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**DETERMINATION OF PHOTOSYNTHETIC PARAMETERS OF PLANTS OF THE
GALIUM VERUM AND *HELICHRYSUM ITALICUM* FROM THE FLORA OF THE
REPUBLIC OF MOLDOVA AND ROMANIA**

**DETERMINAREA PARAMETRILOR FOTOSINTETICI AI PLANTELOR *GALIUM
VERUM* ȘI *HELICHRYSUM ITALICUM* DIN FLORA REPUBLICII MOLDOVA ȘI
ROMÂNIA**

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Summary. This study investigates the photosynthetic responses of *Galium verum* and *Helichrysum italicum*, sourced from Romania and Republic of Moldova, under varying light conditions. Photosynthetic parameters, including assimilation rate, stomatal conductance, and dark respiration, were measured to assess adaptations in response to light intensity. Results reveal that *G. verum* plants exhibit high assimilation rates and stomatal conductance, with minor regional variations likely due to seasonal harvesting differences. *H. italicum* plants, conversely, show lower assimilation and stomatal conductance, indicative of adaptations to bright environments. Both species display similar light saturation points across origins, suggesting stable photosynthetic efficiency under controlled light. This work underscores the intricate adaptations plants utilize to optimize photosynthesis, offering insights into species-specific strategies for light use efficiency and environmental resilience.

Key words: *Galium verum*, *Helichrysum italicum*, photosynthesis.

Rezumat. Studiul investighează răspunsurile fotosintetice ale plantelor *Galium verum* și *Helichrysum italicum* din România și Republica Moldova, în condiții variate de lumină. Parametrii fotosintetici, inclusiv viteza netă de asimilație, conductanța stomatală la diferite intensități luminoase au fost măsurate pentru a evalua adaptarea, ca răspuns la intensitatea luminii. Rezultatele arată că plantele de *G. verum* prezintă viteze de asimilație și conductanță stomatală ridicate, cu variații minore datorită diferențelor sezoniere de recoltare. Plantele *H. italicum*, dimpotrivă, prezintă o viteză netă de asimilație și o conductanță stomatală mai scăzută, indicând adaptări la medii de lumină. Ambele specii au puncte de saturație a luminii similare, sugerând o eficiență fotosintetică stabilă la lumina controlată, iar lucrarea pune în valoare adaptările complexe ale plantelor pentru a optimiza fotosinteza, oferind perspective asupra strategiilor specifice pentru eficiența utilizării luminii, cât și rezistența la diferite medii de creștere.

Cuvinte cheie: *Galium verum*, *Helichrysum italicum*, fotosinteză.

INTRODUCTION

This G3P (glyceraldehyde 3-phosphate) can be converted into glucose, sucrose, or other sugar molecules. These sugars are rich in stored energy in covalent bonds. Organisms use this stored energy by breaking down these sugars, releasing the energy

