

Morphological characteristic of *Nepenthes gymnamphora*'s pitcher in Pasir Cadas Panjang Mountain Ciwidey West Java

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Abstract. Limited information exists on the variation and function of pitcher morphological diversity in *Nepenthes* spp., necessitating further research. Based on a previous study in Mount Pasir Cadas Panjang Ciwidey that identified *N. gymnamphora*, this research aims to characterize pitcher morphology in Mount Pasir Cadas Panjang Ciwidey. The method used purposive sampling from six locations of *N. gymnamphora*, pitchers were categorized into upper and lower types based on size. The study revealed distinct differences in shape between upper and lower pitchers. The upper type exhibited a jug-like enlargement at the bottom, narrowing towards the top and enlarging again at the peristome. In contrast, the lower type featured a uniformly enlarged diameter from the bottom to the peristome. Both types shared similar components, differing mainly in wing characteristics the upper type having unhaired and unembossed wings, while the lower type had embossed and haired wings. Pitcher colors included green, patterned, and reddish, with higher elevations correlating with a greater likelihood of reddish or patterned pitchers. This research provides comprehensive data on the morphological and anatomical variations of *N. gymnamphora*, offering valuable insights for future studies.

1 Introduction

Nepenthaceae is a family of floral species that grows in Indonesia and has only one genus (monogeneric), *Nepenthes*, commonly known as "Kantong Semar." *Nepenthes* is a unique plant that has the ability to prey on insects, often referred to as an "insectivorous plant" or "pitcher plant," so it is classified as a carnivorous plant with a pocket that is a modification of the leaf tip [1]. This plant is generally found on nutrient-poor soils and likes to grow in the open air and moist conditions. *Nepenthes* roots have the same ability to fix nutrients like nitrogen and phosphorus as other plants, but growing in nutrient-poor soils, the roots cannot absorb nutrients optimally. Recognizing this problem, *Nepenthes* has modified the tips of its

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leaves into pitchers or pitcher that act as insect traps, where the trapped insects absorb phosphorus and nitrogen. Another uniqueness of this plant is found in the pitcher of *Nepenthes*, which have different sizes, colors, patterns, and shapes and become morphological characteristics that differentiate the species [2].

The features and variety of the *Nepenthes* pitchers cannot be detached from their existence in a habitat suited for their growth. West Java, an Indonesian province on the island of Java, possesses abundant natural resources, including humid tropical forests and favorable climatic conditions, making it an ideal location for the *Nepenthes* habitat. On Java Island, three *Nepenthes* species exist: *N. mirabilis*, *N. adrianae*, and *N. gymnamphora* [3]. *Nepenthes* that grow in West Java province only *N. gymnamphora* and *Nepenthes mirabilis* [4].

Based on previous surveys conducted at Mount Pasir Cadas Panjang Ciwidey, West Java (2050 masl), a natural habitat of *Nepenthes*, *N. gymnamphora* species was found. *N. gymnamphora* is an endemic species to Java [3-4]. It is closely related to *N. pectinata*, a species found in Sumatra, as their morphological characteristics are almost identical. However, there is a distinction between the two species, as *N. gymnamphora* has a simpler food network. The term *gymnamphora* derives from Greek, specifically, "gymnos," meaning naked or exposed, and "amphorous," meaning flask shaped. *N. gymnamphora* is a protected flora species listed in CITES (Convention on International Trade in Endangered Species) Appendix II due to Least Concern (LC) status on the IUCN Red List.

N. gymnamphora has been found to have differences in pitcher morphological characteristics between individuals. various researchers have examined variations in *Nepenthes* pitcher morphology, study of pitcher morphological characteristics and color variations in pitcher parts [5]. Objective evaluations were used throughout, and technical term abbreviations were explained upon their first use. This investigation analyzed tendril, body, wing, lip, and lid variation in the *N. gracilis* Korth species. The pitchers possessed by various *Nepenthes* types differ. Each pitcher has the capacity and inclination to catch different types of prey [6]. The distinct shape of the pitcher between *Nepenthes* species is a type of adaptation to its surroundings [7]. The alteration in the shape of the pitcher demonstrates the pattern of *Nepenthes*' morphological evolution to survive.

