ABS ACTA BIOLOGICA SIBIRICA
A journal on the biodiversity of Siberia and the adjacent lands

Acta Biologica Sibirica 12: 1179–1190 (2026)
doi: 10.5281/zenodo.22822271
http://journal.asu.ru

RESEARCH ARTICLE

Towards standardization of carotenoid analysis in crustaceans: effects of storage, extraction time, and calculations

Liubov V. Yanygina[1], Egor A. Vorobiev[1], Petr A. Shipunov[1]

1 *Altai State University, 61 Lenina Ave., Barnaul, 656049, Russia*

Corresponding author: Liubov V. Yanygina (yan_lv@mail.ru)

Academic editor: A. Matsyura | Received 5 August 2026 | Accepted 4 September 2026 | Published 19 September 2026

http://zoobank.org/F2CD2703-5B61-4CC0-959C-76506DBB372C

Citation: Yanygina LV, Vorobiev EA, Shipunov PA (2026) Towards standardization of carotenoid analysis in crustaceans: effects of storage, extraction time, and calculations. Acta Biologica Sibirica 12: 1179–1190. https://doi.org/10.5281/zenodo.22822271

Abstract

Carotenoids play an essential role in aquaculture by supporting growth, survival, and stress tolerance in cultured species, while also contributing to product coloration, an important indicator of its quality and market value. Reliable monitoring of carotenoid levels in cultured organisms and feeds, as well as the development of effective feed enrichment strategies, requires standardized analytical procedures. This study provides a comparative evaluation of sample preparation methods and calculation approaches used for spectrophotometric determination of total carotenoids, using *Artemia* as a model organism. The results show that extending acetone extraction from one to three days increases the measured carotenoid concentration in fresh samples, whereas the opposite trend is observed in frozen samples, where prolonged extraction leads to pigment degradation. Additionally, the use of different calculation formulas can produce multi-fold discrepancies in carotenoid concentrations derived from identical absorbance data. These findings highlight the need for unified protocols for sample preparation, extraction, and data processing to ensure consistency and comparability of carotenoid measurements in aquaculture research and practice.

Keywords

Carotenoids, spectrophotometry, *Artemia*, aquaculture nutrition

Introduction

Carotenoids constitute a diverse group of lipid-soluble natural pigments with a wide range of biological functions across aquatic and terrestrial organisms. In aquatic ecosystems, these compounds are synthesized primarily by photosynthetic organisms, where they serve as accessory light-harvesting pigments and provide photoprotection (Maoka 2020). Animals are unable to synthesize carotenoids de novo, but they can metabolically modify dietary pigments into new derivatives. In aquatic animals, carotenoids contribute to ultraviolet protection, antioxidant defense, stress tolerance, and immune function (Tan et al. 2020). They also play a key ecological role by determining body coloration, which influences camouflage, communication, species interactions, and reproductive behavior (Svensson & Wong 2011).

In natural environments, aquatic animals obtain carotenoids through trophic pathways originating from primary producers. In aquaculture, however, artificial feeds often contain insufficient levels of these pigments, which may lead to carotenoid deficiency in cultured species. Numerous studies have demonstrated that dietary carotenoids enhance growth performance, survival, reproductive success, and overall physiological condition in fish and crustaceans (Safari & Atash 2015; Çankırılıgil et al. 2022; Rahman & Shikdar 2025). Moreover, carotenoid-based pigmentation is widely recognized by consumers as an indicator of product quality and can significantly influence the market value of aquaculture products (Steine et al. 2004; Parisenti et al. 2011; de Carvalho & Caramujo 2017; Rosenau et al. 2023). These factors have driven the widespread adoption of carotenoid supplementation in aquaculture feeds (Manikandan et al. 2020; Elbahnaswy & Elshopakey 2024). *Artemia* nauplii are widely used as a starter feed for early life stages of fish and shrimp, which are highly sensitive to feed quality, and they may be enriched with carotenoids to improve their nutritional value.

