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Cell Regeneration Capability in Lizards (*Mabouya multifasiata*) and Flat-Tailed House Gecko (*Hemidactylus platyurus)*

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ABSTRACT

Regeneration signifies a mechanism for regaining the original function through the creation of a new cell or tissue when damage or loss of function results in the failure to play a given role. Regeneration is the ability to recover that is shared by all organisms, from microorganisms to humans. The phenomenon of regeneration, however, manifests very differently depending on the organism. One of the organism can regenerate their body are *Mabouya multifasiata* and *Hemidactylus platyurus*, has the ability to autotomize the tail as a self-defense mechanism. This study aims to obtain cell regeneration during autotomy of lizards (*M.multifasiata*) and flat-tailed gecko (*H.platyurus*). This research was conducted at the Intergrated Laboratory of UIN Raden Fatah and Dyatnitalis Laboratory Palembang. This research method is Quantitative Descriptive, the results of morphometric measurements of morphology and the development of tail cell histology tissue. The models used in these studies have offered a new platform for investigations of the morfology and histology underlying regeneration in Reptiles. *M. multifasiata*, morphometric shown growth development same as *H. platyurus*. From *Hematoxylin-eosin* (HE) staining showed the development of cell regeneration from Day 1 in Lizards which is called the initial sample, the 15th tail results and the 30th tail results days after autotomy showed cell growth on day 15 with the discovery of muscle cells (muscles), fat tissue (perivertebral fat tissue), cartilage (cartilagous tube), central nervous system (meninx) and Ependimal cells (ependymal cell). The ability to regenerate tail cells is compiled from the number of accretions of cartilage cells (cartilagous tube) that are perfect on the 15th and 30th days. Flat-tailed gecko, HE staining, it showed the growth and development of cells in the 15th tail, on the 30th day it showed the presence of muscle cells (muscles), fat tissue (perivertebral fat tissue), cartilage (cartilaginous tube), the central nervous system (meninx) and ependymal cells. The ability to regenerate lizard cells is arranged by the perfect cartilage (cartilaginous tube) on day 30. This results what is known about the roles of regeneration, and compares regenerative with the mechanisms and function of apoptosis in development. Defining the complexity of regenerative will contribute to new knowledge and perspectives for understanding mechanisms of regeneration induction and regulation.

Keyword : cell regeneration, *Mabouya multifasiata*, *Hemidactylus platyurus*

INTRODUCTION

Every living animal has certain ways to avoid enemies and their predators. In *M. multifasiata* and *H. Platyurus* the method of self-protection is by tail autotomy, namely breaking off the tail at certain places along the tail which are called autotomy planes. The autotomy process, apart from surviving and avoiding enemies, can also function for movement, foraging and reproduction. Thus, tail regeneration is an important function for amphibians, reptiles and arthropods (Maginnis, 2006). Cell regeneration is a process that occurs when a cell is degenerative or damaged, the cell will be replaced by new cells that regenerate continuously so that they can form new, complex tissue. The subject of discussion about autotomy was first studied from a term by a scientist named Fredricd (1892) which was originally called "autotmie" which describes an animal adaptation by breaking off the tail because of the animal's ability to defend itself from predators. In lizards, lizards and flat-tailed house gecko, after the tail undergoes autotomy, a regeneration process will follow so that a new tail grows, this tail will replace the old tail, which is called a Regeneral tail. The size and shape of this tail is almost the same as a real tail (Balinsky, 1970). The change that occurs after the tail is broken is that the wound surface will be filled again by the tissue and cells around it. The tail regeneration process begins with the wound healing and dedifferentiation phase, the formation of new embryonal cells for regeneration (Blastema), followed by differentiation, morphogenesis and growth of a new or regeneralized tail. In the process of wound closure, cell proliferation occurs by mitosis, which then migrates or moves to the wound surface, so that the entire wound surface slowly closes (Soesilo, 1992).

