Article

Not peer-reviewed version

Energy Allocation and Ovarian Nutrient Reserves in Japanese Eel (*Anguilla Japonica*) Before and After Seaward Migration: Evidences from Fatty Acid Profiles

Chao Song , Dongyu Song , Chengyao Yang , Yijia Li , Hang Li , Zhiqiang Ye , Junlin Ren , Sikai Wang , Feng Zhao *

Posted Date: 14 May 2026

doi: 10.20944/preprints202605.0915.v1

Keywords: Yangtze River Estuary; offshore; *Anguilla japonica*; fatty acid; energy allocation; nutrient reserve

Preprints.org (www.preprints.org) | NOT PEER-REVIEWED | Posted: 14 May 2026 doi:10.20944/preprints202605.0915.v1

Article

Energy Allocation and Ovarian Nutrient Reserves in Japanese Eel (*Anguilla Japonica*) Before and After Seaward Migration: Evidences from Fatty Acid Profiles

Chao Song [1,2,3,4], Dongyu Song [1,2], Chengyao Yang [2,3], Yijia Li [2,4,*], Hang Li [2,4], Zhiqiang Ye [2,5], Junlin Ren [2], Sikai Wang [2] and Feng Zhao [2,*]

[1] National Engineering Research Center for Marine Aquaculture, Zhejiang Ocean University, Zhoushan 316022, China
[2] East China Sea Fisheries Research Institute, Chinese Academy of Fishery Sciences, Shanghai 200090, China
[3] School of Environment and Architecture, University of Shanghai for Science and Technology, Shanghai 200093, China
[4] College of Fisheries and Life Science, Dalian Ocean University, Dalian 116023, China
[5] College of Marine Sciences, Fujian Agriculture and Forestry University, Fuzhou 350002, China
* Correspondence: zhaof@ecsf.ac.cn

Simple Summary

Anguilla japonica is a catadromous species that migrates to the ocean for spawning. By comparing the fatty acid composition in different tissues of *A. japonica* before and after their entry into the sea, this study elucidated the energy allocation and ovarian nutritional status during the migratory process. The distribution patterns of different fatty acids across tissues were closely associated with the progression of the seaward migration. Prior to entering the sea, *A. japonica* in the Yangtze River Estuary were primarily focused on energy accumulation and hepatic metabolism. After entering the sea, with the progression of gonadal development, the ovaries gradually accumulated polyunsaturated fatty acids (PUFA) such as EPA, DPA+DHA, and n3-PUFA. The selective retention of specific fatty acids in the ovaries represents a physiological regulation that meets the nutritional demands of gonadal development. These findings reveal the energy utilization and conversion mechanisms underlying the eel's adaptation to reproductive migration, offering insights into its reproductive physiology. This study provides foundational data for assessing the reproductive potential of *A. japonica* after entering the sea, and offers theoretical guidance for the selection of wild broodstock and the conservation of reproductive populations.

Abstract

To elucidate the characteristics of fatty acid composition in different tissues of *Anguilla japonica* before and after their seaward spawning migration, this study measured the fatty acid contents in the muscle, liver, and ovary of eels collected from the Yangtze River Estuary and offshore waters, and analyzed the distribution characteristics and transformation patterns of various fatty acids in different tissues. The results showed that the fatty acid composition in different tissues of eels from both the Yangtze River Estuary and offshore waters was essentially similar. Among all fatty acids, C18:1n9c had the highest proportion, accounting for over 31% of the total fatty acids in each tissue. Comparing different tissues, in eels from the Yangtze River Estuary, the muscle had the highest content of EPA, the liver had the highest content of DHA and EPA+DHA, and the ovary had the highest contents of ARA, n6-PUFA and SFA. In offshore eels, the muscle had the highest contents of C16:1, C18:1n9c and MUFA; the liver had the highest content of C16:0; and the ovary had the highest contents of C18:0, DPA, HUFA, n3-PUFA and PUFA. Comparing eels before and after seaward migration, the contents of C16:0, ARA, n6-PUFA, SFA, and the DHA/EPA ratio in the ovary of

Yangtze River Estuary eels were higher than those in offshore eels. Conversely, the contents of C18:0, C16:1, C18:1n9c, EPA, DPA, DHA, EPA+DHA, HUFA, n3-PUFA, MUFA, PUFA, as well as the EPA/ARA, n3/n6 PUFA and PUFA/SFA ratios in the ovary of offshore eels were higher than those in Yangtze River Estuary eels. The ovary of Yangtze River Estuary eels mainly contained fatty acids for energy provision and precursors for long-chain fatty acid synthesis, whereas the ovary of offshore eels had preliminarily accumulated PUFA nutrients required for egg and embryonic development. Thus, the distribution patterns of different fatty acids among tissues are closely related to the seaward spawning migration process of *A. japonica*. Before migration, eels in the Yangtze River Estuary primarily focus on energy accumulation and liver metabolism; after entering the sea, eels gradually accumulate PUFA such as EPA, DPA+DHA and n3-PUFA as gonads develop. The selective reservation of different fatty acids in the ovary represents a physiological regulation in response to the nutritional demands of gonadal development.

Keywords: Yangtze River Estuary; offshore; *Anguilla japonica*; fatty acid; energy allocation; nutrient reserve

1. Introduction

Anguilla japonica is a catadromous fish species that is widely distributed across major river systems in Asia, and is found in coastal waters, large rivers, and their associated water bodies in China [1,2]. Each spring, juvenile eels migrate from the sea into estuaries and ascend rivers to inhabit, grow, and fatten in various river sections and lake-connected water bodies. Mature adult eels begin their seaward spawning migration in autumn [3,4]. Under natural conditions, the gonadal development and reproductive activity of *A. japonica* depend on their physical condition and nutritional status, with nutrition derived from prey organisms in their habitats [3]. *A. japonica* is omnivorous, typically feeding on invertebrates, including various crabs and oligochaete worms, as well as various fish species. The main diet composition and feeding intensity vary across different growth stages, with the highest feeding intensity occurring in summer and autumn. After autumn, eels gradually enter the reproductive migration stage, and it is generally believed that eels do not feed during their seaward spawning migration, with their digestive organs degenerating accordingly [5,6]. Given that the nutritional status of broodstock, particularly the level of essential fatty acids, is a crucial factor affecting reproductive performance and larval quality [7,8], the nutritional reserves of eels before entering the sea are directly related to their subsequent reproductive activities after seaward migration.

For a long time, research on *A. japonica* has mainly focused on nutritional requirements during aquaculture [9,10], eel rearing [11], artificial reproduction [12,13], and larval cultivation [14,15]. In recent years, due to dam construction, water conservancy projects along rivers, environmental degradation, and overfishing, *A. japonica* resources have gradually declined [16]. To better protect *A. japonica* resources, studies have gradually been conducted on wild population resource surveys [17], migratory behavior [18,19], and spawning ecology [20,21]. However, regarding the nutritional status of wild *A. japonica*, only studies on the muscle of adult eels distributed in freshwater [22] and on the tissues of glass eels captured in offshore waters [23] have been reported. No studies have been reported on the fatty acid composition in different tissues of reproductive eels before and after their seaward migration.

The Yangtze River Estuary serves as a critical passageway for the seaward spawning migration of *A. japonica*. Previous studies have found that eels generally have gonads at stage II before entering the sea, with gonads continuously developing and maturing after seaward migration [3]. Currently, direct evidence is lacking regarding the migratory routes and distribution areas of eels from sea entry to spawning grounds, whether and where they stay in offshore waters, how long they remain there, and whether they require further feeding to replenish energy and reserve nutrients. The energy reserves and nutritional status of different tissues in eels with initially developed gonads distributed in the Yangtze River Estuary remain unknown, and there is also a lack of understanding regarding the nutritional transformation and directional enrichment among different tissues after seaward migration. Given that the distribution characteristics of fatty acids in different tissues can directly reflect the functions of these tissues and the roles of various fatty acids in energy provision and nutritional reserve [24,25], this study compared the fatty acid composition in different tissues of *A. japonica* before and after seaward migration. The aim is to reveal the functions of different tissues, elucidate the allocation and transformation patterns of different fatty acids among tissues during the reproductive migration, and thereby infer the energy allocation, fatty acid reserves, and gonadal development status of eels before and after seaward migration. This study provides fundamental data for evaluating the reproductive potential of eels after seaward migration and reveals the energy utilization and transformation mechanisms of *A. japonica* in adapting to reproductive migration, along with their potential reproductive physiological significance.

2. Materials and Methods

2.1. Animals and Sampling

In this study, *A. japonica* samples were collected from the Yangtze River Estuary waters (EW) and offshore waters (OW), respectively (Figure 1). Six female individuals from EW and 6 from OW were randomly selected for analysis. The mean body length of the samples from EW was 60.90±4.78 mm, and the mean body weight was 401.78±158.81 g; the mean body length of the samples from OW was 72.38±4.86 mm, and the mean body weight was 681.49±79.03 g.

