DOI: 10.32615/ps.2019.146 PHOTOSYNTHETICA 58 (SI): 323-330, 2019

Special issue in honour of Prof. Reto J. Strasser

Chlorophyll *a* fluorescence of *Bertholletia excelsa* Bonpl. plantations under thinning, liming, and phosphorus fertilization

K.C. PIRES DA COSTA*, R. KIRMAYR JAQUETTI**, and J.F. DE CARVALHO GONÇALVES**,+

*Faculty of Agronomy, Institute of Studies in Agrarian and Regional Development (IEDAR), Federal University of South and Southeast of Pará (UNIFESSPA), Folha 31, Quadra 07, Nova Marabá, 68507-590 Marabá, PA, Brazil**
*Laboratory of Plant Physiology and Biochemistry, National Institute for Amazonian Research (INPA), André Araújo Ave. 2936, Aleixo, 69011-970 Manaus, AM, Brazil***

Abstract

Phosphorus (P) fertilization and liming can reduce negative effects caused by a sudden increase in light availability after thinning of forest plantations. In this study, immediately after thinning, photochemical performance (quantum yield of PSII and performance index) decreased, however, liming and P fertilization reduced the negative effects caused by thinning and accelerated the recovery of *Bertholletia excelsa* trees. After thinning, the remaining trees increased J-I phase with the formation of a positive K-band which occurred exclusively in unfertilized (no liming or no P) individuals. Additionally, enhanced effective dissipation (DI_0/RC) values (140%) were found in individuals without liming or P after thinning, while plants under liming and P increased DI_0/RC values by 47 and 79%, respectively. Photochemical performance of *B. excelsa* was positively correlated to iron, zinc, and manganese. Thus, P fertilization and liming are recommended for alleviating the stress caused by high irradiance after thinning in *B. excelsa* plantations.

Additional key words: Brazil nut tree; photosynthetic pigments; plant nutrition; silvicultural practices; stress physiology.

Introduction

Silvicultural practices are important for the management of planted forests and such treatments have been recommended for increasing plantation productivity (Forrester *et al.* 2013). In order to increase the primary production of planted forests, practices, which improve tree photosynthesis, are extremely useful since they increase carbon assimilation. But some forestry practices such as thinning may initially have negative effects and decrease biomass gain as observed in a boreal forest in which the leaf area index decreases from 8 to 6 (Vesala *et al.* 2005). In addition, recent study has shown that after thinning, the remaining trees may reduce their photochemical performance [quantum yield of PSII (F_V/F_M) and performance index (PI_{ABS})] as a response to excess energy, thus enhancing photoinhibition, which may decrease the net photosynthetic rates (P_N) and consequently plantation productivity (Wu *et al.* 2018). So, some questions emerge, for example: Can other silvicultural practices mitigate or decrease the impact of thinning on photosynthesis?

Photosynthesis is determined by chlorophyll (Chl) content and the flux of energy in leaves. Plants should adjust Chl (*a*+*b*) content and Chl *a*/*b* ratio to adapt to certain environmental conditions and optimize photosynthesis under high and low irradiance (Ma *et al.* 2015). Liming (material contains calcium and/or magnesium compounds capable of neutralizing soil acidity) and P fertilization treatments, just as in other silvicultural treatments, in turn, can reduce the high-light stress effects on chloroplast pigments and stimulate photosynthesis in high light plasticity plants (Filstrup and Downing 2017, Lopes *et al.* 2019) after thinning, due to the enhancement of the nutrient uptake of trees and the availability of nutrients in soil, mainly calcium (Ca), magnesium (Mg), and P. On the other hand, under conditions, where light is limited, liming can increase leaf Chl (due to the increase of Mg concentrations in leaves and P addition) in order to favor

---

Received 21 September 2019, *accepted* 5 November 2019.

+Corresponding author; phone: +55-92-3643-1938, e-mail: jfc@inpa.gov.br

Abbreviations: Chl – chlorophyll; OJIP – fast chlorophyll fluorescence transients; DI_0/RC – dissipated energy flux per reaction center of PSII; ET_0/RC – electron transport of an active RC; TR_0/RC – maximum trapping rate per RC.

Acknowledgements: We are grateful to the National Institute for Amazonian Research (MCTIC-INPA) and the National Council for Scientific and Technological Development (CNPq, Brazil) for financial support of our study on tree physiology. This study was also supported by funding from Coordination for the Improvement of Higher Level Personnel (CAPES, Brazil), project Pro-Amazonia (grant no. 52) and Amazonas State Research Support Foundation (FAPEAM, Brazil). We also thank Empresa Agropecuária Aruanã S.A. for providing logistical support.

increased light energy flux and use (Filstrup and Downing 2017).

When a Chl molecule absorbs light, approximately 80% of the absorbed light is used in photosynthesis, but under stress conditions, changes in the dynamics of energy flow in leaves may occur, which may alter the shape of the OJIP curve (Strasser *et al.* 2004). Plants in high light environments may increase initial fluorescence (F_0) and reduce all the induction phases (O-J, J-I, and I-P), and show a much flatter curve (Kalaji *et al.* 2012), which may result in the formation of a positive K-band in O-J step, indicating more severe stress. The effects of P deficiency on OJIP curvature seem contradictory and nonspecific, but it was reported that under low P, the curvature of the OJIP transient is affected at around the I-step at 20 to 50 ms that gradually straightens and might be absent (Carstensen *et al.* 2018a).

Changes in the curvature of OJIP transient reflect the alteration in the energy flux in leaves. Plants subjected to stressful conditions may increase effective dissipation (DI_0) of an active reaction center (RC) as a strategy to reduce excessive absorbed energy and enhance protective quenching of photosynthetic apparatus, thus they may also reduce other steps of the energy flux, such as absorption (ABS), trapped energy (TR_0), and electron transport (ET_0) (Strasser *et al.* 2004, dos Santos Junior *et al.* 2015). The increase in nonphotochemical dissipation, in general, results in the decrease of F_V/F_M and a decrease in PI_{ABS} (Strasser *et al.* 2004, Kalaji *et al.* 2012, Stirbet and Govindjee 2012).

