Anatomical characteristics of stomata, mesophyll and petiole of six varieties sweet potatoes (*Ipomoea batatas* L.) after organic fertilizer induction

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Anatomical characteristics of stomata, mesophyll and petiole of six varieties sweet potatoes (*Ipomoea batatas* L.) after organic fertilizer induction

H L Rampe[1] , S D Umboh[1], R Siahaan[1], P V Maabuat[1]

[1]Biology Department of Mathematics and Natural Science Faculty,
Sam Ratulangi University, Indonesia

Corresponding author : hennyrampe@unsrat.ac.id,

Abstract. The study aimed to evaluate the anatomical characteristics of six sweet potato varieties (*Ipomoea batatas* L.) after induction of organic fertilizer. Research used Completely Randomized Block Design with 3 replications. Five sweet potato varieties (J) were used i,e: Antin 3 (J1), Jago (J2), Cilembu (J3), Shi Royutaka (J4), Local Purple (J5) and Local White (J6). Petroganic Organic Fertilizers were applied 20 tons/ha, three weeks before sweet potato cuttings were planted. Observation of anatomical characteristics include: stomata density, length and width opened stomatal pores, mesophyll thickness, diameter of the petiole cortex. All characteristics were observed 60 days after planting (HST) using a microscope with a camera Optilab 2.2 and image capture with the Image Raster 3.0 program. Anatomical characters of sweet potatoes showed stomata density J1, J2, J3, J4, J5 and J6 respectively 8.56; 6.11; 5.11; 6.33; 8.56 and 9.11 in 75796.36 µm. The length of opened stomata pores were 26.99 µm, 32.08 µm, 28.96 µm, 38.72 µm, 39.52 µm and 35.86 µm. The width of opened stomata pores were 7.95 µm, 5.11 µm, 4.67 µm, 5.04 µm, 9.28 µm and 6.98 µm. The mesophyll thickness were 364,986 µm, 280,703 µm, 389,743 µm, 245,749 µm, 261,439 µm and 434,913 µm. The diameter of the petiole cortex were 454,030 µm, 373,453 µm, 456,439 µm, 373,001 µm, 275,647 µm and 318,785 µm. Five sweet potato varieties differ in anatomical characters.

1. Introduction

Sweet potato (*Ipomoea batatas*) is one of the tubers of carbohydrate sources in addition to wheat, rice, corn, potatoes and cassava. Besides containing carbohydrates, sweet potatoes also contain vitamins A and C, minerals potassium, phosphorus, calcium and sodium. Sweet potatoes are not only used as food but also as industrial raw materials and animal feed [1][2]. Sweet potatoes also contain beta carotene, anthocyanin, phenolic compounds, essential amino acids such as lysine and tryptophan [3][4].

Potential of sweet potato as a functional food continues to be developed to support food diversification programs. Various research on plant carbohydrate substitute for rice continues to be developed, including research on sweet potato plants. Research Institute for Various Nuts and Tuber of Malang City, until 2016, has released 24 superior varieties of sweet potatoes, where each variety has specific resistance properties [5]. Plant resistance to pests and pathogens can be antixenosis/non-

preferences, antibiosis and tolerant. The morphoanatomy characteristics of antixenotic resistances of plants are color, shape, cell wall thickness, proliferation rate of plant tissue, hardness, characteristics of the stem, trichomes, magnitude of stomata, cuticle layer and wax [6][7].

The anatomical structure of the leaves is arranged by three tissue systems namely epidermis, mesophyll and vascular tissue. The epidermis is a layer of outer cells and covers the surface of plant organs. In leaf organs, the epidermis is on the adaxial and abaxial side [8]. Epidermal cells in later development are modified and specialized into structures in the form of stomata, trichomes, cuticles and fan cells. Stomata generally occur on the abaxial side of the leaves, but in certain plants the stomata are found on the adaxial and abaxial sides of the leaves. The shape and type of stomata are anomocytic (ranunculaceous), anisocytic (cruciferous), parasitic (rubiaceous), dichitic (caryophyllaceous) and actinositic [9][10]. Stomata together with neighboring cells are called stomata complexes.

