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Drought or Severe Drought? Hemiparasitic Yellow Mistletoe (*Loranthus europaeus*) Amplifies Drought Stress in Sessile Oak Trees (*Quercus petraea*) by Altering Water Status and Physiological Responses

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Abstract: European oak species have long been considered relatively resistant to different disturbances, including drought. However, several recent studies have reported their decline initiated by complex changes. Therefore, we compared mature sessile oak trees (*Quercus petraea* (Matt.), Liebl.) infested versus non-infested by hemiparasitic yellow mistletoe (*Loranthus europaeus Jacq.*) during the relatively dry vegetation season of 2019. We used broad arrays of ecophysiological (maximal assimilation rate A_{sat}, chlorophyll *a* fluorescence, stomatal conductance g_S, leaf morphological traits, mineral nutrition), growth (tree diameter, height, stem increment), and water status indicators (leaf water potential Ψ, leaf transpiration T, water-use efficiency WUE) to identify processes underlying vast oak decline. The presence of mistletoe significantly reduced the Ψ by 1 MPa, and the WUE by 14%. The T and g_S of infested oaks were lower by 34% and 38%, respectively, compared to the non-infested oaks, whereas the A_{sat} dropped to 55%. Less pronounced but significant changes were also observed at the level of photosystem II (PSII) photochemistry. Moreover, we identified the differences in C content, which probably reduced stem increment and leaf size of the infested trees. Generally, we can conclude that mistletoe could be a serious threat that jeopardizes the water status and growth of oak stands.

Keywords: hemiparasite; oak stands dieback; water deficit; mineral nutrition; photosynthesis; growth response

1. Introduction

European oak species are dominant species in temperate European hardwood forests [1] and among the main components of the European forest economy [2]. These woody plants are, therefore, of great environmental, economic and cultural importance [3]. There is high genetic, physiological and morphological differentiation within the genus, but European oaks may be generally considered as one of the most drought-tolerant tree species in comparison with other deciduous trees of European forests [3–5]. With average air temperatures significantly rising and significant changes in precipitation regime in Europe over the last several decades [6], conditions at higher altitudes have become more suitable for these species [7]. However, they have been more often negatively affected and reports about the decline of oak stands suggest that multifactorial processes are responsible for the increased damage [8]. Inappropriate oak forest management [9], cold winters [10] and soil nutrient imbalance [11] may lead to a strong decrease in oak resistance potential. The interaction of various factors occurring on the background of climate changes leads to the more frequent extreme weather events, namely, heat stress, and a lack of precipitation during the beginning of the vegetation season and during the summer, which may affect the growth and survival of trees [12,13]. There is evidence of frequent crown and stand declines, especially at dry sites [14]. Weakened oak forests are more affected by biotic factors which may include infection by pathogenic fungi and microorganisms [15–17], and insect attacks [8,10,18]. Most of the aforementioned factors that negatively affect the vitality of trees can be amplified by an often neglected biotic agent with an increasing significance: hemiparasitic plants [19–21]. Hemiparasitic plants and their hosts form integrated systems that are stable for many years [22]. They occur more often in less dense stands [23] and even though they can assimilate CO_2, their exclusive source of water and minerals is the host plants. Thus, hemiparasites need to keep their stomata open whenever possible to sustain the stream of transpiration and the amount of water lost by hemiparasitic plants may be enormous [24]. By withdrawing water from the host trees even during drought periods, hemiparasites cause the drought effects to become more severe [23,25]. The cumulative effect of these factors leads to a more pronounced decrease in the water potential [26], stomatal conductivity and assimilation rate [23,27,28], which alter the nutrient and carbon status of host trees compared to non-infested trees growing in the same conditions [29]. Moreover, transpiration-mediated cooling of host trees is restricted, and heat stress may also impact light-dependent photosynthetic reactions. The probability of yellow mistletoe spreading is higher with rising temperatures [30], resulting in a feedback loop.

We focused on examining how yellow mistletoe (*Loranthus europaeus*) impacts the physiology of sessile oak trees (*Q. petraea* s.l.). This hemiparasitic plant is widespread in Europe, and its northern distribution border crosses Slovakia [31]. It grows mainly in oak tree crowns and occurs less frequently on stems at the places where the outer bark is cracked, and it mainly infests oaks that are more than 60 years old [19].

Lower branches of infested trees become more massive resulting in changes of overall tree shape. The accelerated growth of lower branches is a rebalancing effort of the tree to maintain the physiological performance as the upper part of the crown is more severely affected by the presence of yellow mistletoes. However, this defence mechanism is effective only when the infection is not spread and has a small extent [32]. Currently, a high population density of mistletoe can be observed and, therefore, the defence mechanism of oak trees is often ineffective. Severely infested trees respond by a premature senescence of the crown, which may eventually result in the death of the whole individual [32,33].

As results from the aforementioned, yellow mistletoe negatively affects the physiological status of host trees in many ways [23,25,26,34]. The goal of this study was to analyse a broad array of ecophysiological properties and processes in oak trees infested by yellow mistletoe during the drought period. We compared indicators of water status, photosynthetic performance, drought-induced stress nutrient status, growth and leaf traits on the infested, and non-infested mature oak trees and yellow mistletoe. We hypothesize that (i) the water potential of infested trees is significantly lower than that of non-infested trees and even lower for mistletoe, seriously influencing photosynthesis. The drought enhanced by the presence of yellow mistletoe leads to drops in both the light-dependent and light-independent stages of photosynthesis. We expect the lowest water-use efficiency (WUE) for yellow mistletoe and the highest for non-infested trees. Another hypothesis is that (ii) infested trees have significantly lower content of minerals due to losses to mistletoe and due to restricted upward transpiration stream, limiting the growth of leaves and the stem increment. Finally, we expect that (iii) higher and thicker individuals are more infested than those of lower diameters.

2. Materials and Methods

2.1. Study Site and Design Description

The research was performed in Slovakia, in the central part of the Považský Inovec Mts. (48°40′02″ N; 18°04′09″ E, 360–400 m a.s.l.). The site is located on a southwest-oriented slope with an inclination of 10–30°. The soils are acid cambisol podzol. The area belongs to a warm, dry region, hilly land and highlands.

For the research, a stand with an age of 95 years consisting exclusively of sessile oak trees (*Q. petraea* (Matt.) Liebl s.l.), was chosen. The stand belongs to managed forests with an area of 12.95 ha, stand density of 0.8 (8) and full canopy. Trees have originated from generative regeneration of 100% sessile oak seeds. The standing volume per hectare is 264 m^3 and the total standing volume is represented by 3419 m^3.

The long-term climatic data (1961–1990) for the study area were taken from the nearest meteorological station Topoľčany. The air temperature and precipitation during 2019 were measured on an open field close to the study site using our own meteorological station with Minikin Tie and Eri sensors (EMS, Brno, Czech Republic) and a built-in datalogger. The annual mean air temperature was higher in 2019 compared to the long-term mean (10.4 °C and 9.4 °C, respectively) and the average air temperature during the vegetation period (from April to September) was higher in 2019 (16.6 °C and 15.9 °C, respectively). The long-term mean annual rainfall was 549 mm, while the rainfall in 2019 was 782 mm. The amount of precipitation during the vegetation season in 2019 was 409 mm and the long-term mean was 326 mm. Even though the total precipitation was quite high compared to the long-term averages, the distribution was uneven (Figure 1) and only 23 mm fell during the 30-day period prior to the physiological measurements.

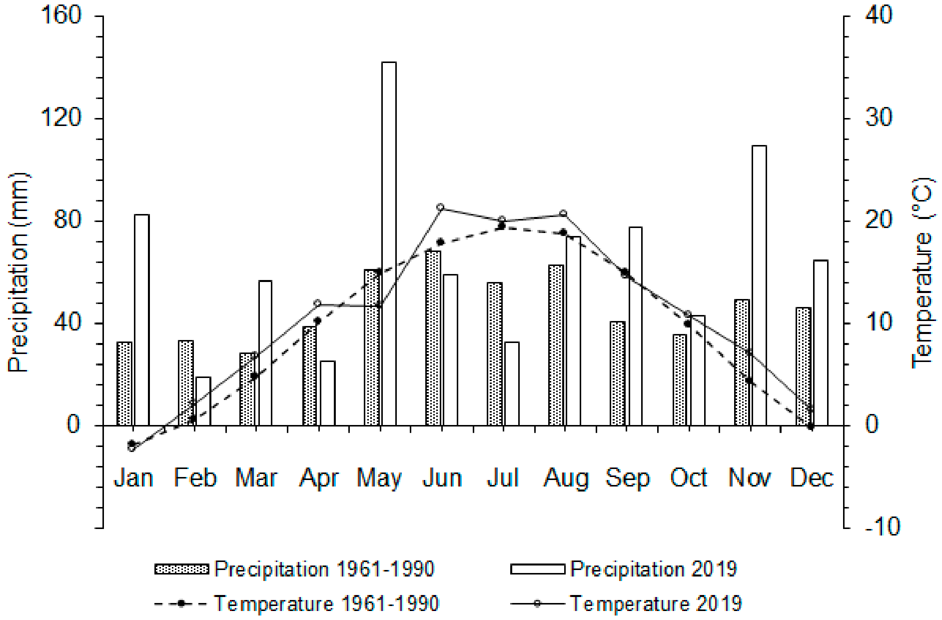

Figure 1. Precipitation and mean air temperatures for years 1961–1990 and 2019.

