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Morphology and Anatomy of Stomata of *Plagiobryoides cellularis* in Galunggung Mount Tourist Area Tasikmalaya

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Abstract. *Plagiobryoides cellularis* is a part of mosses that has stomata. Furthermore, *P. cellularis* has different characteristics compared to the other mosses. These facts prove that some Bryophytes have stomata. But unlike other land plants, Bryophyte stomata are located in a capsule. To understand the characteristic of P. cellularis stomata and all over morphologies, we utilize light microscope and SEM as the observation tools. *Plagiobryoide cellularis* from a morphological perspective indicates special characteristics from other mosses. Besides, the presence of stomata located in capsule, have barely similar to other stomata land plants. With stomata formed from 2 guard cells and surrounded by subsidiary cells that are hard to distinguish from epidermal cells.

Keywords: Morphology · Mosses · Plagiobryoides cellularis · Stomata

1 Introduction

Stomata (singular: stomata) are pores or apertures between 2 specialized cells that are called guard cell in the epidermis [2, 26]. The cell has different characteristics compared to the epidermal cell, expanded and flexed are one of the abilities of guard cell caused by it has a thin area in its cell wall [7, 35]. These abilities support stomata to open and close due to turgor pressure changes in guard cell or sometimes the pressure of neighbor cell affected too [16]. The mechanism of opening and closing in stomata, develop stomata's role to regulate gas exchange [16, 28]. Moreover, stomata can be found in any leaves from land plants, especially flowering plants [28, 33]. However, stomata can be found in Bryophyte too [20, 30].

In Bryophyte, not all of the groups have stomata [11, 15]. Liverworts are the only bryophyte devoid of stomata, but some mosses and hornworts do [4, 11, 23]. In mosses, stomata are located on the base of capsule (sporangium) [3, 20]. However, that information doesn't make all families in mosses have stomata. Several families or genera, stomata are absent, such as Takakia, Andreaea, Andreaeobryum, and the family of Archidaceae [22, 23, 27]. Furthermore, some mosses are losses and lack of stomata, with over 30 families and 74 genera identified [27]. Nevertheless, some of the families are having stomata, for example in Bryaceae [27, 34]. Bryaceae is a family of mosses that have a

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Fig. 1. Location of Galunggung Mountain. Three stations become location sampling for *Plagiobryoides cellularis.*

lot of species that are identified had stomata. For example, *Plagiobryoides cellularis* or most known as Bryum cellulare [14]. In P. cellularis, stomata are easy to find. Besides that, P. cellularis has special morphological characteristics that distinguish it from other mosses [6, 31].

Morphology characteristics are including gametophyte and sporophyte. Gametophyte involves characteristics of rhizoid, leaf, stem, and branch. Meantime sporophyte involves characteristics of seta and capsule (sporangium) [5, 24, 32]. Hence, morphological characteristics are one of the phylogeny determinants' aspects especially the effect of habitat on morphological characteristic changes [13].

The habitat of *Plagiobryoides cellularis* is rock and pantropic [6, 31]. The fact that P. cellularis is distributed in pantropic areas made Indonesia becomes a proper habitat, especially in the tourist area of Galunggung mountain [14]. It is a volcano area located in Tasikmalaya, West Java, Indonesia with 2,168 m above sea level or 1,820 m from the mainland of Tasikmalaya city, and the astronomic location at coordinates 7.25°–7°15′0″N and 108.058°–108°3′30″E [1]. With good and proper habitat, *Plagiobryoides cellu-lars* is abundant in Galunggung mountain.

2 Material and Methods

2.1 Study Area

The tourist area of Galunggung mountain becomes the location for sampling. This location was chosen because it was a proper habitat for mosses especially *Plagiobryoides cellularis* (Fig. 1).

2.2 Procedure Specimen Collection

Plagyobryoides cellularis is sample collected in the tourist area of Galunggung mountain, Tasikmalaya. Purposive sampling is used to collect P. celularis which is part of family Bryaceae, while Bryaceae is a family of mosses that have stomata are located in sporangium [14, 27]. P. cellularis is collected during the rainy season. It makes a significant chance for us to collect samples in a good condition. Accordingly, we can observe the samples from morphological and stomata characteristics.

