ANATOMICAL CHARACTERS AND THEIR TAXONOMIC IMPORTANCE IN ACANTHOLIMON

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ABSTRACT. This paper deals with the anatomical structures of stem, inflorescence axis and leaves of *Acantholimon*. The range of variation in general anatomical features of these organs is described and their taxanomic significance briefly evaluated. The primary aim of the present investigation is to look for neglected endomorphic features which can be useful taxonomic characters when used in conjunction with exomorphic ones.

INTRODUCTION

Acantholimon is one of the most intractable Plumbaginaceous genera in which some sections are delimited on morphological characters lost in herbarium material, and some species are so exceptionally variable that it is easy to place them in the wrong section. The greatest problem at sectional level arises in Sect. *Tragacanthina* in which "heteromorphic" leaves (fleshy, early-evanescent spring leaves and rigid, persistent summer leaves) are a diagnostic character. This character is constant and useful if species are examined in the field in early spring, but the early-evanescent nature of the spring leaves makes it difficult or impossible to refer species to this section from herbarium material. Owing to the paucity of vegetative and floral characters and their notorious variability, the present study sought to explore the possibility of using anatomical diversity to support taxonomic characters. Anatomical investigation has revealed that species belonging to Sect. *Tragacanthina* have a diagnostic summer leaf anatomy, so this section can be safely delimited on leaf anatomy in the absence of spring leaves from the herbarium material. Leaf anatomy has also been found to be useful as a supporting character in delimiting some other sections or even distinguishing some critical species.

MATERIAL AND METHOD

This account is mainly based on a study of herbarium material, except for *A. glumaceum* and *A. androsaceum* of which herbarium and fresh material was studied. The techniques used for preparing stem, inflorescence axis and leaf were the same as those described for *Limonium* (Bokhari, 1970). For the sake of convenience, hand sections were taken at different levels of the inflorescence axis and from the middle of the leaf sheath and lamina. A total of about 100 species was examined in nine sections. The following lists only those mentioned in the text.

I. Sect. *Gontscharovia* Lincz.: *A. inerme* Rech. f. & Koeie; *A.saxifragifolium* Rech. f. & Koeie
II. Sect. *Glumaria* Boiss.: *A. bromifolium* Boiss.
III. Sect. *Pulvinaria* Boiss.: *A. diapensioides* Boiss.

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IV. Sect. *Tragacanthina* Bunge: *A. curviflorum* Bunge; *A. polystachyum* Boiss.; *A. quinquelobum* Bunge
V. Sect. *Dracogyna* Mobayen: *A. tomentellum* Boiss.
VI. Sect. *Acmostegia* Bunge
VII. Sect. *Cymaria* Bunge: *A. cymosum* Bunge
VIII. Sect. *Staticopsis* Boiss.: *A. acerosum* (Willd.) Boiss.; *A. androsaceum* (Jaub. & Sp.) Boiss.; *A. calvertii* Boiss.; *A. confertiflorum* Bokhari; *A. glumaceum* (Jaub. & Sp.) Boiss.; *A. huetii* Boiss.; *A. hypochaerum* Mobayen; *A. saxifragiforme* Mobayen; *A. venustum* Boiss.
IX. Sect. *Acantholimon*: *A. bracteatum* (Girard) Boiss.

The following account deals with the anatomical variation in the vegetative parts of the plant which have been found of special interest and most useful in the classification.

Stem anatomy

In the young stem, collateral bundles of different sizes are quite distinct, are present in a ring and are relatively widely spaced. Metcalf & Chalk (1950) examined *A. glumaceum*; I have also studied it, including the type specimen, and have noticed that besides the characters mentioned by Metcalf & Chalk, there are distinctive groups of fibres (fibrous bundles) scattered in the xylem region of the stem. Such fibrous bundles are also reported by Labbe (1962). In fact, these fibrous bundles in the stem are correlated with some morphological characters. In the species with branched spikes, e.g. *A. curviflorum* (fig. 1C) and *A. glumaceum* the fibrous bundles are present in the stem while in the species with unbranched spikes, e.g. *A. acerosum* (fig. 1B) and *A. bracteatum* (fig. 1D) the fibrous bundles are absent from the stem. Usually these fibrous bundles are confined to the xylem region, but rarely, as in *A. huetii*, they are also in the tissue immediately outside the phloem.