We identified 100 adult dominant oak trees with canopies comparably exposed to sun radiation (excluding extreme phenological forms): 50 individuals of non-infested oaks and 50 individuals infested by the hemiparasitic yellow mistletoe (*Loranthus europaeus Jacq.*), which were comparable for both, the height and diameter. All chosen trees were situated inside the stand and did not show any other visible damage, except the presence of yellow mistletoe. The diameter at breast height (DBH), the tree height, and the height of the first branch were assessed for each of 100 oak trees. The heights were measured using the ultrasonic hypsometer for the measuring of tree height and distance—Vertex IV (Haglöf, Långsele, Sweden) at late autumn of 2018. The DBH was measured in early spring and late autumn (before and after leaf unfolding) of 2018 using the steel manual band dendrometers with an accuracy of 1 mm, which were mounted at a height of 1.3 m. The difference between the two measurements represents the stem increment per year. Moreover, we visually checked each infested oak tree using binoculars, and mistletoes were counted during the winter of 2018/2019. The mean number of mistletoes per infested tree was 8.30. A total of 67% of the infested oak trees had fewer than 10 mistletoes. The maximum number of mistletoes per oak was 19, while the smallest number of mistletoes per infested oak tree was 2 individuals.

For further physiological measurements, the subset of 14 infested and 14 non-infested trees were selected, whereas we chose infested trees with between 8 and 11 mistletoes. The sampling of leaves and the in situ measurements were carried out during two sequential days on 6 and 7 August 2019 from 8:30 to 12:00 and from 15:00 to 18:00. The 2–4 m long branches from the upper, unshaded part of the crown of selected trees were cut in turns during the day by professional climbers. Immediately after cutting, the branches were put into water and measured to prevent the rapid desiccation of tissues.

The more detailed information about the individual measurements and their sample sizes are displayed in Table 1.

Table 1. The description of sample sizes for conducted measurements.

Sampling and Measurements	Oak Trees Per Variant/Repetitions	Yellow Mistletoes/Repetitions
Diameter at breast height	50/1	-
Height of tree	50/1	-
Height of the first branch	50/1	-
Stem increment	50/1	-
Leaf water potential	13/2–4	10/1
Gas exchange	14/8	14/6
Chlorophyll a florescence	14/5	-
Content of nutrients in leaves	14/1 (ca. 30 leaves)	14/1 (ca. 100 leaves)
Leaf morphological traits	14/1 (ca. 20 leaves)	14/1 (ca. 100 leaves)

2.2. *Leaf Water Potential*

The water potential (Ψ) was assessed for a total of 10 to 13 individuals per group. A Scholander-type pressure chamber SAPS II (Soil Moisture Equipment Corp., Goleta, CA, USA) was used and 2–4 repetitions for each tree were measured. Just one value was assessed for the mistletoe, as the variability found for one individual was low and measurements were more demanding regarding the pressure in the portable gas bottle.

2.3. *Gas Exchange Measurements*

Parameters related to photosynthesis and water-use efficiency were compared among infested trees, non-infested trees and yellow mistletoes. Eight and six leaves were recorded per individual oak and mistletoe, respectively. Measurements were carried out using an Li-6400XT gas exchange system connected to a standard 6 cm^2 chamber fitted with a 6400-02B light-emitting diode (LED) light source (LI-COR Biosciences, Lincoln, NE, USA). Inside the chamber, the reference CO_2 concentration and photosynthetically active radiation were maintained at 400 $\mu mol\ mol^{-1}$ and 1400 μmol photons $m^{-2}\ s^{-1}$, respectively. The air temperature inside the chamber was set to 23 °C and the relative humidity of the air was approximately 70%.

Values of the CO_2 assimilation rate (A_{sat}), transpiration rate (T), intercellular concentration of CO_2 (Ci) and stomatal conductance to water vapour (g_S) were recorded after the adaptation of leaves inside the chamber, when the values of CO_2 assimilation rate remained stable (1–2 min). The intrinsic water use efficiency WUEi was calculated as the ratio of the CO_2 fixation rate to the stomatal conductance (A_{sat}/g_S).

2.4. Chlorophyll a Fluorescence Measurements

Chlorophyll *a* fluorescence was assessed using a PAM-2500 fluorimeter (Walz, Effeltrich, Germany). Five leaves per oak tree (infested and non-infested) were measured. The leaves were kept in darkness for 30 min and then illuminated by a saturation pulse with an intensity of approximately 6580 µmol m^{-2} s^{-1} for a duration of 1 s to assess the minimum fluorescence of dark-adapted leaves (Fo) and the maximal fluorescence of dark-adapted leaves (Fm). Then, the leaves were illuminated with an actinic light of an intensity 350 µmol m^{-2} s^{-1} for 1.5 min to obtain the steady-state fluorescence (Fs). The second saturation pulse was used to measure the maximal fluorescence of light-adapted leaves (F′m). The maximal efficiency of the photosystem II (PSII) photochemistry (Fv/Fm), the actual efficiency of the PSII photochemistry (Φ_{PSII}), the electron transport rate (ETR) and the non-photochemical quenching (NPQ) were calculated as follows [35]:

$$\frac{Fv}{Fm} = \frac{Fm - Fo}{Fm} \tag{1}$$

$$\Phi_{PSII} = \frac{F'm - Fs}{F'm} \tag{2}$$

$$ETR = \Phi_{PSII} \times PAR \times 0.84 \times 0.5 \tag{3}$$

$$NPQ = \frac{Fm}{F'm} - 1 \tag{4}$$

2.5. Content of Nutrients in Plants and Soil

Biomass samples consisting of approximately 30 leaves per tree (infested and non-infested oaks) and 100 leaves per mistletoe individual were collected. Subsequently, the leaves were dried for 48 h in a drying oven at a temperature of 105 °C. The dry matter was then milled into a powder in a Fritsch Planetary Micro Mill (Fritsch, Markt Einersheim, Germany). The total nitrogen and sulphur content were determined with a FLASH 1112 Nitrogen, Carbon and Sulphur Analyser (Thermo Fisher Scientific Inc, Hanau, Germany). The Ca, Mg, K, and P contents were determined after mineralization of the samples in concentrated HNO_3 using microwave decomposition (UniClever type, Plazmatronika, Wrocław, Poland). The content of P was measured with an atomic emission spectrometer (AES-ICP, type LECO ICP-3000, LECO, St. Joseph, MI, USA), while the Ca, Mg and K contents were analysed using a SensAA atomic absorption spectrometer (GBC, Dandenong, Victoria, Australia).

The biological absorption coefficient (BAC) defined in 1969 by Kovalevsky [36] was calculated as the ratio between the nutrient content in the plants and the same nutrient in the soil. We utilized this relationship to determine the ability of the species to accumulate nutrients from a soil subsystem into the plant biomass or from the host plant into the parasitic plant during the growing season. Within the research plot, the infested and non-infested trees were randomly integrated, and therefore the calculation of BAC was based on the same soil samples. Sampling of the soil was carried out in August 2019. Ten samples of mineral topsoil (5–10 cm deep) were taken along two line transects (contour and fall line with lengths of 10 and 15 m, respectively) at regular distances of 5 m. These 10 samples from research plot were mixed and analysed. The collected soil sample was dried at 105 °C to a constant weight. Subsequently, the samples were milled into a fine dust using the Planetary Micro Mill. We determined the total N and S content with the FLASH 1112 CNS analyser. Available forms of the other macronutrients (Ca, Mg, K, P) were extracted according to Mehlich II, and then the samples were analysed using an atomic emission spectrometer.

2.6. Leaf Morphological Traits

Leaves from non-infested trees, infested trees, and yellow mistletoe plants were sampled and transported to the laboratory to measure of the mean leaf size (area, cm^2), using ImageJ 1.51 k software (National Institute of Health, Bethesda, MD, USA), and the leaf mass area (LMA, $g\ cm^{-2}$). LMA was calculated as the ratio between dry weight and area of leaves. We sampled approximately 20 leaves per oak tree and 100 leaves per mistletoe.

2.7. Statistical Analysis

Hierarchical analysis of variance was performed to reveal the differences among the studied parameters whenever more repetitions for one individual were measured (more leaves/twigs per oak tree/yellow mistletoe). This was used for the measurements of leaf water potential, gas exchange parameters, and chlorophyll *a* fluorescence parameters (Table 1). The analysis was performed in R (R Core Team 2017) using the lme function from the nlme package. For the measurements, where just one repetition per individual oak tree/yellow mistletoe was conducted (e.g., tree height, DBH, LMA, content of nutrient; Table 1), a one-way analysis of variance was used. Then, the Tukey tests (with significance level $\alpha = 0.05$) were performed for multiple comparisons using the glht function from the multcomp package.

Moreover, relationships between the number of mistletoes and the growth parameters of the infested trees were tested using the linear regression models.

3. Results

3.1. Leaf Water Potential and Photosynthesis

The water potential values were negative, with the lowest value in the yellow mistletoe, higher values in the infested sessile oaks and the highest value in the non-infested sessile oaks. The steps between respective variants were nearly 1 MPa, and the differences among all groups were significant (Figure 2a). Strikingly low variability was found for mistletoe.

There were no significant differences in the assimilation rate between the infested trees and the mistletoes (Figure 2b), but these two groups differed statistically from the best-performing non-infested trees. The average value of the assimilation rate for the non-infested trees was almost twice as high as that of the infested trees or mistletoes.

The yellow mistletoe showed the highest average values of transpiration rate and stomatal conductance which were 3.3 and 4 times higher than values of the infested oaks and 2.2 and 2.5 times higher than the values of the non-infested trees (Figure 2c,d). Even though the differences between the infested and non-infested trees were not as pronounced for the assimilation rate, they were statistically significant between all three groups for both, T and g_S. Despite the lower stomatal conductance in the infested trees, their intercellular concentration of CO_2 was significantly higher than that of the non-infested trees (Figure 2e). The Ci values of the yellow mistletoe was close to the ambient concentration of CO_2, and again, the variability was extremely low.

The differences in the intrinsic water-use efficiency were significant between all three groups. The lowest WUEi value was recorded for mistletoe and the highest value was recorded for the non-infested trees (19.2 and 84.5 $\mu mol\ mol^{-1}$ respectively). The WUEi values of the non-infested trees was roughly 4 times higher than that of the mistletoe (Figure 2f). The differences between the groups of oaks were significant, with higher WUEi values confirmed for the non-infested trees, in particular due to much higher assimilation rates.

