

Morphological, antioxidase, and photosynthetic responses in Liaodong elata (*Aralia elata*) under treatments of light intensity

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Abstract

Light intensity is important for the growth, development, and productivity of shade-tolerant plants. *Liaodong elata* (*Aralia elata*) is a highly nutrient natural organic food widely used in northern China. However, little is known about its cultivation under shading treatments. In this study, we conducted the morphological, physiology, and photosynthetic capacity of *Liaodong elata* under five treatments of 0% (CK), 20% (S1), 35% (S2), 50% (S3), and 60% (S4) shading levels. The results showed that the plant morphology in both S3 and S4 is better than CK, S1, and S2. Furthermore, the antioxidant enzyme activities of CK, S1 and S2 were higher than S3 and S4 at 30 d (T1) and 40 d (T2) shading time. We speculate that a certain light stress could destroy the antioxidant enzyme system. Photosynthetic results showed that the chlorophyll content, Fv/Fm, YII, qP of S3 were higher and the NPQ was lower. It explained that S3 had a more efficient photosynthetic system. At the same time, we measured the physiological indices of leaves at different positions and analyzed the correlation of each index. We found that the Top leaves had more stable antioxidant enzymes, higher chlorophyll content, Fv/Fm, YII, qP, and lower NPQ. And antioxidant enzyme activity was negatively correlated with Fv/Fm, YII, qP. This study proved the effectiveness of using a shading net for *Liaodong elata* cultivation. The 50% shading treatment is the optimal condition for the cultivation of *Liaodong elata*. These results are helpful to improve the reference for the introduction and cultivation of *Liaodong elata*.

1 Introduction

Plant growth and development are affected by temperature, humidity, light and other environmental factors. Light is one of the most important environmental factors for plant growth and development (Xu et al. 2020). Light quality, photoperiod, and luminous flux have profound effects on the morphogenesis and growth of plant cells, tissues, and organ cultures (Reuveni et al. 2007). The adaptability of plant species to the light environment varies greatly (Huang et al. 2011). Due to the adaptation mechanism of plants, the plasticity of plants can be improved under shading conditions, including its morphological and physiological parameters (Xie et al. 2020). Some plants leave chlorophyll content increases and chlorophyll a/b value decreases to improve plant photosynthetic activity with shading, such as *Aglaonema commutatum* and *Phoebe bournei* (Abrams 1987; Lambers and Poorter 1992). When light conditions are not suitable for plant growth, light can become the main abiotic stress factor (Fiorucci et al. 2017). For instance, some plants elongate growth, accelerate fBottoming, early seed production and yield reduction after shading (Franklin et al. 2005). In the molecular aspect, shading also can induce decreased gene expression in soybean light harvesting complex, leading to low physiological photosynthesis. And it also led to the general down-regulation of key genes in starch and sucrose metabolism and hormone signaling pathways (Chen et al. 2020).

Photosynthesis enables plants to absorb energy from light (Pfannschmidt et al. 1999), and is considered as the fundamental physiological basis of crop growth and yield formation. Therefore, it is important to study the photosynthetic characteristics of crops to clarify and fully exploit the yield potential. Higher light intensity reduced the growth and quantum yield efficiency of the photosystem II reaction center

(Souza et al. 2019), but photochemical inactivation can be reduced by shading, and shading can improve photosynthetic electron transfer efficiency as well (Montanaro et al. 2009). Shading also protects plant oxidase activity. Therefore, shading treatment plays an important role in cultivation.

Liaodong elata is also called thorn bud or dragon tooth, which belongs to Araliaceae. It is a perennial deciduous small tree or shrub, mainly distributed in northeast China (Li and Chen 2017). *Liaodong elata* has many applications values, among them, the tender buds of *Liaodong elata* are natural green organic food with a delicious taste and high nutritional value, and the market is short. Due to the feeling of wild *Liaodong elata*, the number of *Liaodong elata* decreased dramatically. Therefore, the cultivation environment of *Liaodong elata* is a more important problem, and this problem has caused extensive research in the field of cultivation of *Liaodong elata*. Recent studies have shown that *Liaodong elata* grows better near the edge of the forest (Li and Chen 2017), and also choose mountain shady slope or half shady and half sunny slope planting. There are relevant reports that the net photosynthetic efficiency of 72.1 % in full is relatively high and is conducive to the growth of commodity buds (Li 2020).

At present, the influence of the environment on the morphology of *Liaodong elata* is only simply considered, but there are few more detailed research reports. To our knowledge, shade treatment has no relevant progress in the physiological direction of *Liaodong elata*. In recent years, the study of tolerance to shade in plants is mostly based on photosynthetic characteristics and chlorophyll fluorescence parameters. Chlorophyll fluorescence is closely related to various reaction processes in photosynthesis, the change of parameters can reflect the process of absorption, utilization, transfer, and dissipation of light energy of PSI and PSII (mainly PSII) (Schreiber et al. 1995). Therefore, we can quickly understand the operation and metabolism of photosynthetic apparatus from chlorophyll fluorescence. Plants under light stress can lead to the production of harmful active intermediates, mainly reactive oxygen species (ROS) (Zavafer et al. 2015; Tikkanen et al. 2014). ROS can lead to oxidative damage of chloroplasts, causing the optical system to be suppressed, and severely limiting plant growth (Barber et al. 1992). To maintain steady state of photosynthetic efficiency and prevent ROS accumulation, plants activate the buffering mechanisms. To scavenge ROS, plants further stimulate antioxidative mechanisms via the enhanced activity of associated enzymes such as catalase (CAT) and peroxidase (POD). CAT and POD are the key enzymes for scavenging O_2 and H_2O_2 in plants. Under normal circumstances, the combined action of POD and CAT can eliminate the damage of active oxygen free radicals to plants. Therefore, it is generally believed CAT and POD are closely related to plant stress resistance and senescence, so it has become an important research object of plant stress physiology.

Active control of the plant growth environment is usually used to optimize plant yield and quality (Dueck et al. 2016). With the adjustment of the industrial structure and the continuous development of facility agriculture, optically selective mesh or shade net has been increasingly used in crop greenhouse technology to control the quality and quantity of ambient light, control the phytochemical composition of cultivated plants, and protect them from pests and physical damage (Nissim-Levi et al. 2014; Selahle et al. 2014; Carvalho et al. 2016; Zare et al. 2019).

Therefore, to better understand the effect of the intensity of light on the morphological, antioxidant system, and photosynthetic performance. We studied four shading treatments of 20% shading (S1), 35% shading (S2), 50% shading (S3), and 60% shading (S4), and compared them with the full light (CK) as the control. This experiment is designed to assess the impact of light intensity on the plant. First, we investigated the physiological response of *Liaodong elata* to different treatments and compared it with CK by measuring morphology, the proportion of blade damage, height, and ground diameter. Second, we tested whether the response of plants to different light intensities is different by measuring antioxidase. Third, we revealed the photosynthetic response mechanism of *Liaodong elata* by testing the changes in chlorophyll and chlorophyll fluorescence parameters under different treatments. Our findings expand the current understanding of the physiological, antioxidant system, and photosynthetic responses of *Liaodong elata* to light intensity. Furthermore, this study provides theoretical support for optimizing light environment conditions in the planting process of *Liaodong elata*.

