

Article

Swimming Performance and Energetics of Japanese Eels Across Selected Temperature, Salinity, and Silvering-Stage Contrasts

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Abstract

Swimming performance and energetics of the Japanese eel (*Anguilla japonica*) across growth and early migratory phases remain poorly quantified. We used swim-tunnel respirometry to compare selected environmental and developmental contrasts in yellow and early silver eels from the Tone River estuary. Critical swimming speed (U_{crit}), optimal swimming speed (U_{opt}), minimum cost of transport ($minCOT$), oxygen consumption at U_{opt} , and low-flow MO_2 measured at 0.1 m s^{-1} were evaluated in sequential freshwater trials at 25 and 18 °C, an exploratory Y2 freshwater–seawater comparison at 18 °C, and a Y2–S1 comparison in seawater at 18 °C. U_{crit} and low-flow MO_2 were numerically higher under FW25 than FW18, but this contrast was confounded with fixed trial order and fasting history. In Y2 eels, SW18 was associated with higher low-flow MO_2 than FW18, whereas active-speed-derived variables were limited by sample size. Under SW18, no clear stage-associated differences were detected between Y2 and S1 eels. Laboratory-derived U_{opt} and U_{crit} exceeded field-estimated active horizontal swimming speeds, but this comparison should be interpreted cautiously.

Keywords: *Anguilla japonica*; swim-tunnel respirometry; optimal swimming speed; critical swimming speed; silvering; salinity; temperature; spawning migration



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Key Contribution: This study provides baseline swim-tunnel respirometry data for Japanese eels across selected environmental and early migratory-stage contrasts. In an exploratory comparison of independent Y2 groups, SW18 was associated with higher low-flow MO_2 than FW18, whereas no clear stage-associated differences were detected between Y2 and S1 eels under SW18.

1. Introduction

Freshwater eels (genus *Anguilla* Schrank, 1798) are catadromous fishes that migrate between tropical oceanic spawning areas and coastal, brackish, and freshwater growth

habitats [1]. The Japanese eel, *Anguilla japonica* Temminck & Schlegel, 1846, is the only species in the genus for which fertilized eggs have been collected, providing direct evidence that its spawning area is located in waters west of the Mariana Islands [2]. Leptocephalus larvae hatched in this area are transported toward East Asian coastal waters by the North Equatorial Current and the Kuroshio [3]. During this northward transport in the Kuroshio, they metamorphose into glass eels and recruit to estuaries throughout East Asia [4]. Following river entry, they remain in a resident growth phase as elvers and subsequently as yellow eels for periods ranging from several years to a few decades.

After completing the yellow eel growth phase, anguillid eels undergo physiological and morphological changes associated with the onset of sexual maturation and transform into silver eels [5]. During this process, known as silvering, conspicuous changes occur, including the development of a silvery body coloration, increases in eye diameter and pectoral fin size [6], and thickening of the swim bladder wall [7]. These changes have generally been interpreted as adaptations for the oceanic spawning migration. Behavioral tracking using ultrasonic transmitters and laboratory experiments have also shown that activity differs between yellow and silver eels [8]. Because yellow eels represent the resident pre-migratory stage, comparisons between yellow and silver eels provide a useful framework for evaluating silvering-associated changes in swimming performance.

Silver-phase Japanese eels descend rivers, enter the sea, and return to their spawning area west of the Mariana Islands to spawn. Tracking studies using pop-up satellite archival tags (PSATs) have shown that they exhibit diel vertical migration during the spawning migration [9–12]. A previous report [11] estimated the horizontal movement speed of tagged individuals to range from 0.097–0.501 m s^{−1}. However, these estimates include both the active swimming component of the eel and the passive transport component driven by ocean currents. Acoustic telemetry studies have also quantified the active swimming speed of silver eels [13,14], reporting values of 0.06–0.16 m s^{−1} during the day and 0.07–0.18 m s^{−1} at night in the Kuroshio region, and 0.05–0.24 m s^{−1} during the day and 0.26–0.30 m s^{−1} at night in subtropical and tropical waters. These field-based estimates from PSAT and acoustic telemetry studies are lower than the physiologically optimal swimming speed reported for the European eel, *A. anguilla* Linnaeus, 1758, in swim-tunnel experiments (0.61–0.68 m s^{−1}) [15]. In addition, the activity of Japanese eels is known to decrease at low water temperatures [16].

