Short Report

Effect of Soil Water Conditions on Biopolymer Accumulation in Root Polyderm of Feijoa sellowiana (Myrtaceae)

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Introduction

Feijoa sellowiana, a subtropical species of Myrtaceae family, native to South Brazil with secondary dispersion in Uruguay (Thorp and Bielek, 2002), has both ornamental and medicinal properties and shows great potential in foods, drugs and cosmetics industry (Hardy and Michael, 1970; Kolesnik et al., 1991). Biopolymers like lignin and suberin are cell wall components and specialized plant cells can locally modify their cell walls by accumulating biopolymers as part of a developmental program or when challenged by environmental conditions (Roppolo and Geldner, 2012). It is interesting to study biopolymer accumulation in feijoa roots because it bears a special type of protective root tissue called polyderm, composed of alternating suberized and non-suberized layers, common in only a few other plant families like Hypericaceae, Onagraceae and Rosaceae (Esau, 1970; Fahn, 1982). In our previous study on root polyderm (Nii et al., 2012), we called the non-suberized layers of polyderm, thick-walled cells (twc) and suberized layers, endodermis-like (el) cell layers.

In non-polyderm bearing roots of loquat (Eriobotrya japonica Lindl.) (Nii et al., 2004; Pan et al., 2006) and red bayberry (Myrica rubra Sieb. et Zucc.) (Song et al., 2011), our data indicated a significant increase in biopolymer accumulation in cortical cell walls under drought conditions. However, in polyderm-bearing roots, studies on biopolymer accumulation are still rare. Recently, we examined the effect of long-term water-logged condition in roots of wax apples (Syzygium samarangense), also belonging to Myrtaceae family, and found that polyderm layers decreased in water-logged roots than normally grown roots (Tuladhar et al., 2015). Decrease in polyderm layers means decrease in the biopolymer accumulation in the cell wall. To understand what factor/s link to restriction of cell division at polyderm, biopolymer accumulation must be studied in detail. This study focuses on the extent of biopolymer accumulation in feijoa polyderm under varying the soil water conditions.

Materials and Methods

In the years 2011 and 2012, 23 year-old feijoa (cultivar ‘Mammoth’) trees propagated from cuttings, were grown under water-logged, normal and drought conditions in a greenhouse located at Meijo University in Nagoya, Japan. They were grown in plastic baskets (diameter and depth 300 mm, stitch size 30 mm) covered with nonwoven fabric and filled with sandy soil. Eight baskets were allotted per treatment batch in both years. Treatment was maintained for more than 4 months starting from early April to late August of each year.

The water-logged batch was watered every day until it saturated the soil in the baskets, and leaked out of its holes and collected in plastic dishes (10 centimeters deep) placed at the bottom of the baskets. Water depth in these dishes was maintained at a depth of 3-5 centimeters. Baskets allotted to normal were watered everyday but no plastic dish was placed under them. Baskets in drought treatment were watered only when the upper surface of the soil became dry and no dish was placed under these baskets as well. Depending on the weather, it took 2-4 days for the upper surface to dry. In the second year, the magnitude of stress during the treatment was assessed by measuring the water potential using a pressure chamber (Plant Moisture Stress Measurements by PMS Instrument Company, Oregon, U.S.A) during the month of July as shown in Fig. 2. A branch with 2 or 3 leaves attached on the second or third node was collected from each plant, before noon, in air-tight
plastic bags and delivered to the laboratory in an icebox for immediate measurement of water potential of xylem sap. The value of the pressure chamber was maintained at approximately -1.0 MPa for water-logged, -1.5 to -2.0 MPa for normal and -3.0 to -3.5 MPa for drought treatment before watering the plants. During the last week of August, roots were collected for observation. At first, the plants were removed from the baskets without disturbing the soil intact with root mass. Then new white roots were collected from the tip to the basal zone. Then they were hand-sectioned using a stainless steel razor blade for observation. A series of cross-sections were prepared at three locations 1) less than 20 mm from the root tip, 2) between 30-50 mm from the root tip, and 3) more than 60 mm from the root tip to examine the development of biopolymer accumulation in polyderm as roots aged. However, in the second year, 2012, the roots elongation was restricted in drought treatment so roots that elongated more than 60 mm are not presented in Fig. 5C. Root specimens were then examined under fluorescent microscopy (excitation wavelength 365 nm) without staining. About 20-30 roots were collected from plants allotted to each treatment and sectioned for observation. Then a representative root was selected for report since the basic anatomical features were similar among root samples. The extent of biopolymer accumulation in thick-walled cell layers that formed between the endodermis and the endodermis-like cell was compared. To simplify comparison, we categorized the results of our observation as - : thick-walled cells developed but biopolymer accumulation was not observed, + : biopolymer accumulation was observed partly in only a few thick-walled cells, ++ : light biopolymer accumulation was observed in all thick-walled cells, +++ : heavy biopolymer accumulation was observed in all thick-walled cells as shown in Fig. 1.