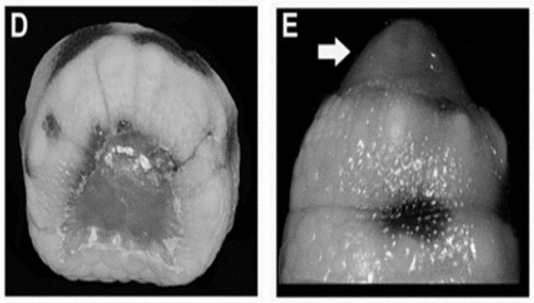

Figure 1. Phases of Blastema
(Gilbert, E., Payne, S., dan Vickaryous, M., 2013)

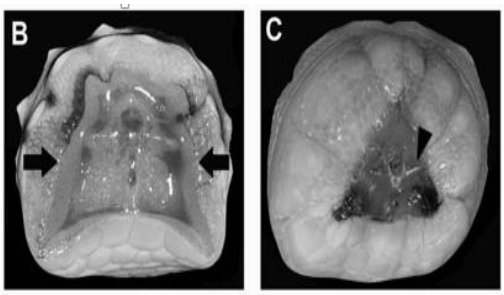

Figure 2. Phases of Wound Healing
(Gilbert, E., Payne, S., dan Vickaryous, M,

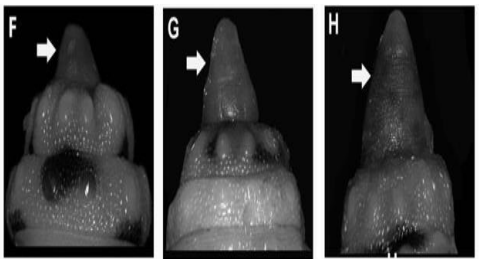

Figure 3. Cell Diferentiation Phase
(Gilbert, E., Payne, S., dan Vickaryous, M, 2013)

The Figure shown about The regeneration process according to 3 phase in Reptiles Ordo Squamata. Based on Figure 1, 2 and 3, the information can be taken to see how the process is like during the regeneration of the tail of a lizard that is undergoing autotomy. This process can be seen by taking samples from the reptile Ordo Squamata, a type of lizard with the scientific names *Mabouya multifasiata* and *Hemidactylus platyurus*. According to (Novianti et al., 2019) flat-tailed gecko have high regeneration power in severed tails (autotomy) and are able to form new tails. Autotomy is a flat-tailed gecko's behavior in its ability to sever its tail to trick predators. The flat-tailed gecko will press its muscles at the autotomy level so that the tail is cut off.

METHOD

This research is a type of research that is Quantitative Descriptive. With a research focus on the characteristics of animals observed using the morphometric method of measuring body length and body width during each autotomy process in *Mambouya multifasiata* and *Hemidactylus platyurus*. The method used in this research is the Histological Morphometric method, this morphomeric histological method aims to measure variations and changes in tail shape and body size of an organism (Harjana, 2011). Morphometry includes length and width measurements, field and outline analysis. The application of morphometry is usually carried out in measuring living things and geographical measurements (Fauzi, 2017). According to Rakhmiyati, et al. (2016) said that histological observations are carried out using a light microscope on histological slides stained with Hematoxylin-Eosin (HE) to determine the shape and condition of the cells on days 1, 3, 5, 8, 10, 13, 17, 21, 25, and 30 (Novianti et al., 2020). Sixteen male of the *Hemidactylus Platyurus* and *Mabouya multifasiata* variety obtained from Central Java, sample bottles, gloves, alcohol, chloroform, formalin and Hematoxylin Eosin (HE).

The first method of work used in this research is the sample acclimation process which aims for the samples to adapt to the new environment by being given food and drink according to their natural habitat. To determine the characteristics of the morfometrics, samples were measured using a caliper on all 4 parts of the body starting from the Head (HeadL), Body, Tail and Total Length (SvL) (Novianti et al., 2020). Meanwhile, micro-observations on the autotomy tail are carried out using the Paraffin Method with the help of Hematoxylin Eosin (HE) staining. On each Longitudinal Slice Preparation and Transverse Slice Preparation, the slices will be examined directly at the Palembang Dyanitalis Laboratory. which was stained with Hematoxylin-Eosin (HE) to determine the shape and condition of the cells on days 1, 3, 5, 8, 10, 13, 17, 21, 25, and 30. All histology data were analyzed descriptively (Novianti et al., 2020).

