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*Short communication*

## **The microstructure of the root of *Euryale ferox* Salisb. (Nymphaeaceae) from the Russian Far East**

Dulmazhab Yu. Tsyrenova 

*Pacific National University, ul. Tikhookeanskaya 136, Khabarovsk, 680035 Russia*

*duma@mail.ru*

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**Abstract.** Roots of *Euryale ferox* Salisb. a rare hydrophyte of the family Nymphaeaceae from the Russian Far East, are anatomically studied. These roots are characterized by a primary structure with typical proportions of histological and topographic zones. There is a general aerenchymatization of tissues due to adaptation to the aquatic environment. Organ-specific characters, including the polyarchstele, endoderm with Casparian strips, and unreduced tracheal xylem elements, are present. The primary structure of the *E. ferox* root indicates its relationship with monocots.

**Key words:** hydrophyte, root anatomy, histological and topographic zones, organ-specific features.

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Краткое сообщение

# Микроструктурные особенности корней *Euryale ferox* Salisb.(Nymphaeaceae) с Дальнего Востока России

Д.Ю. Цыренова 

Тихоокеанский государственный университет, 680035, Россия, г. Хабаровск, ул. Тихоокеанская, д. 136

duma@mail.ru

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**Аннотация.** В статье представлены результаты анатомического исследования корней редкого в Дальневосточном регионе России представителя водной флоры *Euryale ferox* Salisb. из семейства Nymphaeaceae. Корни исследуемого вида характеризуются первичной структурой с типичными пропорциями гистолого-топографических зон. Отмечается общая аэренхиматизация тканей в связи с адаптацией к водной среде обитания. Органоспецифические признаки – полиархная стебла, эндодерма с поясками Каспари, нередуцированные трахеальные элементы ксилемы. Выявленная первичная структура корня *E. ferox* свидетельствует о его родстве с однодольными.

**Ключевые слова:** гидрофит, анатомия корня, гистолого-топографические зоны, органоспецифические признаки

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## Introduction

Problems of anthropogenic transformation of the vegetation cover are currently widely discussed. In the Russian Far East, an unfavorable ecological situation is developing due to the transformation of ecosystems affected by logging, fires, mining and agricultural development, and urbanization (Kryukova, 2013; Schlotgauer, 2007). There are catastrophic zones where 90% of rare plants are no longer present in their former habitats due to the destruction of indigenous vegetation. It has also been shown that over the past ten years, in the Lower Amur region alone, the total number of species listed as rare and threatened has almost doubled (133 to 265). Hence, research aimed at expanding knowledge of rare and threatened species in this region is an urgent task. Studying the features of distribution, ecology, biology, morphology, and adaptability potential of populations to anthropogenic and climatic transformations of the natural environment will allow the assessment of the

current state of vulnerability of populations of these species and, if necessary, will enable measures for their reintroduction.

Currently, the underground organs of most species of higher plants are poorly studied in comparison with their aboveground parts (Tarshis, 2008). The fragmentation of our knowledge about the structural features of underground organs makes it difficult to use them for the purposes of taxonomy and phylogeny, as well as for understanding the adaptive evolution of plants. One of the tasks of modern botany is the study of specific anatomical characters of subterranean organs in representatives of various taxa and ecological groups.

The object of this study is the prickly waterlily (*Euryale ferox* Salisb.) of the family Nymphaeaceae. The rarity of this species in the Far East region is associated with both natural and anthropogenic factors. *E. ferox* is a representative of an ancient South Asian subtropical genus (Tsvelev, 1987). In the temperate

