Effect of shading on the English oak (*Quercus robur* L.) seedlings water regime

A Popova* and V Popova

Department of Botany and Plant Physiology, Voronezh State University of Forestry and Technologies named after G.F. Morozov, 8 Timiryazev street, 394087 Voronezh, Russian Federation

*E-mail: lf_popovaaa@vgltu.ru  https://orcid.org/0000-0003-4711-5377

Abstract. Shading is a common urban stress in the city due to the light block from buildings. Oaks stands are found as part of park areas, forests and in point gardening in many large cities of the world. In this work we examined anatomical and physiological aspects of the adaptation of oak seedlings to the lack of light. To determine the intensity of transpiration, leaf hydration, and water deficiency, the weight indicators of the leaves were measured. Anatomically, the analysis of the leaf epidermis was carried out by microscopy of varnish prints. Changes were revealed in the anatomical structure of the leaf and were irreversible in the water regime undo shading. It was found the correlation between stomatal apparatus of the leaves and incoming solar radiation amount. With the optimal combination of hydrometeofactors, the evaporation rate in the heavily shaded leaves during the day did not exceed 15 ± 0.9 g/(m²h), which was 30…35% of the transpiration intensity in the well-lit control leaves. Then, shading by 80% reduced transpiration by 50…65 %. The influence of the degree of shading on the water regime of oak seedlings is shown for the first time at forest-steppe region.

1. Introduction

A change in any of the environmental factors in the direction of its increase or decrease leads to a change in both plant morphogenesis and individual physiological processes. The formation of the urban environment leads to a change in the place of growth of plants, their inclusion to the biotechnosphere of cities. The ecology of the city, in contrast to natural communities, is characterized by the presence of thermal islands, light noise, salinization and changes in the movement of air masses, air humidity, and precipitation. The growth of plants in urban conditions of tall buildings is associated with a shortage of solar radiation, a decrease in the level of illumination. Trees in the urban environment form the ecological framework, ecosystem services provided by green spaces in cities are diverse and are of great importance - this is the improvement of the temperature regime and microclimate, carbon storage, the weakening of the storm flow, the absorption of pollutants from the air [1-3].

The selection of adapted species and genera for the green architecture of megacities is important from the point of view of forming a long-lasting favorable human habitat. At the same time, great importance is attached to the protective and adaptive reactions of the plant organism that occur in response to adverse external influences, including a lack of light.

Members of the genus Quercus are among the longest-lived woody plants and are distributed throughout the Northern Hemisphere. The genus Quercus has hundreds of species that differ in biology and can be selected for introduction depending on the growing conditions. In many large cities of the world, different types of oak are found in park areas, forests, and in spot gardening [4-9].
Light is one of the most significant abiotic environmental factors and determines the development and metabolism of plants, photosynthesis and photomorphogenesis [10, 11]. Shading is a common abiotic stress in the city due to the light block from the city. Low light has been shown to inhibit physiological and enzymatic processes, including photosynthesis and antioxidant properties, as well as carbon and nitrogen fixation [12-14]. The light mode is an important factor for the growth of young plants and the optimal amount of light for growth depends on the type of plant. For *Aralia elata*, 65% shading stimulated growth [15], for seedlings of *Faidherbia albida*, conditions of high illumination and high soil moisture of 50…100% are optimal [16]. For oaks, different reactions are shown depending on the regions of origin [17]. Leaf dusting has a similar effect as shading; for *Quercus brantii*, regardless of the irrigation regime, the transpiration significantly reduced the relative water content in the leaves and the mass of seedlings [18].

The aim of the study is to determine the effect of shading on water regime English oak seedlings in urban.

2. Methods and materials

The object of the study was the English oak (*Quercus robur* L.), which belongs to the main forest-forming species of broad-leaved forests of Central Russia [18].

Experimental studies were carried out in plantations of Voronezh State University of Forestry and Technologies named after G.F. Morozov: Voronezh (geographical coordinates: 51.735417, 39.188404). The research carried out on 2-year-old seedlings of English oak. For this purpose, we shaded the seedlings for two months during the growing season. The degree of shading provided gauze and dense cotton of 1 and 2, 3 layers with shading up to 40, 60 and 80%, respectively. The scheme of the experiment contained control-full illumination and 0% shading. The light level measured with a luxmeter U-116 (Pribortech, Russia).

In July, the transpiration rate, water content, and stomata in the leaf epidermis measured in two temperature variants: normal temperatures (20…25°C, 70% humidity) and elevated temperatures (25…30°C and 55% humidity).

