

Wood anatomical study of *Osyris quadripartita* Salz.exDecne (African sandalwood) from the Northern Western Ghats, India

Abstract

The family Santalaceae is medicinally important. *Osyris quadripartita* Salz.ex Decne has been used for the medicinal use such as anthelmintic, leishmaniasis like leprosy diseases, abnormalities, kidney disease, forage, hygienic/perfuming, eye pain, cold, anaphylactic shock, epilepsy, circumcision wound, toothache, tonsillitis, abdominal pain, urine problem, breast cancer, mental problems and anti-coronavirus disease, etc. It has also been reported for its activities such as anti-ulcer activity, antiviral activity. *Santalum album* and *O. quadripartita* belong to the same family santalaceae and have many similarities in their morphological characteristics. Both plant species belong to the same family, that's why anatomical character help to resolve this problem and easy to distinguish most wood anatomical characteristics. Therefore, distinguishing one from the other anatomically is very easy.

A study has been carried out to authenticate the identity of this specie based on anatomical characters. The anatomical sections of stem (Transverse Section (TS), Transvers Longitudinal Section (TLS) and Radial Longitudinal Section (RLS) of the species were observed under the bright-field compound binocular microscope, and characters were documented. The aim of the study is to elucidate the means and criteria to identify the anatomical structure of *O. quadripartita*. Though the diagrammatic outline of the stem sections is similar, detailed anatomical characters are significantly different. The characters such as the arrangement of cortical tissues, pericyclic fibres, phloem fibers, and phloem rays are useful in differentiating the stem and petiole of the species. Wood anatomical characteristics of *O. quadripartita* were seriation of rays, type of crystalliferous cells, and abundance of extractives. Ray cells frequently contain rhomboidal crystals. In cortex, and pith region of the stem observed solitary crystals. Based on the preliminary study, the species can be differentiated from other species of the same family based only on anatomical characters.

Keywords: *Osyris quadripartita*, Authentication, Anatomy, Vegetative parts

Introduction:

The Santalaceae family covered three types of habitat like herbs, shrubs, and rare trees. The family Santalaceae is medicinally important. The family widely distributed about 44 genera and 930 species (Simpson, 2010). In the *Osyris* genus, six or seven species were distributed in Asia, Africa, Europe, and China (Nianhe & Gilbert, 2003). *Osyris* genus is the evergreen roothemiparasites, ethnomedicinal shrub plants. Many tribal peoples used this plant leaves for making a herbal tea (Herrera, 1984; Subba & Basnet, 2014; Kefalew et al., 2015; Silva et al., 2016). The taxonomical characteristic of *Osyrisqua dripartita* Salzmänn ex Decaisne, *Ann. Sci. Nat., Bot., sér. 2, 6: 65. 1836*. *O. quadripartita* species are shrub or small tree up to 5- 7 m tall. Leaves are subsessile, rigid, elliptic to elliptic-oblong, 1–7 x 0.5–4 cm, acute or mucronate at the apex, with slightly thickened margins. Flowers yellowish-green; perianth-lobes triangular-ovate, 1–1.5 mm long. Male flowers in 5–13-flowered cymes. Female flowers in 1–3-flowered cymes. Fruit is drupes 5–10 mm in diam., Fruit intially green in colour then becoming ripen bright red colour (Zhen, 2022).

The only two main species used to produce commercial sandalwood oil are *Santalum album* (Indian sandalwood) and *S. spicatum* (Australian sandalwood). In domestic as well as international market true sandalwood oil is very costly. Wrongly or intentionally some peoples sell adulterated low quality cheaper substitutes, the ‘African sandalwood’ (*O. quadripartita*). The heartwood of this plant not as strongly fragrant as sandalwood so, this plant is used as adulterating sandalwood in the market (Khare, 2007; Shyaula, 2012). *O. lanceolata* is a timber like true sandalwood and it contains oil components. This oil is used to make a scent (Mabberley, 2018). *O. quadripartita* coming under least concern IUCN category (Wilson, 2018). In Ethiopia, this plant leaves and root use as medicinal purpose to cure antihelmintic and leishmaniasis (Geyid et al., 2005). Its used for curing skin infection (Mesfin, 2007). Its use as medicinal purpose to cure eye pain, anaphylactic shock and epilepsy (Agize et al., 2013). Disease treated as abdominal pain, kidney disease and urine problem (Getaneh & Girma, 2014; Worku, 2018). Its use as medicinal purpose to cure eye pain (Birhan et al., 2017). Disease treated as tonsillitis (Abdela & Sultan, 2018). Leaf and stem used as circumcision, wound, toothache (Yohannis et al., 2018). Decocted dried leaves are used for the treatment of breast cancer (Graham et al., 2000). Anthrax and pasturolosis disease treated with the help of *O. quadripartita* plant root (Advisor, 2017). *O. quadripartita* bark gave to orally for the treatment of jaundice (Maryo et al., 2015). *O. quadripartita* fruits are consumed as wild edible fruit in the Western Ghats, Tamil