Given the ecological and practical importance of carotenoids, reliable and comparable data are essential for assessing pigment dynamics in natural populations and evaluating feed quality in aquaculture. However, current methodologies for carotenoid determination vary widely in sample preparation, extraction procedures, and calculation approaches, often hindering meaningful comparison across studies. The aim of this work is to critically evaluate existing methods used to quantify carotenoids in aquatic organisms and to propose recommendations for selecting appropriate approaches that ensure data comparability. This study focuses on spectrophotometric determination of total carotenoids, a method widely used in aquaculture research due to its simplicity and accuracy comparable to chromatographic techniques (Casella et al. 2020). The specific objectives were: (1) to assess the effects of freezing and extraction duration on measured total carotenoids in *Artemia* tissues; (2) to compare carotenoid concentrations calculated using the most commonly applied formulas in the literature; and (3) to develop recommendations for standardizing sample preparation and data processing in spectrophotometric carotenoid analysis.

Materials and methods

Sample collection and preparation

Artemia specimens were collected from Lake Bolshoye Yarovoye (Ob-Irtysh interfluve) on September 22, 2025. The crustaceans were captured using a small Jeddi net in the surface layer of the lake, placed into 5-L containers with lake water, and transported alive to the laboratory. Upon arrival, the samples were transferred into Petri dishes, and live individuals of similar size were selected using forceps. After preliminary drying on filter paper to remove surface moisture, the specimens were placed onto Vladipor MFAS-OS-3 membrane filters, 30 individuals per filter. A total of 50 *Artemia* samples were prepared in this manner, each weighed using a VT-500 torsion balance. Prior to the experiments, sample moisture content was determined using a MOC63u moisture analyzer (Shimadzu, Japan), with a mean value of 76.34%. The coefficient of 4.2 was applied to convert wet weight to dry weight.

Experimental design: effect of freezing and extraction duration

To assess the effect of storage conditions on the measured carotenoid concentration, two sample sets were created. For the first set (20 samples), pigment extraction was started on the day of capture. The second set (30 samples) was frozen for 45 days at –20 °C before extraction. Within each set, the influence of extraction time was further evaluated. The fresh samples were divided into two groups of 10 samples each, extracted for either 1 day or 3 days. The frozen samples were split into three groups: extraction for 1 day (10 samples), 3 days (10 samples), or 7 days (10 samples).

Pigment extraction

For pigment extraction, the filters with *Artemia* tissues were placed into 5-mL centrifuge tubes. A first extraction was performed by adding 3.0 mL of 100% acetone to each tube. The tubes were capped to prevent evaporation and stored at 6 °C in darkness for the designated extraction period (1, 3, or 7 days). After the extraction period, the samples were centrifuged at 5000 rpm for 15 min to sediment suspended particles. The supernatant was transferred to a separate tube. A second extraction was then performed for 2 h by adding 2.0 mL of 100% acetone. After centrifugation at 5000 rpm for 15 min, the second supernatant was combined with the first, the total volume of the extract (V) was recorded for each sample and used for calculations.

The absorbance of the samples was measured using an SF-56 spectrophotometer over the wavelength range of 400–950 nm, with a slit width of 6 nm. A blank sample (a filter without *Artemia* tissue, processed identically) served as the control.

Calculation of total carotenoid concentration

Total carotenoid concentration was calculated as astaxanthin equivalents using the Bouguer–Lambert–Beer law:

$$TCC = \frac{A_{470} \times V \times 10^4}{E^{1\%}_{1\text{см}} \times L \times m}$$

where TCC is the total carotenoid content (μg·g^{-1} of tissue); A_{470} is the absorbance of the extract at 470 nm; V is the extract volume (ml); 10^4 is the conversion factor used to express the result in μg·g^{-1}; $E^{1\%}_{1\text{см}}$ is the specific extinction coefficient of a 1% astaxanthin solution in acetone measured at optical path length 1 cm, taken as 2100 dl·(g·cm)$^{-1}$ (Johnson & An 1991; Feizi & Jafari 2025); L is the optical path length (1 cm); m is the sample mass (g). This formulation is widely used for acetone extracts in which astaxanthin is the predominant carotenoid, and it allows direct conversion of spectrophotometric absorbance values into total carotenoid concentration.