Because Japanese eels are thought not to feed during the spawning migration [17], swimming efficiency is likely to be an important determinant of migratory and reproductive success. However, the swimming speed of Japanese eels has not been measured in physiological experiments. Therefore, the relationship between field-observed swimming speeds and physiological swimming capacity remains unresolved in Japanese eels, particularly across migratory stages and environmental conditions. In this study, we used controlled swim-tunnel experiments to compare swimming performance and energetics among selected environmental and developmental contrasts in Japanese eels, and then compared the laboratory-derived swimming parameters with field-estimated swimming speeds reported from tracking studies.

2. Materials and Methods

2.1. Eels Used in the Experiments

Experiments were conducted using 25 Japanese eels collected from the Tone River estuary, Chiba Prefecture, Japan. Fifteen individuals captured by fishers using set nets between September and December 2011 were purchased in December 2011 (total length [TL], 70.0–93.0 cm). The exact capture month and pre-acquisition holding conditions of each individual in this cohort were unavailable. Ten additional individuals were obtained

from the same locality in August 2012 (TL, 53.4–70.3 cm) (Table 1). Capture salinity was not recorded.

Table 1. Biological data for the experimental eels used in this study before the swim-tunnel experiments. TL, total length (cm); BW, body weight (g); SI, silvering stage (Y1, Y2, or S1); EV, eye vertical diameter (mm); EH, eye horizontal diameter (mm); Fin, pectoral fin length (mm). For freshwater trials, the experiment order was FW25 followed by FW18.

Fish Code	TL	BW	SI	EV	EH	Fin	Experiment
Eel 1	53.4	198	Y1	5.3	5.3	24.0	FW25 → FW18
Eel 2	58.9	300	Y1	4.4	5.0	27.8	FW25 → FW18
Eel 3	53.5	183	Y1	3.6	4.5	24.0	FW25 → FW18
Eel 4	56.3	211	Y1	3.6	3.7	25.4	FW25 → FW18
Eel 5	56.8	204	Y1	4.5	3.9	24.9	FW25 → FW18
Eel 6	62.7	347	Y2	5.1	6.9	38.7	FW25 → FW18
Eel 7	70.3	570	Y2	6.3	7.0	39.9	FW25 → FW18
Eel 8	66.8	310	Y2	6.2	6.1	31.5	FW25 → FW18
Eel 9	65.3	440	Y2	5.3	5.9	34.5	FW25 → FW18
Eel 10	69.3	414	Y2	5.5	6.4	34.8	FW25 → FW18
Eel 11	81.0	857	Y2	7.0	6.4	38.0	SW18
Eel 12	75.4	713	Y2	6.2	6.2	37.0	SW18
Eel 13	79.5	862	Y2	7.2	6.5	43.6	SW18
Eel 14	77.6	718	Y2	7.2	6.5	38.0	SW18
Eel 15	78.2	777	Y2	7.4	6.7	44.5	SW18
Eel 16	73.0	615	Y2	6.8	6.5	35.4	SW18
Eel 17	70.0	529	Y2	6.9	6.4	41.2	SW18
Eel 18	78.5	825	S1	7.9	8.0	45.6	SW18
Eel 19	80.3	914	S1	8.5	7.8	45.0	SW18
Eel 20	77.5	747	S1	7.5	7.5	35.8	SW18
Eel 21	73.2	720	S1	7.5	7.5	40.5	SW18
Eel 22	81.5	906	S1	8.2	8.2	47.0	SW18
Eel 23	83.0	977	S1	9.0	9.2	45.4	SW18
Eel 24	93.0	1366	S1	9.1	9.1	52.8	SW18
Eel 25	91.0	1060	S1	8.3	8.3	48.6	SW18

After acquisition, the eels were maintained in light-shielded plastic tanks (140 cm width × 140 cm depth × 60 cm height) under laboratory conditions for approximately one month before swim-tunnel experiments. The eels assigned to freshwater trials were initially held in freshwater at 25 °C before the first FW25 trial, whereas eels assigned to seawater trials were held in seawater at 18 °C before the SW18 trial. The same freshwater individuals were subsequently used for the FW18 trial after the FW25 trial. Feeding was withheld for 3 days before the FW25 and SW18 trials. For the subsequent FW18 trial, fasting had already begun 3 days before the FW25 trial and was continued throughout the transition/acclimation period to FW18. Trial order and fasting duration for each trial are provided in Supplementary Table S1. On the day before the first trials, the eels were anesthetized with a 2 mL L^{−1} eugenol solution, and TL, body weight (BW), pectoral fin length, and eye diameter were measured. Individuals were assigned to the Y1, Y2, or S1 stage based on the silvering index described by [6].