Based on the maximal (Fv/Fm) and the actual photochemical efficiency of PSII (Φ_{PSII}), we did not confirm significant damage to the PSII photochemistry induced by the activity of yellow mistletoe (Figure 3a,c). However, the non-infested sessile oak trees showed a slightly (but significantly) higher efficiency of the electron transport rate (Figure 3b). The dissipation of excess energy as heat (non-photochemical quenching, Figure 3d) differed greatly between the groups of trees, with average values of 1.03 and 1.31 for non-infested and infested oaks, respectively.

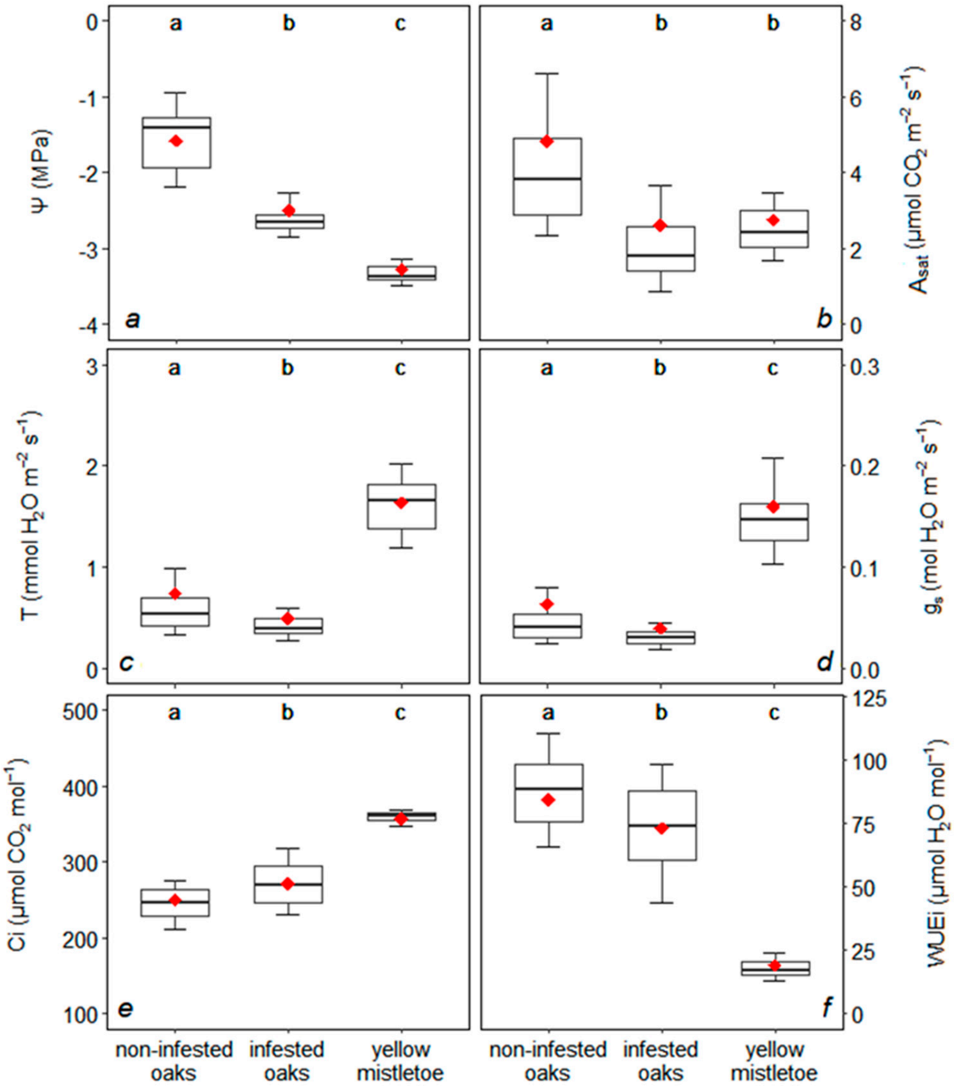

Figure 2. Water potential (Ψ, (**a**)), assimilation rate (A_{sat}, (**b**)), transpiration rate (T, (**c**)) stomatal conductivity (g_S, (**d**)), intercellular concentration of CO_2 (Ci, (**e**)) and intrinsic water-use efficiency (WUEi, (**f**)) of non-infested oaks, infested oaks and yellow mistletoes. The median (black line), 25% quantile (box; 37.5–62.5%), 50% quantile (whiskers; 25–75%) and mean (red diamond) are displayed. Lowercase letters represent statistical significance at the level of $p = 0.05$.

3.2. Content of Nutrients in Plants and Their Absorption

By comparing mistletoe and both groups of sessile oak trees we found that content of Ca, Mg, K and P in the leaves of mistletoe were significantly higher than those of both groups of oaks (Figure 4a–f). In contrast, the contents of C and N were significantly lower in the leaves of mistletoe. Most importantly, the only significant difference between the two groups of oaks was confirmed for C. Non-infested trees had a lower concentration of C in their leaves compare to the infested trees (479.2 and 498.4 g kg^{-1}, respectively).

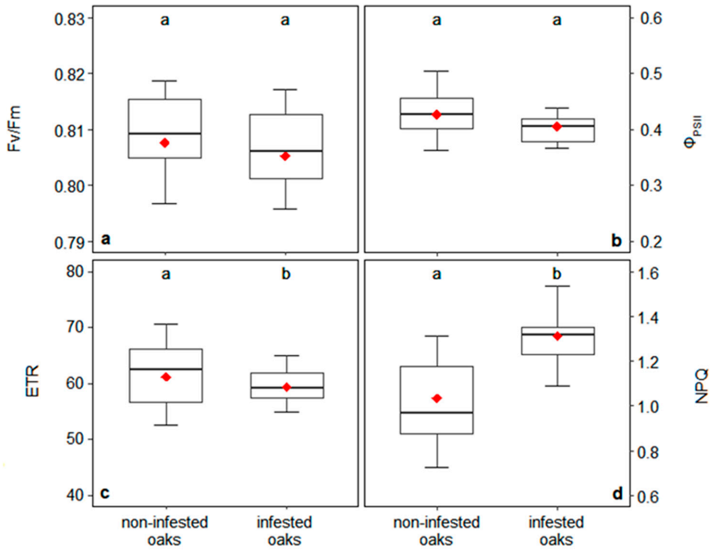

Figure 3. The maximal efficiency of the PSII photochemistry (Fv/Fm, (**a**)), actual efficiency of the PSII photochemistry (ΦPSII, (**b**)), electron transport rate (ETR, (**c**)) and non-photochemical quenching (NPQ, (**d**)) of non-infested oaks and infested oaks. The median (black line), 25% quantile (box; 37.5–62.5%), 50% quantile (whiskers; 25–75%) and mean (red diamond) are displayed. Lowercase letters represent statistical significance at the level of p = 0.05.

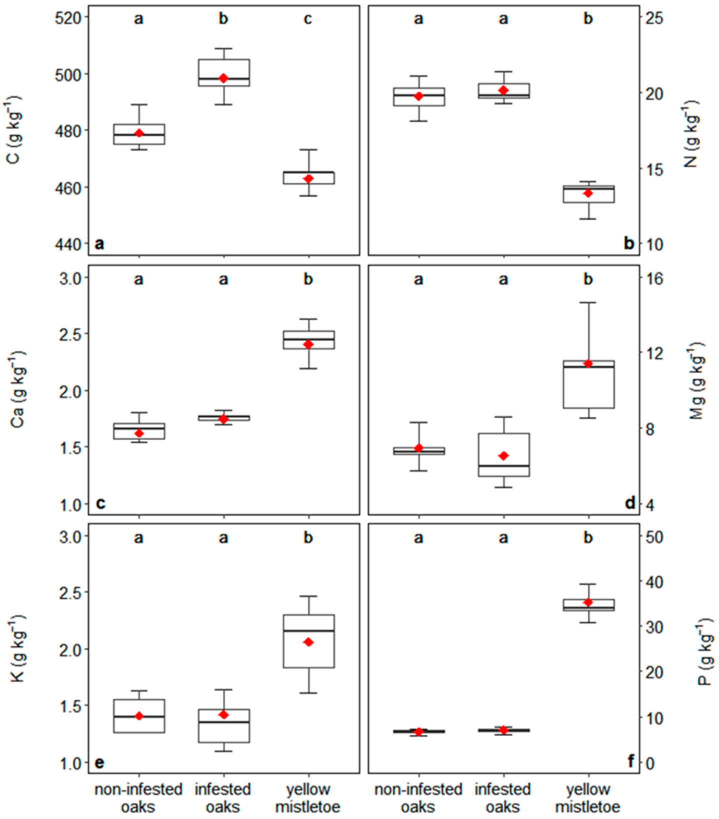

Figure 4. Contents of carbon (**a**), nitrogen (**b**), calcium (**c**), magnesium (**d**), potassium (**e**) and phosphorus (**f**) of non-infested oaks, infested oaks and yellow mistletoes. The median (black line), 25% quantile (box; 37.5–62.5%), 50% quantile (whiskers; 25–75%) and mean (red diamond) are displayed. Lowercase letters represent statistical significance at the level of p = 0.05.

Analyses of the mineral topsoil (5–10 cm) at the study site showed a low base saturation. On the poorest solid rocks we detected acid cambic podzol (pH 4.8). The content of C showed the highest value (22.1 g kg^{-1}) among all nutrient. On the other hand, the contents of other nutrients (N, Ca, Mg, K and P) did not exceed 1.2 g kg^{-1}. Biogeochemical flows of selected nutrients between the soil and the oaks were analysed using the biological absorption coefficient (BAC, Table 2).

Table 2. Biological absorption coefficient of nutrient (in direction from soil to oaks and from oaks to mistletoe). Mean ± standard deviations are displayed. Superscript letters (a,b) represent the statistical significance at the level of $p = 0.05$.

