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BORON DEFICIENCY AFFECTS GAS EXCHANGE AND PHOTOCHEMICAL EFFICIENCY (JPI TEST PARAMETERS) IN GREEN DWARF COCONUT

Leandro G. R. Pinho^a; Eliemar Campostrini^b; Pedro H. Monnerat^a; Alena Torres Netto^b; André Assis Pires^a; Cláudio Roberto Marciano^c; Yaska Janaína Bastos Soares^a

^a Mineral Plant Nutrition Sector, LFI, Agricultural Science and Technology Center, State University of North Fluminense, Campos dos Goytacazes, Brazil ^b Plant Physiology Laboratory, Agricultural Science and Technology Center, State University of North Fluminense, Campos dos Goytacazes, Brazil ^c Soil Physic Sector, LSOL, Agricultural Science and Technology Center, State University of North Fluminense, Campos dos Goytacazes, Brazil

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BORON DEFICIENCY AFFECTS GAS EXCHANGE AND PHOTOCHEMICAL EFFICIENCY (JPI TEST PARAMETERS) IN GREEN DWARF COCONUT

Leandro G. R. Pinho □ *Mineral Plant Nutrition Sector, LFI, Agricultural Science and Technology Center, State University of North Fluminense, Campos dos Goytacazes, Brazil*

Eliemar Campostrini □ *Plant Physiology Laboratory, Agricultural Science and Technology Center, State University of North Fluminense, Campos dos Goytacazes, Brazil*

Pedro H. Monnerat □ *Mineral Plant Nutrition Sector, LFI, Agricultural Science and Technology Center, State University of North Fluminense, Campos dos Goytacazes, Brazil*

Alena Torres Netto □ *Plant Physiology Laboratory, Agricultural Science and Technology Center, State University of North Fluminense, Campos dos Goytacazes, Brazil*

André Assis Pires □ *Mineral Plant Nutrition Sector, LFI, Agricultural Science and Technology Center, State University of North Fluminense, Campos dos Goytacazes, Brazil*

Cláudio Roberto Marciano □ *Soil Physic Sector, LSOL, Agricultural Science and Technology Center, State University of North Fluminense, Campos dos Goytacazes, Brazil*

Yaska Janaina Bastos Soares □ *Mineral Plant Nutrition Sector, LFI, Agricultural Science and Technology Center, State University of North Fluminense, Campos dos Goytacazes, Brazil*

□ *Boron (B) deficiency causes a wide array of symptoms, not only among species of palms, but also within a single species (i.e. Cocos nucifera). A better understanding of the effects of B deficiency in coconut will be important to try optimizing a rational fertilization management in coconut plants. Thus, modification of PSII photochemistry (using a group of fluorescence parameters, called the JIP-test, that quantify the stepwise flow of energy through Photosystem II) and gas-exchange in boron deficient green dwarf coconut plants were investigated. Our results suggest that a modification of PSII photochemistry (non-stomatic effects) and gas-exchange (stomatic effects) were induced by boron deficiency. Such modifications are manifested by (1) increase the ratio of total dissipation to the amount of active reaction centers (RCs) [dissipation (DI)/RC] and (2) leaf-to-air vapor pressure difference ($VPD_{leaf-air}$). These modifications (on PSII photochemistry and gas-exchange) were caused by a decrease in energy absorbed per excited cross-section [absorption flux (ABS)/cross section of the*