2.3 Identification of Specimen

The result of identification specimen process is referred by Spence (2009) dan Lawson & Matthews (2020). Key determination is used as the identification process. The identification process is supported by many references such as e-Flora of South Africa (2018).

2.4 Light Microscope and SEM

Plagiobryoides cellularis with its substrate is put in a container with wet cotton inside, before that the cotton is watered with aquades to make the condition specimens still fresh. And then, part of P. cellularis from gametophyte and sporophyte was observed using microscope binocular CX-22 at magnification of 40×, 100×, and 400×. The capsule (sporangium) is splitting become 2 parts and manually separated from the spore using a toothpick. So we can observe the existence of stomata more clearly. For Scanning Electron Microscopy (SEM), the capsule is coated with gold coating so we can get the figure of surface area from the capsule (sporangium) [21, 27]. Scanning was used at magnification of 1 mm, 50 µm, 20 µm, dan 10 µm with SU2300 SEM.

2.5 Data Analysis

Analysis data using Miles and Huberman's model.The activity of analysis is divided into three concurrent and continuous activities: data condensation, data display, and conclusion/verification. The result of observation is visualized into table and figure that display the morphological characteristics of P. cellularis including gametophyte, sporophyte, and anatomy of stomata that are located in capsule (sporangium).

3 Result and Discussions

3.1 Morphology of Gametophyte

Plagiobryoides cellularis is acrocarpous mosses (Fig. 2A). Furthermore, it's because P. cellularis is part of Family Bryaceae, while Bryaceae is acrocarpous mosses [8, 14]. P. cellularis has rhizoids that attach to the stem. The rhizoids are brownish red (Fig. 2c). In mosses, rhizoids always uniseriate, also rhizoids of P. cellularis branched and multicellular. We can see that several rhizoids are made up of 2 cells or more. Besides that, it has several papillae because the fact that the rhizoids are roughness [34] (Fig. 2c). The stem is sympodial, brownish-red or green, and orthotropic. As a result of the orthotropic, the stem grows spreading or making an angle of >45º and the seta grows as though erecto-patent in its habitat. Not only seta, but leaf also grows erecto-patent in P. cellularis (Fig. 1a).

The leaves of P. cellularis are ovate-lanceolate form with acute apex form in the tip (Fig. 2B). The color of the leaves is green and grows in spiraling lines. Besides that, the leaves of P. cellularis are isophyllous and distichous. Furthermore, the leaves are arranged in two rows and typically the same size and shape for oldest or youngest leaves (Fig. 1B). And it didn't have wax to cover the surfaces of the leaves.

3.2 Morphology of Sporophyte

Sporophyte in *P. cellularis* started with seta that grows from the lateral of the stem and then the capsule grows curves towards seta (Fig. 2a, 3c). Seta isn't branched and the color is brownish red until brownish- -green when it comes with capsule (Fig. 2a, 3f). Seta connected capsule with gametophyte in P. cellularis. The capsule is green or brownish (Fig. 3a). In P. cellularis, the capsule isn't protected with calyptra.

Moreover, for immature capsule, it has an operculum (Fig. 3a–b). In P. cellularis, operculum shaped umbonate or convex with a rounded central point (Fig. 3a). Furthermore, the operculum is yellowish-green and the operculum will separate when the capsule matures (Fig. 3d–e). When the capsule isn't have an operculum, the capsule will release spore for reproduction. This is the reason why the immature capsule has an operculum to protect the capsule that is not ready to release spore (Fig. 3a–b).

