

The anatomical structure of the leaves in *Echeveria* sp.

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ABSTRACT. *Echeveria* is a genus of plants that has distinctive structure namely fleshy leaves. The study of anatomical structure of the *Echeveria* was still limited especially the leaf anatomy. This study aims to determine the anatomical structure of the leaves of several plants of the *Echeveria*. This study used a descriptive method to describe the anatomical structure of the leaves. The leaves were sectioned paradermally on both sides namely adaxial and abaxial and observed using a binocular microscope with 400X magnification. The parameters observed on the both side namely adaxial and abaxial were epidermal cells including shape, length and amount of the cells and stomata including distribution type, shape and type. The results showed that the anatomical structure of the leaf including the epidermal cells and stomata of several plants of the *Echeveria* varies. The shape of the epidermal cells found was square with curved and straight anticlinal anticlinal walls. The longest epidermal cells were found on the abaxial surface of the leaves of *Echeveria* 'Purple Pearl' which was 213 μm and the shortest epidermal cells were found on the adaxial surface of the leaves of *Echeveria* 'Black Chroma' namely 83.5 μm . The number of epidermal cells in each field of view ranged from 3 units/ cm^2 to 16 units/ cm^2 . The distribution type of stomata on the seven leaves observed was amphistomatic. The forms of stomata found were amarylliacae, mniium and helleborus. The type of stomata observed is anisocytic.

Keywords: Crassulaceae; epidermis; free-hand section; leaf anatomy; stomata

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INTRODUCTION

Species of plants *Echeveria* known as the cabbage cactus (Astriani *et al.*, 2020) Based on the growth time, plants of *Echeveria* are included in the type of *Spring/Fall* that grows fast and be fertile in the spring while in the dry season it grows infertile. However, as a country that does not have spring, plants from *Echeveria* could live in Indonesia because the plants are able to adapt well. Moreover, as the belonging of Crassulaceae, *Echeveria* carry out Crassulacean Acid Metabolism (CAM) as a photosynthetic pathway to adapt in dry conditions (Karwowska *et al.*, 2015; Jimeno & Kromer, 2013; Hastilestari, 2015; Ripley *et al.*, 2013) *Echeveria* plants can be cultivated vegetatively by using leaf cuttings. Plants from this genus have leaves that are shaped like flowers with a textured surface like wax (Jessar *et al.*, 2021). In general, plants of *Echeveria* have a short stem, the arrangement of leaves is not scattered and not branched. Plants from this genus are easy to find as well as their distinctive shape and uncomplicated maintenance make plants from this genus *Echeveria* used as an ornamental plant (Astriani *et al.*, 2020).

Previous research on the crassulaceae family had been carried out by observing the anatomical structure of the root of and *Rhodiola rosea* L., leave of *Crassula cordata* and *Echeveria gigantea*, and stem of *Kalanchoe turbiflora* and *Sempervivum tectorum*. The root anatomy of *Rhodiola rosea* L. and *Sempervivum tectorum* L. consist of epidermal tissue, collenchyma, and parenchyma. Both of them have an epidermal tissue that consist of thick parenchyma, a transport tissue namely xylem and floem, and cortex tissue composed of collenchyma (Maftei & Maftei, 2019; Kirilenko, 2016).

MATERIALS AND METHODS

This research is qualitative research using descriptive method. Observations on the anatomical structure of the leaves of several plants of the *Echeveria* genus were carried out at the Biology Education Laboratory, FKIP, Sriwijaya University. The observed leaf parts were the epidermal in the upper (adaxial) and lower (abaxial) paradermal sections. Parameters observed in paradermal sections

were variations in the shape of the epidermal cells, the length of the epidermal cells, the amount of epidermal cells and the characteristics of the stomata namely shape, type, and distribution type.

The sectioning method used is manual or free-hand section. This method is a sectioning method that is often used to observe the anatomical structure of plants because it is fast and inexpensive (Lux *et al.*, 2005). Previous study had done before, part of the leaf that observed were epidermal cells and stomata (Cristanti, 2021). In this research, the observations used paradermal sections on each leaf to observe epidermal cells (shape, amount and size of cells) and stomata (type of distribution, shape and type).

