Morphology Variations of *Hemidactylus frenatus* in Java

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**ABSTRACT**

*Hemidactylus frenatus* is one of the most widely distributed reptile species, almost all over Indonesia, including Java. The distribution is due to the process of merging and separation on some islands, which can lead to new morphological, anatomical and molecular characters that are very different as a result of adaptation. The aim of this study is to determine the distribution or philobiogeography of *Hemidactylus frenatus* by using morphometric analysis especially in the Java region, the data obtained is entered into Principal Component Analysis (PCA) and followed by cluster analysis. The result of this study was it found *Hemidactylus frenatus* samples from several regions which are divided into three provinces, namely East Java, Central Java, and West Java. The morphometric analysis of *H. frenatus* from the three provinces shows the grouping of *H. frenatus* according to the province of each individual, and can be seen also the similarity and difference character based on the biplot line. In the cluster analysis, there are two large *H. frenatus* groupings showed the degree of morphometric proximity among *H. frenatus* individuals from several regions of Java.

**Keywords:** Actinomycetes, antimicrobial, *Nasutitermes* sp., *Streptomyces prasinopilosus*, termite nest

**Introduction**

Indonesia’s biodiversity is one of the largest in the world. The richness of flora and fauna and microbes contained in the soil and water, land sea are distributed in some parts of Indonesia. The diversity is influenced by the climatic conditions and geographical position of Indonesia which is very appropriate with the physiological conditions required by flora and fauna to grow and develop well [1,2]. Indonesia was located in an area bounded by the Wallace line which is the divided into Sundaland and Lesser Sunda. This condition causes some species to be separated or dispersed, so the species must adapt to new environmental, temperature and habitat conditions [3,4].

As a result of these events, it can appear to new morphological, anatomical and molecular characters that are very different from the previous species in one population. This event is caused by the existence of isolation mechanisms that occur between the population and the interaction between the population and its environment as a form of adaptation to be able to maintain the life cycle and its offspring. On the other hand, the event also raises the character differentiation of a species as a form of interaction between the two factors [5].

One type of animal that experienced the impact of sea water fluctuation events and has distributed evenly in almost all areas of Indonesia is a group of reptiles, including *Hemidactylus frenatus* which has a very wide dispersal area, almost all over Indonesia including Java, Sumatra and Borneo. In addition, the distribution of *Hemidactylus frenatus* also covers most of Southeast Asia, including Myanmar, Thailand, Vietnam and Laos and Australia [6]. This research is important to know the distribution or philobiogeography *Hemidactylus frenatus* by using morphometry analysis especially in Java region.
Material and Methods

Sampling Data

Morphological data were recorded from available material of *Hemidactylus frenatus* from Java that of various was distributed from three provinces: East Java, Central Java and West Java.

Morphometric Examination
The following morphometric measurements were taken with a caliper to the nearest 0.1 mm: Snout-Vent Length (SVL), Total Length (TL), Aksial-Length (AGL), Head Length (HeadL), Head Width (HeadW), Eye Diameter (EyeD), Snout Eye (SnEye), Nares Eye (NArEye), and Internasal (IN). Morphometric data were included in principal component analysis (PCA). The PCA was used to determine when populations were morphologically Clustered (Sneath and Sokal, 1973). A hierarchical cluster analysis was performed with PAST v. 2.17c to determine which individuals were the most morphologically similar based on the the Unweighted Pair Group Method with Arithmetic Mean (UPGMA).

Results and Discussion
Based on the analysis of morphometry, the result obtained there are 20 *Hemidactylus frenatus* (Hf) females from Java. This species originated from various areas was distributed over three provinces, including East Java, Central Java, and West Java. The morphological characteristics of *H. frenatus* from each province in Java based on morphometric analysis are presented in below (Table 1).