2 Materials And Methods

2.1 Survey of the research area

Liaodong elata with plant height of about 80cm planted in Taitou garden, Qingdao, Shandong Province, China. The geographical location is between 120° 285' E ~120° 287' E and 36° 475' N ~36° 478' N, and the average altitude is 54- 57 m. The mean annual temperature is 12.7 °C, the mean annual precipitation is 662.1 mm. The annual average relative humidity is 73 %. It is a typical temperate monsoon climate.

2.2 Plant materials and experimental design

The experiment was conducted on 2 September 2022 at the Qingdao Agricultural University, Qingdao, Shandong, China. Eight-twelve *Liaodong elata* were planted in different shade nets (6m×6m) without any other treatment. Three *Liaodong elata* with similar growth vigor were randomly selected from each group as experimental materials. The degree of shading was gradually increased by increasing the number of layers of yarn net. Treatments with full light (CK), 20% shading (S1), 35% shading (S2), 50% shading (S3) and 60% shading (S4) net. The functional leaves with different treatments (CK, S1, S2, S3, S4) and different parts (Top, Central and Bottom) were selected at 30 d, 40 d and 50 d of shading (T1, T2, T3), carefully washed in running water to remove adhering dusts and debris, and then packed individually and stored at -80°C for subsequent analysis (Fig. 2).

2.3 Determination of morphological indicators

In 30 d, 40 d, 50 d, the morphology of *Liaodong elata* were photographed and recorded. Plant height was measured from ground level to the top of the main stem. The diameter 5 cm above the unearthed position was measured as the diameter of stem. The leaf morphological was obtained by taking pictures of the

fresh leaves on a whiteboard. All proportions of leaf damage were calculated using Image J (Image J 1.53t).

2.4 Determination of antioxidant metabolism

For physiological analyses, approximately 100 mg of fresh leaf or root samples were ground in 5 mL of 50 mM sodium phosphate buffer (pH = 7.8) on ice and centrifuged at 10,000 rpm for 25 min at 4°C; the extract was used for enzyme assays. The activity of POD was measured following the methods (Abedi and Pakniyat 2010). The activity CAT was determined following the procedure (Wang 1995). The relative electrical conductivity was analyzed, and using the DDSJ-308A (Leizi, Yidian., China) following the methods (Sterck et al. 2008).

2.5 Determination of photosynthetic fluorescence parameters

The fresh leaves *Liaodong elata* were cut into filaments with a width of less than 1 mm and 0.2 g of the leaves were weighed and immersed in 95 % ethanol until the leaves became white. The absorbance was measured at 470 nm, 649 nm and 665 nm for chlorophyll measurement., respectively (Xu et al. 2020). The chlorophyll fluorescence parameters F_v/F_m , YII, qP and NPQ of *Liaodong elata* leaves were measured at 10: 00-12: 00 on the 30 d, 40 d, and 50 d of shading treatment. The measured *Liaodong elata* leaves were dark adapted for 30 minutes to measure the maximum photochemical rate (F_v/F_m). The actual photochemical efficiency (YII), the photochemical quenching coefficient (qP) and the non-photochemical quenching coefficient (NPQ) of PSII were measured after light recovery. Chlorophyll fluorescence parameters were analyzed, and imaging was conducted using the MAXI-Imaging-PAM Series chlorophyll fluorescence imaging system (Heinz Walz, Effeltrich., Germany) and a Dual-PAM-100 differential fluorescence analyzer (Heinz Walz, Effeltrich., Germany) following the methods (Naranjo et al. 2016). All data were generated via measurements of three biological replicates per treatment.

2.6 Statistical analysis

The morphological, physiological, and biochemical parameters of *Liaodong elata* were analysed by one-way analysis of variance (ANOVA). Duncan range test, and two-factor interaction analysis to compare the differences between different treatments and different leaf positions. Leaf parameters of *Liaodong elata* were analysed in five different treatments. Correlation analysis is used to explain the correlation between the coefficients. All calculations were performed using SPSS (IBM, Inc., 25.0, IL., USA). The difference between mean values was statistically significant at the level of the level of $p < 0.05$ level for the minimum significant difference test.

3 Results

3.1 Effects of shading treatments on morphology of Liaodong elata

To illustrate the light adaptability changes of Liaodong elata under different treatments, we first determine the morphology using five shading treatments (CK, S1, S2, S3, and S4) under three duration stages (T1, T2, and T3). Morphological changes under different treatments were not significant at T1. However, we found that the leaves of Liaodong elata were yellow after CK, S1, and S2 treatments, but the changes of S3 and S4 were not obvious. Therefore, we found different shading treatments can influence the morphology of Liaodong elata (Fig. 3a). In addition, we found that CK, S1, S2, S3 treatments could increase plant height, but S4 treatment had no significant effect on plant height (Fig. 3b). However, shading has little effect on the diameter of the ground (Fig. 3c). More specifically, shading treatments decrease Liaodong elata proportion of blade damage of Liaodong elata. Compared to CK, S1 and S2, we found that the treatments with S3 and S4 treatments could significantly reduce the proportion of leaf damage (Fig. 3d). Moreover, we also investigate the changes of leaves color of three positions (Top, Central, and Bottom) among these treatments. As shown in (Fig. S1), we found the Bottom position of the leaves to turn yellow more quickly than the Top and Central under T2 and T3, especially for T3. These results showed that shading can influence the morphology of Liaodong elata.

3.2 Effects of shading treatments on antioxidant enzyme activity of Liaodong elata

We next examined the physiological effects of shading on Liaodong elata. We found that the POD activity of the S1, S2, S3, and S4 treatments was Bottom than CK, but different shading treatments had no significant effect on the POD activity in T1 and T2. Interestingly, the POD activity of the S3 and S4 treatments was higher than other treatments in T3 (Fig. 4a). In addition, CAT activity was significantly higher under full light in T1, but the CAT activity under the treatments S1, S2, S3, and S4 was higher than CK in T2 and T3 (Fig. 4b). We also found that the Top leaves could maintain higher POD and CAT activities. CAT and POD work together to remove reactive oxygen species in plants to reduce plant damage, showing the same trend as POD.

3.3 Effects of shading treatments on the relative conductivity activity of Liaodong elata

The relative electrical conductivity (REC) of plant leaves can reflect the damage to the plant cell membrane. In the Top leaves, there was no significant change between all treatments in T1 and T2, but the REC of S3 and S4 is significantly higher than CK, S1, and S2. In the Central and Bottom leaves, we found that S1, S2, S3, S4 have Bottom relative conductivity than CK in T1 and T2, and the REC of leaves

under all treatments did not change significantly in T3 (Fig. 4c). It illustrates that light can damage of Liaodong elata cells. It directly proves that Liaodong elata was stressed under the full light.

