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REVEALED CONNECTIVITY THE ENDEMIC SPECIES OF HALMAHERA WALKING SHARK (Hemiscyllium halmahera, Allen 2013) IN THE HALMAHERA SEA, INDONESIA, AS CONSERVATION EFFORTS

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Abstract

Endemic species are animals that are limited by geographic latitude and are not found in other locations. The Halmahera Walking Shark (Hemiscyllium halmahera) is an endemic species found in the sea of the Halmahera Islands. North Maluku, Indonesia. The research objective is the aspect of habitat connectivity that occurs based on morphometric characters. Sampling locations were Morotai Island, Kao Bay, and Tawabi Island. Kayoa. Sample collection was carried out in coral reef and seagrass ecosystems, mangroves, and bycatch products. The morphometrics of each individual are measured with precision, and weight is measured. Analysis Principal component analysis (PCA) was carried out to see the distribution and relationship of morphometric characteristics between locations and coral life forms and morphometrics. Cluster analysis (CA) was performed for proximity between locations. The research results found that there were morphometric variations between locations, although not significant. The distribution of species was found in the coastal areas of each sampling location, with a total of 30 individuals (Morotai Island), 33 individuals (Kao Bay), and 30 individuals (Tawabi Kayoa Island). This study has found a novelty that statistically answers that there is morphometric connectivity of the Halmahera Walking Shark based on habitat in the Halmahera Sea. Our findings have answered that the morphometric characteristics of the Halmahera Walking Shark on Morotai Island and Kao Bay are connected, namely, the larger size compared to the morphome size on Tawabi Island, Kayoa, which is smaller based on habitat. Novelty is also found statistically in habitat preferences in coral life forms that have been descriptively found to be true. Habitat preferences are found to be different in each location and are found to be the habitat of this species. Important information we find to provide data for species and habitat conservation.

Keywords: *Bioecology; Coral triangle; International Union for Conservation of Nature (IUCN); Near Threatened (NT); Population biology*

Introduction

Geographically, Indonesia's eastern region is home to the North Maluku Islands. The Pacific Ocean, Maluku Sea, Halmahera Sea, and Seram Sea all abut these islands. The ring of fire around the planet includes the North Maluku archipelago as well. Pacific Ocean seawater has an impact on the waters in the North Maluku Islands. Distinct marine waters are created by the passage

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of the Indonesian Throughflow (Arlindo), which transports large amounts of warm seawater from the Pacific to the Indian Ocean [1], [2], [3]. Islands are developed in a complex way, as demonstrated by geological formation processes. Halmahera Island's landmass originated differently from Seram Island, with Halmahera Island's geological features pointing east [4]. Parts of the Sorong Island fault plate, which have been created since the Pliocene age, have been discovered to the south of Halmahera Island, which forms a block to the west and is bounded by the Maluku Sea [5], [6]. In the past, seismic activity led to the creation of distinctive habitats with a wide variety of wildlife [7]. The North Maluku archipelago is a biodiversity hot spot due to its extensive biodiversity, which benefits from the diversity and uniqueness of the newly produced marine fauna. Because the North Maluku Islands are a part of the coral reef triangle, the water is home to a variety of rare and abundant natural resources [8].

The North Maluku Islands region is home to a wide diversity of endemic species due to the combination of geological processes related to island formation in the past, oceanographic influences, and the world's active mountain belt, or ring of fire. A variety of biota and endemic species are provided by geographic location, geological events, and marine features [9]. Endemic species are animals that are limited by geographic latitude and are not found in other locations. North Maluku's Halmahera Islands are home to the rare Halmahera Walking Shark (Hemiscyllium halmahera). The North Maluku archipelago's Halmahera Island lies to the south of this species, which was found by [10]. This species is distinct in that it swims using its pelvic and dorsal fins, giving the impression that it is walking rather than swimming [10], [11]. Papua, Papua New Guinea, Australia, the Aru Islands, and Halmahera are home to nine different species of walking sharks [11], [12]. "Phenotype" is typically used to illustrate differences across Walking Shark species [10], [13]-[17]. The phenotypic traits of Walking Shark species are similar in every place, although there were no discernible morphological distinctions. This could be because of the habitats in which they live. According to several studies [10], [13], [15], [18]-[23], the habitat of walking sharks is found in mangrove, seagrass, and coral reef habitats. Coral, seagrass, mangroves, depth, sand substrate, salinity, rock, temperature, pH, currents, sponges, and dead coral are the characteristics of the walking shark's habitat [24], [25].