zone of East Asian countries (China, the Korean Peninsula, Japan) and the south of the Russian Far East, the range of the species is relict (*Euryale ferox*..., 2022; Lee, 1996; Ohwi, 1965). The natural localities of the prickly waterlily in Russia are noted mainly in the valley of the Ussuri River (Tsvelev, 1987). The species was also found in the Middle Amur region within the city of Birobidzhan, Jewish Autonomous Region (Rubtsova, 2009). This is the northernmost locality in the Far Eastern part of the range, located about 300 km north of other known sites. The biomorphology of the species is distinct, in that it is represented by an annual-biennial life form, while most representatives of the Nymphaeaceae are perennials (Bezdelev and Bezdeleva, 2006; Takhtadzhyan, 1987). Anthropogenic factors affecting the frequency of the species include regulation of the flow of rivers in the Amur basin, which causes almost irreversible disturbances to the hydrological regime of floodplain oxbow lakes and shallow backwaters, which are optimal habitats for *Euryale*.

The external morphology, the biology of seed reproduction in culture, and the distribution of the species in the region have been studied in detail (Kryukova, 2013; Murdakhaev, 1976; Pavlenko, 1971). The taxonomic summaries of Angiosperm Phylogeny Website also contain some information on the floral biology and histogenesis of the spermoderm in *E. ferox*. In addition, the results of an anatomical study of the aerial organs of the species have been published (Tsyrenova and Sharin, 2021).

This report presents the results of further anatomical studies of the vegetative organs in *E. ferox* in order to establish the constitutional and organ-specific characters of the root microstructure.

## Material and methods

The material for the study was collected from a stagnant lake in the vicinity of the village of Orenburgskoe (Bikinsky District of the Khabarovsk Region), located in the floodplain of the Bikin River (N 46°47'20", E 134°14'50"). The area of the reservoir is 55.8 km<sup>2</sup>. The maximum depth is about 3 m (local resident, pers. comm.). The bottom is lined with large pebbles covered with loose silty sediments. It experiences an increased recreational load in the summer swimming season. Coastal sediments are trampled by domestic animals. *Euryale* is found throughout the entire length along the southern shore of the lake, forming continuous monodominant communities.

Parts of organs from three different individuals were selected for the study. The material was fixed in 70% alcohol. The studies were carried out according to the generally accepted method of microstructural analysis (Furst, 1979). Sections of organs were made with a razor blade by hand in three repetitions. Staining was with an alcoholic solution of phloroglucinum with a drop of concentrated hydrochloric acid.

The finished micropreparations were examined using a Biolam-LOMO microscope and a digital microcomplex Altami BIO 8. The cell size and the proportion of histological zones were determined using a standard eyepiece-micrometer M-LOMO and an OMO object-micrometer were used. Micropreparations were visualized with a digital camera for a ToupCam microscope using the ToupView software. Primary research materials are housed in the cabinet of botany of the Pacific State University.

## Results and discussion

### Morphological characters of the species

In the surveyed water body, *E. ferox* plants are quite large, reaching a height of 50–60 cm. All parts of the plant are covered with strong, sharp spines. The shoot system of *E. ferox* is vegetative-generative, including a short rosette shoot and flower stalks. Peduncles bear solitary bisexual flowers. No specialized shoots of vegetative renewal are present. The leaves are bifacial with photosynthetic plates floating on the surface of the water. Diameter of leaf blades 30–45(60) cm. The phyllotaxis is spiral. In the course of the main genetic spiral, leaves alternate with peduncles. Scale-like, non-photosynthetic stipules are located at the upper metameres. The root system of *E. ferox* is fibrous. The roots are cord-like, numerous, arising at the base of the petiole of each leaf. The root system is completely immersed in the soil of the basin, fixing the rosette shoots of the plant in a certain area of the water. By the end of the growing season, the root system is completely destroyed as the individual dies.