In *P. cellularis*, the capsule has a hypnoid peristome or double peristome because the peristome separate into endostome and exostome (Fig. 3d). A peristome is a circle of teeth inside the mouth of a mosses capsule (Fig. 3e). Endostome is an inner circle

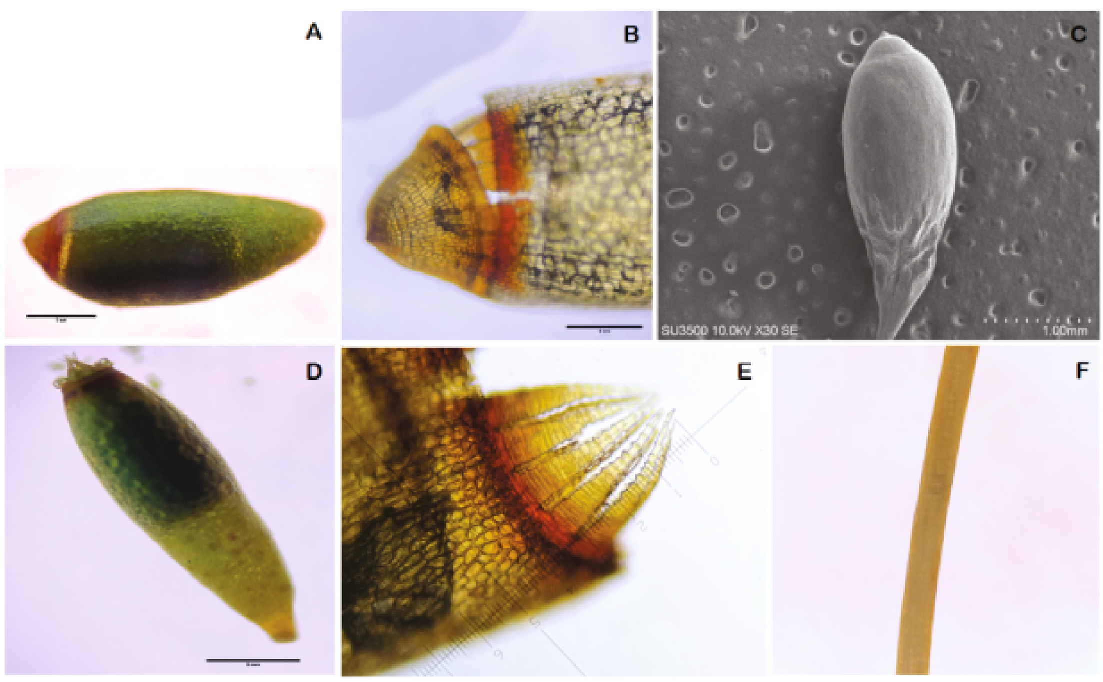

Fig. 2. Sporophyte of *Plagiobryoides cellularis.* **(a)** Immature capsule with operculum. **(b)** Operculum in capsule with magnification 400× in the light microscope. **(c)** Immature capsule in SEM. **(d)** The capsule with operculum that almost separate. **(e)** Exostome with magnification 400× in the light microscope. **(f)** The seta. Scala bars: **(a, b, d)** = 5.00 mm; **(c)** = 1.00 mm.