When it comes to identifying the similarities and differences among species, ecology is crucial. The habitat between sites affects the connectivity of the Halmahera Walking Shark population between areas. A population of marine species' morphology, phenotype, and genetic makeup can all be influenced by the connectivity of their habitats. These three ecosystems are crucial for the connectivity of the fish, mangrove, seagrass, and coral ecosystems [26]. Abiotic elements (stones, water, and sand) as well as plants and animals interact with one another within habitats [27].

Habitat similarity between locations forms habitat connectivity, which influences the morphological similarities and differences of a species. The landscape science approach (seascape ecology) is important to understand the interactions of marine animals with their habitat [26]. Connectivity refers to the extent to which organisms or natural processes can move unhindered across both terrestrial and aquatic habitats. Habitat connectivity addresses the way ecological processes, such as the movement of individual animals, are influenced by the arrangement of elements in the landscape [27]. The interaction between Walking Sharks and their habitat is important to know. Morphology is important for seeing morphometric similarities between populations, important information to support biological data, and information for determining protected endemic species. Connectivity based on morphometric habitat is critical to life history and anthropogenic pressure metapopulation structure [28]. It is important to know the connectivity morphometric based on habitat characteristics to describe associations and the importance of ecological protection. The hypothesis of this research is that there is suspected population connectivity based on morphometric characteristics in different regions. Our research is different from previous research, where we answer a hypothesis about the existence of morphometric connectivity relationships between islands based on habitat. The aim of the research is to reveal connectivity based on the morphometrics of the Halmahera Walking Shark in different areas. This research is very informative and answers previous findings with statistical analysis.

Experimental part

The main objective of this study is to examine the morphometric connectivity between populations in three different regions. Morphometrics is a biological element used to assess relationships based on statistical cluster and morphological tests. This study aims to obtain kinship information based on morphometric and habitat characteristics. This information is scientifically important for addressing the relationship processes caused by past geology. This data is crucial for describing how species and habitats will be protected in the future. It scientifically describes past moments that may have occurred due to geological shifts and similarities in morphology.

Materials and Methods

Sampling box for sample collection, mask, snorkel, wetsuit, Scubapro booties, gloves, cloth ruler, iron ruler, seawater, sampling board, digital scale, cannon camera, underwater cannon camera, and sampling boat.

Sampling

Sample collections of Halmahera Walking Sharks (Hemiscyllium halmahera) on Morotai Island, namely South Morotai, North Morotai, West South Morotai, and East Morotai. Data collection in Kao Bay on the coast, North Kao District, North Halmahera Regency. Halmahera Walking Shark Sampling on Tawabi Island, Kayoa (Latitude 0°) (Fig. 1).

Morphometrics

Samples were taken at each location, targeting 30-33 individuals. Sampling was carried out by diving in shallow water at depths of 1-15 meters. Sampling of Halmahera Walking Sharks was also conducted in mangrove and seagrass ecosystems. Sampling was done using gloved hands. Halmahera Walking Sharks were searched for in the root systems of mangroves and seagrass at low tide. Morphometric characteristics were measured based on previous research guidelines, and weight was measured using a precision digital scale (Fig. 2) [13], [15].