The materials used were *Echeveria agavoides*, *Echeveria chantily*, *Echeveria 'afterglow'*, *Echeveria 'Purple Pearl'*, *Echeveria 'Black chroma'*, *Echeveria 'Peach Pride'*, and *Echeveria laurensis*, safranin, tissue, distilled water and label paper. The steps are by preparing all tools and materials. Then, make paradermal sections using blade of each leaf of plants from the *Echeveria*, namely *E. chantily*, *E. 'Black Chroma'*, *E. 'Peach Pride'*, *E. agavoides*, *E. laurensis*, *E. 'Purple Pearl'* and *E. 'Afterglow'*. Next, put each section to each object glasses. Furthermore, drops amount of safranin to object glasses and cover the sections using a cover glass. Then, adhere a label containing the identity of the section used. Last, observe each samples using a *Bioeco Germany* binocular microscope and do documentation using *Canon* camera on each observation result.

RESULTS AND DISCUSSION

The results showed that the *Echeveria* genus has various epidermal cell shapes namely square with square and wavy anticlinal walls (Table 1). The average length of epidermal cells also varies from 83.5 μm to 213 μm . The longest epidermal cell is found on the abaxial surface of the *Echeveria 'Purple Pearl'* plant, which is 213 μm . The shortest epidermal cells belong to the upper surface of the *Echeveria 'Black Chroma'*, which is 83.5 μm . The number of epidermal cells in each field of view ranged from 3 units/ cm^2 to 16 units/ cm^2 .

Table 1. The Structure of Leaf's Epidermal Cell in *Echeveria* sp.

No	Plant	LS	Average ECL (μm)	ECA (unit/ cm^2 BP)	ECS
1	<i>Echeveria 'Black Chroma'</i>	Ad	83,5	11	SS
		Ab	116,4	10	SS
2	<i>Echeveria 'Peach Pride'</i>	Ad	186	5	WS
		Ab	153	3	SS
3	<i>Echeveria Chantily</i>	Ad	169	3	WS
		Ab	190	3	WS
4	<i>Echeveria 'Purple Pearl'</i>	Ad	160	8	SS
		Ab	213	4	SS
5	<i>Echeveria agavoides</i>	Ad	181	7	SS
		Ab	143	8	SS
6	<i>Echeveria laurensis</i>	Ad	151	6	SS
		Ab	141	4	SS
7	<i>Echeveria 'afterglow'</i>	Ad	112	16	WS
		Ab	130	9	WS

Note: LS: Leaf Surface; ECA: Epidermal Cells's Amount; Ad: Adaksial; Ab: Abaxial; ECL: Epidermal Cells's Length; ECS: Epidermal Cell's Shape; SS: Square; WS: Wavy Square

Based on Table 1, it is known that epidermis cell's shape of leaves of several plants of *Echeveria* are square with straight and wavy anticlinal walls. The straight shape of epidermal cell was found on adaxial and abaxial surfaces of *Echeveria laurensis*, *Echeveria agavoides*, *Echeveria 'Purple Pearl'*, and *Echeveria 'Black Chroma'* and the abaxial of *Echeveria 'Peach Pride'*. Meanwhile, the epidermal cell's shape of the adaxial of *Echeveria 'Peach Pride'*, both surfaces of *Echeveria 'Afterglow'* and *Echeveria chantily* were square with wavy anticlinal walls.

This research shows that the shape of the stomata varies while the type and distribution type are same (Table 2). Observation of stomata on both sides of the leaves showed that the shape of the stomata on each leaf are amarylliaceae, mniium, and helleborus. However, the type of stomata in all species shows the same type, namely anisocytic. Meanwhile, the stomata on the leaves are spread both in adaxial and abaxial surface (amphistomatic) (Table 2).

Table 2. The Characteristics of Stomata of Some Leaves from Echeveria Genus

No	Plant	Distribution type	LS	Stomata's shape	Stomata's type
1	<i>Echeveria</i> 'Black Chroma'	Amphistomatic	Ad	Amarylliaceae	Anisocytic
			Ab	Amarylliaceae	Anisocytic
2	<i>Echeveria</i> 'Peach Pride'	Amphistomatic	Ad	Mniium	Anisocytic
			Ab	Mniium	Anisocytic
3.	<i>Echeveria chantily</i>	Amphistomatic	Ad	Helleborus	Anisocytic
			Ab	Helleborus	Anisocytic
4	<i>Echeveria</i> 'Purple Pearl'	Amphistomatic	Ad	Mniium	Anisocytic
			Ab	Mniium	Anisocytic
5	<i>Echeveria agavoides</i>	Amphistomatic	Ad	Helleborus	Anisocytic
			Ab	Helleborus	Anisocytic
6	<i>Echeveria laurensis</i>	Amphistomatic	Ad	Mniium	Anisocytic
			Ab	Mniium	Anisocytic
7	<i>Echeveria</i> 'afterglow'	Amphistomatic	Ad	Mniium	Anisocytic
			Ab	Mniium	Anisocytic

