The Use of Morphological Measurement Relationships on Sex Differentiation for *Polynemus melanochir* in Cai Rang - Can Tho and Long Phu - Soc Trang

Lam Thi Thao Vo[1], Ton Huu Duc Nguyen[2], Anh Ngoc Tran[3], Quang Minh Dinh[4]*

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

KEYWORDS
Gender Differentiation, Mekong Delta, Morphology, *Polynemus Melanochir*

ABSTRACT
This research examines sex differentiation in Polynemus melanochir through morphometric analyses conducted across two sites in the Mekong Delta: Cai Rang and Long Phu. Over ten months, 520 samples were analyzed for standard length, head length, and body height metrics. Findings indicated that males generally maintain positive or stable relationships between total length and body measurements, while females often exhibit negative growth trends. Seasonal and locational effects were also noted, with the wet season favoring growth. These non-invasive morphometric techniques offer a practical approach to sex differentiation, aiding conservation efforts and enhancing understanding of Polynemus melanochir biology in varying environmental contexts.

1. Introduction:

Polynemus melanochir is one species of the Polynemidae family that is of commercial importance in the Mekong Delta. The Polynemidae family has nine genera with 42 species [1]. According to Rainboth [2], the Polynemidae family has two freshwater genera, *Eleuthronema* (2 species) and *Polynemus* (4 species). In Vietnam, the threadfin family Polynemidae belongs to Perciformes, and the genus *Polynemus* has seven species [3]. In the Mekong Delta, according to Mai et al. [4], the genus *Polynemus* has two species, *Polynemus melanochir* and *Polynemus dubius*. They are primarily distributed in brackish and saline waters of tropical and subtropical regions [5], typically concentrated in estuaries and mainly living in freshwater as bottom-dwelling species, primarily inhabiting muddy or sandy-muddy bottoms, and adapting well to environmental conditions, with suitable temperatures ranging from 30°N to 30°S [6].

To 2024, several studies on *Polynemus melanochir* have been carried out, typical publications by Motomura and colleagues, which discussed the distribution, feeding habits, and migratory behavior of this species, but only briefly [1; 7; 8]. Mustafa et al. [9] provided some biometric indices and population parameters of three polynemid fish species from the Batang Lassa Estuary in East Malaysia, including *Polynemus melanochir*. Vu et al. [10] also published the migratory behavior of several fish species (including *Polynemus melanochir*) in the Mekong Delta. Research on the gene pool of this fish population in the Mekong Delta was conducted by the group of Dang et al. [11].

The fish sex ratio is affected by several variables, including salinity and habitat temperature, according to Vu [12]. This ratio also varies by species, according to Tran et al. [13], depending on their developmental stage and reproductive habits. Fish sex discrimination is a significant topic in the biology and ecology of fish. In the *Periophthalmodon septemradiatus* species, males will have more colorful and longer dorsal fins than females, making it simple to discern between the sexes based on exterior morphological color [14]. However, the secondary genital spine characteristics of several species of the Bagridae family, including the striped pincer *Mystus mysticetus* [15], are used to determine the animal's sex. Females have pink, oval-shaped missing genitals, while males have long genitalia and pointed genital spines that are darker red than females. However, the genital spines of this fish are tiny, so it is difficult to see with the naked eye. Therefore, this study aimed to provide a new way of sex determination in this fish without killing fish by measuring and determining the relationship of morphological features between them. In the world and Vietnam, there have been several studies to implement this method in many different fish species, such as *Zacco koreanus* [16], *Heterotis niloticus* [17], *Glossogobius sparsipapillus* [18], *Glossogobius giuris* [19], *Ellochelon vaigiensis* [20], *Mystus mysticetus* [21], and *Caragobius urolepis* [22]. These studies show that this method can be applied to determine the sex of *Polynemus melanochir*. Limiting the killing of fish when conducting research contributes to better protection of this fish species. Besides, this study supports the validity of this method for determining sex and lays the groundwork for future investigations into the biology of this fish.