needed for various cellular functions. During photosynthesis, the ambient air's carbon dioxide (CO₂) enters the intercellular space through the stomata. From there, the CO₂ dissolves in water in the cell wall and diffuses through the cell wall, plasma membrane, cytoplasm, chloroplast envelope, and stroma until it reaches the Rubisco enzyme [1]. Light intensity and quality are important environmental factors that affect plant physiology and biochemistry. For most plants, even small changes in light intensity can lead to significant changes in leaf structure and morphology [2]. Light provides the energy required for photosynthesis and plant growth, but plants live in an environment where light and dark alternate every 24 hours and day length varies with the seasons. Daily irradiance follows a roughly sinusoidal pattern, with additional, often unpredictable fluctuations due to changes in weather conditions throughout the day and from day to day. In many plant species, starch acts as a buffer and stabilizes metabolism and growth compared to the daily light-dark rhythm [3]. Photosynthesis reductions can occur through two main mechanisms [4]: (i) reduced diffusion of CO₂ into the leaf, which is associated with a reduction in internal and stomatal conductances (GI and GS, respectively), and (ii) metabolic repression, which impairs photosynthesis by slowing leaf growth and limiting cell proliferation. Furthermore, light is the sole energy source for starch biosynthesis [3]. Starch synthesis and degradation rates are adjusted according to light availability, i.e., h. As light intensity increases, starch formation increases while its degradation rate decreases [5]. Increasing photosynthesis is essential for maintaining adequate dry biomass accumulation in plants. It is generally accepted that, in addition to light intensity and duration (photoperiod), the quality of light - precisely the color or wavelength of light - plays a crucial role in influencing plant growth and photosynthesis [6]. Different qualities of light affect plants in various ways. For example, blue light (B) and red light (R) are the wavelengths most efficiently used in photosynthesis due to the absorption peaks of photosynthetic pigments, which mainly absorb blue (400-500 nm) and red (600-700 nm) light spectra [6,7]. Excessive light beyond what is required for photosynthesis can damage the photosystem and reduce productivity, while insufficient light can limit the photosynthetic process. Photosynthesis typically occurs under changing light conditions, with the transition from low light to high light requiring an induction period [8]. Reducing these induction times could present valuable opportunities for enhancing photosynthesis in natural settings [9]. Plants have evolved complex mechanisms to cope with environmental changes, and these adaptive abilities have been strengthened over a long evolutionary process. These mechanisms are active in various fields, from anatomy and physiology to biochemistry, genetics, development, evolution, and molecular biology. Each aspect contributes to the ability of plants to effectively sense and respond to external stimuli, thereby improving their chances of survival and reproduction under different environmental conditions [10]. Anatomically, plants can modify the structure of their leaves, roots, and stems to optimize the absorption of resources, such as light, water, and nutrients. For instance, leaves may become thinner and broader under low-light conditions to increase the surface area for light absorption [11]. Additionally, in nutrient-poor soils, roots can develop more extensive and branched structures to more efficiently access important nutrients. On a physiological level, plants respond to stresses like drought or salinity by adjusting stomata opening, limiting water loss while maintaining the gas exchange necessary for photosynthesis [12]. They also produce osmoregulatory compounds that help maintain water balance under water-stress conditions. Biochemically, plants synthesize a wide range of secondary compounds, such as flavonoids and terpenoids, which help them cope with biotic stress (e.g., pest attacks) and abiotic stress (e.g., UV radiation). These compounds also serve as a defense, protecting plants from pathogens or the damaging effects of intense solar radiation. Genetically and

evolutionarily, plants have mechanisms that enable long-term adaptation by selecting favorable traits in specific environments. At the molecular level, adaptability is supported by gene expression regulation, allowing plants to respond swiftly to environmental changes. For instance, in high-temperature stress, certain genes activate to produce heat-shock proteins that protect cells from damage [13].

AIM OF THE STUDY

Investigating the photosynthetic responses of *Galium verum* and *Helichrysum italicum*, species from Romania and Moldova, under different light conditions, with the measurement of photosynthetic parameters, including assimilation rate, stomatal conductance and dark respiration, to evaluate adaptations in response to light intensity.

MATERIAL AND METHODS

The plant seeds were harvested from plants of the *Galium verum* and *Helichrysum italicum* from the spontaneous flora of the Republic of Moldova (*G. verum* -Edinet and Donduseni; *H. italicum* – Scientific Practical Center of Medicinal Plants of Nicolae Testemitanu State University of Medicine and Pharmacy) and from the spontaneous flora of the Romania (Arad County) during the fruiting period of 2024 year.

The seeds harvested in 2023, were sown in 2024 in 0.5 L plastic pots filled with commercial garden soil and grown in growth chambers under controlled conditions of light ($1000 \mu\text{mol m}^{-2} \text{s}^{-1}$), light period 12/12 h, temperature (30°C), and relative humidity (65%), three to four plants per pot for each experimental species group. Plants were watered every other day, depending on the pot capacity. The studies were conducted after 21 days of sowing. In all experiments, we used the 5th pair of leaves attached to the plants. For the measurement of assimilation rate (A) and stomatal conductance to water vapor (gs), the leaves were enclosed in a portable gas exchange system GFS-3000 (Waltz, Effeltrich, Germany) to determine the photosynthetic parameters. The photosynthetic parameters of the plant leaves were measured under the following conditions: leaf temperature of 25°C , air flow rate cuvette $750 \mu\text{mol/s}$, chamber air humidity of 65%, PARtop $1000 \mu\text{mol m}^{-2} \text{s}^{-1}$, and CO_2 concentration of $400 \text{ mmol mol}^{-1}$. Standard conditions were maintained until the stomata opened and the CO_2 and water vapor exchange rates stabilized at steady-state levels. The steady-state values of assimilation rate (A) and stomatal conductance to water vapor (gs) were calculated using the method described in a previous study [14,15].