At the next stage, total carotenoid concentrations were recalculated using different published formulas. These calculations were based on absorbance values obtained from acetone extracts of *Artemia* tissues collected from Lake Bolshoye Yarovoye, prepared by freezing the samples followed by a 24-hour extraction. Statistical analyses were performed using Statistica 6.0. Data are presented as arithmetic means with their standard errors (M±m). Differences between groups were evaluated using the Mann–Whitney test, and statistical significance was accepted at $p<0.05$. A two way ANOVA was used to test the interaction between storage condition (fresh or frozen) and extraction duration (1 or 3 days). Normality of residuals was assessed using the Shapiro–Wilk test, and homogeneity of variances was confirmed with Levene's test, both of which supported the use of parametric ANOVA.

Results

Effect of extraction duration on carotenoid concentration

The conducted study demonstrated that extraction duration significantly influences carotenoid concentration in *Artemia* tissues, with opposite trends observed for fresh and frozen samples (Fig. 1). The two way ANOVA revealed a statistically significant interaction between storage condition and extraction duration ($F = 23.03$, $p < 0.001$). This confirms that the effect of extraction duration on carotenoid concentration differs significantly between fresh and frozen samples. In fresh samples, carotenoid concentrations increased statistically significantly (Mann–Whitney test, $p < 0.001$) with increasing extraction time. Mean carotenoid concentrations after 3 days of extraction were 45.9% higher than after 1 day. In frozen samples, the maxi-

mum concentration was recorded at the minimum extraction duration (1 day). After 3 days, the mean concentration decreased by nearly 20%, and after one week it amounted to only 59% of the values obtained from frozen samples extracted for 1 day (Fig. 1).

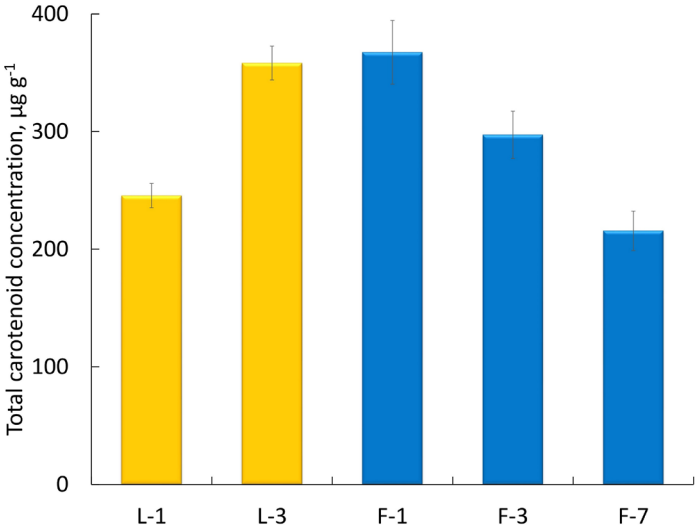

Figure 1. Total carotenoid concentrations (µg g-1) in fresh (L) and frozen (F) *Artemia* tissues after 1, 3, and 7 days of extraction (n=10 per treatment).

Effect of freezing on carotenoid concentration

No statistically significant difference (Mann–Whitney test, p = 1.0) was found between concentrations after 3 days of extraction of fresh samples and 1 day of extraction of frozen samples. These values were the highest among all experimental groups. At the same time, a statistically significant difference (Mann–Whitney test, p = 0.02) was found between carotenoid concentrations from fresh *Artemia* tissues extracted for 1 day and frozen tissues extracted for 3 days. The minimum carotenoid concentrations were observed after 7-day extraction of carotenoids from frozen samples.