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Address correspondence to Eliemar Campostrini, Plant Physiology Laboratory, Agricultural Science and Technology Center, State University of North Fluminense, Av. Alberto Lamego, 2000, 28013-602, Campos dos Goytacazes, RJ, Brazil. E-mail: campost@uenf.br

sample (CS_0), density of active reaction centers (RC/CS), maximal trapping rate of an exciton that will lead to Q_A reduction measured over a cross-section of active and inactive RCs [trapping flux (TR)/ CS_0], electron transport per excited cross-section [electron transport flux (ET_0)/ CS], area above curve (proportional to the pool size of the electron acceptors Q_A on the reducing side of PSII), photosynthesis (A), stomatal conductance (g_s), transpiration (E), chlorophyll concentration (SPAD readings), growth parameters (root DW and height plant). Our results demonstrate that by analyzing fluorescence (JIP test parameters) derived from the polyphasic fluorescence transients measurements were able to estimate the functional changes of PSII in B deficient coconut plants. The results in this study suggest that fluorescence analysis (JIP test) and instantaneous measurements of gas-exchange can be useful tools in assessing the physiological effects of B deficiency in green dwarf coconut.

Keywords: boron, plant physiology, photosynthesis, tropical crops

INTRODUCTION

Boron (B) is a poorly understood micronutrient and at the same time B deficiency is the most widespread micronutrient deficiency affecting the productivity of many important crops (Dordas and Brown, 2005). Boron is considered to be involved in nucleic acid metabolism, carbohydrate and protein metabolism, indole acetic acid metabolism, cell wall synthesis, cell wall structure, membrane integrity and function, and phenol metabolism; however, molecular basis of these roles is mostly unknown (Loomis and Durst, 1992; Marschner, 1995; Goldbach, 1997; Goldbach et al., 2001). Boron cross-links pectins in cell walls, and this cross-linking was shown as essential for normal expansion of leaves.

The symptoms of B deficiency are a consequence of two important features of B physiology. First, the specific structural role of B in the cell wall growth (Kobayashi et al., 1996; O'Neill et al., 1996; Ishii and Matsunaga, 1996; Power and Woods, 1997) and second, the limited mobility of B in the majority of cultivated plant species (Hu et al., 1997). Boron deficiency also causes a wide array of physiological and biochemical changes including altered cell wall structure, altered membrane integrity and function, changes in enzyme activity, e.g., oxidation of phenolics by PPO in B-deficient leaves may lead to production of quinones. Quinones are known to be highly toxic and responsible for production of toxic O_2 species, e.g., hydrogen peroxide (H_2O_2), superoxide radical ($O_2^{\cdot-}$), and hydroxyl radical (OH^{\cdot}) (Shkol'nik et al., 1981; Cakmak and Römheld, 1997). These radicals are very toxic to the membrane proteins and lipids which cause disintegration and destabilization of the cellular membranes and results in membrane leakage.

Photosynthesis can be defined as the physico-chemical process by which photosynthetic organisms use light energy to drive the synthesis of organic

compounds. The photosynthetic process depends on a set of complex protein molecules that are located in and around a highly organized membrane. This process has an intense dependence on the membrane structure. A high degree of organization of the pigment–protein complexes is required for efficient utilization of the solar energy by photosynthetic organisms. Thus, if B deficiency can be associated with structure membrane damage, then a deficiency of this mineral nutrient will affect the photosynthesis process.

The effect of B nutrition on photosynthesis has rarely been studied (Cakmak and Römheld, 1997). There is no evidence for a direct role for B in photosynthesis (Shelp 1993). However, B deficiency reduces chlorophyll (Chl) and soluble protein contents of leaves, which results in an inhibition of Hill reaction and net photosynthetic rate (Sharma and Ramchandra, 1990). Han et al. (2008) showed that B-starved leaves showed decreased carbon dioxide (CO₂) assimilation and stomatal conductance, but increased intercellular CO₂ concentrations, and CO₂ assimilation may be feedback-regulated by the excessive accumulation of starch and hexoses in B-deficient leaves via direct interference with chloroplast function and/or indirect repression of photosynthetic enzymes.