2. Materials And Methods:

Sample collection and analysis

During a period of 10 months from January to October 2024, 520 samples (250 females and 270 males) of *Polynemus melanochir* fish were collected periodically once a month at two sites, including Cai Rang - Can Tho (CRCT) and Long Phu - Soc Trang (LPST) (Figure 1).

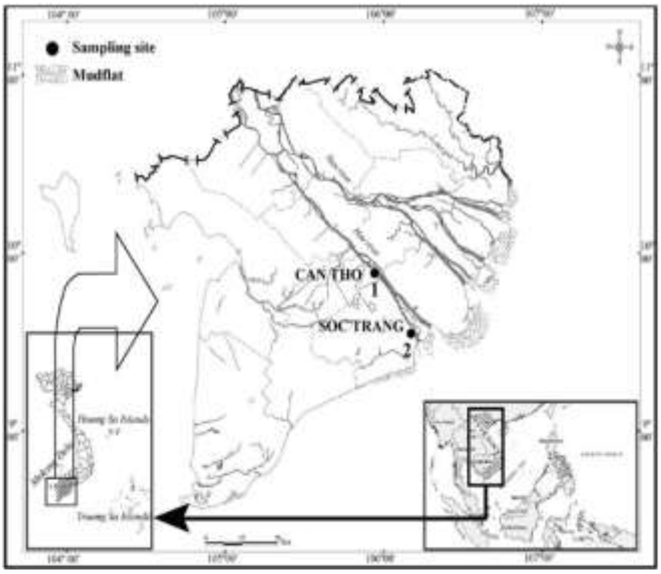

Figure 1. Map of sampling sites (●: Sampling sites; 1: Cai Rang - Can Tho; 2: Long Phu - Soc Trang; this figure was modified from Dinh [23])

Fish samples were collected continuously for 48 hours using fishermen's 1.5 cm trawl nets. Individuals were randomly collected with various sizes. Before being transported to the Animal Laboratory, Department of Biology Teacher Education, Can Tho University, the samples were stored in a 4% formalin solution. Then, fish samples were identified based on the description of Tran et al. (2013). The morphometric features include total length (TL), standard length (SL), eye diameter (ED), eye distance (DE), body height (BH), head length (HL), and mouth diameter (MD) (Figure 2).

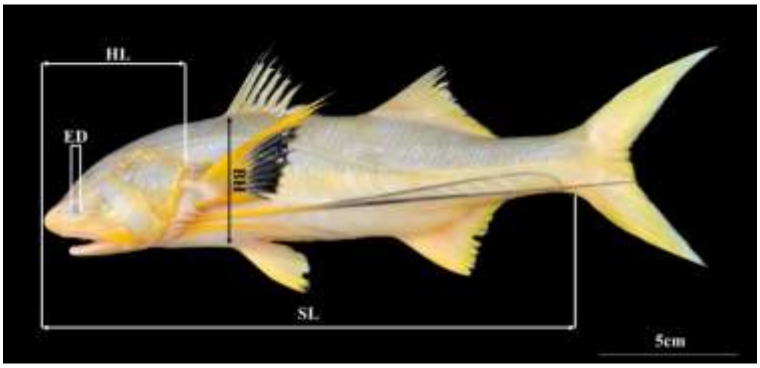

Figure 2. Measuring parameters of *Polynemus melanochir* (TL: total length; SL: standard length; ED: eye diameter; DE: eye distance; BH: body height; HL: head length; Source: Nguyen & Dinh [24]).

Statistical analysis

According to Minos et al. [25], the relationship between total length (TL) and other measurement parameters of fish was determined based on the formula Y=*a* x TL^b. Y was the morphological size (SL, ED, DE, BH, HL, and MD), and *a* and *b* were the regression coefficients (a was the intercept, and b was the slope). Morphological variables were classified into positive growth (A+) if *b* >1, negative growth (A-) if *b* <1, and isometry (I) if *b* ≈1. This test also confirmed if *b* varied by sex, season, and sampling location. The data in this study were analyzed using SPSS v21 with a significance level of 5%.

