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Biomass Estimation Using Length-Weight Regression for the Freshwater Cyclopoida

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Abstract Zooplankton biomass is essential for understanding the quantitative structure of lake food webs and for the functional assessment of biotic interactions. In this study, we aimed to propose a biomass (dry weight) estimation method using the body length of cyclopoid copepods. These copepods play an important role as omnivores in lake zooplankton communities and contribute significantly to biomass. We validated several previously proposed estimation equations against direct measurements and compared the suitability of prosomal length versus total length of copepods to suggest a more appropriate estimation equation. After comparing the regression analysis results of various candidate equations with the actual values measured on a microbalance—using the coefficient of variation, mean absolute error, and coefficient of determination—it was determined that the Total Length-DW exponential regression equation [$W = 0.7775 \times e^{2.0183L}$; W (μg), L (mm)] could be used to calculate biomass with higher accuracy. However, considering practical issues such as the morphological similarity between species and genera of copepods and the limitations of classifying copepodid stages, we derived a general regression equation for the pooled copepod community rather than a species-specific regression equation.

Key words: zooplankton, dry weight estimation, pooled cyclopoids, lake ecosystem assessment, food web analysis

INTRODUCTION

Biomass, which is closely related to an organism's body specification (e.g., length, width, area, etc.), reflects size differences among species and can therefore provide a

more objective assessment of the function of zooplankton in aquatic ecosystem food webs and facilitate relative comparisons among different water bodies (Kane *et al.*, 2009; Beaver *et al.*, 2020; Oh *et al.*, 2023). At the same time, zooplankton biomass is essential for quantifying secondary productivity, which is the intermediary between primary productivity by phytoplankton and the mass of higher trophic level organisms, and can be used to estimate the efficiency of energy transfer between low- and high-trophic level organisms, and to track material circulations and ener-

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gy flows within the food web (Ku *et al.*, 2022; Choi *et al.*, 2023a, b).

Cyclopoida demonstrate a wide feeding spectrum that ranges from omnivorous to carnivorous, feeding on phytoplankton, rotifers, and occasionally even large *Daphnia* in freshwater ecosystems (Chang and Hanazato, 2003, 2005; Chae *et al.*, 2023; Liu *et al.*, 2023). At the same time, they play an important role as intermediaries in aquatic ecosystem food chains, serving as important food sources for fishes (Chang *et al.*, 2001; La *et al.*, 2008). They can sometimes occur in high densities, which makes them numerically significant (Oh *et al.*, 2020; Hong *et al.*, 2023; Kalinowska *et al.*, 2024). Cyclopoid copepods are typically microscopic, ranging in size from 0.5 to 1.0 mm, although some can reach lengths of 2 mm (Genus *Megacyclops* and *Cyclops*). They exhibit a high degree of morphological similarity between species, more so than other zooplankton taxa.

For cyclopoid copepods, due to the consistency of shape between species within the same suborder, researchers have proposed equations to estimate the biomass of pooled copepods. Additionally, generic equations applicable to taxonomic groups—cyclopoids, calanoids, harpacticoids—have been developed (Burgis, 1975; Dumont *et al.*, 1975; Bottrell *et al.*, 1976) based on findings such as those reported by Dumont *et al.* (1975), which noted that littoral species weigh more than limnetic species. Until recently, research has focused on developing species-specific equations for more accurate biomass estimation of copepods. Moreover, the U.S. EPA has proposed not only species-specific equations but also those that are life stage-specific (USEPA, 2016).

In Korea, a total of 388 species of Cyclopoida, spread across 34 phyla and 149 genera, have been reported (National Institute of Biological Resources, <https://species.nibr.go.kr/>). Of these, 59 species across 18 genera are known to occur in freshwater. The national guidelines propose a universally applicable equation, except for certain cyclopoid genera—*Cyclops* spp., *Diacyclops* spp., *Eucyclops* spp., *Mesocyclops* spp., *Thermocyclops* spp., and *Tropocyclops* spp.—for which genus-specific length-weight estimation equations are proposed (NIER, 2017). However, none of these equations have been empirically tested on domestic species; they are all adapted from international studies that were published some time ago. Regrettably, the manual provides only the author's name and year of publication,

omitting the full citation; consequently, it is not feasible to ascertain the precise origin of the equations.

In this study, we evaluated the suitability of existing biomass estimation equations for individual cyclopoids in Korea. We conducted regression analyses on total body length and prosome length data from each individual to derive new regression equations. These new equations are characterized by enhanced statistical robustness and are applicable to pooled cyclopoid copepod communities.