Based on Table 2, it is known that stomata are spread on both leaf surfaces, namely abaxial and adaxial. The forms of stomata obtained are amarylliaceae, mniium, and helleborus. The shape of amarylliaceae stomata is found on the leaves of *Echeveria* 'black chroma'. Meanwhile, the leaves of *Echeveria* 'Peach Pride', *Echeveria laurensis*, *Echeveria* 'afterglow' and *Echeveria* 'Purple Pearl' have Mniium stomata shape. While, helleborus stomata are found in *Echeveria chantily* and *Echeveria agavoides*. The type of the stomata on the several plants of *Echeveria* are anisocytic.

Echeveria '**Black Chroma**'. Observation of the anatomical structure of the leaves of *Echeveria* 'Black Chroma' shows that the adaxial and abaxial surfaces of the leaves have a square with straight anticlinal walls of epidermal cells. The average length of epidermal cells on the adaxial surface is 83.5 μm . However, the average length of epidermal cells on the abaxial surface is 116 μm . The number of epidermal cells on the adaxial surface is 11 units/cm² field of view while on the abaxial is 10 units/cm² field of view. Amarylliaceae-shaped stomata are found on both leaf surfaces. The type of stomata found on both leaf surfaces is anisocytic. The distribution type of stomata is amphistomatic.

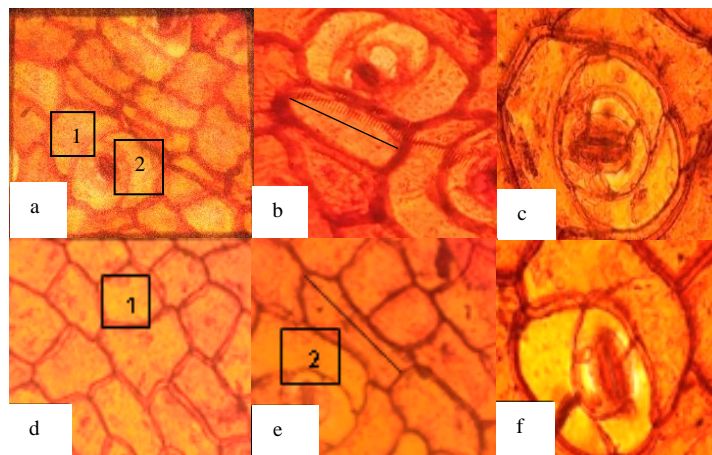


Fig. 1. Epidermal tissue and stomata on adaxial (a-c) and abaxial (d-f) leaf of the *Echeveria* 'Black Chroma'. A. Amount of epidermal cells; B. Measurement of epidermal cell length; 1. Straight square cell's shape; 2. Anisocytic stomata (P. 400X); C. Amarylliaceae stomata (P. 400X)

Echeveria 'Peach Pride'. On the upper surface of *Echeveria 'Peach Pride'* leaves, the epidermal cells are square with wavy anticlinal walls. However, the epidermal cells' on the abaxial are square with straight anticlinal walls. The average length of epidermal cells on the adaxial is 186 μm while on the abaxial surface is 153 μm . The amount of epidermal cells on the adaxial of the leaf is 5 units/cm² field of view while on the lower surface of the leaf is 3 units/cm² field of view. The form of stomata found on both leaf surfaces is Mniium. The type of stomata found on both leaf surfaces is anisocytic. Stomata in this leaves have an amphistomatic distribution type.