Comparative analysis of different carotenoid concentration calculation methods

Total carotenoid concentrations in *Artemia* tissues from Lake Bolshoye Yarovoye, calculated using different formulas, varied substantially (Table 1). Although Formulas 1–3 are based on common calculation principles, the resulting values differed by 1–4 orders of magnitude. Formulas 2 contained an erroneous conversion factor (10^3), apparently due to incorrect interpretation of the physical meaning of the

extinction coefficient, which indicates the absorbance of a 1% extract at 1 cm path length and is measured in $dL \cdot g^{-1} \cdot cm^{-1}$ (not $mL \cdot g^{-1} \cdot cm^{-1}$). The results calculated using Formula 4 were approximately two times lower than values obtained with Formula 1. Formula 5 substantially overestimated the results, which is attributed to a technical error in the formula (missing parentheses). A similar situation is observed when using Formula 6.

Table 1. Comparative analysis of approaches to total carotenoid concentration (TCC) calculation in *Artemia* tissues

No	Formula	This study	TCC, μg g^{-1}	References
1	$TCC = \frac{A_\lambda \times V \times 10^4}{E^{1\%}_{1см} \times W}$	154,3±11,4	200	Elshafey et al. 2023
2	$TCC = \frac{A_\lambda \times V \times 10^3}{E^{1\%}_{1см} \times W}$	15,4±1,1	1,79±0,14	Kanokrung et al. 2024
3	$TCC = \frac{A_\lambda \times V \times Df}{E^{1\%}_{1см} \times W}$	0,015±0,001	82*	Abdollahi et al. 2019
4	$Chl_a = 16.72A_{665.2} - 9.16A_{652.4}$ $Chl_b = 34.09A_{652.4} - 15.28A_{665.2}$ $TCC = \frac{1000A_{470} - 1.63Chl_a - 104.96Chl_b}{221}$	75,18±4,6	180	Cheban et al. 2020
5	$Chl_a = 11.75A_{662} - 2.350A_{645}$ $Chl_b = 18.61A_{645} - 3.960A_{662}$ $TCC = 1,000A_{470} - 2.270Chl_a - \frac{81.4Chl_b}{227}$	21184±1445	112620±1980	Anuevo 2014
6	$TCC = 1 \times 10^4 \times \frac{OD_{450}}{2.62\%} \times \frac{V}{W}$	130801±9943	45900±130	Vahdat et al. 2018

Note: * In the TCC caculation, the dilution factor employed was not provided.

Discussion

The rapid expansion of aquaculture in recent decades has stimulated extensive research aimed at identifying factors that enhance the productivity and resilience of cultured aquatic species. Numerous studies have demonstrated that carotenoids play a critical role in improving growth, survival, stress tolerance, and immune function, as well as in determining the pigmentation and market value of aquaculture products (Steine et al. 2004; Parisenti et al. 2011; Safari & Atash 2015; Çankırılıgil et al. 2022; Rahman & Shikdar 2025). Despite the substantial data available on carotenoid levels in cultured organisms and live feeds, comparative analysis remains challeng-

ing due to considerable variation in sample preparation procedures, extraction protocols, and calculation methods used across studies. This highlights the need for a critical evaluation of existing analytical approaches and the development of standardized methodologies suitable for aquaculture research and practice.

Chromatographic and spectrophotometric techniques are the two most widely used methods for carotenoid analysis in biological samples. Chromatographic approaches enable the quantification of individual carotenoid compounds and are essential for studying metabolic pathways and pigment composition. Spectrophotometry, by contrast, provides an integrated estimate of total carotenoids without separating individual components. Owing to its simplicity, rapidity, and minimal equipment requirements, spectrophotometry is widely applied in practical aquaculture research, particularly for evaluating the efficiency of carotenoid enrichment in live feeds such as *Artemia* (Anuevo 2014; Abdollahi et al. 2019; Cheban et al. 2020; Elshafey et al. 2023; Millán-Almaraz et al. 2023; Kanokrung et al. 2024). Comparative studies have shown that spectrophotometric measurements of total carotenoids can achieve accuracy comparable to chromatographic methods (Casella et al., 2020), making this approach suitable for routine assessments of feed quality and pigment accumulation.