The effects of thinning, liming, and P fertilization on photochemical performance are associated with the photosynthetic capacity of plants, since it is a stress characteristic, and also demonstrates the species ability to acclimate to new environmental conditions. In this context, *Bertholletia excelsa* Bonpl. (Brazil nut tree) is a singular species and studies have indicated the capacity of this species to tolerate and recover from stress conditions (Ferreira *et al.* 2009, Lopes *et al.* 2019, Schimpl *et al.* 2019). But nowadays, it has been a common theme of discussion in the literature that fertilization can increase stress tolerance, and that plant's ability to adapt to stressful environments can be detected and monitored using the Chl *a* fluorescence (Kalaji *et al.* 2017, Tariq *et al.* 2017).

B. excelsa is a highly valuable commercial Amazonian tree species that has occupied prominent position in the silvicultural scenario in this region (Scoles *et al.* 2011, 2014; Ferreira *et al.* 2015, Schroth *et al.* 2015). In a *B. excelsa* forest plantation for wood production, we evaluated the effects of thinning, liming, and P fertilization on photosynthetic pigments and Chl *a* fluorescence (ChlF). The two main hypotheses were: (*1*) thinning may affect photochemical efficiency of individuals and (*2*) liming and P fertilization may alleviate the effects of high light stress by causing enhanced light energy uptake and usage.

Materials and methods

Study site, thinning, and fertilization treatments: The *Bertholletia excelsa* Bonpl. (Brazil nut tree) plantation studied was located in Itacoatiara, Amazonas, Brazil (3°0'30.63"S, 58°50'1.50"E), which is 120 to 170 m a.s.l., has a mean annual temperature of 31.2°C, and an annual rainfall of 2,539.6 mm (INMET 2018). Two well-defined seasons exist; the rainy season with precipitation of > 300 mm from January to April and the dry season with precipitation of < 100 mm from August to September. The dominant soil type is ferrasol. The plantation was established in 2000, on an area of 2.69 hectares. Seven-month-old seedlings with 45 cm of height were used; the spacing between seedlings was 2.5 × 1.5 m, resulting in a stocking density of 2,666 trees per hectare. Seedlings were not fertilized.

A randomized complete block design was established in this study. The number of blocks was defined from the variable diameter (measured at 1.3 m above ground), obtained by a preliminary pilot inventory. We used the sample size for the infinite population equation in order to determine how many sample units were needed to have a degree of confidence of 95% and 10% precision. Thus, eight blocks (1,102.5 m^2) were established and in each block, the six treatments were randomly applied: (*1*) control; (*2*) liming; (*3*) liming + phosphorus; (*4*) thinning; (*5*) thinning + liming; and (*6*) thinning + liming + phosphorus. The treatments plots were separated by a buffer zone of approximately 4 m. Thus, the measurements were performed in a plot of 12.5 m × 7.5 m (93.75 m^2) with 25 trees per plot (a total of 1,200 trees).

Liming treatments were 0 and 2,000 kg ha^{-1}, applied to the soil in the high precipitation season (14 March to 1 April 2016), with applications of 824 g of limestone (relative power of total neutralization of 91%) per tree. Phosphorus treatments were 0 and 150 kg ha^{-1} of triple superphosphate (46% of P_2O_5), applied 60 d after the liming was assigned, and when the soil pH increased to 5.5, then another 122 g per tree were applied. The estimate for the liming necessity was performed based on the methodology proposed by Batista (2014), while the requirement for P_2O_5 was extrapolated from suggestions for *Eucalyptus* ssp. (CFSEMG 1999).

Liming and P were applied at a distance of 50 cm from the stem base and 20-cm depth from the soil surface, forming a circle around the plant (soil amendment circle). The distance from stem base, depth of liming, and P application was defined according to studies regarding root distribution, which showed that 70% of the total root biomass of adult trees are in 0–20 cm depth from the soil surface and 61% are at a distance of 50 cm from the stem base (Sudmeyer *et al.* 2004). Thus, all liming and P treatments were applied within the influence of the root absorption zones.

After the P fertilizer was applied, a low level thinning of high intensity was performed, where 50% of the basal area was removed from the stand and suppressed, tortuous, and forked trees were eliminated. The thinning intensity was defined using the recommendations made by Costa (2015).

After application of the treatments, thinning increased the light availability from 51 to 423 μmol(photon) m^{-2} s^{-1} and, from rainy to dry season, the daily PAR in unthinned plots increased by 70%, while in thinned plots the increase was 200% (Fig. 1*A*). However, the treatments applied did not change the availability of water in the soil, but we observed a progressive reduction of soil moisture from rainy to dry season (Fig. 1*B*). Sixty days after lime application, the soil pH increased from 4.3 to 5.8, and 30 d after P application, the P in soil increased from 2.1 to 88.2 mg g^{-1} (Fig. 1*C*,*D*). Values of pH and P remained greater in these plots throughout the experimental period (Fig. 1*C*,*D*).

Data collection and tree sampling: Data collection was performed between 2016–2017, during historical peaks of high precipitation (precipitation > 300 mm) and low precipitation (precipitation < 100 mm), as well as in the transition between these seasons (Fig. 2). Due to the impossibility of taking measurements from all the trees of the experiment (1,200 trees in total), three sample trees were selected in each plot for data collection. The criteria for selected trees were representativeness in diameter, height, and crown form in each plot.