MATERIALS AND METHODS

This study was conducted using individuals of various cyclopoid species collected from Lake Shingal (37°14'27.5"N 127°05'34.9"E), South Korea. Specimens were gathered in the field using a zooplankton net with a diameter of 22 cm and a mesh size of 90 µm. To gather a sufficient number of individuals for biomass estimation and measurement, the net was towed several times in an oblique direction. The cyclopoid species were then sorted from these samples in the laboratory using a light microscope (CKX41, Olympus, Japan) and a dissecting microscope (SZ, Olympus, Japan). For the review of biomass estimation methods applicable to pooled cyclopoid species, individuals were selected to cover a broad range of size information across different species. Selection of individuals for measurement encompassed all life stages of copepods, excluding nauplii due to their significant differences in shape and size.

Total length (mm) and prosome length (mm) of selected individuals were measured under an inverted microscope (CKX41, Olympus, Japan) (Fig. 1). The presence or absence of egg sacs can influence the weight variations among individuals of the same size. Therefore, the measurements of individuals with and without egg sacs were recorded separately. Length measurements were performed using a digital microscope camera (5.1 MP Aptina CMOS sensor, China) and an image viewing program (ImageView, Bestscope, China).

Dry weight measurements were conducted using tin capsules (Tin capsules pressed ultra-light weight, 6×4 mm; Elemental Microanalysis, UK) and a microbalance (MYA 2.4Y; RADWAG, Poland - limit of quantification 0.001 mg). For more accurate weight determination, clusters of individuals with the same total length were grouped and

placed in a single tin capsule, ensuring that the sum of the theoretical weights, based on the same body length, was approximately 5 to 10 μg or more, as observed in previous studies (Table 1).

Clusters of individuals were initially pre-dried in a 60°C oven and then placed into pre-weighed tin capsules. They were subsequently dried for an additional 24 hours under the same conditions and weighed again to ascertain the dry weight of the individuals. To ensure accurate calculation of dry weight, each tin capsule was weighed three times before and after drying, and the average of these measurements was utilized. The standard deviation of the three measurements was between 0.000 and 0.001 mg. The total weight

was divided by the combined number of individuals in each group (n) to calculate the average dry weight per individual, as follows:

$$\text{Measured dry weight } (\mu\text{g}) = \frac{(W_b)_{avg.} - (W_a)_{avg.}}{n} / 1000$$

Measured dry weight = Average dry weight of n individuals used in the weight measurement (μg)

W_a = Weight of tin capsule pre-weighed (mg)

W_b = Weight of tin capsule weighed after drying the individuals (mg)

$W_{avg.}$ = Weight average of 3 measurements

n = Number of individuals used for weight measurement

To validate the existing length-weight (body length-dry weight) regression equation for estimating the biomass of cyclopoid individuals without species distinction, we calculated the error between the biomass estimate derived from the equation and the actual measured value obtained using a microbalance.

Three equations were selected as comparison equations: Eq. (1) and Eq. (2), proposed by Dumont *et al.* (1975), are for biomass estimation of adult cyclopoid species—Eq. (1) includes ovigerous females, and Eq. (2) excludes them. Eq. (3), developed by Burgis (1975), is tailored for pooled stages of *Cyclops* spp., a representative cyclopoid species. Additionally, Eq. (4) by Bottrell *et al.* (1976), which includes ovigerous females, is suited for pooled copepod species and

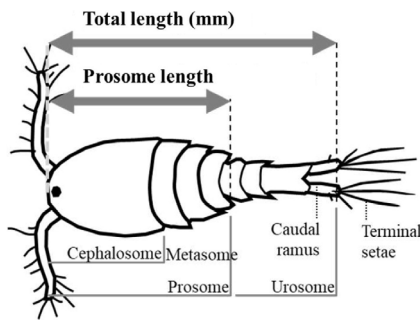


Fig. 1. Measurements of body specifications [Total length (mm) = prosome length + urosome length, Prosome length (mm) = cephalosome length + metasome length] of various cyclopoid species' individuals.

Table 1. Dry weight information by body length of cyclopoid copepods based on literature and number of individuals collected for weight measurement (No. of individuals per cluster) in this study.

| Length (mm) | Cyclopoida | | | |
|-------------|------------------------------|-------|-------------------|--------------------------------|
| | Dumont <i>et al.</i> , 1975 | | In this study | |
| | Dry weight (μg) | | Length range (mm) | No. of individuals per cluster |
| Copepodites | Adult | | | |
| 0.5 | 1.41 | – | 0.5~0.6 | 5 |
| 0.6 | 1.99 | – | 0.6~0.7 | 5 |
| 0.7 | 2.66 | – | 0.7~0.8 | 4 |
| 0.8 | 3.42 | 4.95 | 0.8~0.9 | 3 |
| 0.9 | 4.27 | 6.84 | 0.9~1.0 | 2 |
| 1.0 | 5.22 | 9.16 | | 1 |
| 1.1 | 6.24 | 11.88 | 1.0~1.2 | 1 |
| 1.2 | – | 15.13 | 1.2~1.4 | 1 |
| 1.4 | – | 23.14 | 1.4~1.6 | 1 |
| 1.6 | – | 33.45 | 1.6~ | 1 |

Table 2. Length-weight regression equations used to estimate the biomass (dry weight) of Cyclopoid copepods.

