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Jia-En Zhang [a], Guo-Ming Quan [a], Zhao-Xiang Huang [a], Shi-Ming Luo [a] & Ying Ouyang [b]
[a] Key Laboratory of Ecological Agriculture of Ministry of Agriculture of the People's Republic of China, Key Laboratory of Agro-ecology and Rural Environment of Guangdong Regular Higher Education Institutions, South China Agricultural University, Guangzhou, P.R., China
[b] USDA Forest Service, Center for Bottomland Hardwoods Research, Mississippi State, Mississippi, USA
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Evidence of Duck Activity Induce Anatomical Structure Change and Lodging Resistance of Rice Plant

JIA-EN ZHANG,[1] GUO-MING QUAN,[1] ZHAO-XIANG HUANG,[1] SHI-MING LUO,[1] AND YING OUYANG[2]

[1] Key Laboratory of Ecological Agriculture of Ministry of Agriculture of the People's Republic of China, Key Laboratory of Agro-ecology and Rural Environment of Guangdong Regular Higher Education Institutions, South China Agricultural University, Guangzhou, P.R. China

[2] USDA Forest Service, Center for Bottomland Hardwoods Research, Mississippi State, Mississippi, USA

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Address correspondence to Jia-En Zhang, Key Laboratory of Ecological Agriculture of Ministry of Agriculture of the People's Republic of China, Key Laboratory of Agro-ecology and Rural Environment of Guangdong Regular Higher Education Institutions, South China Agricultural University, Guangzhou 510642, P.R. China. E-mail: jeanzh@scau.edu.cn; and Ying Ouyang, USDA Forest Service, Center for Bottomland Hardwoods Research, Mississippi State, MS 39762, USA. E-mail: youyang@fs.fed.us

The system of rice cultivation associated with duck raising is known as an integrated rice-duck (RD) farming system and has a long history in China and Asia. However, the roles of duck activities on rice physiological characteristics are unknown. Field and laboratory experiments were conducted to investigate mechanical stimulations of duck activities on rice physiological

characteristics, and hence on morphology and anatomy. Here we found that there were variations in anatomical structure of the rice culm internodes and in lodging resistance of rice plants with and without the duck activities. Anatomical analysis showed a distinct difference between the RD and RND (rice-no-duck) treatments in culm wall thickness of the rice For Internode N4, the culm wall from the RD treatment was more than 15% thicker than that the RND treatment. In general, the epidermal layer of the rice culm was significantly denser for the internodes and the thickness of the culm walls increased from N1 to N4 under the RD system. Additionally, the areas of vascular bundles were larger and were better organized and structured in the RD treatment. Our results further revealed that the breaking-resistant strength increased whereas the lodging index of the rice culm decreased with duck activities.

Keywords anatomical structure; lodging resistance; rice duck farming

1. Introduction

The system of rice cultivation associated with duck raising is known as an integrated rice-duck (RD) farming system (Wang, 2003; Huang et al., 2005). This system is a form of organic farming that yields two crops simultaneously, one for rice as the main crop and the other for ducks as the subsidiary crop. The RD farming has a long history in China (since 1368 A.D.) and is a complex planting and breeding model of paddy fields in Asia (Zhang et al., 2007). The integrated RD

farming is known to have numerous economical, environmental, and ecological benefits. Ducks eat weed seeds, tender weeds, insects and crabs, and thus reduce pests in the paddy field. Due to the frequent movement, ducks improve the physical structure of the paddy soil that enhances the root growth and ultimately produce higher yields. The RD farming system can also reduce the costs of weeding, insecticides and chemical fertilizers, and therefore higher net returns could be achieved. In other words, the RD farming system can reduce agricultural input costs, produce organic grains and ducks, and promote more sustainable agroecosystems.