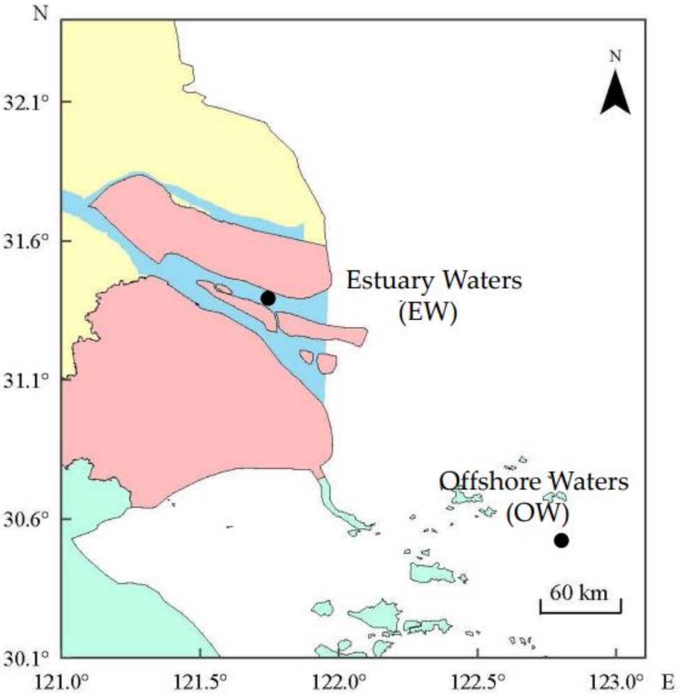

Figure 1. *A. japonica* sampling waters.

2.2. Sample Processing and Analysis

Samples collected in the field were preserved in self-sealing bags, transported back to the laboratory under refrigeration, and dissected under low-temperature conditions. Muscle, liver, and ovary tissues were taken from *A. japonica* samples collected from EW and OW, respectively. Each tissue was cut into small pieces, homogenized by grinding, and used for fatty acid determination and analysis. Fatty acids were extracted and fatty acid methyl esters (FAMEs) were prepared according to the ISO 5509 method: first, Soxhlet extraction and then, saponification, followed by esterification

doi:10.20944/preprints202605.0915.v1

and finally, extraction of FAMEs in hexane. FAMEs were subsequently analyzed by capillary gas chromatography (column: 30 m × 0.25 mm I.D., 0.5 μm film thickness; Supelco. Flame ionisation detected temperature at 210 °C; carrier gas N_2 at 1.0 ml/min; injector temperature at 210 °C; oven temperature programmed from 180 to 250 °C) using an Agilent 6890 capillary gas chromatograph. Quantitative data were calculated using the peak area ratio (% total fatty acids) [26].

2.3. Statistical Analysis

Experimental data were analyzed and processed using SPSS 25.0 statistical software. An independent samples t-test was used to compare samples from the two sources (EW and OW), and One-way ANOVA was used to test for significant differences among three tissues (muscle, liver and ovary), followed by SNK multiple comparisons. Descriptive statistics are presented as means and standard deviations (SD), and $p<0.05$ was considered statistically significant.

3. Results

3.1. Difference of Fatty Acid Composition Before and After Seaward Migration

Table 1 shows the fatty acid composition in the muscle of *A. japonica* from EW and OW. A total of 32 fatty acids were detected, including 12 SFA, 9 MUFA and 11 PUFA. The fatty acid composition in the muscle of *A. japonica* from the two sources was generally consistent, with significant differences observed only in five fatty acids (C12:0, C15:1, C18:1n9t, C22:1n9 and C20:2), PUFA and the PUFA/SFA ratio ($p<0.05$). Among the SFA, C16:0 was the most abundant in both groups, with its content being higher in *A. japonica* from EW than in those from OW. Among the MUFA, C18:1n9c was the most abundant in both groups, with its content being lower in *A. japonica* from EW than that from OW. Regarding PUFA, the predominant fatty acids in the muscle of *A. japonica* from EW were C18:2n6c and DHA, whereas in *A. japonica* from OW, they were DPA and C18:2n6c. The contents of important fatty acid combination indices, including EPA+DHA, HUFA, n3-PUFA, and n6-PUFA, were all higher in the muscle of *A. japonica* from EW than that from OW. Among different types of fatty acids, the SFA and PUFA contents in the muscle of *A. japonica* from EW were slightly higher than those from OW, while the MUFA content was lower. Among the key fatty acid ratios, the DHA/EPA, EPA/ARA and PUFA/SFA ratios in the muscle of *A. japonica* from EW were higher than those from OW, whereas the n3/n6 PUFA ratio was lower.

Table 1. Fatty acid composition in the muscles of A. japonica from EW and OW (%).

Fatty Acid	EW	OW
C10:0	0.01±0.00	0.01±0.00
C12:0	0.16±0.04^{a}	0.08±0.03^{b}
C13:0	0.08±0.04	0.05±0.02
C14:0	4.00±1.17	3.60±0.38
C15:0	0.88±0.33	0.99±0.68
C16:0	24.58±7.01	22.42±0.51
C17:0	0.78±0.21	0.81±0.08
C18:0	5.67±1.73	5.38±0.56
C20:0	0.25±0.05	0.22±0.03
C21:0	0.08±0.01	0.11±0.06
C22:0	0.11±0.03	0.10±0.07
C23:0	0.14±0.05	0.19±0.12
SFA	36.75±9.36	33.96±1.62
C14:1	0.23±0.07	0.19±0.06
C15:1	0.05±0.01^{a}	0.01±0.00^{b}
C16:1	9.81±1.59	10.13±1.85
C17:1	0.91±0.18	0.83±0.43

C18:1n9t	0.32±0.07^{a}	0.44±0.11^{b}
C18:1n9c	35.74±2.74	37.80±5.58
C20:1n9	1.79±0.56	1.58±0.36
C22:1n9	0.13±0.03^{a}	0.08±0.02^{b}
C24:1n9	0.05±0.03	0.05±0.02
MUFA	42.70±13.52	51.11±3.79
C18:2n6c	4.99±3.08	2.67±1.35
C20:2	0.91±0.26^{a}	0.60±0.14^{b}
C22:2	0.06±0.02	0.05±0.04
C18:3n6	0.14±0.12	0.10±0.06
C18:3n3(ALA)	2.11±0.41	1.70±0.60
C20:3n6	0.45±0.22	0.38±0.17
C20:3n3	0.51±0.05	0.36±0.18
C20:4n6(ARA)	1.83±0.33	2.05±0.25
C20:5n3(EPA)	2.88±0.71	2.12±1.28
C22:5n3(DPA)	2.56±0.47	2.68±0.93
C22:6n3 (DHA)	4.01±2.24	2.21±1.28
PUFA	20.36±4.26^{a}	14.93±2.43^{b}
EPA+DHA	6.89±2.18	4.34±2.04
HUFA	14.39±1.75	11.60±2.79
n3-PUFA	11.98±1.81	9.08±2.72
n6-PUFA	7.42±3.20	5.20±1.35
DHA/EPA	1.52±1.00	1.17±0.30
EPA/ARA	1.62±0.46	1.05±0.62
n3/n6	1.83±0.74	1.90±0.92
PUFA/SFA	0.56±0.03^{a}	0.44±0.06^{b}

Values are expressed as mean value ± SD; Values in the same row with different superscript letters mean significant difference ($p<0.05$). 3. Tables 2 and 3 are the same.

Table 2 shows the fatty acid composition in the liver of *A. japonica* from EW and OW. A total of 30 fatty acids were detected in the liver, including 12 SFA, 7 MUFA and 11 PUFA. The fatty acid composition in the liver of *A. japonica* from the two sources was generally consistent, with significant differences observed only in four fatty acids (C16:0, C18:1n9t, C20:1n9 and C22:1n9), as well as in SFA content and the DHA/EPA ratio ($p<0.05$). Among the SFA, C16:0 was the most abundant in both groups, with its content being significantly lower in the liver of *A. japonica* from EW than in that from OW. Among the MUFA, C18:1n9c was the most abundant in both groups, with its content being higher in the liver of *A. japonica* from EW than that from OW. Regarding PUFA, the predominant fatty acids in the liver of *A. japonica* from EW were DHA and C18:2n6c, whereas that from OW were ARA and DHA. The contents of important fatty acid combination indices, including EPA+DHA, HUFA, and n3-PUFA, were all higher in the liver of *A. japonica* from EW than that from OW, while the n6-PUFA content was lower. Among different types of fatty acids, the SFA content in the liver of *A. japonica* from EW was lower than that from OW, while the MUFA and PUFA contents were higher. Among the key fatty acid ratios, the DHA/EPA, EPA/ARA, n3/n6 PUFA and PUFA/SFA ratios in the liver of *A. japonica* from EW were all higher than those from OW.

Table 2. Fatty acid composition in the liver of *A. japonica* from OW and EW (%).