Anatomical characters are antixenotic resistance parameters, can be in the form of color and plant shape, cell wall thickness and proliferation rate of plant tissue, hardness and characteristics of the stem, trichomes, magnitude of stomata, cuticle layer and wax [11][12][13]. The anatomical characteristics of the leaves can be seen through the length and width of the stomata, the density of the stomata, the thickness of the mesophyll and the length of the petioles [14][15]. Six varieties of sweet potatoes show specific resistance to pests and pathogens, characterized by qualitative and quantitative morphology, the ability to synthesize primary and secondary metabolites including the anatomical characteristics of leaves. Description of anatomy and morphology is a product of the response to the conditions of the environmental factors in which the plant grows. Application of organic fertilizer will provide mineral nutrients [16-17], so that plants can grow optimally and can induce the nature of antixenosis resistance, antibiosis and tolerance.

Six varieties of sweet potatoes showed specific resistance to pests and pathogens, characterized by qualitative and quantitative morphology, the ability to synthesize primary and secondary metabolites including the anatomical characteristics of leaves. Description of anatomy and morphology is a product of the response to environmental factors in which the plant grows. Application of organic fertilizer will provide mineral nutrients [16], so that plants can grow optimally and can induce the nature of antixenosis resistance, antibiosis and tolerance.

In Tomohon City, there are two local varieties of sweet potatoes with purple and white bulbs, with local names Kekeruten. These types of sweet potatoes grow wild on agricultural land. Their leaves are often used as animal feed. Their growth properties with creeping stems provide distinctive morphological structure, even showed specific properties of antibiotic resistance, antixenosis, and tolerance. Morphological hostage to find out the diversity of local cultivars will provide benefits to improve germplasm conservation.

The study of morphological diversity in sweet potatoes is the morphological diversity of local sweet potato from Muna [17], sweet potato morphology and kinship, and the correlation of anatomical characteristics of disease resistant sweet potato cultivars [18][19]. Morphological characteristic study of six sweet potato varieties was carried out to obtain morphological descriptions as scientific information that could be used to develop resistant varieties to pests and diseases. The study aimed to evaluate the anatomical characteristics of six sweet potato varieties (*Ipomoea batatas*) after the induction of organic fertilizer.

2. Research Methods

The study was conducted in the experimental garden in Lansot Village, Tomohon City. Research used Completely Randomized Block Design with three replications. Treatment of sweet potato varieties (J): Antin 3 (J1), Jago (J2), Cilembu (J3), Shi Royutaka (J4), Local Purple (J5) and Local White (J6). Petroganic organic fertilizer (20 tons/ha) were applied in three weeks before sweet potato cuttings were planted. The observed anatomy characteristics include: stomata density, length and width of opened stomata pores, thickness of mesophyll, 5) diameter of the petiole cortex. The observations were obtained at 60 days after planting (HST) using a microscope with a camera Optilab 2.2 and image capture with the Image Raster 3.0 program.

3. Results and Discussion

The results of longitudinal slices of the epidermal surface on the abaxial side of sweet potato leaves were shown in Figure 1. The stomata of sweet potato had paracytic kidney type, marked by closed cells surrounded by two neighboring cells with their longitudinal axis parallel to closed cell axis.