The studies were carried in the field and in the laboratory. The leaves weighed in the field on a torsion scale VT-500 (TSNIHBI, USSR, accuracy ± 0.01 g) to calculate the transpiration rate by “rapid weighing”: to do this, after you tear off the sheet, weigh it and re-weigh it after 5 min, the weight indicators used to calculate the transpiration intensity (equation (1)).

The transpiration rate (*Tr*) was calculated using the following equation (1):

\[ Tr = \frac{10^3 \times (A - B)}{S \times t}, \]

where, *A* is the weight of the sheet at the beginning of the measurement, g; *B* – weight of the sheet after 5 min, g; *S* – area of the sheet, m²; and *t* – time of the experiment, h.

The analysis of tissue hydration, water deficiency, dry matter content, microscopy of the leaf epidermis were carried in laboratory. The water content in the leaf tissues and the dry matter content was assessed by weighing on a scale MWP-300 (CAS, Korea) and further drying in a dry-heating oven GP-20-Ox (Amedis Engineering, Russia). The water deficit in the leaf tissues was determined by measurements of weighing at the beginning after full saturation them in Petri dishes (1 h at 25°C) and after further drying in a dry-heating oven until the leaf material completely dried. The measurements were used in equation 2.

The water deficit (*Wd*) was calculated using the following equation (2):

\[ Wd = \frac{(b - a)}{(b - c)} \times 100 \]

where, *a* is the initial weight of the leaves, g; *b* is the weight of the leaves after saturation, gl; *c* is the weight of the leaves after drying, g.

The number of stomata calculated in 10 fields of view on preparations was obtained by the fingerprint method (Carl Zeiss microscope, Austria, magnification 10×40). For each variant of illumination, three
repetitions were laid, from each of which 20 samples were taken. At the same time, the temperature and humidity of the air were measured using a psychrometer and a thermometer, respectfully.

3. Results and discussion
It turned out that almost all the processes associated with it detect self-oscillations when observing the kinetics of water exchange of the sheet. Diurnal (circadian) rhythms are important for the intensity of transpiration. The closing of the stomata at noon can be associated with both an increase in the level of CO2 in the leaves with an increase in air temperature (due to increased respiration and photorespiration), and a possible deficiency that occurs in the tissues at high temperature, low humidity, and especially in windy weather. The decrease in air temperature in the afternoon promotes the opening of stomata and the enhancement of photosynthesis [1]. Setting up an experiment with shading in conditions of high and normal temperatures is important for studying the oak growth possibilities in urban conditions.

The data obtained on the intensity of seedling transpiration, depending on the degree of shading, are presented on figures 1 and 2. The course of daytime transpiration differs under normal and elevated temperatures. At temperatures of 20-25°C the restoration of the transpiration course after 15 h to 28 g/(m²h) is observed in conditions of shading of 40%. When shading was 60 and 80%, the transpiration intensity decreases and has a difference with zero dimming of more than 20 units.

At the temperature of 25…30°C, the initial course of transpiration coincides with any shading. At 40% shading the maximum of transpiration is observed at 15 h (34±0.9 g/(m²h)), while for shading 60 and 80%, the maximum of transpiration occurs at 13 h with 20±0.7 and 15±0.9 g/(m²h), respectively. There is no second peak on the intensity curve of the daily transpiration of English oak seedlings. This indicates that the leaves of the English oak are xeromorphic. In comparison with other deciduous woody plants, oaks show low transpiration activity.

The heavily shaded leaves, receiving only 20% of the sunlight, caused changes of daily course of the transpiration process (figure 2, line 80%). In the control and 40% shading during the day shaded leaves the curves had a single-vertex, dome-shaped character with maxima at 10 and 13 h – in moderately warm weather and at 13…15 h – in hot weather. With a strong lack of light (80% shading), the intensity of transpiration fell to a minimum value (9±0.6; 12±1.2; 15±0.9; 11±0.7; and 10±1.0; in accordance with the study time from 9 to 18 h) and acquired monotonous character, reacting little to the temperature and humidity of the air. With the optimal combination of hydrometeofactors, the evaporation rate in the heavily shaded leaves during the day did not exceed 15 ± 0.9 g/(m²h), which was 30…35% of the transpiration intensity in the well-lit control leaves. Shading by 80% reduced transpiration by 50…65%.