Nadu regions, and Delanta region of northern Ethiopia (Arinathan et al., 2007; Alemayehu et al., 2015; Meragiaw et al., 2015).

Wood anatomical characteristics of *O. quadripartita* were observed different characters such as seriation of rays, type of crystalliferous cells, and abundance of extractives. Ray cells frequently contain rhomboidal crystals. In cortex, and pith region of the stem observed solitary crystals. Based on the preliminary study, the species can be differentiated from other species of the same family based only on anatomical characters.

Table 1: Classification of *Osyris quadripartita*

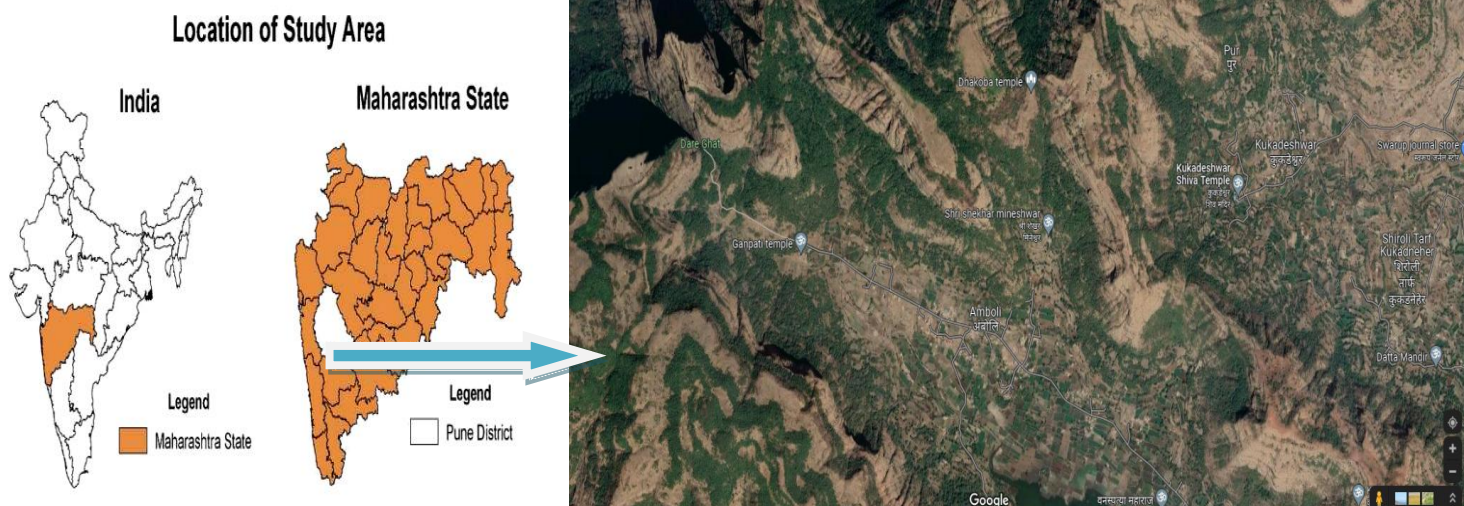
Kingdom	Plantae
Class	Magnoliopsida
Order	Santalales
Family	Santalaceae
Genus	<i>Osyris</i>
Species	<i>O. quadripartita</i> Salzm. ex Decaisne
Common names	Pulluruvi, Popali, Dalmia, Paral and wild tea plant

(Singh et al., 2001).