In cotton, results indicated that B deficiency during vegetative growth significantly affected membrane integrity, decreased leaf CO₂ exchange rate and chlorophyll photosynthetic efficiency, suppressed plant growth and dry matter accumulation, and changed photo-assimilate partitioning among plant tissues and other physiological processes, resulting in increased fruit abscission (Zhao and Oosterhuis, 2003). In sunflower, B deficiency appreciably decreased photosynthetic oxygen evolution by leaves, the apparent quantum yield and quantum efficiency of photosystem two electron transport (Kastori et al., 1995). These authors related that the diminished rate of photosynthesis in B-deficient sunflower leaves could be correlated with the diminished efficiency of electron transport and to increased sugar content in the leaves. However, B deficiency caused a dramatic restriction of growth, but did not have any negative effect on parameters related to photosynthesis (such as stomatal density, chlorophyll concentration, photosynthetic capacity and intrinsic photochemical efficiency of PS II) in *Dittrichia viscosa* (Stavrianakou et al., 2006).

Boron deficiency is widespread in Brazilian plantations (Malavolta, 1986; Malavolta et al., 2001), resulting in a reduced root system, flower abortion, fruit malformation and consequently low yields (Franco, 1982). In palm, B deficiency can cause leaves to appear small and crumpled. It may cause a sharp bend in the trunk and horizontal growth and can kill the bud (Worden et al., 2002). Boron deficiency causes a wide array of symptoms, not only among species of palms, but also within a single species (Broschat, 2005). Symptoms always occur on newly emerging leaves, and remain visible on

these leaves as they mature and are replaced by younger leaves (Broschat, 2005). Boron deficiency can cause premature fruit drop in *Cocos nucifera* (Broschat, 2005).

In Brazil, Macedo et al., (1999) related that B is an important micronutrient in *Cocos nucifera*, and this mineral nutrient deficiency can be found in this species. The authors related that the deficiency can promote inhibition of growth and death of growing meristems. In coconut, the deficiency symptoms increase over time, with initial symptoms of reduced leaf area, and after days or months, increased leaf senescence, and finally, necrosis of the apical meristem (Macedo et al., 1999).

Worldwide, coconut (*Cocos nucifera* L.) is cultivated in more than 90 countries, and is the most versatile tree in the world. From this species come many natural products, including foods, drinks, fibers, building materials and chemicals, and coconut palms are an excellent source of food for native peoples in the tropics. In the literature, there are few studies on B deficiency in coconut, and these do not discuss the effects of B deficiency on photosynthetic capacity in coconut palms. A better understanding of the effects of B deficiency in coconut will be important in developing a rational fertilization management program for in coconut palms. The objective of this research was to study the effects of soil B deficiency on gas exchange and photochemical efficiency in green dwarf coconut.

MATERIAL AND METHODS

Plant Material and Experimental Conditions

Seedlings of green dwarf coconut with seven leaves, root washed and without fruit, were cultivated in 100 L pots with sand during 05 Jan 2006 to 29 May 2007 (~17 months) in greenhouse conditions [50% photon photosynthetically flux density (PPFD), maximum PPFD = 1100 $\mu\text{mol m}^{-2} \text{s}^{-1}$; average relative humidity (RH) of 59.6%; average temperature 29.6°C; minimum temperature 17.4°C and maximum temperature 45.5°C] at State University of North Fluminense Darcy Ribeiro, Campos dos Goytacazes city (21°45'39.99" S; 41°17'20.93" W), Rio de Janeiro state]. Before the sand was used, it was purified using hydrochloric acid (HCl): water (1:4) (4h flooded). After the use of the acid, the sand was washed using deionized water until pH to adjust for value 5.0.

Treatments were a) control [complete nutrient solution (CNS); B sufficient (B+)]. The CNS had: macronutrients [3 mmol L⁻¹ calcium nitrate, Ca(NO₃)₂·2H₂O; 1 mmol L⁻¹ ammonium nitrate (NH₄NO₃); 0,25 mmol L⁻¹ potassium phosphate (KH₂PO₄); 4 mmol L⁻¹ potassium nitrate (KNO₃); 1 mmol L⁻¹ magnesium sulfate (MgSO₄·7H₂O)] and micronutrients 40 $\mu\text{mol L}^{-1}$ until 23 August 2006 and 80 $\mu\text{mol L}^{-1}$ after 23 August 2006 iron (Fe)-ethylenediaminetetraacetic acid (EDTA); 5 $\mu\text{mol L}^{-1}$