There is a distinct periderm which is subepidermal in origin. The pith and cortical cells are filled with some substances of unidentified nature. These contents were formerly regarded as of the nature of plumbagin (Solereder, 1908), but recently Harborne (1967) has shown that plumbagin is uniformly absent from all the genera belonging to the Tribe *Staticeae*.

Anomalous structural feature of the stem. Kruger (1884) was the first to discover the interxylary phloem (included phloem of concentric type) in the wood of unspecified species of *Acantholimon*. According to the same author, the appearance of successive rings of growth with included phloem was accompanied by medullary bundles which are simply collateral and inversely oriented. In the present investigation, the included phloem was found to be of quite common occurrence and hence of no taxonomic value, but medullary bundles were found only in a few species, like *A. hypochaerum* (fig. 1A). In this respect *A. hypochaerum* differs from its close ally *A. calvartii*. In *A. hypochaerum* some of the medullary bundles are completely encircled by lignified fibres.

Anatomy of the inflorescence axis

In this account, the part of the inflorescence axis below the spike is called a scape, while the inflorescence axis constituting the spike is referred to as rachis and the individual internodes of the rachis are termed joints.

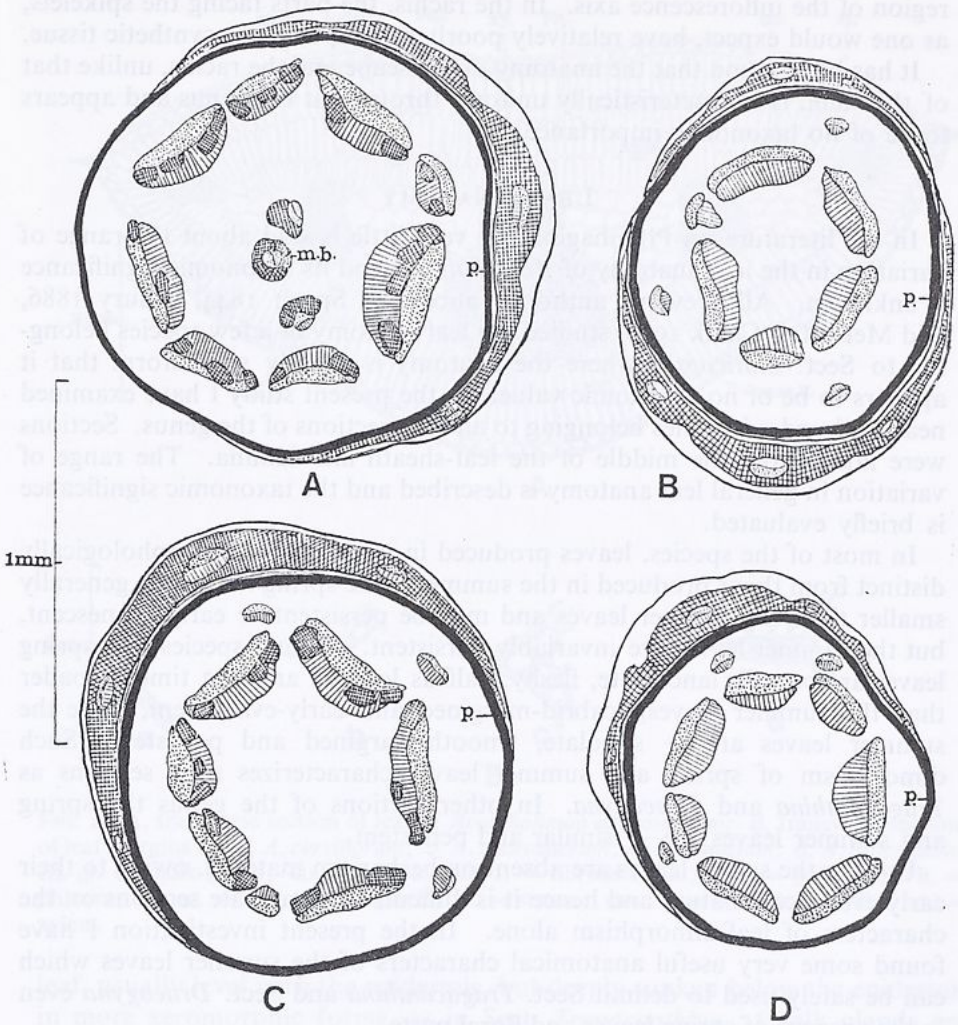

FIG. 1. Diagrams of transverse sections of stems of: A, *Acantholimon hypochaerum;* B, *A. acerosum;* C, *A. curviflorum;* D, *A. bracteatum.* m.b. = medullary bundle; p. = periderm; fibrous tissue cross-hatched; phloem dotted; xylem hatched.