Fatty Acid	**EW**	**OW**
C6:0	0.02±0.03	0.01±0.01
C11:0	0.03±0.01	0.05±0.06
C12:0	0.07±0.02	0.06±0.04
C13:0	0.05±0.03	0.03±0.02
C14:0	2.72±0.84	2.39±0.74

C15:0	0.40±0.22	0.75±0.46
C16:0	21.35±2.67^{a}	25.21±2.88^{b}
C17:0	0.48±0.22	0.73±0.17
C18:0	7.13±1.88	7.09±1.65
C20:0	0.22±0.09	0.22±0.05
C21:0	0.05±0.02	0.09±0.06
C22:0	0.05±0.02	0.10±0.07
SFA	32.59±2.86^{a}	37.04±3.94^{b}
C14:1	0.12±0.04	0.10±0.05
C16:1	6.02±1.87	7.56±1.71
C17:1	0.48±0.19	0.75±0.43
C18:1n9t	0.30±0.05^{a}	0.49±0.09^{b}
C18:1n9c	36.81±7.36	33.79±3.05
C20:1n9	1.80±0.41^{a}	1.23±0.26^{b}
C22:1n9	0.15±0.05^{a}	0.09±0.03^{b}
MUFA	45.70±8.43	44.03±4.56
C18:2n6c	4.27±1.86	2.48±1.06
C20:2	0.71±0.20	0.75±0.20
C22:2	0.07±0.04	0.05±0.02
C18:3n6	0.09±0.05	0.06±0.03
C18:3n3(ALA)	0.90±0.51	1.17±0.26
C20:3n6	0.53±0.26	0.37±0.09
C20:3n3	0.24±0.17	0.35±0.10
C20:4n6(ARA)	2.72±2.05	4.32±1.72
C20:5n3(EPA)	1.76±0.93	2.15±0.90
C22:5n3(DPA)	1.79±0.46	2.93±1.93
C22:6n3 (DHA)	8.88±7.20	4.32±2.23
PUFA	21.44±8.51	18.93±5.61
EPA+DHA	10.64±8.01	6.47±3.02
HUFA	16.42±9.51	15.67±6.46
n3-PUFA	13.53±7.89	10.92±4.98
n6-PUFA	7.16±2.12	7.23±1.32
DHA/EPA	5.03±2.29^{a}	2.02±0.49^{b}
EPA/ARA	0.70±0.26	0.53±0.23
n3/n6	1.97±1.18	1.51±0.60
PUFA/SFA	0.66±0.27	0.53±0.20

Table 3 shows the fatty acid composition in the ovary of *A. japonica* from EW and OW. A total of 24 fatty acids were detected in the ovary, including 8 SFA, 7 MUFA and 9 PUFA. The fatty acid composition in the ovary of *A. japonica* from the two sources was generally consistent, with significant differences observed only in seven fatty acids (C14:0, C16:0, C18:0, C20:0, C20:1n9, C20:2 and DPA), as well as HUFA, n3-PUFA and the n3/n6 PUFA ratio (p<0.05). Among the SFA, C16:0 was the most abundant in the ovary of *A. japonica* from EW, whereas C18:0 was the most abundant in that from OW. Among the MUFA, C18:1n9c was the most abundant in both groups, with its content being lower in the ovary of *A. japonica* from EW than that from OW. Regarding PUFA, the predominant fatty acids in the ovary of *A. japonica* from EW were C18:2n6c and DHA, whereas in that from OW were DHA and ARA. The contents of important fatty acid combination indices, including EPA+DHA, HUFA and n3-PUFA, were all lower in the ovary of *A. japonica* from EW than that from OW, while the n6-PUFA content was higher. Among different types of fatty acids, the SFA content in the ovary of *A. japonica* from EW was higher than that from OW, while the MUFA and PUFA contents were lower. Among key fatty acid ratios, the DHA/EPA ratio in the ovary of *A. japonica* from EW was higher than that from OW, whereas the EPA/ARA, n3/n6 PUFA and PUFA/SFA ratios were all lower.

Table 3. Fatty acid composition in the ovary of *A. japonica* from EW and OW (%).

Fatty Acid	EW	OW
C12:0	0.21±0.24	0.02±0.01
C13:0	0.56±0.50	0.03±0.03
C14:0	2.83±0.35^{a}	1.24±0.45^{b}
C15:0	1.11±0.87	0.49±0.30
C16:0	24.38±3.93^{a}	11.25±2.91^{b}
C17:0	0.99±0.75	0.41±0.09
C18:0	8.53±2.93^{a}	15.40±3.93^{b}
C20:0	0.31±0.21^{a}	3.33±0.14^{b}
SFA	39.63±9.96	31.62±1.09
C14:1	0.52±0.45	0.06±0.01
C15:1	0.32±0.43	0.61±0.22
C16:1	6.08±1.34	8.52±1.65
C17:1	0.96±0.04	1.30±0.34
C18:1n9t	0.40±0.23	0.49±0.10
C18:1n9c	31.58±8.74	34.34±4.18
C20:1n9	1.69±0.26^{a}	0.99±0.31^{b}
MUFA	40.75±8.42	46.23±2.28
C18:2n6c	4.69±2.76	2.50±1.01
C20:2	0.84±0.08^{a}	0.57±0.15^{b}
C18:3n6	0.19±0.04	0.16±0.13
C18:3n3 (ALA)	1.29±0.29	2.01±0.63
C20:3n6	0.53±0.16	0.50±0.17
C20:4n6(ARA)	4.42±2.50	3.88±0.84
C20:5n3(EPA)	1.87±0.29	2.73±0.66
C22:5n3(DPA)	1.50±0.09^{a}	3.87±1.29^{b}
C22:6n3 (DHA)	4.54±2.31	5.49±1.92
PUFA	19.13±3.98	22.16±3.10
EPA+DHA	6.41±2.33	8.22±2.48
HUFA	13.60±1.77^{a}	18.99±3.24^{b}
n3-PUFA	8.70±2.55^{a}	14.45±3.55^{b}
n6-PUFA	9.59±2.30	7.09±1.78
DHA/EPA	2.46±1.32	2.00±0.34
EPA/ARA	0.54±0.30	0.76±0.34
n3/n6	0.93±0.26^{a}	2.22±1.07^{b}
PUFA/SFA	0.52±0.24	0.70±0.12

3.2. *Difference of Fatty Acid Composition Among Different Tissues*

Figure 2 presents the composition of different types of fatty acids in different tissues. For *A. japonica* from EW, SFA content was highest in the ovary and lowest in the liver, with no significant differences among tissues ($p>0.05$). In contrast, for that from OW, SFA content was highest in the liver and lowest in the ovary, with the content in the ovary being significantly lower than that in the liver ($p<0.05$). For MUFA, in *A. japonica* from EW, the content was highest in the liver and lowest in the ovary, with no significant differences among tissues ($p>0.05$); whereas in that from OW, MUFA content was highest in the muscle and lowest in the liver, with the content in the muscle being significantly higher than that in the liver and ovary ($p<0.05$). Regarding PUFA, in *A. japonica* from EW, the content was highest in the liver and lowest in the ovary, with no significant differences among tissues ($p>0.05$); while in that from OW, PUFA content was highest in the ovary and lowest in the muscle, with the content in the muscle being significantly lower than that in the ovary ($p<0.05$).

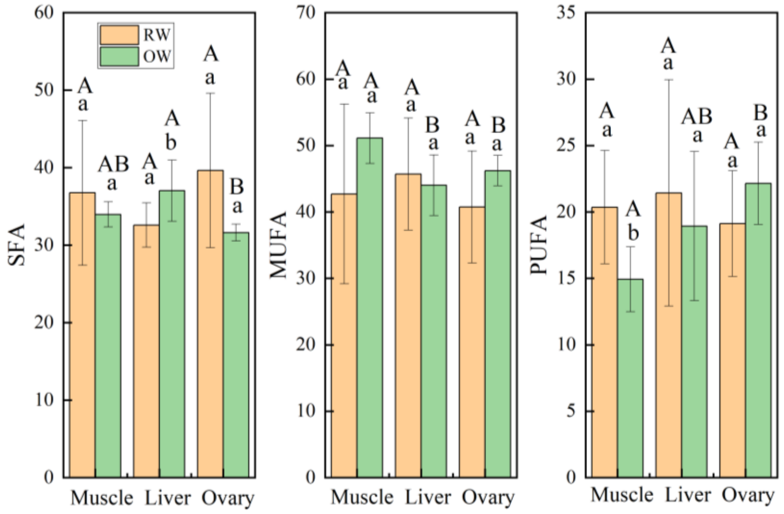

Figure 2. Comparison of different types of fatty acids in different tissues of *A. japonica* from EW and OW. EW: estuary waters; OW: offshore waters. Different small letters (a, b) denote significant differences between samples from different waters of EW and OW. Different capital letters (A, B, C) denote significant differences among different tissues of muscle, liver and ovary. Figures from 3 to 6 are the same.

Figure 3 presents the main fatty acids contents of SFA and MUFA in different tissues. In *A. japonica* from EW, C16:0 content was highest in the muscle and lowest in the liver, with no significant differences among tissues (p>0.05). In contrast, in that from OW, C16:0 content was highest in the liver and lowest in the ovary, with the content in the ovary being significantly lower than that in the liver and muscle (p<0.05). For C18:0, the content was highest in the ovary in both groups, with the distribution across tissues showing the highest level in the ovary and the lowest in the muscle. Moreover, the C18:0 content in the ovary of *A. japonica* from OW was significantly higher than that from EW and in other tissues (p<0.05). For C16:1, the content was highest in the muscle in both groups, with the distribution across tissues showing the highest level in the muscle and the lowest in the liver. Additionally, the C16:1 content in the muscle of *A. japonica* from EW was significantly higher than that in the liver and ovary (p<0.05). For C18:1n9c, in *A. japonica* from EW, the content was highest in the liver and lowest in the ovary, whereas in that from OW, the content was highest in the muscle and lowest in the liver. No significant differences were observed among tissues in either group (p>0.05).