3.4 Effects of shading treatments on chlorophyll content of *A.elata*

In plant photosynthesis, the chlorophyll content plays an important role in the absorption, transmission, and transformation of light energy, so chlorophyll can determine the efficiency of plant photosynthesis. We found that regardless of the top, central and bottom leaves, the chlorophyll content of S3 and S4 was higher than CK, S1, and S2 in T1, T2, and T3. In more detail, we found that the S3 treatment had a higher chlorophyll content (Fig. 4d). Therefore, we obtained that strong light inhibited the chlorophyll content of Liaodong elata leaves and a certain shade treatment can increase the chlorophyll content of Liaodong elata leaves. It illustrates light has a certain effect on the chlorophyll of Liaodong elata.

3.5 Effects of shading treatments on the chlorophyll fluorescence of Liaodong elata

Through analysis of chlorophyll fluorescence parameters, we found that shading had a significant effect on Fv/Fm, YII, qP, and NPQ (Fig. S2). And there are the highest chlorophyll fluorescence parameters in T1. Different parts of the leaves values are different. The Fv/Fm of the Top leaves changes a little under different treatments. The Fv/Fm of the Central and Bottom leaves under the S3 and S4 treatments is higher than the CK, S1, and S2 treatments. Therefore, we found that Fv/Fm always maintain a high level under the S3 and S4 treatments in T1, T2, and T3, but the Fv/Fm of the CK, S1 and S2 treatments gradually decreased in T1, T2, and T3. It indicates S3 and S4 treatments can maintain the maximum photochemical amount of PSII in leaves, ensure photosynthetic potential. YII and qP showed the same result as Fv/Fm, the YII and qP of S3 and S4 treatments were higher than CK, S1, and S2 treatments T1, T2, and T3. NPQ shows the opposite result with other parameters. The NPQ of CK, S1 and S2 treatments is higher than S3 and S4 treatments in T1, T2, and T3 (Figs. 5, S3).

3.6 Analysis of leaves in different parts

Under the same shade and time treatment, we compared the leaves of Liaodong elata in different positions. There was no significant difference in POD, CAT activity, REC, and chlorophyll content in leaves at different positions under full light treatment. However, we found that there were significant differences in leaf indexes at different positions in S1, S2, S3, and S4. We also found that the Top leaves had higher antioxidant enzyme activity, chlorophyll content and lower REC. The antioxidant enzyme activity and chlorophyll content of the Bottom leaves were the lowest (Table S1). We also analyzed the chlorophyll fluorescence of the different parts of the leaves. We found that the Top leaves had higher Fv/Fm, YII, qP,

and lower NPQ. And the result was Top leaves > Central leaves > Bottom leaves (Table S2). This is consistent with Table S1 from the physiological point of view.

3.7 Interaction between shading treatment and leaf position in different periods

The shading gradients, the leaves position and their interaction have significant effects on most physiological indices ($P < 0.05$). But the degree of influence will change over time, such as the activity of POD, REC, Fv/Fm, YII (Table S3).

3.8 Correlation analysis

In order to study the physiological effects of shading treatment on *Liaodong elata* and the relationship between physiological indexes, we evaluated the relationship between antioxidant enzyme activity, REC, chlorophyll content, and chlorophyll fluorescence parameters. We consider that most of the leaves have been damaged in T3. Therefore, we analyzed the correlation between *Liaodong elata* in T1 and T2. As demonstrated in (Fig. 6), there was a significant positive correlation between processing time and POD and CAT activities, the Pearson coefficient are 0.81 and 0.82 ($P < 0.01$), and there was a significant negative correlation with chlorophyll content, Fv/Fm, YII and qP, the Pearson coefficient are -0.46, -0.29, -0.69 and -0.56 ($P < 0.01$). The shading gradient was negatively correlated with POD activity, the Pearson coefficient is -0.21 ($P < 0.05$). And it was negatively correlated with REC and NPQ, the Pearson coefficient are -0.499 and -0.80 ($P < 0.01$). The shading gradient was positively correlated with Fv/Fm, the Pearson coefficient is 0.33 ($P < 0.01$) and qP, the Pearson coefficient is 0.22 ($P < 0.05$). We conclude that all indicators have a negative correlation between the leaves. Furthermore, we also explored the relationship between antioxidant enzyme activity, REC, chlorophyll, and chlorophyll fluorescence parameters. We found that POD activity was significantly negatively correlated with chlorophyll content, Fv/Fm, YII and qP, the Pearson coefficient are -0.46, -0.78 and -0.65 ($P < 0.01$), and there was a significant negative correlation between CAT activity and chlorophyll content, YII, qP, the Pearson coefficient are -0.32, -0.47 and -0.44 ($P < 0.01$). There was a significant negative correlation between REC and Fv/Fm, the Pearson coefficient is -0.29 ($P < 0.01$), and it has a significant positive correlation with NPQ, the Pearson coefficient is 0.38 ($P < 0.01$). Chlorophyll content has a significant positive correlation with Fv/Fm, YII, qP, the Pearson coefficient are 0.38, 0.60 and 0.64 ($P < 0.01$).

4 Discussion

4.1 Morphological effects of shading on *Liaodong elata*

The Photoperiod can not only affect plant flowering induction and flower differentiation, but can also affect its vegetative growth and physiological and biochemical characteristics (Hayama et al. 2004;

Arena et al. 2016). Plants can sense subtle changes in light quality through photoreceptors, which in turn regulate the growth and development of plants through exciting signaling pathways (Ward et al. 2005). As a plant photoreceptor organ, leaf traits are partly the result of plant response to the external environment (Vendramini et al. 2002) and its change is one of the most important strategies that the plant has developed to cope with adverse environments. Furthermore, leaf is the main source of light interception and carbon assimilation. Therefore, it is of great significance to study the influence of environment on leaf morphological characteristics for measuring the growth of *Liaodong elata*. The effects of low light and shade on morphological parameters have been reported extensively (Wu et al. 2017). Shading promoted the distribution of photosynthetic photon capture potential and higher chlorophyll content. Thus improving plant growth. Specifically, this study showed that shading, time and their interaction imposed significant impacts on leaf traits. We found that leaves had a higher proportion of damage in CK, S1, S2. The main reason is that the high light intensity causes the leaves not to lose the absorbed excess light energy in time, and then the leaves accumulate a large amount of reactive oxygen species, resulting in damage to the photosynthetic system. S4 treatment leaves less damage, but too low light intensity cannot provide enough material and capacity for normal growth of the plant due to the failure of the plants to obtain enough light energy for photosynthesis, resulting in its growth being inhibited. Moreover, there are research reports *Liaodong elata* grows best in the forest with canopy density of 0.2-0.3 (Li and Chen 2017). However, our results are not consistent with former studies that S1, S2, S3 promoted the growth of *Liaodong elata*, but S3 treatment had the most significant effect on the plant height of *Liaodong elata*. Furthermore, shading may help to maintain soil water quality and improve air humidity, which potentially providing the necessary water for the growth of *Liaodong elata*. Generally, the ground diameter is a measure of the diameter of the tree trunk and the thicker ground diameter indicates that the plant has strong growth. In this study, we found that shading has no significant effect on the ground diameter of *Liaodong elata*, we speculate that mainly because the test processing time was short, we cannot clearly observe the ground diameter changes of *Liaodong elata*.