2.2. Swim-Tunnel Apparatus and Respirometry Quality Control

Four Blazka-type swim tunnels developed at Leiden University (working volume, 127 L; see Figure 1 in van den Thillart et al. 2004 [18], and Supplementary Figure S1 in this paper) were installed in an air-conditioned room, with the room temperature adjusted to approximate the experimental water temperature. Each tunnel was connected by piping to

a 250-L filtered and aerated reservoir tank and could be operated as a closed recirculating system isolated from the reservoir by valve control. Water within each tunnel was circulated by an internal propeller. When necessary, the valves were opened to exchange the water in the tunnel with water from the filtered and aerated reservoir. Dissolved oxygen concentration was measured using an oxygen sensor (InPro 6050, Mettler Toledo, Urdorf, Switzerland) installed in the swimming chamber of each tunnel.

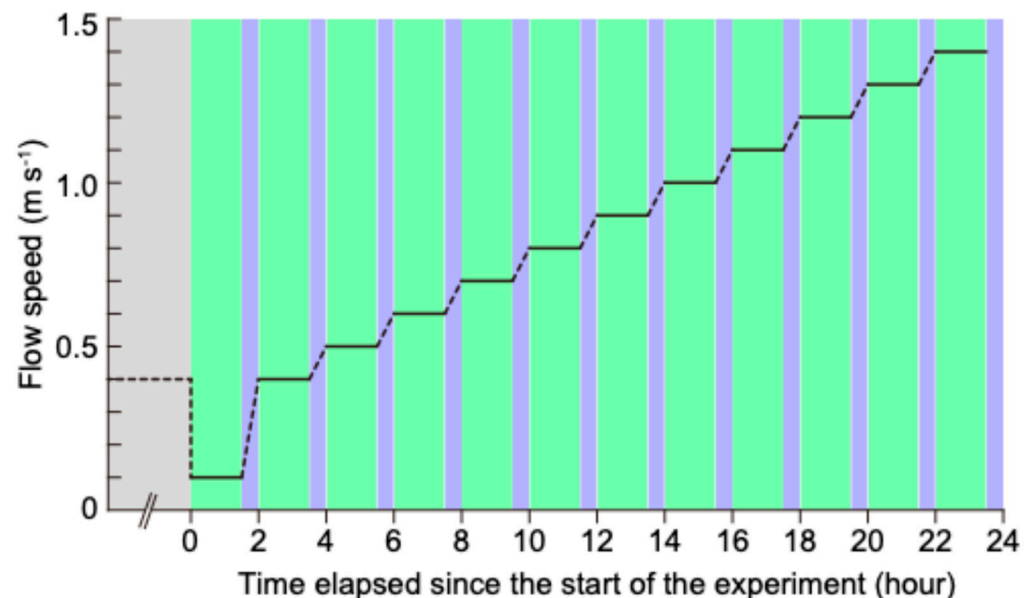


Figure 1. Flow-speed schedule used in the swim-tunnel experiments during a single experimental sequence. After morphometric measurements, anesthetized eels were placed in a swim tunnel and allowed to recover and acclimate overnight at a flow velocity of 0.4 m s^{-1} (gray shaded area). On the following day, oxygen consumption was first measured at 0.1 m s^{-1} as an operational low-flow condition, after which the experiment alternated between a 1.5 h measurement period at a constant flow velocity (green shaded area) and a 0.5 h water-exchange period, during which flow velocity was increased stepwise (blue shaded area). The black line indicates flow velocity; solid and dashed segments represent measurement and non-measurement periods, respectively.