Based on the issues mentioned above, various aspects of *N. gymnamphora* require further observation, primarily concerning the varying morphological characteristics of the pitcher among individuals. Additional data regarding the morphological traits of *N. gymnamphora* in its natural habitat is notably deficient, particularly within Mount Pasir Cadas Panjang Ciwidey, West Java. Based on this, a study will conducted to characterize morphological differences of pitchers in *N. gymnamphora* species at Mount Pasir Cadas Panjang Ciwidey, West Java.

2 Materials and methods

2.1 Methods

This study employed a survey methodology with purposive sampling. The method involved systematically traversing a specified observation area and directly recording plant observations as they were encountered. In this research, the focus was on *N. gymnamphora*. *N. gymnamphora* found was then marked by GPS (Global Positioning System) on the device used to determine the observation point. The observation area was divided into six *Nepenthes* distribution areas. Ten pitchers were randomly selected for observation in each area, and then 60 samples were taken for this study. Each *Nepenthes* specimen was observed, recorded, and photographed before being categorized by color variation and pitcher type. The dimensions of the pitcher parts were then measured and described to determine the largest and smallest

pitchers' sizes for each color variation and pitcher type. Habitat observations were conducted to gather data on biotic and abiotic factors. Biotic factors studied included the vegetation surrounding *Nepenthes*. The abiotic factors assessed are related to climate and soil [4-8].

2.2 Times and location

The study was conducted over one month, in October 2023, at Mount Pasir Cadas Panjang Ciwidey. This site was chosen due to the presence of a *Nepenthes* community and its altitude of 2050 meters above sea level and managed by PT. Perhutani West Java, the project is located in Alam Endah village, Bandung Regency, West Java [9]. The study area covers a route beginning at the base of the mountain or low-lying region and ending at the highest area - the summit. The research site is situated at an altitude of 1.700-2.050 meters above sea level. The study area covers a route beginning at the base of the mountain or low-lying region and ending at the highest area, the summit. Data collection took place between 9 am and 3 pm (WIB).

The survey method was to explore the study site following the path from the low area to the peak area at Cadas Panjang Sand Mountain. Observations were made on three different paths (Figure 1) to reach the entire area of *Nepenthes* presence. These three paths ended at the same peak [4,5].

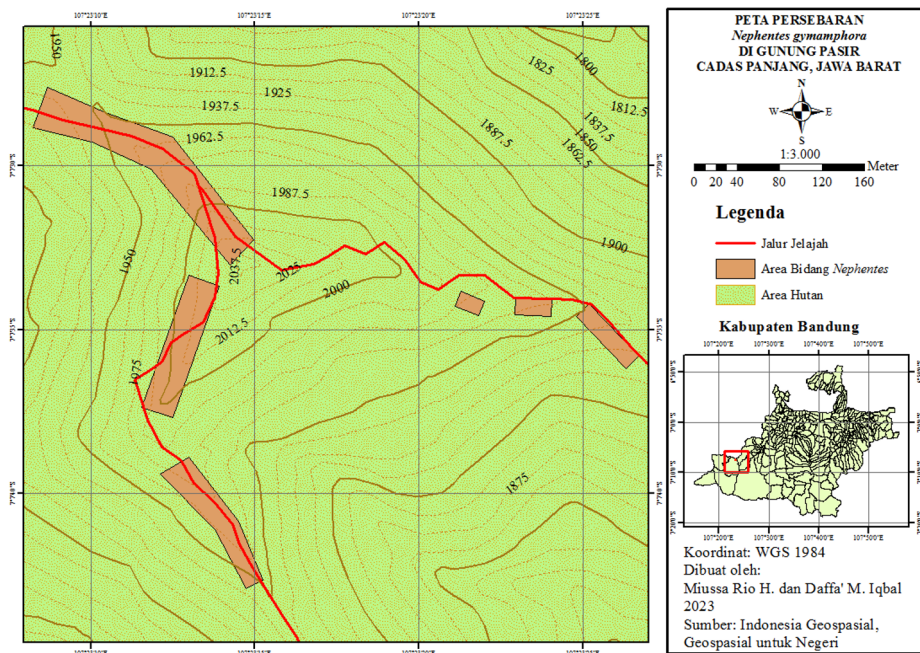


Fig. 1. Map of *N. gymnamphora* trails at Cadas Panjang, Ciwidey, West Java.