Regardless of the analytical method used, sample preparation is a critical step that strongly influences the accuracy of carotenoid measurements. One of the key methodological decisions concerns the use of frozen versus fresh samples. Freezing is often preferred because it facilitates workflow organization and enables the analysis of organisms collected far from laboratory facilities. Previous studies have shown that freezing at −20 °C does not lead to pigment degradation and allows longterm preservation (Tan et al. 2024). Freezing also disrupts cellular structures, thereby improving pigment extraction efficiency. In the present study, carotenoid concentrations in frozen *Artemia* samples after 24 hours of extraction were comparable to those in fresh samples only after three days of extraction. This confirms that freezing can significantly accelerate pigment release. Freezing is widely used in carotenoid studies involving *Artemia* (Anuevo 2014; Vikas et al. 2014) and other aquatic organisms (Hooshmand et al. 2017), including cultured species (Maoka et al. 2018).

Extraction solvent and extraction duration represent additional methodological factors that influence carotenoid quantification. Acetone was selected in this study due to its widespread use for carotenoid extraction from biological tissues, including *Artemia* (Hsu et al. 1970; Abdollahi et al. 2019; Cheban et al. 2020; Huang & Hui 2020; MillánAlmaraz et al. 2023; Kanokrung et al. 2024). A notable finding of our work is that maximum carotenoid concentrations in fresh *Artemia* samples were achieved only after three days of extraction. This suggests that intact tissue structures in unfrozen organisms may hinder solvent penetration and pigment release, resulting in incomplete extraction within 24 hours. Nevertheless, the use of fresh *Artemia* remains common in enrichment experiments (Abdollahi et al. 2019; Cheban et al. 2020; Elshafey et al. 2023; Wang et al. 2024) and in studies of natural crustacean populations (Hsu et al. 1970; Nelis et al. 1988; Kumar & Marian 2006;

Xue et al. 2024). Our results indicate that this practice may lead to substantial underestimation of carotenoid concentrations, particularly when absolute values are compared across studies. Conversely, prolonged extraction of frozen samples (three days or more) possibly resulted in pigment degradation and reduced carotenoid concentrations, suggesting that extended exposure to acetone may negatively affect pigment stability. Thus, the most reliable approach for *Artemia* appears to be freezing followed by a 24hour acetone extraction. However, optimal extraction duration and sample preparation procedures may vary among taxa and tissue types, warranting further investigation.

Even greater discrepancies in carotenoid measurements arise from differences in calculation methods. Several formulas based on the Bouguer–Lambert–Beer law are used in the literature. The most common approach involves converting absorbance values into carotenoid concentrations using tabulated extinction coefficients. Errors may arise when incorrect conversion factors are used, for example, substituting 10^3 for 10^4, as reported in some studies (e.g., Kanokrung et al. 2024). Such errors likely stem from misinterpretation of the physical meaning of the extinction coefficient, which is expressed in $dl \cdot (g \cdot cm)^{-1}$ rather than $ml \cdot (g \cdot cm)^{-1}$, and can lead to a tenfold underestimation of carotenoid concentrations.

