

Ecophysiology of forest species in south-west Romania in the context of climate change

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Abstract

In the context of intensifying climate change, evaluating the ecophysiology of forest species becomes essential for understanding adaptation mechanisms and ecological resilience. This paper investigates the ecophysiological responses of five native forest species – black locust (*Robinia pseudoacacia*), hornbeam (*Carpinus betulus*), field maple (*Acer campestre*), sycamore (*Acer pseudoplatanus*), and ash (*Fraxinus excelsior*) – in two contrasting ecosystems: the "Porțile de Fier" Natural Park and the Pădurea Verde Forest in Timișoara, during the 2021–2024 period. The study was based on seasonal measurements of net photosynthesis and leaf chlorophyll content (SPAD values), correlated with local climate data (temperature, precipitation). The analyses highlighted significant differences between species and sites. Black locust and hornbeam exhibited superior capacity to maintain photosynthetic activity under water stress conditions, while ash and sycamore showed pronounced sensitivity, especially in years with precipitation deficits. In the Pădurea Verde Forest, the impact of thermo-hydric stress was more pronounced, suggesting a combined influence of climatic and anthropogenic pressures. The results support the importance of continuous monitoring of ecophysiological parameters in forest areas, providing relevant data for the implementation of adaptive management strategies aimed at maintaining ecosystem functionality in the face of future climate scenarios.

Keywords: chlorophyll, photosynthesis, climate change, *Robinia pseudoacacia*, *Carpinus betulus*, *Acer campestre*, *Acer pseudoplatanus*, *Fraxinus excelsior*

Introduction

Climate change represents one of the most urgent and multifaceted ecological challenges of the 21st century, exerting profound and far-reaching effects on the functionality, stability, and regenerative capacity of forest ecosystems worldwide [1,2,8]. The intensification of extreme weather events, alterations in precipitation regimes, rising average temperatures, and the increasing frequency of drought periods generate substantial abiotic stress on forest vegetation, directly impacting fundamental physiological processes such as photosynthesis, transpiration, nutrient uptake, and pigment synthesis. These disruptions lead to a decline in primary productivity, alterations in biogeochemical cycles, and a diminished capacity of ecosystems to deliver essential ecological services, including carbon sequestration, microclimate regulation, and soil protection. [4,7,15]

In this context, ecophysiology—an integrative discipline situated at the intersection of plant physiology, functional ecology, and applied climatology—emerges as a critical methodological tool for investigating the adaptive mechanisms and phenotypic plasticity of forest species in response to climate-induced abiotic stress [3,9,14]. By analyzing physiological parameters in correlation with environmental factors, ecophysiology enables the identification of adaptive response patterns, the assessment of stress tolerance, and the anticipation of species behavior under future climatic scenarios. [3,9]

The present study aims to contribute to the advancement of knowledge regarding the ecophysiological responses of native Romanian forest species through a comparative analysis conducted

in two contrasting ecosystems located in the southwestern region of the country: the “Iron Gates” Natural Park, characterized by a high degree of naturalness, floristic diversity, and ecological integrity, representative of sub-Mediterranean forest types; and the “Pădurea Verde” peri-urban forest in Timișoara, subject to intense anthropogenic pressure, habitat fragmentation, atmospheric pollution, and socio-economic interference. The selection of these two sites provides a relevant framework for exploring the influence of ecological context and anthropogenic impact on the physiological performance of forest species, allowing for the analysis of interspecific and intersite variability.[14]

Covering the period 2021–2024, the study is based on the monitoring of key ecophysiological parameters, such as net photosynthetic rate, foliar chlorophyll content (SPAD), stomatal conductance, and water potential, correlated with local climatic data (temperature, relative humidity, solar radiation, and precipitation regime). Through this integrative approach, the research seeks to elucidate how environmental factors and anthropogenic pressures shape the physiological functioning of trees, with the aim of identifying mechanisms of ecological resilience and vulnerability.

The findings of this study are expected to inform the development of adaptive forest management strategies focused on biodiversity conservation, functional restoration of degraded ecosystems, and enhancement of forest resilience to climate change. Furthermore, the research provides scientific foundations for the formulation of public policies in the field of sustainable forestry and promotes an ecologically integrated vision for the management of forest resources in Romania.