In recent years, with an increasing understanding of the benefits of the RD farming system, several efforts have been devoted to characterizing the morphological characteristics of rice due to the impacts of duck activities (Masahara and Hideomi, 1993; Koji et al., 1998; Teo, 2001). Ma et al (2004) showed that with the presence of ducks, the rice stalk becomes thicker or wider; the rice plant forms a strong and dwarf harpoon shape; and the growth and grain-bearings of the rice are enhanced in the tillering stage. Huang et al (2003) reported that the height, leaf area index, and biomass of rice stalks in the RD farming system decrease during the tillering and heading stages. Zhang et al. (2007) found that the length and width of flag leaf, the number of spikelets per panicle, the panicle length, and the mass of panicles of the rice plant increased in the RD farming system. Although these morphological phenomena were observed, the underlying mechanisms and the impacts of duck activities upon rice anatomical structure are unknown.

Lodging resistance is one of the most important mechanisms for producing high grain yields of rice (Terashima et al., 1994; Khanna, 1991). Zuber et al. (1999) reported that, among the morphological characteristics of the rice plant, stem diameter and weight are directly correlated to lodging resistance and the breaking strength of the stem. Mulder (1954) demonstrated that the cause of lodging is the lack of proportionality between the weight of the upper part and the sturdiness of the basal part. Ma et al (2004) indicated that the morphological traits of the N1, N2, and N4 internodes, the culm wall, and the vascular bundles were related with the lodging index of the rice plant. However, little attention has been devoted to investigating the impacts of duck activities on the lodging resistance of the rice plant. We examined here the impacts of duck activities (including pecking, diving, and dabbling) in a paddy field upon the anatomical structure and lodging resistance of the rice plants by comparing the results with and without duck activities.

1.Materials and methods

1.1 Study site and experimental design

The experiment was conducted at the Zengcheng Research and Educational Station located about 40 km east of the campus of the South China Agricultural University, Guangzhou City, China. This station has a subtropical climate with an average annual rainfall of 1800 mm and a mean annual temperature of 22^{o}C. The paddy soil in the station is developed from Latosol.

The rice variety used in this study was *Oryza sativa* cv. Simiaoxuan, which was provided by South China Agricultural University, Guangzhou City, China. The following two cultivation treatments each with four replicates were chosen for the experiment: (1) rice with duck (RD); and (2) rice with no duck (RND), which resulted in a total of eight experimental plots. Each experimental plot had an area of 67 m^2 and was separated from each other by clay ridges with a height of 0.5m and width of 0.3 m around the plot boundaries to prevent water and nutrient exchanges between the plots. To prevent the ducks from escaping, the plots were contained by the nylon-net with a height of 0.8 m. These plots were randomly distributed in the study site. All of the experimental plots received the same amount of chemical and organic fertilizers.

The experimental plots in the paddy field were tilled, fertilized, and planted on August 15, 2005. After 10 days from rice transplanting, an average of 3 ducks with the ages ranging from 10 to 15 days were introduced into each experimental plot for the RD treatment. The ducks were retrieved and transferred on October 18, 2005 to other places after the heading stage of the rice growth.

1.1.Sampling and analysis

Plant sampling was collected 20 days after full heading of the rice. Four representative hills of the rice plant were sampled from each plot and the 12 largest tillers with three from each hill were used to measure characteristics of the lodging resistance. The culm internodes were cut transversely with a scalpel to measure the inner and outer diameters of the internodes with a vernier caliper. The averaged culm wall thickness was then calculated by the following equation: culm wall thickness = (outer diameter - inner diameter)/2. The breaking resistant strength of Internodes N2 or N3 was measured and the lodging index was calculated according to the procedures reported by Islam, et al. [14].

Anatomical structure variations between the two treatments were analyzed by cutting the Internodes of N1, N2 and N4 with a scalpel and by observing the samples under an electron microscope (Model XL30). The pictures were taken only at these representative positions of the rice culm. Statistical analyses were conducted with SPSS (SPSS, Inc., Chicago, IL, USA) and a χ^2-test was performed using Microsoft Excel 2000. The significance level was set at $\alpha = 0.05$ with a confidence interval of 95%.

1.1.Mechanical touching experiment

Four treatments were employed for the mechanical touching on rice stalk experiment, including (1) control (without mechanical touching), (2) mechanical touching for five seconds, (3) mechanical touching for 10 seconds, and (4) mechanical touching for 15 seconds. The mechanical touching was conducted by a brush and was started once a day at 5:00 pm and continued for 40 days after the rice planting. The contents of abscisic acid (ABA) in the rice leaves were analyzed by HPLC (Agilent HP1100).

