



## Comparative bark anatomical studies of *Tecomella undulata* (Rohitaka) with its adulterant taxa

V. Nagaraju<sup>1,2</sup>, K. Manipal<sup>1</sup> and K. Madhava Chetty<sup>1\*</sup>

Department of Botany, P.C.R Government Junior College, Chittoor 517001, Andhra Pradesh, India.

Department of Botany, Sri Venkateswara University, Tirupati 517502, Andhra Pradesh, India.

**Abstract:** This study aimed to compare and characterize stem bark anatomy of *Tecomella undulata* (Rohitaka), a potent medicinal plant with its adulterant taxa. Our field survey and the microscopic study revealed that Rohitaka is being adulterated and substituted with its allied taxa in the crude drug market. Photographs and Comparative note of original taxon (Rohitaka - *Tecomella undulata*) with habitat, Flowers, Bark sections were presented here for ease of identification.

**Keywords:** Rohitaka; *Tecomella undulata*; Bark anatomy; adulteration

### Introduction

Rohitaka, a potent ayurvedic drug, possesses extraordinary multidimensional therapeutic potency described in ayurvedic literature (Thakur *et al.*, 2020; Keshari *et al.*, (2018) and Sharma 2002). Rohitaka is botanically known as *Tecomella undulata* (Sm.) Seem. (Family: Bignoniaceae) is a deciduous medium sized ornamental tree generally habitats in tropical regions in India. *T. undulata* is considered a genuine source of Rohitaka mentioned in Ayurveda classics (Various samithas) for liver spleen disorders and other ailments (Keshari *et al.*, 2018; Kirtikar & Basu, 1984; Jain *et al.*, 2012).

Due to its abundant presence in arid zone and rare occurrence in South India, this essential species undergoing substitution, incorrect taxonomical identification, accidental contamination or intentional adulteration of original plant species with other allied morphologically similar species which are easily available became a common practice in trading of raw drugs. (Quilho *et al.*, 2020; Arun Suma *et al.*, 2014).

This scenario creates a high frequency of adulteration which cannot be detected without performing microscopic nor chemical investigation as guided by Quality control suggested by WHO and Ayurvedic literature (Shaheen *et*

*al.*, 2019; Prakash *et al.*, 2013; Santhosh Kumar *et al.*, 2018; Shanmughanandhan *et al.*, 2016; Srirama *et al.*, 2017; Ichim 2019; Ichim *et al.*, (2020) Kannangara *et al.*, 2020). Therefore, authentication at various stages from the collection and authentic identification of the plant material to the final product marketing of the original drug, is a need of the hour (Khan *et al.*, 2011; Schieber 2020). In this present situation and high rate of adulteration and substitution in the drug market at different local markets, there had been a negative impact on the ayurvedic drug in society, so we intended to focus on "Rohitaka" an important ayurvedic drug used adulterated in high incidence. Our previous research (Ramesh *et al.*, 2013 a & b, 2014; Sivaji *et al.*, 2012) was on controversial drugs used in the local markets.

This article focuses on the morpho-anatomical differentiation of bark for the ease of identifying the original plant taxa against its adulterant taxa sold under Rohitaka.

### Materials and Methods

The specimens of fresh barks of *Tecomella undulata* and allied adulterant taxa (Table 1) were collected from different areas around Tirupati in Andhra Pradesh (Figure 1. A-F). The botanical identification of the taxa was carried

