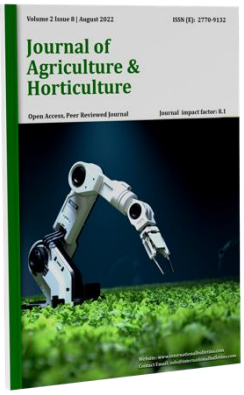

ANATOMICAL STRUCTURE OF DWARF CHERRY ROOTSTOCKS WIDELY USED IN UZBEKISTAN: MORPHOLOGICAL FEATURES AND ADAPTIVE TRAITS

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https://doi.org/10.5281/zenodo.17424414

Abstract

The anatomical structure of cherry (Prunus avium L.) dwarf rootstocks remains insufficiently described in the scientific literature, particularly regarding stem tissue organization and its relationship to growth characteristics. This study provides the first detailed anatomical assessment of the stems of dwarf cherry rootstocks—specifically the *VSL-2* (Krymsk-5) and *Prunus mahaleb* (Mahaleb cherry, used as control). The objective was to investigate structural features and adaptive traits associated with the dwarfing phenotype and cambial activity. Transverse stem sections were analyzed using light microscopy, with quantitative morphometric parameters statistically processed. The findings reveal both commonalities and significant differences between rootstocks in terms of periderm structure, vascular tissue development, and calcium oxalate crystal formation. These anatomical distinctions reflect adaptive strategies and may underlie key physiological mechanisms responsible for dwarfism and environmental resilience. Results contribute novel insights into the functional anatomy of cherry rootstocks and support their further use in breeding and horticultural practices.

Keywords: cherry, rootstocks, stem anatomy, secondary growth, dwarfing, adaptive traits, sclerenchyma, cambium activity, calcium oxalate crystals

1. Introduction

Dwarf rootstocks play a vital role in modern intensive fruit production systems, offering key advantages such as tree size control, increased productivity, and improved orchard management efficiency [5], [1], [9]. In sweet cherry (*Prunus avium* L.), the development and utilization of dwarf and semi-dwarf rootstocks have received significant attention in recent decades. Detailed anatomical studies of various cherry rootstocks under European growing conditions have been conducted by Zoric Lana et al. [8] and S.L.C. Garibay et al. [6]. Nevertheless, the anatomical mechanisms underlying dwarfism in these rootstocks remain insufficiently understood. While most existing research emphasizes agronomic traits and field performance, detailed studies of internal structural organization – particularly stem anatomy, which is fundamental to water transport, mechanical support, and phytohormonal regulation – are still limited.

Given that stem anatomy reflects both genetic background and adaptive responses to environmental conditions, its analysis can provide critical insights into the physiological behavior of rootstocks. Moreover, anatomical traits may underlie key differences in growth vigor, mechanical stability, and resistance to abiotic stressors.

This study addresses this gap by presenting a **detailed anatomical characterization** of stem tissues in two contrasting cherry rootstocks: the dwarf *VSL-2* (Krymsk-5) and the vigorous *Prunus mahaleb* (Mahaleb cherry). The aim was to identify diagnostic anatomical markers of dwarfism and evaluate potential adaptive features relevant to plant performance under varying environmental conditions.

2. Materials and Methods

2.1 Plant Material

Two rootstock genotypes of cherry were selected for anatomical study:

- **VSL-2 (Krymsk-5):** A dwarf rootstock of vegetative origin known for its reduced tree size and compatibility with many sweet cherry cultivars.
- **Prunus mahaleb L.:** Used as a control; a vigorous, widely used rootstock in cherry cultivation, especially in semi-arid regions.

Fresh stem segments were collected from the basal portion of current-year shoots during the active growth period. At least 10 biologically independent plants per genotype were sampled.

2.2 Fixation and Sectioning

Plant samples were fixed in **70% ethanol** to preserve cellular integrity. Transverse sections (~20–30 µm thick) were hand-cut using a sterilized safety razor. Sections were stained with **safranin**, which selectively stains lignified and suberized tissues, and mounted in **glycerin-gelatin medium** following the protocol of Barykina et al. [2].

2.3 Microscopy and Imaging

Prepared slides were examined under a **Motic B1-220A-3 light microscope** equipped with a **Canon A123 digital camera** and microphoto attachment. Digital micrographs were taken at various magnifications for documentation and quantitative analysis.

2.4 Quantitative Analysis

Anatomical measurements were performed using a calibrated **ocular micrometer**, converted to micrometers (µm). Each parameter was measured in **30 replicates** per tissue type and plant. The following traits were analyzed [3], [4]:

- Diameter of parenchyma cells
- Diameter and number of vessels in secondary xylem
- Thickness and diameter of cortical parenchyma
- Diameter and wall thickness of sclerenchymatous fibers
- Diameter of the central pith

2.5 Statistical Processing

Data were statistically analyzed using **Microsoft Excel** based on the principles of experimental statistics [7]. For each trait, **mean values and standard deviations** (±SD) were calculated. Differences between genotypes were assessed descriptively and graphically.