The same apparatus (GFS-3000, Heinz Walz GmbH, Effeltrich, Germany) recorded light-response curves for the photosynthetic parameters. To assess the influence of the light intensity on the photosynthetic parameters, the following sequence (light intensities in $\mu\text{mol m}^{-2} \text{s}^{-1}$) was used to make the light-response curve measurements: 0, 50, 100, 200, 400, 500, 600, 800, 1000, 1200, 1500. Other parameters remained unchanged. The gas exchange rates were maintained at each light intensity until the steady-state values were observed. Three separate measurements of the light-response curves were made. The rectangular hyperbola of Smith [16], as previously described [16, 17], was used to fit the light response curves, yielding the maximum net assimilation rate (A_{max}), initial quantum yield (α), and dark respiration rate (Rd).

RESULTS AND DISCUSSION

Species of the genus *Galium* (Rubiaceae family) have a special role in the traditions and spirituality of many people. Although, some species have been used for hundreds of years in traditional medicine, their therapeutic properties are still little highlighted, and in the Republic of Moldova they are not studied until the present [18]. Of the more than 600 species of g. *Galium* in the world, the European flora includes more than 145 species. The Romanian flora counts 38 species, while 20 species grow in the Republic of Moldova, 12

of which are common to both countries. *G. verum* L., also known as „Lady’s bedstraw”, bloom at the summer solstice with golden yellow flowers. Its chemical composition is various and consist of: polyphenols and flavonoids, hydroxycinnamic acids, iridoids, tannins and essential oils [19], for which properties it is considered to be a medicinal plant.

Helichrysum genus (Asteraceae family) also includes approximate 600 species spread mainly in Europe, Asia, North America, in dry forest-steppe areas. According to the determinator, the spontaneous flora of Romania includes 3 species of g. *Helichrysum*: *H. bracteatum*, *H. petiolare* and *H. arenarium* with subspecies *H. ponticum*, found in steppe areas, oak forests, dry, sandy places [20]. On the territory of the Republic of Moldova only one species - *H. arenarium* is found spontaneously. *H. italicum*, a cultivated species, is rich in essential oils: approximately 67 compounds have been identified, which in larger quantities: α -cedrene, α -curcumen, geranyl acetate, limonene, nerol, neryl acetate and α -pinene [21]. Light response curves provide valuable insights into how plants optimize photosynthesis at different light intensities. They show critical thresholds for maximum efficiency and potential points of photoinhibition [22]. Figure 1 shows the water vapor assimilation rate and stomatal conductance of *Galium verum* plants as a function of photosynthetically active radiation (PAR). The graphs show that all plants, regardless of their origin (Romania or Moldova), exhibited high assimilation rates and stomatal conductance, indicating robust photosynthetic activity in both populations. In addition, dark respiration rates were quite high, indicating a high basal metabolic rate in the absence of light. Interestingly, plants from Romania showed a slight increase in all measured photosynthetic parameters, probably due to their summer harvest, when environmental conditions may have optimized photosynthetic capacity.