Ecologically, *E. ferox* is a typical hydatophyte. Shoots fixed at the bottom of reservoirs carry leaves with plates floating on the water surface. The tops of the flowers are also advanced into the air. In the water column there are young, unfolded leaves and unopened flowers; as they develop later, all of them are exposed to an insolated situation. When describing the aquatic vegetation of the surveyed water body, it was found that *E. ferox* occurs in the coastal overgrowth zone among submerged macrohydrophytes. This community includes the following species: *Ceratophyllum demersum* L. (abundant), *Trapa maximowiczii* Korsh. (occasional), *Potamogeton maakianus* A. Benn. (occasional), *Potamogeton berchtoldii* Fieb. (rare), *Hydrilla verticillata* (L. f.) Rich. (rare), *Sagittaria natans* Pall. (rare), *Utricularia vulgaris* L. (rare), *Trapa sinensis* Oliv. (rare).

### Anatomical features of the roots

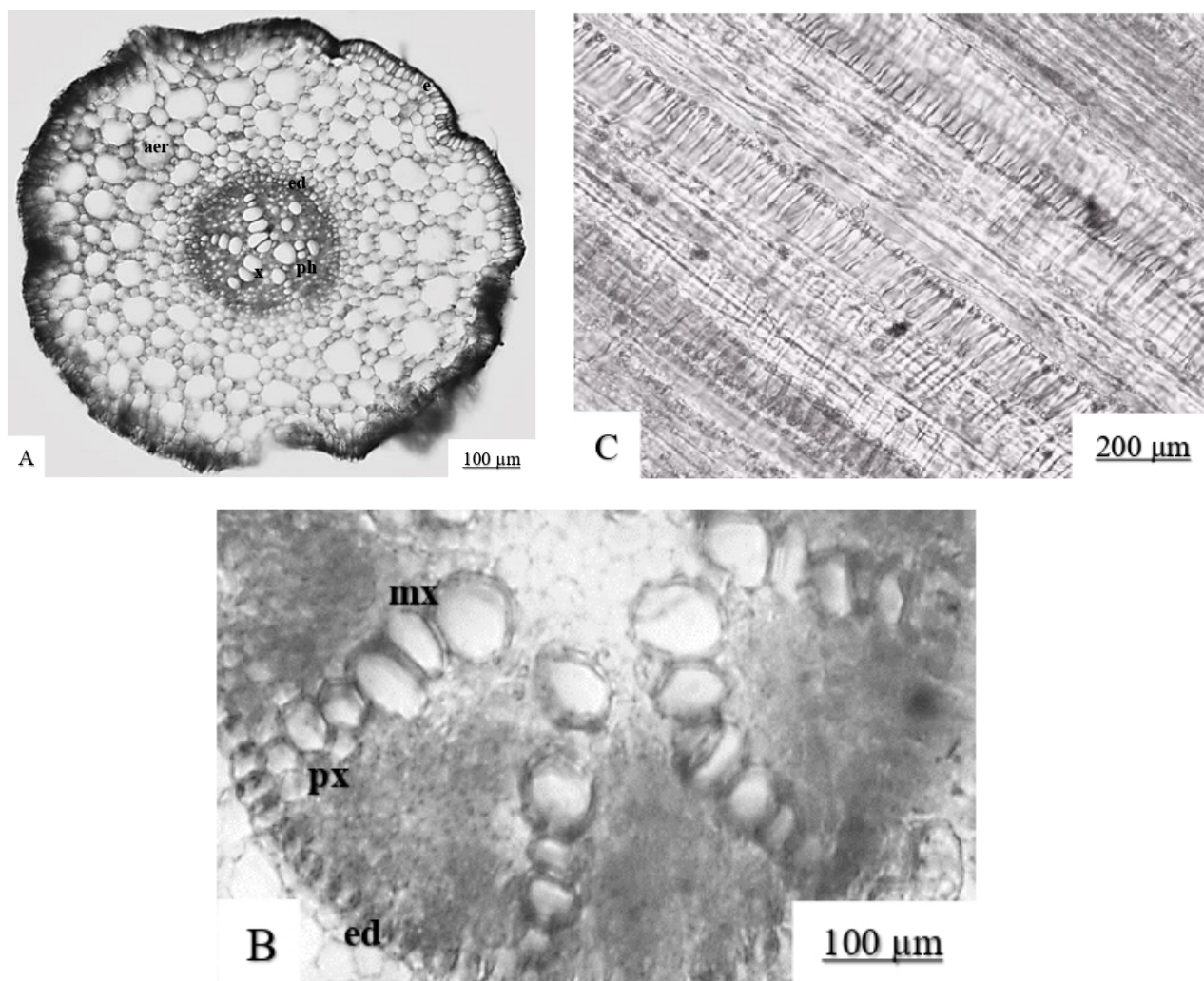
The main histological-topographic zones in the roots of *E. ferox* are well differentiated (Fig. 1A). On transverse sections of the root in the absorption zone, a single-layered epiblema (e). The epiblema is formed mainly by atrichoblasts, trichoblasts are