Materials and methods:

Study area and collection: Stem of *O. quadripartita* were collected from Pune, district during 2020-2021 of Maharashtra, India. The plant was identified using Flora of Maharashtra & Flora of China and authenticated by myself and deposited in BSI 829420.

Anatomical study of stem: Make a thin sliced from fresh mature stem samples for anatomical study (8 mm to 10 mm) from plant species. Prepare hand sections stained were stain using 0.1% Safranin and (98 % grade Alka Trading Corporation, Nagpur) mount in glycerin (98 % LOBA CHEM PVT LTD) and examined under bright-field compound microscope (Chamberlain, 1920; Khandelwal & Sethi, 2019). Measure a morphometric observations of length, diameter, breadth and number of layers (μm). (Salisbury's, 1927; Evert, 2006).



Map 1: STUDY AREA FROM JUNNAR TEHASIL (Table 3)

Results and Discussion:

During the study of *Osyris* species we observed many of anatomical variation TS of stem (Cork layer, Cortex cell layer, Pericyclic fiber cell number, Phloem cell layer, Phloem medullary layer, Xylem medullary layer, Xylem vessels) of the species were observed under the bright-field light compound microscope and their characters were documented (Metcalf & Chalk, 1965; Khandelwal & Sethi, 2019). This species produces an annual ring and observed a secondary growth. These plants show annual rings because of well developed secondary growth. In stem woody anatomy observed a broad range arrangement of vessels and fibers produces a porous wood (Metcalf & Chalk, 1965). Xylem is a complex tissue system composed of different cell types. The vessel elements adjoining to each other, at their end possess perforations, whereas tracheid elements lack these perforations. Tracheid elements are elongated, lignified, thick walled and pointed at both ends. The perforations several openings which are scalariform perforation plates (Fig.2). Xylem vessels showing simple pits in their lateral walls. To observe xylem structure, mature stems are sectioned transversely (transverse section: TS) and longitudinal planes: along the rays (radial longitudinal section: RLS and tangential longitudinal section: TLS). Rays are termed one cell wide tangentially called uniseriate and more than one cell wide called multiseriate observed in both species, viewed in TS and TLS. The vascular cambium produces secondary xylem and phloem of stem. Prismatic calcium oxalate crystals observed in *O. quadripartita* in the pith region of stem (Evert, 2006; Rudall, 2007; Khandelwal & Sethi, 2019). Starch grains are one of the non-nitrogenous stored forms observed in studied plant species. Tapioca starch grain type observed in both plant species (Stevens, 1907; Wallis, 1939). Wood anatomical characteristics of *O. quadripartita* were seriation of rays, type of crystalliferous cells and abundance of extractives. Ray cells frequently contain rhomboidal crystals. In cortex and pith region of stem observed solitary crystals (fig. 2 b, e). (Rao & Das, 1979; Zobel, 1985; Evert, 2006). The sclerenchyma found in the pericyclic region of stem. The stone cells are observed in both species. Stone cells important function is thickening and lignification of the walls of thin-walled parenchyma cells. Stone cells providing a strengthening and toughness of parenchyma cells. Sclerenchyma cells are classified into two categories fibers and sclereids (Rao, 1957; Evert, 2006).

Anatomical study of stem: Transverse Section (T.S.) of stem of *O. quadripartita* (Figs. 2) (Table 4,5 and 6)

Corck: Superficial and strongly thickened cork layer observed in the species. It was 14-16 cork layer.

Cortex: The cortex cells arranged in compact form. The total cortex layer is 10-18 (fig.2a).

Pericyclic fiber: Pericycle is a part of ground tissue. Ground tissue primary function is mechanical support. Pericyclic fiber are sclerenchyma in nature. Pericyclic fiber were observed in this species. It was observed in each clump number of cells are 02-06

Vascular tissues:

Phloem: The phloem stratified into soft and hard portions. In which hard portions is nothing but primary phloem fibers, or proto-phloem fibers and secondary phloem fibers. Phloem observed in this species 10-15 layered. Phloem medullary rays observed in this species there are 2-4 layered. Various cluster of crystalline structures deposition were present in pith cells.