2.3 Morphological characterization of pitchers

Pitcher samples were observed for morphology either visually or using a camera. Measurements of the pitchers and their components were taken with a ruler or tape measure. The pitcher samples were selected in the field, where the largest and smallest pitchers of each type and color were selected [5].

Pitcher morphology was observed by taking measurements of various parts of the pitcher, including its length, upper and lower circumference, mouth length and width, and pitcher lid

length and width. Vine length was also measured from the end of the lamina to the lowest point of the pitcher body. The pitcher height was measured from the lowest point of the pitcher body (at the vine attachment point) to the end of the peristome [10]. The largest part of the upper pitcher body was used to measure the upper circumference of the pitcher, while the lower circumference was measured at the largest part of the lower pitcher body. The longest part of the mouth was used to measure mouth length, while mouth width was measured at the widest part of the mouth. The longest and widest parts of the flap were used to measure flap length and width, respectively [11-15].

The visual of pitcher color is conducted through observation. The process is initiated by the primary color and followed by the secondary color. This study defined the primary color as the predominant color encompassing a larger portion of the pitcher. In contrast, the secondary color refers to the color that covers a smaller section of the pitcher. If a part of the pitcher is covered by only one dominant color; the color of that part of the pitcher is named with a single color, such as green or dark red. If there is more than one color, the pocket section is named by the combination of colors present, such as purplish red [5].

2.4 Habitats

Habitat conditions were assessed based on the environmental conditions of the study site. The study evaluated both abiotic and biotic factors [16]. Climatic abiotic factors such as temperature, humidity, and light intensity were measured, as well as soil-related abiotic factors, including soil organic matter or Materi Organik Tanah (MOT) testing, temperature, and soil moisture. Climatic abiotic factors such as temperature, humidity, and light intensity were measured, as well as soil-related abiotic factors, including MOT testing, temperature, and soil moisture. Technical abbreviations will be explained when first used to ensure clarity. Soil samples weighing 200 grams each were collected from the field, and a total of three replicates were taken for each sample. The testing procedure was conducted at the environmental laboratory of FPMIPA UPI. During the study, the biotic factors were observed included vegetation in the surrounding area and plants commonly found with *N. gymnamphora*.

3 Results and discussion

Observations and data collected on Mount Pasir Cadas Panjang Ciwidey have provided information on the morphological characteristics of *N. gymnamphora* growing at different altitudes on the mountain. On this mountain, *N. gymnamphora* was found with two pitcher types: upper and lower. Nearly all *Nepenthes* species develop two types of pitchers: lower (lower type) and upper (upper type) [17]. Lower pitchers are small leaf structures on young saplings or stems still in the rosette stage. These structures are typically found near or attached to the soil surface. It is important to note that the technical term "pitcher" refers to these structures and should be used when discussing them. Occasionally, during stem growth and the growth of seated rosette leaves, the lower pitcher may be located on the upper stem. The upper pitcher are formed by the leaves on mature or climbing stems with spiral-seated leaves. The lower and upper pitchers comprise various parts, including tendrils, pitcher body consisting of the wax zone and digestive zone, wings, peristome/pitcher lip, and pitcher lid [14-16]. Based on these distinct features, observations were conducted, and the resulting data is presented in Table 1.