manganese sulfate (MnSO_4); $2 \mu\text{mol L}^{-1}$ zinc sulfate ($\text{ZnSO}_4 \cdot 7\text{H}_2\text{O}$); $0.5 \mu\text{mol L}^{-1}$ copper sulfate ($\text{CuSO}_4 \cdot 5\text{H}_2\text{O}$); $0.086 \mu\text{mol L}^{-1}$ ammonium molybdate [$(\text{NH}_4)_6\text{MoO}_7\text{O}_{24}$]; $50 \mu\text{mol L}^{-1}$ potassium chloride KCl; $25 \mu\text{mol L}^{-1}$ until 29 August 2006 and $50 \mu\text{mol L}^{-1}$ after 29 August 2006 boric acid (H_3BO_3); and b) solution without B [boron deficiency (B $-$)]. The experimental design was the completely randomized design with 6 replications in each treatment. The software SAEG (Federal University of Viçosa, Viçosa, MG, Brazil) was used and the data were analyzed statistically at 5% probability.

During first 10 days after planting (DAP), each plant of the each treatment received 2.5 L day^{-1} of the CNS $\frac{1}{4}$ strength ($\text{pH} = 5.5 \pm 0.2$). At 10 DAP all plants received CNS $\frac{1}{2}$ strength for five days. From 16 to 60 days after planting (DAP) the treatments (control and $-B$) received full strength CNS. After 61 DAP the $-B$ treatment did not receive H_3BO_3 in CNS.

Plants were irrigated using an automatic system that applied deionized water eight times per day; 6:00 to 20:00h, and used three drip emitters (8 L h^{-1} each drip emitter) per plant. In four pots (two per treatment; tensionmeters were inserted at 0–15 and 0–25 cm deep). The soil potential pressure (ψ_{sp}) was measurement using Hg tensiometers with 1mm of precision and for each day the ψ_{sp} was calculated using $\psi_{\text{sp}} = -12,6H + Z + h_c$ equation. ψ_{sp} is soil potential pressure, H is height of Hg column and h_c is the distance of the Hg column to soil surface and Z is the depth of the tensiometer capsula. The soil moisture among treatments was monitored and differences between B treatments were insignificant.

Gas-Exchange Measurements

In 26 June 2006 the measurements of the photosynthesis (A), stomatal conductance (g_s), transpiration and leaf-to-air vapor pressure difference ($\text{VPD}_{\text{leaf-air}}$) ($n = 18$) were performed from 8:00 to 10:00h a.m. at 171 DAP (initial B deficiency symptoms were observed 3 months after withdraw this mineral nutrient of the soil), using a Portable Photosynthesis System (LI-6200, LI-COR, Lincoln, NE, USA). For gas exchange measurements middle leaflet from leaf number 2 (counting from the top, taking the spindle as zero) of six different plants randomly selected at each time from each treatment. Measurements were made at the middle of each leaflet. The infrared gas analyzer was equipped with an artificial irradiance source red (LEDs, $1000 \mu\text{mol m}^{-2} \text{ s}^{-1}$). The CO_2 flux was adjusted to maintain a concentration of $370 \mu\text{mol m}^{-2} \text{ s}^{-1}$ inside the chamber.

Chlorophyll a Fluorescence Measurements

Instant light response curves of fluorescence were measured on the same attached leaflets ($n = 24$) used for the gas exchange measurements, using the non-modulate plant efficiency analyzer (PEA) fluorometer (Plant Efficiency

Analyzer-Hansatech Ltd., Norfolk, UK). The sensor unit consists of an array of ultra-bright red LED's optically filtered to a peak wavelength of 650 nm, which is readily absorbed by the chloroplasts of the leaf, at a maximum intensity of $>3000 \mu\text{mol m}^{-2} \text{s}^{-1}$ at the sample surface. The LED's are focused via lenses onto the leaf surface to provide even illumination over the area of leaf exposed by the leafclip (4 mm diameter). LEDs have the advantage of being rugged, emitting low levels of heat, and of rising to full intensity very rapidly (typically microseconds) after being switched on. The leaflets were dark adapted for 30 min before the light, when the actinic light intensity was increased from 0 to $\sim 3500 \mu\text{mol m}^{-2} \text{s}^{-1}$.