Another widely used approach involves formulas that account for the specific absorption coefficients of different pigments. The Lichtenthaler and Welburn (1983) formula is widely used for total carotenoids calculation in both plant and animal tissues. This method assumes that absorbance at any wavelength is the sum of absorbances of individual components, allowing correction for chlorophyll interference. In some studies (e.g., Anuevo 2014), the Lichtenthaler and Welburn formula is presented with a technical error (missing parentheses), resulting in substantial overestimation of total carotenoids. Importantly, when whole individuals are extracted together with gut contents containing undigested algae, the Lichtenthaler and Welburn formula helps avoid overestimation by correcting for chlorophyll absorption. In contrast, calculations based solely on extinction coefficients may yield inflated carotenoid values that reflect gut fullness rather than tissue pigment content. In our study, carotenoid concentrations calculated using the extinction coefficient were two times higher than those obtained using the Lichtenthaler and Welburn formula.

Conclusion

This study demonstrates that the choice of calculation method can influence the estimated carotenoid concentration to a greater extent than the tested sample preparation procedures or extraction durations. Although each individual study typically applies a consistent analytical protocol internally ensuring the validity and reliability of its own conclusions differences in storage conditions, sample preparation, extraction time, and calculation formulas limit the comparability of absolute carotenoid values across publications. Consequently, data obtained using different methodo-

logical approaches should be interpreted with caution when used for cross-study comparisons or meta-analyses.

Our findings highlight the importance of methodological consistency in carotenoid research. Standardization is needed not only for sample preparation and extraction procedures but also for the selection and transparent reporting of calculation formulas. For whole Artemia individuals (without gut removal), the most consistent results were obtained when samples were frozen prior to analysis, pigments were extracted in acetone for 24 hours.

Overall, the results underscore the need for continued refinement of analytical protocols and encourage the development of standardized guidelines that can be adopted across laboratories. Such efforts will strengthen both fundamental research on carotenoid metabolism and its practical applications in modern aquaculture.

Acknowledgments

The research was supported by Russian Science Foundation (project No 25-16-00099, https://rscf.ru/project/25-16-00099).

References

Abdollahi Y, Ahmadifard N, Agh N, Rahmanifarah K, Hejazi MA (2019) βCaroteneenriched *Artemia* as a natural carotenoid improved skin pigmentation and enhanced the mucus immune responses of platyfish *Xiphophorus maculatus*. Aquaculture International 27: 1847–1858. https://doi.org/10.1007/s10499-019-00437-8

Anuevo GM (2014) Evaluation of green tea enriched *Artemia* sp. as food and antioxidant activity enhancer for freshwater ornamental fishes. PhD dissertation, University of Tsukuba.

Çankırılıgil EC, Berik N, Çakmak E, Özel OT, Erbay EA (2022) Dietary carotenoids influence growth, fillet pigmentation, and quality characteristics of Black Sea trout (*Salmo labrax* Pallas, 1814). Thalassas 38:793–809. https://doi.org/10.1007/s41208-022-00407-7

Casella P, Iovine A, Mehariya S, Marino T, Musmarra D, Molino A (2020) Smart method for carotenoids characterization in *Haematococcus pluvialis* red phase and evaluation of astaxanthin thermal stability. Antioxidants 9: 422. https://doi.org/10.3390/antiox9050422

Cheban L, Khudyi O, Prusińska M, Duda A, Khuda L, Wiszniewski G, Kushniryk O, Kapusta A (2020) Survival, proximate composition, and proteolytic activity of *Artemia salina* bioencapsulated with different algal monocultures. Fisheries & Aquatic Life 28: 205–215. https://doi.org/10.2478/aopf-2020-0025

De Carvalho CC, Caramujo MJ (2017) Carotenoids in aquatic ecosystems and aquaculture: a colorful business with implications for human health. Frontiers in Marine Science 4: 93. https://doi.org/10.3389/fmars.2017.00093

Elbahnaswy S, Elshopakey GE (2024) Recent progress in practical applications of a potential carotenoid astaxanthin in aquaculture industry: a review. Fish Physiology and Biochemistry 50: 97–126. https://doi.org/10.1007/s10695-022-01167-0