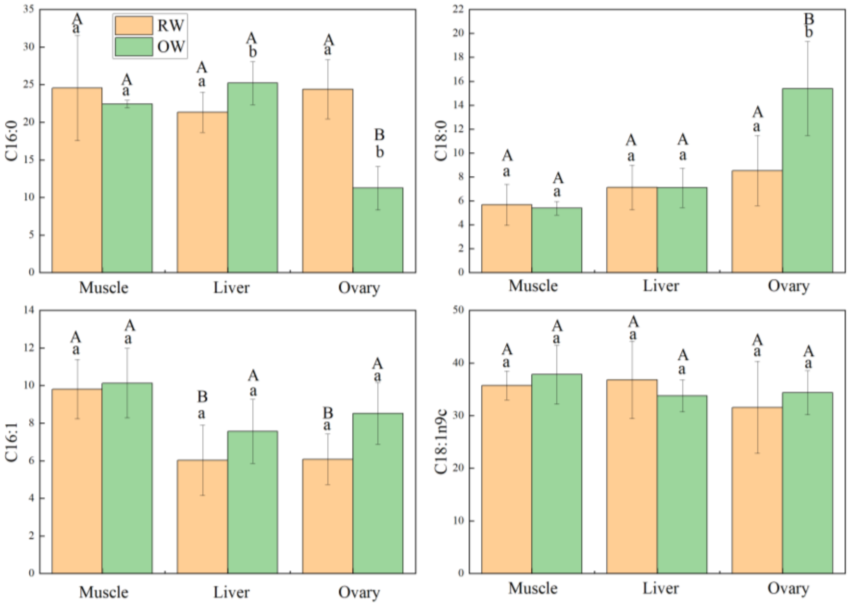

Figure 3. Comparison the main fatty acids contents of SFA and MUFA in different tissues of *A. japonica* from EW and OW.

Figure 4 presents the main fatty acids contents of PUFA in different tissues. In *A. japonica* from EW, ARA content was highest in the ovary and lowest in the muscle, with no significant differences among tissues (*p*>0.05). In contrast, in that from OW, ARA content was highest in the liver and lowest in the muscle, with the content in the muscle being significantly lower than that in the liver and ovary (*p*<0.05). For EPA, in *A. japonica* from EW, the content was highest in the muscle and lowest in the liver, whereas in that from OW, EPA content was highest in the ovary and lowest in the muscle. No significant differences in EPA content were observed among tissues in either group (*p*>0.05). For DPA, in *A. japonica* from EW, the content was highest in the muscle and lowest in the ovary, with the content in the muscle being significantly higher than that in the liver and ovary (*p*<0.05). In that from OW, DPA content was highest in the ovary and lowest in the muscle, and the DPA content in the ovary of *A. japonica* from OW was significantly higher than that from EW (*p*<0.05). For DHA, in *A. japonica* from EW, the content was highest in the liver and lowest in the muscle, whereas in that from OW, DHA content was highest in the ovary and lowest in the muscle, with the content in the ovary being significantly higher than that in the muscle (*p*<0.05).

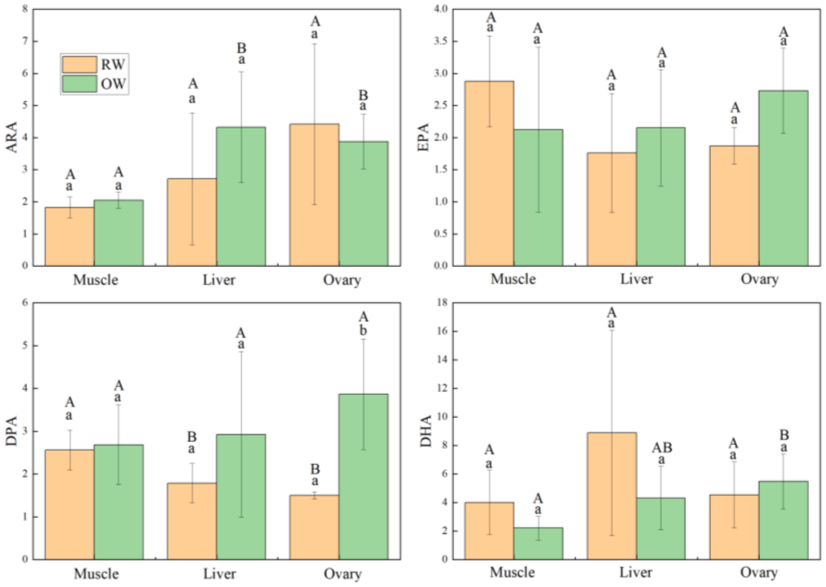

Figure 4. Comparison the main fatty acids contents of PUFA in different tissues of *A. japonica* from EW and OW.

Figure 5 presents the content of the important fatty acid combinations in different tissues. In *A. japonica* from EW, DHA+EPA content was highest in the liver and lowest in the ovary, with no significant differences among tissues (*p*>0.05). In contrast, in that from OW, DHA+EPA content was highest in the ovary and lowest in the muscle, with the content in the muscle being significantly lower than that in the ovary (*p*<0.05). For HUFA, in *A. japonica* from EW, the content was highest in the liver and lowest in the ovary, with no significant differences among tissues (*p*>0.05), whereas in that from OW, HUFA content was highest in the ovary and lowest in the muscle, with the content in the muscle being significantly lower than that in the ovary (*p*<0.05). For n3-PUFA, in *A. japonica* from EW, the content was highest in the liver and lowest in the ovary, with no significant differences among tissues (*p*>0.05), while in that from OW, n3-PUFA content was highest in the ovary and lowest in the muscle, and the n3-PUFA content in the ovary of *A. japonica* from OW was significantly higher than that from EW (*p*<0.05). For n6-PUFA, in *A. japonica* from EW, the content was highest in the ovary and lowest in the liver, whereas in that from OW, n6-PUFA content was highest in the liver and lowest in the muscle. No significant differences in n6-PUFA content were observed among tissues in either group (p>0.05).

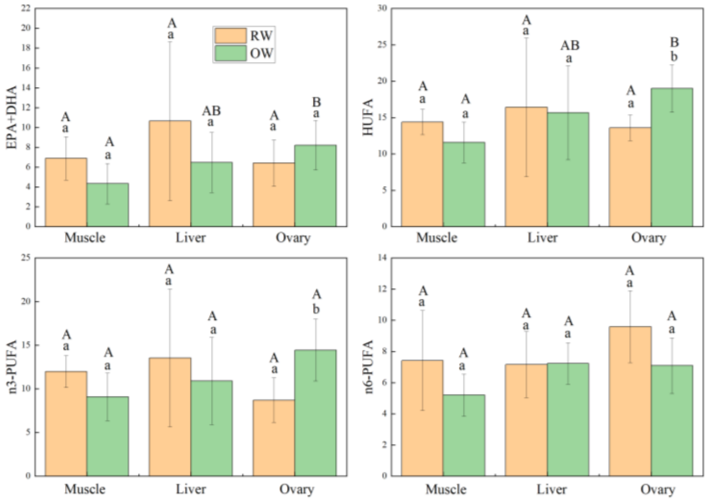

Figure 5. Comparison the content of the important fatty acid combinations in different tissues of *A. japonica* from EW and OW.

Figure 6 presents the key fatty acid ratios in different tissues. The DHA/EPA in both groups of *A. japonica* was highest in the liver and lowest in the muscle. In *A. japonica* from EW, the DHA/EPA in the liver was significantly higher than that in the ovary and muscle (p<0.05), whereas in that from OW, the DHA/EPA in the muscle was significantly lower than that in the liver and ovary (p<0.05). The EPA/ARA in both groups was highest in the muscle. In *A. japonica* from EW, the EPA/ARA in the muscle was significantly higher than that in the liver and ovary (p<0.05), while in that from OW, the EPA/ARA was lowest in the liver (p<0.05). For the n3/n6 PUFA, in *A. japonica* from EW, it was highest in the liver and lowest in the ovary, with no significant differences among tissues (p>0.05); whereas in that from OW, the n3/n6 PUFA was highest in the ovary and lowest in the liver, and the ratio in the ovary of *A. japonica* from OW was significantly higher than from EW (p<0.05). For the PUFA/SFA, in *A. japonica* from EW, it was highest in the liver and lowest in the ovary, with no significant differences among tissues (p>0.05); whereas in that from OW, the PUFA/SFA was highest in the ovary and lowest in the muscle, and the ratio in the ovary was significantly higher than that in the liver and muscle (p<0.05).

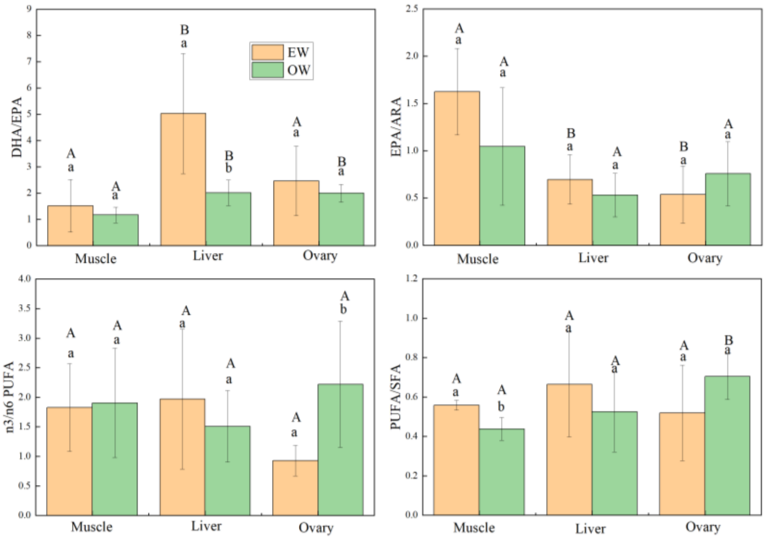

Figure 6. Comparison the key fatty acids ratios in different tissues of *A. japonica* from EW and OW.