| Species | SVL (mm) | TL (mm) | AGL (mm) | HeadL (mm) | HeadW (mm) | EyeD (mm) | SnEye (mm) | NArEye (mm) | IN (mm) |
|---------|----------|---------|----------|------------|------------|-----------|------------|-------------|--------|
| *Hf* Malang | 52.30 | 79.21 | 21.00 | 12.50 | 11.59 | 3.52 | 4.48 | 3.28 | 4.09 |
| *Hf* Malang | 54.20 | 88.51 | 23.09 | 11.70 | 10.32 | 3.76 | 3.14 | 2.52 | 4.79 |
| *Hf* Bondowoso | 44.77 | 80.60 | 22.37 | 13.84 | 9.34 | 2.79 | 5.31 | 4.19 | 3.82 |
| *Hf* Bondowoso | 49.03 | 95.58 | 20.27 | 14.55 | 10.56 | 2.78 | 5.78 | 4.30 | 3.15 |
| *Hf* Bondowoso | 47.80 | 66.80 | 23.40 | 13.80 | 9.00 | 3.00 | 6.00 | 4.60 | 2.30 |
| *Hf* Mojokerto | 48.48 | 88.53 | 18.77 | 13.87 | 10.39 | 1.98 | 5.70 | 4.20 | 3.99 |
| *Hf* Mojokerto | 54.90 | 121.20 | 24.70 | 21.20 | 10.80 | 3.10 | 6.80 | 5.10 | 2.50 |
| *Hf* Banyuwangi | 45.40 | 92.60 | 18.40 | 14.80 | 8.60 | 2.70 | 5.50 | 4.00 | 1.90 |
| *Hf* Banyuwangi | 47.50 | 88.90 | 16.90 | 12.10 | 10.00 | 2.70 | 5.70 | 4.60 | 2.90 |
| *Hf* Banyuwangi | 55.75 | 93.50 | 25.85 | 16.80 | 11.20 | 2.90 | 7.00 | 5.30 | 2.00 |
| *Hf* Madura | 45.20 | 94.20 | 19.10 | 15.30 | 9.00 | 2.60 | 6.10 | 4.60 | 2.80 |
| *Hf* Madura | 50.50 | 105.50 | 20.90 | 16.20 | 10.20 | 3.10 | 5.90 | 4.90 | 1.70 |

Continue.....
**Central Java**

| Sample Region | Hf Ns. Kambangan | Hf Cilacap | Hf Karimun Jawa | Hf Karimun Jawa | Hf Karimun Jawa | Hf Karimun Jawa |
|---------------|------------------|------------|----------------|----------------|----------------|----------------|
|               | 31.46            | 60.31      | 13.30          | 10.53          | 7.09           | 1.91           |
|               | 44.69            | 102.11     | 17.08          | 12.76          | 9.15           | 1.70           |
|               | 53.50            | 116.00     | 29.70          | 15.70          | 9.80           | 3.40           |
|               | 43.20            | 45.30      | 18.60          | 13.40          | 7.80           | 2.50           |
|               | 52.00            | 108.80     | 19.40          | 16.70          | 9.90           | 2.30           |

**West Java**

| Sample Region | Hf Sukabumi | Hf Bogor | Hf Cibinong |
|---------------|-------------|----------|-------------|
|               | 54.30       | 85.30    | 53.40       |
|               | 49.10       | 106.90   | 112.40      |

*Hemidactylus frenatus* sample from each region in Java includes east Java which is represented by Malang, Bondowoso, Mojokerto, Banyuwangi, and Madura. Then the area of Central Java is Nusa Kambangan, Cilacap, and Karimun Java, while for the West Java region includes Sukabumi, Bogor, and Cibinong can be seen in the picture 1 below.

![Figure 1. Study of Location in Hemidactylus frenatus research](image_url)
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Figure 2. Results of PCA Hemidactylus frenatus (Hf) East Java, Central Java, and West Java. Description: HFMAL= Hf Malang, HFBON= Hf Bondowoso, HMO= Hf Mojokerto, HFMA= Hf Madura, HF MAD= Hf Madiun, HFBAN= Hf Banyuwangi, HFNK= Hf Nusa Kambangan, HFCIL= Hf Cilacap, HFKJ= Hf Karimun Jawa; HFS= Hf Sukabumi, HFB= Hf Bogor, HFCIB= Hf Cibinong.

Based on the results of PCA of H. frenatus sample in Figure 1., it can be seen that the navy blue line shows the collected samples from East Java, which amounted to 15 individuals; the dark blue line shows the collected samples from Central Java, with a total of 5 individuals; While the red line shows the collected samples from West Java which amounted to 3 individuals.