According to Table 1 findings, the distribution of *N. gymnamphora* on Mount Pasir Cadas Panjang Ciwidey (2050 masl) can be categorized into distinct altitude areas. Specifically, it can be divided into six distribution areas with varying altitudes. The shared characteristics of these six distribution areas are the type of pitcher and leaf growth phase, both of which consist

of sitting leaves. Lower and Upper pitcher types with sitting leaves in the rosette and spiral formations are present across these six distribution areas. This statement aligns with previous research that indicates *Nepenthes* typically possess two types of pitchers that mature differently depending on the type of pitcher [17-19]. These are lower types (rosette) and upper types (spiral). Color variants in *Nepenthes* were analyzed from six distinct distribution areas. Results showed that the color of *Nepenthes* pitchers tended to be green in the lower areas and reddish towards the higher areas. The reddish hue progressed into full red as the altitude increased. The diversity of color variations was also evident at stations 4, 5, and 6, which are in higher distribution areas than stations 1, 2, and 3. This is probably influenced by distinct chemical and physical environmental factors at each altitude of the *Nepenthes* distribution region [20-22]. Observations about the parts of the pitcher were made by taking a sample of the pitcher and being shown the name of the part, as shown below.

Table 1. Variations in pitcher colour from each altitude region

Areas	Colors	Pitchers type	Leafs type	Number of Individuals	Altitudes (masl)	Coordinates
1	Green	Lower	Rosette	97	1959-1968	S07°07'35.4" E107°23'26.3"
		Upper	Spiral			
2	Pattern	Lower	Rosette	50	1971-1990	S07°07'34.2" E107°23'23.7"
		Upper	Spiral			
3	Full Red/Reddish	Lower	Rosette	25	1994-2002	S07°07'34.2" E107°23'21.8"
		Upper	Spiral			
4	Green	Lower	Rosette	130	2006-2048	S07°07'32.3" E107°23'14.0"
		Upper	Spiral			
	Pattern	Lower	Rosette			
		Upper	Spiral			
	Full Red/Reddish	Lower	Rosette			
		Upper	Spiral			
5	Pattern	Upper	Spiral	85	2012-2015	S07°07'39.2" E107°23'12.5"
	Full Red/Reddish	Upper	Spiral			
6	Green	Lower	Rosette	150	1969-2009	S07°07'39.2" E107°23'12.5"
		Lower	Rosette			
	Pattern	Lower	Rosette			
		Lower	Rosette			
	Full Red/Reddish	Lower	Rosette			
		Lower	Rosette			

N. gymnamphora have been found with differences in pitcher morphological characteristics between individuals (Figure 2). Based on the observations of the pitcher parts depicted in Figures 3 and 4, it is evident that lower and upper types of pitchers share similar components. However, the wings of the upper type contain fewer hairs than those of the lower type pitchers. The abundant hair growth on the wings of the lower type pitcher serves as a foothold for soil insects to climb toward the mouth of the pitcher, leading them to get trapped inside. The upper type pitcher is designed to capture flying insects because its placement above the stem of the plant it supports via tendrils creates an environment where

wing hair modification is often reduced or absent. This agree with a previous study of the morphological characteristics of *N. gracilis* Korth in Samboja Kuala Village, Samboja District, Kutai Kartanegara Regency, East Kalimantan Province, which found that lower pitcher wings are crucial for both attracting prey and providing a foothold for pitcher climbing [5].



Fig. 2. Pitchers with colors variation.

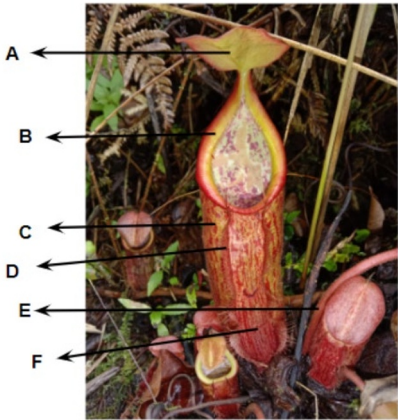


Fig. 3. A lower pitcher type of *N. gymnamphora* which divided into six parts : (A) A leaf-like lid, (B) Peristome, (C) Upper part, (D) Wings, (E) Tendril, and (F) Bottom part.