	Carbon	**Nitrogen**	**Calcium**	**Magnesium**	**Potassium**	**Phosphorus**
Non-infested tree	21.7 ± 0.92 ^{b}	17.3 ± 2.31 ^{a}	12.3 ± 1.96 ^{a}	190.9 ± 37.11 ^{a}	18.4 ± 4.77 ^{a}	55.7 ± 9.57 ^{a}
Infested tree	22.6 ± 0.60 ^{a}	17.7 ± 1.77 ^{a}	13.2 ± 1.28 ^{a}	179.8 ± 69.83 ^{a}	18.5 ± 5.46 ^{a}	59.2 ± 12.71 ^{a}
Mistletoe	0.9 ± 0.02	0.7 ± 0.13	1.4 ± 0.23	1.9 ± 0.58	1.5 ± 0.27	5.2 ± 1.44

We confirmed that there were no significant differences for nutrient accumulation between groups of trees (except C). The nutrients with the highest accumulation were Mg and P, while Ca had the lowest accumulation in the leaves of trees. The accumulation of Mg was 14 times higher than the accumulation of Ca for infested oak trees and almost 16 times higher for the non-infested oak trees. We found the following BACs for the nutrients in the leaves of the tested trees: Mg > P > C > K > N > Ca.

Hemiparasitic yellow mistletoe gains nutrients exclusively from its host (oak). For this reason, we calculated the BAC of the leaf nutrients ratio (mistletoe/infested oak tree). The highest values of the BAC among all the nutrients were detected for P (5.16 ± 1.44), while the lowest values were found for N (0.66 ± 0.13). By comparing the ability of mistletoes to accumulate nutrients with that of oaks, we found much lower values (Table 1) and a markedly different order: P > Mg > K > Ca > C > N. We assume that the values of the coefficients reflect the bioaccumulation potential of mistletoe and the oak–mistletoe interaction.

3.3. Growth and Rate of Infestation

We confirmed marginally significant differences in leaf area between infested and non-infested oaks. Infested oaks had significantly smaller and slightly (but not significantly) thicker leaves (Figure 5a,b).

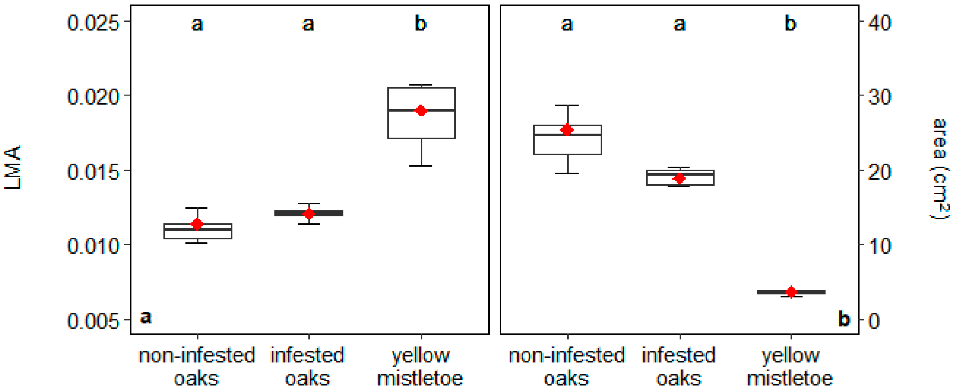

Figure 5. Leaf mass area (**a**) and leaf area (**b**) of non-infested oaks, infested oaks and yellow mistletoes. The median (black line), 25% quantile (box; 37.5–62.5%), 50% quantile (whiskers; 25–75%) and mean (red diamond) are displayed. Lowercase letters represent statistical significance at the level of $p = 0.05$.

From the point of view of tree growth, we found significant differences in all studied traits, except diameter at the breast height (Table 3). The mean height of the infested sessile oak trees was 2 m lower compared to non-infested trees with the lower-lying crown as well. Although, the differences in DBH were not significant, analysis of the average stem increment per year showed that the stem increment of the infested oaks was approximately 34% lower than of non-infested trees.

Table 3. Growth traits of non-infested and infested oak trees. Mean ± standard deviations are displayed. Superscript letters (a,b) represent the statistical significance at the level of $p = 0.05$.

	DBH (cm)	**Height (m)**	**Height of the First Branch (m)**	**Stem Increment (mm)**
Non-infested trees	36.6 ± 3.5^{a}	25.2 ± 1.7^{a}	9.1 ± 3.3^{a}	1.75 ± 1.4^{a}
Infested trees	39.2 ± 5.1^{a}	23.2 ± 1.9^{b}	6.7 ± 2.6^{b}	2.38 ± 1.7^{b}

Moreover, we calculated the correlation coefficient between the height of oak trees and the frequency of mistletoe in the infested oaks and found no significant relationship. On the other hand, we found a significant positive relationship between the diameter at breast height of the infested oak trees and the frequency of mistletoe ($r^2 = 0.234$, Figure 6a,b).

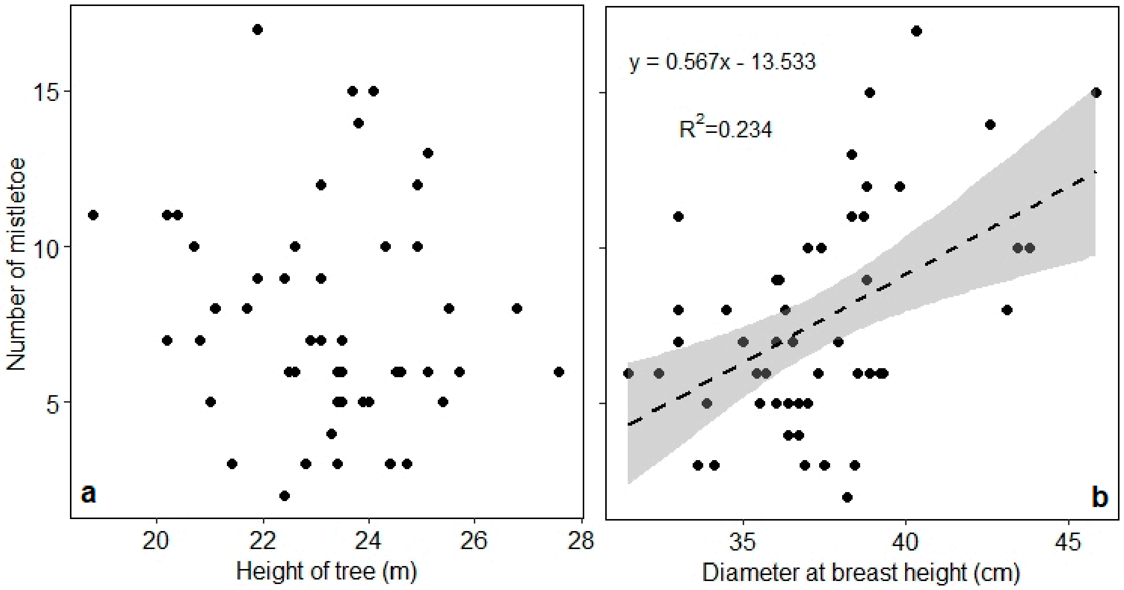

Figure 6. Relationship between the number of mistletoes and the tree height (**a**) and the tree diameter at breast height (**b**). For statistically significant relationships (**b**), the regression line (dashed line) and the 95% confidence interval (grey area) are displayed.

4. Discussion

4.1. Impact of Mistletoe on Photosynthesis and Water Relationships

Our results showed that yellow mistletoe infestation alters the water status of its host plant. Infested trees showed a more negative leaf water potential Ψ (−2.5 MPa) than did non-infected trees (−1.58 MPa). This result was expected because part of the water is drained by mistletoe, amplifying the drought stress of the host trees in our experiment. Moreover, plants try to avoid water shortage and wilting caused by drought and hemiparasite by lowering the osmotic potential via production of osmoprotective compounds, in addition to closing their stomata [24,37]. There is a close relationship between the severity of drought stress and the leaf water potential. Breda et al. [4] reported a predawn leaf water potential of −2 MPa and midday leaf water potential of −3.3 MPa as a threshold values of *Q. petraea* when the number of embolised vessels significantly increase. Cochard et al. [38] estimated the threshold value for midday water potential of −3 MPa in a 30-year-old *Q. petraea* stand. While the non-infested trees in our study were, during the observations (from 8:30 to 12:00 and from 15:00 to

18:00), in the "safe zone", the infested ones were threatened in the long term by water shortage. The high water demands of mistletoe are guaranteed by a greater negative leaf water potential (−3.5 MPa), which causes the hydrostatic pressure in the cells between the host and the hemiparasite to favour the hemiparasite and the xylem sap to flow from the host to the mistletoe [39]. The average differences in Ψ during observation hours between the infested oaks and the mistletoes was 0.75 MPa, which is a smaller value than the published values for yellow mistletoe parasitizing *Q. robur* (from −1.0 to −1.5 MPa, [40]). Davidson et al. [41] pointed out that the Ψ difference between a host and mistletoe depends on the air temperature and vapour pressure deficit (VPD). Higher air temperatures and VPDs lead to larger differences. During our experiment, the VPD ranged from 1.54 to 2.48 kPa, and the air temperature ranged from 22.0 to 26.0 °C indicating rather mild conditions. However, drought was present, and the rainless period lasted for nearly one month prior to the experiment; therefore, we assume that under more severe climate conditions (warmer and drier), the difference in Ψ between a hemiparasite and its host might be higher.

As European oaks are considered as an anisohydric species in general, which are characterized by relatively low stomatal control and under drought conditions they try to retain the CO_2 assimilation rate at the costs of water loses [42,43], we expected that the presence of yellow mistletoe would enhance the anisohydric behaviour of the oak–mistletoe system. However, based on our results as we observed the decrease in stomatal conductance and transpiration rate, we now suppose that behaviour of infested oak trees is more isohydric compared to the non-infested trees. Transpiration in the infested trees was 66% of that in the non-infested ones. Reduced transpiration of the infested trees was connected to an increased stomatal closure. In our case, the closing of stomata is a water-saving tool that prevent hydraulic failure in host trees suffering from the presence of hemiparasites. The activity of mistletoe leads to a reduced water supply in the host tree, a lower transpiration rate due to preventive stomatal closure and an eventual reduction in the cooling ability at high air temperatures [44]. Unlike host trees, mistletoe controls its own transpiration rate at very low levels and keeps stomata open allowing it to transpire large amounts of water [26]. However, several studies have described the coupled regulation of mistletoe stomata based on the host tree water status [28,40,45], light and air humidity [46] to some extent. In our study, we did not observe any of these regulatory mechanisms. Ullmann et al. [45] reported that the ratio between the transpiration rate of most mistletoe species and their hosts usually ranges between 1.5 and 7.9, but only a few studies have dealt with yellow mistletoe and *Quercus* sp. relationships. The ratio of transpiration between the mistletoe and its host in our study was 3.3, which is in good accordance with the findings published by Schultze et al. [40] and [28]. Urban et al. [47] reported that the transpiration rate of *Loranthus europaeus* was 5 times higher than that of oak. The differences in the transpiration rates might be caused by a different duration of observations and the presence of drought in our study.