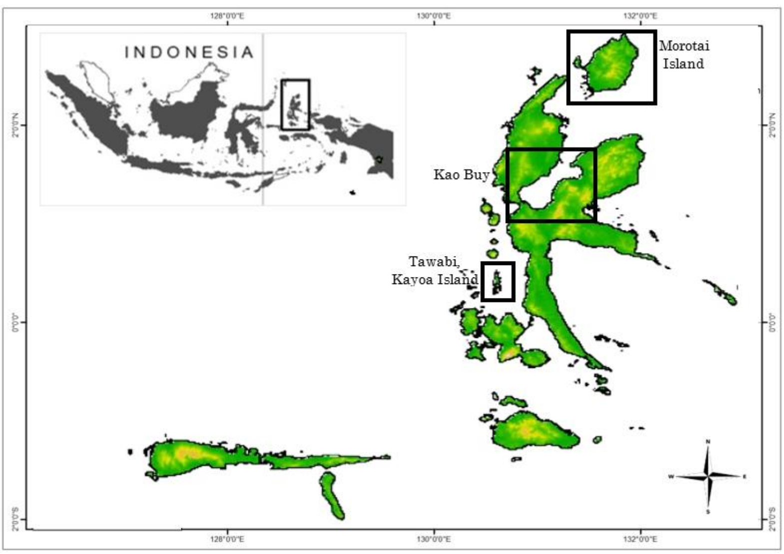

Fig. 1. Site sampling Halmahera Walking Shark on Morotai Island, Kao Bay and Tawabi Island

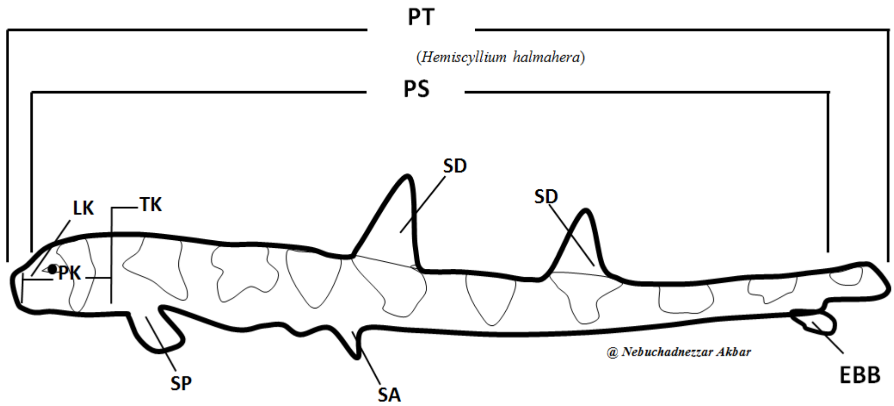

Fig. 2. Morphometrics of the Halmahera Walking Shark. PT (total length), PS (standard length), TK (head height), PK (head length), LK (head width), LT (body circumference), SP (pectoral fin), SA (anal fin), SD (dorsal fin), and EBB (back tail) [10], [13], [15]

Data analysis

The relationship between morphometrics and habitat (coral life forms) was analyzed using *Principal Component Analysis* (PCA) with XLSTAT software [29]. *Principal Component Analysis* (PCA) in the scientific implementation of the field of ecology is used to characterize the relationship between environmental components that affect the species and research site; besides that, it aims to simplify variables into different components so that it can explain simple correlations between variables [29], [30].

Results and discussion

Morphometrics

The *H. halmahera* population has a range of morphometric sizes, according to morphometric measures (Tables 1, 2 & 3). The distribution of the Halmahera Walking Shark population in the waters surrounding Morotai Island can be explained by a variety of size-based classes, as indicated by the different morphometric sizes (Table 1). Because it has many effects, the research station's location can have an impact on the morphometric size distribution of fish. The Halmahera Walking Shark's morphology is greatly influenced by the circulation of Pacific Ocean currents and the richness of food sources, as evidenced by the shark's overall length. The Halmahera Walking Shark's total body length is believed to be influenced by the strength of the Pacific Ocean currents. Strong surface and deepwater currents may make it more difficult for the Halmahera Walking Shark to migrate. The amount of work the fish must put in to swim against the stream or to cross it may have an impact on its size. Fish must expend energy throughout the migration phase, which is followed by muscle action that allows them to grow larger. Since Kao Bay is a semi-enclosed location with a high total length, it is likely that there are plenty of food sources nearby, reducing food rivalry (Table 2). The Walking Shark of Halmahera on Tawabi. In comparison to the two previous locations (Morotai Island and Kao Bay), Kayoa Island is smaller this is believed to be a result of variations in the area's food and external energy supply (Table 3). On Tawabi Island, oceanic dynamics (currents, waves, and tides) are generally present. Kayoa's health is normal.