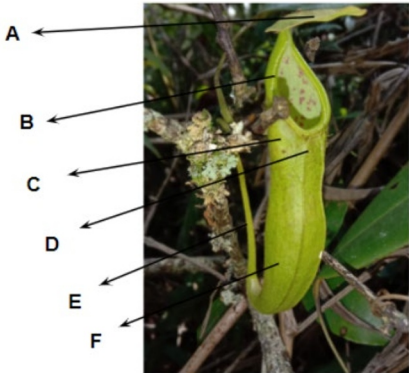


Fig. 4. An upper pitcher type of *N. gymnamphora* that can be divided into six parts : (A) A leaf-like lid, (B) Peristome, (C) Upper part, (D) Wings, (E) Tendril, and (F) Bottom part.

The prey composition of *Nepenthes* can be influenced by environmental conditions that affect insect diversity as well as field and *Nepenthes* conditions during sampling [17]. Variations in prey composition can be influenced by pitcher age and affecting trapping efficiency. Another influence is the level of digestibility of prey in the pitcher. Some taxa of prey, especially small ones such as Diplura and Diptera [23-25]. The enzymes chitinase and protease secreted by *Nepenthes* can digest part or all of the body of these small organisms. The study previously stated that they successfully isolated protein from the liquid of the upper and lower pitchers of *N. gymnamphora* from Gunung Halimun National Park. One milliliter of protein could be purified from each 800 mL of collected liquid. Activity tests on the purified protein indicated it was a protease enzyme, most likely Nepenthesin I and *Nepenthesin* II [3,4].

The structure of the pitcher influences the composition of prey in the pitcher type. The upper and lower pitchers of *N. gymnamphora* have different structures, especially in shape and color. The upper pitcher has a dominant green color with a purplish red pattern with green or reddish lips, while the lower pitcher is predominantly purplish red with green lips. The contrast pattern in the upper pitcher of *Nepenthes* attracts insects that perceive light in the ultraviolet spectrum, especially flying insects. An unattractive lower pitcher reduces the risk of the overabundance of prey, which can trigger pitcher rot and protect pitchers from herbivores. Other factors for attracting prey include the diameter of the pitcher opening, the cone of the pitcher, aroma, nectar, and the structure of the pitcher's lip, which have not been studied much in *N. gymnamphora*. Pitchers of *N. gymnamphora* are also microhabitats for Diptera larvae, namely mosquitoes (Culicidae) and hoverflies (Syrphidae). Most of the Diptera larvae live in the upper pitcher, namely 117 Culicidae larvae and 18 Syrphidae larvae, while 22 Culicidae larvae and 7 Syrphidae larvae are in the lower pitcher [17]. Culicidae larvae as organic particle feeders and Syrphidae as scavengers and predators for Culicidae larvae help decompose *Nepenthes*' prey more quickly [25].

Once the parts of the pitcher were known, measurements were made of the pitcher parts based on their size, pitcher type, and color variation. According to the measurements taken from various pitcher components (Table 2), it has been determined that the size of pitcher parts is not distinguished by color variations but instead by the type and growth stage (leaf seat) of the pitcher. Pitchers with the upper type (seated spiral leaves) have a tendency to possess longer pitcher bodies than the lower type (seated rosette leaves). On the other hand, the lower pitcher's body and mouth diameter are broader than the upper pitcher's. This elongated pitcher with a smaller diameter is most likely a result of geotropism, as the Upper pitcher's growth response is more sensitive to the earth's gravitational force. The top of the Upper pitcher continues to grow upwards while the bottom binds to the stem of the associated plant. Additionally, the location of the upper pitcher above has a greater gravitational pull from the earth. On the contrary, lower-type pitchers exhibit low sensitivity to the Earth's gravitational pull due to their elevated location and absence of tendrils that bind to other plants' stems [26-30].