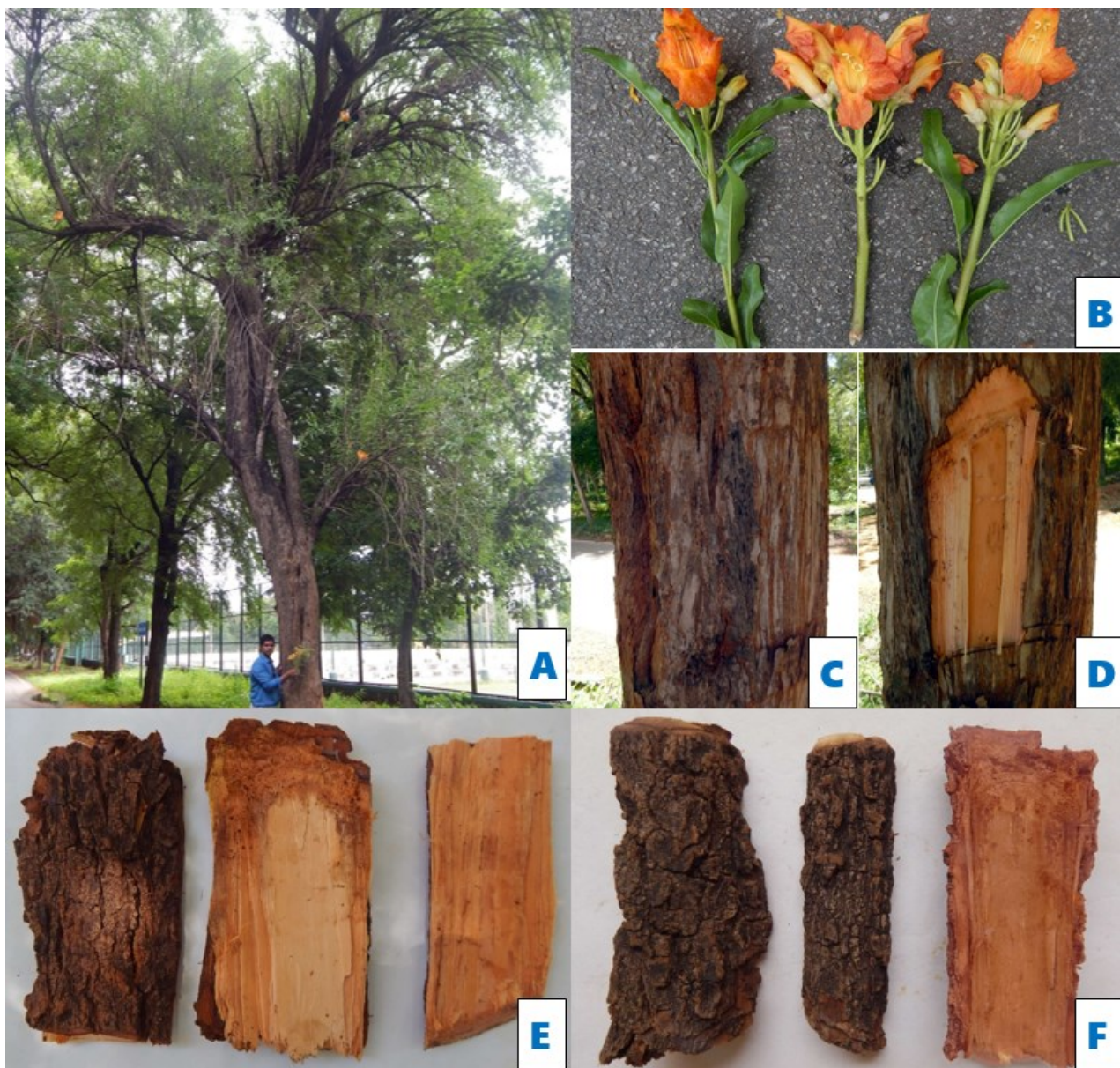
\*Corresponding Author:

Dr. K. Madhava Chetty,

E-mail: [madhavachetty@gmail.com](mailto:madhavachetty@gmail.com)

out by using regional and local floras (Pullaiah, 2018; Gamble 1918; and Chetty *et al.*, 2019). Pertinent literature review on anatomy and controversial drugs was done. The bark specimens were collected from the field and local market and deposited in the Herbarium of Department of Botany, Sri Venkateswara University, Tirupati, Andhra Pradesh. The Plant specimens (SVUTY/ BIG/ 4899) were authenticated by comparing voucher herbarium specimens with relevant virtual herbaria (K, NYBG). Photographs of authentic taxon *T.undulata* habitat, flowering stage, Stem,

chopped Bark were taken. Barks from the local markets were also collected for studying comparative anatomical studies on the adulterant taxa. Healthy bark was chopped and fixed in the field immediately in FAA (Formalin - 5ml + Acetic acid - 5ml + 70% ethyl alcohol - 90ml). Bark anatomical studies and morphometric data collection were performed following the standard microtomy methods (Ruffinatto and Crivellaro, 2019; Schweingruber *et al.*, 2019). Methods for Comparative Wood Anatomy Studies were followed according to Carlquist (2001).



**Figure 1.** *Tecomella undulata*. A. Author with Tree habitat; B Flowering twig; C: Bark; D: Chopped Bark E: Bark in fresh condition: F: Dried Barks

## Results and Discussion

In this research, collections of various *Tecomella undulata* bark samples and the substituted barks sold under the name Rohitaka were obtained from the herbal vendors and Ayurvedic raw drug sellers and then compared with the original plants collected from the field. We have noticed adulteration and substitution of other taxa which were traded and sold as Rohitaka in the markets in different places in Chittoor district. Table 1 represents the identified adulterant taxa (*Aphanamixis polystachya*, *Ventilago maderaspatana*, *Diospyros ferrea* var. *buxifolia* and *Polygonum glabrum*) as Rohitaka. Hitherto, no survey has ever conducted to document the comparative anatomical account intensity of the adulteration problem on Rohitaka.

In our observation, *Aphanamixis polystachya* (A.1) and *Diospyros ferra* var. *buxifolia* (A.3) tree barks look alike when powdered. Abundantly grown *Ventilago maderaspatana* (A.2) barks were also being adulterated. Since *Polygonum glabrum* (A.4) being an herb with red coloured stem, is being adulterated in dry form. we here did not compared *P. glabrum* with the other taxa. But these substituted tree taxa showed some taxonomic similarities and differences at generic and species level (Table 2). we claim that interpretation of the anatomy of bark show one unique anatomical feature, which is the key to its identification of original plant material to the adulterants enlisted in Table 1. The internal micromorphological anatomical structures and magnifications are evaluated by the scale bars.

**Table 1.** Details of adulterated taxa of Rohitaka

S. No.	Adulterant taxa	Habitat	Family	Voucher specimen
1.	<i>Aphanamixis polystachya</i> (A.1)	Tree	Meliaceae	SVUTY/MEL-4082
2.	<i>Ventilago maderaspatana</i> (A.2)	Liana	Rhamnaceae	SVUTY/RHM-4766
3.	<i>Diospyros ferra</i> var. <i>buxifolia</i> (A.3)	Tree	Ebenacea	SVUTY/EBE-4143
4.	<i>Polygonum glabrum</i> (A.4)	Herb	Polygonaceae	SVUTY/POL-4419

**Table 2:** Comparative morphoanatomical characters of bark anatomy of different taxa used as an Ayurvedic drug Rohitaka (genuine taxa comparison with adulterant taxa)

Different parts of the bark	<i>Tecomella undulata</i> (Original taxon)	<i>Aphanamixis polystachya</i> (Adulterant 1)	<i>Ventilago maderaspatana</i> (Adulterant 2)	<i>Diospyros ferrea</i> var. <i>buxifolia</i> (Adulterant 3)
Texture	Outer surface is hard and dark brown, inner is reddish orange	Outer surface is dark brown, inner is bright red	Outer bark is dark grey, inner is dark brown	Outer bark is grey to reddish black, inner is light red
Periderm	The bark consists of outer zone of periderm and inner zone of secondary phloem	The bark consists of outer zone of periderm and inner zone of secondary phloem	The bark consists of outer zone of periderm and inner zone of secondary phloem	The bark consists of outer zone of periderm and inner zone of secondary phloem
	It is not divided into phellem and phelloderm cells	It is not divided into phellem and phelloderm cells	Divided into equally broad phellem and phelloderm cells	Divided into phellem and phelloderm cells
Phellem cells	Absent	Absent	Two types of cells: a layer of squarish cells which alter-nate with a layer of narrow tabular cells,	Homogenous tabular thin walled suberised cells