In the current global context marked by the intensification of climate change and the growing anthropogenic pressure on natural ecosystems, temperate forests across Europe—including those in Romania—are facing unprecedented ecological challenges. Global climatic shifts, manifested through rising average temperatures, increased frequency and intensity of extreme meteorological events, and altered precipitation regimes, are generating significant abiotic stress on forest vegetation. This stress translates into disruptions of fundamental physiological processes in trees, affecting photosynthesis, transpiration, nutrient uptake, and the synthesis of photosynthetic pigments, with direct implications for tree vitality, regenerative capacity, and the overall resilience of forest ecosystems[4,7,15].

Concurrently, peri-urban forests are subjected to intense anthropogenic stress, driven by accelerated urban expansion, atmospheric pollution, habitat fragmentation, and recreational pressure[5,6,16]. The interaction between abiotic and anthropogenic stressors creates a complex ecological environment in which trees are compelled to activate adaptive mechanisms to survive and maintain functional integrity. Within this framework, ecophysiology emerges as a critical scientific discipline for understanding how forest species respond to climatic variability and environmental disturbances. By analyzing physiological parameters in correlation with ecological factors, ecophysiology provides valuable insights into phenotypic plasticity, stress tolerance, and the adaptive potential of tree species.

The present research aligns with this scientific endeavor by proposing a comparative analysis of the ecophysiological responses of native Romanian forest species, monitored across two distinct ecosystems in the southwestern region of the country. The first study site is the Iron Gates Natural Park, a nationally and internationally significant protected area located at the confluence of continental, Balkan, and sub-Mediterranean climatic influences. The region’s topographic and edaphic diversity supports a wide range of forest vegetation types, composed of species adapted to varied ecological conditions. In recent years, this area has experienced increasing climatic pressure, evidenced by reduced soil water availability, elevated maximum temperatures, and intensified summer precipitation deficits[2,15]. These changes have negatively impacted tree physiological processes, diminishing their regenerative capacity and necessitating a reassessment of forest conservation and management strategies.

The second ecosystem under investigation is the Green Forest, located in the peri-urban zone of Timișoara. This forest plays a vital role in maintaining local microclimatic balance, reducing air pollution, and providing green spaces for the urban population. Its location within an urban-periurban matrix exposes it to a range of anthropogenic stressors, including the urban heat island effect, elevated levels of atmospheric pollutants, habitat fragmentation, and recreational pressure. Additionally, abiotic stressors such as drought, high temperatures, and fluctuating precipitation patterns compound these anthropogenic influences, creating a highly complex ecological setting. Previous studies conducted in this area have revealed significant seasonal variations in physiological processes, particularly in photosynthesis and

chlorophyll content, confirming the relevance of the Green Forest as a model site for analyzing the cumulative effects of environmental stress on forest vegetation[10,14,17].

Against this ecological backdrop, the proposed research aims to advance the understanding of the ecophysiological mechanisms by which forest species respond to combined abiotic and anthropogenic stress. The study spans the period 2021–2024 and is based on the monitoring of key physiological parameters, such as net photosynthetic rate and foliar chlorophyll content (SPAD), correlated with local climatic data (temperature, relative humidity, solar radiation, and precipitation regime). The analysis of these parameters enables the identification of seasonal variations in ecophysiological performance, the comparison of species behavior across the two sites, and the assessment of the influence of climatic and anthropogenic factors on tree physiological functioning.

Through this integrative approach, the research seeks to identify species with high adaptive potential that may be suitable for reforestation, ecological restoration, and biodiversity conservation programs. Correlating ecophysiological parameters with local climatic data will allow for the evaluation of species sensitivity to thermo-hydric stress and the anticipation of their behavior under future climate scenarios. The findings are expected to contribute to the consolidation of fundamental knowledge regarding tree physiological adaptability and to provide scientific support for the development of public policies in the field of sustainable forestry, in alignment with European objectives for ecological transition and climate resilience.

Materials and Methods

The present study was designed as a longitudinal comparative investigation conducted over a four-year period (2021–2024) within two ecologically distinct forest ecosystems located in southwestern Romania. The selection of study sites was based on ecological and climatic criteria, aiming to capture physiological response variability across environments characterized by differing degrees of naturalness and anthropogenic pressure. This design enables the assessment of species-specific ecophysiological performance under contrasting ecological conditions, thereby enhancing the relevance of the findings for adaptive forest management.