4.2 Effects of shading on the antioxidant system of *Liaodong elata*

Under normal circumstances, the production and elimination of active oxygen in plants are in a dynamic equilibrium. When plants are under adverse conditions, this dynamic equilibrium is destroyed. Decreased ability of the antioxidant system to scavenge reactive oxygen species, leading to a large accumulation of reactive oxygen species, causing damage to cells (Moran et al. 2017). Research shows that many plants have increased antioxidant enzyme activity under strong light induction (Logan et al. 1998). And furthermore, ROS accumulation in plants can lead to inhibition of photosynthesis and respiration, which then affect plant growth. As *Liaodong elata* leaves contain a considerable amount of water, this will be an important radiation target that will result in the production of reactive oxygen species (ROS) such as superoxide anion (O_2^-), hydroxyl radical ($\cdot OH$), hydrogen peroxide (H_2O_2), hypochlorite anion (ClO^-) and singlet oxygen (1O_2) (Neill et al., 2002), which will cause *A.elata* cellular damage. Catalase (CAT) and

other antioxidases such as peroxidases (POD), ascorbate peroxidase (APX) and guaiacol peroxidase (GPX) function as important hydrogen peroxide scavengers (Bonet et al. 2012; Vanhoudt et al. 2014). The increase of proportion of blade damage ratio mainly suggested that Liaodong elata at CK, S1, S2 were sensitive to photoinhibition and can cause the formation of ROS. To get an overview of the effects of different light irradiances on these ROS scavenging enzymes, activities of POD and CAT were determined. We found under CK, S1 and S2, the activities of CAT and POD are higher than other treatments in T1 and T2. A significant increase in POD and CAT activity in Liaodong elata may have resulted due to the formation of H_2O_2 , which allows Liaodong elata to protect themselves against oxidative stress, whereas there is a slight reducing of POD and CAT activity in T3, which may be due to excessive reactive oxygen species leading to leaves death. Evidently, the greater increases in the activities of these two enzymes, together with the increase of proportion of blade damage ratio, resulting in less production of ROS in the Liaodong elata. According to the report, plants have developed several strategies to minimize these effects and protect themselves against oxidative damage in the process of adaptation to the environment, including protective effects of carotenoids and events associated with activation of the xanthophyll cycle (Lindahl et al. 1997). Therefore, our results show that under light stress, enhanced antioxidant metabolism detoxifies excessive ROS accumulation, so the antioxidant system plays an important role in alleviating light stress, such S1 and S2. However, for instance, for CK, higher light intensity will damage the antioxidant system of Liaodong elata and eventually lead to the death of plant leaves. However, we found that the leaves of Liaodong elata treated with S3 and S4 had relatively stable antioxidant enzyme activities at T1, T2 and T3, so S3 and S4 treatments could maintain the stability of antioxidant enzyme system, improve resistance, and reduce the damage of ROS to itself. We finally verified by measuring the REC, and the results were consistent with our speculation. CK, S1, S2 treatment had higher REC and S3, S4 treatment had Bottom REC at T1, T2 and T3.

4.3 Effect of shading on the photosynthetic mechanism of Liaodong elata

Plant photosynthesis is the physiological basis for the formation of crop yields. The chlorophyll in leaves is the material basis for the absorption, transmission, and conversion of light energy in plant photosynthesis. Plants utilize photosynthetic pigments (such as chlorophyll a, chlorophyll b, and carotenoids) in chloroplasts and convert the energy of the Sun into its own energy source to grow (Choi 2021). Therefore, the chlorophyll content is an important physiological index that reflects the photosynthetic intensity of crops. Studies have shown that shading can change the chlorophyll a, chlorophyll b, total chlorophyll content and chlorophyll a/b values of plants (Mendes et al. 2001). But too high shading will also reduce the chlorophyll content in leaves. This experiment was the same as the above conclusion, the total chlorophyll content of Liaodong elata under S3 were higher than CK, S1, S2 and S4. It shows that chloroplast can move in a certain environment to avoid short-term absorption of strong light and then to avoid light-induced damage to PSII and any subsequent photoinhibition (Kong et al. 2013; Szymańska et al. 2017). The increase in chlorophyll content may be related to the decrease of

photooxidation damage under low light conditions, the enlargement of the grana in the chloroplast and the increase in the degree of stacking of the grana lamella (Atanasova et al. 2003). Chlorophyll fluorescence parameters are widely used to reflect the intrinsic characteristics of light energy absorption, transmission, dissipation, and distribution during photosynthesis (Baker et al. 2004), the maximum chemical efficiency of PSII (F_v/F_m) is a measure of light energy conversion efficiency of the PSII reaction centre, under normal circumstances rarely changing. But when plants are stressed, the value of this parameter will be significantly reduced (Jiao et al. 2001). In the present study, F_v/F_m was significantly decreased at CK, S1, S2 coinciding with the sharp decline in chlorophyll content, implying that decrease in photosynthetic rate, which partly explains the mechanism of the inhibitory effect of light irradiance stress on the photosynthetic of *Liaodong elata*. Nevertheless, *Liaodong elata* has higher F_v/F_m under S3, and the increase of F_v/F_m is beneficial to enhance the speed and efficiency of photosynthetic pigments to convert light energy into chemical energy, and accumulate more energy for carbon assimilation, then improve the photosynthetic function of plant leaves, increase the photosynthetic rate (Shi et al. 2007). Y_{II} reflects the actual primary light energy capture efficiency of PSII reaction centre under light conditions. Photochemical quenching (qP) is a fluorescence quenching induced by photosynthesis, corresponding to the high or low photosynthetic activity of plants, the value of qP conveys the redox, state of QA (Original electron acceptor) the main electron acceptors and the number of PSII open centres. The higher the value of qP , the stronger the electron transfer activity of PSII. In this experiment, the Y_{II} and qP of T3 and T4 were significantly higher than those of CK, S1, and S2. It indicated that full light and low shading treatment reduce the reoxidation ability of QA during illumination. This effect may be due to the inhibition of Calvin cycle activity, proving certain light can reduce the irradiation induced pressure on PSII, and lead to the closure of PSII reaction center (Calatayud and Barreno 2001). On the contrary, the actual light energy capture efficiency, the number of PSII open centers, the number of oxidized QA and PSII electron transfer activity all increased of *Liaodong elata* after certain shading treatment, increasing reduced coenzyme nicotinamide adenine dinucleotide phosphate (NADPH) and to improve the formation of photosynthetic assimilation. At the same time, a certain shading also promotes the dark reaction in the chloroplast matrix, resulting in a significant increase in photosynthetic rate. NPQ is a self-protection mechanism of photosynthetic apparatus. It allows plants to absorb excess light energy and dissipate it in the form of heat, preventing excess light energy from damaging the photosynthetic apparatus. We found that S3 had Bottom NPQ than other treatments, mainly because *Liaodong elata* consumed excess light energy in the photosystem by reducing heat dissipation through its own adjustment mechanism, and improved the efficiency of captured light energy. At this time, the light energy quota for photochemical processes increased, and the energy allocated to non-photochemical processes decreased accordingly. In our investigation, the above changes might indicate that PSII function enhanced and excitation energy from reaction center increased, which was conducive to improving photosynthetic efficiency. Furthermore, we also speculate PSII reaction center under S3 open more than CK, S1, S2, and absorbed more light energy for photosynthesis to a greater extent, and the plantlets have higher electron transfer efficiency and stronger photosynthetic capacity. This may explain the better growth and leaf morphology of *Liaodong elata* under S3 treatment. Therefore, this conclusion verifies under appropriate light conditions, plants can improve their CO_2 fixation capacity and net photosynthetic rate by increasing light energy