Xylem: The arc type vascular bundles observed. Diffuse porous wood observed because of tracheary elements presents (tracheids and vessels). Medullary rays broader and expanded at the distal end. Vessels are arranged in vertical rows. Vessels varying in size 144.31 μm . Medullary rays mostly are 1-2. Well developed xylem fibers were arranged in vertical rows and in group. Fiber both end are pointed. Diffuse, aliform and confluent of parenchymatous cell observed in both species.

Pith: Pith are un lignified, large pitted parenchymatous cell with thin walled. Pith region of stem observed solitary crystals.

CONCLUSION:

The study undertaken to preliminary stem anatomical study of *O. quadripartita* showed the significant wood anatomy reliably on the basis of anatomical structure.

Microscopically preliminary observation reported ray cells contain rhomboidal crystals are the unique diagnostic characters reported in *O. quadripartita*. In Cortex and pith region of stem observed solitary crystals. On the basis of preliminary study the species can be differentiated from other species of same family based only on anatomical characters.

Table 2: Morphology of *Osyris quadripartita*

Characteristics	
Hight	2-5 m tall
Bark	Brown in colour
Leaves	Simple
Flower	Yellow in colour
Inflorescence	Cymes axillary
Fruit	Drupe orange to redwhen ripe
Occurrence	Bhutan, Cambodia, India, Laos, Myanmar, Nepal, Sri Lanka, Thailand, Vietnam; Africa, S Europe
Altitude from sea level	600-2700 m

(Singh et al., 2000;Zhen, 2022).

Table 3.*Osyris quadripartita* studied from northern Western Ghats of Maharashtra.

Taxon	Place of Collection	Geographic coordinates	Elevation (m)
<i>Osyris quadripartita</i>	19°14'22.2"N 73°41'35.4"E	Amboli, Junnar, Maharashtra 410502	1088
	19°13'14.3"N 73°43'37.0"E	Keldare, Junnar, Maharashtra 410502	325
	19°13'56.2"N 73°42'42.7"E	Uchhil, Junnar, Maharashtra 410502	277
	19°30'42.9"N 73°41'42.8"E	Ratangad, Maharashtra 421601	1126

Table 4.Stem anatomical characteristics of *Osyris quadripartita*.

Characters	<i>O. quadripartita</i>
Cork layer	14- 16
Cortex layer	10 - 18
Pericyclic fiber cell number	02-06
Phloem cell layer	10-15
Phloem medullary layer	2-4
Xylem medullary layer	1 - 2
Xylem vessels	simple and perforation
Pith cell	Parenchyma cell- rosette solitary crystals present

Table 5.T.S stem anatomical characteristics of *Osyris quadripartita*

Characters	<i>O. quadripartita</i>
Cork layer length 10 X	53.033±9.95
Pericyclic fiber cell diameter (cell number in each pach 18-25) 10 X	35.32±14.81
Vessele diameter 40 X	114.07±22.19
Medullary rays length 40 X	51.37±16.32
Pith cell diameter 40 X	56.66±16.98

*Each value expressed as mean± S.D of 10 replicate

Table 6.T.L.Sstem anatomical characteristics of *Osyris quadripartita*

Characters	<i>O. quadripartita</i>
Vessele Length 10 X	144.31 ±31.36
Bredth10 X	25.98±3.42
Medullary rays length(Medullary rays cell layer number 2-4) 10 X	406.29±99.56
Bredth10 X	13.40±1.31