After dark adapting for 30 min, and an increase of the actinic light, it was possible to obtain F_0 (represent emission by excited chlorophyll a molecules in the antennae structure of Photosystem II); F_v/F_{max} is a parameter widely used to indicate the maximum quantum efficiency of Photosystem II (F_v/F_{max}). In addition, based on the analysis of how the data from the 0-J-I-P fluorescence transient can be processed, a test has been developed and called the "JIP-test" after the steps of the transient. This test can be used as a tool for rapid screening of many samples providing adequate information about the structure, conformation and function of their photosynthetic apparatus (Strasser and Strasser, 1995; Strasser et al., 1999, 2000). From the data stored during the first second [$F_1(t = 50 \mu\text{s})$, $F_2(t = 100 \mu\text{s})$, $F_3(t = 300 \mu\text{s})$, $F_4(t = 2 \text{ ms})$, $F_5(t = 30 \text{ ms})$], the following values are selected to be used by the JIP-test for the calculation of several phenomenological and biophysical expressions leading to the dynamic description of a photosynthetic sample at a given physiological state: the total number of photons absorbed by the antenna molecules of active and inactive PS II RCs over the sample cross-section (ABS/CS_0), electron transport in a PS II cross-section (ET_0/CS_0), concentration of the active reaction centres (RC/CS_0), the maximal trapping rate of an exciton that will lead to Q_A reduction measured over a cross-section of active and inactive RCs (TR_0/CS_0), the ratio of the total dissipation of untrapped excitation energy from all RCs with respect to the number of active RCs (DI_0/RC) (Force et al., 2003). The area over the chlorophyll fluorescence curve is used to determine the plastoquinone pool size of PS II (Govindjee, 1995) i.e. this area is proportional to the pool size of the electron acceptors Q_A on the reducing side of PS II.

SPAD Readings

The leaf 'greenness' measurements were made on the same region of the leaflets used for the gas-exchange and chlorophyll a fluorescence measurements using the Minolta SPAD-502 chlorophyll meter (Minolta Corp., Tokyo, Japan). Ten measurements were performed on each leaflet.

Boron Analysis

After taking physiological measurements, the same leaflets were used for determination of the B concentration. Wet cotton was used to remove dirt from leaflet surfaces, the central vein was removed from each leaflet, and the lamina samples were dried in paper envelopes at 70°C for two days, weighed for dry weight determination, ground in a Wiley mill to pass a 20 mesh screen, and finally, 250 mg samples of the dried material were dry digested in a muffle furnace at 550°C for four hours for B determination using the azomethine-H method (Malavolta et al., 1997). SAEG software was used to evaluate and the data were analyzed statistically at 5% probability.

RESULTS AND DISCUSSION

The B concentration in leaflets of the plants growing in B deficient (–B) sand was reduced compared to the complete nutrient solution (CNS, B+), and the decrease was ≈65% (21.1 to 7.4 mg kg⁻¹ DW (Table 1). The reduction of the B concentration in leaflets was correlated with growth characteristics root dry weight and height of the coconut plants (Table 1). Those growth characteristics were reduced in ~39%, i.e., 2.62 to 1.61 m (height) and 4.26 to 2.99 Kg root DW, respectively. In *Dittrichia viscosa* significant differences in B concentration between B+ and B– plants were observed among the different treatments (Stavrianakou et al., 2006). The leaves, shoots and roots of B– plants showed 93, 54, and 39% lower B concentration, respectively, than that of the corresponding organs of B+ plants, and at plant harvest, B deficiency caused a dramatic restriction of growth, which was accompanied by profound changes in key morphological characteristics (total leaf area per plant, leader shoot length, total number of leaves per plant) of *D. viscosa* plants (Stavrianakou et al., 2006). Han et al. (2008) reported that B deficiency in citrus resulted in a decrease in growth and an increase in

