

# Morphometrics and Meristics of *Tubuca rhizophorae* in Ca Mau and Bac Lieu Provinces, Vietnam

# Anh Ngoc Tran<sup>1</sup>, Ton Huu Duc Nguyen<sup>2</sup>, Lam Thi Thao Vo<sup>3</sup>, Quang Minh Dinh<sup>4\*</sup>

- <sup>1</sup> Faculty of Biology, School of Education, Can Tho University, Xuan Khanh ward, Ninh Kieu district, Can Tho, Vietnam.
- <sup>2</sup> Faculty of Biology, School of Education, Can Tho University, Xuan Khanh ward, Ninh Kieu district, Can Tho, Vietnam.
- <sup>3</sup> Faculty of Biology, School of Education, Can Tho University, Xuan Khanh ward, Ninh Kieu district, Can Tho, Vietnam.
- <sup>4</sup> Faculty of Biology, School of Education, Can Tho University, Xuan Khanh ward, Ninh Kieu district, Can Tho, Vietnam. Email: dmquang@ctu.edu.vn

#### **KEYWORDS**

#### Mekong Delta, Morphometrics, meristics, <u>Tubuca</u> rhizophorae

#### **ABSTRACT**

This study investigates the morphological variations in the claw and walking leg characteristics of <u>Tubuca rhizophorae</u>, a fiddler crab species from the Ocypodidae family, collected from two coastal sites in the Mekong Delta, Vietnam. Monthly samples of <u>T. rhizophorae</u> were taken between December 2023 and September 2024 from Dong Hai, Bac Lieu, and Dam Doi, Ca Mau, with environmental parameters such as temperature, pH, and salinity recorded for each site. Morphological indices, including propodus length, manus length, pollex length, and dactyl length, were measured and analyzed. Results show that while the morphological index did not significantly vary between the two sites or across seasons, significant sex-based differences were observed, with male individuals displaying larger indices than females. Principal Component Analysis (PCA) indicated that temperature and salinity were substantial environmental factors influencing the morphological traits of <u>T. rhizophorae</u>. This research contributes to a deeper understanding of the species' ecological adaptations and morphological plasticity in response to environmental factors in mangrove ecosystems, providing valuable insights into the species' ecological role in the Mekong Delta.

#### 1. Introduction:

Ocypodidae family is one of the dominant groups of fiddler crabs found in tropical and near-tropical intertidal zones [1]. These crabs are known for their high species diversity and population density, and they play important ecological roles [2, 3]. Their diet consists of mangrove leaves, detritus on the substrate, and decaying organic matter from the detrital chain, which helps initiate food chains through organic fragments. By burrowing into the substrate for shelter and defense, they modify the pH levels between sediment layers and the surface, promoting sediment deposition, water retention, and substrate aeration after the tide recedes [4, 5].

Recent studies have identified 14 species of fiddler crabs within the Ocypodidae family in Vietnam [6], with *Tubuca rhizophorae* being commonly found in the Mekong Delta [6]. According to Shih (2022) [7], *Tubuca rhizophorae* is characterized by a relatively flat carapace, a small body size (with a carapace width of up to ~20 mm), a large manus with small tubercles, slender fingers, and a row of slightly enlarged pedestal teeth along approximately half the length of the inner margins of the pollex and dactyl. Crane (2015) [1] also noted that the sides of the carapace in this species are less convergent, and both sexes display a dark carapace with a distinctive marbled pattern in lighter tones, ranging from olive green to yellow or light brown.

These crabs are crucial to tropical and subtropical mangrove ecosystems and represent a diverse group living across various habitats [8]. However, limited information is available on species within the Ocypodidae family, particularly *Tubuca rhizophorae*, in the coastal regions of the Mekong Delta. Previous research on this species has primarily focused on taxonomy and morphological descriptions [1, 9]. Additionally, the specific morphological features of this species have not been extensively studied. Researchers are interested in examining differences in morphological traits across regions, as these variations can help address broader questions, including relationships between species or populations [10-13]. This study aims to provide new insights into the morphological parameters of the claw and walking legs of *Tubuca rhizophorae* in the Mekong Delta, Vietnam.