4. Discussion

4.1. Fatty Acid Composition Characteristics in Different Tissues and the Manifestation of Tissue-Specific Functions

The distribution of fatty acids in different tissues of fish is not uniform but is rather tailored to the functional demands of each tissue, exhibiting a high degree of regularity. The content and ratios of different fatty acids can reflect the physiological functions of different tissues, such as energy reserve, nutrient metabolism, and accumulation [27,28]. Muscles is the primary tissue for storing energy, and SFA and MUFA are considered the preferred fatty acids for oxidation to provide energy [29]. Studies have shown that, in both freshwater and marine fish, the most abundant fatty acids in muscle are typically C16:0 and C18:1n9c [30]. The fatty acid composition of the muscle for eels from both EW and OW in this study followed this pattern, indicating the fundamental roles of these two fatty acids in energy provision and maintenance of muscle cell membrane function. Comparing different tissues, the C16:0 content was highest in the muscle of eels from EW, while the C18:1n9c content was highest in the muscle of eels from OW, suggesting that before seaward migration, the eels in Yangtze River Estuary primarily use muscle for energy storage, whereas after entering the sea, the offshore eels undergo adaptive changes such as osmoregulation, exhibiting a stronger role in maintaining cell membrane function. Additionally, during the early stages of gonadal development, muscle also stores considerable amounts of PUFA such as EPA+DHA and n3-PUFA. Studies have found that in the muscle of *Illex argentinus*, DHA is the most abundant PUFA, followed by EPA [31], which is consistent with the distribution of DHA and EPA contents in the muscle of the eels from EW. This indicates that before seaward migration, Yangtze River Estuary eels store substantial amounts of DHA and EPA in their muscle. Research on European whitefish has shown that DHA and EPA contents in muscle decrease by approximately 60% from late summer to the spawning period [32]. In this study, the DHA/EPA in the muscle of offshore eels decreased by 23% compared with that of Yangtze River Estuary eels, indicating that after entering the sea, the key fatty acids such as DHA stored in the muscle are extensively mobilized during the reproductive period to support gonadal development.

The liver is the central organ for lipid metabolism in fish, responsible for the synthesis, transformation and distribution of fatty acids to other tissues [33]. Therefore, its fatty acid composition can better reflect the metabolic status and nutritional level of fish. The MUFA content in the liver of eels from EW was the highest among all tissues and significantly higher than that in muscle and gonad, suggesting that MUFA in the liver serve not only as energy storage forms but also as substrates for active metabolism and synthesis. The liver is not only rich in MUFA such as C18:1n9c, but rich in substrates of ARA for synthesis and metabolism. During fish reproductive migration, the ARA metabolic pathway in the liver is significantly activated, participating in environmental adaptation such as osmoregulation and gonadal development [34]. In this study, the ARA content in the liver of eels from OW was higher than that from EW, indicating that after seaward migration, the liver of eels undergoes active ARA metabolism, which can supply more unsaturated fatty acids to the ovary. Compared with muscle, the liver often contains a higher proportion of PUFA, particularly EPA and DHA [35]. In this study, the contents of ARA, DHA, EPA+DHA, HUFA, n3-PUFA and PUFA in the liver of both groups of eels were higher than those in their respective muscles, with DHA content in the liver being 2.21 and 1.95 times that in the muscle, respectively. As the processing and reserve center for DHA, the liver often contains the highest DHA content, exhibiting tissue specificity [36,37]. In *Tridentiger trigonocephalus*, the DHA/EPA ratio in the liver was 1.75, higher than that in the ovary and muscle [38]. In *Paralichthys olivaceus*, the DHA/EPA ratio in the liver of stage III to IV gonadal development of broodstock was significantly higher than that in stage V broodstock, suggesting that the liver reserves DHA in the early stages of gonadal development to provide sufficient DHA to the gonads later [35]. In this study, the DHA/EPA ratio in the liver of eels from EW was the highest and significantly higher than that from OW, which is consistent with the characteristics of gonadal development during the migration to the sea of the eels. As seaward migration and gonadal maturation proceed, DHA is gradually transferred to the ovary, resulting in a gradual decrease of this ratio in the liver. The n3/n6 PUFA ratio can reflect tissue functional orientation and metabolic homeostasis [39,40]. In *Tridentiger trigonocephalus* distributed in the Yangtze River Estuary, the n3/n6 PUFA ratio in the liver reached 8.05, higher than that in the ovary (7.00) and muscle (6.44) [38], which is similar to the pattern in the liver of Yangtze River Estuary eels, where this ratio was highest. In contrast, in *Pampus argenteus* distributed in offshore waters, the n3/n6 PUFA in the gonad was significantly higher than that in the liver and muscle [41], which is similar to the pattern in the liver of offshore eels. The differences in the n3/n6 PUFA among different tissues at different migratory stages of eels are closely related to the metabolic functions of tissues during different migration processes, reflecting the physiological differences of eels at different gonadal development stages. The Yangtze River estuary eels at the early stage of gonadal development, the n3/n6 PUFA was highest in the liver. As gonads develop, the n3/n6 PUFA in the ovary gradually increases, and that in the ovary of offshore eels was significantly higher than that in Yangtze River Estuary eels. The high ratio of n3/n6 PUFA in the liver of Yangtze River Estuary eels is related to its function as a metabolic center for n3-PUFA, while the high ratio in the gonad of offshore eels reflects their priority accumulation demand for n3-PUFA.

The fatty acid composition of the gonad has distinct characteristics related to reproductive demands. In the process of reproduction, fish preferentially transport and store the most critical HUFA in the developing gonad [42]. In *Takifugu flavidus*, the gonad has the highest relative PUFA content, significantly higher than that in the liver and muscle [43]. Studies on *Siganus guttatus* have also reached the same conclusion, with the highest contents of PUFA, n3-PUFA, and n6-PUFA in the ovary [44]. Research on wild *Paralichthys olivaceus* broodstock found that the fatty acid composition of stage V eggs contains a large amount of n3-HUFA, and the proportions of EPA, DPA, and DHA were also significantly higher than those in muscle and liver at the same developmental stage [35]. This clearly indicates that during the critical period of gonadal maturation, fish comprehensively regulate fatty acids across tissues to transfer HUFA into the eggs. In this study, the contents of EPA, DPA, DHA, EPA+DHA, HUFA, n3-PUFA and PUFA in the ovary of eels from OW were all the highest and higher than those from EW, indicating that with the seaward migration process and continuous gonadal development, the ovary has begun to directionally accumulate various important fatty acids required for ovarian development. The DHA/EPA can reflect the tissue’s demand for DHA [45]. In *Tridentiger trigonocephalus*, the DHA/EPA in the ovary was 1.59, higher than that in muscle [38]. This characteristic is even more extreme in bluefin tuna, where the DHA/EPA ratio in the ovary reaches 4.5. This indicates that during gonadal development, fish preferentially transport and retain DHA in reproductive tissues to support the nervous system development of offspring [46]. In this study, the DHA/EPA in the ovary of both groups of eels was higher than that in muscle, being 1.6 and 1.7 times that of the latter, respectively, indicating that eels have begun to retain DHA in the gonad at the onset of reproduction, with more pronounced DHA enrichment in the ovary of offshore eels after sea entry. The n3/n6 PUFA in the gonad is relatively high, and is significantly higher than that in the liver and muscle [38,41], indicating that developing germ cells have a specific demand for n3-PUFA. A relatively low n6-PUFA level helps maintain an appropriate n3/n6 PUFA balance, ensuring reproductive success. In this study, the n3/n6 ratio in the ovary of offshore eels was the highest, higher than that in muscle and liver, and significantly higher than that in the ovary of Yangtze River Estuary eels, indicating that after migrating into the sea, offshore eels have gradually accumulated n3-PUFA in their ovaries.

4.2. Fatty Acids Distribution Pattern in Different Tissues and Its Roles in Energy Allocation and Gonadal Development

Different fatty acids play distinct and well-defined roles in the energy supply and reproductive processes of fish [8,47]. SFA and MUFA, as the primary energy-providing fatty acids, are mainly stored in the form of triglycerides in the muscle, liver, and peritoneal fat of adult fish. When fish face periods of high energy demand, such as starvation, migration or reproduction, these fatty acids are

doi:10.20944/preprints202605.0915.v1

preferentially mobilized to provide energy through β-oxidation [43]. Gonadal development requires substantial energy consumption, and major fatty acids such as C16:0 and C18:1n9c can provide large amounts of energy and carbon skeletons for the growth and maturation of oocytes [30]. Particularly during the early life stages of fish, they rely entirely on nutrients from the yolk for energy. At this stage, C16:0, C18:0 and C18:1n9c were used preferentially to provide necessary ATP for embryo development and larval hatching [48]. In this study, compared with offshore eels, Yangtze River Estuary eels had a higher C16:0 content in the ovary, while offshore eels had higher contents of C18:0, C16:1 and C18:1n9c in the ovary. This indicates that the ovary of offshore eels contains a greater variety and higher contents of major saturated and monounsaturated fatty acids, which can provide energy security for later embryonic development and larval hatching. The PUFA/SFA reflects the energy allocation strategy of fish. A high PUFA/SFA gives the oocyte membrane optimal fluidity and functionality, which is the basis for cell division, signal transduction, and material exchange. At the same time, these stored PUFA provide essential structural components and energy sources for early embryonic development [46]. The low PUFA/SFA in muscle and liver reflects their functional roles as energy storage and metabolic organs, while the high PUFA/SFA in the gonad reveals the nutritional allocation strategy of fish during the critical life process of reproduction, preferentially transferring precious PUFA to the gonad while retaining more SFA in tissues such as muscle and liver for basal metabolism [38,44,49]. During the embryonic development of *Takifugu flavidus*, SFA and MUFA are preferentially used as energy sources, while PUFA such as DHA are highly retained for constructing the larval nervous system and cell membranes [48]. This indicates that between energy supply and structural construction, fish prioritize the latter. In this study, the PUFA/SFA in the muscle and liver of offshore eels was relatively low, while it was highest in the ovary, which is more consistent with the energy allocation and reproductive nutritional demands of eels after sea entry, indicating that the ovary of eels has stored a certain amount of PUFA nutrients after migrating into the sea.

