VB	5	45.45
central tri branched VB+2lateral VB	1	9.09
central VC + 2 lateral VB	1	9.09
no petiole	1	9.09
N= 11		



Figure(13) Position of lateral Vascular bundle in relation to central vascular bundle in leaf petiole

4. Discussion

Vascular tissue in both leaf blade and petiole are proved to be the most significant characters among the investigated taxa. It is to be noted that the vascular system is composed of two whorls the inner whorl with main or central vascular bundle or bundles accompanied with number of lateral vascular bundles and the outer one with number of peripheral vascular bundles representing the veinlets. Comparing the leaf blabe characters of both *Zygophyllum* species of section Bipartita: *Z. simplex* (Figure. 1) have only one central vascular bundle while *Z. decumbens* (Figure. 2a) have one central vascular bundle accompanied by 6 lateral vascular bundles 3 in each wing . In species of section Mediterranea vascular tissue show high variation: *Z. mandavillei* (Figure. 3a)and *Z. boulosii* has (Figure. 4a) one central vascular bundle accompanied by 2 lateral vascular bundles, while the central vascular bundle in *Z.album* (Figure. 5a), *Z. migahidii* (Figure. 6a) and *Z. propinquum* (Figure. 7a) is dichotomously branched. In *Z. coccineum* (Figure. 8a) and *Z. hamiense* (Figure. 9a) the central vascular tissue is subdivided into number of vascular bundles. *Z. qatarense* (Figure. 10a) which *belong* also to section *Mediterranea* take a cress shape central vascular bundle. *Z. fabago* (Figure. 11a) (section Fabago of subgenus *Zygophyllotypus*) shows a distinct complete vascular cylinder in both leaf blade and petiole

Vascular tissue in petiole of the *Zygophyllum* species show a less variation even *Z. simplex* is sessile all the other species has one central vascular bundle accompanied by 2 laterals. Shape of the central vascular tissue range between an asymmetric cylinder in *Z. decumbens*, (Figure. 2b) tribranched in *Z. boulosii* (Figure. 3b) and a cress shape in all other lifted species. Table (2)&(3) also show other characters viz. petiole and leaf blade out line, type of mesophyll and presence or absence of crystals, which are of less systematic value and can be used for the distinction of certain species. Leaf blade out line ranging between winged linear in *Z. decumbens* and *Z. fabago* and linear in, *Z. mandavillei; Z. migahidii* and *Z. qatarense*, while it is elliptical in *Z. simplex*, *Z.album* and *Z. propinquum* , it takes an irregular kidney shape in *Z. coccineum* and *Z. hamiense* .Petiole out line takes an irregular cylinder shape with wavy margin in all species except elliptical in *Z. mandavillei* (Figure.3b) and linear in *Z. boulosii*(Figure.4b) . Type of mesophyll in both leaf blade and petiole is identical, unilateral in *Z. decumbens* (Figure. 2b) and *Z. fabago* (Figure.11b) and bilateral in all other species. Druses crystals can be seen in *Z. simplex, Z. qatarense* (Figure. 10b) *Z. coccineum* (Figure 8b) , *Z. hamiense* and *Z. propinquum.* (Figure. 7b) Utilization of artificial key based on anatomical diagnosis is specially recommended in seasons where the plants are devoid of flowers

5. Conclusion

In view of the previous results differentiating the various anatomical characters on leaf blade and petiole the following key shows the efficiency of such characters between the 11 investigated species of *Zygophyllum.*

1.a. Leaf blade vascular tissue of one central vascular bundle----------2
b Leaf blade vascular tissue of separated vascular bundles ----------10
2. a.Central vascular bundle not accompanied with lateral V.B. ----------3
b. Central vascular bundle accompanied with lateral V.B. ----------5
3.a. Central vascular bundle dichotomous branching ---------- *Z. propinquum*
b. Central vascular bundle simple ----------4
4.a.. cress shape---------- *Z. qatarense*
b .Not as above ---------- *Z. simplex*
5.a. Central vascular bundle dichotomous branching----------6
b. . Central vascular bundle simple----------7
6.a. Central vascular bundle accompanied with 2 lateral V.B-------- *Z.album*
b. Central vascular bundle accompanied with 1 lateral V.B---- *Z. migahidii*
7 .a.Central vascular bundle accompanied with 2 lateral V.B----------8
b. Central vascular bundle accompanied with more than 2 lateral V.-------9
8.a. Central petiole vascular bundle tribranched---------- *Z. boulosii*
b. Central petiole vascular bundle not as above---------- *Z. mandavillei*
9.a.. Asymmetric vascular cylinder in petiole---------- *Z. decumbens*
b. Complete vascular cylinder in petiole---------- *Z. fabago*
10.a. Central vascular bundle accompanied with lateral V.B-- *Z. coccineum*
b Central vascular bundle not accompanied with lateral V.B- *Z. hamiense*

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