1. Results

The internode of the rice culm is composed of coat, mechanical tissue, thin-wall tissue, and vascular bundle. These internodes are named as first node (N1), second node (N2), third node (N3), and fourth node (N4) counting downward from the top of a rice plant. Comparison of the light electron microstructures of the N1, N3, and N4 internodes for the rice culm between the RND and RD treatments showed that the vascular bundles were much better organized and structured in the RD treatment than in the RND treatment (Fig. 1). There are two vascular bundle rings inside the epidermal layer of the rice culm, namely the outer ring and the inner ring. Figure 2 shows the outer ring of vascular bundles for N4, N3, and N1 internodes between the two treatments. The sizes and areas of the vascular bundles for the internodes were larger in the

RD treatment than in the RND treatment. Table 1 shows the culm wall thicknesses for all of the four internodes between the two treatments. There was a distinct difference in the culm wall thickness between the RD and RND treatments except the Internode N3. The former treatment was more than 15% thicker than the latter treatment for Internode N4.

The outermost part of the rice culm is the epidermis that is composed of a thin but dense layer made up of various thick-walled cells in a remarkably uniformed pattern. Figure 3 shows the cross-section view of the epidermis of the rice culm. This photograph demonstrates that the layer of epidermis of the rice culm for the RD treatment was significantly denser (or more compact) than the RND treatments, especially for the N4 and N3 internodes.

Within the epidermis are the small, thick-walled, sclerenchyma that form the mechanical tissue and provide a major mechanical support for non-elongating regions of the rice plant. Figure 3 further reveals that the mechanical tissue of the four internodes was thicker for the RD treatment than for the RND treatment.

Table 1 also shows that the breaking resistance strength and lodging index of Internodes N2 and N3 of the rice culm between the RD and RND treatments. The breaking resistance strengths of Internodes N2 and N3 were, respectively, 101 and 119 g larger in the RD treatment than in the

RND treatment, while the lodging index of Internodes N2 and N3 were, respectively, 21 and 15 smaller in the RD treatment than in the RND treatment.

Figure 4 shows a field photograph comparing the real lodging status at the ripening and maturing stages of the rice plants between the RD and RND treatments. The patch isolated with a plastic barrier was a plot for RND treatment surrounded by the RD treatment. This field picture shows that the lodging resistance of the rice plant in the ripening stage was obviously weaker in the RND treatment than in the RD treatment (Fig. 4a). Such a trend became prominent as time elapsed to the maturing stage (Fig. 4b).

1. Discussion

The large size, well organized, and compacted structure of the vascular bundles of the rice culm from the RD treatment demonstrated that the impacts of duck activities on anatomical structure of the rice were profound, which have not been reported elsewhere. The decrease in internode length for N4 as well as the increase in culm wall thickness for all internodes except for N3 from the RD treatment further confirmed this finding. Although the exact reasons for such anatomical structure changes remain unknown, a possible explanation would be the mechanical stimulations of the rice culm and the changes of paddy field microclimate environments due to the duck activities. In our other mechanical touching study, we found that

the artificial mechanical stimulation or touch on the rice stalks with a brush increased the content of ABA in the leaves of the rice plant (Table 2). The increase in ABA content inhibited stem elongation and thus resulted in thicker culm wall for improving lodging resistance. Additionally, duck activities may improve potassium and silicon influxes into the rice plant because of the larger vascular bundles. These two elements increase the stiffness of the rice culm.

The denser layer and thicker mechanical tissue of epidermis of the rice culm in the N4 and N3 internodes from the RD treatment suggest that duck activities promoted the stronger internode lodge-resistance strength, especially near the base of the rice plant and thereby enhances the lodging resistance of the rice plant [10, 13].

Difference in lodging indexes of a rice culm was largely dependent on the breaking resistance strength of the basal internodes of the rice plant. The larger breaking resistance strengths and the lower lodging indexes of Internodes N2 and N3 in the RD treatment proved that the lodging resistance of rice culm was greatly enhanced with duck activities. Comparison of the lodging status at the ripening and maturing stages of the rice plant between the RD and RND treatments (Fig. 4) further confirmed that duck activities enhanced the lodging resistance of the rice plant.