RESULT AND DISCUSSION

Characteristics of Morphometrix *Mabouya multifasiata*

These characteristics were measured and observed using a caliper and measuring mat, on a morphometrix in the sections, Head (HeadL), Body (Body), Tail (Taile) and Total length (SvL).

Table 1. Morphometric Characteristics of Lizards (*Mabouya multifasiata*)

Sample	Morfometrics *Mabouya multifasiata*				
	HeadL	AGL	TaiL	*SVL*	Average
S1	2,6 cm	7,6 cm	10 cm	21 cm	41,2
S2	2,5 cm	6,5 cm	9 cm	17, 2 cm	35,2
S3	3,5 cm	8,5 cm	12,4 cm	24,3 cm	48,7
S4	2,5 cm	8 cm	9,5 cm	20 cm	40
S5	3 cm	8 cm	10,5 cm	22 cm	43,5
S6	2,5 cm	7,5 cm	11,5 cm	21,5cm	43
S7	2,6 cm	7,8 cm	11,2 cm	21,6 cm	43,2

- SVL : Length from mouth to tail
- HeadL : Head to neck length
- AGL : Body length to cloaca
- TaiL : Length of pankal tail to tip

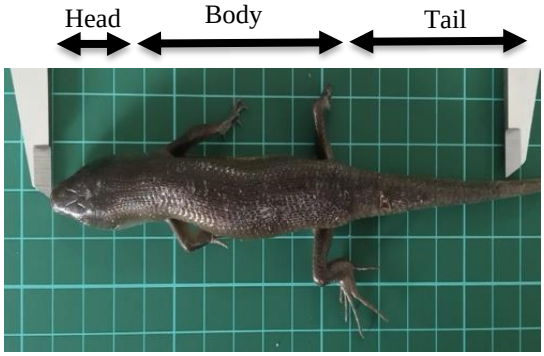

Figure 4. *Mabouya multifasiata* Morphology

2. Histology of *Mabouya multifasiata* Tail Cells

Based on the results of research that has been carried out, autotomy samples were cut from the tail of the *Mabouya multifasiata* which had been taken and divided into three tail samples consisting of the initial tail sample before autotomy, the sample on the 15th day and the last sample after autotomy, namely the 1st day. 30. Then from these three tail pieces, slice preparations were made using the Hematoxylin Eosin (HE) staining method. The slices of this preparation are divided into two, namely longitudinal slices (longitudinal) and transverse slices (transversal), then continued with a histological examination under a microscope which will be carried out at the Palembang Dyatnitalis Special Anatomical Pathology Laboratory. The Results showed if positive (+) there is the cell that part of regeneration, negative(-) if there is no cell or part of tissue. Part of cell and tissue the growth showed significant in the mass of cell from histology parameters there are 3 abbreviation : many, medium and few tissues.

Table 2. Sections (Longitudinal) of Lizard cells (*Mabouya multifasiata*)

No.	No Sampel	Longtudinal				
		Muscles	Perivertebral Fat Tissue	Cartilaginous Tube	Menix	Epindymal Cell
1	Sample 1	(+)	(+)	(+)	(-)	(+)
2	Sample 2	(+)	(-)	(++)	(+)	(+)
3	Sample 3	(+)	(-)	(+)	(+)	(-)

Table 3. Cross section of lizard cells (*Mabouya multifasiata*)

No.	No Sample	Transversal				
		Muscles	Periver tebral Fat Tissue	Cartila ginous Tube	Menix	Epindymal Cell
1	Sample 1	(+)	(+)	(++)	(+)	(++)
2	Sample 2	(+)	(+) Many	(++)	(-)	(+)
3	Sample 3	(+)	(-)	(+)	(+)	(+) Little

The Table 2 and 3 above shows the development of cells in the tail of the *Mabouya multifasiata* in the initial or original tail, the 15th day and the 30th day tail with 3 samples each with transverse (Longitudinal) and longitudinal (Transversal) incisions.