The PUFA content in the fish gonad is usually significantly higher than that in tissues such as muscle and liver. This directional transfer of nutrients required for later oocyte development to the ovary is termed tissue-specific accumulation, which is the result of fish actively and preferentially transporting and storing valuable PUFA such as ARA, EPA, and DHA in the gonad to ensure successful offspring development [30]. Among these, ARA is a precursor for the synthesis of eicosanoids such as prostaglandins, directly participating in the regulation of reproductive processes such as ovulation and spawning [8]. Studies on *Cynoglossus semilaevis* have found that the dietary ARA level significantly affects the synthesis of estradiol (E2) in females and testosterone (T) in males, and this regulation is achieved by modulating the expression of key genes such as steroidogenic acute regulatory protein (StAR) and 3β-hydroxysteroid dehydrogenase (3β-HSD) in the gonad. This explains why the ARA pathway is significantly enriched in fish during reproductive migration [50]. A systematic study on *Plectorhynchus cinctus* broodstock showed that there is an optimal dosage range of dietary n3-HUFA content for reproductive performance. When the dietary n3-HUFA content was 2.40% and 3.70%, indicators such as spawning amount, egg fertilization rate, oil globule diameter, larval survival rate, and larval body length at first feeding performed best; whereas insufficient (1.12%) or excessive (5.85%) n3-HUFA content reduced spawning performance and sex steroid hormone (17β-estradiol and testosterone) levels [51]. n3-HUFA affect hormone synthesis and egg quality by regulating the production of sex steroid hormones, the sensitivity of isolated ovarian follicles to chorionic gonadotropin, and the fatty acid composition of eggs and larvae [52]. It is evident that n3-PUFA, particularly DHA, are irreplaceable in physiological functions, and the ovary has a strong selective retention effect on them.

The DHA/EPA ratio is also crucial for reproductive success [45]. Studies on the artificial reproduction of *A. japonica* found that the gonadosomatic index (GSI) showed the strongest negative correlation with the DHA content in muscle, indicating that during gonadal development, DHA stored in muscle is extensively and preferentially mobilized and transferred to the gonad [53]. In this study, the distribution of DHA content in different tissues of both Yangtze River Estuary and offshore eels followed a similar pattern. Before seaward migration, Yangtze River Estuary eels had the lowest DHA content in muscle and the highest in the liver, indicating that DHA in muscle is being continuously consumed and transferred through the liver before sea entry. After sea entry, offshore eels had the highest DHA content in the ovary and the lowest in muscle, indicating that DHA from muscle has been gradually transferred and enriched in the ovary. During early fish development, fish actively regulate the levels of ARA, EPA, and DHA, indicating that they play irreplaceable roles in early growth and sexual maturation [8]. DHA is an essential fatty acid for the development of the fish brain and retina [54]. A high DHA/EPA ratio ensures the development of the larval brain and retina, which is directly related to larval survival rate, growth rate, and predatory ability [55]. Therefore, during gonadal development, fish regulate the DHA/EPA to ensure that more DHA is transferred to the ovary, providing nutritional support for later larval development [56]. For example, the high ratio of 4.5 in the ovary of Atlantic bluefin tuna is a manifestation of this preferential retention [46]. In *Paralichthys olivaceus* broodstock, the DHA/EPA ratio in the liver of stage III–IV broodstock was significantly higher than that in stage V broodstock, while the EPA/ARA in the eggs was significantly lower than that in stage V broodstock [35]. This result is similar to the situation in the ovary of Yangtze River Estuary and offshore eels. The DHA/EPA ratio in the liver of Yangtze River Estuary eels at the early stage of gonadal development was significantly higher than that in offshore eels, while the EPA/ARA ratio in the ovary was significantly lower than that in offshore eels. These changes in ratios reveal that during different stages of gonadal development, fish have dynamic adjustments in their demands for DHA, EPA, and ARA. The extremely low EPA/ARA ratio in the gonad is to ensure sufficient ARA for the synthesis of eicosanoids such as prostaglandins, as excessive EPA can inhibit the conversion of ARA into active hormones [49]. Therefore, maintaining an appropriate EPA/ARA ratio is crucial for ensuring normal ovulation and improving fertilization and hatching rates [8]. The ratio of n3/n6 PUFA fundamentally affects the quality of eggs and larvae [57]. The different requirements of fish for the compositional ratios of n3-PUFA and n6-PUFA are essentially determined by the competitive relationships of DHA and EPA, as well as EPA and ARA, in metabolism and enzyme actions [58]. An appropriate n3/n6 ratio is crucial for maintaining normal reproductive cycles, stable sex hormone synthesis, and the healthy development of eggs and sperm [57]. The n3/n6 PUFA was highest in the liver of Yangtze River Estuary eels, while it was highest in the ovary of offshore eels, reflecting the characteristics of the n3/n6 ratio in the liver and ovary before and after seaward migration, respectively, which is closely related to the migration process.

In this study, ARA content was highest in the ovary of Yangtze River Estuary eels, EPA content was highest in muscle, DHA content was highest in the liver, while HUFA, n3-PUFA, and PUFA contents were highest in the ovary of offshore eels. Comparing Yangtze River Estuary and offshore eels, ARA content in the ovary of Yangtze River Estuary eels was higher than that in offshore eels, while the contents of PUFAs such as EPA, DPA, and DHA in the ovary of offshore eels were higher than those in Yangtze River Estuary eels. The DHA/EPA and n3/n6 ratios in the ovary of offshore eels were significantly higher than those in Yangtze River Estuary eels. It is evident that before seaward migration, the ovary of Yangtze River Estuary eels mainly reserves precursors such as ARA in preparation for initiating seaward migration, whereas after entering the sea, the gonad of offshore eels has gradually accumulated PUFAs such as EPA, DPA, and DHA required for egg and embryonic development. These eels are in the process of gonadal maturation, and the PUFA nutrients (EPA, DPA, and DHA) stored in their gonads can support ovarian maturation and subsequent larval development.

5. Conclusion

In this study, *A. japonica* from the Yangtze River Estuary and offshore waters were selected as research subjects. The fatty acid composition in the muscle, liver, and ovary of eels from both sites was determined using national standard methods, and the differences in corresponding components among different tissues were comparatively analyzed. The results showed that, comparing different tissues, EPA; DHA and EPA+DHA; ARA, n6-PUFA, and SFA were highest in the muscle, liver, and ovary of Yangtze River Estuary eels, respectively; while C16:1, C18:1n9c, and MUFA; C16:0; C18:0, DPA, HUFA, n3-PUFA and PUFA were highest in the muscle, liver, and ovary of offshore eels, respectively. The DHA/EPA was highest in the liver of Yangtze River Estuary eels, while the n3/n6 PUFA and PUFA/SFA were highest in the ovary of offshore eels. The distribution of different fatty acids in different tissues exhibited tissue specificity, which is related to the functions of different tissues. Comparing Yangtze River Estuary and offshore eels, the contents of C16:0, ARA, n6-PUFA, SFA, and the DHA/EPA ratio in the ovary of Yangtze River Estuary eels were higher than those in offshore eels, while the contents of C18:0, C16:1, C18:1n9c, EPA, DPA, DHA, EPA+DHA, HUFA, n3-PUFA, MUFA, PUFA, and the n3/n6 PUFA and PUFA/SFA in the ovary of offshore eels were higher than those in Yangtze River Estuary eels. The ovary of Yangtze River Estuary eels mainly contained energy-providing fatty acids and precursor substances, whereas the ovary of offshore eels had preliminarily accumulated fatty acid nutrients required for egg and embryonic development. The distribution patterns of corresponding components among different tissues are closely related to the migration process of eels before and after seaward migration. In particular, the abundant PUFA nutrients accumulated in the ovary can promote gonadal development after sea entry and provide sufficient nutrition for later egg and larval development. The selective accumulation of polyunsaturated fatty acid nutrients in the ovary is an adaptation to the environmental adaptation and nutritional demands of gonadal development after eels enter the sea. In this study, the selected eels were individuals before and after seaward migration, respectively. The findings can lay a foundation for further elucidating the relationship between ovarian fatty acid composition and gonadal development status at different migratory stages of *A. japonica*. The relevant data can enrich the knowledge of the reproductive ecology and biology of *A. japonica*, and provide theoretical guidance for the selection of wild broodstock and the protection of reproductive populations.