In this study, we showed that the evidence of duck activities induced the anatomical structure changes of the rice culm and enhanced the lodging resistance of the rice plant. This finding would

shed the light on exploring the role of mechanic stimulation of animal (e.g., duck, fish) activities on plant growth and would intrigue researchers' interests to develop a novel, organic, and substantial rice farming pattern for economical, environmental, and ecological benefits. Further study is warranted to investigate the plant physiological and biochemical mechanisms causing the anatomical structure change and lodging resistance enhancement of the rice plant by duck activities.

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FIGURE CAPTIONS

Figure 1. Microstructures of Internodes N4 (a1), N3 (a2) and N1 (a3) of the rice culm for the RD treatment (a). Microstructure of Internodes N4 (b1), N3 (b2) and N1 (b3) of rice culm for the RND treatment (b). Amplifying factor of the microscopes is 80 for a1, a2, b1 and b3, and is 150 for a3 and b3.

Figure 2. Microstructures in the outer ring of the vascular bundles for N4 (a1), N3 (a2) and N1 (a3) of the rice culm for the RD treatment (a). Microstructures in the outer ring of the vascular bundles for N4 (b1), N3 (b2) and N1 (b3) of the rice culm for the RND treatment (b). Amplifying factor of the microscopes is 600 for a1, a2, b1 and b2, and is 800 for a3 and b3.

Figure 3. Epidermis layers for Internodes N4 (a1), N3 (a2) and N1 (a3) of the rice culm for the RD treatment (a). Epidermis layers for Internodes N4 (b1), N3 (b2) and N1 (b3) of the rice culm for the RND treatment (b). Amplifying factor of the microscopes is 2560 for a1, a2, b1, and b2, and is 1280 for a3 and b3.

Figure. 4. Lodging resistance status of the early rice in ripening (a) and maturing (b) stages. Plots isolated with a barrier were for the RND treatment surrounded by the RD treatment.

Table 1. Stem wall thickness, breaking resistance strength, and lodging index of Internodes N1, N2, N3, and N4 of the rice culm. Same letters after the values in the same column in the table indicate no statistical significance.

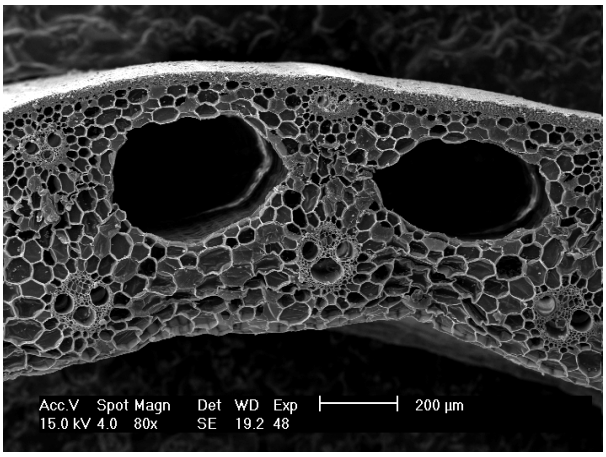
Treatment	N1	N2	N3	N4
Stem wall thickness (mm)				
RD	0.44±0.02a	0.65±0.04a	0.79±0.05a	0.91±0.06a
RND	0.31±0.02b	0.54±0.02b	0.66±0.03a	0.67±0.03b
Breaking resistance strength (g)				
RD		560.18±16.38a	725.09±12.40a	
RND		458.61±37.62b	605.47±39.32b	
Lodging index				
RD		116.34±6.05b	91.91±3.45b	

RND		137.41±7.84a	107.22±4.37a	
Internode length (cm)				
RD	31.25±0.76a	15.78±0.36a	10.42±0.21a	5.65±0.10b
RND	30.85±1.05a	15.52±0.29a	10.93±0.27a	6.85±0.56a