absorption and electron transport rate (Stirbet et al. 2018), Moreover, we found that Liaodong elata leaves from up to low have different responses to resistance. It may be due to the different nutrient transport structure and life span of leaves at different positions, and we speculate that there are different anatomical structures, inclusions, and photosynthetic capacity in leaves at different positions of Liaodong elata, such as thickness, porosity, palisade tissue and spongy tissue. However, the current results are limited. To make the results more convincing, we will conduct multi-year studies. Furthermore, a follow-up study (i.e., structure botany, transcriptomics, metabonomics, and proteomics) should be conducted to obtain further insights into the mechanisms underlying the tolerance advantages. Meanwhile, an effective potential strategy for enhancing the plant tolerance and mitigating injure from lighting stress should be exported in the future sustainable agricultural production.

5 Conclusion

This experiment describes the effect of different light intensity on the growth of Liaodong elata in physiological level. In conclusion, this experiment showed Liaodong elata can significantly reduce the effect of light stress under 50% shading treatment, and maintain the stability of the antioxidant system, increase the total chlorophyll content, maintain high photochemical conversion efficiency and photosynthetic structure self-protection ability. And from the different positions of the leaves, whether it is the external morphology and internal physiological characteristics of the leaves, we found that the Top leaves perform best, and there is a trend of Top leaves > Central leaves > Bottom leaves. Therefore, we provide a cultivation method to ensure the normal growth of Liaodong elata after transplanting, and then improve the quality of Liaodong elata buds and ensure periodic production after transplanting.

Declarations

Author contributionsAll authors contributed to the study conception and design. Material preparation were performed by Jing Zhang and Zhenying Zhang. Data collection and analysis were performed by Boda Liu, Yong Sui, Hengxin Zhu, Yaru Shi, Xiuyu Sui. The first draft of the manuscript was written by Boda Liu and Xinqiang Jiang, and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

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Data availabilityThe datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

Competing interest The authors have no relevant financial or nonfinancial interests to disclose.

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Figures

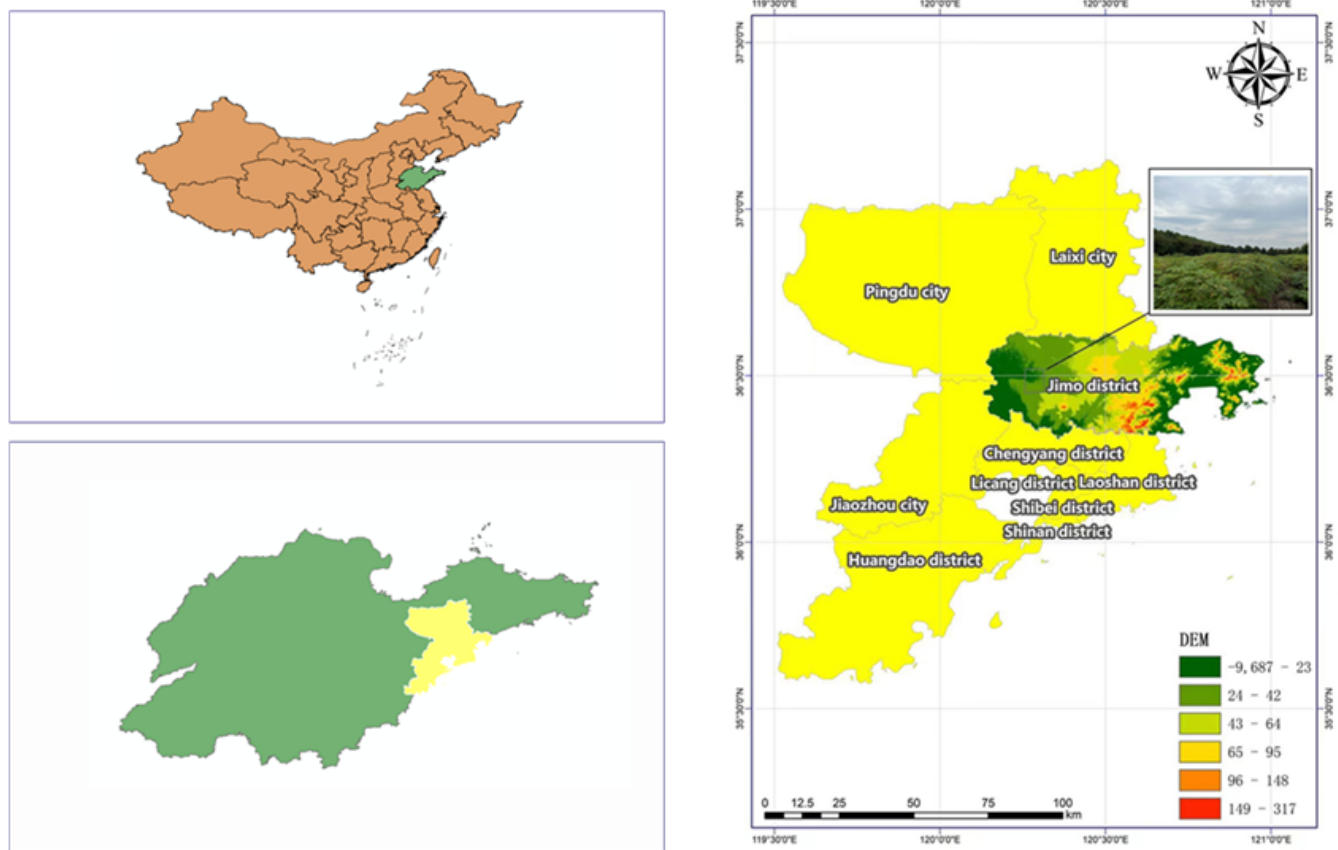


Figure 1

Geographical location diagram of *Liaodong elata* planting (generated using ArcGIS 10.8). China in brown color, Shandong province in green color, Qingdao in yellow color, and the inset map represents Taitou garden test field.