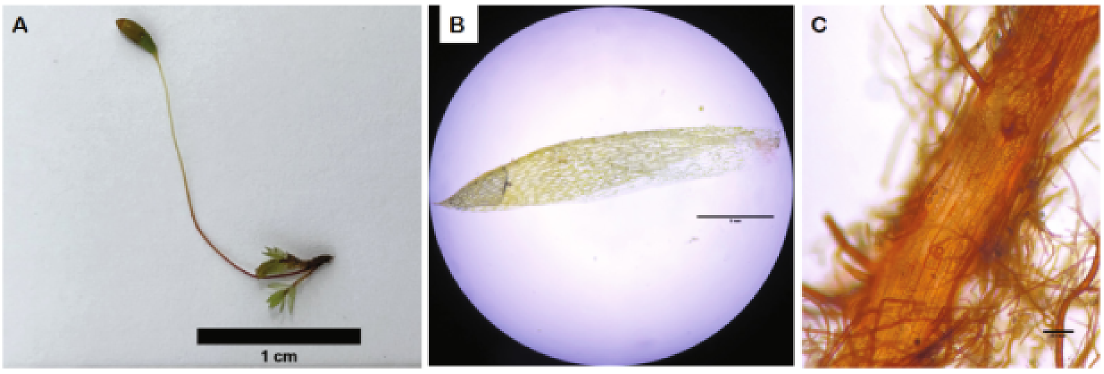

Fig. 3. Morphology of *Plagiobryoides cellularis*. **(a)** *Plagiobryoides cellularis*. **(b)** leaf with magnification 400× in the light microscope. **(c)** Rhizoids attach to the stem. Scala bars: (a) = 1 cm; (b) = 5.00 mm; (c) = 0.1 mm.

of teeth and the exostome is the outer circle of teeth. Exostome are yellowish-brown (Fig. 3e). Moreover, exostome are 92.5 µm from the top layer of the capsule (Fig. 3e).

3.3 Anatomy of Stomata

Stomata are located in the sporophyte of mosses, especially in the base of the capsule (sporangium) [19, 29]. However, *Plagiobryoides cellularis* stomata are located in the base of the capsule (Fig. 3d). Stomata in mosses can open and close like stomata in general [16] (Fig. 4a–c). When stomata are open, we can see the pore that creates because of the guard cell is swelling [9] (Fig. 4a, c). Otherwise, the guard cell is shrinking which make stomata close (Fig. 4b). Nevertheless, the contribution from the mechanism of open and close stomata still become a further study for researchers [3, 17]. Hence, *Plagiobryoides cellularis* has stomata in its capsule is important information. The diameter of stoma size (plural:stomata) is 45 µm and the size of the guard cell is 45 × 25 µm (Fig. 4k–l).

Besides the size of stomata and their mechanism, stomata in mosses can classify based on their position from the epidermis. Stomata can be at the same level as epidermis, raised, or sunken [23]. In *Plagiobryoides cellularis,* stomata are raised from the epidermis. The epidermis will look blurry when we observe the stomata using the light microscope (Fig. 4d). Furthermore, stomata spread randomly in the base of capsule, it can be solitary or stick with the other stoma [23] (Fig. 4d, f). The stick stomata happened when the same neighbor cell is directly side by side with 2 different guard cells (Fig. 4f). Moreover, stomata in mosses are anomocytic [19, 29]. The anomocytic is the type of stomata when the neighbor cells are hard to distinguish from epidermal cell or don't have neighbor cell either [10, 25, 26]. In P. cellularis, the stomata are anomocytic too (Fig. 4d–f, 4i–l).

In *Plagiobryoides cellularis*'s case, the neighbor cell is slightly distinguishable from the epidermal cell by shape and size. However, the neighbor cell often looks like epidermal cell but for the size of several neighbor, cells are bigger or smaller from epidermal cell (Fig. 4i). The size of neighbor cell is 25 × 37.5 µm while the size of epidermal cell is 35 × 30 µm (Fig. 4g–j) (Table 1). The epidermal cells contain chlorophyll (Fig. 4g–h). Not only the epidermal cells, but the guard cells of P. cellularis also contain chlorophyll