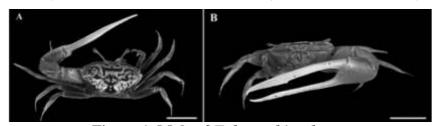


# 2. Materials And Methods:

## Sample Collection and Analysis

Tubuca rhizophorae (see Fig.1) were collected manually during the day in the intertidal areas along the riverbanks. The sampling was conducted from December 2023 to September 2024 with a monthly frequency at two sites in Dong Hai, Bac Lieu (DHBL) and Dam Doi, Ca Mau (DDCM). Common plant species at two sites were Acanthus ebracteatus Vahl., Avicen-nia marina (Forssk.) Vierh., Bruguiera gymnorrhiza (L.), Nypa fruti-cans Wurmb., Savigny Sonneratia caseolaris (L.) A. Engl., and Rhizophora apiculata Blume. Bruguiera gymnorrhiza were dominant at DHBL, respectively, whereas both Avicennia marna and Bruguiera gymnorrhiza were dominant at DDCM. An average of 30 samples per month were collected at each site. At each site, the samples were collected manually. The samples were randomly collected in different sizes. Subsequently, the samples were fixed and stored in 70% ethanol before being transported to the laboratory.

In total, all individuals were collected at two sites, of which at DDCM, there were 438 individuals (312 males: 126 females) and DHBL had 263 individuals (178 males: 85 females).



**Figure 1. Male of** *Tubuca rhizophorae* (A: top view; B: front view; scale bar: 10 mm)

In the laboratory, *Tubuca rhizophorae*, after being identified based on the claw morphological characteristics described by Tweedie (1950) [14], had the following morphological parameters determined: propodus length (PL), manus length (M), pollex length (P), dactyl length (D) (see Fig. 2). Measurements are taken with a caliper that was accurate to the nearest millimeter. The environmental parameters include temperature, pH, and salinity, which were 30.46°C, 7.79, and 26.02 in DDCM and 30.94°C, 7.74, and 32.97‰ in DHBL. The average pH (measured with a pH meter (Hanna HI98127)) and salinity (measured with a Refractometer 0-32 Brix (95000-002)) in each study site were presented; pH and temperature were determined once a month at each sampling sites at high tide [15].

The claw morphological ratios (M/PL, P/PL, D/PL) were used to determine the morphological characteristics of the species (see Fig. 2).

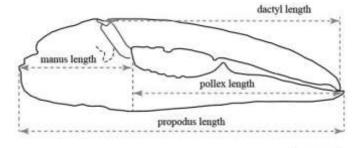


Figure 2. Illustration depicting the annotations of the morphological characteristics of the *Tubuca rhizophorae* 

(Scale bar: 10 mm (Dao et al., 2024 [16]))

#### **Data Analysis**

The t-test determined differences in propodus length, manus length, pollex length, dactyl length, and walking leg indices between male and female individuals and between two sites. Principal component analysis (PCA) was used to identify the factors (such as pH, salinity, or water temperature) that



influence these variations based on the research methodology of Nguyen et al., (2024) [17]. The tests were performed at a 95% confidence interval and using Jamovi 2.4.11.

#### 3. Results

# **Morphometric variation**

The study results on 701 individuals of *Tubuca rhizophorae* collected from DHBL and DDCM was not showed significant differences in the measured morphological indices PL, M, D, P, and L between the two sampling sites (see Table I).