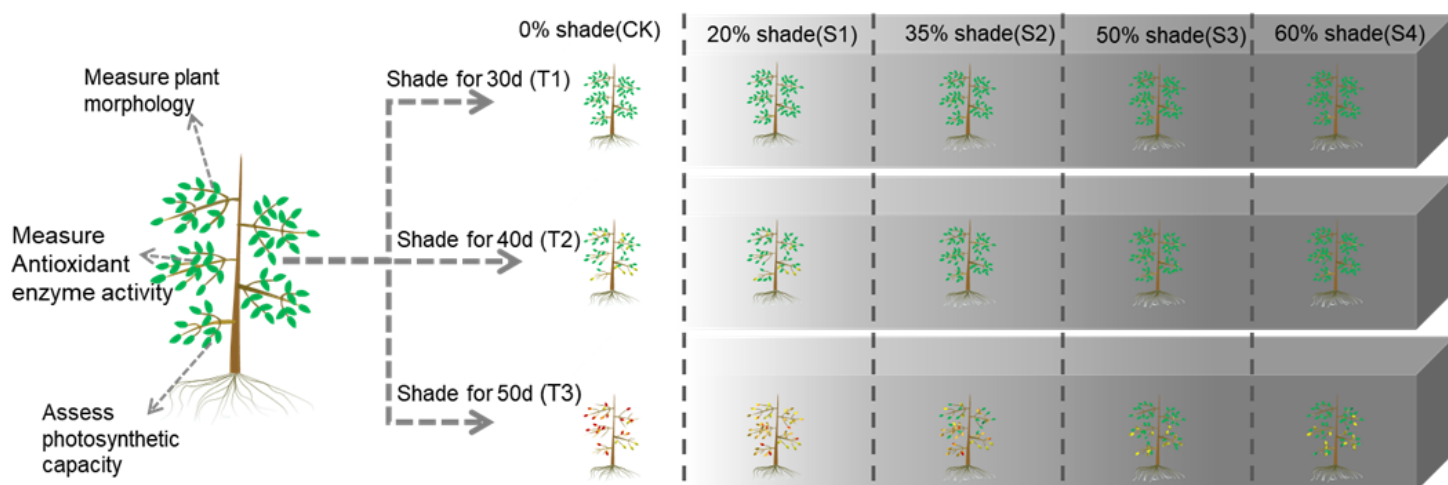


Figure 2

Schematic diagram of *Liaodong elata* under different shading and time treatments.

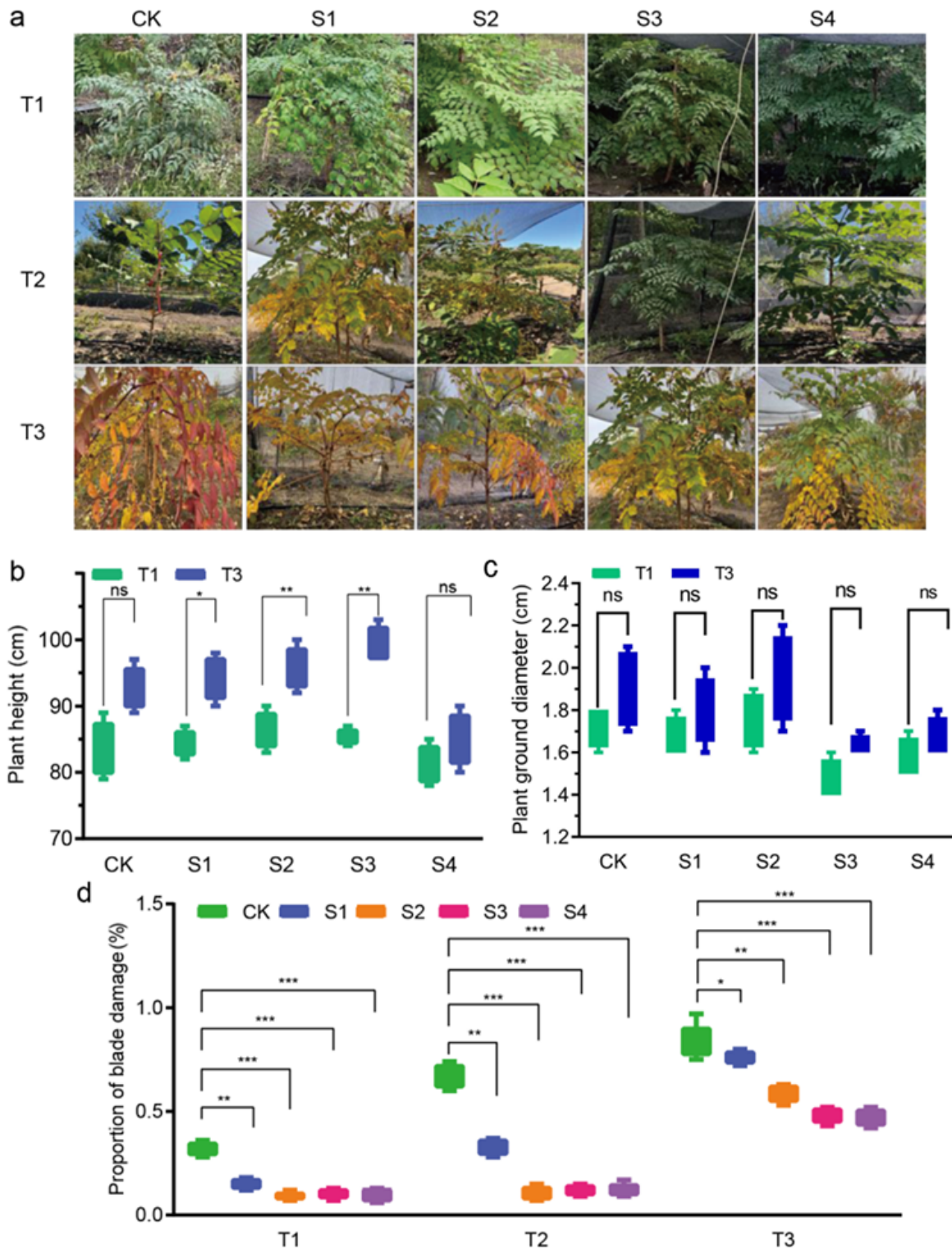


Figure 3

Morphological characteristics and statistical analysis of the shade conditions in *Liaodong elata*. (a) Morphological changes of *Liaodong elata* under different treatments. (b) leaf morphological changes under different treatments. (c) height change analysis. (d) Ground diameter change analysis. (e) leaf damage change analysis. Data are mean $\pm$ SD (n = 3). Asterisks indicate significant differences within each column (Duncan's multiple range test, * p 0.05, ** p < 0.01, *** p < 0.001). CK, full light; S1, 20%

shading; S2, 35% shading; S3, 50% shading; S4, 60% shading; T1, 30 d of shading; T2, 40 d of shading; T3, 50 d of shading.