Author Contributions: Conceptualization and writing—original draft, C.S. D.S. and C.Y.; writing—review and editing, C.S., D.S., C.Y. and F.Z.; investigation and methodology, Y.L., H.L. and Z.Y.; resources and funding acquisition, C.S. and F.Z.; validation and supervision, J.R. and S.W. All authors have read and agreed to the published version of the manuscript.

Funding: This research was supported by the National Key R&D Program Key Special Project (2024YFD2401000), Shanghai Science and Technology Innovation Action Plan (24N12800500), Fuzhou Science and Technology Program Project (2025-ZD-012) and Central Public-interest Scientific Institution Basal Research Fund, CAFS (2023TD14).

Institutional Review Board Statement: The animal study protocol was approved by the Animal Care Welfare Committee of Chinese Academy of Fishery Sciences East China Sea Fisheries Research Institute (protocol code ECSFRI25).

Data Availability Statement: The data presented in this study are available in the article. Further information is available upon request from the corresponding author.

Conflicts of Interest: The authors declare no conflict of interest.

References

1. Tesch, F.W. *The Eel*; Wiley-Blackwell: Oxford, UK, 2003; PP. 1–436.
2. Song, C.; Yang, C.; Li, Y.; Song, D.; Huang, X.; Wang, S.; Zhao, F.; Tao, H. Geographical Traceability of *Anguilla japonica* from Different Habitats Successfully Achieved Using Muscle Elemental Fingerprint Analysis. *Fishes* **2026**, 11, 68.
3. Zhuang, P.; Zhang, T.; Li, S.F.; Ni, Y.; Wang, Y.H.; Deng, S.M.; Zhang, L.Z.; Ling, J.Z.; Hu, F.; Yang, G.; et al. *Fishes of the Yangtze Estuary*; China Agriculture Press: Beijing, China, 2018; pp. 91–99.
4. Tsukamoto, K.; Kuroki, M.; Watanabe, S. *Eel Science*; Springer: Singapore, 2023; pp. 37–50.

5. Huang, X.; Liu, J.; Song, C.; Liu, R.; Wang, S.; Zhang, T.; Yang, G.; Zhao, F. Ultrastructural Evidence for Dual Sperm Morphotypes in Hormone-Induced Japanese Eel (*Anguilla japonica*): Implications for Sperm Maturation. *Biology* **2026**, 15, 437.
6. Aida, K.; Tsukamoto, K.; Yamauchi, K. *Eel Biology*; Springer: Tokyo, Japan, 2003; pp. 55–58.
7. Guo, H.Y.; Zheng, L.; Ye, Y.M.; Tang, W.Q.; Zhang, Y. Zhang, X.G. Silvering indexes for spawning migratory population of the Japanese eel *Anguilla japonica* in the Yangtze River estuary, China. *Acta Hydrobiol. Sin.* **2019**,43,133–141.
8. Furuita, H.; Hori, K.; Suzuki; Sugita, T.; Yamamoto, T. Effect of n-3 and n-6 fatty acids in broodstock diet on reproduction and fatty acid composition of broodstock and eggs in the Japanese eel *Anguilla japonica. Aquaculture* **2007,** 267, 55–61.
9. Thiruvasagam, T.; Chidambaram, P.; Ranjan, A.; Komuhi, N.B. Significance of fatty acids in fish broodstock nutrition. *Anim. Reprod. Sci.* **2024**, 268, 107573.
10. Xu, Q.Y.; Zhang, M.L.; Qu, Q.; Chen, X.H.; Zhai, S.W. Requirement of dietary essential amino acid for Japanese eel (*Anguilla japonica*). *Nutr. Res.* **2019**, 12, 55–57.
11. Zhang, X.; Wang, Z.F. Progress in research on nutritional requirements and feed additives for Japanese eel (*Anguilla japonica*): a review. *J. Dalian Ocean Univ.* **2023**, 38, 543–552.
12. Wang, H.; Jiang, X.L.; Wang, Z.X.; Liu, Y. Ecological treatment of intensive eel aquaculture tail water. *J. Hydroecol.* **2024**, 45, 204–210.
13. Xia, Y.; Huang, X.R.; Liu, J.Y.; Zhang, T.; Zhuang, P.; Zhao, F. Effects of salinity acclimation on morphological characteristics and gonadal development of female *Anguilla japonica*. *J. Fish. Sci. China* **2024**, 31, 29–39.
14. Zhang, M.M.; Huang, X.R.; Wang, S.K.; Zhang, Y.C.; Zhang, T.; Song, C.; Yang, G.; Zhao, F. Study on estradiol-induced sexual differentiation in glass eels of Japanese eel (*Anguilla japonica*). *J. Fish. Sci. China* **2026**, 33, 012601.
15. Jang, W.J.; Kim, S.K.; Park, S.Y.; Kim, D.P.; Heo, Y.J.; Kim, H.; Lee, S.J.; Shin, M.G.; Lee, E.W.; Lee, S.; et al. Effect of host-associated bacillus-supplemented artificial diets on growth, survival rate, and gene expression in early-stage eel larvae (*Anguilla japonica*). *Fishes* **2023**, 8,247.
16. Furuita, H.; Jinbo, T.; Higuchi, M.; Nomura, K.; Sudo, R.; Matsunari, H.; Murashita, K.; Oku, H.; Yamamoto, T.; Tanaka, H. Diets comprising hen egg yolk and milk proteins as potential alternatives to shark egg-based diets for larvae of the Japanese eel *Anguilla japonica*. *Fish. Sci.* **2024**, 90, 295–305.
17. Zhang, T.T.; Zhao, F.; Zhang, T.; Wang, S.K.; Song, C.; Zhuang, P.; Lu, Y.N. Development of eel aquaculture industry and its resource conservation in China. *Fish. Inf. Strateg.* **2019**, 34, 235–243.
18. Wang, S.K.; Song, C.; Zhang, T.T.; Gao, Y.; Zhuang, P.; Zhao, F. Spatiotemporal distribution and catching status of elver (*Anguilla japonica*) in the Yangtze River Estuary. *J. Fish. China* **2023**, 47, 171–182.
19. Song, C.; Zhang, T.; Zhao, F.; Liu, J.Y.; Zhang, T.T.; Wang, S.K.; Yang, G.; Zhuang, P. Preliminary study on the tagging release of *Anguilla japonica* parents in the Yangtze Estuary. *Mar. Fish.* **2020**, 42, 699–710.
20. Jia, Q.J.; Shuai, F.M.; Li, J. Migration characteristics of Japanese eel in the Pearl River system based on otolith microchemistry. *Acta Hydrobiol. Sin.* **2024**, 48, 1758–1765.
21. Tsukamoto, K.; Chow, S.; Otake, T.; Kurogi, H.; Mochioka, N.; Miller, M.J.; Aoyama, J.; Kimura, S.; Watanabe, S.; Yoshinaga, T.; et al. Oceanic spawning ecology of freshwater eels in the western North Pacific. *Nat. Commun.* **2011**, 2, 21285957.
22. Miller, M.J.; Tsukamoto, K. The behavioral ecology and distribution of leptocephali: marine fish larvae with unforeseen abilities. *Mar. Biol.* **2020**, 167, 1–21.
23. Jin, X.C.; Li, K.; Ji, S.; Luo, X.N.; Shen, Z.Q.; Zeng, X.B.; Xie, B.; Han, H.; Zhang, J.M.; Ni, G.B.; et al. Comparative analysis of muscle quality and flavor of Japanese eel (*Anguilla japonica*) from two culture systems and wild. *J. Fish. China* **2026**, doi: 10.11964/jfc.20241014720.
24. Li, T.Z.; Li, K.; Wu, J.M.; Liu, L.P. Comparative study on the nutrient composition, amino acids, and fatty acids composition of glass eel tissues of Japanese eel (*Anguilla japonica*) from offshore of China. *J. Fish. China* **2023**, 47, 105–117.
25. Weber, J.M.; Brichon, G.; Zwingelstein, G. Fatty acid metabolism in rainbow trout (*Oncorhynchus mykiss*) tissues: differential incorporation of palmitate and oleate. *Can. J. Fish. Aquat. Sci.* **2003**, 60, 1281–1288.