			both the cells are thin walled and suberised	
Phelloderm cells	Absent	Absent	Absent	Homocellular, squarish and thin walled
Rhytidome	Not evident	Not evident	Rhytidome is present	Not evident
Secondary phloem	Divided into outer collapsed phloem and inner non-collapsed phloem	Divided into outer collapsed phloem and inner non-collapsed phloem	Divided into outer collapsed phloem and inner non-collapsed phloem	Divided into outer collapsed phloem and inner non-collapsed phloem
Collapsed phloem	It is several layered, consists of phloem sclerenchyma, phloem parenchyma and calcium oxalate crystals	It is many layered, consists of phloem sclerenchyma, phloem parenchyma, calcium oxalate crystals, starch grains and tannins	It is numerous layered, consists of phloem sclerenchyma, phloem parenchyma, gelatinous fibers and calcium oxalate crystals	It is numerous layered, consists of phloem sclerenchyma, phloem parenchyma and calcium oxalate crystals
Sieve elements	Crushed into tangential darker streaks	Crushed into several tangential darker layers	Crushed into tangential darker streaks	Crushed into tangential darkly stained straight arc, concave lines
Phloem rays	Wide, dilated and undulate	Undilated	Narrow at the base and gradually dilated at top	Wide, highly dilated and wavy
Non-collapsed phloem	Consists of phloem sclerenchyma, sieve elements and their companion cells	It consists of phloem parenchyma, sieve elements and their companion cells	It lacks phloem sclerenchyma, crystals, tannins and other cell inclusions	It consists of sieve elements and their companion cells
Sieve elements	Wall are angular	Walls are radial	Walls are polygonal	Walls are reticulate
Companion cells	Present along the lateral corners of the sieve elements	Present along the lateral corners of the sieve elements	Present along the lateral corners of the sieve elements	Present along the lateral corners of the sieve elements
Secondary xylem	In the secondary xylem the phloem rays are multi rarely uni or biseriate, heterocellular, non-storied	Phloem rays are narrow, uni- or biseriate, heterocellular, non-storied	Phloem rays are narrow, uni-, rarely biseriate, homocellular, non-storied	Phloem rays are polyhedral, multiseriate, homocellular, non-storied
Phloem rays	Phloem rays with marginal upright cells and middle procumbent cells  short Phloem rays, spindle shaped	Phloem rays with marginal upright cells and middle procumbent cells  Phloem rays are narrow	Phloem rays with only oblong upright cells  Phloem rays are narrow	In the phloem rays there is no differentiation of upright and procumbent cells.  Phloem rays are polyhedral
Calcium oxalate crystals	Spindle shaped spiny druses abundant in phloem parenchyma cells	Rhomboidal, cuboidal and pyramidal prismatic types are spar-	Rectangular to cuboidal prismatic type of several tangential lines are	Cuboidal small prismatic types are present in the phloem

		sely present in phloem parenchyma cells	abundant in the parenchyma cells	parenchyma cells
--	--	---	----------------------------------	------------------