Nutrients: Five healthy and fully expanded leaves were taken from the middle third of the sample trees. The leaf samples were dried in an oven at 65°C for mass constant evaluation. The total N was determined by the Kjeldahl method (Bremner and Mulvaney 1982). Macronutrients (Ca, Mg, P, and K) and micronutrients (Fe, Zn, Cu, and Mn) were extracted with a 3:1 nitric-perchloric solution, the concentrations of these nutrients were determined by atomic absorption spectrometry (*Perkin-Elmer 1100B*, Uberlingen, Germany) and P was determined by spectrophotometry (UV/VIS spectrophotometer *Ultrospec 2100 pro*, *Amersham Biosciences*) at 725 nm (Miyazawa *et al.* 1999).

Photosynthetic pigments: Chl contents were estimated from samples with 0.1 g of healthy and fully expanded leaves (fresh material) collected between 7:00–9:00 h and taken from the middle third of the sample trees. The pigments were extracted with 10 ml of 80% (v/v) acetone and 0.5% (w/v) magnesium carbonate ($MgCO_3$). The suspension was filtered (filter *Quanty JP42*) and the absorbance was determined using a spectrophotometer (*Ultrospec 2100 pro*, *Armesham Biosciences*) with wavelengths set at 663 nm (Chl *a*), 645 nm (Chl *b*), and

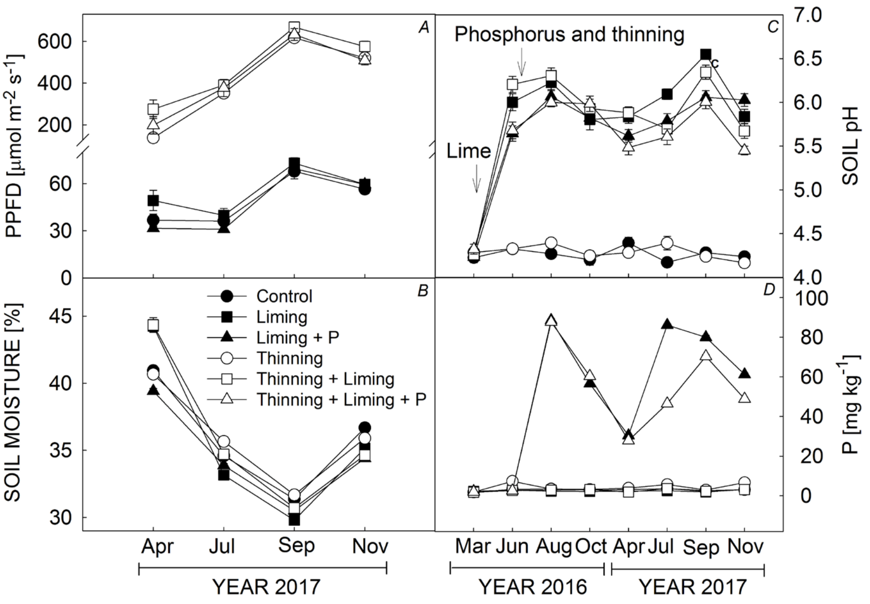


Fig. 1. Changes in light (*A*), soil moisture (*B*), soil pH (*C*), and soil phosphorus (*D*) in *Bertholletia excelsa* plantations under thinning, liming, and phosphorus fertilization.

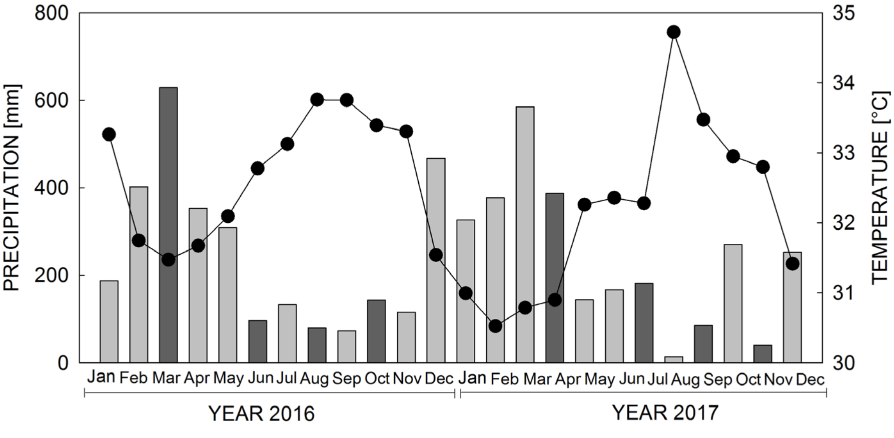


Fig. 2. Precipitation (*bars*) and temperature (*line*) over the period in which the data were collected in the field (2016–2017). *Dark grey bars* indicate the months in which data were collected.

total chlorophyll Chl_{total} = Chl (*a*+*b*) (Arnon 1949). The calculations for the mass (μmol g^{-1}) of Chl pigments were obtained using the equations of Hendry and Price (1993).

Chl *a* fluorescence and JIP-test: The Chl *a* fluorescence induction curve was measured using a *Handy-PEA* fluorimeter (*Hansatech Instruments Ltd.*, UK). The data were collected between 9:00–12:00 h in healthy and fully expanded leaves that were taken from the middle third of the sample trees. The selected leaves were subjected to a 30-min period of adaptation to darkness. Immediately after the dark-adaptation period, the leaves were exposed to a pulse of saturated light at an intensity of 3,000 μmol(photon) m^{-2} s^{-1} (peak at 650 nm) for 1 s, thus, fast fluorescence transients were obtained and were analyzed by the so-called 'JIP-test' (Strasser *et al.* 2004).

Data analysis: A linear mixed model was performed to evaluate the effects of treatments and seasonality on leaf pigment content and Chl *a* fluorescence. Block and plots were included in the models as nested random effects in order to control potential spatial autocorrelation. *Pearson*'s product-moment correlations were used to assess the influences of nutrients (mass basis) on Chl *a* fluorescence.