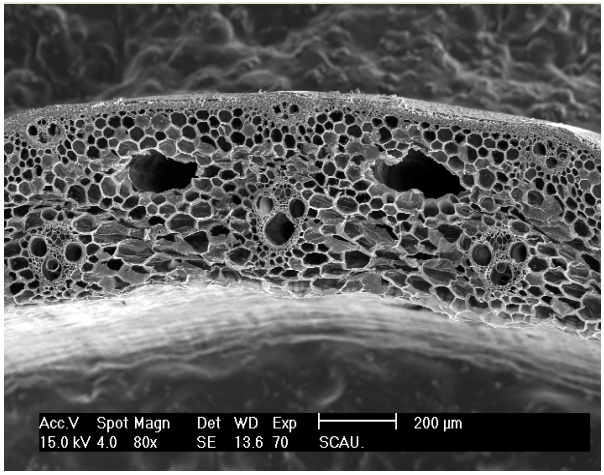
Table 2. Contents of leaf abscisic acid (ABA) (ng/g) under different treatments. The experiment started on September 5, 2004 and ended on October 15, 2004. Same letters after the values in the same column in the table indicate no statistical significance.

Treatment	Measured on 9/25/2004	Measured on 10/5/2004	Measured on 10/15/2004
Control (no touch)	4440.02±466.64b	4041.68±296.75a	4484.22±371.09b
Touch for 5 seconds per day	4852.13±569.83b	5030.8±1024.27a	6009.33±1447.33b
Touch for 10 seconds per day	6576.36±323.13a	5191.14±1526.31a	6083.97±1067.12b
Touch for 15 seconds per day	4963.42±119.68b	5382.3±947.60a	7486.23±744.11a

Figure 1.



a1



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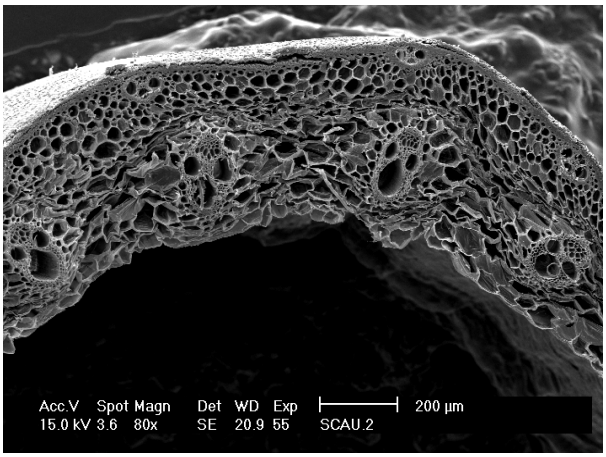
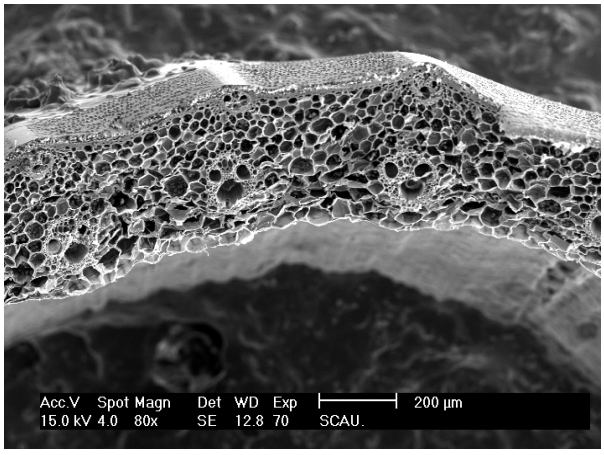


a2



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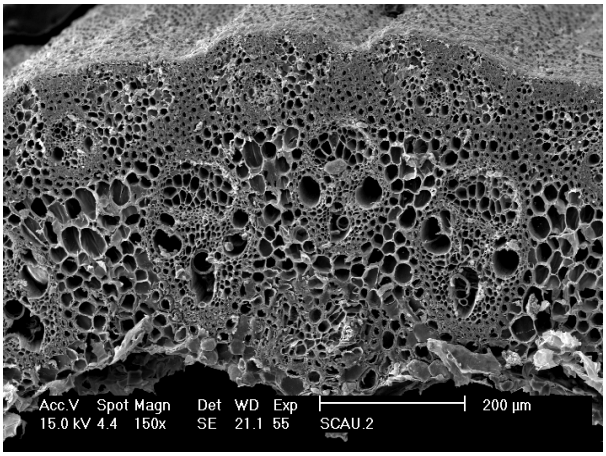