Table I. Claw morphometrics' variation in *Tubuca rhizophorae* regarding sampling site

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Claw morphometric parameters	Sites	N	Mean	SE	t	р
Propodus length	DDCM	438	18.30	0.44	-0.173	0.48
	DHBL	263	17.77	0.62	-0.173	
Manus length	DDCM	438	8.02	0.18	1.113	0.27
	DHBL	263	7.68	0.25	1.113	0.27
Pollex length	DDCM	438	10.24	0.27	-0.349	0.73
	DHBL	263	10.08	0.38	-0.349	0.73
Dactyl length	DDCM	438	11.98	0.31	-0.284	0.79
	DHBL	263	11.83	0.44	-0.284	0.78

In Figure 3, data was divided by site: DHBL (blue circles) and DDCM (orange triangles). The distribution showed a clear separation between the two sites, with DHBL and DDCM data points clustering apart from each other. This indicates that sites had a significant influence on the measurements. The environmental vectors indicated that salinity (Sal) and pH had stronger influences at one site (DHBL) than the other (DDCM), while temperature (Tem) seems to play a more prominent role at DDCM. The divergence in the vectors' directions across the two sites suggested that each site had unique environmental conditions that impact the measurements differently. Site had a strong effect on the fiddler crab measurements. The differences in environmental impacts between sites underscore the role of geographic sites in shaping environmental conditions and, subsequently, the fiddler crab characteristics.

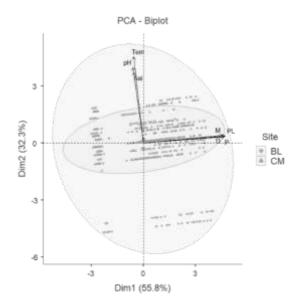


Figure 3. PCA illustrates relationship between some claw morphometrics and environmental factors of *Tubuca rhizophorae* species according to sampling sites (morphological measurements: PL: propodus length; M: manus length; P: pollex length; D: dactyl length; Tem: temperature; Sal: salinity)



The morphological indices of *Tubuca rhizophorae*, such as PL, M, P, D, and L, showed significant variations according to gender. Specifically, the PL, M, P, D, and L values in males were significantly higher than in females (t-test, p<0.001).

Table II. Claw morphometrics' variation in *Tubuca rhizophorae* regarding sex

Claw morphometric parameters	Sex	Number	Mean	SE	t	p
Propodus length	Male	490	23.5		-45.98	< 0.001
	Female	211	5.54	0.06	-43.96	
Manus length	Male	490	10.2	0.03	-58.19	< 0.001
	Female	211	3.02	0.03	-38.19	
Pollex length	Male	490	13.3	0.03	26.95	< 0.001
	Female	211	3.02	0.04	-36.85	< 0.001
Dactyl length	Male	490	15.6	0.03	-38.89	< 0.001
	Female	211	3.47	0.03	-38.89	< 0.001

The plot clearly illustrates the dispersion of data based on the two principal components: Dim1 (55.8%): This axis explains 55.8% of the total variance in the data, which means it represented the primary dimension of differentiation among the fiddler crabs measurement indices; Dim2 (32.3%): This axis explains 32.3% of the variance, which was also significant. Together, Dim1 and Dim2 account for 88.1% of the total variance, indicated that most of the data's structure was captured within these two dimensions.

Temperature (tem): The "Tem" vector was notably long and aligns mostly with Dim1. This suggestsed that temperature was a major factor influencing the variation along Dim1, and it significantly impacts the crabs' measurement indices. The longer vector length showed temperature's dominant role in explaining variance. Salinity (Sal) and pH: Both vectors were shorter and do not align as closely with Dim1 or Dim2, indicating that salinity and pH had a more moderate impact on the crabs' indices compared to temperature. Their directions suggested that they contribute partially along both dimensions, but their impact was less pronounced.

The data points were divided into two groups, female (F) and male (M), represented by circles and triangles, respectively. The ellipses showed the distribution of each gender in the PCA space. In this plot, there was a clear separation between female and male groups along Dim1. This separation implied that gender plays a significant role in the way fiddler crab respond to environmental factors, especially temperature, as Dim1 correlates strongly with temperature. Interpretation: This gender-based separation suggested that male and female fiddler crab might had physiological or behavioral differences that result in distinct responses to temperature variations. For example, females might be more sensitive to temperature changes than males, or vice versa, leading to differentmeasurement outcomes.