The diversity of bark morphology due to selective pressures encountered by tree species is huge, and can provide a very important identification feature, particularly at the family level in the tropics and at the species level (Morris and Jansen, 2016). Nahida *et al.*, (2015) carried out the quality standards and safety profile of stem bark of *Tecomella undulata* and comparative evaluation with its adulterant *Aphanamixis polystachya* as per WHO guidelines. Keshari *et al.*, (2018) studied the comparative pharmacognostic evaluation of *Tecomella undulata* and *Rhododendron arboreum* as two different sources of Rohitaka. Patil *et al.*, (2016) worked on the ayurvedic drug "Punarnava". Pundir *et al.*, (2015) worked on the pharmacogenetic standardization and chromatographic evaluation of *Tecomella undulata* stem bark. Patel *et al.*, (2011) observed the hepatoprotective effect of ethanolic extract of *T.undulata* bark significantly restored physiological integrity of hepatocytes in rats. Thakur *et al.*, (2020) reviewed the multidimensional pharmacological properties of Rohitaka and documented the adulterated species traded of Rohitaka. Despite of seasonal variation in the life cycle stages, confusing vernacular names, and unauthorized or fraudulent substitutions might have contributed to species adulteration in the raw drugs (Nithaniyal *et al.*, 2017). Only small differences between species were observed that cannot be used alone for taxonomic purposes (Mota *et al.*, 2017). The complex structure of the bark can be utilized for botanical identification to maintain the quality and purity of the drug (Brinda *et al.*, 2000). In this investigation, our approach is to obtain the visual knowledge on the main comparative features of barks by the measure of macroscopic and microscopic Anatomical bark characters. We are here with report that the availability of the crude drug Rohitaka genuine source (*Tecomella undulata*) is diminishing, and this has resulted in the sale of adulterants.

## Conclusion

In this present research, we characterized and compared in relation to anatomical features of different plant barks for the originality of the genuine taxa (*Tecomella undulata*) and the adulterant taxa (*Aphanamixis polystachya*, *Ventilago maderaspatana*, *Diospyros ferra* var. *buxifolia*, *Polygonum glabrum*) being sold under ayurvedic drug Rohitaka. We have proved the adulteration, and we suggest Researchers, ayurvedic physicians to undertake Pharmaco-vigilance and scientific validation is essential on any drug sold in the herbal drug markets.

## Acknowledgments

Authors acknowledge the Department of Botany, Sri Venkateswara University, Tirupati for providing research facilities.

## References


1. Arun Suma., E. M. Muralidharan., P. Sujanalal M. Balasundaran. "Identification of market adulterants in East Indian sandalwood using DNA barcoding". *Annals of Forest Science*, 71.4: (2014). 517-522. Online.
2. Brinda P., A. Saraswathy and P. Jayaraman. "Micromorphological identification of the medicinal bark of *Ventilago maderaspatana*". *Journal of Medicinal and Aromatic Plant Science*. 23. (2000): 619-622. Online.
3. Carlquist S. "Comparative Wood Anatomy". Springer-Verlag Berlin Heidelberg. 1 (2001): 1-7. Print.
4. Chetty K.M., Ramesh L., Rajalkar K. M., & Manipal K. "Flowering plants of Chittoor district", Andhra Pradesh, India. Students offset Printers, Tirupati. (2019). pp.358. Print.
5. Gamble J.S. "Flora of the presidency of Madras", Adlard & Sons, Ltd., London, (1918). 1-3. Print.
6. Ichim M.C. The DNA-Based Authentication of commercial herbal products reveals their globally widespread adulteration. *Frontiers in pharmacology*, 10. (2019): 1227. Online.
7. Ichim M.C., A. Häser., P. Nick. "Microscopic Authentication of Commercial Herbal Products in the Globalized Market: Potential and Limit-