a3

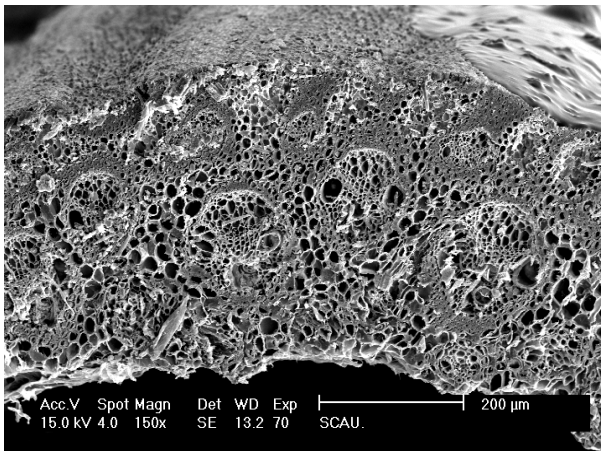

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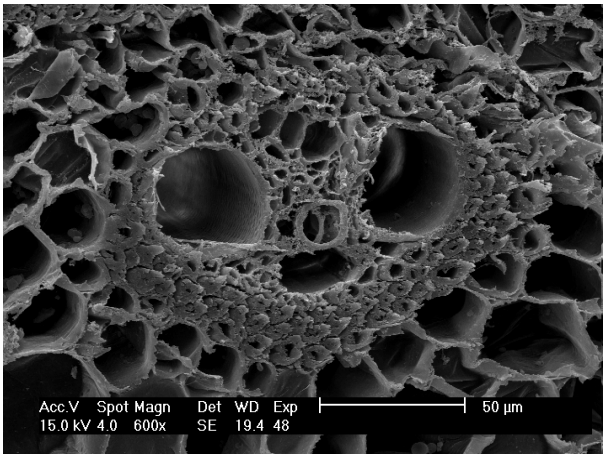
Figure 2.



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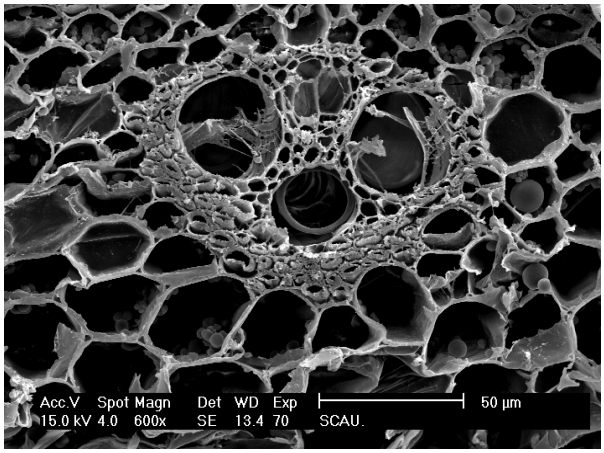

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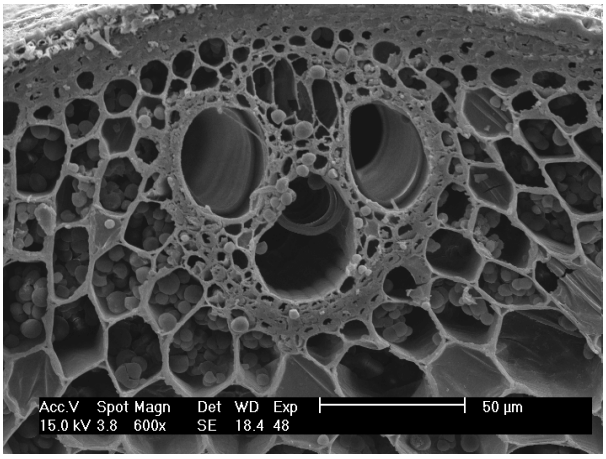