Five native forest species were selected for analysis, based on their ecological prevalence within the two ecosystems, their functional significance, and their documented physiological behavior in the scientific literature. The chosen species represent a spectrum of ecological strategies and stress tolerances, as follows:

Robinia pseudoacacia – a pioneer species with high drought tolerance and the capacity for atmospheric nitrogen fixation, often colonizing degraded habitats;

Carpinus betulus – a mesophytic species exhibiting moderate ecological plasticity, commonly found in temperate deciduous forests;

Acer campestre – a highly adaptable species with stable physiological performance in disturbed environments, frequently used in urban and periurban forestry;

Acer pseudoplatanus – a montane species sensitive to hydric stress, with ecological requirements linked to cooler and more humid conditions;

Fraxinus excelsior – a species with elevated ecological demands, vulnerable to drought and pollution, yet ecologically important in riparian and lowland forest systems.

To evaluate ecophysiological performance, two core physiological parameters were selected, both widely recognized as indicators of photosynthetic efficiency and foliar health:

Net photosynthetic rate, expressed in $\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$, measured using a portable gas exchange analysis system under standardized light and temperature conditions;

Foliar chlorophyll content (SPAD index), determined with a portable chlorophyll meter, serving as an indirect indicator of photosynthetic capacity and leaf vitality.

Sampling procedures were carefully standardized to ensure statistical robustness and ecological relevance. Measurements were conducted seasonally—during spring, summer, and autumn—on mature, healthy individuals of each species. For each species and location, a minimum of ten trees were selected, evenly distributed across representative habitat zones while avoiding edge effects and localized disturbances. Climatic data corresponding to each sampling period were obtained from local meteorological stations and included:

Daily mean temperature;

Relative air humidity;

Precipitation totals;

Aridity index, calculated from temperature–precipitation ratios.

Statistical analysis was performed using SPSS software, employing robust techniques to identify significant differences and correlations. To detect interspecific, intersite, and seasonal variations, one-way ANOVA (Analysis of Variance) was applied, supplemented by post hoc tests where appropriate. Relationships between ecophysiological parameters and climatic variables were examined through:[12]

Pearson correlation coefficients, to assess the strength and direction of linear associations;

Multiple linear regression models, to evaluate the combined influence of climatic factors on physiological performance.

This methodological framework provides a rigorous basis for interpreting species-specific responses to abiotic and anthropogenic stressors, contributing to the development of evidence-based strategies for forest conservation, ecological restoration, and climate-resilient silvicultural practices in Romania.

Climatic data corresponding to the study period (2021–2024) were obtained from the meteorological stations in Drobeta-Turnu Severin (representing the Iron Gates Natural Park) and Timișoara (representing the Green Forest). The data include mean annual and seasonal temperature, precipitation, and relative humidity. Table 1 summarizes the annual climatic conditions, while Table 2 presents the seasonal averages. The results indicate that the Iron Gates area is characterized by a sub-Mediterranean influence, with higher thermal amplitudes and lower precipitation, whereas the Green Forest in Timișoara shows slightly higher humidity but is affected by pronounced summer aridity, particularly during 2022 and 2023. These climatic contrasts provide a relevant environmental context for interpreting the ecophysiological differences observed among the studied forest species.

Tabel 1. Mean annual climatic data for the study period (2021–2024)

Year	Location	Mean annual temperature (°C)	Mean summer temperature (°C)
2021	Iron Gates NP	11.2	23.8
2021	Green Forest (Timișoara)	11.8	24.2
2022	Iron Gates NP	11.5	24.9
2022	Green Forest (Timișoara)	12.1	25.6
2023	Iron Gates NP	10.9	23.1
2023	Green Forest (Timișoara)	11.6	24
2024	Iron Gates NP	11.3	24.5
2024	Green Forest (Timișoara)	11.9	25.1

Tabel 2. Seasonal climatic averages (2021–2024) for both sites

Season	Location	Mean temperature (°C)	Precipitation (mm)	Relative humidity (%)
Spring (Mar–May)	Iron Gates NP	14.1	180	72
Spring (Mar–May)	Green Forest	14.6	165	73
Summer (Jun–Aug)	Iron Gates NP	24.1	172	63
Summer (Jun–Aug)	Green Forest	24.8	158	65
Autumn (Sep–Nov)	Iron Gates NP	13.7	160	70
Autumn (Sep–Nov)	Green Forest	14.0	150	72

Season	Location	Mean temperature (°C)	Precipitation (mm)	Relative humidity (%)
Winter (Dec–Feb)	Iron Gates NP	3.2	110	78
Winter (Dec–Feb)	Green Forest	3.8	120	80

Results and Discussion

Net photosynthetic rates exhibited significant variation depending on species, location, and season. Within the Iron Gates Natural Park, *Robinia pseudoacacia* (black locust) and *Carpinus betulus* (hornbeam) maintained elevated photosynthetic activity even during drought periods, indicating a superior adaptive capacity to hydric stress. In contrast, *Fraxinus excelsior* (European ash) and *Acer pseudoplatanus* (sycamore maple) recorded marked declines in photosynthetic performance during years characterized by pronounced precipitation deficits, suggesting a heightened sensitivity to climatic constraints.