Differences in growth stage or leaf arrangement have an impact on the dimensions of *Nepenthes* leaves. As revealed in Tables 2 and 3, leaf width and length measurements show discrepancies between the rosette and spiral leaf arrangements. Rosette leaves display relatively smaller dimensions compared to spiral leaves. Leaves with a rosette type are sapling leaves and have a small size, whereas spiral-type leaves have matured [5, 16].

The environmental conditions where *N. gymnamphora* was located in the diverse Pasir Cadas Panjang Mountain forest displayed an inclined soil structure owing to its situation at the mountain's peak. *N. gymnamphora* was observed on either side of the exploration course, growing on the slopes of the mountain's ravine. Furthermore, *N. gymnamphora* was detected to be growing on the ground and trailing on the forest shrubs.

Table 2. The pitcher's size measurement

Colors	Pitcher s type	Leafs type	Length pitcher s (cm)	Wide bottom s part (cm)	Wide Peristo m (cm)	Length Peristo m (cm)	Length Lids (cm)	Wide lids (cm)	Wide e leafs (cm)	Length h Leafs (cm)
Green	Lower	Rosett e	3.6- 9.5	1.1- 2.5	0.8- 2.1	1.1- 3	1.3- 3.5	1.1- 2.6	1.3- 3.2	2.6- 8
	Upper	Spiral	2- 12	0.5- 1.4	0.4- 2.7	0.5- 3.6	0.7- 3.3	0.5- 2.8	3.7- 7.1	9- 30
Pattern	Lower	Rosett e	3.8-13.5	1- 3.3	1- 3.7	1- 4.6	1.1- 4.7	1- 3.3	1.7- 3.9	4.3- 10.4
	Upper	Spiral	6.7-16.6	1.5- 2.6	1.6- 3.4	1.7- 4	1.7- 3.6	1.6- 3.5	3.6- 4.8	8-18.2
Full Red/Reddis h	Lower	Rosett e	2.7-10.3	0.9- 2.8	0.7- 3.6	0.8- 4.5	0.8- 4.4	0.7- 3	0.6- 4	2.4- 10.5
	Upper	Spiral	4.8-13.1	0.8- 2.1	1- 3.4	1.3- 5	1.3- 4.1	1.1- 3	5.9- 3	21.5- 13.5

Abiotic factors were discovered during this analysis in air temperature at the location where *N. gymnamphora* grew. The location's air temperature ranges between 17.9-21.5°C with an air humidity of 62.73-92.76%. This provides evidence that *N. gymnamphora* grows in bunches when the environmental conditions are suitable. The air temperature reduces, and the humidity increases as the altitude increases. These findings align with the previous report, which indicates that *Nepenthes* plants in highland regions grow in temperatures ranging from 10-30°C, and the other study suggests that *Nepenthes* can thrive in air humidity levels of 70-95% to meet their survival requirements [31-35]. Another abiotic factor affecting *N. gymnamphora* growth is light intensity. The observation location recorded a range of 1122-6615 lux. Tree vegetation affects the light intensity, with higher altitude regions having less vegetation and becoming more open. Consequently, open areas experience higher intensities of light. The high light intensity at the research site is due to the moderately to very open canopy cover, resulting in favorable light conditions. This high intensity significantly influences the adaptation level of *Nepenthes* plant species. Some *Nepenthes* types are capable of thriving in full sunlight and prefer direct sunlight exposure [8].

Soil condition constitutes a key factor that impacts the development of *N. gymnamphora*. The research site demonstrated a widened range of 19-21.5°C soil temperature, corresponding to 14-24% average soil moisture values, with higher locations exhibiting escalating moisture levels. It should also be noted that the pH degree of the soil plays a critical role in the plant's development. It should also be noted that the pH degree of the soil plays a critical role in the plant's development. It should also be noted that the pH degree of the soil plays a critical role in the plant's development. The tested soil exhibited mildly acidic or neutral values from 6.8- 6.9. Previous research suggests that forest soils tend to be acidic, with a pH below 7 and values between 4 and 4.6 [36-38]. It should be noted that each species of *Nepenthes* plant displays varying degrees of adaptation to soil acidity. The other research showed that *N. tentaculata* hook. Has the ability not only to thrive in acidic soils but also to prefer places with an alkaline soil pH, specifically pH 7 [39-41]. Consequently, it can be inferred that *N. gymnamphora*, found in the diverse forest of Mount Pasir Cadas Panjang, can grow in environments with a soil pH ranging from slightly acidic to neutral.