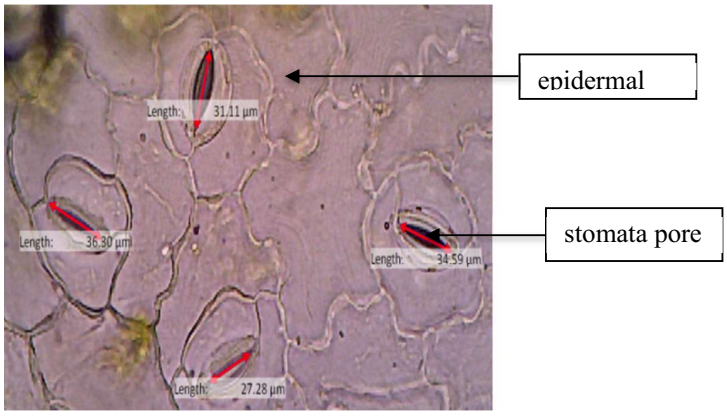

Figure 1. Anatomical structure of epidermal and stomata cells (40 times magnification)

The leaf epidermis consists of one cell layer, not plastid except in the stomata cover cell. The epidermal cell wall contains a cuticle layer. The epidermal tissue is the outermost tissue of plant organ, where the cell wall that builds it has undergone structural development according to the plant habitat. The cell wall consists of 3 layers, middle lamella, primary wall and secondary wall. The primary wall contains 25-50% hemicellulose, 10-35% pectic substances, about 10% of extensine proteins and lectin proteins[9,20]. The cell wall consists of cellulose microfibrils and nonselulose matrix. The matrix generally consists of pectin and hemicellulose. In the cell wall lignin, suberin, warts, wax, tannin, calcium carbonate and calcium oxalate, silica and other ingredients are found [20][21][22].

The results of measured anatomical characteristics of J1, J2, J3, J4, J5 and J6 were 8.56, 6.11, 5.11, 6.33, 8.56 and 9.11 respectively in 75796.36 µm. The length of opened stomata pores were 26.99 µm, 32.08 µm, 28.96 µm, 38.72 µm, 39.52 µm and 35.86 µm. The width of opened stomata pores were 7.95 µm, 5.11 µm, 4.67 µm, 5.04 µm, 9.28 µm and 6.98 µm. The highest average stomata density on J6, the highest average length of stomata pore on J5, and the highest average width of opened stomata pore on J5.

The anatomical structure of stomata is strongly associated with ontogeny stomata. Stomata are usually found in plant organs associated with air. Stomata distribution, length and width pore are closely related to plant physiological processes, namely transpiration, photosynthesis and respiration. The flexibility of the stomata pore wall allows the stomata to open and close [23]. The number and density of stomata between cultivars are influenced by the environment in which they grow. Light intensity, water availability and temperature are factors that determine stomata density [24].

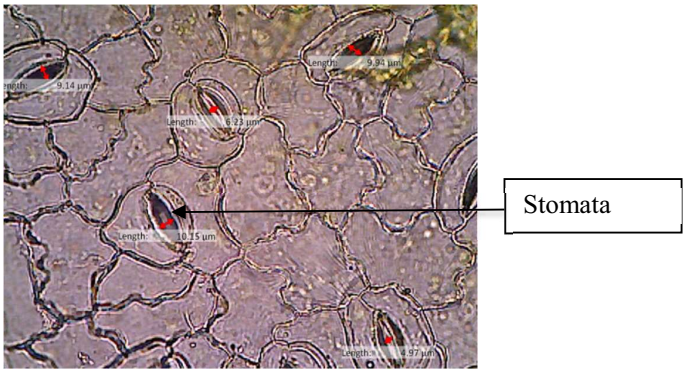

Figure 2. The width of opened stomata pore (40 times magnification)

Figures 1 and 2 show the length and width of opened stomata pore. The opening and closing the stomata is affected by closing cell turgor. Stomata open when the cell closure is high. The changing of osmotic potential value in the closing cell is caused by chemical changes that occur in the cell closure which will then change the potential of the water. Environmental factors that influence the opening of the stoma cover are the concentration of CO_2, light, water stress, temperature and wind [25].