TABLE 1 Growth parameters, boron concentration and gas exchanges parameters in leaflets green dwarf coconut under boron sufficient (B+) and boron deficient (B–) conditions at plant

Parameter	+B	–B	% Change	Statistical significance
Root dry weight (kg)	4.26*	2.99*	–29.81	0.05
Plant height (m)	2.62*	1.61*	–38.55	0.05
Leaflets B concentration (mg kg ⁻¹ DW)	21.1*	7.4*	–64.9	0.05
Photosynthesis (μmol m ⁻² s ⁻¹)	11.2*	7.2*	–35.7	0.05
Transpiration (m mol m ⁻² s ⁻¹)	11.4*	7.5*	–34.2	0.05
Stomatal conductance (g _s) (mol m ⁻² s ⁻¹)	0.46*	0.25*	–45.6	0.05
Leaf to air vapor pressure difference (DPV) (kPa)	3.2*	4.2*	+31.3	0.05
SPAD readings	59.6*	42.2*	–29.2	0.05

*Indicate significantly average differences, $P < 0.05$, Tukey's test 5%.

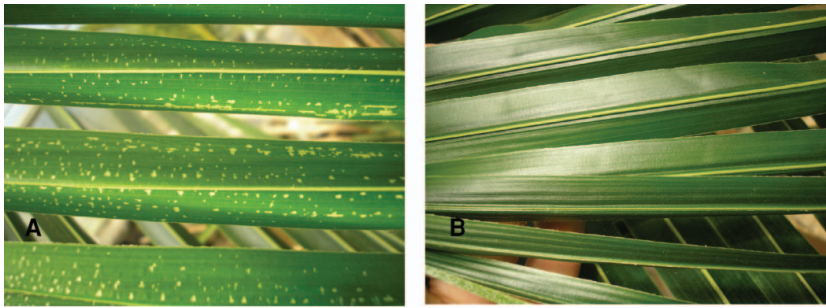


FIGURE 1 Leaflets of green dwarf coconut cultivated as (A) boron deficiency (B⁻) and as (B) boron sufficient (B⁺) in Campos dos Goytacazes, Brazil.

specific leaf weight, and these results also can be explained by the role of B in the formation of primary cell walls. Boron deficiency inhibits root elongation through limiting cell enlargement and cell division in the growing zone of root tips (Dell and Huang, 1997), and inhibition of root tip elongation occurs within hours after the removal of B (B⁻) from nutrient solution (Cohen and Lepper, 1977; Hirsch and Torrey, 1980). In sunflower, Kastori et al. (1995) reported that B deficiency decreased dry matter yield of the roots, shoots and leaves. In coconut palm our results showed that B concentration of the tissue is very important in leaflets, and reduced concentrations in plant tissue can result in reduced growth.

Visual B symptoms appeared on coconut leaflets of the young leaves three months after withdrawal of B (06/26/2006, ~171 days after planting) (Figure 1). The symptoms showed leaflets with white translucent punctuations (“white punctuations”) that coalesce forming parallel lines to the midrib (Figure 1). The leaflet tissue was intact without holes. This symptom has been described for coconut and other palm species by Broschat (2007), Corrado et al., (1992), Dufour and Quencez (1979), and Rajaratnam (1972). The control plant leaflets were completely green and the mesophyll was intact (Figure 1). Worden et al., (2002) showed that the B deficiency in palms can cause leaves to appear small and crumpled. Broschat (2005) reported that in many species including coconut palm (*Cocos nucifera*) mild B deficiency is manifested as sharply bent leaflet tips, commonly called “hook-leaf”. These sharp leaflet hooks are quite rigid and cannot be straightened out without tearing the leaflets. In some species, these “hooks” drop off. Other symptoms show inverted “V” shaped truncations on each leaf and multiple unopened spear leaves were observed on a chronically B-deficient coconut palm (Broschat, 2005).