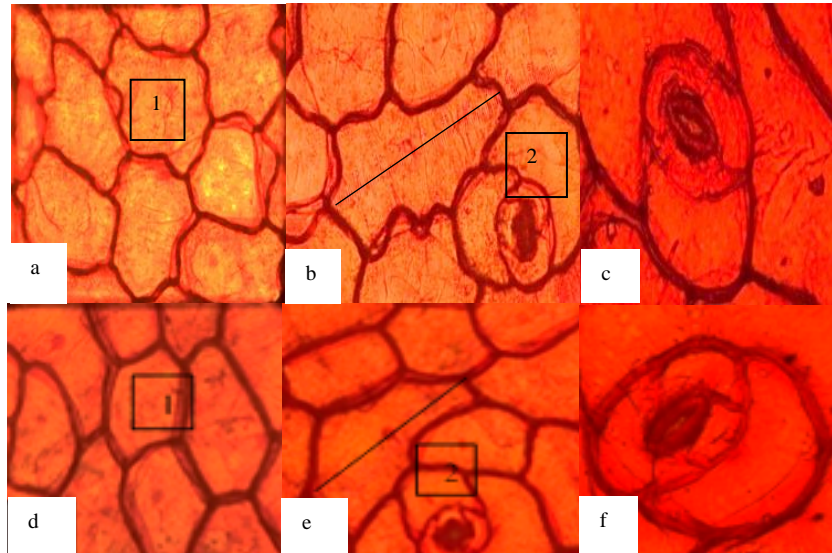


Fig. 2. Epidermal tissue and stomata on adaxial (a-c) and abaxial (d-f) leaf of the *Echeveria 'Peach Pride'*. A. Amount of epidermal cells; B. Measurement of epidermal cell length: 1. Wavy square cell's shape; 2. Anisocytic stomata (P. 400X); C. Mniium stomata (P. 400X)

Echeveria chantily. On the adaxial and abaxial surfaces of the leaves of the *Echeveria chantily*, there are square epidermal cells with wavy anticlinal walls. The average length of epidermal cells found on the adaxial surface is 169 μm . The average length of epidermal cells on the abaxial surface of the leaves is 190 μm . In one field of view, 4 units/cm² of epidermal cells are found on the adaxial surface. The amount of epidermal cells found on the abaxial surface was 3 units/cm². The shape of the stomata found on both leaf surfaces is helleborus. Leaves have anisocytic stomata on both adaxial and abaxial surfaces. Leaves have an amphistomatic type of distribution of stomata.

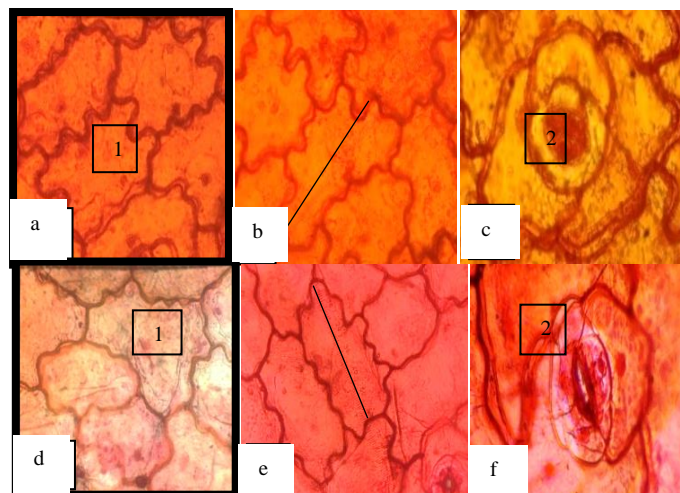


Fig. 3. Epidermal tissue and stomata on adaxial (a-c) and abaxial (d-f) leaf of the *Echeveria chantily*. A. Amount of epidermal cells; B. Measurement of epidermal cell length: 1. Wavy square cell's shape; 2. Anisocytic stomata (P. 400X); C. Stomata Form of Helleborus (P. 400X)

Echeveria 'Purple Pearl'. *Echeveria* 'Purple Pearl' have a square epidermal cell with straight anticlinal walls on adaxial and abaxial surfaces. Epidermal cell length on the adaxial surface is 160 μm while on the abaxial surface is 213 μm . On the adaxial, there are 4 units/cm² field of view epidermal cells and 7 units/cm² field of view on the abaxial surface. The shape of the stomata on both surfaces is mniium. The type of stomata in *Echeveria* 'Purple Pearl' is anisocytic that distribute amphistomatic.