In addition to the reduced transpiration in the infested oaks, the stomatal conductance was also significantly lower compared to the non-infested trees (0.039 mmol m^{-2} s^{-1} and 0.062 mmol m^{-2} s^{-1}, respectively). Low stomatal conductance on infested trees prevents water loses, but on the other hand, it causes limited CO_2 assimilation. Hence, the presence of yellow mistletoe increases the risk of carbon starvation. Under a sufficient water supply, this is not an issue for a host [24], but carbon starvation remains a challenge under drought conditions. The activity of mistletoe reduced the stomatal conductance to the same extent as that of the transpiration rate, as these processes are tidily coupled. This is in accordance with Orcutt and Nilsen [48], who demonstrated a decline in the conductance in tree leaves infested by yellow mistletoe to 68% when compared with non-infested oaks. The stomatal conductance of mistletoe (0.159 mmol m^{-2} s^{-1}) was significantly higher than in both groups of oaks confirming the huge water withdrawal from the host tree xylem sap. A high rate of water loss due to stomata openness is the most common feature among mistletoes either from temperate, tropical or arid regions [40,41,45,46,49,50].

As is apparent from the aforementioned results, we found a difference in the photosynthetic performance between the infested and non-infested trees. The close relationship between the severity of drought stress and the decline in photosynthetic activity was earlier described by Wang et al. [51], who mainly attributed the decline of photosynthesis during mild and severe drought stress to the stomatal limitation and PSII photoinhibition with accumulation of reactive oxygen species (ROS), respectively. Queijeiro-Bolaños et al. [52] stated that the presence of mistletoes impacts the photosynthetic performance of host according to the severity of infestation. The maximal rate of CO_2 assimilation (A_{sat}) in our study dropped to 55%. A similar decline (drop to 68%) was found by Johnson and Choinski [44]. For oaks infested by mistletoe, Schulze et al. [40] found A_{sat} values between 4 and 7 μmol CO_2 m^{-2} s^{-1}. Generally, the A_{sat} values of non-infested oaks are much higher. Epron et al. [53] found A_{sat} values for *Q. petraea* ranging from 12 to 14 μmol CO_2 m^{-2} s^{-1} and Morecroft and Roberts [54] published a value of 10.4 μmol CO_2 m^{-2} s^{-1} for *Q. rubra*. Osuna et al. [55] published values from 7 to 15 μmol CO_2 m^{-2} s^{-1} for *Q. douglasii* in dry years, but the A_{sat} values shifted to 15–23 μmol CO_2 m^{-2} s^{-1} during the peak of the growing season for precipitation rich years. Differentiation between our results and those mentioned above suggests pronounced drought stress during the measurements. Typically, most mistletoes have lower rates of CO_2 assimilation rate than the trees they infest, but this is not an absolute rule [56–58]. Davidson et al. [41], as in our case, observed that the A_{sat} values of a hemiparasite and its host were nearly equal. In addition to reducing the CO_2 assimilation rate of the host tree, yellow mistletoe can gain carbon from host xylem sap, as there is no phloem bridge between the host tree and hemiparasite [59]. This heterotrophic carbon gain by mistletoe species could range from 5% to 62% [57,60,61], which might lead to different impacts on host trees according to the abundance of the parasite. To date, no such data are available for yellow mistletoe.

Coupled processes of CO_2 assimilation and water transpiration on the leaf level are described by the intrinsic water use efficiency (WUEi), which is the ratio of carbon assimilation to the rate of stomatal conductance. Lower WUEi values generally means that more water is required for carbon fixation. In our study, the WUEi values of infested trees were significantly lower (73 μmol H_2O mol^{-1}) than that of non-infested trees (84 μmol H_2O mol^{-1}). This corresponds well with the results of Sangüesa-Barreda et al. [62], who reported the substantial decrease in WUEi of Scots pine (*Pinus sylvestris*) severely infested by European mistletoe (*Viscum album*) based on long-term observation. Similar results were recorded by Queijeiro-Bolaños et al. [52] for the same species. The mistletoes in our study had WUEi values 4 times lower than those of non-infested oaks, which also support the strongly anisohydric behaviour of this hemiparasitic plant.

In addition to the above, drought stress could be identified with the intercellular concentration of CO_2 (Ci). Cornic and Massachi [63] identified drought stress by declining CO_2 concentration inside the leaf. We found significantly higher Ci values accompanied by lower stomatal conductance in infested oaks. A situation in which Ci increases with declining stomatal conductance was described by Flexas and Medrano [64], who identified an inflexion point beyond which Ci starts to increase with declining conductance. The inflexion point usually lies close to the value of 50 mmol H_2O m^{-2} s^{-1}. In general, stomatal conductance below 150 causes metabolic limitation; for g_S values below 100 mmol H_2O m^{-2} s^{-1}, photochemical activity is reduced; and for g_S values below 50 mmol H_2O m^{-2} s^{-1}, permanent photoinhibition frequently occurs [65]. The stomatal conductance values in our study were notably low, at 60 mmol H_2O m^{-2} s^{-1} in non-infested tree leaves and 39 mmol H_2O m^{-2} s^{-1} in infested tree leaves. According to the association of higher Ci values with lower conductance values in infested trees, we assume that the Ci values were beyond the inflection point of the Ci curve, and photoinhibition is present. In contrast, non-infested trees show lower Ci values and higher g_S values. This points out that the inflection point of the Ci curve lies between these two values and that infested individuals undergo severe heat-light stress, which could lead to permanent photoinhibition.

The method of chlorophyll *a* fluorescence can be a sensitive indicator of plant photochemical performance under a wide range of stress factors and it is suitable for assessing photoinhibition [66]. Even though PSII has been considered relatively resistant to drought and is more suitable as an indicator of heat stress than drought [67,68], several studies have used this method and confirmed significant reduction in the photochemical performance under drought conditions [35,68,69]. Moreover, oaks in field conditions may suffer from additive effects, such as heat stress or a hindered ability to regenerate photosystems II [70]. Therefore, we expected that the unsuitable conditions intensified by yellow mistletoe ($\Psi = -1.58$ MPa in non-infested trees and $\Psi = -2.5$ MPa in infested oaks) will alter the PSII photochemistry yield in both the dark-adapted and light-adapted states. However, the Fv/Fm and Φ_{PSII} values (ranging in the interval 0.8–0.81 and 0.4–0.45, respectively) showed no significant differences between non-infested and infested oak trees. Φ_{PSII} values in the range of 0.3–0.6 were described for two oaks species by [71], whereas the values were the same during wet and dry summers, suggesting a relatively high resistance of PSII photochemistry to drought. However, the electron transport rate (ETR), which is closely coupled with the maximum velocity of carboxylation ($V_{c,max}$), can be used as a surrogate for leaf biochemistry and reflects gross photosynthesis [72,73], was significantly reduced in infested trees. This suggests that non-infested oaks utilize light energy in carboxylation more efficiently than infested trees, which corresponds well with the carbon assimilation rate results. On the other hand, we observed substantially upregulated non-photochemical quenching NPQ in infested oak trees. The higher values of NPQ means that the infested trees probably tried to avoid possible damage caused by the ROS overproduction by dissipating excess energy through the xanthophyll cycle, as they were under more severe drought stress [74,75]. Such a protective behaviour to maintain normal photochemistry has been described in several studies dealing with drought stress [51,76]. However, Wang et al. [51] described the upregulated NPQ under moderate drought stress caused by reversible photosynthetic adjustment (e.g., limited stomatal conductance, Ribulose-1-5-bisphosphate (RuBP) regeneration, adenosine triphosphate (ATP) synthesis, and/or decreases in Rubisco activity). On contrast, under a severe water deficit (with a Ψ values of approximately -3 MPa in apple tree leaves), the sharp drop of NPQ was observed, which suggests the irreversible impairment of PSII units. As we observed the higher ability of NPQ in the infested trees, we can conclude that the changes in photosynthesis of infested oaks were caused by reversible photosynthetic adjustment rather than irreversible damages of PSII.

4.2. Relationships between Oaks and Mistletoes: Growth and Mineral Nutrition

Long-term reduced photosynthesis may alter growth as well. The results of our study show significant differences in the growth parameters between non-infested and infested oak trees. We found differences in the stem increment (ca. 0.63 mm) and height (2.6 m) of non-infested oaks. Infested trees were significantly lower compare to the non-infested ones as well as their radial growth was slower.

We found that the height of trees has no significant effect on the number of mistletoes, while the relationship between the diameter at breast height and the number of mistletoes on the oaks was significant ($r^2 = 0.234$). Similar to Aukema and Martinez del Rio [77], we suppose that the tree height, crown density and position of oaks in the forest stand play important roles for infestation of the oak by mistletoe. However, the frequency and density of mistletoe are not related just to a character of the oaks and the forest stand. In general, the distribution of mistletoes is affected by climate conditions such as winter and spring frosts [31] and the presence and frequency of common bird species that feed on mistletoe fruits [78–80]. Birds usually prefer higher oak trees with a high crown density to smaller oak trees with crown lift in the forest stand [77]. According to Overton [81], larger trees are older and have more branches (space) for birds. This could be a reason why they have a higher probability of being colonized by mistletoe [82]. Matula et al. [23] found that the probability of mistletoe infection increased significantly as the stem diameter of oak increased (>27 cm). Our results confirm this observation, because mistletoe infestation occurred (Figure 6b) on trees with a stem diameter above 31 cm. If mistletoe attacks an individual oak tree, the probability of mistletoe frequency will increase due to its own reproduction and to the behaviour of birds, which disperse mistletoe seeds. This means that the most infested oaks could be those that are the nearest to other infested trees. If the distance to an infested oak tree is greater than 5 m, the likelihood of infection is much smaller [23].