Dissolved oxygen sensors were calibrated to 100% air saturation before the start of the experiments for each cohort. During the trial period for each cohort, the sensors were kept connected to the transmitters and remained powered. Before measurements on each experimental day, the sensors were removed from the water, cleaned, dried, and checked in air to confirm that they indicated 100% air saturation. If the reading deviated from 100% saturation, the sensor was recalibrated according to the transmitter procedure. If power loss or disconnection occurred, the sensors were checked or recalibrated after allowing the polarization time required by the manufacturer's manual.

Oxygen concentration was recorded continuously during each closed-circuit measurement period, and the rate of oxygen decline was estimated from the slope of a linear regression of dissolved oxygen concentration against time. No formal R^2 -based slope-quality threshold was applied. However, oxygen-decline slopes were estimated from measurement periods in which oxygen concentration decreased approximately linearly. Measurement periods showing obvious disturbance, abrupt oxygen fluctuations, or interruption due to water exchange were excluded from slope estimation.

Background microbial respiration was not measured directly and was therefore not subtracted from MO_2 estimates. Thus, the reported MO_2 values represent the combined oxygen consumption of the fish and any chamber background respiration, normalized to fish body mass, rather than fish metabolic oxygen consumption alone. Because COT was

calculated from MO_2 , the reported COT values were likewise not blank-corrected. The use of filtered and aerated reservoir water and repeated water exchange during trials was intended to minimize background respiration; however, the absence of blank correction is acknowledged as a limitation.

Flow speeds were set according to the calibrated propeller-control settings of the swim-tunnel system. Solid-blocking correction was not applied; therefore, possible effects of fish size on effective flow speed are acknowledged as a potential source of uncertainty. Oxygen concentration measurements were temperature-compensated by the sensor system, and salinity settings were adjusted according to the experimental water medium when calculating dissolved oxygen concentration.

2.3. Experimental Design

Experimental conditions were designed to compare swimming performance and energetics among selected environmental and developmental contrasts while approximating the conditions likely to be encountered by eels at each silvering stage in the wild (Table 1). The design was not fully factorial. Temperature-associated differences were assessed using sequential FW25 and FW18 trials in the same Y1- and Y2-stage yellow eels. The FW25 trial was conducted first, followed by the FW18 trial. Freshwater–seawater differences at 18 °C were explored by comparing Y2-stage eels under FW18 and SW18. Stage-associated differences under seawater conditions were assessed by comparing Y2- and S1-stage eels under SW18. Five Y1-stage individuals (fish code: Eels 1–5) were tested under both freshwater conditions, at 18 °C (Exp. FW18) and 25 °C (Exp. FW25). Five Y2-stage individuals (Eels 6–10) were also tested under the same two freshwater conditions. Seven additional Y2-stage individuals (Eels 11–17) were tested in seawater at 18 °C (Exp. SW18), and eight S1-stage individuals (Eels 18–25) were likewise tested under Exp. SW18.

Swimming trials were conducted following the general protocol of Palstra et al. (2008) [15], with modifications to fit the present experimental setup. After being anesthetized for morphometric measurements, each individual was transferred while still anesthetized into the swimming chamber of a swim tunnel filled with water adjusted to the relevant experimental condition. Each eel was then allowed to recover from anesthesia under a 0.4 m s^{-1} current to provide oxygen and acclimate to the experimental environment overnight (gray-shaded phase in Figure 1).

On the following day, low-flow MO_2 was measured at a flow velocity of 0.1 m s^{-1} . Because the dissolved oxygen sensor was fixed inside the swimming chamber, some water circulation was required to obtain representative oxygen measurements. Accordingly, a flow velocity of 0.1 m s^{-1} was used as an operational low-flow condition for oxygen measurement.

Swimming trials were subsequently initiated at a flow velocity of 0.4 m s^{-1} . At each test velocity, one experimental cycle consisted of a 1.5-h measurement period under temporarily closed-circuit conditions (green-shaded phase in Figure 1), followed by a 0.5-h water-exchange period, during which the tunnel water was replaced with aerated water to restore dissolved oxygen levels (blue-shaded phase in Figure 1). After each cycle, flow velocity was increased stepwise by 0.1 m s^{-1} . This procedure was repeated until the eel could no longer maintain its position against the current. At the start of each measurement period, dissolved oxygen concentration in the swimming chamber was adjusted to 100% air saturation. If dissolved oxygen declined below 75% during the measurement period, the trial was temporarily suspended, the tunnel water was exchanged, and measurements were resumed after dissolved oxygen had recovered to 80–90% air saturation.

