

Postprint: Stomatal Changes in *Populus euphratica* Leaves Under Drought Stress

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Abstract

Investigating the stomatal characteristics of *Populus euphratica* leaves facilitates the interpretation of water use traits and drought resistance mechanisms under drought stress conditions. This study selected three typical sections in the lower reaches of the Tarim River—Yingsu, Alagan, Yiganbu, and Ma—using *Populus euphratica* as the research subject to explore variation patterns in leaf stomatal density and stomatal length along different drought stress gradients, and to analyze response differences between female and male *Populus euphratica* plants under drought stress. The results demonstrate: Stomatal density in *Populus euphratica* leaves exhibits an increasing trend with intensifying drought stress, whereas leaf stomatal length shows a decreasing trend under the same conditions;

Under drought stress, the rates of change in stomatal density and stomatal length differ between female and male *Populus euphratica* leaves, specifically manifested as: the growth rate of abaxial epidermal stomatal density in male plants is relatively small, while the negative growth rate of abaxial epidermal stomatal length in female plants is significantly greater than that in male plants;

Changes in stomatal density and length of *Populus euphratica* leaves reflect the drought resistance capacity of female and male plants; based on these variations, male *Populus euphratica* plants exhibit stronger water retention capacity and tolerance than female plants under drought stress conditions.

Full Text

Preamble

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Abstract:

Investigating stomatal characteristics of *Populus euphratica* leaves is essential for understanding water use patterns and drought resistance mechanisms under water stress. This study examined three typical *P. euphratica* stands (Yengisu, Aragan, and Yikanbujima) in the lower reaches of the Tarim River, analyzing variations in stomatal density and stomatal length under different drought stress gradients and comparing responses between male and female individuals. The results revealed: (1) Stomatal density in *P. euphratica* leaves increased while stomatal length decreased with intensifying drought stress; (2) Under drought conditions, significant differences in stomatal density and length emerged between male and female plants, with male individuals exhibiting slower rates of stomatal density increase in leaf epidermis; (3) Female plants displayed significantly longer stomata in lower epidermis compared to male plants. These changes in stomatal density and length reflect sex-specific drought resistance strategies, with male *P. euphratica* demonstrating superior water retention and drought tolerance relative to female plants.

Keywords: *Populus euphratica*; stomatal density; stomatal length; lower reaches; Tarim River

2. Methods

2.1 Data Collection

In August 2017, leaf samples were collected from three representative *P. euphratica* profiles (Yengisu, Aragan, and Yikanbujima) in the lower Tarim River region. Sampling locations were established at two distances from the river channel: 50 m and 500 m. At each site, healthy, mature leaves were harvested from both male and female trees. Soil samples were collected from depths of 15-40 cm at three points per location. Leaf specimens were fixed in FAA solution (38% formaldehyde:70% ethanol:distilled water = 5:5:90) for subsequent anatomical analysis.

2.2 Stomatal Measurements

Stomatal density (SD) and stomatal length (SL) were measured using a ZEISS optical microscope at 63 \times magnification. For each sample, ten fields of view were randomly selected. Stomatal density was calculated as the number of stomata per unit area ($n \cdot \text{mm}^{-2}$), counting all stomata within each field. Stomatal length

was measured for 25 randomly selected stomata per sample. All measurements were recorded in Excel and statistically analyzed using SPSS software to examine differences among treatments, distances, and sexes.

3. Results and Discussion

3.1 Stomatal Density Response

Stomatal density across all samples ranged from 56 to 152 $\text{n} \cdot \text{mm}^{-2}$, with a mean of 91 $\text{n} \cdot \text{mm}^{-2}$. Under control conditions, mean density was 102 $\text{n} \cdot \text{mm}^{-2}$; under moderate drought stress, 80 $\text{n} \cdot \text{mm}^{-2}$; and under severe stress, 79 $\text{n} \cdot \text{mm}^{-2}$ at 50 m distance and 102 $\text{n} \cdot \text{mm}^{-2}$ at 500 m distance. Statistical analysis revealed significant differences in stomatal density among drought stress treatments ($P < 0.01$). Both stress level and distance from the river channel significantly affected stomatal density, with a significant interaction between these factors ($P < 0.01$). As shown in Fig. 2, stomatal density exhibited clear differentiation across treatments, with moderate and severe stress inducing significant reductions compared to control conditions.

3.2 Stomatal Length Response

Stomatal length ranged from 23.26 to 28.93 μm , averaging 25.81 μm . Control plants averaged 25.03 μm , moderate stress 26.58 μm , and severe stress 26.86 μm at 50 m versus 24.76 μm at 500 m. Significant differences in stomatal length were detected among treatments ($P < 0.01$), with stress level, sampling distance, and their interaction all exerting significant effects. Fig. 3 illustrates that stomatal length tended to increase under moderate drought stress but decreased at the 500 m distance under severe stress, suggesting plastic responses to water availability.

3.3 Sex-Specific Drought Adaptation Strategies

The divergent responses of stomatal traits between sexes reflect different drought resistance mechanisms. Similar patterns have been observed in other desert species such as *Sophora moorcroftiana* and *Ammodendron argenteum*. The greater stomatal length in female plants may enhance gas exchange efficiency under moderate stress, while the higher stomatal density and water retention capacity in male plants confer superior drought tolerance. These findings align with previous research demonstrating that *P. euphratica* employs multiple adaptive strategies, including stomatal regulation, to cope with extreme aridity. The sex-based differences suggest that male individuals are better adapted to severe water deficit, making them potentially more suitable for afforestation in hyper-arid regions.

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