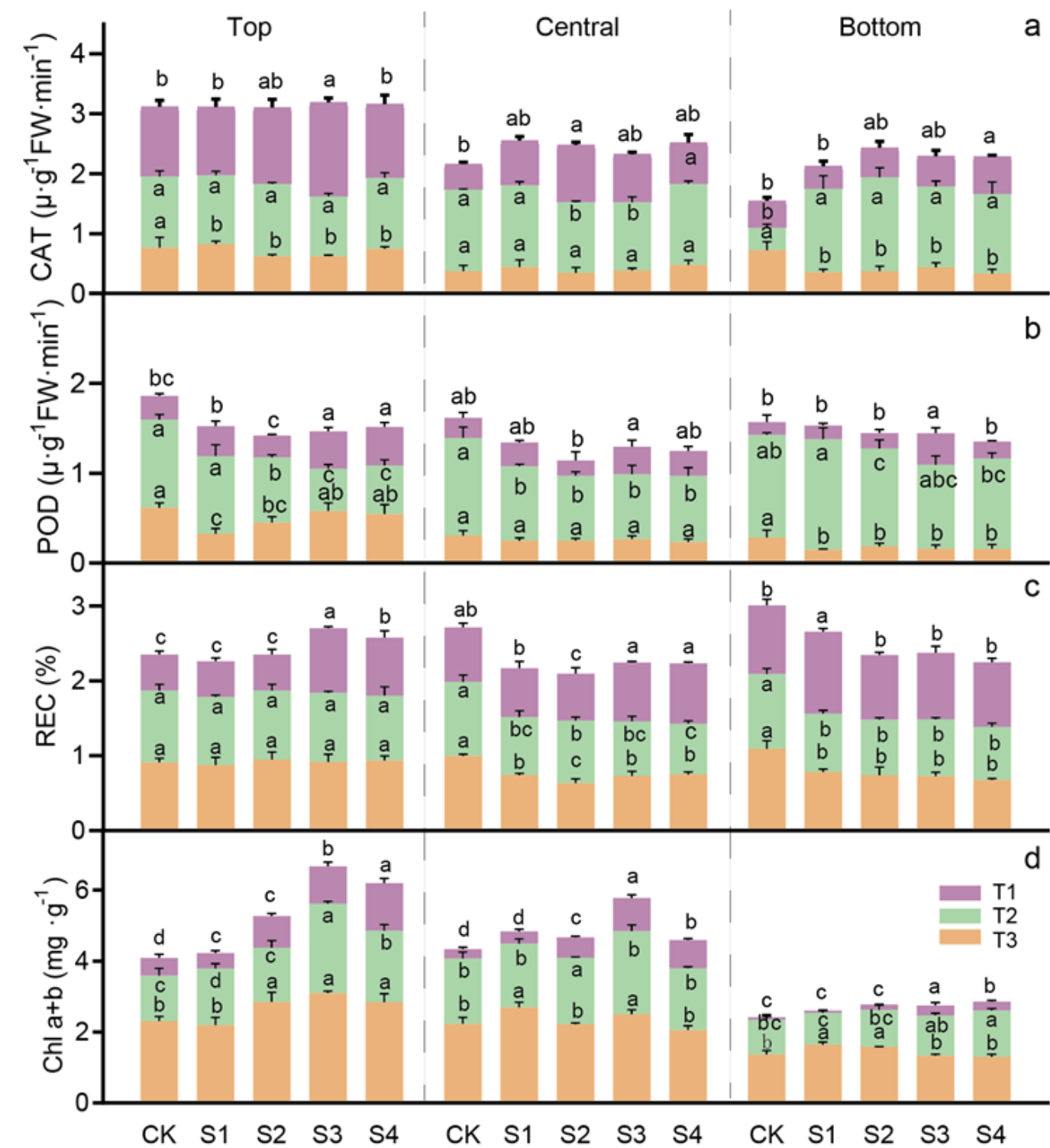


Figure 4

Analysis of physiological indexes of alder leaves in Liaodong elata at different times and treatments. (a) Analysis of POD change leaves. (b) Analysis of CAT change of leaves. (c) Analysis of relative

conductivity variation of leaves. (d) Analysis of chlorophyll content of leaves. Data are mean $\pm$ SD (n = 3). Different letters (a, b, c) in the same column indicate significant differences between treatments at $p \leq 0.05$ according to Duncan's new multiple range test. CK, full light; S1, 20% shading; S2, 35% shading; S3, 50% shading; S4, 60% shading; T1, 30 d of shading; T2, 40 d of shading; T3, 50 d of shading; POD, peroxidase; CAT, catalase; REC, relative electrical conductivity; Chl a+b, total chlorophyll a+b.