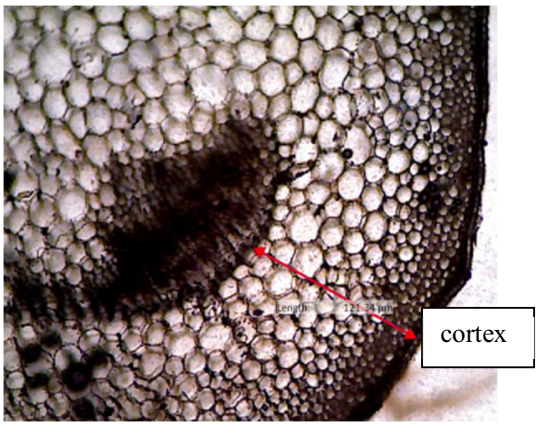

Figure 3. Petiole transverse slices (4 times magnification)

Figure 3 showed a cross section of sweet potato petiole. Petiole is composed of three regions namely epidermis, cortex and stele. The petiole epidermis is the outer layer of cells, covering the organ with a protective tissue function. The shape, size and composition of the epidermal cells are very tight to each others to form layers without space between cells. The epidermis is composed by a layer of cells, in the form of living cells, the cells have the power to divide and do not carry secondary thickening growth[22-25]. The petiole cortex is composed by rounded parenchymal tissue with real intercellular space. The results of average diameter of the petiole cortex of sweet potatoes J1, J2, J3, J4, J5 and J6 were 454,030 µm, 373,453 µm, 456,439 µm, 373,001 µm, 275,647 µm and 318,785 µm respectively. The highest petiole cortex diameter on J3.

Mesophyll leaves are built by the parenchymal tissue with a transport file. Palisade parenchyma forms elongated cells, arranged in solid bonds and containing chloroplasts. Parenchymal sponges are composed of cells that are irregular in shape, contain chloroplasts and have large interstellar space. Each cultivar has different thickness variation[26]. The average mesophyll thickness of J1, J2, J3, J4, J5 and J6 were 364,986 µm, 280,703 µm, 389,743 µm, 245,749 µm, 261,439 µm and 434,913 µm respectively. The highest mesophyll thickness was J6. The leaves that have thick mesophyll indicated the availability of chloroplast in the palisade tissue and the high parenchymal sponge caused high effectiveness of photosynthesis.

Lamina thickness is influenced by the composition of leaf mesophyll, namely parenchymal palisade and parenchymal sponge. In the growth and development of plants, the availability of essential water and nutrients is the main factor for optimal growth, including growth and development of leaves. In mesophyll leaves, there is a network of xylem and phloem transport which is a continuation of the rod transporting file. Morphological characteristic such as lamina thickness is closely related to resistance to suckling pests *Amrasca biguttula* in tobacco[26]. The thickness of the lamina is affected by the thickness of the cell wall that made up organ. The cell wall contains proteins and enzymes that actively work for the growth of thickened cell walls and strengthen cell walls during inductive resistance. When cells detect the presence of potential pathogens, enzymes catalyze and produce reactive oxygen molecules that can damage the cells of the invading organism.

The relationship with antixenotic resistance to cell wall thickness and leaf cuticle layer affect leaf hardness. The length and width of the stomata pores are related to insects that have a sucker-type mouth tool that can insert the stylet through the stomata opening. Stomata diameter does not have direct effect on movement and feeding activity but is related to the product of plant cell metabolism which is needed by insects for growth and development. Stomata diameter affects the amount of CO_2 diffusion for the synthesis of primary metabolite products, namely carbohydrates, proteins and fats,

and the synthesis of metabolites secondary to plant resistance. Stomata pore size is tightly regulated by plants, and cover cells participate in resistance by closing in response to the molecular pattern of microbial association.

4. Conclusion

Six sweet potato varieties showed differences in anatomical characteristics in terms of stomata density, length and width of opened stomata pores, mesophyll thickness and petiole cortex diameter. The highest mean stomata density was local white (J6) which is 9.11 in 75796.36 μm2. The average length and width of opened stomata pore was the highest in local purple (J5) which was 39.52 μm and 9.28 μm. The highest petiole cortex diameter in Cilembu (J3) was 456,439 μm.

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