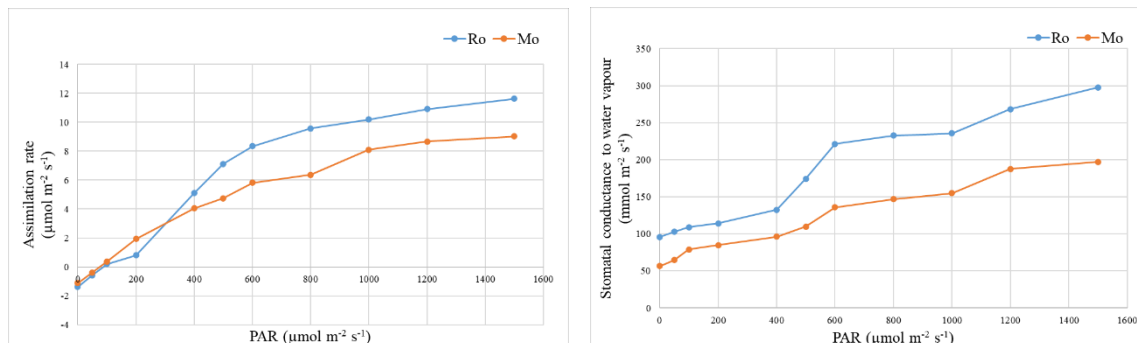


Figure 1. Assimilation rate (a) and stomatal conductance to water vapour (b) function of PAR for *Galium verum* plants.

Furthermore, the light saturation points of plants from Romania and Moldova were relatively consistent, indicating similar light intensity requirements for maximum photosynthetic performance. This consistency in saturation light values suggests that, despite geographic and potentially climatic differences, both groups show comparable adaptation to light conditions and support efficient photosynthetic performance over a range of light intensities.

In plants of *Helichrysum italicum*, the assimilation rate was very low, reflecting the special adaptations of this species (Fig. 2). However, the dark respiration rate was high, consistent with the plant's preference for shaded environments, where higher basal respiration could be beneficial. The light saturation point was also low, indicating that these plants reach maximum photosynthetic efficiency at lower light intensities. Furthermore, the stomatal conductance was low, probably due to the presence of smaller

stomata that could limit gas exchange and reduce water loss. This trait is advantageous in shaded or low-light conditions.

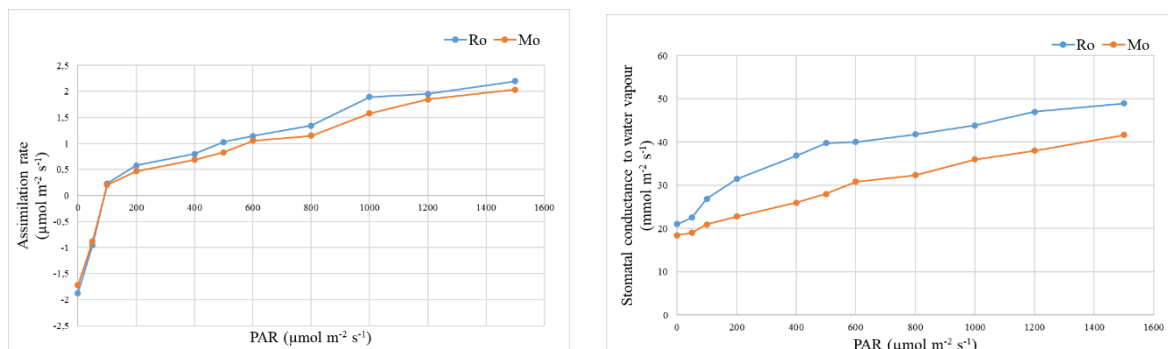


Figure 2. Assimilation rate (a) and stomatal conductance to water vapour (b) function of PAR for *Helichrysum italicum* plants.

CONCLUSIONS.

This research demonstrates the complexity and versatility of photosynthesis as plants respond to different environmental conditions. Through photosynthesis, plants derive oxygen and energy from their carbohydrates and utilize environmental signals, such as light intensity and quality, to alter their growth and storage capacity. The results indicate that, although photosynthesis is primarily dependent on optimal CO₂ diffusion and availability of light, plant species have evolved complex mechanisms to cope with fluctuations in light and temperature, these include structural changes in leaves and chemical responses to stress. The efficiency of photosynthesis and the adaptability of species like *Galium verum* and *Helichrysum italicum* are demonstrated, the physiological and anatomical properties of these species influence their response to light conditions, which can differ depending on the species' origin and environmental exposure.

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