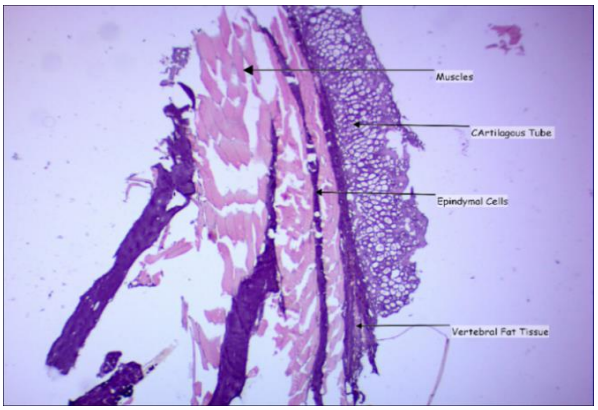

Figure 5. Longitudinal cross section of a real lizard's tail, Hematoxylin eosin (HE), magnification (4x10). (a) Vertebal tissue, (b) Ependymal cells, (c) Cartilage, and (d) Muscle.

Based on the histology image of lizard cells (*Mabouya multifasiata*) in the original or original tail, Figure 5 and Figure 7. Longitudinal incision, cells were found in muscle tissue which is shown by red fibers, cartilaginous tubes, epindymal cells and perivertebral fat tissue. Meanwhile In Figure 6. transverse section, several cells were found whose arrangement and location were very clear, namely, muscle tissue, vertebral tissue, menix tissue, ependymal cell tissue and cartilage tissue.

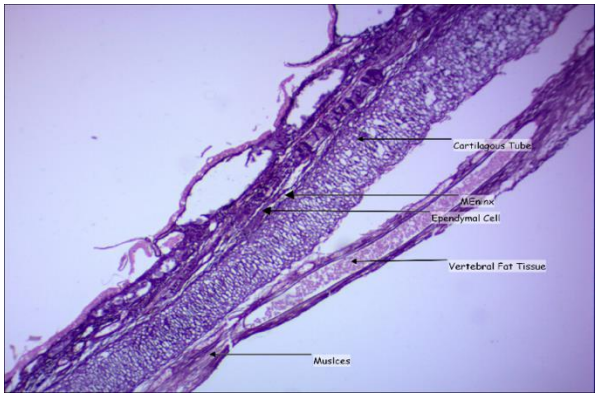

Figure 6. Longitudinal cross section of the original tail, Hematoxylin eosin (HE), magnification (4x10). (a) Ependymal cells, (b) Cartilage Tube, (c) Menix (d) vertebral tissue and (e) muscle

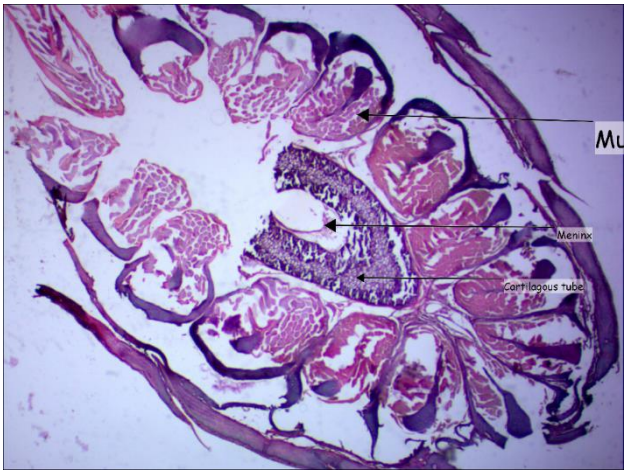

Figure 7. Transverse (transverse) section of the original tail, Hematoxylin eosin (HE), magnification (4x10). (a) Ependymal cells, (b) Cartilage Tube, (c) Menix (d) vertebral tissue and (e) muscle.