3. Results And Discussion:

With females, most indicators such as SL(standard length), ED(eye diameter), DE, BH, HL(head length), and MD have *b* values ranging from 0.28 to 0.50. All indicators showed a negative growth pattern (A-), meaning they grew slower than TL. For example, SL had a highly negative ts value (-7.97), suggesting a significant decline in standard length as TL increases (Table 1). This result may be due to physiological and structural differences, where female bodies showed a trend of decreasing indicator values as TL increases, possibly because of inherent biological constraints.

With males, indicators such as SL had *b* values around 1.00, indicating *a* strong relationship between standard length and TL in males. Some indicators like BH (body height) and HL (head length) exhibit positive (A+) or isometry (I) growth patterns, with values greater than one and P < 0.05, meaning these indicators increase as TL increases (Table 1). The stability in male indicators compared to females may be due to differences in body structure, as male bodies display a more synchronous increase in indicators like standard length and head length as TL rises.

General pattern by gender. Males showed a strong positive relationship between TL and indicators like SL, HL, and BH, while females tend to see a decline in indicators as TL increases. The data reveal marked differences in morphometric growth between male and female *Polynemus melanochir*. Males exhibited stable or positive growth relationships with total length in traits such as standard length and body height, consistent with observed patterns in other species like *Ellochelon vaigiensis* [20], where males tend to support traits suited to reproductive roles and competitive behaviors. Females, in contrast, often displayed decreasing trait values with increasing total length, reflecting potentially different adaptive pressures that prioritize energy allocation for reproductive output.

Table 1. Regression relationship of the total length (TL) to standard lengths (SL), eye diameter (ED), eye distance (DE), body height (BH), head length (HL), and mouth diameter (MD) of *Polynemus melanochir* regarding sex

Index	*b*	SE	*a*	SE	n	Growth pattern	t_s	P
Female								
SL	0.41	0.03	4.46	0.43	250	A-	-17.97	0.00
ED	0.40	0.05	4.18	0.62	250	A-	-11.76	0.00
DE	0.28	0.09	1.60	0.41	250	A-	-8.17	0.00
BH	0.38	0.09	3.33	0.89	250	A-	-6.75	0.00
HL	0.50	0.09	6.34	1.71	250	A-	-5.38	0.00
MD	0.36	0.14	11.03	4.54	250	A-	-4.53	0.00
Male								
SL	1.00	0.03	0.80	0.06	270	I	-0.15	0.88
ED	1.04	0.05	0.64	0.09	270	I	0.86	0.39
DE	1.19	0.14	0.12	0.05	270	I	1.37	0.17
BH	1.64	0.17	0.10	0.05	270	A+	3.84	0.00
HL	1.19	0.05	0.85	0.12	270	A+	4.00	0.00
MD	0.73	0.12	3.35	1.16	270	A-	-2.21	0.03

Furthermore, the analysis of the relationship between seasonal morphology indicators (dry season and wet season) showed that *Polynemus melanochir* had significant differences, as shown in Table 2.

In the dry season, most indicators had *b* values below 0.50, indicating that in the dry season, indicators showed a minimal increase as TL changes. All indicators exhibited a negative growth pattern (A-) with negative ts values and $P < 0.05$, indicating a negative relationship where indicators decrease as TL increases. For example, SL sharply decreases in the dry season with $t_s = -18.48$, indicating that the conditions might negatively impact body growth. This could be due to less favorable environmental conditions (such as limited food and water), which lead to reduced body size or hindered growth as TL increases.

In the wet season, indicators like SL, ED, BH, and HL had higher *b* values (greater than 1), indicating a strong positive relationship with TL. These indicators showed a positive (A+) or stable (I) growth pattern, suggesting that under wet season conditions, indicators increase or remain stable as TL rises. This could be because favorable wet season conditions (abundant food resources) support the growth of indicators as TL increases.