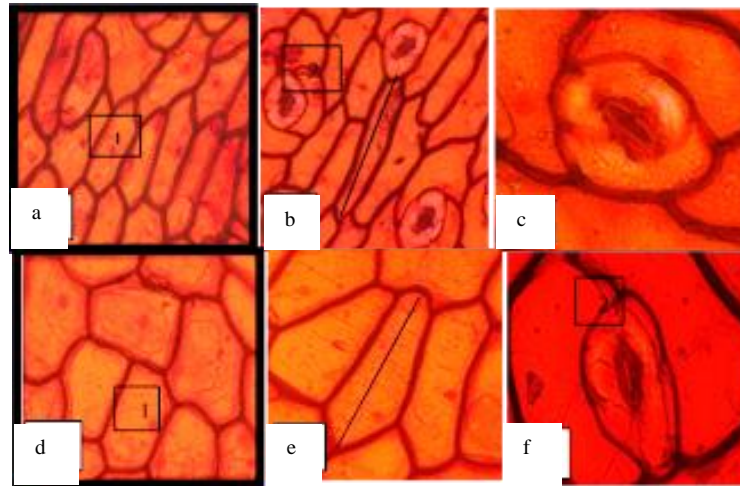


Fig. 4. Epidermal tissue and stomata on adaxial (a-c) and abaxial (d-f) leaf of the *Echeveria* 'Purple Pearl'. A. Amount of epidermal cells; B. Measurement of epidermal cell length: 1. Straight square cell's shape; 2. Type of Anisocytic (P. 400X); C. Shape of Stomata Mniium (P. 400X)

Echeveria agavoides. On the leaves of *E. agavoides* there are square-shaped epidermal cells with straight anticlinal walls on the abaxial and adaxial surfaces. The average length of epidermal cells on the adaxial surface is 182 μm while on the abaxial surface it is 143 μm . On the adaxial surface there are 7 units/cm² field of view epidermal cells and 8 units/cm² field of view on the abaxial surface. The shape of the stomata present on both surfaces is helleborus. The type of stomata scattered on both surfaces is anisocytic. Stomata are found on the abaxial and adaxial or amphistomatic.

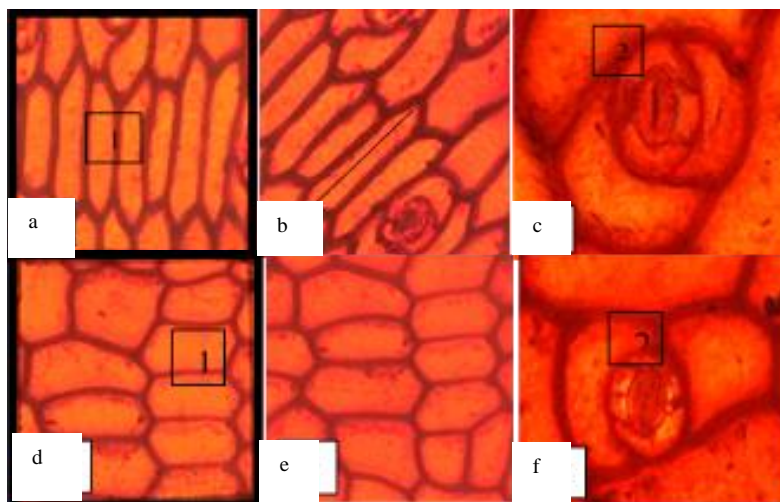


Fig. 5. Epidermal tissue and stomata on adaxial (a-c) and abaxial (d-f) leaf of the *Echeveria agavoides*. A. Amount of epidermal cells; B. Measurement of epidermal cell length: 1. Straight square cell's shape; 2. Anisocytic stomata (P. 400X); C. Helleborus stomata (P. 400X)

Echeveria laurenensis. Anatomical observations of the leaves of the *Echeveria laurenensis* plant show that the epidermal cells are square-shaped with square anticlinal walls on both surfaces. On the adaxial surface there are epidermal cells with an average length of 151 μm . on the abaxial surface there are epidermal cells with an average length of 142 μm . The amount of epidermal cells is 3 units/cm² field of view on the adaxial surface. As for the abaxial surface, there are epidermal cells as much as 7 units/cm² field of view. The shape of the stomata on both surfaces is mniium. Stomata scattered on both surfaces have the same type, namely anisotic. On the abaxial and adaxial surfaces are scattered or amphistomatic stomata.