Growth of oak trees has focused not only on the carbon and water cycle mechanisms, but also on the critical role of soil-available nutrients [83]. Our results show that the soil under oak forests is an important reservoir of organic C. In the soil of our study area, we found 22.1 g kg^{-1} of C. Soils from European oak forests show similar carbon concentrations [84]. On the other hand, we detected a relatively low content of Mg. According to Nilsson [85], this is not especially surprising because acidic, respectively cambic podzols are characteristic of a low availability of nutrients (particularly Mg). The ability to accumulate nutrients (other than Mg) was slightly higher in infested oaks than in non-infested oaks (Table 2), but this difference was not significant suggesting that mistletoe does not reduce the nutrient content of host trees. The reason for the low differences between infested and non-infested oaks could be the stock of nutrients that oaks have, similar to other long-living plants. We suppose that the infested trees acclimated their metabolism and mineral acquisition processes to mitigate the negative effect of the hemiparasites. On the other hand, this phenomenon may be related to the number and age of mistletoes. We expect that a low number of young mistletoes will not cause as much damage as a high number of older mistletoes. However, as there is no data about the length of time for which this particular mistletoe infestation has been in the study area, the present study can only describe one snap shot in time. Mistletoes derive nutrients from their hosts [86], so we expected lower nutrient contents in the leaves of infested trees. However, the nutrient content in leaves did not differ significantly between infested and non-infested trees in our study. Infested oaks had a relatively higher nutrient accumulation than non-infested oaks. However, the overall biomass of leaves may differ between infested and non-infested oaks. The formatting of smaller and thicker leaves and the overall reduction of growth is probably a reaction to competition for water, inorganic ions, and metabolites [87], and, at the same time, could be a mechanism to maintain a relatively high efficiency of light-dependent photosynthetic reactions. We confirmed that infested trees formed smaller leaves without significant differences in the thicknesses of the leaves between oaks. Thus, the total amounts of allocated nutrients in leaves probably differ too. The high hemiparasite to host phosphorus ratio (5× higher concentration in mistletoe) is in agreement with data published by Lamont [88]. Ca and Mg are in the plausible range, but potassium was much lower in our study. According to Hosseini et al. [86] and Glatzel and Geils [26], nutrient flow through the transpiration stream is predominantly a one-way flow (from host to mistletoe, not the opposite direction). This may be the main reason why the content of some mineral nutrients is higher in mistletoe leaves than in infested and non-infested oak trees. Many other authors have reported similar findings on various host and mistletoe species [89–91] Mistletoe parasitism is a strategy for N acquisition [61,88]. In our study, the N content was markedly lower in mistletoe than in both infested and non-infested oaks. Schulze and Ehleringer [22] and Ehleringer et al. [50] reported N availability as a limiting factor for mistletoe growth. In the study by Schultze et al. [40], the ratio of parasite/host N content was 1.08. Additionally, in the study by Bowie and Ward [92], the N content was higher in mistletoe than in host trees. In our case, the ratio is opposite (0.66), and we found higher N contents in the host tree leaves. The same findings were published Hosseini et al. [86]. However, we discarded the leaf petiole prior to determination of the elemental contents, which could cause a discrepancy with the aforementioned studies.

5. Conclusions

According to our results, we can conclude that yellow mistletoe (*Loranthus europaeus*) infestation seriously threatens the physiological and growth performance of sessile oak (*Quercus petraea* s.l.) trees. We observed a significant reduction of leaf water potential, transpiration rate and stomatal conductance in infested oak trees, which resulted in the decrease of their CO_2 assimilation rate as well. Moreover, significant changes of PSII photochemistry were observed in infested trees. Considering the nutrient status, infestation by yellow mistletoe caused a reduction in the accumulation of C, which probably led to a lower stem increment and leaf size of infested oak trees.

For this reason, we suppose that yellow mistletoe may represent a great ecological and economic problem in Europe oak forest stands, which could also reduce expected ecosystem services in these unique habitats. To determine the most effective way to combat yellow mistletoe infestation, intensive monitoring of both hemiparasite and its host is required.

Author Contributions: D.K., J.G., and P.F. conceived the design of the study, planned and coordinated the experiment; J.R., M.L., S.R., J.P. collected plant material, conducted the measurements of growth traits, and processed an overall description of stand and level of mistletoe infestation; M.K., P.F.J., J.R., D.K., A.K., H.H., and P.F. conducted the ecophysiological measurements and processed the data; P.F.J., A.K., M.K. performed the statistical analyses; M.K., P.F.J. and D.K. wrote the first version of the manuscript; all the authors contributed critically to the drafts and gave final approval for publication. All authors have read and agreed to the published version of the manuscript.

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References

1. Dimopoulos, P.; Bergmeier, E.; Chytrý, M.; Rodwell, J.; Schaminée, J.H.J.; Sykora, K.V. European oak woodlands: Past, present and future. *Bot. Chron. Patra* **2005**, *18*, 1–316.
2. Rock, J.; Gockel, H.; Schulte, A. Vegetationsdiversität in Eichen-Jungwüchsen aus unterschiedlichen Pflanzschemata. *Beitr. Für Forstwirtsch. Landschaftsökologie* **2003**, *37*, 11–17.
3. Kunz, J.; Löffler, G.; Bauhus, J. Minor European broadleaved tree species are more drought-tolerant than Fagus sylvatica but not more tolerant than *Quercus petraea*. *For. Ecol. Manag.* **2018**, *414*, 15–27. [CrossRef]
4. Breda, N.; Cochard, H.; Dreyer, E.; Granier, A. Water transfer in a mature oak stand (*Quercus petraea*): Seasonal evolution and effects of a severe drought. *Can. J. Forest Res.* **1993**, *23*, 1136–1143. [CrossRef]
5. Pretzsch, H.; Schütze, G.; Uhl, E. Resistance of European tree species to drought stress in mixed versus pure forests: Evidence of stress release by inter-specific facilitation. *Plant Biol.* **2013**, *15*, 483–495. [CrossRef] [PubMed]
6. Pachauri, R.K.; Mayer, L.; Core Writing Team. (Eds.) *IPCC Climate Change 2014: Synthesis Report*; Intergovernmental Panel on Climate Change: Geneva, Switzerland, 2014.
7. Mert, A.; Özkan, K.; Şentürk, Ö.; Negiz, M.G. Changing the potential distribution of Turkey oak (IL.) under climate change in Turkey. *Pol. J. Environ. Stud.* **2016**, *25*, 1633–1638. [CrossRef]
8. Sallé, A.; Nageleisen, L.-M.; Lieutier, F. Bark and wood boring insects involved in oak declines in Europe: Current knowledge and future prospects in a context of climate change. *For. Ecol. Manag.* **2014**, *328*, 79–93. [CrossRef]
9. Olano, J.M.; Laskurain, N.A.; Escudero, A.; De La Cruz, M. Why and where do adult trees die in a young secondary temperate forest? The role of neighbourhood. *Ann. For. Sci.* **2009**, *66*, 105. [CrossRef]
10. Thomas, F.M.; Blank, R.; Hartmann, G. Abiotic and biotic factors and their interactions as causes of oak decline in Central Europe. *For. Pathol.* **2002**, *32*, 277–307. [CrossRef]
11. Bréda, N. Water shortage as a key factor in the case of the oak dieback in the Harth Forest (Alsatian plain, France) as demonstrated by dendroecological and ecophysiological study. In *Recent Advances on Oak Health in Europe. Selected Papers from a Conference Held in Warsaw, Poland, 22–24 November 1999*; Instytut Badawczy Leśnictwa (Forest Research Institute): Warsaw, Poland, 2000; pp. 157–159.
12. Doležal, J.; Mazůrek, P.; Klimešová, J. Oak decline in southern Moravia: The association between climate change and early and late wood formation in oaks. *Preslia* **2010**, *82*, 289–306.
13. Sohar, K.; Helama, S.; Läänelaid, A.; Raisio, J.; Tuomenvirta, H. Oak decline in a southern Finnish forest as affected by a drought sequence. *Geochronometria* **2014**, *41*, 92–103. [CrossRef]
14. Sonesson, K.; Drobyshev, I. Recent advances on oak decline in southern Sweden. *Ecol. Bull.* **2010**, *11*, 197–208.
15. Jung, T.; Blaschke, H.; Oßwald, W. Involvement of soilborne Phytophthora species in Central European oak decline and the effect of site factors on the disease. *Plant Pathol.* **2000**, *49*, 706–718. [CrossRef]
16. Marçis, B.; Caël, O.; Delatour, C. Relationship between presence of basidiomes, above-ground symptoms and root infection by *Collybia fusipes* in oaks. *For. Pathol.* **2000**, *30*, 7–17. [CrossRef]
17. Hajji, M.; Dreyer, E.; Marçais, B. Impact of *Erysiphe alphitoides* on transpiration and photosynthesis in *Quercus robur* leaves. *Eur. J. Plant Pathol.* **2009**, *125*, 63–72. [CrossRef]