General pattern by season: During the wet season, indicators tend to increase significantly and positively as TL changes, highlighting the importance of seasonal factors on body growth. Seasonal change also influenced these morphometric patterns. During the dry season, most growth indicators showed a negative trend, possibly due to limited resources, whereas in the wet season, fish displayed increased values, particularly in head length and body height. This growth contrast across seasons aligns with studies on species such as *Glossogobius sparsipapillus* [26; 27], which also experience enhanced growth in more resource-abundant conditions.

Table 2. Regression relationship of the total length (TL) to standard lengths (SL), eye diameter (ED), eye distance (DE), body height (BH), head length (HL), and mouth diameter (MD) of *Polynemus melanochir* regarding season

Index	*b*	SE	*a*	SE	n	Growth pattern	t_s	P
Dry season								
SL	0.39	0.03	4.68	0.45	331	A-	-18.48	0.00
ED	0.42	0.06	3.97	0.66	331	A-	-10.25	0.00
DE	0.29	0.11	1.76	0.55	331	A-	-6.57	0.00
BH	0.40	0.13	3.61	1.38	331	A-	-4.55	0.00
HL	0.50	0.08	6.44	1.57	331	A-	-5.96	0.00
MD	0.35	0.15	10.25	4.37	331	A-	-4.42	0.00
Wet season								
SL	1.02	0.01	0.74	0.02	189	A+	3.29	0.00
ED	1.01	0.01	0.70	0.03	189	I	1.00	0.32
DE	0.85	0.06	0.25	0.04	189	A-	-2.76	0.01
BH	1.31	0.06	0.21	0.04	189	A+	4.80	0.00
HL	1.14	0.04	0.95	0.12	189	A+	3.27	0.00
MD	0.91	0.04	2.29	0.24	189	A-	-2.49	0.01

The regression relationship of morphological values varied not only by sex and season but also by sampling site (Table 3). The analysis of morphological parameters in the Cai Rang, Can Tho area showed that indicators such as SL and ED had *b* values close to 1, indicating a stable relationship between these indicators and TL. Other indicators like DE and BH showed a negative growth pattern (A-) with low correlation coefficients, suggesting that these indicators decrease slightly as TL increases. This may be due to the environmental conditions in Cai Rang, which support stable growth in key indicators, though some indicators may be slightly impacted by local ecology. In addition, at Long Phu, Soc Trang, All indicators had *b* values below 0.52, reflecting a weak relationship between indicators and TL at this site. All indicators display a negative growth pattern (A-) and a substantial decrease as TL rises, as SL with t_s = -19.10 showed a significant reduction as TL increases. This could be attributed to less favorable environmental conditions in Long Phu, leading to a more pronounced indicator decrease as TL grows.

Table 3 Regression relationship of the total length (TL) to standard lengths (SL), eye diameter (ED), eye distance (DE), body height (BH), head length (HL), and mouth diameter (MD) of *Polynemus melanochir* regarding sampling sites

Index	*b*	SE	*a*	SE	n	Growth pattern	t_s	P
Cai Rang, Can Tho								
SL	1.02	0.04	0.73	0.08	215	I	0.65	0.52
ED	0.96	0.07	0.81	0.18	215	I	-0.49	0.62
DE	0.09	0.22	3.26	2.09	215	A-	-4.19	0.00
BH	0.47	0.24	3.36	2.36	215	A-	-2.22	0.03
HL	0.92	0.05	1.96	0.28	215	I	-1.55	0.12
MD	1.23	0.19	0.73	0.40	215	I	1.20	0.23
Long Phu, Soc Trang								
SL	0.43	0.03	4.04	0.35	305	A-	-19.10	0.00
ED	0.44	0.05	3.56	0.47	305	A-	-12.26	0.00
DE	0.51	0.05	0.74	0.11	305	A-	-9.61	0.00
BH	0.49	0.06	2.16	0.36	305	A-	-8.84	0.00
HL	0.52	0.09	5.53	1.34	305	A-	-5.67	0.00
MD	0.47	0.11	8.06	2.61	305	A-	-4.63	0.00

General pattern by sampling sites: In Cai Rang, key indicators tend to maintain a more stable relationship with TL, while in Long Phu, indicators showed a marked decrease as TL increases, possibly due to environmental and ecological factors unique to each site. Comparisons of sites further illustrated environmental impact, with fish in Cai Rang maintaining stable trait relationships while those in Long Phu exhibited reduced growth, potentially reflecting differences in habitat quality or resource availability. This geographic influence was similar to *Heterotis niloticus* patterns, suggesting habitat variability as a critical factor affecting morphometrics in the Mekong Delta. These findings confirm the applicability of morphometric measurements for non-lethal sex identification, supporting efforts in species conservation and sustainable management.