Histological images of Mabouya multifasiata cells in the 15th tail growth can be found in the image above in cross section and longitudinal section. In Figure 6 (cross section), after observing under a microscope to see the shape and size of the cells, several tissues were found that were visible, although not very perfect, namely bone tissue, muscle tissue, and vertebral tissue. Meanwhile, the second cut is a transverse incision (Fig. 9). It is clear that several tissues were stained with Hematoxylin-Eosin (HE) to determine the shape and condition of the cells on days 1, 3, 5, 8, 10, 13, 17, 21, 25, and 30 (Novianti et al. , 2020).

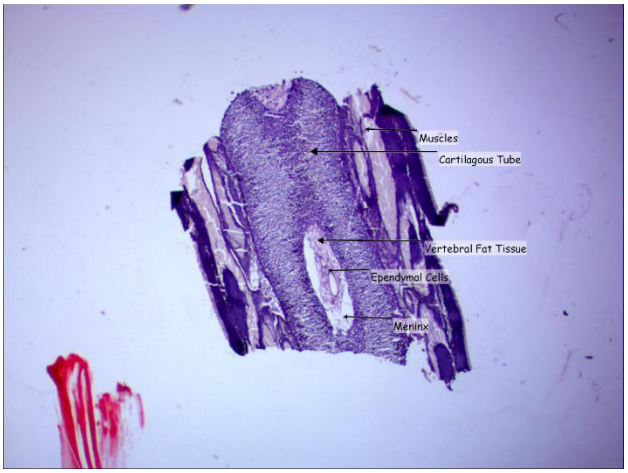

Figure 8. Longitudinal cross section of the original tail, Hematoxylin eosin (HE), magnification (4x10). (a) Ependymal cells, (b) Cartilage Tube, (c) Menix (d) vertebral tissue and (e) muscle.

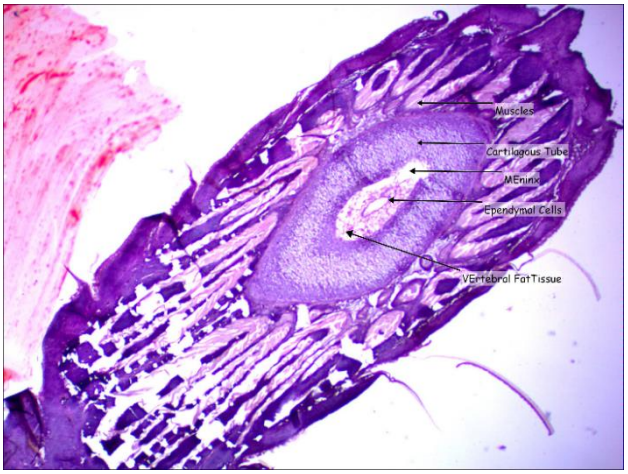

Figure 9. Transverse (transverse) section of the original tail, Hematoxylin eosin (HE), magnification (4x10). (a) Ependymal cells, (b) Cartilage Tube, (c) Menix (d) vertebral tissue and (e) muscle.

Morphometry of *Hemidactylus Platyurus*

From the results of the morphometric research carried out, the results obtained were that during the identification process, there were 7 individual house lizards (Hemidactylus platyurus) that were male. In the observations made on morphometry, namely; head, body, tail which have different sizes for each individual.

Table 4.1. Morphometric Characters of *H.platyurus*

Sample	Hemidactylus Platyurus Characteristic				Size		
	HeadL	AGL	Tail	SVL	Mean	Maks	Min
S1	1,8 cm	3 cm	5,5 cm	5,9 cm	4,25	6,00	2,00
S2	2,3 cm	4,10 cm	8 cm	6 cm	5,25	8,00	2,00

Sample		Hemidactylus Platyurus Characteristic				Size	
S3	1,9 cm	4 cm	3,9 cm	5, 4 cm	3,75	5,00	2,00
S4	1,9 cm	3,5 cm	5,2 cm	5,4 cm	4	5,00	2,00
S5	1,7 cm	3,4 cm	4,8 cm	5,3 cm	3,75	5,00	2,00
S6	1,8 cm	3,4 cm	4,2 cm	5, 6 cm	3,75	6,00	2,00
S7	1,7 cm	1,9 cm	4 cm	5,7 cm	3,5	6,00	2,00