18. Galko, J.; Nikolov, C.; Kimoto, T.; Kunca, A.; Gubka, A.; Vakula, J.; Zúbrik, M.; Ostrihoň, M. Attraction of ambrosia beetles to ethanol baited traps in a Slovakian oak forest. *Biologia (Bratisl.)* **2014**, *69*, 1376–1383. [CrossRef]
19. Millaku, F.; Sahiti, G.; Abdullahu, K.; Krasniqi, E. The spread and the infection frecuency of the Golliak (Kosovo) forest with the species hemipariastoc mistletoe (*Loranthus europaeus* L.). *Int. Multidiscip. Sci. GeoConf. SGEM* **2011**, *2011*, 1035–1040.
20. Kumbasli, M.; Keten, A.; Beskardes, V.; Makineci, E.; Özdemir, E.; Yilmaz, E.; Zengin, H.; Sevgi, O.; Yilmaz, H.C.; Caliskan, S. Hosts and distribution of yellow mistletoe (*Loranthus europaeus* Jacq. (Loranthaceae) on Northern Strandjas Oak Forests-Turkey. *Sci. Res. Essays* **2011**, *6*, 2970–2975.
21. Gebauer, R.; Volařík, D.; Urban, J. Seasonal variations of sulphur, phosphorus and magnesium in the leaves and current-year twigs of hemiparasitic mistletoe *Loranthus europaeus* Jacq. and its host *Quercus pubescens* Willd. *J. For. Sci.* **2018**, *64*, 66–73.
22. Schulze, E.D.; Ehleringer, J.R. The effect of nitrogen supply on growth and water-use efficiency of xylem-tapping mistletoes. *Planta* **1984**, *162*, 268–275. [CrossRef]
23. Matula, R.; Svátek, M.; Pálková, M.; Volařík, D.; Vrška, T. Mistletoe infection in an oak forest is influenced by competition and host size. *PLoS ONE* **2015**, *10*, e0127055. [CrossRef]
24. Zweifel, R.; Bangerter, S.; Rigling, A.; Sterck, F.J. Pine and mistletoes: How to live with a leak in the water flow and storage system? *J. Exp. Bot.* **2012**, *63*, 2565–2578. [CrossRef]
25. Rigling, A.; Eilmann, B.; Koechli, R.; Dobbertin, M. Mistletoe-induced crown degradation in Scots pine in a xeric environment. *Tree Physiol.* **2010**, *30*, 845–852. [CrossRef] [PubMed]
26. Glatzel, G.; Geils, B.W. Mistletoe ecophysiology: Host-parasite interactions. *Botany* **2009**, *87*, 10–15. [CrossRef]
27. Press, M.C.; Phoenix, G.K. Impacts of parasitic plants on natural communities. *New Phytol.* **2005**, *166*, 737–751. [CrossRef]
28. Glatzel, G. Mineral nutrition and water relations of hemiparasitic mistletoes: A question of partitioning. Experiments with *Loranthus europaeus* on *Quercus petraea* and *Quercus robur*. *Oecologia* **1983**, *56*, 193–201. [CrossRef] [PubMed]
29. Mathiasen, R.L.; Nickrent, D.L.; Shaw, D.C.; Watson, D.M. Mistletoes - ecology, systematics, ecology and management. *Plant Dis.* **2008**, *92*, 987–1006. [CrossRef]
30. Szmidla, H.; Tkaczyk, M.; Plewa, R.; Tarwacki, G.; Sierota, Z. Impact of Common Mistletoe (*Viscum album* L.) on Scots Pine Forests—A Call for Action. *Forests* **2019**, *10*, 847. [CrossRef]
31. Eliáš, P. Úhyn imelovca (*Loranthus europaeus* Jacq.) na severnej hranici rozšírenia v Európe: Slovensko (in Slovak)/Mistletoe (Loranthus europaeus Jacq.) mortality at northern geographical distribution in Europe: Slovakia. In *Dreviny v Mestskom Prostredí a v Krajine. Aktualne Trendy Dendrologického Výskumu a Praxe*; Slovak University of Agriculture: Nitra, Slovakia, 2007.
32. Kubíček, J.; Martinková, M.; Špinlerová, Z. Hemiparazité a provozní bezpečnost jimi napadených stromů (in Czech)/Hemiparasites and operating safety of infested trees. In *Provozní Bezpečnost Stromů*; Mendel University in Brno: Brno, Czech Republic, 2011; pp. 44–50.
33. McDowell, N.G. Mechanisms linking drought, hydraulics, carbon metabolism, and vegetation mortality. *Plant Physiol.* **2011**, *155*, 1051–1059. [CrossRef] [PubMed]
34. Doležal, J.; Lehečková, E.; Sohar, K.; Altman, J. Oak decline induced by mistletoe, competition and climate change: A case study from central Europe. *Preslia* **2016**, *88*, 323–346.
35. Maxwell, K.; Johnson, G.N. Chlorophyll fluorescence—A practical guide. *J. Exp. Bot.* **2000**, *51*, 659–668. [CrossRef] [PubMed]
36. Brooks, R.R. *Biological Methods of Prospecting for Minerals*; Wiley: Chichester, UK; New York, NY, USA, 1983.
37. Griebel, A.; Watson, D.; Pendall, E. Mistletoe, friend and foe: Synthesizing ecosystem implications of mistletoe infection. *Environ. Res. Lett.* **2017**, *12*, 115012. [CrossRef]
38. Cochard, H.; Bréda, N.; Granier, A. Whole tree hydraulic conductance and water loss regulation in Quercus during drought: Evidence for stomatal control of embolism? *Ann. Sci. For.* **1996**, *53*, 197–206. [CrossRef]
39. Zuber, D. Biological flora of Central Europe: *Viscum album* L. Flora—Morphol. Distrib. *Funct. Ecol. Plants* **2004**, *199*, 181–203. [CrossRef]
40. Schulze, E.D.; Turner, N.C.; Glatzel, G. Carbon, water and nutrient relations of two mistletoes and their hosts: A hypothesis. *Plant Cell Environ.* **1984**, *7*, 293–299.

41. Davidson, N.J.; True, K.C.; Pate, J.S. Water relations of the parasite: Host relationship between the mistletoe *Amyema linophyllum* (Fenzl) Tieghem and *Casuarina obesa* Miq. *Oecologia* **1989**, *80*, 321–330. [CrossRef]
42. Steckel, M.; del Río, M.; Heym, M.; Aldea, J.; Bielak, K.; Brazaitis, G.; Černý, J.; Coll, L.; Collet, C.; Ehbrecht, M.; et al. Species mixing reduces drought susceptibility of Scots pine (*Pinus sylvestris* L.) and oak (*Quercus robur* L., *Quercus petraea* (Matt.) Liebl.)—Site water supply and fertility modify the mixing effect. *For. Ecol. Manag.* **2020**, *461*, 117908. [CrossRef]
43. Sade, N.; Gebremedhin, A.; Moshelion, M. Risk-taking plants: Anisohydric behavior as a stress-resistance trait. *Plant Signal. Behav.* **2012**, *7*, 767–770. [CrossRef]
44. Johnson, J.M.; Choinski, J.S. Photosynthesis in the *Tapinanthus-Diplorhynchus* Mistletoe-Host Relationship. *Ann. Bot.* **1993**, *72*, 117–122. [CrossRef]
45. Ullmann, I.; Lange, O.L.; Ziegler, H.; Ehleringer, J.; Schulze, E.-D.; Cowan, I.R. Diurnal courses of leaf conductance and transpiration of mistletoes and their hosts in Central Australia. *Oecologia* **1985**, *67*, 577–587. [CrossRef]
46. Küppers, M.; Küppers, B.I.L.; Neales, T.F.; Swan, A.G. Leaf gas exchange characteristics, daily carbon and water balances of the host/mistletoe pair *Eucalyptus behriana* F. Muell. and *Amyema miquelii* (Lehm. ex Miq.) Tiegh. at permanently low plant water status in the field. *Trees* **1992**, *7*, 1–7.
47. Urban, J.; Gebauer, R.; Nadezhdina, N.; Čermák, J. Transpiration and stomatal conductance of mistletoe (*Loranthus europaeus*) and its host plant, downy oak (*Quercus pubescens*). *Biologia (Bratisl.)* **2012**, *67*, 917–926. [CrossRef]
48. Orcutt, D.M.; Nilsen, E.T. *Physiology of Plants under Stress: Soil and Biotic Factors*; John Wiley & Sons, Inc.: Hoboken, NJ, USA, 2000.
49. Goldstein, G.; Rada, F.; Sternberg, L.; Burguera, J.L.; Burguera, M.; Orozco, A.; Montilla, M.; Zabala, O.; Azocar, A.; Canales, M.J.; et al. Gas exchange and water balance of a mistletoe species and its mangrove hosts. *Oecologia* **1989**, *78*, 176–183. [CrossRef]
50. Ehleringer, J.R.; Schulze, E.-D.; Ziegler, H.; Lange, O.L.; Farquhar, G.D.; Cowar, I.R. Xylem-tapping mistletoes: Water or nutrient parasites? *Science* **1985**, *227*, 1479–1481. [CrossRef] [PubMed]
51. Wang, Z.; Li, G.; Sun, H.; Ma, L.; Guo, Y.; Zhao, Z.; Gao, H.; Mei, L. Effects of drought stress on photosynthesis and photosynthetic electron transport chain in young apple tree leaves. *Biol. Open* **2018**, *7*, bio035279. [CrossRef]
52. Queijeiro-Bolaños, M.E.; Malda-Barrera, G.X.; Carrillo-Angeles, I.G.; Suzán-Azpiri, H. Contrasting gas exchange effects on the interactions of two mistletoe species and their host Acacia schaffneri. *J. Arid Environ.* **2020**, *173*, 104041. [CrossRef]
53. Epron, D.; Dreyer, E.; Bréda, N. Photosynthesis of oak trees [*Quercus petraea* (Matt.) Liebl.] during drought under field conditions: Diurnal course of net CO_2 assimilation and photochemical efficiency of photosystem II. *Plant Cell Environ.* **1992**, *15*, 809–820. [CrossRef]
54. Morecroft, M.D.; Roberts, J.M. Photosynthesis and stomatal conductance of mature canopy oak (*Quercus robur*) and sycamore (*Acer pseudoplatanus*) trees throughout the growing season. *Funct. Ecol.* **1999**, *13*, 332–342. [CrossRef]
55. Osuna, J.L.; Baldocchi, D.D.; Kobayashi, H.; Dawson, T.E. Seasonal trends in photosynthesis and electron transport during the Mediterranean summer drought in leaves of deciduous oaks. *Tree Physiol.* **2015**, *35*, 485–500. [CrossRef]
56. Strong, G.L.; Bannister, P.; Burritt, D. Are mistletoes shade plants? CO_2 assimilation and chlorophyll fluorescence of temperate mistletoes and their hosts. *Ann. Bot.* **2000**, *85*, 511–519. [CrossRef]
57. Marshall, J.D.; Dawson, T.E.; Ehleringer, J.R. Integrated nitrogen, carbon, and water relations of a xylem-tapping mistletoe following nitrogen fertilization of the host. *Oecologia* **1994**, *100*, 430–438. [CrossRef] [PubMed]
58. Lüttge, U.; Haridasan, M.; Fernandes, G.W.; de Mattos, E.A.; Trimborn, P.; Franco, A.C.; Caldas, L.S.; Ziegler, H. Photosynthesis of mistletoes in relation to their hosts at various sites in tropical Brazil. *Trees* **1998**, *12*, 167–174. [CrossRef]
59. Tennakoon, K.U.; Pate, J.S. Heterotrophic gain of carbon from hosts by the xylem-tapping root hemiparasite *Olax phyllanthi* (Olacaceae). *Oecologia* **1996**, *105*, 369–376. [CrossRef] [PubMed]
60. Marshall, J.D.; Ehleringer, J.R. Are xylem-tapping mistletoes partially heterotrophic? *Oecologia* **1990**, *84*, 244–248. [CrossRef]