2.4. Calculations of Swimming Parameters

To compare swimming performance among experimental conditions, oxygen-consumption-based physiological parameters were calculated for each eel following Palstra et al. (2008) [15]. The mass-specific oxygen consumption rate (MO_2 ; $\text{mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$) and cost of transport (COT ; $\text{mg O}_2 \text{ kg}^{-1} \text{ km}^{-1}$) were calculated using Equations (1) and (2), respectively:

$$MO_2 = (127 \times \Delta[O_2] / \Delta t) / (BW), \quad (1)$$

$$COT = MO_2 / (3.6 \times U), \quad (2)$$

where 127 is the working volume (in liters) of the swim tunnel, $\Delta[O_2] / \Delta t$ is the rate of decline in dissolved oxygen concentration in the swimming chamber, and U is swimming speed (m s^{-1}). Dissolved oxygen concentration ($[O_2]$) in the swimming chamber was recorded continuously during each measurement period, and $\Delta[O_2] / \Delta t$ was obtained from the slope of the linear regression of dissolved oxygen concentration against time (t). COT was calculated from MO_2 by normalizing oxygen consumption to swimming speed, with the factor 3.6 converting U from m s^{-1} to km h^{-1} . MO_2 measured at 0.1 m s^{-1} was treated as an operational low-flow MO_2 measurement. Because a small flow velocity was required for oxygen-sensor measurements inside the swimming chamber, this value was not assumed to represent true resting metabolic rate or standard metabolic rate.

Critical swimming speed (U_{crit}) was calculated according to Brett (1964) [19] as follows:

$$U_{crit} = U_f + (t_f / 120) \times \Delta U, \quad (3)$$

where U_f is the highest swimming speed (m s^{-1}) that the eel was able to maintain for the full 2-h period, t_f is the time (in minutes) spent swimming at the final, fatiguing speed, and ΔU is the velocity increment between successive swimming steps (0.1 m s^{-1}). Fatigue was defined as the point at which the eel could no longer maintain its swimming position and remained in contact with the downstream grid despite continued flow. The time spent at the final, fatiguing speed was recorded as t_f . The optimal swimming speed (U_{opt}) was determined using a discrete-minimum approach. For each trial, U_{opt} was defined as the observed active swimming speed ($\geq 0.4 \text{ m s}^{-1}$) at which COT was minimized, and $minCOT$ was defined as the corresponding COT value. MO_2 at U_{opt} was obtained from the same speed step. Curve fitting was not applied because the number of available speed steps differed among individuals and decreased at higher swimming speeds, making fitted individual COT -speed relationships unstable for several trials. Both U_{crit} and U_{opt} were subsequently normalized by TL and expressed as TL s^{-1} . U_{opt} , $minCOT$, and MO_2 at U_{opt} were determined from active swimming speeds ($\geq 0.4 \text{ m s}^{-1}$), whereas 0.1 m s^{-1} was used only for low-flow MO_2 . The 0.1 m s^{-1} step was excluded from COT -based estimation of U_{opt} and $minCOT$ because it was used as an operational low-flow measurement rather than as an active swimming step. The 0.4 m s^{-1} step was therefore treated as the first active swimming speed in the experimental protocol. Individual-level swimming and metabolic endpoints are provided in Supplementary Table S1, and speed-step-level MO_2 and COT values are provided in Supplementary Tables S2 and S3, respectively. We acknowledge, however, that 0.4 m s^{-1} may already represent a relatively high sustained speed for smaller individuals. This limitation was considered when interpreting U_{opt} and $minCOT$.

2.5. Statistical Analysis

Because the experimental design was not fully factorial, statistical analyses were conducted as exploratory analyses of predefined contrasts rather than as a single factorial test of temperature, salinity, and silvering stage effects. The following individual-level

response variables were analysed: critical swimming speed (U_{crit}) and optimal swimming speed (U_{opt}), each expressed both in absolute units (m s^{-1}) and normalized by total length (TL s^{-1}); minimum cost of transport (minCOT); MO_2 at U_{opt} ; and MO_2 measured at the low-flow condition of 0.1 m s^{-1} . The latter variable was treated as an operational low-flow MO_2 measurement rather than as a true standard or resting metabolic rate.