Results

Phosphorus and liming did not change the Chl and Chl *a*/*b* ratio in leaves of unthinned trees in the plots (Fig. 3), but thinning decreased the Chl in leaves. P and liming minimized these reductions and significant differences were not observed for the Chl and Chl *a*/*b* ratio between trees from thinned plots that received liming and P, when compared to the trees from unthinned plots (Fig. 3*A*). Accentuated differences in Chl and Chl *a*/*b* ratio between treatments were observed during the rainy–dry transition (July 2017) (Fig. 3). In this period, the trees in unthinned plots showed the Chl content 60% higher and the Chl *a*/*b* ratio 25% lower than that in trees of thinned plots, regardless of liming or P fertilization (Fig. 3).

Liming and P fertilization did not influence the F_V/F_M ratio and PI_{ABS} of *B. excelsa* trees of unthinned plots (Fig. 4). However, immediately after thinning, the F_V/F_M and PI_{ABS} decreased; liming and P fertilization helped minimize the stress caused by thinning and accelerated the recuperation (Fig. 4). After thinning, the reductions of F_V/F_M in trees under thinning + liming treatments were 3% and thinning + liming + phosphorus were 7%, while, in trees without liming or P, the reductions were in the order of 12% (Fig. 4*A*). Seasonality only influenced trees under thinning without liming or P fertilization. In these trees, we found a decrease in F_V/F_M from 0.82 to 0.80 (for rainy and dry season, respectively) (Fig. 4*A*).

In order to understand factors associated with stress and recuperation of *B. excelsa* after thinning, the OJIP transients were measured during the most severe stress stage (June 2016) and after the complete recovery from stress in all treatment regimes (April 2017). During the period of the most severe stress, we observed a decrease in the I-P phase in all trees under thinning, irrespective of liming or P. The increase in the J-I phase and positive K-band formation was also observed, but only in trees under thinning without liming or P (Fig. 5*A*,*B*). After recuperation from stress, no differences were found between treatments for OJIP transients and the formation of positive K-band for treatments was not observed (Fig. 5*C*,*D*).

Additionally, we found that during stress conditions, after thinning, there was an increase of 140% in photochemical dissipation (DI_0/RC) in trees without liming or P, though the increase for trees under liming was only 47% and under liming + P it was only 79% (Table 1). We also found that stress conditions caused by thinning increased the absorption flux per PSII reaction center (ABS/RC), trapped energy flux per PSII reaction center (TR_0/RC), nonphotochemical dissipation per PSII reaction center (DI_0/ABS), but no differences were found for electron transport flux from Q_A to Q_B per PSII (ET_0/RC).

During investigation of the effects of nutrients on photochemical performance of *B. excelsa*, we found positive effects for Fe and Zn micronutrients, on mass basis, on F_V/F_M, and PI_{ABS} (Fig. 6). Macronutrients (N, P,

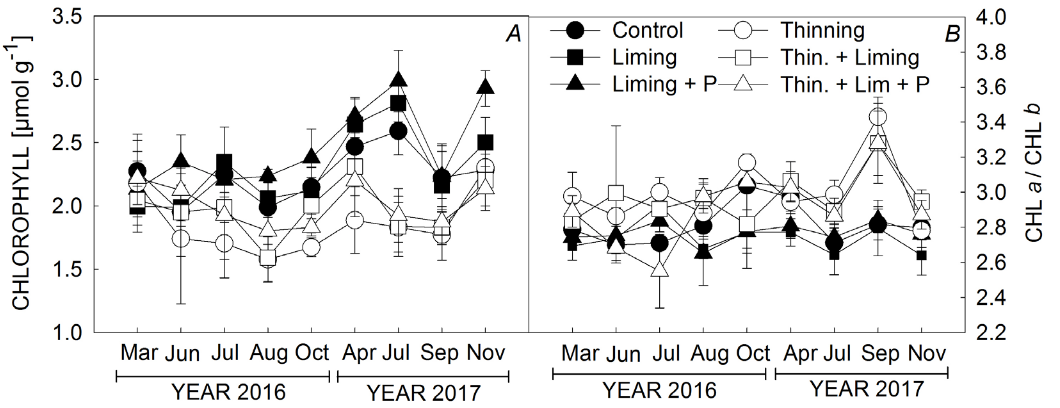


Fig. 3. Seasonal content of chlorophyll (*A*) and chlorophyll *a*/*b* ratio (*B*) in leaves of *Bertholletia excelsa* trees under thinning, liming, and phosphorus fertilization. Values are means ± SE, *n* = 8.

K, Ca) were not significantly correlated to F_V/F_M, but N and P positively influenced PI_{ABS} (Fig. 6).

Discussion

We found that thinning decreased Chl and the Chl *a*/*b* ratio, but application of P and liming minimized these reductions. The reduction of Chl concentrations in plants after thinning probably occurred due to high incident light on thinned trees. The degradation of Chl under these conditions is due to the formation of reactive oxygen species (ROS) which are responsible for a faster degradation of Chl (Croft and Chen 2017). In addition, the reduction of Chl content suggests a reduction in the light-harvesting complex, which may prevent the absorption of excessive light energy and thus avoid oxidative damage.

Chl *b* is mainly present in the light-harvesting complexes of PSI and PSII, however, Chl *a* is present in both the light-harvesting complexes and also in the reaction centers of the photosystems. Leaves under high irradiance tend to degrade the light-harvesting complexes in order to avoid damage to the photosynthetic apparatus due to the excess irradiance and tend to increase the number of PSII in order to increase the photosynthetic capacity of the plants (Živčák *et al.* 2014). The increase in ABS/RC values immediately after thinning indicates an increasing amount of the absorbing Chl *a* molecules per active RC. In this case, this response of ABS/RC after thinning may reflect a strategy that *B. excelsa* possesses for increasing the use of available energy.