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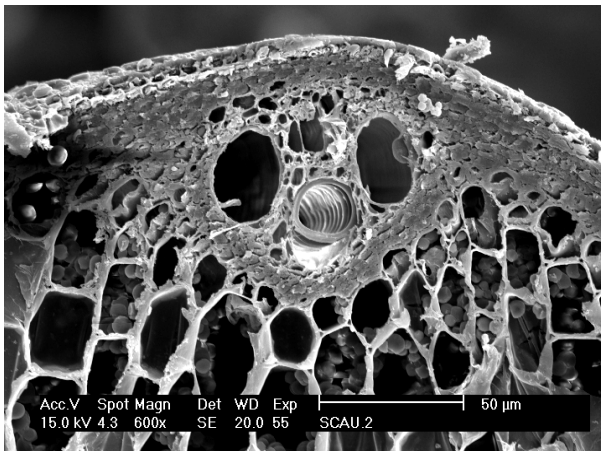

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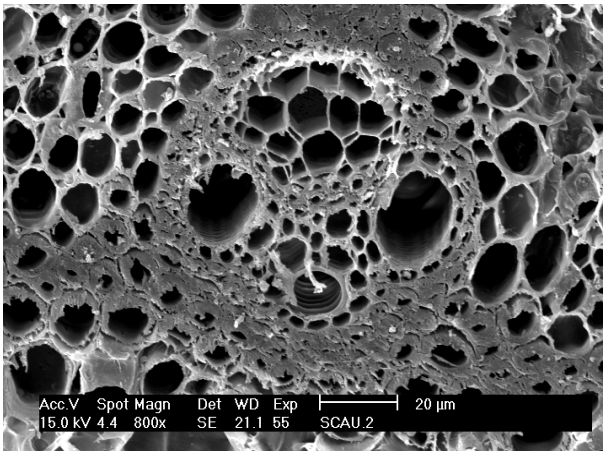

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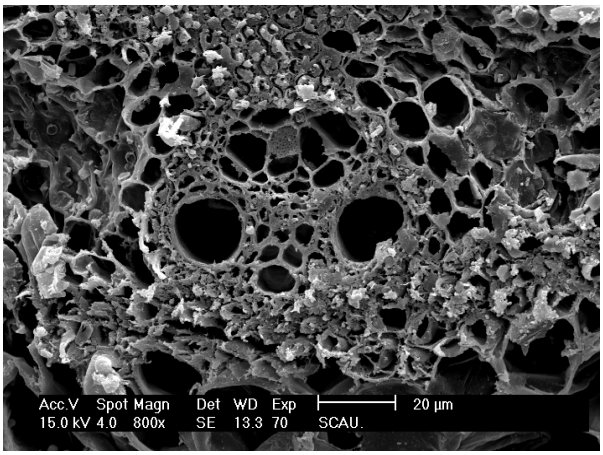

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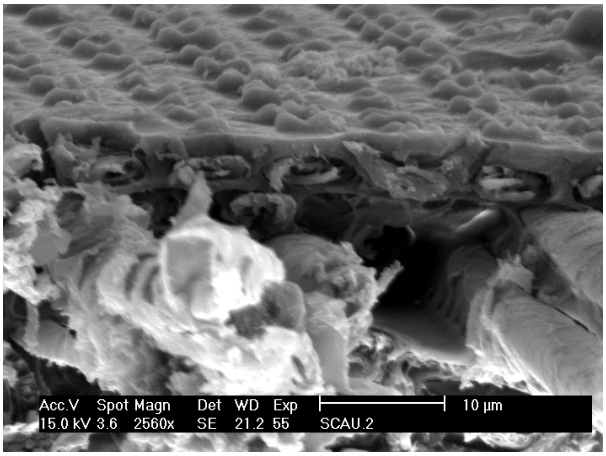
Figure 3.