| Equation | Features | Reference |
|---|--|-------------------------------|
| (1) $W = 4.9 \times 10^{-8} \times L^{2.75}$ - W : μg , L : μm | Derived using adult cyclopoid species including females with eggs | Dumont <i>et al.</i> , 1975 |
| (2) $W = 1.1 \times 10^{-7} \times L^{2.59}$ - W : μg , L : μm | Derived using adult cyclopoid species excluding females with eggs | |
| (3) $W = 2.2 \times 10^{-8} \times L^{2.82}$ - W : μg , L : μm | Derived using pooled stages of <i>Cyclops</i> species (<i>C. vicinus</i> , <i>C. vernalis</i> , <i>C. viridis</i>); including females with/without eggs - excluding the furcal (caudal) rami | Burgis, 1975 |
| (4) $\ln(W) = 1.9526 + 2.3990 \times \ln(L)$ - W : μg , L : mm | Derived using pooled copepod species including females with eggs - total lengths including rami, but not terminal setae * Equation for unidentified Cyclopoida suggested by 『Biomonitoring Survey and Assessment Manual』 of Korea | Bottrell <i>et al.</i> , 1976 |

is documented in the Korean Biomonitoring Survey and Assessment Manual-Weir section monitoring (NIER, 2017) as a method for estimating the biomass of unidentified Cyclopoida (Table 2). These equations were used to evaluate their applicability to the domestic cyclopoid population, encompassing various genera and species.

Comparisons were made between dry weight estimates calculated using existing biomass estimation equations for various cyclopoid species and actual measurements obtained in this study. This was achieved through the application of the mean absolute error (MAE), which involves converting the sum of error values (\pm) to absolute values and then averaging them. Additionally, body measurements (total length and prosome length) along with dry weights from individuals of both species were used to develop optimal regression equations. These equations are designed to accurately estimate the biomass of each species.

We conducted regression analysis using average body measurements and dry weight data for individuals with similar body sizes. Both linear and non-linear regression analyses were carried out. To evaluate the goodness of fit for the regression models, we employed two metrics: the mean absolute error (MAE) and the coefficient of determination (multiple R-squared). Further, we implemented the K-means clustering algorithm to investigate discrepancies between the estimated and actual dry weights across various body size ranges. This analysis was performed using the RStudio packages ‘factoextra’ and ‘caret’ (Kuhn, 2008; Kassambara and Mundt, 2020). Based on these results, we proposed the optimal regression equation for estimating the biomass of a pooled sample of various genera and species

of cyclopoids in Korea.

RESULTS

1. Trends in size distribution of pooled cyclopoid species in this study

In this study, the total length of the pooled individuals from various cyclopoid species ($n = 143$) ranged from 0.510 to 1.777 mm, while their prosome length spanned from 0.331 to 1.055 mm. Analysis of frequency distributions revealed that total length did not exhibit significant variation across most size intervals, except for the ~ 1.6 mm interval. Conversely, prosome length displayed a prominently high frequency, approximately 42.7%, in the 0.4~0.5 mm and 0.8~0.9 mm intervals.

The analysis of the overall prosome length (PL) to total length (TL) distribution indicated a positive correlation, with an increase in PL typically accompanied by an increase in TL (Fig. 2). To investigate differences in PL among individuals with similar TL, we categorized TL into five 0.25 mm bins. Within each bin, we calculated the coefficient of variation (CV%) for both measurements. Our findings revealed no significant difference in the variability of TL and PL among individuals in bin 1 ($TL < 0.75$ mm)—a category likely containing a mix of copepodites and small adults. In contrast, for adult individuals, the variability of PL was found to be greater than that of TL (Fig. 2; Table 3). This trend is likely attributable to the fact that as copepodids develop into their adult forms, morphological differences between species (or genera) become more pronounced. As a result, the observed

increase in variability of PL relative to TL in adult specimens reflects these noticeable morphological distinctions.

Cyclopoid individuals with similar body lengths, irrespective of their genus and species, were weighed in clusters of one, two, or five individuals, depending on their total length (TL). The representative body measurements for these clusters are documented in Appendix 1. To ensure that these weights were representative of the individual body

measurements, the variability in body measurements among individuals within these clusters was analyzed. No clusters were found to have a coefficient of variation (CV) exceeding 10%, indicating a high level of consistency in body size within each cluster (Appendix 1).

2. Examining existing biomass estimation formulas through biomass measurements of pooled cyclopoid species

To assess the suitability of existing biomass estimation equations for species in the domestic Cyclopoida family, Fig. 3 illustrates the error between estimated values and actual measurements. The total length (prosome length + urosome length) of the collected individuals was used as the L value in each equation. When comparing the Mean Absolute Errors (MAEs) across all intervals, the equations ranked from lowest to highest error were: Eq. (2) < Eq. (3) < Eq. (4) < Eq. (1), as shown in Table 4.