Fig. 1. Illustration on the extent of lignin accumulation in thick-walled cells at polyderm.
- : thick-walled cells developed but lignin accumulation was not observed.
+ : lignin accumulation was observed in some thick-walled cells.
++ : lignin accumulation was observed in all thick-walled cells.
+++ : heavy lignin accumulation was observed clearly in all thick-walled cells.
en : endodermis, twc : thick-walled cell, el : endodermis-like cell.

Fig. 2. Water-potential measurement of feijoa feijoa branch with 2 or 3 leaves grown in drought, normal and water-logged treatments in the year 2012. The arrows show the days when water-potential measurement was highest in normal in normal and drought conditions.
Results and Discussion

In every treatment, the extent of biopolymer accumulation at the polyderm increased as distance from the root tip increased (Figs. 4, 5). In Fig. 4, roots grown in normal and drought conditions showed similar data at 30-50 mm distance from the root tip but in water-logged treatment, biopolymer accumulation was - level. Between the endodermis and the endodermis-like cell layer, although formation of thick-walled cell layer could be observed under the fluorescent microscope (Figs. 3B, B1, white arrow), there was a difference in the extent of biopolymer accumulation in thick-walled cells according to soil water condition. Referring to the illustration in Fig. 1, lignin-like accumulation in roots of the drought treatment reached ++ level (Figs. 3C, C1, white arrow) whereas, in water-logged roots, biopolymer accumulation was - level (Figs. 3B, B1, white arrow) when cross-sections of roots with similar xylem vessel number (and same distance from the root tip) were compared. It is important to note that the distance from the root tip was not considered as the only barometer to determine root age. This is because, root elongation is restricted but secondary development of roots occurs faster in roots under drought treatment. In the second year, new roots that grew more than 60 mm were fewer in number in the drought treatment. Therefore only two graph bars are presented in Fig. 5C.

In roots of water-logged treatment, the formation of twc was observed but the accumulation of biopolymers

Fig. 3. Difference in biopolymer accumulation in thick-walled cells (twc) in roots grown in normal (panels A, A1), water-logged (panels B, B1) and drought (panels C, C1) treatments in the consecutive years 2011 (panels A, B, C) and 2012 (panels A1, B1, C1). Arrows show the extent of biopolymer accumulation in thick-walled cells. en: endodermis, Cs: Casparian strip, twc: thick-walled cells, el: endodermis-like cells. Scale bar = 50 μm.
occurred further away from the root tip. In contrast, heavy accumulation of biopolymers was visible in drought treatment. This result showing biopolymer accumulation to be earlier and more heavy in roots under drought treatment, was similar to roots of loquat (Nii et al., 2004; Pan et al., 2006) and red bayberry (Song et al., 2011) in which lignin content in cell wall of cortex cells increased significantly under drought treatment. When stained with phloroglucinol-HCl, the thick-walled cells stained red so it was thought to contain mostly lignin. However, further study using chemical analysis is necessary to distinguish whether biopolymer accumulating in thick-walled cells is solely lignin.

The objective of this study was to confirm whether lignin-like biopolymer accumulation in thick-walled cells, got affected under different soil water conditions. Based on this observation, we know that encountering drought can trigger earlier and heavier deposition of biopolymer at thick-walled cells of feijoa root polyderm. Recently biopolymer in root tissues is gaining more attention because lignin was detected in the Casparian strip of arabidopsis previously thought to be mostly suberin (Naseer et al., 2012). A detailed knowledge on biopolymer accumulation may hold the key to further understand the role of polyderm. In this study, we also noticed that many thick-walled cell layers existed one after another, between the endodermis and endodermis-like cell layer in roots under drought treatment (Figs. 3C, C1) than normal treatment (Figs. 3A, A1). Cell layers existing alternately in the order of “suberized - non-su-
berized - suberized” were disturbed under drought treat-
ment. This is because the thick-walled cells increased 
consecutively between endodermis-like cells instead of 
existing alternately, as described by Esau (1970) and 
Fahn (1982). Further studies must be conducted to elu-
cidate how thick-walled cell layers increase in roots of 
drought treatment because it differed from our previous 
conclusion that each cell in the pericycle divided into 3 
cell layers; the outermost cell layer accumulated lignin to 
form the el and the innermost cell layer retained the pericyclic 
characteristic and divided further in the same way to 
form the polyderm layers (Nii et al., 2012). Irregularity 
in biopolymer accumulation at polyderm layers under 
drought condition must be investigated further.

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