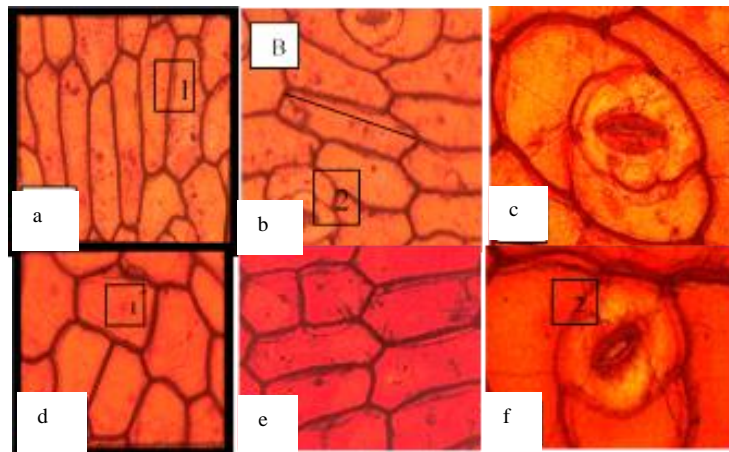


Fig. 6. Epidermal tissue and stomata on adaxial (a-c) and abaxial (d-f) leaf of the *Echeveria laurenensis*. A. Amount of epidermal cells; B. Measurement of epidermal cell length: 1. Straight square cell's shape; 2. Anisocytic stomata (P. 400X); C. Helleborus stomata (P. 400X)

Echeveria 'afterglow'. Epidermal cells found on the adaxial and abaxial surfaces of the leaves of the *Echeveria 'afterglow'* plant are square with wavy anticlinal walls. On the adaxial surface, the average length of epidermal cells was 112.5 μm , while on the abaxial surface it was 130 μm . The amount of epidermal cells found on the adaxial surface is 13 units/cm² field of view while on the abaxial surface it is 8 units/cm² field of view. On the abaxial and adaxial surfaces are scattered stomata with a mniium shape and anisocytic type. Stomata are scattered on adaxial and abaxial surfaces or are called amphistomatic.

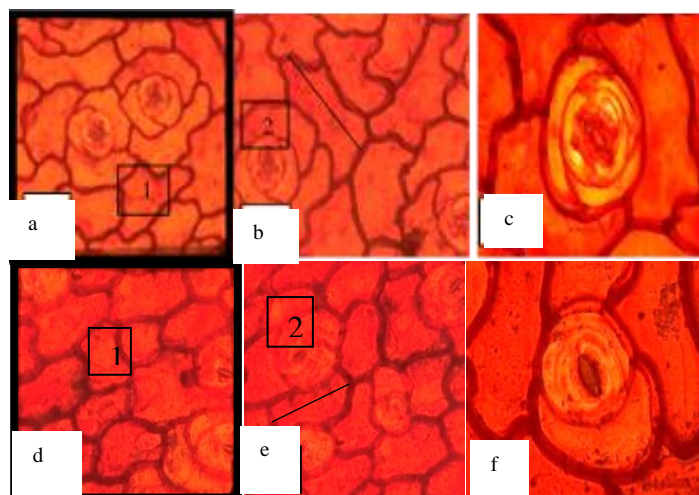


Fig. 7. Epidermal tissue and stomata on adaxial (a-c) and abaxial (d-f) leaf of the *Echeveria 'afterglow'*. A. Amount of epidermal cells; B. Measurement of epidermal cell length: 1. Wavy square cell's shape; 2. Anisocytic stomata (P. 400X); C. Mniium stomata (P. 400X)

Based on the results of observations about the anatomical structure of the leaves of several plants from *Echeveria* genus, namely *E. chantily*, *E.* 'Black Chroma', *E.* 'Peach Pride', *E. agavoides*, *E. laurensis*, *E.* 'Purple Pearl' and *E.* 'afterglow', it can be seen that the shape of the epidermal cells, the length of the epidermal cells, and the amount of epidermal cells as well as the shape of the stomata have variations. Meanwhile, the type of stomata and the distribution type of stomata obtained were same namely anisocytic and amphistomatic.