- ations". *Frontiers in pharmacology*, 11 (2020): 876. Online.
8. Jain M., R. Kapadia., R.N Jadeja., M.C Thounajam, R.V Devkar., S.H Mishra. "Hepatoprotective potential of *Tecomella undulata* stem bark is partially due to the presence of betulinic acid". *Journal of Ethnopharmacology* 143. (2012): 194-200. Online.
9. Kannangara S., S Karunarathne., L. Ranawee-ra., K Ananda., D Ranathunga., H Jayarathne., C Weebadde and S Sooriyapathirana. "Assessment of the applicability of wood anatomy and DNA barcoding to detect the timber adulterations in Sri Lanka". *Scientific Reports*. 10.(2020): 4352. Online.
10. Keshari P., Pradeep, Prabhu SN. "Comparative pharmacognostic evaluation of *Tecomella undulata* and *Rhododendron arboreum* as two different sources of *Rohitaka*". *International Journal of Green Pharmacy*. 12.4 (2018): 242 -252. Online.
11. Khan S., K.J. Mirza., F. Al-Qurainy., M.Z. Abdin. "Authentication of the medicinal plant *Senna angustifolia* by RAPD profiling". *Saudi Journal of Biological Sciences*, 18.3 (2011): 287-292. Online.
12. Kirtikar K.R., and B.D. Basu. "Indian Medicinal Plants", (Dehradun, India: *Bishan Singh Mahendra Pal Singh*) (1984). 724-1841. Print.
13. Morris H and S. Jansen. Bark: its anatomy, function and diversity. *International Dendrology Society*. Year Book (2016): 51-61. Print.
14. Mota G.S., C.J. Sartori., I. Miranda., T. Quilhó., F.A. Mori., H. Pereira. "Bark anatomy, chemical composition and ethanol-water extract composition of *Anadenanthera peregrina* and *Anadenanthera colubrina*". *PLoS ONE*. 12.12 (2017): e0189263. Online.
15. Nahida P., Dharya S., Vidhu A. "Quality control and comparative study of ayurvedic plant *Tecomella undulata* (Sm.) Seem. With its adulterant *Aphanamixis polystachya* (Wall.)." *International Journal of Pharmacognosy and Phytochemical Research*. 7.5. (2015): 917-922. Online.
16. Nithaniyal S., S.L Vassou., S. Poovitha., B. Raju and M. Parani. "Identification of species adulteration in traded medicinal plant raw drugs using DNA barcoding". *Genome*. 60.2 (2017): 139-146. Online.
17. Patel K.N., G. Gupta., M. Goyal and B.P. Nagori. "Assessment of hepatoprotective effect of *Tecomella undulata* (Sm.) Seem., Bignoniaceae, on paracetamol-induced hepatotoxicity in rats *Revista Brasileira de Farmacognosia*, 21.1. (2011): 133-138. Online.
18. Patil V.S., K.S Rajput., N.P. Malpathak. "Comparative study on morphoanatomy of leaf, stem and root of *Boerhavia diffusa* L. (Nyctaginaceae) and its adulterant plants". *Brazilian Journal of Pharmaceutical Sciences*. 52.3. (2016): 433- 442. Online.
19. Prakash Om., Jyoti., A. Kumar., P. Kumar and N.K. Manna "Adulteration and substitution in Indian medicinal plants: an overview". *Journal of Medicinal Plants Studies*. 1.4 (2013): 127-132. Online.
20. Pullaiah T., Chennaiah E and Sandhya rani S. "Flora of Andhra Pradesh"-2<sup>nd</sup> Rev.Ed. Scientific Publishers. 5. (2018):1932p. Print.
21. Pundir S., S.K. Tomar., S.H. Mishra., S Albert., M. Jain. "Pharmacognostical standardization and chromatographic evaluation of *Tecomella undulata* stem bark". *International Journal of Biology, Pharmacy and Allied Sciences*. 4: (2015): 4499-4511. Online.
22. Quilho T., F. Bessatwo., A.I Ribeiro-Barros., N Ribeiro. "The anatomy as a tool for the identification of the bark of *Pterocarpus angolensis* and *Terminalia sericea*". *Advances in Forestry Science*. 7.1. (2020): 925-930. Online.
23. Ramesh L, D.Muniswamy, A.Veerabhadrappe, K. Sreedevi and K. Madhavachetty. "Comparative pharmacognostical studies of *Asclepias curassavica* used in ayurvedic drug "kakanasa" with its adulterant *Trichosanthes cucumerina*". *Archives of Pharmacy and Biological Sciences*, 1. 3. (2013): 85-93. Online.
24. Ramesh L., M. Mahendra Nath and K. Madhava Chetty. "Comparative Pharmacognostical Studies of original Taxa with substituents used in Ayurvedic drug Kakanasa". *Indian Journal of Fundamental and Applied Life Sciences*, 3.1 (2013): 278- 288. Online.
25. Ramesh L., M. Sudhakar., K. Madhava Chetty., M. Mahendranath. "Comparative Pharmacognosy of *Asclepias curassavica* used in Ayurvedic Drug "Kakanasa" with its Adulterant *Lepta-denia reticulata*". *International Journal of Pharma Science Review Research*. 26(1). (2014): 43 -49. Online.