4. Conclusion

The results of the analysis of morphological parameters of *Polynemus melanochir* played a role in sex determination. By gender, males tend to maintain or increase indicator values as TL rises, whereas females generally show decreased indicators, highlighting physiological differences. By season, the wet season promotes a more positive and significant increase in indicators compared to the dry season, reflecting seasonal environmental impacts on growth. By sampling sites, key indicators remain more stable in Cai Rang, Can Tho, whereas in Long Phu, Soc Trang, they showed a notable decrease with TL growth, likely due to local environmental conditions. These results indicate that the growth of body indicators is highly influenced by gender, seasonal conditions, and sites, showing a complex relationship between TL and other biological indicators.

Acknowledgements

Lam Thi Thao Vo was funded by the Master, PhD Scholarship Programme of Vingroup Innovation Foundation (VINIF), code VINIF.ThS.2023.ThS.071.

Reference

[1] Motomura H. 2004. *Threadfins of the world (Family Polynemidae): An annotated and illustrated catalogue of polynemid species known to date*, Food & Agriculture Org.
[2] Rainboth W. J. 1996. *Fishes of the Cambodian Mekong*, Roma, FAO.
[3] Nguyen V. H. 2005. [*Freshwater fish of Viet Nam*], Vol. III, Ha Noi, Agriculture Publishing House.