Since this study included females with egg-sacs, we anticipated the smallest estimate-observation error when applying Dumont *et al.*'s (1975) Eq. (1), designed specifically for adult cyclopoid species, including females with eggs. However, it was found to have the largest overall error, displaying low MAE values only for smaller individuals. Conversely, Burgis' (1975) Eq. (3) was derived using body length measurements

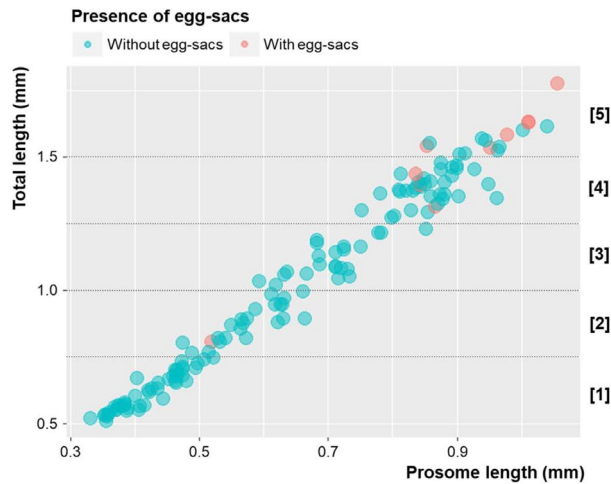


Fig. 2. Trends in total length (y-axis, in mm) as a function of individual prosome length of pooled cyclopoid species (x-axis, in mm). Color points indicate reproductive status: blue for individuals without egg-sacs and red for individuals with egg-sacs.

Table 3. Summary of body specifications (Total length, and Prosome length) by total length interval in Fig. 2.

| The Section in Fig. 2 | Total length (mm) | | Prosome length (mm) | |
|-----------------------|------------------------------------|-------|------------------------------------|-------|
| | Avg. \pm Std. (Min~Max) | CV% | Avg. \pm Std. (Min~Max) | CV% |
| [1] n=47 | 0.620 \pm 0.070 (0.510~0.748) | 11.31 | 0.421 \pm 0.049 (0.331~0.522) | 11.71 |
| [2] n=23 | 0.878 \pm 0.067 (0.764~0.997) | 7.62 | 0.577 \pm 0.052 (0.474~0.664) | 9.08 |
| [3] n=22 | 1.117 \pm 0.062 (1.022~1.230) | 5.58 | 0.706 \pm 0.057 (0.593~0.851) | 8.03 |
| [4] n=36 | 1.382 \pm 0.055 (1.272~1.478) | 3.98 | 0.856 \pm 0.044 (0.751~0.962) | 5.12 |
| [5] n=15 | 1.579 \pm 0.066 (1.510~1.777) | 4.17 | 0.959 \pm 0.058 (0.853~1.055) | 6.06 |
| Total n=143 | 1.030 \pm 0.356 (0.510~1.777) | 34.59 | 0.656 \pm 0.204 (0.331~1.055) | 31.15 |