Elshafey AE, Khalafalla MM, Zaid AAA, Mohamed RA, Abdel-Rahim MM (2023) Source diversity of *Artemia* enrichment boosts goldfish (*Carassius auratus*) performance, βcarotene content, pigmentation, immunephysiological and transcriptomic responses. Scientific Reports 13: 21801. https://doi.org/10.1038/s41598-023-48621-4

Feizi P, Jafari SM (2025) Optimization of astaxanthin extraction from red (*Gracilaria corticata*) and brown (*Sargassum polycystum*) macroalgae through ultrasonication and microwave processing. Ultrasonics Sonochemistry 121: 107556. https://doi.org/10.1016/j.ultsonch.2025.107556

Hata M, Hata M (1969) Carotenoid metabolism in *Artemia salina* L. Comparative Biochemistry and Physiology 29: 985–994. https://doi.org/10.1016/0010-406x(69)91000-7

Hooshmand H, Shabanpour B, Moosavi-Nasab M, Golmakani MT (2017) Optimization of carotenoids extraction from blue crab (*Portunus pelagicus*) and shrimp (*Penaeus semisulcatus*) wastes using organic solvents and vegetable oils. Journal of Food Processing and Preservation 41: e13171. https://doi.org/10.1111/jfpp.13171

Hsu WJ, Chichester CO, Davies BH (1970) The metabolism of βcarotene and other carotenoids in the brine shrimp, *Artemia salina* L. (Crustacea: Branchiopoda). Comparative Biochemistry and Physiology 32: 69–79. https://doi.org/10.1016/0010-406x(70)90156-8

Huang J, Hui B (2020) Feedinduced variation in the carotenoid composition of brine shrimp. eFood 1: 247–253. https://doi.org/10.2991/efood.k.200522.001

Johnson EA, An GH (1991) Astaxanthin from microbial sources. Critical Reviews in Biotechnology 11: 297–326. https://doi.org/10.3109/07388559109040622

Kanokrung A, Ratananikom K, Watanadilok R (2024) Effect of dietary carotenoids on survival, growth, and carotenoid accumulation in mandarinfish, *Synchiropus splendidus* (Herre, 1927). Journal of Fisheries and Environment 48: 38–47. https://doi.org/10.34044/j.jfe.2024.48.2.03

Kumar PA, Marian MP (2006) Studies on carotenoid in *Artemia parthenogenitica*. Romanian Biotechnological Letters 11: 2733–2737.

Lichtenthaler HK, Wellburn AR (1983) Determinations of total carotenoids and chlorophylls a and b of leaf extracts in different solvents. Biochemical Society Transactions 11: 591–592. https://doi.org/10.1042/bst0110591

Manikandan K, Felix N, Prabu E (2020) A review on the application and effect of carotenoids with respect to canthaxanthin in the culture of fishes and crustaceans. International Journal of Fisheries and Aquatic Studies 8: 128–133. https://doi.org/10.22271/fish.2020.v8.i5b.2314

Maoka T (2020) Carotenoids as natural functional pigments. Journal of Natural Medicines 74: 1–16. https://doi.org/10.1007/s11418-019-01364-x

Maoka T, Kawashima Y, Takaki M (2018) Structures of yellow xanthophylls and metabolism of astaxanthin in the prawn *Penaeus japonicus*. Journal of Oleo Science 67: 1425–1433. https://doi.org/10.5650/jos.ess18103

Millán-Almaraz MI, López-Peraza DJ, Nieves-Soto M, Peraza-Yee MM (2023) Effect of 4 microalgal diets on the proximal composition, chlorophyll concentration, and total carotenoid content in *Artemia franciscana*. Ciencias Marinas 49: e3381. https://doi.org/10.7773/cm.y2023.3381

Minakova AA, Bazarnova NG, Minakov DV, Mikushina IV, Dubrovina AE (2022) Obtaining astaxanthin from *Artemia salina* cysts for the development of functional food products. Polzunovskiy Vestnik 1: 167–173. https://doi.org/10.25712/astu.2072-8921.2022.04.022 [In Russian]