Temperature was the most influential factor impacting the fiddler crab measurement indices along Dim1. Gender differentiation was strong, especially along Dim1, suggesting temperature influences males and females differently. Salinity and pH had a more modest impact but may still contribute to the variation in fiddler crab indices, though they were not primary factors in this plot (see Fig. 4).



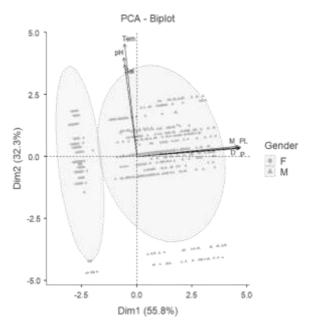


Figure 4. PCA illustrates relationship between some claw morphometrics and environmental factors of *Tubuca rhizophorae* species according to sex (morphological measurements: PL: propodus length; M: manus length; P: pollex length; D: dactyl length; Tem: temperature; Sal: salinity)

Morphological indices of *Tubuca rhizophorae*, limited to PL, M, P, D and L, were found to not vary according to the season of the year (see Table III).

Table III. Claw morphometrics' variation in *Tubuca rhizophorae* regarding season

Table 111. Claw morphometries variation in Tubuca Interpretate regarding season								
Claw morphometric parameters	Season	Number	Mean	Standard error of mean	t	p		
Dropodus langth	Dry	502	17.94	0.41	-0.72	0.47		
Propodus length	Wet	199	18.51	0.72	-0.72	0.4/		
Manus length	Dry	502	7.82	0.17	-0.81	0.42		
	Wet	199	8.08	0.29	-0.81	0.42		
Dollar langth	Dry	502	10.09	0.25	0.71	0.49		
Pollex length	Wet	199	10.43	0.44	-0.71	0.48		
Dactyl length	Dry	502	11.8	0.29	0.76	0.45		
	Wet	199	12.23	0.51	-0.76	0.43		

In this plot, data points were divided by season: dry (blue circles) and wet (orange triangles). The distribution of data points showed a clearer separation between the dry and wet seasons. The points for the wet season tend to cluster on one side, while the dry season points were distributed on the opposite side. Temperature (Tem) and salinity (Sal) vectors suggested a stronger influence during the dry season, while pH appears to had a more significant effect during the wet season. The orientation of the environmental factor vectors indicated seasonal differences, suggesting that changing environmental conditions between seasons affect the crab measurements. Season had a marked impact on the distribution of crab measurements. The dry and wet seasons display distinct environmental effects, particularly noticeable in temperature and salinity changes.



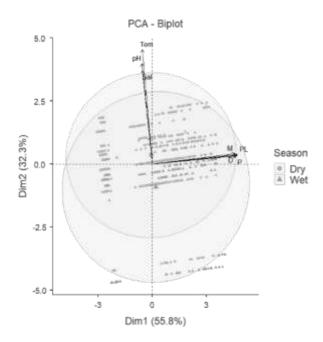


Figure 5. PCA illustrates relationship between some claw morphometrics and environmental factors of *Tubuca rhizophorae* species according to season (morphological measurements: PL: propodus length; M: manus length; P: pollex length; D: dactyl length; Tem: temperature; Sal: salinity)

#### **Meristics' Variation**

The study used the ratio of the manus length, pollex length, and dactyl length sizes to the propodus length. Based on the statistical results, the values of M/PL, P/PL, and D/PL at DDCM and DHBL for the species do not vary according to the research sites (see Table IV) and the season (see Table V).