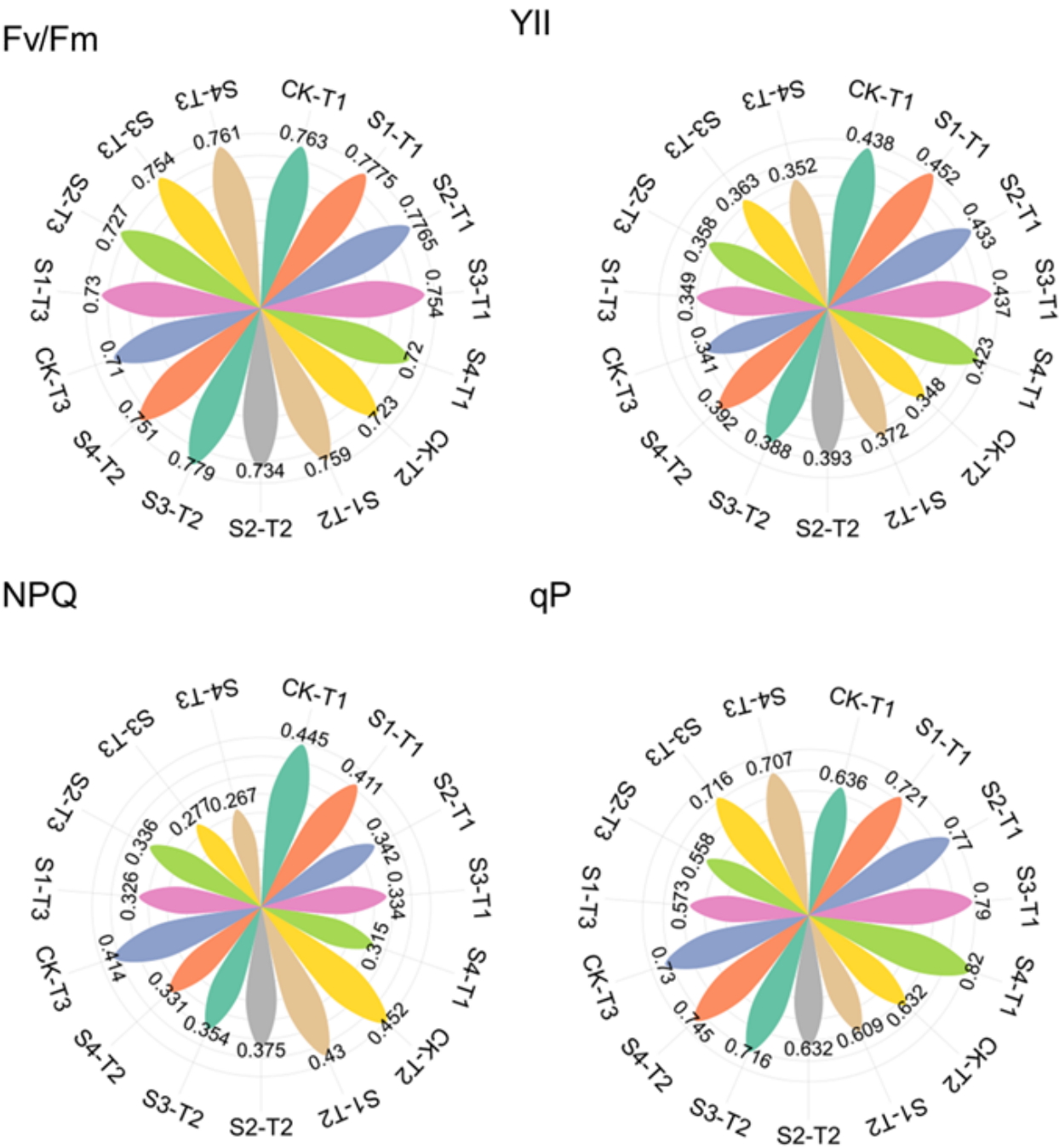


Figure 5

Chlorophyll fluorescence parameters of Top leaves of *Liaodong elata*. CK, full light; S1, 20% shading; S2, 35% shading; S3, 50% shading; S4, 60% shading; T1, 30 d of shading; T2, 40 d of shading; T3, 50 d of shading; Fv/Fm, the maximum photochemical rate; YII, the actual photochemical efficiency; NPQ, the non-photochemical quenching; and qP, the photochemical quenching coefficient. Data are mean $\pm$ SD (n = 3).

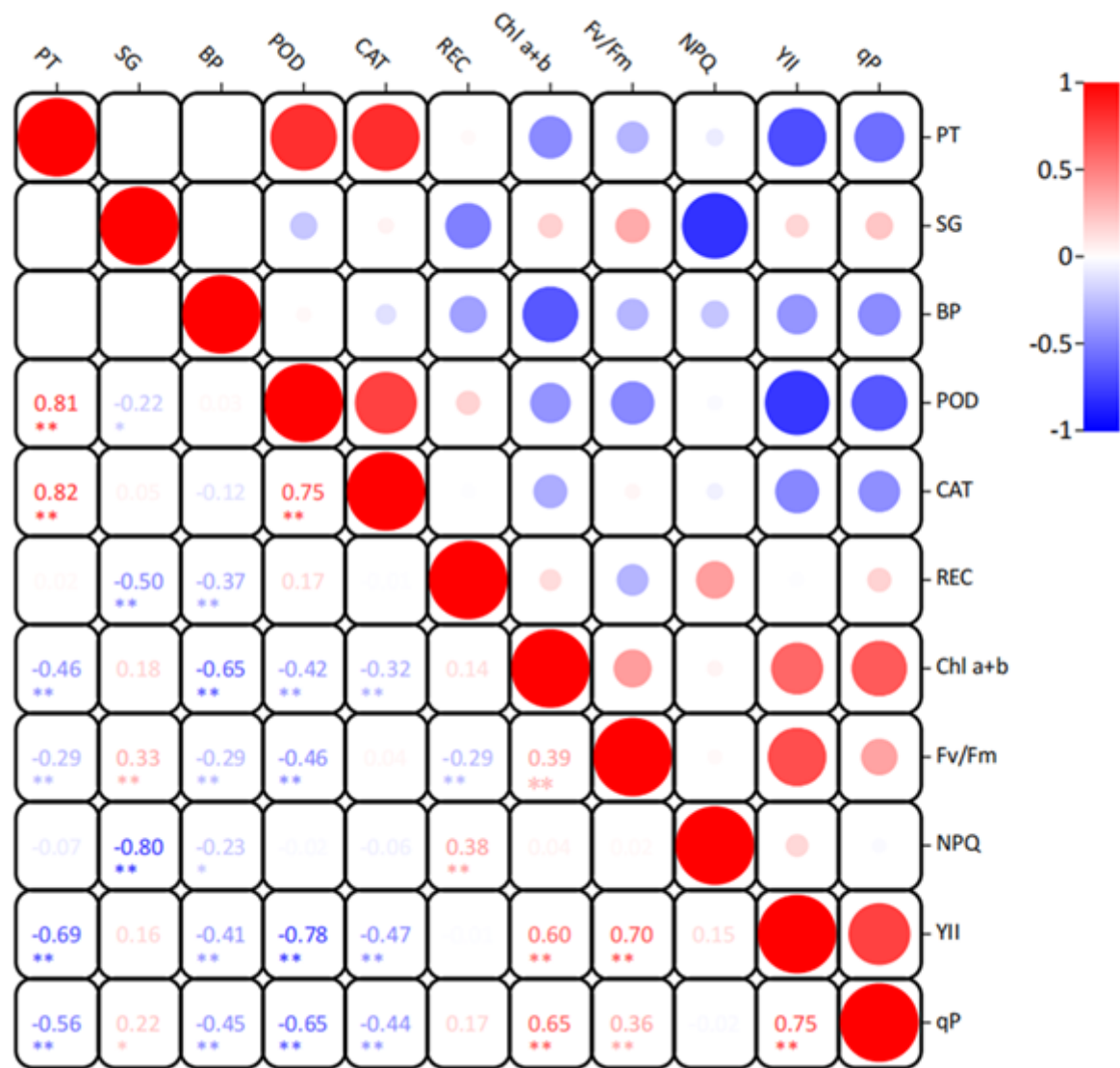


Figure 6

Correlation analysis of physiological indexes under different treatments. Data are mean $\pm$ SD (n = 3). Asterisks indicate significant differences within each column (Duncan's multiple range test, * p 0.05, ** p < 0.01). respectively using Pearson correlation coefficients. red ** represents positive correlation of 99%, blue ** represents negative correlation of 99%, red * represents positive correlation of 95%, blue * represents negative correlation of 95%, white represents non-significant correlation. PT, processing time; SG, shading gradient; BP, blade position; POD, peroxidase; CAT, catalase; REC, relative electrical conductivity; Chl a+b, total chlorophyll a+b; Fv/Fm, maximum quantum yield of PSII photochemistry;

NPQ, non-photochemical quenching of maximal fluorescence; qP, photochemical quenching of variable fluorescence; YII, primary light energy capture efficiency.

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