Sections of the inflorescence axis were taken from the first internode, in the middle of the scape and in the middle of the joint of the rachis. In the scape as well as in the rachis, the pith is surrounded by varying numbers of large collateral vascular bundles forming an inner ring. Outside this ring there is a broad zone of lignified fibres in the pericycle region. On the periphery of this lignified zone, and sometimes even within it, are smaller bundles (cortical bundles) of normally oriented xylem and phloem constituting the outer ring. The cortex is composed of parenchymatous cells with intercellular spaces and is evidently assimilatory in function. In the first internode of the scape, the cortex has 4-6 layers of rounded parenchymatous cells with a few patches of lignified tissue, but above the first internode and in the region of the rachis, the outermost layer of the cortical cells (2–3) becomes clearly differentiated into a palisade which constitutes the photosynthetic

region of the inflorescence axis. In the rachis, the parts facing the spikelets, as one would expect, have relatively poorly developed photosynthetic tissue.

It has been found that the anatomy of the scape and the rachis, unlike that of the stem, is characteristically uniform throughout the genus and appears to be of no taxonomic importance.

Leaf anatomy

In the literature on Plumbaginaceae very little is said about the range of variation in the leaf anatomy of *Acantholimon* and its taxonomic significance is unknown. All previous authors (Jaubert & Spach 1844, Maury 1886, and Metcalf & Chalk 1950) studied the leaf anatomy of a few species belonging to Sect. *Staticopsis*, where the anatomy is usually so uniform that it appears to be of no taxonomic value. In the present study I have examined nearly a hundred species belonging to all nine sections of the genus. Sections were taken from the middle of the leaf-sheath and lamina. The range of variation in general leaf anatomy is described and the taxonomic significance is briefly evaluated.

In most of the species, leaves produced in the spring are morphologically distinct from those produced in the summer. The spring leaves are generally smaller than the summer leaves and may be persistent or early-evanescent, but the summer leaves are invariably persistent. In some species, the spring leaves are broadly lanceolate, fleshy, half as long as and 2–3 times broader than the summer leaves, scabrid-margined and early-evanescent, while the summer leaves are ± subulate, smooth-margined and persistent. Such dimorphism of spring and summer leaves characterizes such sections as *Tragacanthina* and *Dracogyna*. In other sections of the genus the spring and summer leaves are ± similar and persistent.

Usually the spring leaves are absent on herbarium material, owing to their early-evanescent nature and hence it is difficult to demarcate sections on the characters of leaf-dimorphism alone. In the present investigation I have found some very useful anatomical characters of the summer leaves which can be safely used to delimit Sect. *Tragacanthina* and Sect. *Dracogyna* even in the absence of spring leaves and floral parts.

Leaf sheath. The study of a large amount of herbarium material revealed that the number and position of vascular bundles and the shape of the fibrous sheath do exhibit variation, and when correlated with other morphological and anatomical characters they may usefully serve as auxiliary criteria for delimiting some sections. In *A. saxifragifolium* and *A. inerme* of Sect. *Gontscharovia*, there are five free bundles in the sheath, each accompanied by a cap of fibres on both sides. In the remaining sections of the genus vascular bundles in the leaf sheath are never free but are always encircled by a group of lignified fibres called the fibrous sheath. In Sect. *Tragacanthina* the fibrous sheath is well-developed and ± smooth in its outline; in the remaining sections, the fibrous sheath is deeply undulate.