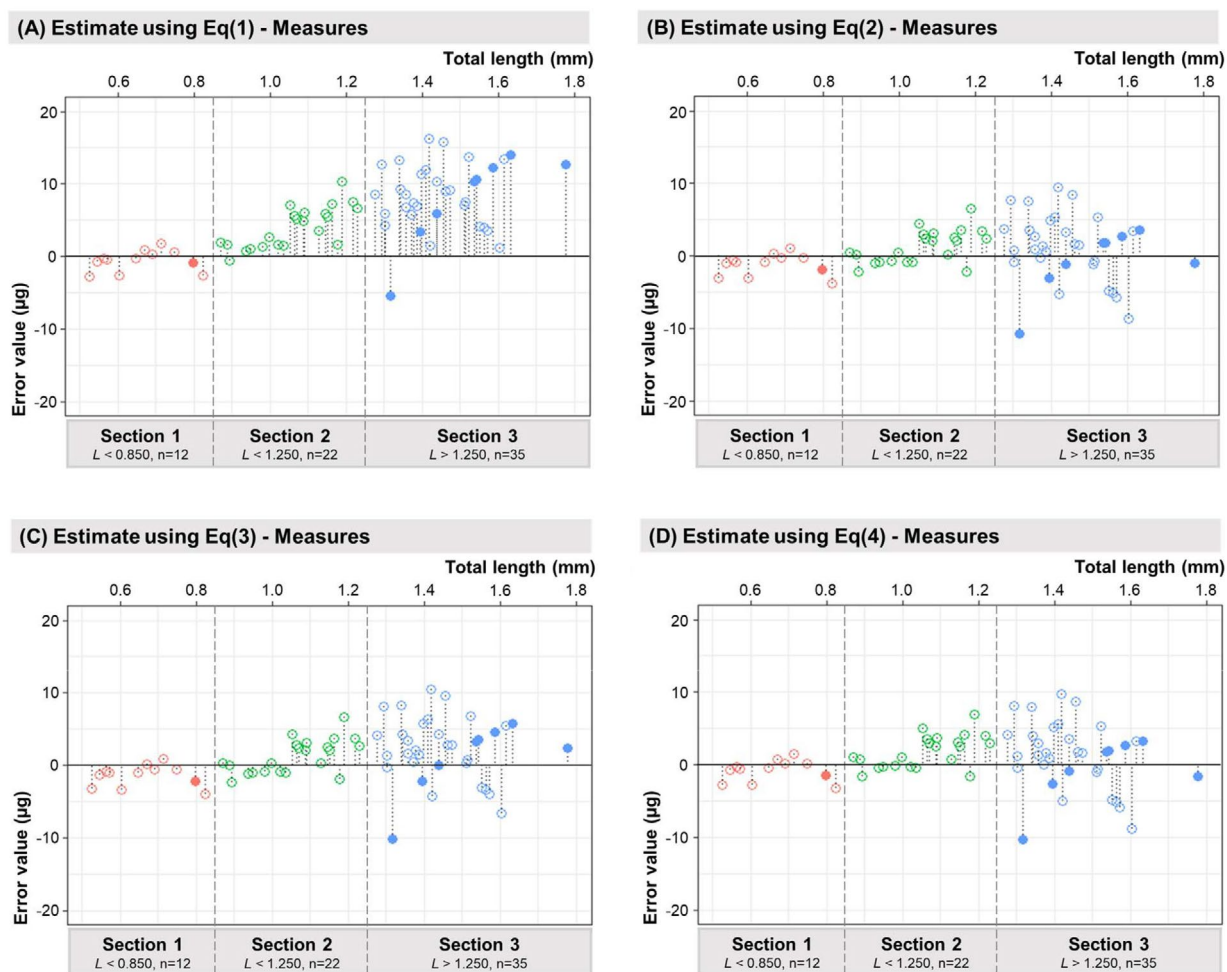


Fig. 3. Trends in the degree of error between the estimated and measured dry weights (biomass) of pooled cycloids varied with body length. These estimates were calculated using length-weight regression equations proposed by (A)~(B) Dumont *et al.* (1975), (C) Burgis, 1975 and (D) Bottrell *et al.* (1976). A hollow dot represents individuals without egg-sacs, while a filled dot denotes those with egg-sacs.

that excluded the caudal rami for *Cyclops* spp.

Bottrell *et al.*'s (1976) Eq. (4), proposed for estimating unidentified cyclopid biomass in the Korean Biomonitoring Survey and Assessment Manual-Weir section, exhibited no significant estimate-observation error, both overall and by section. However, it was relatively more erroneous compared to Dumont *et al.*'s (1975) Eq. (2), which is designed for adult cycloids excluding females with eggs, as shown in Fig. 3 and Table 4. Concerns were raised that Eq. (2), which was derived specifically for non-egg-bearing female individuals, might significantly underestimate the biomass of females with egg-sacs. However, when comparing the estimate-to-observation errors of various equations, Eq. (2) demonstrated relative adequacy. The mean absolute errors (MAEs) in dry weight for females with egg-sacs were as

follows: Eq. (1) resulted in an MAE of 8.41, Eq. (2) had an MAE of 3.07, Eq. (3) showed an MAE of 3.80, and Eq. (4) recorded an MAE of 2.96.

Meanwhile, when the four previously proposed biomass estimation equations were applied to individuals in this study, differences were observed depending on the size of the individuals. For smaller individuals (bin 1; TL < 0.850 mm), the biomass estimates tended to be underestimated, with negative error rates as follows: Eq. (1) approximately 67%, Eqs. (2) and (3) about 83% each, and Eq. (4) around 67%. On the other hand, for larger individuals (bins 2 and 3; TL > 0.850 mm), the estimates generally showed overestimations, with positive error rates: about 96% for Eq. (1), approximately 65% for Eq. (2), around 70% for Eq. (3), and about 67% for Eq. (4).

Table 4. Summary of error information between biomass (dry weight) estimates and measures of pooled cyclopoids by total length bin shown from Fig. 3; see Table 2 for a detailed description of Eqs. (1)~(4). Shading: the lowest mean absolute error (MAE).

| Error value (Estimates – Measures) | | Total (n=49) | | |
|--|-----------------|---------------------|---------------------|---------------------|
| | | Section 1 (n=12) | Section 2 (n=22) | Section 3 (n=35) |
| (A) Eq. (1); Dumont <i>et al.</i> , 1975 | Range (Min~Max) | – 5.46~16.21 | | |
| | | – 2.79~1.86 | – 0.60~10.34 | – 5.46~16.21 |
| | MAE | 1.21 | 4.12 | 8.70 |
| (B) Eq. (2); Dumont <i>et al.</i> , 1975 | Range (Min~Max) | – 10.81~9.47 | | |
| | | – 3.76~1.12 | – 2.15~6.46 | – 10.81~9.47 |
| | MAE | 1.44 | 2.05 | 3.72 |
| (C) Eq. (3); Burgis, 1975 | Range (Min~Max) | – 10.23~10.45 | | |
| | | – 4.01~0.87 | – 2.37~6.66 | – 10.23~10.45 |
| | MAE | 1.58 | 2.11 | 4.12 |
| (D) Eq. (4); Bottrell <i>et al.</i> , 1976 | Range (Min~Max) | – 10.38~9.75 | | |
| | | – 3.25~1.55 | – 1.61~7.00 | – 10.38~9.75 |
| | MAE | 1.23 | 2.25 | 3.83 |