Table 1. Morphometric characteristics of H. halmahera on Morotai Island [9]

Characteristics	Maximum	Minimum	Averages	Deviation Standar (SD)
PT	81	21.5	51.3	42.07
PS	72	16.5	44.3	39.24
TK	6.4	2.3	4.4.	2.90
PK	15.4	2.1	8.8	9.40
LK	8.6	3.8	6.2	3.39
SP	7.7	3.7	5.7	2.83
SA	6.7	2.2	4.5	3.18
SD	6.1	2.3	4.2	2.69
EBB	3.8	1.5	2.7	1.63
LT	19.7	9.2	14.5	7.42
B	613	120	366.5	348.60

Note: PT-EBB = cm, and Weight (B) = gr

Table 2. Morphometric characteristics of H. halmahera on Kao Buy [25]

Characteristics	Maximum	Minimum	Averages	Deviation Standar (SD)
PT	76	22	49	38.2
PS	72	55	38.5	47.4
TK	6.2	2.3	4.25	2.8
PK	16	6	11	7.1
LK	8	4.8	6.4	2.3
SP	8	3.1	5.5	3.5
SA	6.3	3.2	4.7	2.2
SD	7.2	3.6	5.4	2.5
EBB	35	14	18	16
LT	31	7	19	17
B	550	200	375	247.5

Note: PT-EBB = cm, and Weight (B) = gr

Table 3. Morphometric characteristics of H. halmahera on Tawabi Island. Kayoa [25]

Characteristics	Maximum	Minimum	Averages	Deviation Standar (SD)
PT	65	48	56.5	12
PS	62	44	53	12.7
TK	5.6	3.6	4.6	1.4
PK	15	8	11.5	4.9
LK	7	5.5	6.2	1.1
SP	6,7	4.9	5.8	1.3
SA	5.8	4	4.9	1.3
SD	6.4	4.5	5.4	1.3
EBB	35	2.6	18.8	22
LT	26	12	19	9.9
B	510	300	405	148.5

Note: PT-EBB = cm, and Weight (B) = gr

Sex Ratio

The east, west, and transition monsoons are the periods (seasons) that cause changes in ocean dynamics. The fish's efforts to move by using extra energy are most likely unaffected by this typical condition; hence, the size is normal. The physical traits of the Halmahera Walking Shark species are often not very different from one another. The morphology, physiology, and kinematics of the Hemiscyllium family are comparable; yet, the phenotype serves as a unique indicator or benchmark among the species [15], [31]. According to [32], [33], [34], the total length varies and is believed to be impacted by isolation factors, geographic location, genetic structure and habitat conditions, nutrients, water topography, migrating patterns, and diet.

Three research locations, Morotai Island (30 individuals), Kao Bay (33 individuals), and Tawabi Island, saw the discovery of Halmahera Walking Sharks (30 individuals) in Kayoa (Table 4). According to data, the Halmahera Walking Shark is located around the coasts of Tawabi Island, Kao Bay, and Morotai Island, with an average of 5-6 individuals per location. Since the species is solitary, there is little chance that one individual will be found in a given area, so the frequency of individual presence is low. Despite having a sinusoidal movement pattern, meaning it rotates around its surroundings [35], the Halmahera Walking Shark's appearance is unpredictable due to its constantly nomadic mobility. The Halmahera Walking Shark has an unexpected distance between individuals and a random distribution pattern, according to [36]. Although there are one or two individuals of this species in coastal environments, each individual's movements are unique. Walking sharks seem to be opportunistic hunters who use electrical receptacles, scent, and barbels to bring down their prey [10], [37].