General histology of the lamina (figs. 2, 3)

Epidermis. The epidermal cells are thick-walled with a broad lumen. The cuticle is thick and usually striated. Stomata are found on all sides of the

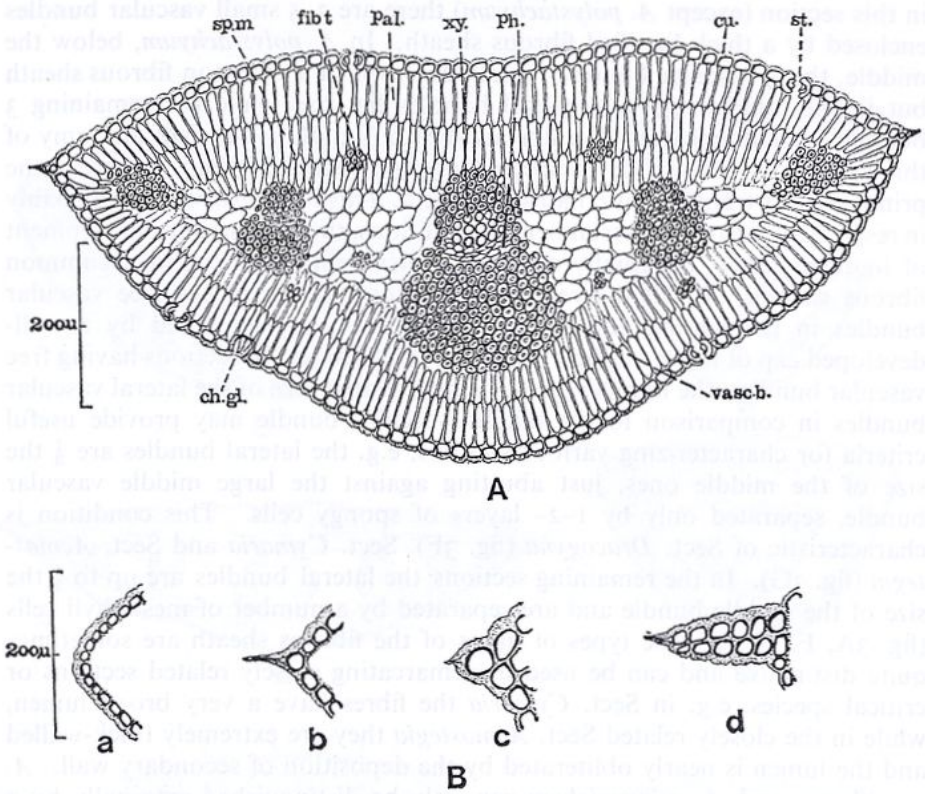

FIG. 2. A, transverse section of leaf of *Acantholimon androsaceum;* B, transverse sections of leaf margins of: a, *A. curviflorum*; b, *A. androsaceum*; c, *A. venustum*; d, *A. bracteatum.* ch. gl. = chalk gland; cu. = cuticle, fib. t. = fibrous tissue; Pal. = palisade; Ph. = phloem; sp. m. = spongy mesophyll; st. = stoma; vasc. b. = vascular bundle; xy. = xylem.

leaf, usually level with the epidermis, but deeply sunken below the epidermis in more xeromorphic forms. as in Sect. *Tragacanthina.* Chalk glands are found depressed below the epidermis.

Palisade tissue. It is usually concentric but, except in Sect. *Tragacanthina,* it tends to become isobilateral in the late summer leaves. It is generally strongly developed and varies from 2–4 cells in thickness (figs. 2A, 3A–I).

Spongy tissue. In comparison with the palisade tissue, the spongy tissue is rather poorly developed. In the majority of the species it is composed of rounded parenchymatous cells with few intercellular spaces. In Sect. *Tragacanthina* it is characteristically reduced to 1–2 layers of small cells around the bundle sheath.

Vascular bundles. There is much variation in the number and position of vascular bundles in the lamina, and usually there is a relative consistency with respect to these features. These characters of leaf anatomy can be very usefully employed to distinguish some sections of the genus. As has been mentioned earlier, Sect. *Tragacanthina* is delimited on the basis of dimorphic leaves, where the spring leaves are early-evanescent. In this section the anatomy of the summer leaf is found to be quite distinctive, and on this basis alone, the section may be reasonably demarcated. In all the species examined