Boron deficiency affected gas-exchange in coconut leaflets (Table 1). Photosynthesis was reduced from 11 to 7 $\mu\text{mol m}^{-2} \text{s}^{-1}$ in plants grown without B, i.e., ~36% reduction. The reduction of g_s and E values were 45.6 and 34.2%, respectively. Because B deficiency reduced stomatal conductance

(g_s), water loss was reduced (reduced E), and the water loss was not sufficient to remove that heat of the coconut leaflets. Thus, the leaf-to-air vapor pressure difference ($VPD_{\text{leaf-air}}$) increased by 31.3% (Table 1). The reduction of g_s may have resulted in a reduction of the CO_2 concentration on Rubisco carboxylation sites. Sharma and Ramchandra (1990) reported that leaves developed under B deficiency can also have lower stomatal frequencies and smaller stomatal apertures than those with sufficient B, resulting therefore, in a decrease in stomatal conductance to CO_2 . These results showed the stomatal effects on photosynthesis in coconut plants grown in B-deficient sand. Possibly, in coconut leaflets those stomatal effects may be related to the damage observed on mesophyll tissue (Table 1 and Figure 1).

Chlorophyll content (estimated using SPAD meter) of coconut plants leaflets grown in B-deficient sand declined by $\sim 30\%$ (~ 60 to ~ 42 SPAD readings) compared to B-sufficient (CNS) (Table 1). Boron-sufficient leaflets were dark-green, whereas leaflets of B-deficient plants were yellow-green. In addition to the stomatal effects on photosynthesis, reduced chlorophyll may have resulted in a reduction in photosynthetic capacity. In sunflower, B deficiency induced a significant reduction in chlorophyll contents (chlorophyll a of 16.5% and chlorophyll b 22.5%) compared to B-sufficient (El-Shintinawy, 1999). The author suggested that the chlorophyll synthesizing system and/or chlorophyllase activity were affected by B deficiency. Kastori et al. (1995) reported the chlorophyll content in the sunflower leaves was reduced under B deficiency.

If the change in chlorophyll content, common in plants under stress, alters the composition and disorganizes the structure of the light-harvesting protein complexes of thylakoid membranes (Kastori et al., 1995) which leads to an altered chloroplast architecture (Khavari-Nejad and Mostofi, 1998), then, these damages can promote alteration in chlorophyll fluorescence emission (Strasser et al., 2000). In fact, in B-deficient coconuts palms the chlorophyll fluorescence was significantly altered (Table 2). The total number of photons absorbed by the antenna molecules of active and inactive PS II RCs over the sample cross-section (ABS/CS) was reduced 6% in B-deficient coconut palms. In addition, the concentration of the active reaction centers (RC/CS_0), electron transport in a PS II cross-section (ET/CS_0), the maximal trapping rate of an exciton that will lead to Q_A reduction measured over a cross-section of active and inactive RCs (Reaction Centers) (TR/CS_0), and the area over the chlorophyll fluorescence curve is used to determine the plastoquinone pool size of PS II (area above the fluorescence induction curve) were reduced 15.2, 12.5, 7.8, and 16.8% in leaflets of B-deficient coconut palms (Table 2).