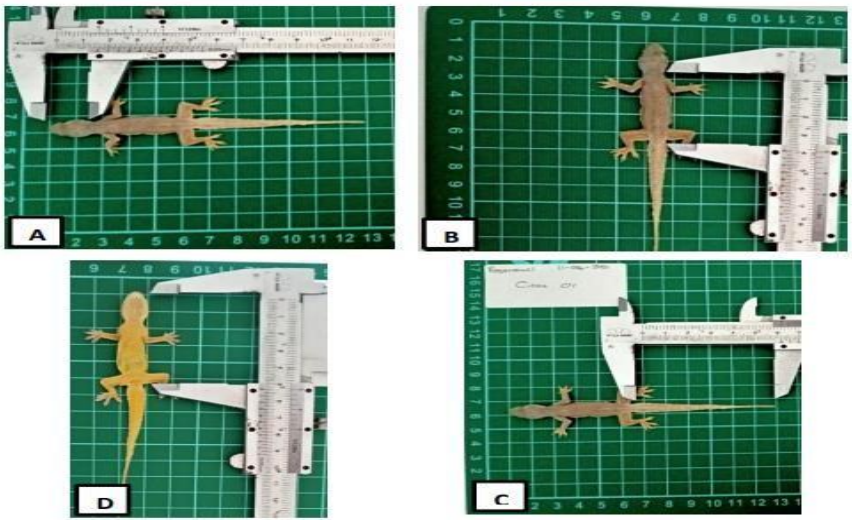

Figure 10. (a). Head, (b) Body, (c). Tail, (d).Mouth and Cloaca

Measurement of morphometry in lizards (Cosymbotus platyurus) based on head, body and tail. Based on research results, seven *H. platyurus* have different head, body and tail lengths.

Growth of the tail of *H. platyurus*.

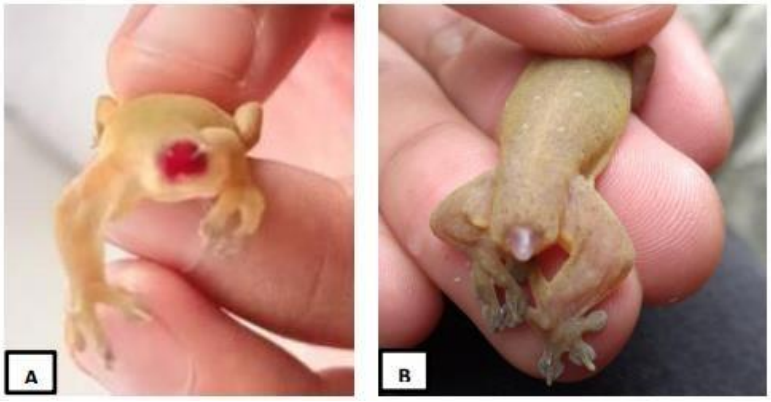

Figure 11. Cosymbotus platyurus 15th tail

From the results of the tail growth of the Cosymbotus playurus lizard during the 15th and 30th days. The picture above shows the condition of the 15th tail and the bleeding condition of the tail of the lizard (Cosymbotus platyurus) when it undergoes autotomy : a. Experiencing bleeding in the tail, b. 15th day of tail growth.

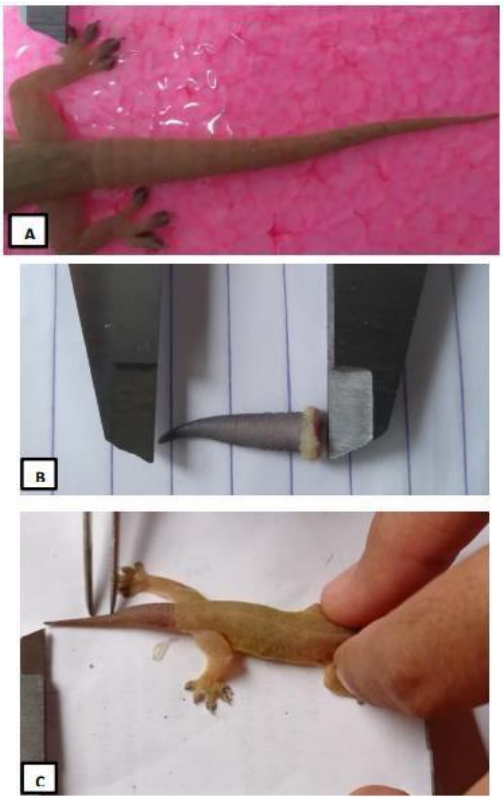

Figure 12. Growth of the lizard's tail

The image above shows the growth of a lizard's tail (Hemidactylus platyurus) and differences in tail color on the first, 15th and 30th days.