[4] Mai Y. D., Nguyen T. V., Nguyen T. V., Le Y. H.,. Hua L. B, 1992. [*Identification of freshwater fishes of South Vietnam*], Ha Noi, Science and Technology Publishing House.
[5] Nelson J., Grande T., Wilson M., 2016. *Fishes of the world*, New York, United States, John Wiley & Sons.
[6] Truong T. K., Tran T. T .H., 1993. *Identification of freshwater fish in Mekong Delta*, Can Tho University, Can Tho University.
[7] Motomura H., Sabaj M. H., 2002. A new subspecies, *Polynemus melanochir dulcis*, from Tonle Sap Lake, Cambodia, and redescription of *P. m. melanochir* Valenciennes in Cuvier and Valenciennes, 1831 with designation of a neotype. *Ichthyological Research*, 49, 181-190.
[8] Motomura H., Mikschi E., Iwatsuki Y., 2001. Galeoides Günther, 1860, a monotypic genus of the family Polynemidae (Perciformes). *Cybium*, 25 (3), 269-272.
[9] Mustafa M. G., Rajaee A. H., Hamli H., Rahim K. A. A., 2021. Biometric indices and population parameters of three polynemid fishes from Batang Lassa Estuary of East Malaysia. *PeerJ*, 9, e12183.
[10] Vu A. V., Baumgartner L. J., Mallen-Cooper M., Doran G. S., Limburg K. E., Gillanders B. M., Thiem J. D., Howitt J. A., Kewish C. M., Reinhardt J., 2022. Diverse migration tactics of fishes within the large tropical Mekong River system. *Fisheries Management and Ecology*, 29 (5), 708-723.
[11] Dang B., Vu Q., Biesack E., Doan T., Truong O., Tran T., Ackiss A., Stockwell B., Carpenter K., 2019. Population genomics of the peripheral freshwater fish *Polynemus melanochir* (Perciformes, Polynemidae) in a changing Mekong Delta. *Conservation Genetics*, 20, 961-972.
[12] Vu T. T., 2000. Ecological foundations
[13] Tran D. D., Shibukawa K., Nguyen T. P., Ha P. H., Tran X. L., Mai V. H., Utsugi K., 2013. *Fishes of Mekong Delta*, Vietnam, Can Tho, Can Tho University Publisher.
[14] Dinh Q. M., Tran L. T., Nguyen N. T. Y., 2018. The flexibility of morphometric and meristic measurements of *Periophthalmodon septemradiatus* (Hamilton, 1822) in Hau river. *Journal of Science and Technology* 187 (11), 81-90.
[15] Nguyen T. N. T., Duong T. Y., 2020. Comparing morphological characteristics of striped dwarf catfish (*Mystus mysticetus*) distributed in U Minh Thuong with other populations in the Mekong Delta. *Can Tho University Journal of Science*, 56 (1), 192-199.
[16] Kim Y. J., Zhang C. I., Park I. S., Na J. H., Olin P., 2008. Sexual dimorphism in morphometric characteristics of Korean chub *Zacco koreanus* (Pisces, Cyprinidae). *Journal of Ecology and Environment*, 31 (2), 107-113.
[17] Obi A., 2010. Sex determination in *Heterotis niloticus* (Cuvier 1829) based on morphometric features. *SSET: An International Journal (Series B)*, 6 (1), 22-30.
[18] Dinh Q. M., Nguyen T. H. D., Nguyen T. T. K., 2021. Allometry variation in morphometrics of *Glossogobius sparsipapillus* caught along Hau river, from Can Tho to Soc Trang provinces. *TNU Journal of Science and Technology*, 226 (05), 3-7.
[19] Dinh Q. M., Nguyen T. H. D., Phan G. H., Nguyen V. L. T., 2021. The morphological variation and sex determination of tank goby *Glossogobius giuris* distributing in some provinces in Mekong Delta. *Science and Technology Journal of Agriculture & Rural Development*, 10, 141-145.
[20] Nguyen D. L., Nguyen T. H. D., Dinh Q. M., 2022. Sex discrimination based on morphological traits in squaretail mullet *Ellochelon vaigiensis* (Quoy & Gaimard, 1825). *TNU Journal of Science and Technology*, 227 (5), 132-136.
[21] Phan T. Q., Tran A. N., Vo L. T. T., Dinh Q. M., 2022. Sex discrimination based on morphological traits in *Mystus mysticetus* Roberts, 1992. *Veterinary Integrative Sciences*, 20 (3), 659-667.
[22] Nguyen P. L. H., Tran L. T. C., Phan T. T. A., Nguyen T. K., Vo L. T. T., Dinh Q. M., 2023. Morphometrics allometry changes and sexual dimorphism in *Caragobius urolepis* (Gobiiformes: Gobiidae). *Veterinary Integrative Sciences*, 21 (1), 29-36.
[23] Dinh Q. M., 2018. Aspects of reproductive biology of the red goby *Trypauchen vagina* (Gobiidae) from the Mekong Delta. Journal of *Applied Ichthyology*, 34 (1), 103-110.
[24] Nguyen T. H. D., Dinh Q. M., 2023. Morphometric and meristic variations of *Mystus albolineatus Roberts*, 1994 in the Mekong Delta, Vietnam. *Veterinary Integrative Sciences*, 21 (3), 705-716.
[25] Minos G., Kokokiris L., Kentouri M., 2008. Allometry of external morphology and sexual dimorphism in the red porgy (*Pagrus pagrus*). *Belgian Journal of Zoology*, 138 (1), 90-94.
[26] Nguyen T. H. D., Dinh Q. M., 2020. Otolith dimensions and their relationship with the size of *Glossogobius sparsipapillus* fish along the coastline of Mekong Delta, Vietnam. *Egyptian Journal of Aquatic Biology and Fisheries*, 24 (2), 525-533.
[27] Truong N. T., Phan G. H., Dinh Q. M., Nguyen T. H. D., Nguyen T. T. K., 2021. Growth and condition factor of the commercial goby *Glossogobius sparsipapillus* living along Bassac River, Vietnam. *Aquaculture, Aquarium, Conservation & Legislation*, 14 (3), 1695-1701.