The greater availability of light associated to the increase of ABS/RC and TR_0/RC, did not change ET_0/RC. The increase in TR_0/RC and the consistency in ET_0/RC indicate that the electron transport from pheophytin (Pheo) to Q_A (reduction of Q_A to Q_A^-) during photosynthesis in the leaves of *B. excelsa* increased, while the rates for transporting electrons were minimally affected by the sudden increase in light availability. The increase in TR_0/RC

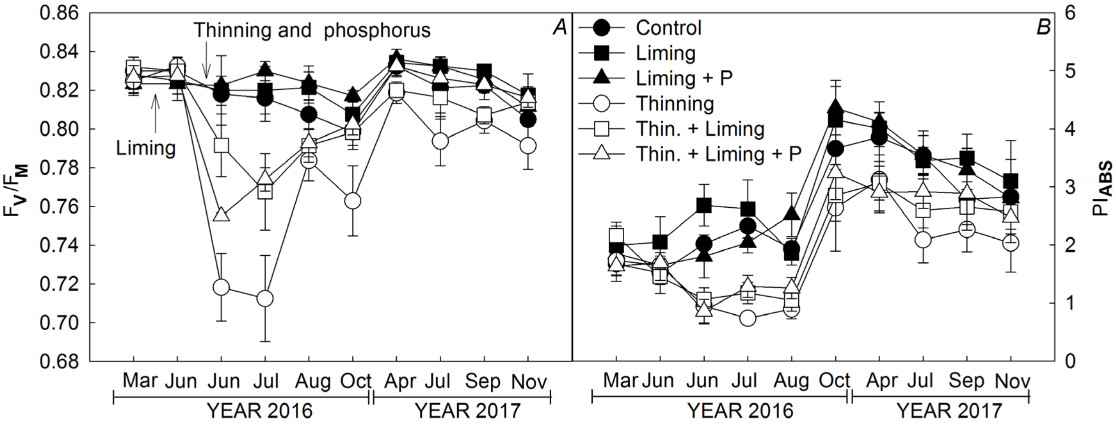


Fig. 4. Seasonal variation of quantum yield of PSII (F_V/F_M) (*A*) and performance index (PI_{ABS}) (*B*) of *Bertholletia excelsa* plantations under thinning, liming, and phosphorus fertilization. Values are means ± SE, *n* = 8.

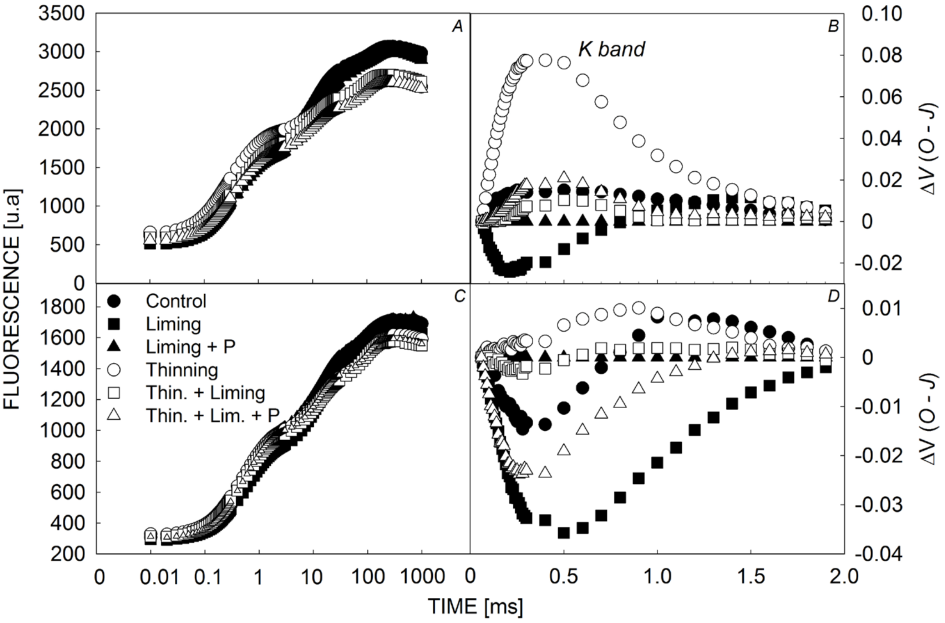
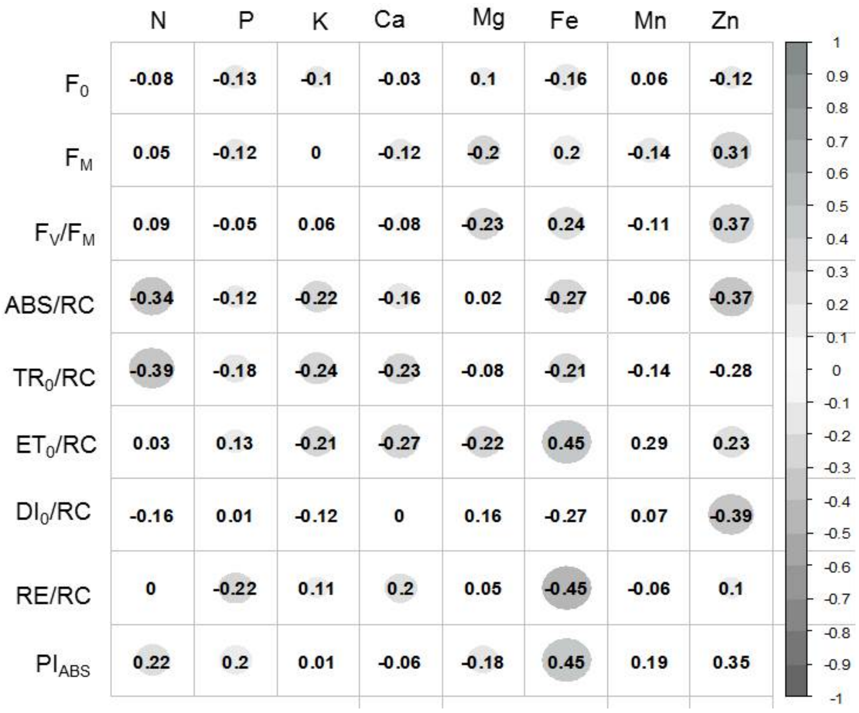


Fig. 5. OJIP transient and K-band performance under stress after thinning (*A*,*B*) and after recuperation (*C*,*D*) of *Bertholletia excelsa* under thinning, liming, and phosphorus fertilization.