Histology of *H. platyurus* cells

Table 2. Regeneration (longitudinal) of *H. platyurus* cells.

No	Sample					
		Muscles	Perivertebral Fat Tissue	Cartilaginous Tube	Menix	Epindymal cells
1.	S1	(+)	(++) Many	(+) Few	(-)	(+) Few
2.	S2	(+)	(++) Many	(+) Few	(-)	(+) Few
3.	S3	(+)	(++) Many	(++) Many	(++) Many	(++) Few

The results of the histology examination of the tail cells of the lizard (Cosymbtus platyurus) with Hematoxylin-Eosin (HE) staining.

Table 3. Regeneration (transverse) of *H. platyurus* cells

No	Sample					
		Muscles	Perivertebral Fat Tissue	Cartilaginous Tube	Menix	Epindymal cells
1.	S1	(+)	(++) Many	(+) Little	(-)	(+) Little
2.	S2	(+)	(++) Many	(+) Ring shape	(-)	(+) Little
3.	S3	(+) many	(++) Many	(++) Many	(-)	(++) Many

The table above shows the development of cells in the tail of the Cosymbotus platyurus lizard in the initial tail, 15th and 30th tail with 3 samples each in longitudinal and transverse sections.

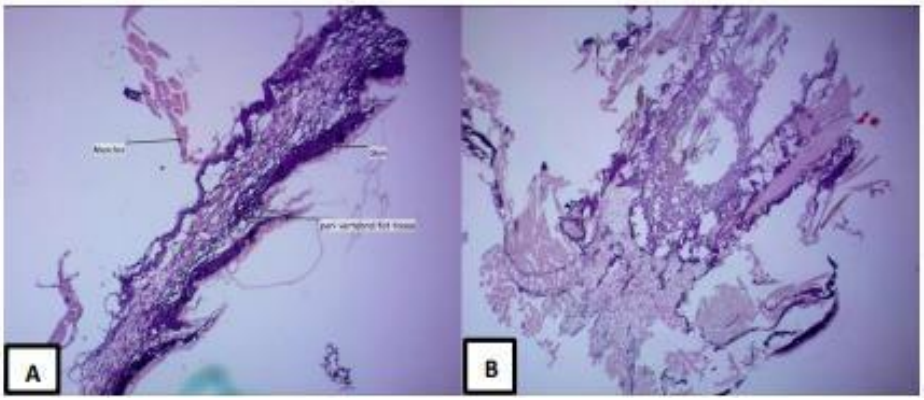

Figure 13. Hematoxylin eosin (HE), magnification (4x10). Initial tail cells (A) elongated, (B) transverse

In the histology image, the initial tail was damaged due to delays in the preservation process (formalin. See Figure 4. In the longitudinal section, perivertebral fat tissue, skin and muscle cells were found. Meanwhile, in the transverse section, no cell parts could be found due to cell damage.