3. Derive an equation for pooled cyclopoid species' biomass using body measurements from domestic individuals

Considering that zooplankton samples collected from lakes contain a mixture of several copepod species and accurate identification is difficult, the relationship between body measurements (total length, TL; prosome length, PL) and dry weight was analyzed using both exponential and power regression functions (Fig. 4). This analysis was conducted to develop a more accurate equation for estimating the dry weight (DW) of cyclopoid individuals from different genera in Korea. The linear function resulted in negative dry weight values for individuals with $TL < 0.604$ mm and $PL < 0.396$ mm, making it unsuitable for estimating pooled cyclopoid

biomass. Consequently, these data were excluded from this analysis. In the TL-DW regression analysis, the exponential and logarithmic models yielded multiple R-squared (R^2) values of 0.7713 and 0.7288, respectively (Fig. 4A). Similarly, the PL-DW regression resulted in R^2 values of 0.6727 for the exponential form and 0.6561 for the logarithmic form (Fig. 4B). The higher coefficient of determination values observed in the exponential models for both length measurements suggest that the exponential function more accurately captures the relationship between increasing measurements of pooled cyclopoids and their dry weight increase (Fig. 4).

The mean absolute error (MAE) was utilized to assess the goodness of fit for the regression equations correlating body measurements with the dry weight of pooled cyclopoids. It was determined that the deviations between the estimated and

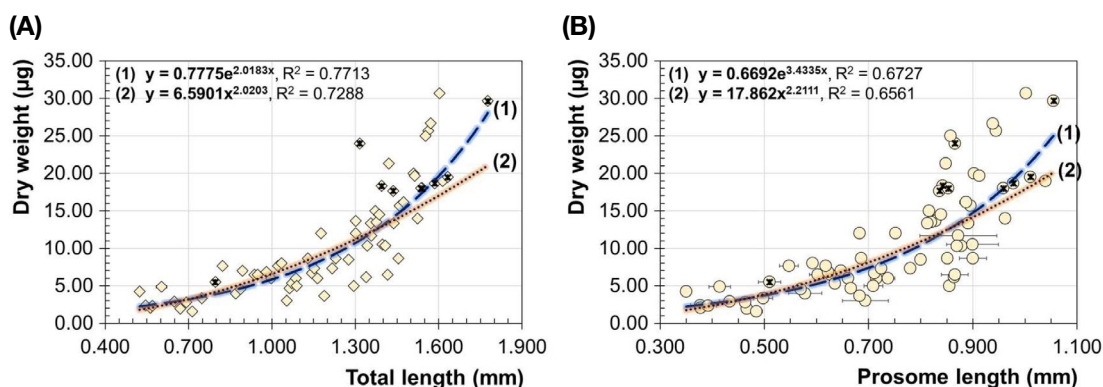


Fig. 4. Regression analyses and fitting results between the body specifications of pooled cyclopoids’ individuals and their biomass (dry weight) measurements; (A) Total length, and (B) Prosome length / (1) exponential function, and (2) power function. * represents individuals with egg-sacs.

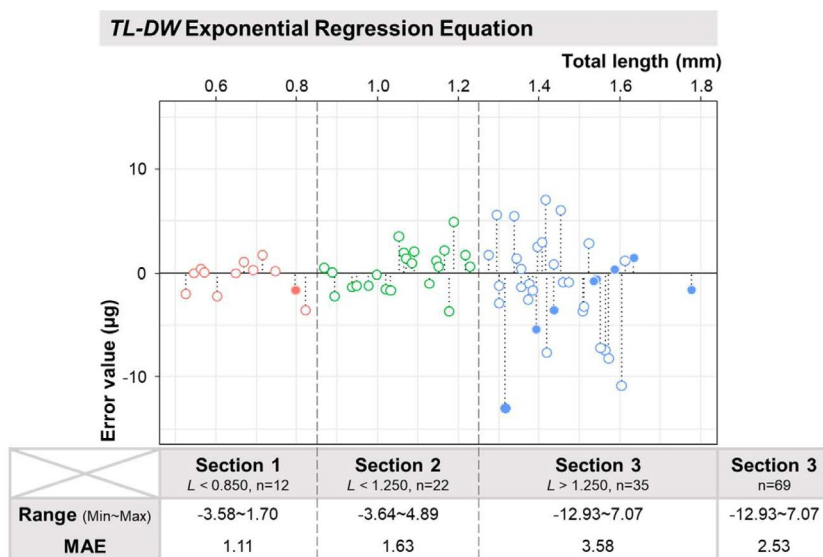


Fig. 5. Trends in the degree of error between estimated and actual biomass (dry weight) of pooled cyclopoids from total length-dry weight exponential regression equation newly derived in this study: A hollow dot represents individuals without egg-sacs, while a filled dot represents individuals with egg-sacs. The table contains the range of error (estimates-measures) and MAE (mean absolute error) for each bin and overall.