Table 4. Population distribution of Halmahera walking sharks in the North Maluku Archipelago Sea. Indonesia

Location	Sex		Totals
	Male	Famale	
Morotai Island	16	14	30
Kao Buy	13	20	33
Tawabi, Kayoa Island.	21	9	30
Totals	50	43	93

In Morotai Island (16 males and 14 females), Kao Bay (13 males and 20 females), and Kayoa Island (21 males and 9 females), the sexes were present in every location (Table 4 and Fig. 3). There was balance in the overall number of sexes discovered. 50 people are male and 43 are female, making up the proportion (Table 4). Given that H. halmahera still finds two sex partners, its sex percentage is within normal limits. The existence of two sex pairs suggests that the population's reproductive mechanism is operating normally. The physical features of the sexes are located in the lower abdomen, with the males having an extended torpedo-like shape and the females having a small hole near the dorsal fin. Similar findings have been made about the distribution and ratio of the sexes in studies [10], [11], [13], [15], [17], and [36].

Fig. 3. Sex of *Hemiscyllium halmahera*. a). Male and b). Female. Photo by Nebuchadnezzar Akbar (point a) and Abdurrachman Baksir (point b)

PCA analysis for Connectivity

Two groups were identified by the PCA analysis results, which examined the distribution and morphometric features at three stations: Morotari Island, Kao Bay, and Tawabi Island (Figure 4). The first group demonstrates how the morphometric measurements of PT (total length), TK (head height), PK (head length), LK (head width), LT (body circumference), SP (pectoral fin), SA (anal fin), SD (dorsal fin), and EBB (back tail) define the Halmahera Walking Shark on Morotai Island and Kao Bay on the F1+ axis (Fig. 4). Halmahera Walking Sharks were discovered by the second group on the F2-axis on Tawabi Island. These sharks are distinguished by low PS (standard length) morphometric measures (Fig. 4).