in this section (except *A. polystachyum*) there are 3–5 small vascular bundles enclosed by a thick lignified fibrous sheath. In *A. polystachyum*, below the middle, the 5 vascular bundles remain enclosed in a common fibrous sheath but in the middle region 2 lateral bundles lie free, while the remaining 3 bundles are still enclosed in a fibrous sheath (fig. 3C). The leaf anatomy of this species appears to be of special interest, because it may represent the primitive and transitional condition in Sect. *Tragacanthina* where, probably in response to extreme environmental conditions, there is a strong development of lignified tissue, eventually all the bundles being enclosed in a common fibrous sheath. In all the remaining sections there are 1–5 free vascular bundles in the lamina and these are invariably accompanied by a well-developed cap of fibres on their upper and lower sides. In sections having free vascular bundles, the number, position and relative size of the lateral vascular bundles in comparison to the middle vascular bundle may provide useful criteria for characterizing various sections, e.g. the lateral bundles are $\frac{1}{3}$ the size of the middle ones, just abutting against the large middle vascular bundle, separated only by 1–2– layers of spongy cells. This condition is characteristic of Sect. *Dracogyna* (fig. 3F), Sect. *Cymaria* and Sect. *Acmostegia* (fig. 3G). In the remaining sections the lateral bundles are up to $\frac{1}{2}$ the size of the middle bundle and are separated by a number of mesophyll cells (fig. 3A, E, H, I). The types of fibres of the fibrous sheath are sometimes quite distinctive and can be used in demarcating closely related sections or critical species, e.g. in Sect. *Cymaria* the fibres have a very broad lumen, while in the closely related Sect. *Acmostegia* they are extremely thick-walled and the lumen is nearly obliterated by the deposition of secondary wall. *A. curviflorum* and *A. quinquelobum* can only be distinguished externally by a very tricky character of the position of the calyx nerves in relation to the lobes, but the lumen of the fibres appears to have specific significance. In *A. curviflorum* the fibres have a very narrow lumen while in *A. quinquelobum* they have a comparatively broad one.

Fibrous bundles. The distinctive feature of the *Acantholimon* leaf anatomy is the occurrence of longitudinal groups of fibres at the lateral side (towards the margin), as well as in the mesophyll. The two lateral groups are usually accompanied by vascular elements in which xylem is facing inwards, probably representing atrophied vascular bundles. The fibrous bundles in the mesophyll are entirely made up of fibres in the majority of the species, but I have observed two species (*A. polystachyum* (fig. 3C) and *A. saxifragifolium* (fig. 3A)) where a very few of the fibrous bundles on the upper side are associated with vascular elements showing inverse orientation. The number of palisade layers, and the position and number of these fibrous strands are sometimes characteristic features of species. In *A. confertiflorum* there are 3–4-layers of palisade on the upper side and a number of fibrous bundles in the lower part of the upper palisade (fig. 3I), while in related species the palisade is usually 2-layered on both sides and there are fewer fibrous bundles present in the middle of the palisade.

Leaf margin. In species with smooth-margined leaves, the margin is usually indistinct in transverse section (fig. 2Ba), e.g. in Sect. *Tragacanthina*. In other sections, leaves are linear-triquetrous to linear-lanceolate and accordingly the margin differs. In linear-triquetrous leaves, as in *A. androsaceum*, the margin is 1-celled in thickness (fig. 2Bb), while in linear-lanceolate