61. Schulze, E.D.; Gebauer, G.; Ziegler, H.; Lange, O.L. Estimates of nitrogen fixation by trees on an aridity gradient in Namibia. *Oecologia* **1991**, *88*, 451–455. [CrossRef]
62. Sangüesa-Barreda, G.; Linares, J.C.; Julio Camarero, J. Drought and mistletoe reduce growth and water-use efficiency of Scots pine. *For. Ecol. Manag.* **2013**, *296*, 64–73. [CrossRef]
63. Cornic, G.; Massacci, A. Leaf photosynthesis under drought stress. In *Photosynthesis and the Environment*; Advances in photosynthesis and respiration; Baker, N.R., Ed.; Springer: Dordrecht, The Netherlands, 1996; pp. 347–366.
64. Flexas, J.; Medrano, H. Energy dissipation in C3 plants under drought. *Funct. Plant Biol.* **2002**, *29*, 1209. [CrossRef]
65. Flexas, J.; Escalona, J.M.; Medrano, H. Down-regulation of photosynthesis by drought under field conditions in grape vine leaves. *Aust. J. Plant Physiol.* **1998**, *25*, 893–900.
66. Björkman, O.; Demmig-Adams, B. Regulation of photosynthetic light energy capture, conversion, and dissipation in leaves of higher plants. In *Ecophysiology of Photosynthesis*; Schulze, E.-D., Caldwell, M.M., Eds.; Springer Berlin Heidelberg: Berlin/Heidelberg, Germany, 1995; pp. 17–47.
67. Aziz, A.; Larher, F. Osmotic stress induced changes in lipid composition and peroxidation in leaf discs of Brassica napus L. *J. Plant Physiol.* **1998**, *153*, 754–762. [CrossRef]
68. Brestic, M.; Zivcak, M. PSII fluorescence techniques for measurement of drought and high temperature stress signal in crop plants: Protocols and applications. In *Molecular Stress Physiology of Plants*; Rout, G.R., Das, A.B., Eds.; Springer India: New Delhi, India, 2013; pp. 87–131.
69. Konôpková, A.; Húdoková, H.; Ježík, M.; Kurjak, D.; Jamnická, G.; Ditmarová, L'.; Gömöry, D.; Longauer, R.; Tognetti, R.; Pšidová, E. Origin rather than mild drought stress influenced chlorophyll a fluorescence in contrasting silver fir (*Abies alba* Mill.) provenances. *Photosynthetica* **2020**, *58*, 549–559. [CrossRef]
70. Awasthi, R.; Kaushal, N.; Vadez, V.; Turner, N.C.; Berger, J.; Berger, K.H.M.; Nayyar, N. Individual and combined effects of transient drought and heat stress on carbon assimilation and seed filling in chickpea. *Funct. Plant Biol. FPB* **2014**, *41*, 1148–1167. [CrossRef] [PubMed]
71. Forner, A.; Valladares, F.; Aranda, I. Mediterranean trees coping with severe drought: Avoidance might not be safe. *Environ. Exp. Bot.* **2018**, *155*, 529–540. [CrossRef]
72. Galmés, J.; Ribas-Carbó, M.; Medrano, H.; Flexas, J. Rubisco activity in Mediterranean species is regulated by the chloroplastic CO_2 concentration under water stress. *J. Exp. Bot.* **2011**, *62*, 653–665. [CrossRef] [PubMed]
73. Galmés, J.; Flexas, J.; Savé, R.; Medrano, H. Water relations and stomatal characteristics of Mediterranean plants with different growth forms and leaf habits: Responses to water stress and recovery. *Plant Soil* **2007**, *290*, 139–155. [CrossRef]
74. Demmig-Adams, B.; Adams, W.W. Xanthophyll cycle and light stress in nature: Uniform response to excess direct sunlight among higher plant species. *Planta* **1996**, *198*, 460–470. [CrossRef]
75. Jahns, P.; Holzwarth, A.R. The role of the xanthophyll cycle and of lutein in photoprotection of photosystem II. *Biochim. Biophys. Acta BBA Bioenerg.* **2012**, *1817*, 182–193. [CrossRef]
76. Kościelniak, J.; Filek, W.; Biesaga-Kościelniak, J. The effect of drought stress on chlorophyll fluorescence in *Lolium-Festuca* hybrids. *Acta Physiol. Plant.* **2006**, *28*, 149–158. [CrossRef]
77. Aukema, J.E.; Martinéz del Rio, C. Where does a fruit-eating bird deposit mistletoe seeds? Seed deposition patterns and an experiment. *Ecology* **2002**, *83*, 3489–3496. [CrossRef]
78. Monteiro, R.F.; Martins, R.P.; Yamamoto, K. Host specificity and seed dispersal of *Psittacanthus robustus* (Loranthaceae) in Southeast Brazil. *J. Trop. Ecol.* **1992**, *8*, 307–314. [CrossRef]
79. Skórka, P.; Wójcik, J.D. Winter bird communities in a managed mixed oak-pine forest (Niepolomice Forest, Southern Poland). *Acta Zool. Crac.* **2003**, *46*, 29–41.
80. Eliáš, P. Quantitative ecological analysis of a mistletoe *Loranthus europaeus* jacq. population in an oak hornbeam forest space continuum approach. *Ekologia* **1987**, *6*, 359–372.
81. Overton, J.M.C. Dispersal and infection in mistletoe metapopulations. *J. Ecol.* **1994**, *82*, 711–723. [CrossRef]
82. Arruda, R.; Fadini, R.F.; Carvalho, L.N.; Del Claro, K.; Mourão, F.A.; Jacobi, C.M.; Teodoro, G.S.; Berg, E.; Caires, C.S.; Dettke, G.A. Ecology of neotropical mistletoes: An important canopy-dwelling component of Brazilian ecosystems. *Acta Bot. Bras.* **2012**, *26*, 264–274. [CrossRef]
83. Gressler, A.; Schaub, M.; McDowell, N.G. The role of nutrients in drought-induced tree mortality and recovery. *New Phytol.* **2017**, *214*, 513–520. [CrossRef] [PubMed]

84. Michopoulos, P.; Kaoukis, K.; Karetsos, G.; Grigoratos, T.; Samara, C. Nutrients in litterfall, forest floor and mineral soils in two adjacent forest ecosystems in Greece. *J. For. Res.* **2020**, *31*, 291–301. [CrossRef]
85. Nilsson, L.-O. Forest biogeochemistry interactions among greenhouse gases and N deposition. *Water. Air. Soil Pollut.* **1995**, *85*, 1557–1562. [CrossRef]
86. Hosseini, S.M.; Kartoolinejad, D.; Mirnia, S.K.; Tabibzadeh, Z.; Akbarinia, M.; Shayanmehr, F. The effects of *Viscum album* L. on foliar weight and nutrients content of host trees in Caspian forests (Iran). *Pol. J. Ecol.* **2007**, *55*, 579–583.
87. Hosseini, S.M.; Kartoolinejad, D.; Mirnia, S.K.; Tabibzadeh, Z.; Akbarinia, M.; Shayanmehr, F. The European mistletoe effects on leaves and nutritional elements of two host species in Hyrcanian forests. *Silva Luisiana* **2008**, *16*, 9.
88. Lamont, B. Mineral nutrition of mistletoes. In *The Biology of Mistletoes*; Calder, M., Bernhardt, P., Eds.; Academic Press Australia: Sydney, Australia, 1983; pp. 185–204.
89. Kutbay, H.; Karaer, F.; Kilinç, M. The Relationships of Some Nutrients between *Cuscuta epithymum* (L.) L. var. *epithymum* and *Heliotropium europaeum* L. Available online: /paper/The-Relationships-of-Some-Nutrients-Between-Cuscuta-Kutbay-Karaer/cf9b77e40ef40542f6de9a39ef0e5a4f1d69a1ca (accessed on 1 September 2020).
90. Malicki, L.; Berbeciowa, C. Content of basic macroelements in common parasitical weeds. *Acta Agrobot.* **1986**, *39*, 123–128. [CrossRef]
91. Türe, C.; Böcük, H.; Aşan, Z. Nutritional relationships between hemi-parasitic mistletoe and some of its deciduous hosts in different habitats. *Biologia (Bratisl.)* **2010**, *65*, 859–867. [CrossRef]
92. Bowie, M.; Ward, D. Water and nutrient status of the mistletoe *Plicosepalus acaciae* parasitic on isolated Negev Desert populations of Acacia raddiana differing in level of mortality. *J. Arid Environ.* **2004**, *56*, 487–508. [CrossRef]