ABS refers to the flux of photons absorbed by the Chlorophyll antenna pigments. Part of this excitation energy is dissipated, mainly as heat and less as fluorescence emission, and another part is channeled as trapping flux TR to the reaction centre RC and converted to redox energy by reducing the

TABLE 2 Fluorescence parameters in leaflets green dwarf coconut under boron sufficient (B+) and boron deficient (B-) conditions at plant

Parameter	+B	-B	% Change	Statistical significance
Total number of photons absorbed by antenna molecules of active and inactive PS II (ABS/CS ₀)	682*	639*	-6.3	0.05
Ratio of total dissipation of untrapped excitation energy from RCs (DI ₀ /RC)	0.49*	0.58*	+18.4	0.05
Concentration of active reaction centers (RC/CS ₀)	299*	253.5*	-15.2	0.05
Maximal trapping rate of an exciton leading to Q _A reduction per cross section of active and inactive RCs (TR ₀ /CS ₀)	536*	493.9*	-7.8	0.05
Area over the chlorophyll fluorescence curve (estimate of plastoquinone pool size of PS II)	55922*	46516*	-16.8	0.05
Electron transport in PS II cross section (ET ₀ /CS ₀)	324.6*	283.9*	-12.5	0.05
Maximum quantum efficiency of PS II (F _V /F _{max})	0.813*	0.803*	-1.1	0.05

*Indicate significantly average differences, $P < 0.05$, Tukey's test 5%.

electron acceptor Q_A to Q_A⁻, which is then reoxidized to Q_A thus creating an electron transport ET that leads ultimately to CO₂ fixation (Strasser et al., 2000). If B deficiency results in a decrease in chlorophyll concentration (Table 1), then the reduction of the ABS/CS₀ (Table 2) was caused by reduced chlorophyll concentration in leaflets of B deficient palm.

The area above the fluorescence curve was the fluorescence parameter that was most affected by B deficiency (16.8%) (Table 2). This parameter represent the area over the curve between F₀ and F_m which has been shown to be proportional to the pool size of the electron acceptors Q_A on the reducing side of PSII. If the transfer of electrons from the reaction centers to the quinone pool is blocked, the area will be dramatically reduced. Thus, the results showed that B deficiency resulted in damage to electron transport, and the reduced values of the ET/CS₀ and TR/CS₀ fluorescence parameter showed this response. Reduced quinone pool will reduce maximal trapping rate of an exciton that will lead to Q_A reduction measured over a cross-section of active and inactive RCs (TR/CS₀). In sunflower, Kastori et al. (1995) reported that B deficiency appreciably decreased the apparent quantum yield and quantum efficiency of photosystem II electron transport, and B deficiency induced a pronounced inhibition (31%) of the whole chain electron transport compared to B-sufficient plants. El-Shintinawy (1999) reported that the activity of PSII declined in B-deficient sunflower chloroplasts by 42% compared to that in B sufficient sunflower chloroplast.

Thus, B deficiency in coconut leaflets significantly affected the structure, conformation and function and modified the energy fluxes in the photosynthetic apparatus. These effects resulted in a 18.4% increase in the effective dissipation in active RC (DI_0/RC). Dissipation in this context refers to the loss of absorbed energy through heat, fluorescence and energy transfer to other systems (Strasser et al., 2000). Therefore, dissipation can be thought of as the absorption of photons in excess of what can be trapped by the RC. In this research, this trapped problem by RC can be showed by reduced values of the RC/CS_0 (density of the active reaction centers per excited cross-section) (Table 2).

However, the maximum quantum efficiency of Photosystem II (F_v/F_{max}) did not vary between treatments (Table 2). Strasser et al., (2000) also noted the relative insensitivity of the F_v/F_{max} ratio to change. The F_v/F_m ratio, often the only fluorescence parameter used to gauge the occurrence and extent of stress in plants, can be quite insensitive to change (Force et al., 2003). Thus, we hypothesized that F_v/F_m is not a good parameter to assess of B deficiency in coconut palms.

Our results demonstrate that by analyzing fluorescence (JIP test parameters) derived from the polyphasic fluorescence transients and gas-exchange measurements, we are able to estimate the functional changes of PSII and gas-exchange in B deficient coconut plants. The results in this study suggest that fluorescence analysis (JIP test) and instantaneous measurements of gas-exchange can be useful tools in assessing the physiological effects of B deficiency in green dwarf coconut.

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