According to the observations that had been done, it is known that the anatomical structure of the leaves of seven plants of *Echeveria*, namely *E. chantily*, *E.* 'Black Chroma', *E.* 'Peach Pride', *E. agavoides*, *E. laurensis*, *E.* 'Purple Pearl' and *E.* 'Afterglow' has a distinctive character. The shape of the epidermal cells found in the seven leaves of the *Echeveria* are square with straight and wavy anticlinal walls. The straight shape of epidermal cell was found on adaxial and abaxial surfaces of *E. laurensis*, *E. agavoides*, *Echeveria* 'Purple Pearl', and *Echeveria* 'Black Chroma' and the abaxial of *Echeveria* 'Peach Pride'. Meanwhile, the epidermal cell's shape of the adaxial of *Echeveria* 'Peach Pride', both surfaces of *Echeveria* 'Afterglow' and *E. chantily* were square with wavy anticlinal walls. Previous research also showed that the epidermal cell's shape can be square or polygonal with straight and wavy anticlinal walls such as *Echeveria gigantea* and *Echeveria gibbiflora* (Sandoval-Zapotitla *et al.*, 2019). The presence of epidermal cells with stomata can loss the inner leaf temperature lower than the environment (Sandoval-Zapotitla *et al.*, 2019). As morphologically, the epidermal cells in each plant have a different shape (Anu *et al.*, 2017). While, ontogenically, the epidermal is a tissue which is arranged equally (Anu *et al.*, 2017). Epidermal cells are the outermost layer found in leaves, flowers, fruit, seeds, roots and stems before secondary thickening occurs (Sabandar *et al.*, 2021; Muthi'ah & Ayun, 2022).

The length of the epidermal cells in each plant also varies, as in the seven leaves of the *Echeveria* plant. On the adaxial surface of *Echeveria* 'Black Chroma' leaves, the average length is 83.5 μm while on the abaxial surface it is 116.4 μm . The length of the adaxial surface epidermal cells on *Echeveria* 'Purple Pearl' leaves is 160 μm while on the abaxial surface it is 213 μm . The average length of epidermal cells on the adaxial of *Echeveria* 'Peach Pride' leaves is 186 μm while on the adaxial surface is 153 μm . The average length of epidermal cells found on the adaxial surface of *Echeveria chantily* is 169 μm while the average length of epidermal cells on the abaxial surface of leaves is 190 μm . The average length of epidermal cells on the adaxial surface of *Echeveria agavoides* is 181 μm while on the abaxial surface it is 143 μm . On the adaxial surface of *Echeveria laurensis*, there are epidermal cells with an average length of 151 μm while on the abaxial surface there are epidermal cells with an average length of 141 μm . On the adaxial laves of *Echeveria* 'afterglow', the average epidermal cell length was 112 μm while on the abaxial surface it was 130 μm . Variations in the size of epidermal cells are influenced by genetic factors as well as environmental factors. According to (Sabandar *et al.*, 2021), the size of epidermal cells is greatly influenced by the environment. The increase in the size of the epidermal cells is related to the length and width of the leaf blade (Jessar *et al.*, 2021).

CONCLUSION

The anatomical structure of the leaves of several plants of *Echeveria*, including *Echeveria chantily*, *E.* 'Black Chroma' *E.* 'Peach Pride', *E. agavoides*, *E. laurensis*, *E.* 'Purple Pearl' and *E.* 'Afterglow'. The shape of the epidermal cells, the size of the epidermal cells, and the amount of epidermal cells as well as the shape of the stomata varies greatly. Meanwhile, the type of stomata and the type of distribution of stomata obtained were same. Epidermal cells of the leaves of several plants of the genus *Echeveria* were found to be in the shape of square with straight and wavy anticlinal walls with an average length ranging from 83,5 μm to 245 μm and the amount of epidermal cells ranged from 3 units/cm² field of view to 16 units/cm² field view. Stomata found on the leaves of several plants of the genus *Echeveria* have the shape of amarylliacae, helleborus and mniium with anisocytic stomata type and amphistomatic distribution type. Researchers suggest observing the anatomical

structure of the leaves of several plants of *Echeveria* sp. using a microtome and conducting further research with samples of other organs, including the roots and stems of *Echeveria* sp.

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