The comparative mean values of net photosynthetic rate and chlorophyll content (SPAD) for the analyzed species are presented in Table 3, illustrating the physiological variability between the two study sites and among species.

Table 3. Mean values of net photosynthetic rate ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) and chlorophyll content (SPAD) for five forest species (2021–2024)

Species	Site	Photosynthesis (mean \pm SD)	SPAD (mean \pm SD)
<i>Robinia pseudoacacia</i>	Iron Gates	12.4 \pm 1.2	48.6 \pm 2.3
<i>Carpinus betulus</i>	Iron Gates	11.8 \pm 1.0	47.2 \pm 2.5
<i>Acer campestre</i>	Green Forest	10.9 \pm 1.5	45.3 \pm 2.7
<i>Acer pseudoplatanus</i>	Green Forest	8.6 \pm 1.4	39.8 \pm 2.9
<i>Fraxinus excelsior</i>	Green Forest	7.9 \pm 1.6	37.4 \pm 3.2

Figure 1 illustrates the comparative mean values of net photosynthesis ($\mu\text{mol CO}_2 \text{ m}^{-2} \text{ s}^{-1}$) for the five analyzed species—Ac, Cc, As, Av, and Fe—across two distinct ecological settings: the Iron Gates Natural Park and the periurban Green Forest of Timișoara. Overall, the data reveal that photosynthetic rates recorded in the natural park are consistently higher than those observed in the urban environment, suggesting a positive influence of natural site conditions on physiological functioning. The differences are particularly pronounced for species Cc (*Carpinus betulus*) and Fe (*Fraxinus excelsior*), highlighting their increased sensitivity to anthropogenic stressors.

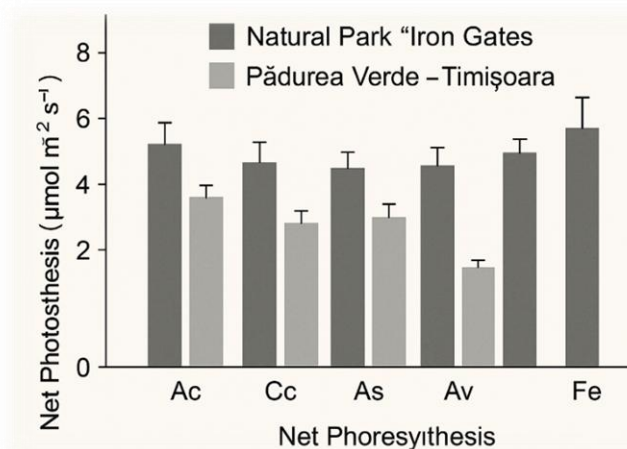


Figure 1. Comparative values of net photosynthetic rates for five forest species between Iron Gates Natural Park and the periurban Green Forest of Timișoara

For clarity in the interpretation of results, the forest species included in the study were coded as follows:

Ac – *Acer campestre* (field maple)

Cc – *Carpinus betulus* (hornbeam)

As – *Acer pseudoplatanus* (sycamore maple)

Av – *Acer platanoides* (Norway maple)

Fe – *Fraxinus excelsior* (European ash)

These abbreviations are consistently used in comparative graphs to facilitate visual representation and ecophysiological analysis of each species within the context of the two studied environments.

In the Green Forest of Timișoara, net photosynthetic values exhibited greater instability, reflecting the cumulative influence of both abiotic and anthropogenic stressors. *Acer campestre* demonstrated relatively high tolerance to thermal stress, maintaining active physiological functions during drought periods. In contrast, *Fraxinus excelsior* was the most adversely affected species, showing significant reductions in photosynthetic parameters.