26. Gao, X.; Li, M.; Guan, J.; Cheng, Z.; Xie, D.; Li, Y. Transcriptomic analysis reveals regulatory responses of fatty acid positional distribution in triacylglycerols and lipid composition to dietary n-3 HUFA in the muscle of *Trachinotus ovatus*. *Animals* **2025**, 15, 2427.
27. Song, C.; Zhao, F.; Zhang, T.; Zhang, T.T.; Huang, X.R.; Yang, G.; Zhuang, P. Proximate composition and fatty acid profile in different tissues of wild adult female *Coilia mystus* in Yangtze Estuary. *Indian J. Anim. Res.* **2020**, doi: 10.18805/ijar. B-1255.
28. Péron, M.; Gonzalvez, R.; Hue, S.; Soudant, P.; Le Grand, F.; Mazurais, D.; Vagner, M. Spatial and ontogenetic modulation of fatty acid composition in juvenile European sea bass (*Dicentrarchus labrax*) from two French estuaries. *Mar. Environ. Res.* **2024**, 197, 106456.
29. Fokina, N.; Ruokolainen, T.; Efremov, D.; Murzina, S.A. Changes in the composition of lipids and their fatty acids in pink salmon, *Oncorhynchus gorbuscha*, during the spawning migration. *Environ. Biol. Fish.* **2025**, 108, 109–130.
30. Lazo-Andrade, J.; Guzmán-Rivas, F.A.; Barría, P.; Urzúa, Á. Variability in the energy reserves of swordfish (*Xiphias gladius*) of the southeastern Pacific Ocean: A temporal and intra-individual perspective. *Mar. Environ. Res.* **2023**, 190, 106081.
31. Zhu, D.G. Analysis of fatty acid composition in red tilapia parent fish. *Southwest China J. Agric. Sci.* **2012**, 25, 713–717.
32. Lin, D.M.; Sun, C.J.; Xuan, S.P.; Han, F.; Chen, X.J. Fatty acid composition and its changes during sexual maturation in female *Illex argentines*. *J. Shanghai Ocean Univ.* **2019**, 3, 409–418.
33. Keva, O.; Tang, P.; Käkelä, R.; Hayden, B.; Taipale, S.J.; Harrod, C.; Kahilainen, K.K. Seasonal changes in European whitefish muscle and invertebrate prey fatty acid composition in a subarctic lake. *Freshw. Biol.* **2019**, 64, 1908–1920.
34. He, A.Y.; Ning, L.J.; Chen, L.Q.; Chen, Y.L.; Xing, Q.; Li, J.M.; Qiao, F.; Li, D.L.; Zhang, M.L.; Du, Z.Y. Systemic adaptation of lipid metabolism in response to low- and high-fat diet in Nile tilapia (*Oreochromis niloticus*). *Physiol. Rep.* **2015**, 3, e12485.
35. Wang, S.Y.; Huang, J.; Liang, L.Q.; Su, B.F.; Zhang, Y.; Liew, H.J.; Sun, B.; Zhang, L.M.; Chang, Y.M. Distinctive metabolite profiles in migrating Amur ide (*Leuciscus waleckii*) reveal changes in osmotic pressure, gonadal development, and energy allocation strategies. *Front. Environ. Sci.* **2022**, 10, 997827.
36. Wang, J.Y.; Miao, S.Y.; Li, B.S.; Wang, S.X.; Huang, B.S.; Zhang, L.M. A comparative study on lipid and fatty acid compositions of wild *Paralichthys olivaceus* broodstocks during different ovary development stages. *J. Shanghai Ocean Univ.* **2011,** 20, 238–243.
37. Morais, S.; Mourente, G.; Martínez, A.; Gras, N.; Tocher, D.R. Docosahexaenoic acid biosynthesis via fatty acyl elongase and Δ4-desaturase and its modulation by dietary lipid level and fatty acid composition in a marine vertebrate. *BBA - Mol. Cell Biol. Lipids* **2015**, 1851, 588–597.
38. Jin, Y.; Harvey, T.N.; Bartosova, Z.; Hassani, S.; Bruheim, P.; Sandve, S.R.; Vik, J.O. Diet and life stage-associated lipidome remodeling in Atlantic salmon. *J. Agric. Food Chem.* **2021**, 69, 3787–3796.
39. Song, C.; Zhuang, P.; Zhang, L.Z.; Zhao, F.; Wang, Y. Analysis of fatty acid composition in different tissues of adult female *Tridentiger trigonocephalus* collected from the Yangtze Estuary. *J. Fish. China* **2012**, 36, 1725–1730.
40. Haliloglu, H.I.; Bayr, A.; Sirkecioglu, A.N.; Aras, N.M.; Atamanalp, M. Comparison of fatty acid composition in some tissues of rainbow trout (*Oncorhynchus mykiss*) living in seawater and freshwater. *Food Chem.* **2004**, 86, 55–59.
41. Dong, Y.; Wei, Y.; Wang, L.; Song, K.; Zhang, C.; Lu, K.; Rahimnejad, S. Dietary n-3/n-6 polyunsaturated fatty acid ratio modulates growth performance in spotted seabass (*Lateolabrax maculatus*) through regulating lipid metabolism, hepatic antioxidant capacity and intestinal health. *Anim. Nutr.* **2023**, 14, 20–31.
42. Huang, X.X.; Wen, W.; Wei, L.K.; Feng, L.F.; Lv, W.Q. Characteristics of lipid and fatty acid accumulation in wild-caught broodstocks of *Pampus argenteus* from Mindong Seazone. *J. Fish. China* **2014**, 38, 99–108.
43. Yu, J.H.; Li, S.G.; Chang, J.; Niu, H.X. Effects of n-3 highly unsaturated fatty acids on reproductive performance of fish broodstock and its mechanism. *Chin. J. Anim. Nutr.* **2018**, 3, 813–819.

44. Xu, J.B.; Shi, Y.H.; Zhang, G.Y.; Liu, Y.S.; Zhang, H.M.; Lu, G.H.; Deng, P.P.; Zhang, Z.F. Analysis of lipid and fatty acid composition in different tissues of adult female and male *Takifugu flavidus*. *Food Sci.* **2014**, 35, 133–137.
45. Zhao, F.; Zhuang, P.; Zhang, L.Z.; Huang, X.R.; Feng, G.P.; Yan, W.G. Analysis of fatty acid composition in different tissues of adult female *Siganus gutattus*. *Mar. Fish.* **2009**, 31, 288–292.
46. Gu, J.; Zhang, Y.; Zhao, W.; Bao, Y.; Wang, X.; Zhu, T.; Sun, P.; Zhou, Q.; Jin, M. Dietary ratios of docosahexaenoic acid and eicosapentaenoic acid modulate osmoregulation and lipid metabolism in juvenile black seabream (*Acanthopagrus schlegelii*) under low salinity environment. *Aquaculture* **2026**, 613, 743424.
47. Mourente, G.; Megina, C.; Díaz-Salvago, E. Lipids in female northern bluefin tuna (*Thunnus thynnus thynnus* L.) during sexual maturation. *Fish Physiol. Biochem.* **2002**, 24, 351–363.
48. Burns, C.M.; Fuiman, L.A. Determining the position of southern flounder *Paralichthys lethostigma* on the reproductive energy allocation spectrum using an essential fatty acid as a maternal dietary biomarker. *Environ. Biol. Fish.* **2020**, 103, 1137–1148.
49. Shi, Y.H.; Xu, J.B.; Liu, Y.S.; Zhang, H.M.; Deng, P.P.; Lu, G.H.; Zhang, Z.F. Changes in fatty acid composition during early developmental stage of tawny puffer (*Takifugu flavidus*). *J. Fish. China* **2017**, 41, 1203–1212.
50. Fei, S.; Liu, C.; Xia, Y.; Liu, H.; Han, D.; Jin, J.; Yang, Y.; Zhu, X.; Xie, S. The effects of dietary linolenic acid to linoleic acid ratio on growth performance, tissues fatty acid profile and sex steroid hormone synthesis of yellow catfish *Pelteobagrus fulvidraco*. *Aquac. Rep.* **2020**, 17, 100361.
51. Cao, L. The Effects of Long Chain-polyunsaturated Fatty Acids on Nutritional Function of Tongue Sole (*Cynoglossus semilaevis*) Broodstock and Juveniles. *Shanghai Ocean Univ. Master's Thesis* **2017**.
52. Chen, W.Z.; Li, Y.Y.; Sun, Z.W.; Huang, J.H.; Du, Y.B.; Tan, Y. Effects of dietary n-3 HUFA levels on lipid content and fatty acid composition in tissues of *Plectorhynchus cinctus* broodstock. *J. South China Agric. Univ.* **2006**, 27, 96–100.
53. Li, Y.Y.; Chen, W.Z.; Sun, Z.W.; Chen, J.H.; Wu, K.G. Effects of n-3 HUFA content in broodstock diets on reproductive performance and seasonal changes of plasma sex steroid levels in *Plectorhynchus cinctus*. *Zool. Res.* **2004**, 25, 249–255.
54. Liu, L.; Li, R.; Zhang, J.M.; Guo, F.; Zhang, T. Composition and metabolizing of lipids and fatty acids in muscle of female *Anguilla japonica* during the period of artificial inducing ovarian maturation. *Acta Hydrobiol. Sin.* **2009**, 33, 1011–1019.
55. Koven, W.; Yanowski, E.; Gardner, L.; Nixon, O.; Block, B. Docosahexaenoic acid (DHA) is a driving force regulating gene expression in bluefin tuna (*Thunnus thynnus*) larvae development. *Sci. Rep.* **2024**, 14, 23191.
56. Shields, R.J.; Bell, J.G.; Luizi, F.S.; Gara, B.; Bromage, N.R.; Sargent, J.R. Natural copepods are superior to enriched artemia nauplii as feed for halibut larvae (*Hippoglossus hippoglossus*) in terms of survival, pigmentation and retinal morphology: relation to dietary essential fatty acids. *J. Nutr.* 1999, 129, 1186–1194.
57. Chaguaceda, F.; Eklöv, P.; Scharnweber, K. Regulation of fatty acid composition related to ontogenetic changes and niche differentiation of a common aquatic consumer. *Oecologia* **2020**, 193, 325–336.
58. Simopoulos, A.P. The importance of the ratio of omega-6/omega-3 essential fatty acids. *Biomed. Pharmacother.* **2002**, 56, 365–379.
59. Castell, J.D.; Blair, T.; Neil, S.; D'Abramo, L.R.; Cahu, C.; Harmon, P.; Ogunmoye, K. The effect of different HUFA enrichment emulsions on the nutritional value of rotifers (*Brachionus plicatilis*) fed to larval haddock (*Melanogrammus aeglefinus*). *Aquac. Int.* **2003**, 11, 109–117.