actual dry weights from the exponential regression equations were minor for both TL and PL (MAE: TL-DW exponential function, 2.53; TL-DW power function, 2.93; PL-DW exponential function, 3.21; PL-DW power function, 3.33). However, the PL-DW exponential regression equation exhibited a higher MAE value (3.21) compared to the Eq. (2)-Dumont *et al.* (1975) equation (MAE = 2.79; Table 4), which displayed the smallest error among the tested equations. Consequently, the newly derived TL-DW exponential regression equation is anticipated to provide the most accurate estimates of dry

weight, closely aligning with actual measurements.

To assess the enhancement in estimating the dry weight of pooled cyclopoids using the newly derived TL-DW exponential regression, total length (TL) was segmented into intervals based on the K-means clustering algorithm to evaluate error levels in each size interval (Fig. 5). The analysis revealed that although the estimated-to-observed error increased with increasing TL—similar to observations with existing biomass estimation equations—the magnitude of error using the new equation was consistently lower, both within each defined

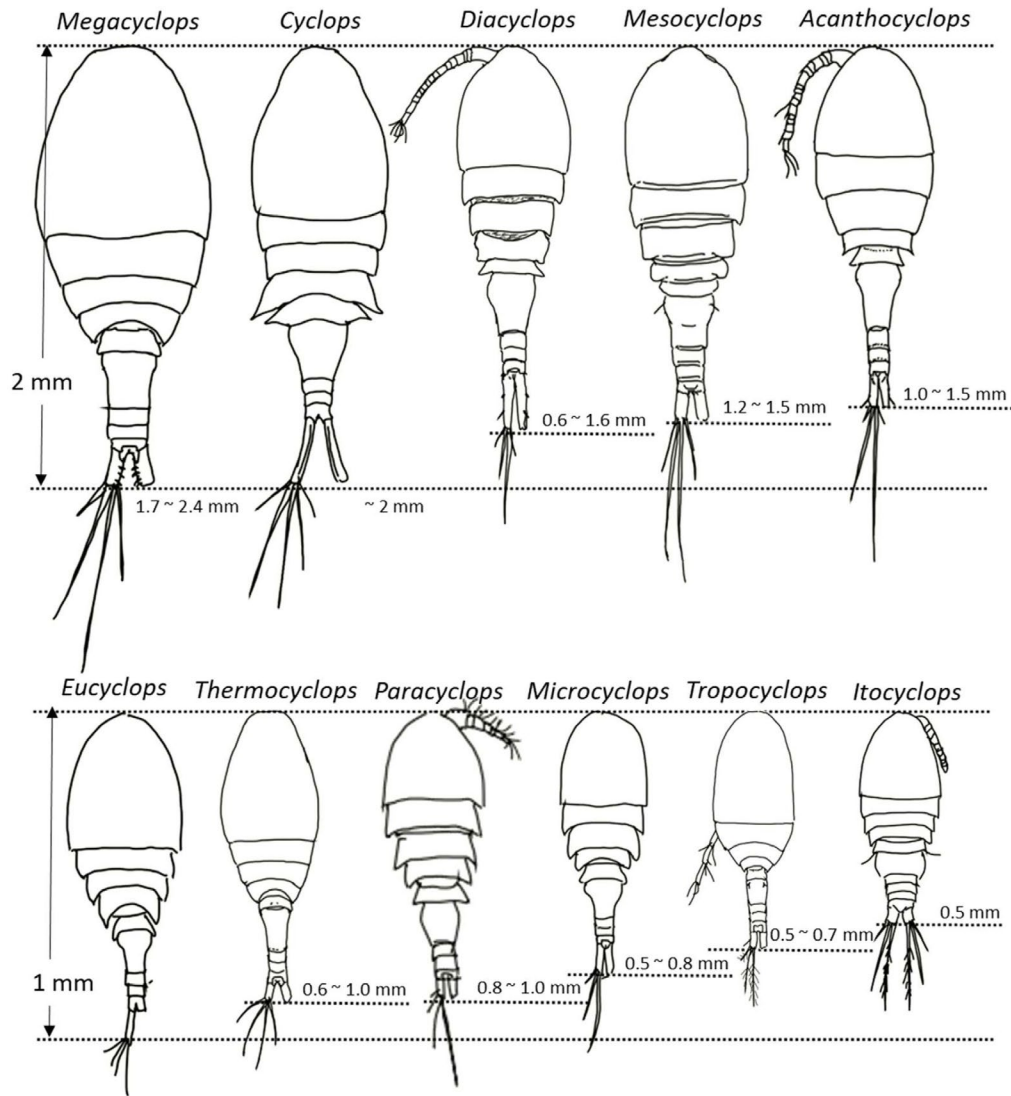


Fig. 6. Major genera of cyclopoid copepods found in Korea: morphology and total length (TL) (The figure was redrawn based on the cited references (Suárez-Morales *et al.*, 2005; NIBR, 2012, 2013)).