*Each value expressed as mean± S.D of 10 replicates



Figure 1.*Osyris quadripartita* from Northern Western Ghats of Maharashtra

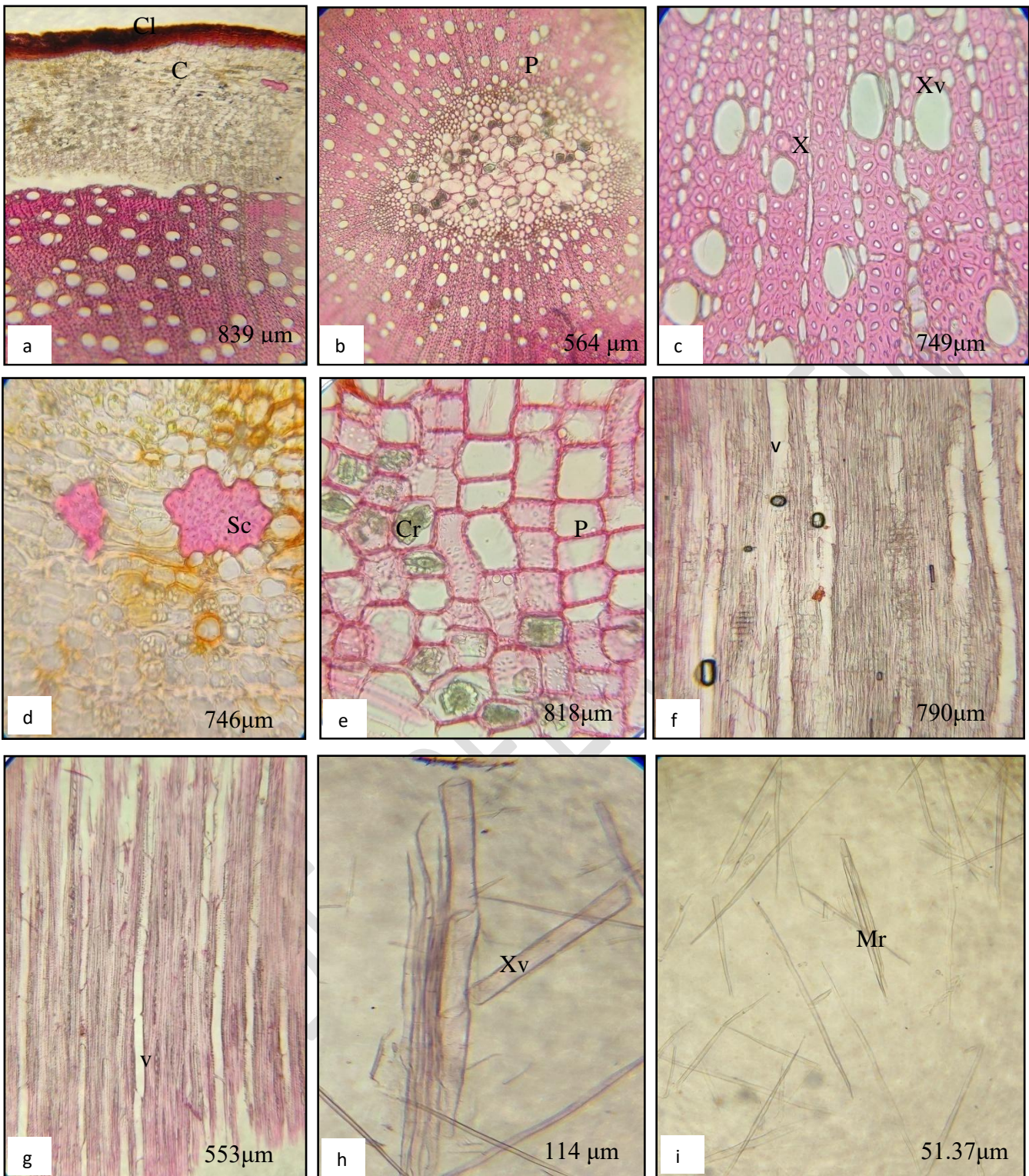


Figure 2.(a-i) Stem anatomy of *O. quadripartita* (μm)

Cl= Cork layer, C=Cortex layer, E = epidermis, P = pith, Ph = phloem, X = xylem, Xylem vessels (40x), Mr= Medullary rays length (40 X), Xm= Xylem medullary layer, Pm= Phloem medullary layer, v= vessels, Sc= Sclerides, Cr= Crystalliferous cells

References:

- Abdela, G., & Sultan, M. (2018).** Indigenous Knowledge , Major Threats and Conservation Practices of Medicinal Plants by Local Community in Heban Arsi District , Oromia , South Eastern Indigenous Knowledge , Major Threats and Conservation Practices of Medicinal Plants by Local Community i. *Advances in Life Science and Technology*, 68, 8–26.
- Advisor, K. T. (2017).** *Ethnobotanical Study of Medicinal Plants Used by the Local People of Akaki District, East Shewa Zone, Oromia Regional State, Ethiopia.*
- Agize, M., Demissew, S., & Asfaw, Z. (2013).** Ethnobotany of Medicinal Plants in Loma and Gena Bosa Districts (Woredas) of Dawro Zone , Southern Ethiopia. *Topclass Journal of Herbal Medicine*, 2(9), 194–212.
- Alemayehu, G., Asfaw, Z., & Kelbessa, E. (2015).** Plant diversity and ethnobotany in Berehet District, North Shewa Zone of Amhara Region (Ethiopia) with emphasis on wild edible plants. *Journal of Medicinal Plants Studies*, 3(6), 93–105.
- Arinathan, V., Mohan, V. R., Britto, A. J. De, & Murugan, C. (2007).** Wild edibles used by Palliyars of the western Ghats , Tamil Nadu. *Indian Journal of Traditional Knowledge*, 6(1), 163–168.
- Bayissa, K. D. (2015).** *Ethnobotanical study of medicinal plants used to treat human and livestock ailments in and around Chato natural Forest, Horo District, Horo Guduru Wollega Zone, Western Oromia, Ethiopia.*
- Birhan, Y. S., Kitaw, S. L., Alemayehu, Y. A., & Mengesha, N. M. (2017).** Ethnobotanical Study of Medicinal Plants used to treat Human Diseases in Enarj Enawga District , East Gojjam zone, Amhara region, Ethiopia. *SM Journal of Medicinal Plant Studies*, 1(1), 1–20.
- Chamberlain, Charles, J. (1920).** *Methods in plant histology* (3rd ed.).
- Dires, H. (2017).** *An ethnobotanical study of medicinal plants of anuak and nuer peoples in Itang district, Gambella region, Ethiopia.*
- Evert, R. F. (2006).** Esau's Plant Anatomy Meristems, Cells, and Tissues of the Plant Body: Their Structure, Function, and Development. In *A John Wiley & Sons, Inc., Publication* (3rd ed.).
- Gemedo-dalle, T., Maass, B. L., & Isselstein, J. (2005).** Plant biodiversity and ethnobotany of Borana pastoralists in southern Oromia, Ethiopia. *Economic Botany*, 59(1), 43–65.
- Getaneh, S., & Girma, Z. (2014).** An ethnobotanical study of medicinal plants in Debre Libanos Wereda, Central Ethiopia. *African Journal of Plant Science*, 8(7), 366–379.
- Geyid, A., Abebe, D., Debella, A., Makonnen, Z., Aberra, F., Teka, F., Kebede, T., Urga, K., Yersaw, K., Biza, T., Mariam, B. H., & Guta, M. (2005).** Screening of some medicinal plants of Ethiopia for their anti-microbial properties and chemical profiles. *Journal of Ethnopharmacology*, 97(3), 421–427.
- Giday, M., Teklehaymanot, T., Animut, A., & Mekonnen, Y. (2007).** Medicinal plants of the Shinasha , Agew-awi and Amhara peoples in northwest Ethiopia. *Journal of*

Ethnopharmacology, 110, 516–525.