Soil organic matter content ranged from 1.26 to 5.17% throughout the study area, with a negative correlation observed between altitude and soil organic matter content. In the

heterogeneous forest of Mount Pasir Cadas Panjang, where *N. gymnamphora* was discovered, soil organic matter content varied from low to high. The other research was found that soil organic matter is considered low if it is less than 2%, moderate if it ranges from 2-3%, and high if it exceeds 3% [12]. This contradicts to previous research that claim *Nepenthes* is exclusively found in nutrient-poor areas with low nutrient content [18]. Therefore, the presence of *N. gymnamphora* in the varied forest of Pasir Cadas Panjang Mountain is not limited to low soil organic matter content; it also occurs in soil conditions with high organic matter content.

Table 3. Range of abiotic factors

Air temperature (°C)	Light intensity (Lux)	Air humidity (%)	Soil temperature (°C)	pH Soil	Soil humidity (%)	Soil Organic Matter (%)
17.9-21.5	112-6615	62.73-92.76	19-21.5	6.8-6.9	14-24	1.26-5.17

Tall trees mark the Pasir Cadas Panjang Mountain's diverse forest, while towards the summit, the area becomes increasingly open and dominated by shrubs. The tree species include *Engelhardia spicata*, *Schima wallichii*, *Podocarpus neriifolius*, *Lithocarpus elegans*, *Lithocarpus indutus*, *Prunus javanica*, *Weinmannia blumei*, *Neolitsea javanica*, *Castanopsis argentea*, *Lithocarpus indutus*, *Litsea cubeba*, *Vaccinium varingiaefolium*, *Syzygium antisepticum*, and *Polyosma sp.* *Acronychia pedunculata*, *Turpinia sphaerocarpa*, *Dacrycarpus imbricatus*, *Sloanea sigun*, and *Macropanax concinnus* are among the plant species found in the heterogeneous forest of Pasir Cadas Panjang Mountain. Additionally, *N. gymnamphora* was observed browsing on shrubs such as ferns *Dicranopteris linearis*, *Dipteris conjugata*, *Pteridium sp.*, and *Selaginella sp.* in association with these plants. This study also identified several plant species, including *Ageratina riparia*, *Scleria bancana*, *Chromolaena odorata*, *Nephrolepis sp.*, *Brachiaria sp.*, *Panicum sp.*, *Curculigo capitulata*, *Melastoma malabatricum*, *Rubus molucanus*, *Rubus rosifolius*, *Austroeupatorium sp.*, and *Hedychium roxburghii*, which were found to be associated with *N. gymnamphora*. This finding is consistent with research conducted by Mansur at Gunung Halimun National Park, where *N. gymnamphora* exhibited associations and creeping behaviors in shrubs and ferns, specifically *Gleichenia linearis*, *Oleandra pistillaris*, and *Dipteris conjugate* [16]. This study serves as a scholarly resource for future research, providing detailed research data on the morphological and anatomical variations of *N. gymnamphora*.

4 Conclusion

The results showed that the shape of the lower and the upper pitchers differed, namely in the upper type; the bottom of the pitcher was enlarged like a jug, then narrowed towards the top, then enlarged again to the peristome or lip of the pitcher. In the lower type, the diameter of the lower part is enlarged to the peristome. The parts of the pitchers in either the upper or lower type have general similarities, the only difference being in the wings. Colors found include green, patterned green, and full red, with the information that the higher the range, the more likely the pitcher is to be reddish or patterned. Based on the results of characterizing differences in pitcher morphology, it is known that these differences have respective roles in prey attraction and photosynthesis. The green on the pitcher is used in photosynthesis, while the patterned green and full red are used to attract insects.

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