The analysis of foliar chlorophyll content, expressed through SPAD values, revealed notable differences between the two studied ecosystems. In the Iron Gates Natural Park, SPAD values remained relatively stable throughout the monitoring period, suggesting a superior capacity of trees to maintain foliar pigmentation under environmental stress. This stability reflects the buffering effect of natural site conditions, which mitigate the impact of thermal and hydric fluctuations. In contrast, the Green Forest of Timișoara exhibited more pronounced SPAD variability, particularly during the summer season, indicating increased sensitivity to thermo-hydric stress and atmospheric pollutants commonly associated with urban environments.

Among the five analysed species, *Robinia pseudoacacia* and *Acer campestre* recorded the highest mean SPAD values, confirming their physiological robustness and adaptive potential under fluctuating environmental conditions. Conversely, *Fraxinus excelsior* consistently exhibited the lowest SPAD values, reinforcing its vulnerability to drought and pollution stressors. These findings align with previous literature highlighting the differential pigment retention capacities among species subjected to abiotic and anthropogenic pressures.

Statistical analysis further substantiated the relationship between ecophysiological parameters and climatic variables. Significant correlations were identified, underscoring the influence of environmental factors on physiological functioning:

Maximum air temperatures were negatively correlated with net photosynthetic rate ($r = -0.68$, $p < 0.01$), indicating that elevated thermal conditions inhibit photosynthetic activity, likely due to stomatal closure and enzymatic limitations under heat stress.

Relative humidity showed a positive correlation with chlorophyll content ($r = +0.72$, $p < 0.01$), suggesting that water availability directly supports foliar physiological integrity and pigment synthesis.

Precipitation levels exhibited species-specific effects, with mesophytic species such as *Carpinus betulus* and *Fraxinus excelsior* being more responsive to hydric fluctuations, reflecting their ecological dependence on consistent moisture regimes.

The results obtained highlight significant physiological divergences between the two ecosystems. The natural environment of the Iron Gates Natural Park provides more stable conditions for tree functioning, supporting sustained photosynthetic and pigment-related activity. In contrast, the urban-periurban context of the Green Forest imposes additional stress factors, including elevated temperatures, reduced humidity, and increased pollution, which collectively challenge the physiological resilience of forest species.

Species such as *Robinia pseudoacacia* and *Carpinus betulus* demonstrated enhanced resilience and adaptability, positioning them as suitable candidates for climate-adaptive reforestation initiatives, particularly in degraded or urbanized landscapes [13]. On the other hand, species like *Fraxinus excelsior* and *Acer pseudoplatanus* require targeted conservation measures and continuous physiological monitoring, especially in urban settings where environmental stressors are amplified. These insights contribute to the refinement of species selection criteria for ecological restoration and support the development of site-specific forest management strategies under changing climatic conditions [11,16].

Net photosynthetic rates and foliar chlorophyll content (SPAD) exhibited marked seasonal and interspecific variability, influenced by both climatic conditions and site-specific ecological context. Across

the 2021–2024 monitoring period, photosynthetic activity generally peaked in spring, declined moderately in summer, and reached its lowest values in autumn, with more pronounced fluctuations observed in the peri-urban Green Forest of Timișoara. *Robinia pseudoacacia* maintained relatively stable photosynthetic rates throughout all seasons, including during summer droughts, confirming its high tolerance to thermal and hydric stress. *Carpinus betulus* also demonstrated consistent physiological performance, although a moderate decline was observed during peak summer temperatures. *Acer campestre* exhibited notable seasonal plasticity, with minimal variation in both photosynthesis and SPAD values, suggesting a robust adaptive capacity. In contrast, *Acer pseudoplatanus* and *Fraxinus excelsior* showed significant reductions in photosynthetic activity and pigment content during the summer months, particularly in years with below-average precipitation, indicating a heightened sensitivity to thermo-hydric stress.

Comparative analysis between the two ecosystems revealed that trees in the Iron Gates Natural Park consistently recorded higher mean values of both net photosynthesis and SPAD, regardless of species or season. This trend underscores the buffering effect of natural site conditions, which mitigate the impact of climatic extremes and support physiological stability. In the Green Forest of Timișoara, however, the combined influence of elevated temperatures, reduced humidity, and anthropogenic pressures—such as atmospheric pollution and habitat fragmentation—contributed to greater physiological instability. The differences were particularly evident in species such as *Fraxinus excelsior* and *Carpinus betulus*, which exhibited significantly lower photosynthetic rates and pigment levels in the urban environment compared to the natural park.