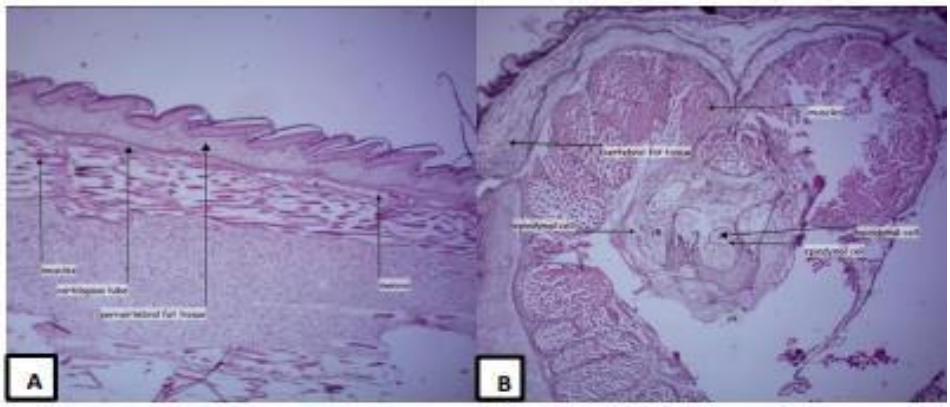

Figure 14. Hematoxylin eosin (HE), magnification (4x10) (A). Longitudinal, (B) transverse

On the 15th day, tail growth was found in longitudinal cuts the presence of muscle cells, cartilaginous tubes, perivertebral fat tissue and menix. Meanwhile, in cross section there are muscle cells, vertebral fat tissue and epindymal cells.

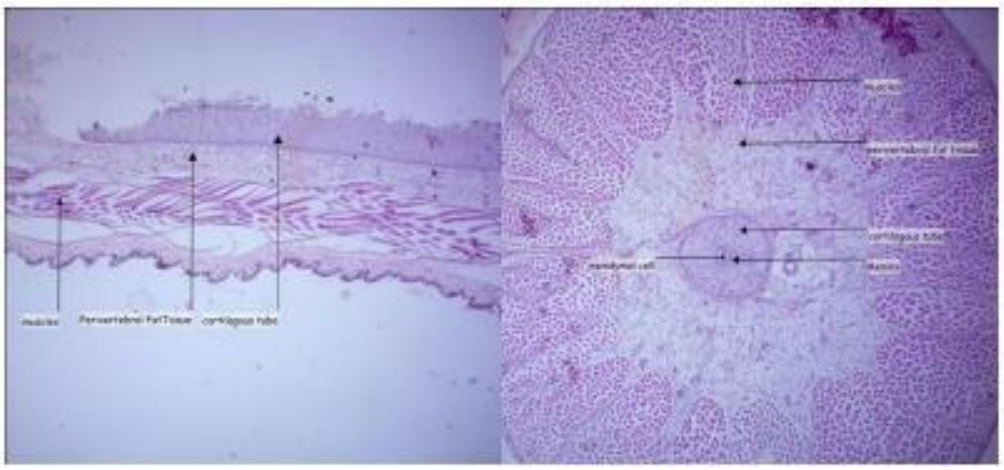

Figure 15. Hematoxylin eosin (HE), magnification (4x10)30th tail, (a).Longitudinal, (b). Tranverse

In the 30th day tail histology results there was significant tail growth perfect and returns to its original tail. A longitudinal section shows the presence of muscle cells, perivertebral fat tissue and catillary tube. Meanwhile, in the transverse section, muscle cells, perivertebral fat tissue, epidermal cells, cartilage and menix were found.

CONCLUSION

Morphometric measurements obtained the *M. multifasiata* morphological shape of the head (HeadL) with a head length of 2-3 cm, the body (AGL) is generally blackish brown with a yellowish green pattern with a length of 8 cm, and the tail (TaiL) is black from the base cloaca to the tip of the tail with a length of 10 -12 cm with a skin surface covered in fine scales. *H. platyurus*, the morphology is head shape (HeadL), the snout is longer than the distance from the eyes to the ears. The shape of the ear canal is oval. Morphometric body parts of H. platyurus (AGL), in general the body (AGL) is gray with varying light and dark patterns on the dorsal side, average 41,2- 43,2 cm.

Histological observation showed after autotomy regeneration in *M. multifasiata* bone tissue, muscle tissue, vertebral tissue on the 15th day. On day 15 as shown, Muscle cells are found in the tail autotomy plate of the *H.platyurus* which is located lengthwise on the caudalis vertebra which is covered by the central nervous system (meninx). Cartilage (Cartilagous tube). The 30th day shows that the cells have formed normally. Cells and tissues are completely visible compared to the tail of the *M. multifasiata* and *H. platyurus* on day 15.

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