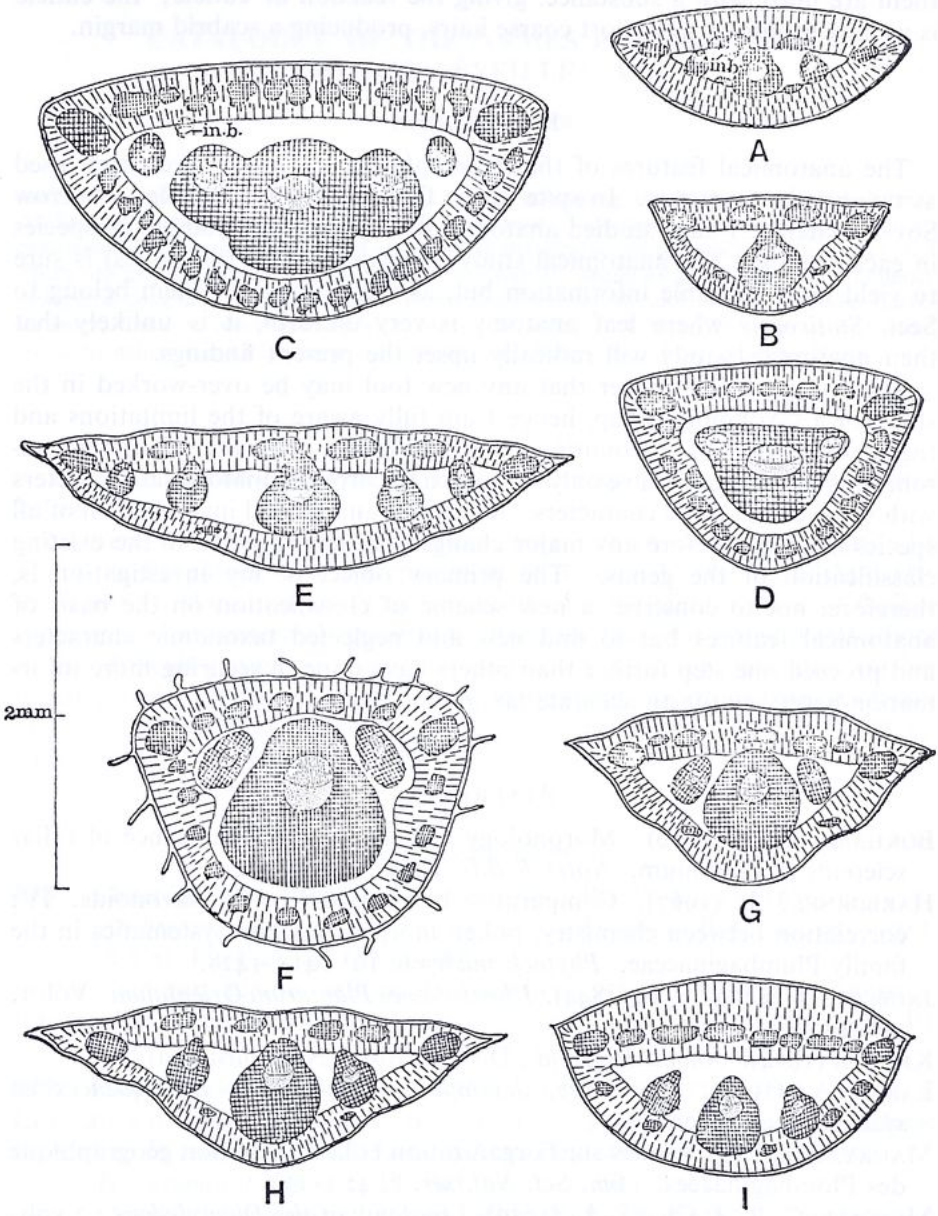

FIG. 3. Diagrams of transverse sections of leaves of: A, *Acantholimon saxifragifolium;* B, *A. diapensioides*; C, *A. polystachyum*; D, *A. curviflorum;* E, *A. bromifolium*; F, *A. tomentellum*; G, *A. cymosum*; H, *A. bracteatum*; I, *A. confertiflorum.* in. b. = inverted bundles.

leaves as in *A. venustum* (fig. 2Bc) and *A. bracteatum* (fig. 2Bd), the margin is ± 2-celled in thickness. These cells have thick walls and the angles between them are filled with a substance, giving the reaction of cuticle; the cuticle is usually produced into short coarse hairs, producing a scabrid margin.

Discussion

The anatomical features of the *Acantholimon* leaf have never been used as taxonomic characters. In spite of the fact that I was not able to borrow Soviet material, I have studied anatomically a reasonable number of species in each section. The anatomical study of Soviet species (endemics) is sure to yield more valuable information but, as the majority of them belong to Sect. *Staticopsis* where leaf anatomy is very uniform, it is unlikely that their anatomical study will radically upset the present findings.

There is always a danger that any new tool may be over-worked in the study of a taxonomic group, hence I am fully aware of the limitations and the inadvisability of discounting other characters in favour of useful anatomical ones. In fact, I have only attempted to correlate anatomical characters with some exomorphic characters. A detailed anatomical investigation of all species is needed before any major changes can be suggested in the existing classification of the genus. The primary object of my investigation is, therefore, not to construct a new scheme of classification on the basis of anatomical features but to find new and neglected taxonomic characters and proceed one step further than others have done in securing more information necessary for an accurate taxonomic study.

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