interval and across all intervals (Fig. 3; Table 4; Fig. 5). However, when this new TL-DW regression equation was applied specifically to females with egg-sacs, the mean absolute error (MAE) of 3.13, although lower than some existing equations, did not show a significant improvement.

Despite potential discrepancies between actual measurements and estimates derived from the regression equation in each interval and overall, the TL-DW exponential regression equation [$W = 0.7775 \times e^{2.0183L}$; W (μg), L (mm)] proposed in this study offers a more accurate method for estimating the body mass of pooled cyclopoids in Korea (Figs. 4~5). Nevertheless, given that the cyclopoid species utilized in this study primarily consist of those from the

genera *Cyclops*, *Thermocyclops*, and *Mesocyclops*, further studies are essential to verify the validity and applicability of this new equation across the wide spectrum of cyclopoid species found throughout Korea.

DISCUSSION

This study critically reviewed existing body length-weight regression equations for estimating the biomass of cyclopoids in Korea, which were not species-specific, and endeavored to derive a more suitable regression model by correlational analysis of dry weight with body measurements, specifically total length (TL) and prosome length (PL). For our experimental

process, precision was verified through the consistency in weight measurements obtained from triplicate weighings of the tin capsules before and after drying, in which the average coefficient of variation was maintained below 0.01%. This high level of precision of the micro-scale ensured reliability in our data collection process.

Furthermore, the methodology was refined by grouping individuals of similar body size for weighing, based on comprehensive data from previous studies, to minimize potential errors in estimating dry weight using the microscale. As a result, the range of measurements for the cyclopoid specimens in this study spanned from 0.524 mm to 1.777 mm in total length and from 0.350 mm to 1.055 mm in prosome length, with their dry weights varying between 1.58 µg and 30.67 µg.

When the equation for pooled copepod species (Eq. (4) in Table 2) by Bottrell *et al.* (1976), which is proposed in the national guidelines to estimate the biomass of unidentified cyclopoida, was applied to individuals, the estimate-to-estimate error was not large, but the estimate using the equation for adult cyclopoid species (Eq. (2) in Table 2) by Dumont *et al.* (1975), excluding females with eggs, was more similar to the actual value. Regression analyses were performed using TL and PL to derive new length-weight regression equations that minimize the estimated-observed error, but the PL-DW regression explained less dry weight with length growth than the TL-DW regression. This is likely due to the fact that even within cyclopoids with similar morphology, prosome length can vary between species. On the other hand, width showed high explanatory power for dry weight increase with individual growth in other crustacean species of the same phylum (Oh *et al.*, 2023), but in the case of cyclopoids, it was considered inappropriate as a regressor due to the difficulty in accurately measuring the width of small individuals during sample identification.

The TL-DW regression showed the lowest estimated-to-observed error in the exponential form, and its MAE was also lower than that of the conventional formulas. The TL-DW exponential regression equation derived in this study for the pooled cyclopoid species provided more accurate dry weight estimates than the previously suggested biomass estimation equations for individuals between 0.5 and 1.8 mm, but did not improve the accuracy of dry weight estimation for females with egg-sacs.

In conclusion, the refinement of the regression equation methodology has improved the accuracy of biomass estimations for cyclopoids in Korea. Cyclopoid copepod species

found in Korea exhibit a wide range of body length but are morphologically conserved (Fig. 6, NIBR, 2012, 2013). For a more accurate calculation of biomass, an equation that considers the subtle morphological differences between species or genera is necessary. However, due to the aforementioned morphological similarities among genera of copepods, species identification requires considerable effort and time, constraining the potential to calculate biomass using specific equation for each species in an environment where multiple species are present. Additionally, the occurrence of unidentifiable copepodids at high population densities further complicates a species-specific approach. Therefore, in this study, a generalized equation, ignoring species differences, is suggested that can be used to estimate biomass using total length (TL) for pooled copepods.

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Author contribution HJO, YSP, YJK and KHC contributed to the research conception and design. Field sampling was performed by HJO, GHH, YC, DHL, and KHC. Length and dry weight measurements were conducted by HJO, GHH, and HLW. Data management and statistical analyses were conducted by HJO, and GHH. The illustration of copepods genera was drawn by KHC. HJO wrote the first draft of the manuscript. HJO and KHC revised the final version of the manuscript. The research project was managed by YSP.

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