Table IV. Meristics' variation in Tubuca rhizophorae regarding sampling site

Table 17. Wellsties variation in Tubuca Phizophorae regarding sampling site							
Claw morphometric parameters	Site	Number	Mean	Standard error of mean	t	p	
Manus length / Propodus	DDCM	438	44.57	0.22	-0.33	0.74	
length	DHBL	263	44.45	0.29	-0.55	0.74	
Pollex length / Propodus	DDCM	438	55.29	0.22	0.72	0.47	
length	DHBL	263	55.55	0.29	0.72	0.47	
Dactyl length / Propodus	DDCM	438	64.82	0.37	0.94	0.40	
length	DHBL	263	65.33	0.47	-0.84	0.40	

Note: DDCM: Dam Doi, Ca Mau; DHBL: Dong Hai, Bac Lieu

Table V. Meristics' variation in Tubuca rhizophorae regarding sampling site

Claw morphometric parameters	Site	Number	Mean	Standard error of mean	t	p
Manus length / Propodus	Dry	502	44.37	0.21	-1.36	0.18
length	Wet	199	44.90	0.34	-1.50	0.10
Pollex length / Propodus	Dry	502	55.51	0.21	1.04	0.29
length	Wet	199	55.10	0.34	1.04	0.29
Dactyl length / Propodus	Dry	502	65.08	0.34	0.26	0.72
length	Wet	199	64.85	0.55	0.36	0.72

Across all three plots, temperature (Tem), salinity (Sal), and pH consistently emerge as key environmental factors. However, their relative influence varied depending on the grouping variable (gender, season, or site). Notably, season and site had more pronounced effects on the separation of



data points compared to gender, suggesting that environmental conditions linked to seasonal changes and geographical differences were more impactful.

The plots for season and site exhibited clearer separations, while the plot for gender showed more overlapping clusters. This implied that seasonality and sites were stronger determinants of variance in crab measurements compared to gender. This pattern suggested that changes in environmental conditions across time (seasonal variation) and space (location differences) drive distinct changes in the measured variables, while gender had a more minimal impact.

The environmental factor vectors (Tem, Sal, pH) generally maintain similar orientations across the three plots. This indicated that, despite the different groupings, the main components (Dim1 and Dim2) consistently capture the primary directions of environmental impact on the measurements. This consistency of vector orientation implies that the primary dimensions (Dim1 and Dim2) effectively capture the variation caused by environmental factors, regardless of the grouping factor.

Temperature, salinity, and pH consistently impact the measurements, but their relative influence shifts based on the seasonal or locational context. Gender was not significantly impact the data distribution, indicating that crabs of different genders respond similarly to environmental conditions within the dimensions captured by PCA.

In summary, these PCA analyses suggested that environmental factors linked to season and site play a dominant role in shaping crab measurements, while gender had minimal influence. The data highlight that temperature, salinity, and pH were influential factors, but their effects were context-dependent, varying with both place (site – see Fig. 6a) and time (season – see Fig. 6b).

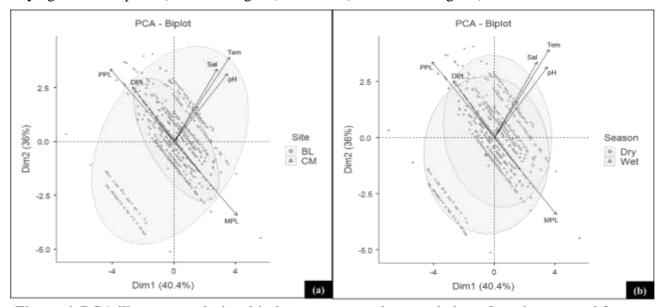


Figure 6. PCA illustrates relationship between some claw meristic and environmental factors of *Tubuca rhizophorae species* (morphological measurements: PL: propodus length; M: manus length; P: pollex length; D: dactyl length; Tem: temperature; Sal: salinity/a – according to site; b – according to season)

The values of M/PL and P/PL showed no variation when considered by sex (male and female). However, D/PL varied by sex (see Table VI and Fig. 7).