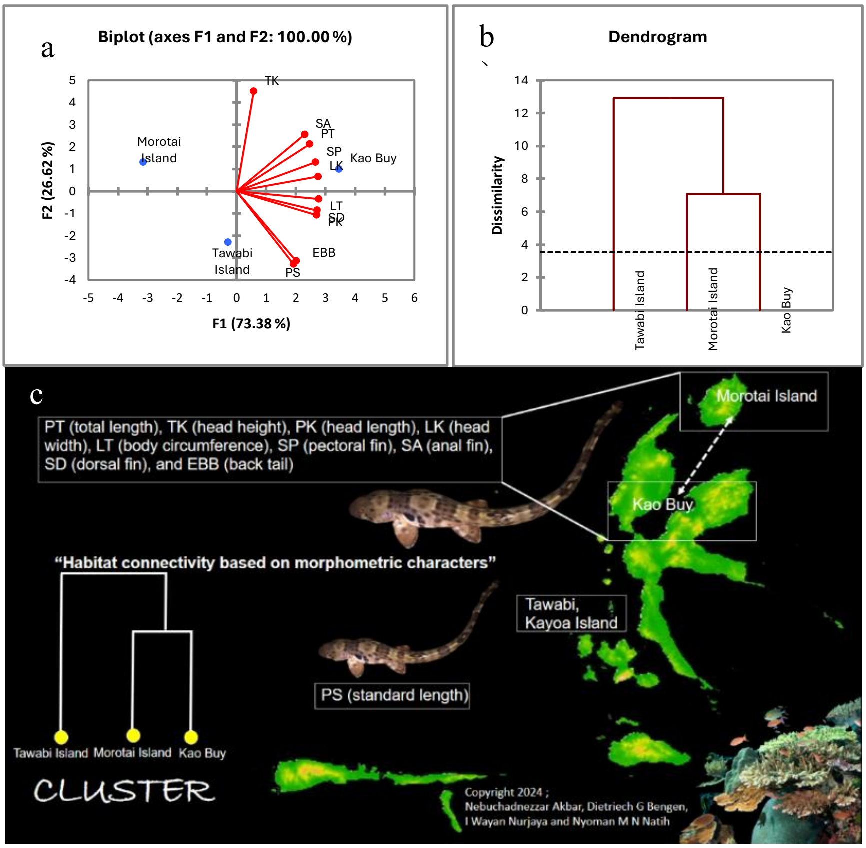

Fig. 4. Principal component analysis (PCA) of the Halmahera Walking Shark population on Morotai Island, Kao Bay, and Tawabi Island. a) Correlation between morphometric variables and research stations; b) Dendrogram of morphometric variables based on research stations, and c) Description of habitat connectivity based on morphometric characters (Illustration of PCA and Cluster analysis results)

According to the multivariate analysis results on ten morphometric measurements, component one's (F1) and component two's (F2) percentages of variance were 73.38% and 26.62%, respectively, with a total variance of 100% (Fig. 4a). According to [38], the component values obtained indicate that the two components, F1 and F2, have furnished information and clarification concerning the correlation between fish morphometric attributes and stations. A method for lowering a data set's dimensionality and improving interpretability while also avoiding information loss is principal component analysis (PCA) [39]. Among the earliest and most popular is principal components analysis (PCA) [39]. Based on cluster analysis, the population of Halmahera Walking Sharks in Kao and Morotai Bays is divided from Tawabi Island and forms a cluster (Fig. 3b). The outcomes of the PCA analysis are reinforced by the cluster analysis results (Fig. 4b). The clusters are connected and adjacent, indicating that the populations are morphologically connected [41]. Adjacent clusters are possible due to morphological similarities, and there is an exchange of genetic material with each other [41], [42].

The similar morphometric features of Morotai Island and Kao Bay may be attributed to the interconnectedness of the Halmahera Walking Shark population and habitat between the two sites (Fig. 4c). Morotai Island and Kao Bay are connected by a body of water, specifically the Indonesian Throughflow (Arlindo), which connects the waters of the Halmahera Sea to the Pacific Ocean. The Indonesian Throughflow (Arlindo), which reaches the western region of the North Maluku archipelago, and the waters of the Maluku Sea connect Morotai Island and Tawabi Island. Comparing the two places geographically to Tawabi Island, which is located in the southern portion of the Halmahera archipelago, reveals that the northern portion of the two locations is closer together. The distance between Morotai Island and Tawabi Island is 285 km, while the distance between Morotai Island and Kao Bay is 165 km. Because Tawabi Island and Kao Bay are on separate areas of the island and are separated by Halmahera Island's mainland, their waters are not connected. Due to previous geological episodes where island formation occurred adjacent to one another, the Halmahera Walking Shark on Morotai Island and Kao Bay have strong morphometric size connections. It was discovered that 20 million years ago, the landmasses of Halmahera Island and Morotai Island appeared together in nearby geographic locations [12]. According to [12], this emergence is also connected to the establishment and dispersal of new Hemiscyllium species and populations, known as Hemiscyllium halmahera.

Based on morphometric characteristics that showed a bigger size than the Halmahera Walking Shark (H. halmahera) habitat on Tawabi Island, we discovered the interconnectedness of the habitat in the waters of Morotai Island and Kao Bay. Kayoa (Fig. 4c). Based on the distance of the location of Morotai Island and Kao Bay, which is 165 km, compared to the distance between Morotai Island and Tawabi Island, which is 285 km, and the distance between Kao Bay and Tawabi Island, which is 122 km but is limited by the Halmahera mainland. Tawabi and Kayao are very far apart due to sea connectivity being blocked by the mainland. Morphological similarities and closeness have been reported [13], but not statistically, but using a manual morphometric approach. The relationship between morphometric characteristics was also obtained [15] between Maitara Island, Central Halmahera, South Halmahera, and Tidore Island but did not show and explain the existence of cluster proximity.