- Graham, J. G., Quinn, M. L., Fabricant, D. S., & Farnsworth, N. R. (2000).** Plants used against cancer – an extension of the work of Jonathan Hartwell. *Journal of Ethnopharmacology*, 73, 347–377.
- Herrera, C. M. (1984).** The Annual Cycle of *Osyris Quadripartita*, A Hemiparasitic Dioecious Shrub of Mediterranean Scrublands. *Journal of Ecology*, 72, 1065–1078.
- Kefalew, A., Asfaw, Z., & Kelbessa, E. (2015).** Ethnobotany of medicinal plants in Ada'a District, East Shewa Zone of Oromia Regional State, Ethiopia. *Journal of Ethnobiology and Ethnomedicine*, 11(25), 1–28.
- Khandelwal, K. R., & Sethi, V. (2019).** *Practical Pharmacognosy techniques and Experiments* (3rd ed.).
- Khare, C. P. (2007).** *Indian Medicinal Plants*.
- Mabberley, D. J. (2018).** *Mabberleys Plant- Book*.
- Maryo, M., Nemomissa, S., & Bekele, T. (2015).** An ethnobotanical study of medicinal plants of the Kembatta ethnic group in Enset-based agricultural landscape of Kembatta Tembaro (KT) Zone, Southern Ethiopia. *Asian Journal of Plant Science and Research*, 5(7), 42–61.
- Meragiaw, M., Asfaw, Z., & Argaw, M. (2015).** Indigenous Knowledge (IK) of Wild Edible Plants (WEPs) and Impacts of Resettlement in Delanta , Northern Ethiopia. *Research and Reviews: Journal of Herbal Science*, 4(3), 8–26.
- Mesfin, F. (2007).** *An ethnobotanical study of medicinal plants in Wonago Woreda, Snnpr, Ethiopia*.
- Metcalf, C. R., & Chalk, L. (1965).** *Anatomy of the Dicotyledons- Vol. II*. Oxford University Press, USA.
- Mouhajir, F., Hudson, J. B., Rejdali, M., & Towers, G. H. N. (2001).** Multiple Antiviral Activities of Endemic Medicinal Plants Used by Berber Peoples of Morocco. *Pharmaceutical Biology*, 39(5), 364–374.
- Nianhe, X., & Gilbert, M. G. (2003).** Flora of China. In *England: Science Press (Beijing) and Missouri Botanical Garden Press*.
- Rao, T. A., & Das, S. (1979).** Typology of foliar tracheoids in angiosperms. *Proceedings of the Indian Academy of Sciences*, 88 B(5), 331–345.
- Shyaula, S. L. (2012).** A review on genus *Osyris*: Phytochemical constituents and traditional uses. *Journal of Natural Pharmaceuticals*, 3(2), 61–70.
- Silva, J. A. T. Da, Kher, M. M., Soner, D., & Nataraj, M. (2016).** African Sandalwood or Nepalese Sandalwood: a Brief Synthesis. *Notulae Scientia Biologicae*, 8(1), 57–61.
- Simpson, M. G. (2010).** Diversity and Classification of Flowering Plants: Eudicots. In *Plant Systematics* (Vol. 315).

- Singh, N. P., Lakshminarasimhan, P., Karthikeyan, S., & Prasanna, P. V. (2000).** *Flora of Maharashtra State, Dicotyledons* (Vol. 1).
- Singh, N. P., Lakshminarasimhan, P., Karthikeyan, S., & Prasanna, P. V. (2001).** Flora of Maharashtra State: Dicotyledones. In *Flora of India Series 2* (Vol. 2).
- Stevens, W. C. (1907).** *Plant anatomy*.
- Subba, B., & Basnet, P. (2014).** Antimicrobial Activity of Some Medicinal Plants from East and Central Part of Nepal. *International Journal of Applied Sciences and Biotechnology*, 2(1), 88–92.
- Wallis, T. E. (1939).** *Textbook Of Pharmacognosy*.
- Wilson, B. (2018).** *Osyris lanceolata*. *The IUCN Red List of Threatened Species*, 8235, 1–8.
- Woldemichael, G. M., & Wink, M. (2002).** Concomitant occurrence of pyrrolizidine and quinolizidine alkaloids in the hemiparasite *Osyris alba* L. (Santalaceae). *Biochemical Systematics and Ecology*, 30(2), 139–149.
- Worku, A. (2018).** Ethnobotanical study on the use and knowledge of medicinal plants at three kebeles of Fedis district of Oromiya Regional state; adjacent to the Babile Elephant Sanctuary, Eastern Ethiopia. *Journal of Medicinal Plants Studies*, 6(5), 15–20.
- Yohannis, S. W., Asfaw, Z., & Kelbessa, E. (2018).** Ethnobotanical study of medicinal plants used by local people in Menz Gera Midir District , North Shewa Zone , Amhara Regional State , Ethiopia. *Journal of Medicinal Plants Research*, 12(21), 296–314.
- Zhen, S. (2022).** Flora of China. In *Flora of China* (p. 211).
- Zobel, A. M. (1985).** Ontogenesis of tannin coenocytes in *Sambucus racemosa* L. I. Development of the Coenocytes from Mononucleate Tannin Cells. *Annals of Botany*, 55(1), 765–773.