Table VI. Meristics' variation in *Tubuca rhizophorae* regarding sex

Claw morphometric parameters	Sex	Number	Mean	SE	t	p
Manus length / Propodus length	Male	296	44.44	0.27	2.64	0.009
	Female	111	44.78	0.49	2.64	
Pollex length / Propodus length	Male	296	55.56	0.27	-2.42	0.016
	Female	111	55.22	0.49		
Dactyl length / Propodus length	Male	296	62.20	0.50	2 66	<0.001
	Female	111	63.30	0.86	-3.66	< 0.001

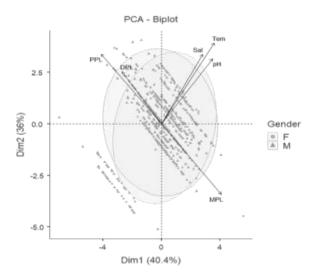


Figure 7. PCA illustrates relationship between some claw meristic and environmental factors of *Tubuca rhizophorae* species according to sex (morphological measurements: PL: propodus length; M: manus length; P: pollex length; D: dactyl length; Tem: temperature; Sal: salinity)

Temperature (Tem): The "Tem" vector was again relatively prominent, but its orientation was not as strongly aligned with Dim1 or Dim2 as in the first plot. This indicated that temperature still plays a role but may interact with other unaccounted-for factors or dimensions in this dataset. Salinity (Sal) and pH: These vectors were slightly more prominent than in the first plot. While they were still shorter than the "Tem" vector, their influence may be more balanced with temperature in this dataset, suggested a potential interaction effect among environmental factors. Temperature, salinity, and pH showed a more balanced influence on the measurement indices of crabs in this dataset. The temperature effect was still present but was less dominant. Weaker gender differentiation compared to the first plot. The male and female groups had more overlap, suggesting that environmental impacts may be more uniform across genders or that additional factors were in play.

#### 4. Discussion

In research conducted by Pramithasari, Butet [18] on the species *Albunea symmysta*, the findings showed that the Aceh population had a longer dactylus length than the Bengkulu population, with notable differences in propodus length. Similarly, a study by Thurman, Alber [19] on the species *Minuca burgersi* revealed morphological variations between populations from the Caribbean Sea and the Atlantic coast of South America. Several researchers suggest that differences in morphometric traits of a species can be attributed to various factors, including geographical region [20], with influences such as elevation and latitude. Environmental conditions also play a significant role, as noted [13, 21, 22], who demonstrated that the morphological differences observed in the pile ark cockle (*Anadara pilula*) were due to environmental factors. These findings support Barría, Sepúlveda [23], who hypothesized that adaptive responses to environmental conditions lead to morphological variations. Overall, the study suggests that each variable contributed individually but insignificantly to the main components. According to Thurman, Alber [19], important factors such as habitat,



substrate type, and salinity significantly influence specific morphological traits.

The study results further indicated that male individuals had larger claw and walking leg characteristics than females, suggesting that the traits of PL, M, P, D, and L varied by sex. Araújo, Coelho [24] also found that adult males tend to be larger than females in mangrove forests, likely because males focus more on body growth, while females allocate more energy to reproduction. Therefore, it can be inferred that morphological ratios between sexes differ, and the study by Thurman, Alber [19] on *Minuca burgersi* also noted differences between male and female individuals in terms of their morphological ratios.

#### 5. Conclusion

Tubuca rhizophorae was not display significant differences in the claw and walking leg morphological indices propodus length (PL), manus length (M), pollex length (P), dactyl length (D), between the two sampling sites and the season. However, there were significant variations in these traits based on sex, with males having higher PL, M, P, D. Analysis of the morphological ratios (M/PL, P/PL, D/PL) found no variations by sampling site, season or sex. However, D/PL varied by sex. The principal component analysis revealed that environmental factors such as pH, temperature, and salinity contribute to variations in some morphological traits, with temperature and salinity show the most significant multivariate correlation based on the three principal components studied. The study highlights how the morphological characteristics of *Tubuca rhizophorae* can vary according to site and sex, providing information on this species of fiddler crabs in the Mekong Delta region. Understanding morphological variations is vital to address taxonomic and ecological issues related to this species.

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