Our research has answered statistically, not descriptively, that there is morphometric connectivity based on habitat between islands. Adjacent characteristics and forming a kinship have been answered with PCA analysis. Morphometric kinship between the Halmahera Walking Shark in Kayoa Bay and Morotai Island, which is possible due to the relationship of habitat and proximity of location, has been found. This discovery has never been done and proven morphometrically or analyzed for association with habitat. This novelty is very important, so we conclude firmly that the Halmahera Walking Shark in Kao Bay and Morotai Island is related morphometrically, based on large size, to connectivity that occurred in the habitat in the past. We are very confident that the statistics have supported the morphometric measurements that we found, where the similarity between the Halmahera Walking Shark in Kao Bay and Morotai Island lies. The large morphometric measurements in both locations were supported by the proximity of the habitat. Morphometric connectivity based on habitat is a key and the main answer to be proved in research using the DNA molecular method. *Principal component analysis* (PCA) was used to summarize genetics and describe similarities in population structure and admixture between populations [42]. Genetic cluster information has been reported [22], which explains that there is genetic isolation so that closeness differs between populations even though there is genetic gene flow.

Three groups emerged from the principal component analysis (PCA) results used to characterize the distribution of features between coral population, location, and life form at three stations (Morotai Island, Kao Bay, and Tawabi Island) (Fig. 5a). On Morotai Island, the first group of sharks are known to inhabit the life form habitats of Acropora encrusting (ACE), Coral foliose (CF), Acropora submassive (ACS), and Coral submassive (CS) (Fig. 5a). These sharks are the small, medium, and giant Halmahera Walking Sharks. The primary feature of Tawabi Island's ecosystem, which is the coral life forms Acropora branching (ACB), Acropora digitate (ACD), Coral branching (CB), and Coral massive (CM), is often the second group of medium- and big Halmahera Walking Sharks (Fig. 5a). The third group, which includes small, medium, and giant Halmahera Walking Sharks in Kao Bay, is located on the F2+ axis and is distinguished by two key features of the habitat of coral life forms: Acropora tubulate (ACT) and coral mushroom (CMR) (Fig. 5a). Multivariate analysis results showed a high total variance value of 69.14% for component one (F1) and 30.86% for component two (F2), with a total variance of 100% (Fig. 5a). According to cluster analysis, Morotai Island belongs to a different group while Tawabi Island and Kao Bay form one (Fig. 5b).