almost absent. The primary cortex is made of aerenchyma (aer) with rounded polygonal intercellular spaces. The parenchyma of the septa is large-celled, anisocytic. Cells are thin-walled and colorless, contain starch grains. Endoderm (ed) monolayered with Casparian strips. The pericycle is formed by a layer of thin-walled tabular cells. The stele is polyarchal (Fig. 1A, B). It is represented by six to nine rays of exarch xylem (x) with well-defined protoxylem (px) and metaxylem (mx). Between the xylem rays are poorly developed sieve elements of the phloem (ph). The tracheal elements are tracheids in their prosenchymal shape and oblique ends (Fig. 1C).

The results of our studies demonstrate that the roots of *E. ferox* are characterized by a primary anatomical structure with typical proportions of histological and topographic zones. The above ground shoot axes of this plant also have a primary structure (Tsyrenova and Sharin, 2021). The reduction of cam-

bial activity in the studied species may be associated with its annual life form. It may also be interpreted as an evolutionary character of Nymphaeaceae, which are a hydrophytic lineage of terrestrial plants.

The general aerenchymatization of the main tissues of the organs is a result of the aquatic habitat of the species. However, in the roots of this plant, there are no signs of structural reduction associated with living in the aquatic environment. A normally developed endoderm with Casparian strips is present. The xylem has unreduced tracheal elements. While the tracheal elements of the xylem are completely replaced by air canals in aboveground shoots (Tsyrenova and Sharin, 2021), they are not subject to reduction in roots. In this regard, the root system of *E. ferox* is characterized by greater tissue differentiation than shoot systems. At the same time, no astrosclereids were found in the roots of the studied species, which are present in the aboveground parts.



**Fig. 1.** A – cross section of the root: e – epiblema, aer – aerenchyma, ed – endoderm, ph – phloem elements, x – xylem elements; B – fragment of the stele: px – protoxylem, mx – metaxylem; C – longitudinal section of the root.

Other organ-specific characters of the root microstructure of *E. ferox* include the presence of a polyarchic stele. Aboveground shoot axes of plants of the species are characterized by atactostelia (Tsyrenova and Sharin, 2021). These microstructural features of the vegetative organs of *E. ferox* suggest its monocot affinity.

## Conclusions

The anatomical structure of the underground organs of *E. ferox*, a rare relictual taxon in the aquatic flora of the Russian Far East, is studied. The obtained data supplement the general characteristics of the species and provide grounds for interpreting its relationship in the context of taxonomy and phylogeny. Probably, the internal anatomical organization of the species, developed during its long evolution, contributes to its ability to resist anthropogenic transformations of the natural environment. This is supported by the development of cenoses including the studied species in water bodies with a significant anthropogenic load near settlements (for example, the city of Birobidzhan, the village of Orenburgskoe), as well as the occurrences of the species in the more northern regions of the Far East (for example, in the floodplain of the Bira River in the Jewish Autonomous Region). It can be assumed that the current climatic conditions in the region contribute to the northward expansion of the range of this subtropical species.

## ORCID

D.Yu. Tsyrenova  [0000-0003-3892-309X](https://orcid.org/0000-0003-3892-309X)

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