Table 1. Specific flux of energy per reaction center (RC) in leaves of *Bertholletia excelsa* plantations under thinning, liming, and phosphorus fertilization. Inicial fluorescence (F_0), maximum fluorescence (F_M), effective antenna size expressed as absorbance per reaction center (ABS/RC), amount of energy dissipated per active reaction center (DI_0/RC), trapping efficiency per reaction center (TR_0/RC), electron transport per reaction center (ET_0/RC).

Fluorescence parameters		Liming	Liming + P	Thinning	Thinning + liming	Thinning + liming + P	R^2	*p*
F_0	Stress	6.45	–0.57	36.56	7.56	10.75	0.22	0.05
	Recupered	–3.47	–1.09	1.46	–5.49	–5.22	0.01	0.50
F_M	Stress	8.11	1.32	–9.13	–4.49	–14.3	0.26	0.03
	Recupered	–1.79	1.16	–5.23	–10.74	–5.21	0.09	0.11
ABS/RC	Stress	–5.16	10.64	39.66	22.74	28.9	0.42	0.00
	Recupered	–6.85	–0.82	1.84	–0.86	–5.51	0.05	0.75
DI_0/RC	Stress	–5.61	9.69	140.65	47.11	78.57	0.38	0.00
	Recupered	–8.86	–2.11	10.97	5.61	–4.22	0.01	0.40
TR_0/RC	Stress	–5.19	10.68	18.29	17.66	18.51	0.38	0.00
	Recupered	–6.45	–0.49	0.33	–1.99	–5.61	0.06	0.79
ET_0/RC	Stress	2.2	4.63	0.95	–2.35	–8.87	0.01	0.76
	Recupered	–9.13	3.17	–8.08	–11.77	–19.77	0.20	0.01



	N	P	K	Ca	Mg	Fe	Mn	Zn
F_0	-0.08	-0.13	-0.1	-0.03	0.1	-0.16	0.06	-0.12
F_M	0.05	-0.12	0	-0.12	-0.2	0.2	-0.14	0.31
F_V/F_M	0.09	-0.05	0.06	-0.08	-0.23	0.24	-0.11	0.37
ABS/RC	-0.34	-0.12	-0.22	-0.16	0.02	-0.27	-0.06	-0.37
TR_0/RC	-0.39	-0.18	-0.24	-0.23	-0.08	-0.21	-0.14	-0.28
ET_0/RC	0.03	0.13	-0.21	-0.27	-0.22	0.45	0.29	0.23
DI_0/RC	-0.16	0.01	-0.12	0	0.16	-0.27	0.07	-0.39
RE/RC	0	-0.22	0.11	0.2	0.05	-0.45	-0.06	0.1
PI_{ABS}	0.22	0.2	0.01	-0.06	-0.18	0.45	0.19	0.35

Fig. 6. Correlations between nutrients on mass basis and specific flux of energy per reaction center (RC) of *Bertholletia excelsa* plantations under thinning, liming, and phosphorus fertilization. Values highlighted in gray are significant correlations. Inicial fluorescence (F_0), maximum fluorescence (F_M), quantum yield of PSII (F_V/F_M), effective antenna size expressed as absorbance per reaction center (ABS/RC), trapping efficiency per reaction center (TR_0/RC), electron transport per reaction center (ET_0/RC), amount of energy dissipated per active reaction center (DI_0/RC), flux reduction of end-electron acceptors (RE/RC), and performance index (PI_{ABS}).

and maintenance of ET_0/RC in trees of *B. excelsa* seems to be associated with the Fe concentration in leaves. Typically, approximately 80% of Fe is found in photosynthetic apparatus, where it is essential for the biosynthesis of cytochromes and other heme molecules, including Chl, the electron transport system, and the construction of Fe-S clusters. It is also an active cofactor of many enzymes that are necessary for synthesis of plant hormones (Rout and Sahoo 2015).

The increase in light availability after thinning also resulted in an enhancement in effective dissipation of an active RC (DI_0/RC). The increase in DI_0/RC values in plants submitted to high-light environments is common and usually represents a photoprotective mechanism of the photosynthetic apparatus to counter photoinhibition damage (Derks *et al.* 2015, Gururani *et al.* 2015). Thus, the increase in DI_0/RC after thinning may be the main factor that led the remaining trees to reduce F_V/F_M ratio from 0.83 to 0.72 and the PI_{ABS} from 1.7 to 0.8. This association between DI_0/RC and photochemical efficiency (F_V/F_M and PI_{ABS}) has been previously demonstrated by Boisvert *et al.* (2006) and Souza *et al.* (2010). However, our results indicate that liming and P fertilization alleviate the effects of high light stress after thinning.

Pi deficiency renders the light income excessive for their actual capacity to process it, thereby leading to photoinhibition. ATP production is affected under Pi deficiency. The photophosphorylation of ADP is impaired under Pi limitation due to low availability of Pi in the stroma, therefore causing limited ribulose 1,5-bisphosphate (RuBP) regeneration. Furthermore, decreased utilization of NADPH and reduced ferredoxin due to decreased activity of the Calvin cycle, together with enhanced acidification of the lumen pH due to restrained ATP synthesis, lead to the accumulation of reduced electron transporters such as plastoquinone or ferredoxin. As a consequence of the feedback inhibition in photosynthesis, Pi deficiency causes Chl loss and a decrease in F_V/F_M, which are symptoms of photooxidative stress (Hernández and Munné-Bosch 2015).