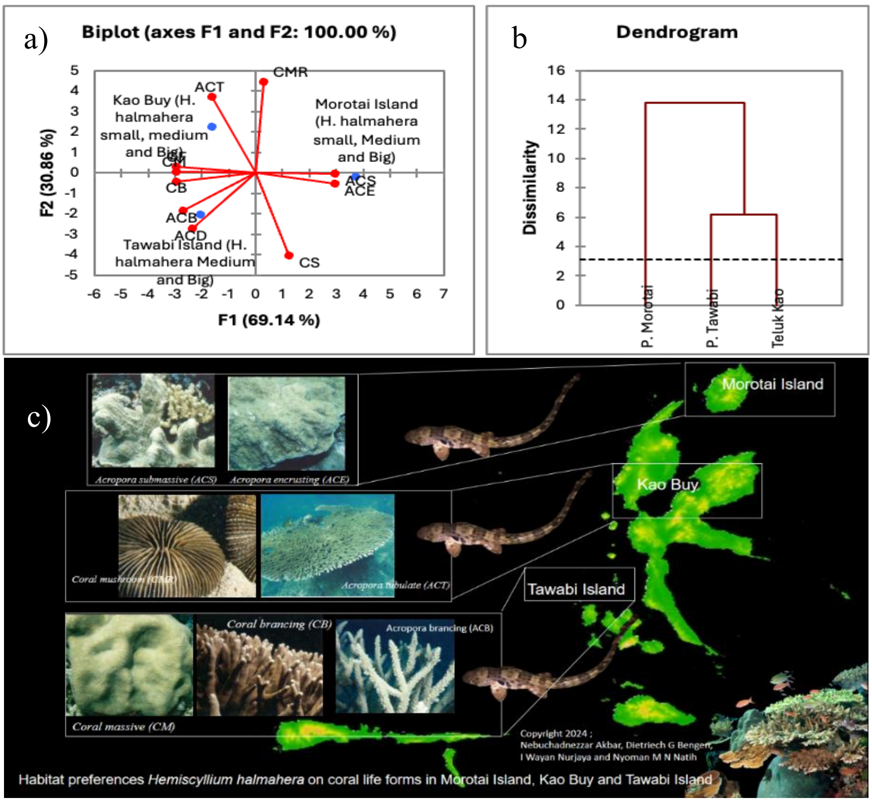

Fig. 5. Principal component analysis (PCA) of the Halmahera Walking Shark population on Morotai Island, Kao Bay, and Tawabi Island. a) Correlation between population size, research station, and life form; b) Dendrogram of morphometric variables based on life form at the research station and c) Habitat preferences of the Halmahera Walking Shark on coral life forms at the observation location

Small to large Halmahera Walking Sharks show a strong association with the coral life form Acropora encrusting (ACE) because this coral life form has dominance and a high percentage of cover in the waters of Morotai Island (Fig. 5). This life form is used as the main habitat for shelter during the day, resting and looking for food, because it has gaps and holes at the bottom of the reef (Fig. 5). The medium and large Halmahera Walking Sharks were specifically found to have the main characteristics of the coral life forms Acropora branching (ACB) and coral massive (CM), allegedly due to having sufficient gaps and space for hiding, resting, sheltering, looking for food, and growing (Fig. 5). Small, medium, and large Halmahera Walking Sharks in Kao Bay have the main characteristics of the Acropora tubulata (ACT) life form habitat with high cover values (Fig. 5).

Because it has gaps and openings beneath the reef, this coral life form has a strong link with environmental characteristics and can use it as a place to eat, lay eggs, and find shelter. Although they are found in mangrove and seagrass ecosystems, Halmahera Walking Sharks are found in coral reef locations [9], [10], [11], [13], [15], [17], [21], [36], [40]. The Halmahera Walking Shark species prefers this ecology because coral reef areas offer an abundance of invertebrate and invertebrate resources [15].

We answer previous findings that describe that the Halmahera Walking Shark is associated with the coral life forms Acropora tubulata, Acropora encrusting, Acropora branching, Acropora massive, Acropora submassive, coral mushroom, and coral branching. PCA statistics have supported the description of previous studies. Statistics that answer the existence of a strong association between the Halmahera Walking Shark and coral life forms are a novelty of this study. Previous studies only mention where the Halmahera Walking Shark was found without statistical analysis. This research is a novelty that has just been scientifically proven in this research. Previous information has not found morphometrics that are analyzed differently by finding similarities based on PCA analysis. This information is important to strengthen the findings and field visualizations when the Halmahera Walking Shark association is found with coral life forms. The scientific evidence that we found statistically is a key to understanding the importance of coral life forms to the Halmahera Walking Shark.

Conclusions

The observed variation in the morphometric size of Hemiscyllium halmahera is believed to be the result of different environmental features found at each site. Based on morphometric size, significant differences in the Halmahera Walking Shark have been observed. This species is sporadically distributed in coastal areas with a normal distribution of male to female sex ratios. The fact that both sexes can still reproduce has shown the viability of the population. We found a research novelty that statistically concluded morphometric connectivity based on habitat. This novelty is very helpful in answering how past geological processes resulted in kinship based on morphometric size due to habitat. The discovery of the proximity between the Halmahera Walking Shark on Morotai Island and Kao Bay based on morphometric connectivity is an advancement in science due to the statistical support carried out. Spatially, the distance between the two adjacent locations allows for habitat connectivity in the past. The geographical distance and historical island geological processes between Morotai Island and Kao Bay and Tawabi Island allow there to be no habitat relationship in the past. The novelty of our next research is answering statistically that the preference of the Halmahera Walking Shark has a tendency towards several coral life forms in our research location. It is important for science because this information can be a reason why the conservation of the Halmahera Walking Shark habitat must be done.

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