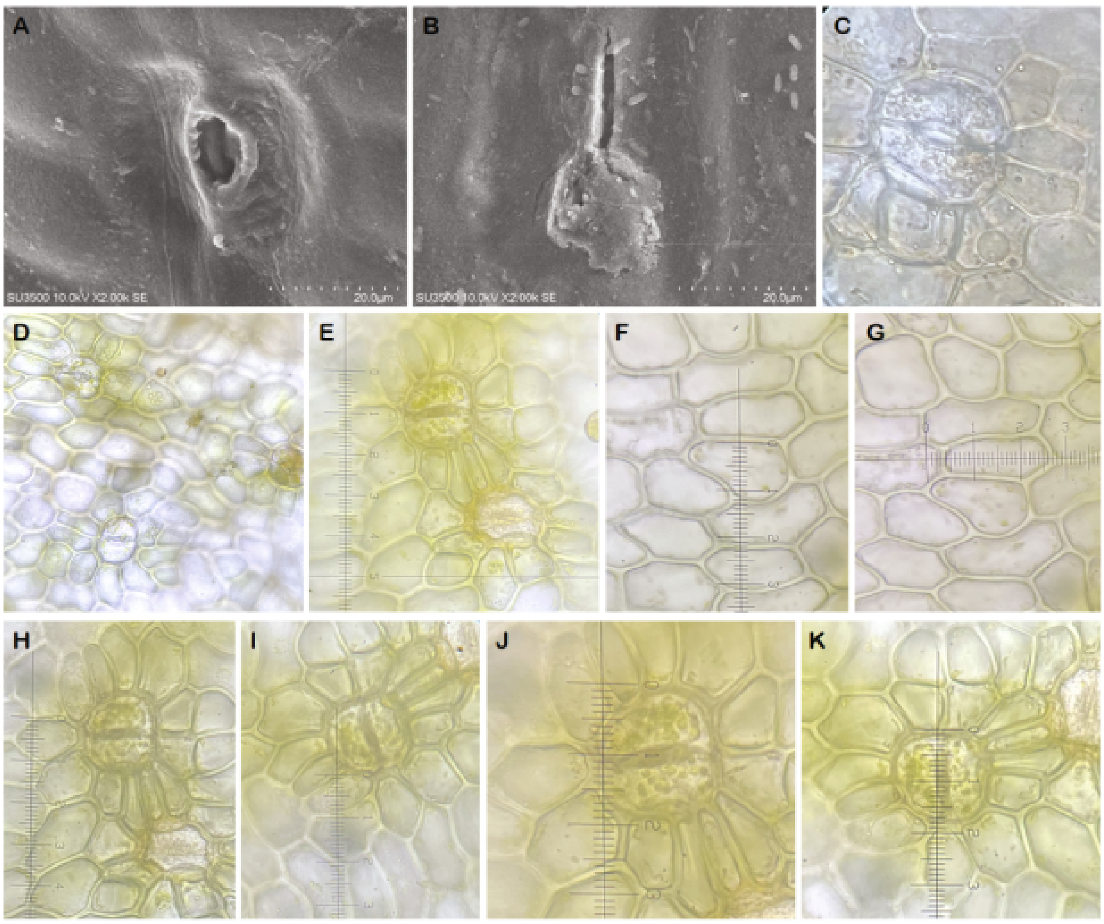

Fig. 4. Anatomy of Stomata. **(a)** The open stomata with magnification 20.0 µm in SEM. **(b)** The close stomata with magnification stomata 20.0 µm in SEM. **(c)** The open stomata with magnification 1000× in the light microscope. **(d)** The raises stomata from the epidermis. **(e)** stomata sticked. **(f)** The epidermal cell. **(g)** the epidermal cell. **(h)** The neighbour cell. **(i)** The neighbour cell. **(j)** The guard cell. **(k)** The guard cell.

Table 1. The Size of Stomata *Plagiobryoides cellularis*

	Stoma	Guard cell	Neighbor cell	Epidermal cell
Diameter	45 µm	–	–	–
Length	–	45 µm	25 µm	25 µm
Width	–	25 µm	37.5 µm	30 µm

too (Fig. 4f, k). Instead, whether chlorophyll in the guard cell is used from photosynthesis or not is still become a further study for researchers [18].

4 Conclusions

Plagiobryoides cellularis is a part of mosses that has stomata. Furthermore, *P. cellularis* has different characteristics compared to the other mosses. These facts prove that some Bryophytes have stomata. But unlike other land plants, Bryophyte stomata are located in a capsule. To understand the characteristic of *P. cellularis* stomata and all over morphologies, we utilize light microscope and SEM as the observation tools. *Plagiobryoide cellularis* from a morphological perspective indicates special characteristics from other mosses. Besides, the presence of stomata located in capsule, have barely similar to other stomata land plants. With stomata formed from 2 guard cells and surrounded by subsidiary cells that are hard to distinguish from epidermal cells.

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