26. Ruffinatto F., A. Crivellaro. "Atlas of Macroscopic Wood Identification (With a Special Focus on Timbers Used in Europe and CITES - listed Species)". Springer Nature Switzerland AG. (2019). pp 439. Print.
27. Santhosh Kumar J.U., V. Krishna, G.S. Seethapathy., R. Ganesan., G. Ravikanth and R.U. Shaanker. "Assessment of adulteration in raw herbal trade of important medicinal plants of India using DNA barcoding". 3 *Biotech.* 8.3. (2018): 135. Online.
28. Schieber A. "Botanicals – challenges abound, solutions in sight?". *Current Opinion in Food Science*, 32, (2020): 144-148. Online.
29. Schweingruber F.H., S Peter., B Annett. "Bark Anatomy of Trees and Shrubs in the Temperate Northern Hemisphere". Springer Nature Switzerland AG. VI. (2019). pp 394. Print.
30. Shaheen S, Ramzan S, Khan F and Ahmad M. "Adulteration in Herbal Drugs: A Burning Issue". Springer International Publishing, Switzerland. (2019). 179pp. Print.
31. Shanmughanandhan D., Ragupathy S., Newmaster S. G., Mohanasundaram S., Sathishkumar R. "Estimating Herbal Product Authentication and Adulteration in India Using a vouchered, DNA-Based Biological Reference Material Library". *Drug safety*, 39.12 (2016):1211–1227. Online.
32. Sharma P.C., M.B Yelne., T.J Dennis. "Rohit-aka. In: Database on Medicinal Plants used in Ayurveda". New Delhi: Central Council for Research in Ayurveda and Siddha. 6:(2002): 321-324. Online.
33. Sivaji K., M. Mahendra Nath., L. Ramesh., K. Madhava Chetty. "Comparative Pharmacognostical Studies of *Terminalia arjuna* used in Ayurvedic drug Arjuna, with its adulterant *Kavalama urens*". *Indian Journal of Plant Sciences*. 1.2. (2012). :229-238.
34. Srirama R., J.U. Santhosh Kumar., G.S Seethapathy., S.G. Newmaster., S. Ragupathy., K. N. Ganeshaiah., R. Uma Shaanker and G. Ravikanth. "Species Adulteration in the Herbal Trade: Causes, Consequences and Mitigation". *Drug safety*, 40. 8. (2017): 651–661. Online.
35. Thakur SK, Natthani S, Kotecha M. "Primitive therapeutic bids of medicinal plant Rohitaka *Tecomella undulata* (SM.) Seem.: a comprehensive appraisal based on classical ayurveda texts".9.8. (2020):17-20. Online.

**Cite this article as:**

V. Nagaraju, K. Manipal and K. Madhava Chetty. Comparative bark anatomical studies of *Tecomella undulata* (Rohitaka) with its adulterant taxa. *Annals of Plant Sciences*. 9.11 (2020) pp. 4067-4073.

 <http://dx.doi.org/10.21746/aps.2020.9.11.2>

**Subject Editor:** Dr. Sateesh Suthari, Telangana, India.

**Source of support:** Nil; **Conflict of interest:** Nil.