These results indicate that the samples from East Java, Central Java, and West Java have similarities to each other, this can be known from blocked lines that are interconnected. In addition to the interconnected lines, the similarity of H. frenatus characters from the three provinces can be known from the samples of HFBON (East Java) and HFKJ (Central Java) which has the same character ie the distance from the nose to the eye (NarEye) and the Aksial-Length (AGL). While HFMAL has distinguishing character with Hf from other area that is based on the highest of headw character. Then, a sample of HFBAN (East Java) and HFCIB (West Java) which have the same character in terms of head length and total length (TL). HFMAL and HFCIL samples appear to have significant differences based on the result of biplot line, thus indicating a negative correlation relationship. Where as the eye diameter (EyeD) is higher, the Snout Eye (SnEye) is lower. Morphometric characters are then used for cluster analysis as shown in Fig. 3.
In this study, morphometric analysis was performed on *Hemidactylus frenatus* from Java. The morphometric results of *H. frenatus* obtained before being analyzed using PCA, were first incorporated into the following formula:

\[
(\text{standardization of characters}) = \frac{\text{character}}{\text{SVL}}
\]

From the formula, it can be seen that the standarization of characters uses SVL as a divisor of each character being measured. This is based on Kurniawan *et al.* [7]. SVL is a common character and is representative of herpetofauna character, as not all herpetofauna groups have complete character for analysis, such as the tail.

Based on PCA results, it can be seen that there are three stations or areas that each show the distribution of *Hemidactylus frenatus* located on the island of Java, with each character produced. The three stations are indicated by the differences in color that have been analyzed, where the blue color tends to be a sample from East Java, dark blue tends to describe samples from Central Java, while red color shows the samples from West Java. According to Carranza *et al.* [2], PCA data provides an explanation of the morphometric differences of each species or individual being analyzed.

PCA analysis results also show that in each region, *Hemidactylus frenatus* has a different character. The different characters can be seen from the direction of the resulting biplot line. The larger direction formed shows the greater difference, so it can be negatively correlated. For example, samples from West Java have high TL and HeadL characters compared to HFBON samples from East Java and HFNK from central Java. Then the most striking different was seen in the HFKJ (Central Java) sample with HFMAL (East Java) seen from HeadL and HeadW characters. If the HeadW character is high, then the HeadL character is low. The HFCIB (West Java) sample has characteristics closer to the HFMAD (East Java) sample with the same TL characteristics.

Figure 3. Dendrogram that describes the level of morphological similarity of *H. frenatus* from East Java, Central Java and West Java
Other characteristic equations like SnEye are seen in HFCIL and HFMO samples.

The existence of difference and similarity character in *Hemidactylus frenatus* sample above is influenced by the existence of external factor in the form of environment which cause the species must adaptation with new environment and internal factor that is event of polymorphism, gene expression, and epigenetic. According to Allis et al. [1], epigenetic events are one of the processes of phenotypic changes or gene expression caused by the mechanisms other than changes in DNA sequences, which are highly influential to the morphological form and structure of an organism, but genetically do not have the difference is very significant or can be said still in one group of organisms or the same species.

The obtained morphometric character was then used for cluster analysis based on UPGMA. The cluster results show that there are two major clade groups, namely clade a and clade b, with 73% for clade a, and 87% for clade b. Clade a consists of samples from East Java (except HFMAL), Central Java, and West Java. Clade a has two subclasses, namely subclade a1 and subclade a2. Subclade a1 consists of samples of HFBON, HFBAN, HFMO, HFMA, HFMA (East Java); HFNK, HFCIL, HFKJ (Central Java); and HFCIB, HFS and HFB (West Java), whereas subclade a2 consists of HFKJ (Central Java) and HFBO (East Java) samples. The level of character similarity between subclade a1 and subclade a2 is 24% and 72% respectively, so there is still a very high difference between each sample and the potential to form a new sub species. As for Clade b, consisting only of HFMD samples, this indicates that the samples from Malang have different characteristics from samples originating from other regions. Bootstrap values on branches of phylogenetic trees of> 70% are considered significant, whereas between 50-70% is considered a tendency [8, 9, 10].

**Conclusion**

The results of morphometric analysis on *Hemidactylus frenatus* found in various regions in Java showed the difference and similarity character in each sample. It is based on PCA results, where interconnected blocked lines indicate the same character, while the direction on the biplot line indicates a character difference. The cluster analysis results form two clade groups (clade a and clade b), with 73% for clade a, and 87% for clade b.

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