In addition, we found increases in the J-I phase and positive K-band formation only in trees under thinning without liming or P. Increases in the J-I phase and positive K-band formation are considered indicative of stress in plants (Frydenvang *et al.* 2015, Luo *et al.* 2016). Changes in the J-I phase have been associated with responses of plants under low phosphorus availability, and K-band formation is normally observed in plants subjected to stress factors such as increased temperature (Sengupta *et al.* 2013, Carstensen *et al.* 2018a,b). However, plants under other stress factors, such as high irradiance, water and nutrient limitation, are also associated with alteration of the shape of the J-I phase and positive K-band formation (Cendrero-Mateo *et al.* 2015).

The increase in the J-I phase can occur due to the reduction of ATP synthase and therefore accumulate protons in the thylakoid lumen which causes acidification, and this can decrease the oxidation of the plastoquinone pool (PQ) at the cytochrome b_6f complex in the electron transport chain. The decreased oxidation changes the flow of electrons towards PSI, which is reflected by the shape of the J-I phase (Carstensen *et al.* 2018a,b). The presence of the positive K-band, in turn, indicates an imbalance between the donation of electrons from the oxygen-evolving complex (OEC) to the oxidized PSII reaction center chlorophyll (P_{680}^+) and the reoxidation of reduced PSII acceptors (Q_A^-). This is usually due to the impaired OEC, which is very sensitive to suboptimal conditions (Strasser 1997).

Conclusions: Thinning reduces the photochemical efficiency of *B. excelsa*, but liming and phosphate fertilization alleviate the effects of high irradiance stress and accelerate tree recovery. Thus, phosphorus fertilization and liming are recommended for attenuating the stress caused by high irradiance after thinning in *B. excelsa* trees because they increase plant tolerance to stress in this species when established on plantation conditions in central Amazonia.

References

Arnon D.I.: Copper enzymes in isolated chloroplasts: Polyphenoloxidase in *Beta vulgaris*. – Plant Physiol. **24**: 1-15, 1949.

Batista I.M.P.: [Liming recommendation for some soils of the State of Amazonas.] Pp. 46. Federal University of Amazonas, Manaus 2014. [In Portuguese]

Boisvert S., Joly D., Carpentier R.: Quantitative analysis of the experimental O-J-I-P chlorophyll fluorescence induction kinetics. Apparent activation energy and origin of each kinetic step. – FEBS J. **273**: 4770-4777, 2006.

Bremner J.M., Mulvaney C.S.: Nitrogen – Total. – In: Page A.L., Miller R.H., Keeney D.R. (ed.): Methods of Soil Analysis. Part 2. Chemical and Microbiological Properties. Pp. 595-624. American Society of Agronomy, Madison 1982.

Carstensen A., Herdean A., Schmidt S.B. *et al.*: The impacts of phosphorus deficiency on the photosynthetic electron transport chain. – Plant Physiol. **177**: 271-284, 2018b.

Carstensen A., Szameitat A.E., Frydenvang J., Husted S.: Chlorophyll *a* fluorescence analysis can detect phosphorus deficiency under field conditions and is an effective tool to prevent grain yield reductions in spring barley (*Hordeum vulgare* L.). – Plant Soil **434**: 79-91, 2018a.

Cendrero-Mateo M.P., Carmo-Silva C.A., Porcar-Castell A. *et al.*: Dynamic response of plant chlorophyll fluorescence to light, water and nutrient availability. – Funct. Plant Biol. **42**: 746-757, 2015.

CFSEMG: [Recommendations for the use of concealers and fertilizers in Minas Gerais.] Pp. 359. SBCS, Viçosa 1999. [In Portuguese]

Costa K.C.P. [Ecophysiological characteristics and growth of *Bertholletia excelsa* Bonpl. in forest planting submitted to thinning.] Pp. 49. National Institute of Amazonian Research, Manaus 2015. [In Portuguese]

Croft H., Chen J.M.: Leaf Pigment Content. Pp. 117-142. Elsevier, Toronto 2017.

Derks A., Schaven K., Bruce D.: Diverse mechanisms for photoprotection in photosynthesis. Dynamic regulation of photosystem II excitation in response to rapid environmental change. – BBA-Bioenergetics **1847**: 468-485, 2015.

dos Santos Junior U.M., Gonçalves J.F.C., Strasser R.J., Fearnside P.M.: Flooding of tropical forests in central Amazonia: What do the effects on the photosynthetic apparatus of trees tell us about species suitability for reforestation in extreme environments created by hydroelectric dams? – Acta Physiol. Plant. **37**: 166, 2015.

Ferreira M.J., Gonçalves J.F.C., Ferraz J.B.S.: Photosynthetic parameters of young Brazil nut (*Bertholletia excelsa* H. B.) plants subjected to fertilization in a degraded area in Central Amazonia. – Photosynthetica **47**: 616-620, 2009.

Ferreira M.J., Gonçạlves J.F.C., Ferraz J.B.S., Correâ V.M.: [Nutritional characteristics of young plants of *Bertholletia excelsa* Bonpl. under fertilization treatments in a degraded area in the Amazon.] – Sci. For. **43**: 863-872, 2015. [In Portuguese]

Filstrup C.T., Downing J.A.: Relationship of chlorophyll to phosphorus and nitrogen in nutrient-rich lakes. – Inland Waters **7**: 385-400, 2017.

Forrester D.I., Elms S.R., Baker T.G.: Relative, but not absolute, thinning responses decline with increasing thinning age in a *Eucalyptus nitens* plantation. – Aust. Forestry **76**: 121-127, 2013.

Frydenvang J., van Maarschalkerweerd M., Carstensen A. *et al*.: Sensitive detection of phosphorus deficiency in plants using chlorophyll *a* fluorescence. – Plant Physiol. **169**: 353-360, 2015.