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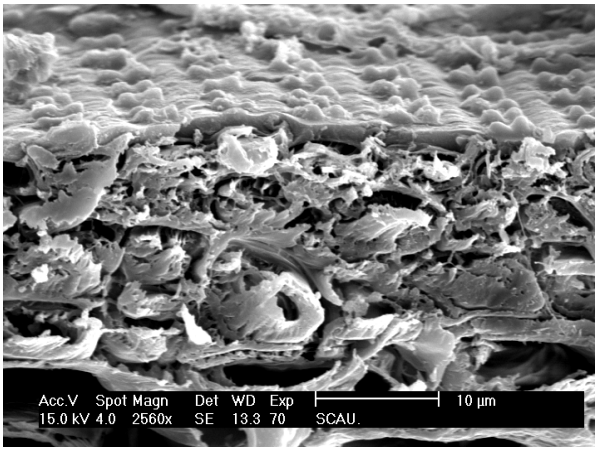
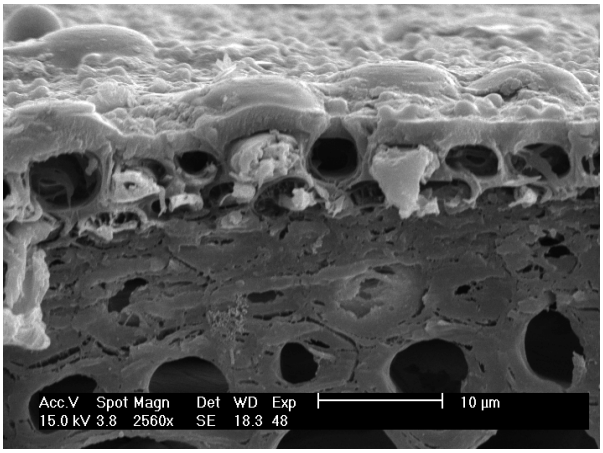


b1



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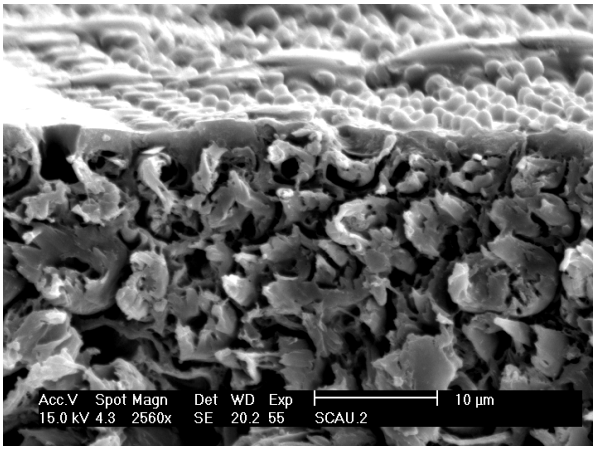

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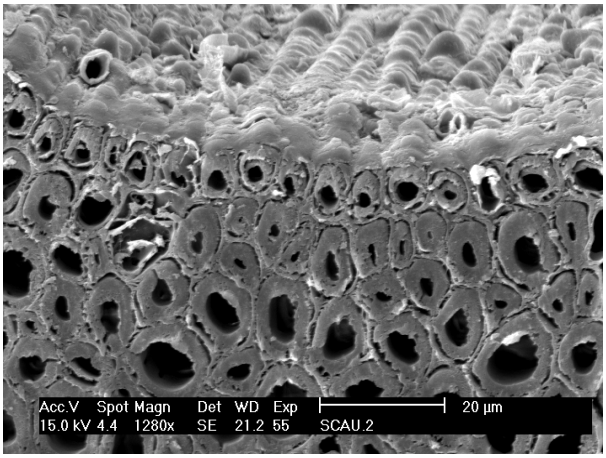

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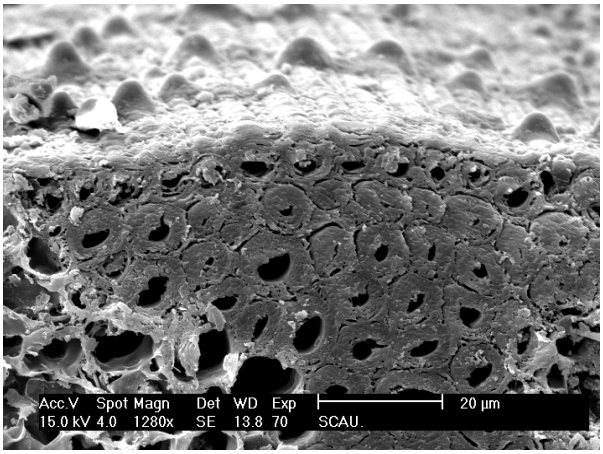

b3

Figure. 4.

a

b