3. Results

3.1 General Stem Anatomy

In both rootstocks, transverse stem sections exhibited a **circular outline** and a **non-fascicular organization**. The vascular tissues formed a nearly continuous **secondary cylinder**, indicative of robust cambial activity.

The **periderm** was well-developed, consisting of multiple layers of radially oriented cork cells (phellem) with thick, suberized walls. These cells appeared dark-brown under the

microscope and formed a dense, compact protective barrier. The phellogen was active, producing phellem cells outward and phelloderm cells inward.

Beneath the periderm, the **primary cortex** was composed of oval parenchymatous cells, which were retained along the entire stem circumference. **Sclerenchyma fibers** (primary phloem fibers) formed discrete clusters within the cortex, marking the transition to the phloem zone.

3.2 Vascular Tissue Organization

The **secondary phloem** lay internal to the cortex and included both thick-walled fibers (hard phloem) and thin-walled elements (soft phloem). The **secondary xylem** formed the bulk of the stem, arranged in a solid concentric cylinder surrounding the pith. Cambial activity produced **annual growth rings**, evident as alternating bands of earlywood and latewood.

The secondary xylem consisted of **tracheary elements arranged in radial files**, with clearly discernible **multiseriate medullary rays**, contributing to the mechanical and conductive functions of the stem.

The **pith** occupied a central position and comprised large, round to oval parenchyma cells. The peripheral pith zone (perimedullary region) contained smaller, compact cells. **Hydrocytic cells** were also observed, suggesting a role in water storage or redistribution.

3.3 Comparative Anatomical Traits

Trait	VSL-2 (Krymsk-5)	Prunus mahaleb
Stem diameter (µm)	12988.5 ± 118.53	Lower values observed
Width of xylem-phloem cylinder (µm)	4870.73 ± 45.15	Moderately developed
Thickness of cortical parenchyma (µm)	Moderate	523.80 ± 5.13
Abundance of calcium oxalate crystals	Rare or absent	Numerous, variable morphology
Crystal morphology	Mostly absent	Prismatic, cubic, druse forms
Sclerenchymatous fiber development	Extensive	Moderate
Cambial activity (growth ring clarity)	High, distinct annual rings	Moderate to high
Pith structure	Narrow, compact	Looser, larger parenchyma

3.4 Crystal Formation and Adaptive Significance

In *Prunus mahaleb*, calcium oxalate crystals were frequently observed in cortical parenchyma cells, occurring as **monocrystals and druses**. These structures were absent or infrequent in *VSL-2*. The presence of crystals may represent a defense mechanism or an adaptive response to nutrient imbalance and environmental stress (e.g., drought or salinity). Their role in regulating cellular calcium and detoxification should not be overlooked.

4. Discussion

Our findings reveal that **both rootstocks share a conserved stem anatomical framework**, with significant **quantitative differences** that likely underlie functional

divergence in growth habit and environmental adaptation.

The **dwarf phenotype of VSL-2** correlates with:

Thicker xylem and phloem regions;

Higher density of sclerenchyma fibers;

Narrower pith and reduced crystal formation.

These features suggest a structural optimization for **mechanical support and limited vertical growth**, likely contributing to dwarfing behavior through modulation of hydraulic conductivity and mechanical resistance.

Conversely, *Prunus mahaleb* exhibits:

Greater cortical parenchyma development;

Higher frequency and diversity of oxalate crystals;

Less robust lignification.

This anatomical profile may reflect **greater plasticity and adaptability**, allowing vigorous growth under variable environmental conditions. The abundance of crystals could also reflect metabolic detoxification mechanisms.

These anatomical features may serve as **morphological markers** for the selection of rootstocks with desirable traits in breeding programs.

5. Conclusion

This study provides the first comprehensive anatomical assessment of stems in dwarf and vigorous cherry rootstocks. Key conclusions include:

VSL-2 (Krymsk-5) exhibits stem anatomical traits associated with dwarfism, including extensive secondary growth and reduced crystal formation.

Prunus mahaleb shows greater cortical complexity and higher oxalate crystal abundance, suggesting enhanced environmental adaptability.

Anatomical characteristics of stems, particularly vascular development and secondary tissue organization, may serve as **diagnostic indicators of rootstock vigor** and resilience.

Future research should explore the **functional implications** of these traits in relation to water transport efficiency, mechanical stability, and interactions with scions under field conditions.

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