Nelis HJ, Lavens P, Van Steenberge MMZ, Sorgeloos P, Criel GR, de Leenheer A (1988) Qualitative and quantitative changes in the carotenoids during development of the brine shrimp *Artemia*. Journal of Lipid Research 29: 491–499. https://doi.org/10.1016/S0022-2275(20)38522-9

Ownagh E, Agh N, Noori F (2015) Comparison of the growth, survival and nutritional value of Artemia using various agricultural byproducts and unicellular algae *Dunaliella salina*. Iranian Journal of Fisheries Sciences 14: 358–368.

Parisenti J, Beirão LH, Tramonte VLCG, da Silva FO, da Silveira Brito CC, Moreira CC (2011) Preference ranking of colour in raw and cooked shrimps. International Journal of Food Science and Technology 46: 2558–2561. https://doi.org/10.1111/j.1365-2621.2011.02781.x

Rahman MM, Shikdar IK (2025) Assessing growth performance and flesh quality of silver carp (*Hypophthalmichthys molitrix*) with natural carotenoids in feeds. Sustainable Aquatic Research 4: 248–259. https://doi.org/10.5281/zenodo.18005874

Rosenau S, Wolgast T, Altmann B, Risius A (2023) Consumer preference for altered color of rainbow trout (*Oncorhynchus mykiss*) fillet induced by Spirulina (*Arthrospira platensis*). Aquaculture 572: 739522. https://doi.org/10.1016/j.aquaculture.2023.739522

Safari O, Atash MMS (2015) The effects of dietary supplement of annatto (*Bixa orellana*) seed meal on blood carotenoid content and fillet color stability in rainbow trout (*Oncorhynchus mykiss*). Aquaculture 437: 275–281. https://doi.org/10.1016/j.aquaculture.2014.12.012

Steine G, Alfnes F, Rora MB (2004) The effect of color on consumer WTP for farmed salmon. Marine Resource Economics 20(2): 211–219. https://doi.org/10.1086/mre.20.2.42629470

Svensson PA, Wong BBM (2011) Carotenoid-based signals in behavioural ecology: a review. Behaviour 148: 131–189. https://doi.org/10.1163/000579510x548673

Tan K, Zhang H, Lim LS, Ma H, Li S, Zheng HT (2020) Roles of carotenoids in invertebrate immunology. Frontiers in Immunology 10: 3041. https://doi.org/10.3389/fimmu.2019.03041

Tan K, Zhang H, Zheng H (2024) Carotenoid content and composition: A special focus on commercially important fish and shellfish. Critical Reviews in Food Science and Nutrition 64: 544–561. https://doi.org/10.1080/10408398.2022.2106937

Vahdat S, Esmaeili Fereidouni A, Khalesi MK (2018) Longterm effects of vermicompost manure leachate (powder) inclusions on growth and survival, biochemical composition, total carotenoids, and broodstock reproductive performance of *Artemia francis-*

cana (Kellogg, 1906). Aquaculture International 26: 569–588. https://doi.org/10.1007/s10499-017-0232-0

Vikas PA, Chakraborty K, Sajeshkumar NK, Thomas PC, Sanil NK, Vijayan KK (2014) Quality of six indian populations of *Artemia franciscana* for larval finfish culture. Journal of Applied Aquaculture 26: 271–291. https://doi.org/10.1080/10454438.2014.942124

Wang W, Ma Z, Li W, Xue Y, Moss AS, Wu M (2024) Impact of βcarotene enrichment on carotenoid composition and gene expression in *Artemia* metanauplii. Metabolites 14: 676. https://doi.org/10.3390/metabo14120676

Xue Y, Jiang G, Shu H, Wang W, Huang X (2024) Effects of temperature and salinity on the growth, reproduction, and carotenoid accumulation in *Artemia sinica* and transcriptome analysis. Fishes 9: 437. https://doi.org/10.3390/fishes9110437