![Figure 1. Transpiration intensity at 20...25°C at different shading.](image1)

![Figure 2. Transpiration intensity at 26...30°C at different shading.](image2)

When the temperature reached 30°C, under sufficient humidification conditions, all leaves had a minimum of transpiration. Intensity of transpiration was decreased at 0 and 60% shading after 15 h and at 80% shading from 13 h. Comparing the daily transpiration curves at moderate and high temperatures, we can note an increase in transpiration in hot weather, sharper peaks. Shading of 80% disrupts the natural course of transpiration in both temperature regimes. In fact, the lack of light had the same effect
as drought, reducing transpiration to a minimum value without significant changes during the day (figure 2) and reducing the number of stomata (figure 3, table 1).

The analysis of the number of stomata showed, that in the field of view of the microscope, the leaves shaded (80%) had only $13.6 \pm 0.6$ stomata, while the well-lit leaves (0 % of shading) had $23.0 \pm 1.8$ or more of them in the same area (figure 3), that is, 1.7 times more. The level of 14 stomata was shown at the shading of 40 and 60 % (table 1).

**Table 1.** The number of stomata in the leaves of the English oak when shaded (in the field of view of the microscope).

| Shading, % | 0     | 40    | 60    | 80    |
|------------|-------|-------|-------|-------|
| Number of stomata | 23.0±1.8 | 14.8±0.6 | 14.2±0.7 | 13.6±0.5 |

![Microphotography of stomata of English oak](image)

**Figure 3.** Microphotography of stomata of English oak (lacquer prints from the leaves of well-lit seedlings), magnification of $10 \times 40$.

Shading led to an increase in the water content of the leaves. The water content in the leaves when the degree of 80% shading of more than 10% higher water content in the leaves of seedlings is well lit (figure 4, where the differences in the degree of 80% shading reliable).

![Leaves moisture content at different time](image)

**Figure 4.** Leaves moisture content at different time.

![Water scarcity, %](image)

**Figure 5.** Water scarcity, %.
The water content changed in English oak seedlings leaves during the day. So in well-lit control leaves, in the morning, the water content was maximum, and it decreased by 13 and 15 h. As the shade increases, the hydration of the leaves increased both in the morning and throughout the day. This difference was especially clear in the afternoon. So, if the illuminated leaves had a water content of just over 68% at 15 h, then as the shading progressed, this value increased to 74-75% when less than half of the solar radiation was received. And, finally, with strong shading up to 80%, the humidity of the leaves of the English oak was almost 81%. The air temperature during the sampling period ranged from 26.5 to 28.5°C.

Calculation of water deficit showed a decrease of water flow and increasing water scarcity seedling under conditions of a high degree of shading and reduce the intensity of transpiration. The change in the water deficit of the leaf tissues of the English oak with different shading shows in figure 5. The differences with the degree of shading 80% are reliable.

Mediavilla et al. [19] showed the effect of higher temperatures and the minimum daily water potential on the anatomy of the epidermis of oak leaves. On the western side, where high temperatures are noted, there is a high density of stomata. With an increase in the degree of shading, the water deficit of leaf tissues increases and reaches 30.38 ± 0.07 % (80% shading). Similar changes are observed for the leaves in the tree crown. Thus, 80% shading leads to a change in the water regime, the level of water deficiency increases and exceeds 25 %, changes in the morphology of stomata, the intensity of transpiration. Taking into account that different reactions of the water regime to shading and temperature are shown for different plant species [15-17], the obtained data are valuable for growing oak under shading conditions and the possibility of comparing the data with other woody plants.

4. Conclusion
As a result of field, when trees are growing in an urban environment, and laboratory studies of oak petiolate in different lighting conditions it was found that the lack of light leads to a violation of the water balance of plants. This is evidenced by the following processes: when shading, the activity of transpiring processes decreases, the water content of the leaves increases and their water deficit increases. It was found that the morphogenesis of the stomatal apparatus of the leaves of the petiolate oak has a direct correlation with the amount of incoming solar radiation, as evidenced by the change in the total number of stomata. Shading by 80% leads to a significant increase in the water deficit of oak seedlings, changes in the anatomical structure of the leaf epidermis by reducing the number of stomata are irreversible in the water regime. Thus, for young oak plants, it is necessary to select growing conditions with a sufficient level of illumination, shading should not exceed 60 %. The novelty of the study is to establish changes in the transpiration activity of seedlings in the urban environment, depending on the temperature and illumination of the environment, which can be used in the design of landscaping areas, the selection of planting sites in the urban environment.

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