Gururani A.M., Venkatesh J., Tran L.-S.P.: Regulation of photosynthesis during abiotic stress-induced photoinhibition. – Mol. Plant **8**: 1304-1320, 2015.

Hendry G.A.S., Price A.H.: Stress indicators: Chlorophylls and carotenoids. – In: Hendry G.A.S., Grime J.P. (ed.): Methods in Comparative Plant Ecology. Pp. 148-152. Chapman & Hall, London 1993.

Hernández I., Munné-Bosch S.: Linking phosphorus availability with photo-oxidative stress in plants. – J. Exp. Bot. **66**: 2889-2900, 2015.

INMET.: National Institute of Meteorology. *Available at*: http://www.inmet.gov.br/portal/. (Accessed on 23 December 2018)

Kalaji H.M., Bąba W., Gediga K. *et al.*: Chlorophyll fluorescence as a tool for nutrient status identification in rapeseed plants. – Photosynth. Res. **136**: 329-343, 2017.

Kalaji H.M., Carpentier R., Allakhverdiev S.I., Bosa K.: Fluorescence parameters as early indicators of light stress in barley. – J. Photoch. Photobio. B **112**: 1-6, 2012.

Lopes J.D.S., Costa K.C.P., Fernandes V.S., Gonçalves J.F.C.: Functional traits associated to photosynthetic plasticity of young Brazil nut (*Bertholletia excelsa*) plants. – Flora **258**: 151446, 2019.

Luo H.-H., Tsimilli-Michael M., Zhang Y.-L., Zhang W.-F.: Combining gas exchange and chlorophyll *a* fluorescence measurements to analyze the photosynthetic activity of drip-irrigated cotton under different soil water deficits. – J. Integr. Agr. **15**: 1256-1266, 2016.

Ma X., Song L., Yu W. *et al*.: Growth, physiological, and biochemical responses of *Camptotheca acuminata* seedlings to different light environments. – Front. Plant Sci. **6**: 321, 2015.

Miyazawa M., Paiva M.A., Muraoka T. *et al*.: [Chemical Analysis Manual for Soils, Plants and Fertilizers.] Pp. 172-223. Embrapa Solos, Brasília 1999. [In Portuguese]

Rout G.R., Sahoo S.: Role of iron in plant growth and metabolism. – Rev. Agr. Sci. **3**: 1-24, 2015.

Schimpl F.C., Ferreira M.J., Jaquetti R.K. *et al*.: Physiological responses of young Brazil nut (*Bertholletia excelsa*) plants to drought stress and subsequent rewatering. – Flora **252**: 10-17, 2019.

Schroth G., Mota M.S., Elias M.E.A.: Growth and nutrient accumulation of Brazil nut trees (*Bertholletia excelsa*) in agroforestry at different fertilizer levels. – J. Forestry Res. **26**: 347-353, 2015.

Scoles R., Gribel R., Klein G.N.: [Growth and survival of Brazil nuts (*Bertholletia excelsa* Bonpl.), in different environmental conditions in region River Trombetas, Oriximiná, Pará, Brazil.] – Bol. Mus. Para. Emilio Goeldi. Cienc. Nat. **6**: 273-293, 2011. [In Portuguese]

Scoles R., Klein G.N., Gribel R.: [Performance and survival of Brazil nut tree (*Bertholletia excelsa* Bonpl., Lecythidaceae), in different light conditions after six years to planting, in Trombetas River region, Oriximiná, Pará, Brazil.] – Bol. Mus. Para. Emílio Goeldi. Cienc. Nat. **9**: 321-336, 2014. [In Portuguese]

Sengupta D., Guha A., Reddy A.R.: Interdependence of plant water status with photosynthetic performance and root defense responses in *Vigna radiata* (L.) Wilczek under progressive drought stress and recovery. – J. Photoch. Photobio. B **127**: 170-181, 2013.

Souza G.M., Sato A.M., Ribeiro R.V., Prado C.H.B.A.: Photosynthetic responses of four tropical tree species grown under gap and understorey conditions in a semi-deciduous forest. – Braz. J. Bot. **33**: 529-538, 2010.

Stirbet A., Govindjee: Chlorophyll *a* fluorescence induction: a personal perspective of the thermal phase, the J–I–P rise. – Photosynth. Res. **113**: 15-61, 2012.

Strasser B.J.: Donor side capacity of photosystem II probed by chlorophyll *a* fluorescence transients. – Photosynth. Res. **52**: 147-155, 1997.

Strasser R.J., Tsimilli-Michael M., Srivastava A.: Analysis of the chlorophyll *a* fluorescence transient. – In: Papageorgiou G.C., Govindjee (ed.): Chlorophyll *a* Fluorescence: A Signature of Photosynthesis. Advances in Photosynthesis and Respiration. Pp. 321-362. Springer, Dordrecht 2004.

Sudmeyer R.A., Speijers J., Nicholas B.D.: Root distribution of *Pinus pinaster*, *P. radiata*, *Eucalyptus globulus* and *E. kochii* and associated soil chemistry in agricultural land adjacent to tree lines. – Tree Physiol. **24**: 1333-1346, 2004.

Tariq A., Pan K., Olatunji O.A. *et al*.: Phosphorous application improves drought tolerance of *Phoebe zhennan*. – Front. Plant Sci. **8**: 1561, 2017.

Vesala T., Suni T., Rannik Ü. *et al*.: Effect of thinning on surface fluxes in a boreal forest. – Global Biogeochem. Cy. **19**: 303-311, 2005.

Wu J.W., Su Y., Wang J.H. *et al*.: Morphological and physiological acclimation of *Catalpa bungei* plantlets to different light conditions. – Photosynthetica **56**: 537-548, 2018.

Živčák M., Brestič M., Kalaji H.M., Govindjee: Photosynthetic responses of sun- and shade-grown barley leaves to high light: Is the lower PSII connectivity in shade leaves associated with protection against excess of light? – Photosynth. Res. **119**: 339-354, 2014.