The correlation analysis confirmed the strong influence of climatic variables on ecophysiological performance. Maximum air temperatures were negatively correlated with net photosynthesis ($r = -0.68$, $p < 0.01$), suggesting that elevated thermal conditions inhibit photosynthetic processes, likely through stomatal closure and enzymatic limitations. Relative humidity showed a positive correlation with SPAD values ($r = +0.72$, $p < 0.01$), indicating that water availability supports pigment synthesis and foliar integrity. Precipitation levels had species-specific effects, with mesophytic species such as *Carpinus betulus* and *Fraxinus excelsior* being more responsive to hydric fluctuations, while drought-tolerant species like *Robinia pseudoacacia* maintained stable physiological parameters regardless of rainfall variability.

The seasonal dynamics of SPAD values further illustrated species-specific pigment retention strategies. In the Iron Gates Natural Park, SPAD values remained relatively stable across seasons, reflecting the resilience of trees to environmental fluctuations. In contrast, the Green Forest exhibited more pronounced seasonal declines, particularly during summer, highlighting the cumulative impact of abiotic and anthropogenic stressors. *Robinia pseudoacacia* and *Acer campestre* recorded the highest mean SPAD values in both ecosystems, confirming their physiological robustness and suitability for reforestation and ecological restoration initiatives. Conversely, *Fraxinus excelsior* consistently exhibited the lowest SPAD values, reinforcing its vulnerability and the need for targeted conservation measures in urban and peri-urban settings.

Overall, the results emphasize the importance of ecological context in shaping the physiological responses of forest species. Natural ecosystems provide more favourable conditions for maintaining photosynthetic efficiency and pigment stability, while urban environments impose complex stress regimes that challenge tree vitality. The identification of species with high adaptive potential—such as *Robinia pseudoacacia* and *Acer campestre*—offers valuable guidance for the selection of planting material in climate-resilient forestry programs. At the same time, the observed sensitivity of species like *Fraxinus excelsior* and *Acer pseudoplatanus* underscores the necessity of continuous monitoring and adaptive management, particularly in areas exposed to intensified climatic and anthropogenic pressures. These findings contribute to a more nuanced understanding of forest species' ecophysiological behaviour and support the development of evidence-based strategies for sustainable forest management in the context of climate change.

Conclusions

Climate change, through its increasingly pronounced and unpredictable manifestations, profoundly influences the ecophysiology of forest species by disrupting the essential physiological processes that underpin tree functioning and survival. Processes such as photosynthesis, transpiration, nutrient uptake, and the synthesis of photosynthetic pigments are directly affected by thermal fluctuations, hydric deficits,

and alterations in precipitation regimes. These disruptions lead to a decline in tree vitality, a reduction in regenerative capacity, and, consequently, a weakening of forest ecosystem stability. Importantly, these effects are not uniform across species but vary according to each species' ecological traits, physiological plasticity, and the environmental context in which they develop.

The forest species analysed in this study exhibit distinct ecophysiological responses depending on the ecosystem and the cumulative stress generated by the interaction between abiotic and anthropogenic factors. In natural ecosystems, where environmental conditions are more stable and less disturbed, trees demonstrate a greater ability to maintain physiological functions. In contrast, periurban environments—characterized by habitat fragmentation, atmospheric pollution, and heightened climatic variability—are associated with increased sensitivity and more pronounced physiological instability. These differences underscore the critical role of ecological context in shaping species' adaptive responses and highlight the need for differentiated approaches in forest management.

In this regard, ecophysiological studies prove to be indispensable tools for informing sustainable forest management strategies. By monitoring physiological parameters in correlation with climatic and ecological variables, such research enables the identification of species with high adaptive potential, the assessment of ecological risks, and the development of appropriate conservation and restoration measures. Integrating ecophysiological findings into silvicultural planning contributes to enhancing the resilience of forest ecosystems and supports their adaptation to changing climatic conditions, in alignment with European policy objectives concerning ecological transition and biodiversity protection.

Moreover, periurban forests must be explicitly recognized and integrated into ecological urban planning, given their crucial role in maintaining ecosystem health, regulating urban microclimates, and providing essential ecosystem services to local populations. These forests serve as true buffer zones between urban and natural environments, helping to mitigate the urban heat island effect, reduce pollution, and improve overall quality of life. Therefore, their protection and responsible management should become a strategic priority within sustainable urban development frameworks, involving active participation from policymakers, local communities, and the scientific research sector.

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