Three predefined contrasts were analysed. First, sequential FW25 and FW18 trials in the same yellow eels were compared to evaluate temperature-associated differences under a fixed trial order. Because the same individuals were used in both trials, this contrast was analysed as a paired comparison, with the estimated difference defined as FW25 minus FW18. Second, Y2-stage eels under FW18 and SW18 were compared at 18°C as an exploratory freshwater–seawater comparison, with the estimated difference defined as SW18 minus FW18. This comparison involved independent groups and was interpreted as a salinity-associated comparison rather than as a definitive salinity effect. Third, Y2- and S1-stage eels under SW18 were compared to evaluate stage-associated differences under seawater conditions, with the estimated difference defined as S1 minus Y2.

For paired contrasts, two-sided p -values were obtained by exact enumeration of all possible sign flips of the paired differences. Unadjusted 95% permutation confidence intervals were obtained by inversion of the same sign-flip tests. For independent-group contrasts, all possible reallocations of group labels were enumerated, and unadjusted 95% permutation confidence intervals were obtained by inversion of the corresponding tests under an additive location-shift framework. Exact confidence sets that were unbounded because of very small sample size were reported as unbounded. Confidence intervals were unadjusted, whereas the Benjamini–Hochberg procedure was applied only to p -values.

Inclusion rules were defined before analysis. For U_{crit} , trials were included when U_{crit} was available, including boundary values and protocol-confirmed low-speed cases; trials with non-estimable or missing U_{crit} were excluded from the corresponding analysis. For U_{opt} , minCOT , and MO_2 at U_{opt} , the main analysis included values classified as valid or boundary estimates, because U_{opt} was determined as the observed active swimming speed at which COT was minimized. Trials with only a single active-speed point or non-estimable active-swimming metrics were excluded. A sensitivity analysis was also conducted using only valid, non-boundary U_{opt}/minCOT estimates. Low-flow MO_2 analyses included only trials with valid MO_2 measurements at 0.1 m s^{-1} .

To assess whether stage-associated differences under SW18 were sensitive to body size, exploratory body-size-adjusted analyses were conducted for the Y2 versus S1 comparison. For each response variable, ordinary least-squares models were fitted with silvering stage and either total length or body weight as predictors. Total length and body weight were analysed in separate models because of the small sample size and collinearity between body-size variables. These models were used as sensitivity analyses and were not interpreted as definitive tests of silvering-stage effects.

Speed-step-level MO_2 and COT data were summarized descriptively by experimental condition, silvering stage, and swimming speed. For each speed step, the number of contributing trials, mean, standard deviation, median, and standard error were calculated. These summaries were used for graphical presentation of MO_2 –speed and COT –speed relationships. Because the number of individuals decreased at higher speeds, speed-specific trends were interpreted descriptively.

All analyses were performed in Python 3 using pandas, NumPy, SciPy, and statsmodels. The processed individual-level and speed-step-level datasets, together with the Python scripts used to generate the statistical summaries and figures, are available in the GitHub repository at https://github.com/TakatoshiHiguchi/swim-tunnel_respirometry_experiments_in_Japanese_eels (version 1.0.0, accessed on 21 July 2026).

2.6. Comparison with Field-Observed Swimming Speeds

Laboratory-derived swimming speeds were compared with active swimming speeds reported from field-tracking studies of Japanese silver eels. In particular, values reported by Fukuda et al. (2022) [14] were used as representative estimates of active horizontal swimming speed during oceanic migration. Field values expressed in body lengths per second were compared with TL-standardized laboratory values.

3. Results

3.1. Morphometric Characteristics of Experimental Eels

Morphometric data for the eels used in this study, including TL, BW, eye diameters (EV and EH), and pectoral fin length (*Fin*), are summarized by silvering stage (Y1, Y2, and S1) in Table 2. These traits showed progressive changes across stages. Among them, the mean difference in pectoral fin length (ΔFin) was 12.9 mm between Y1 and Y2, whereas the corresponding difference between Y2 and S1 was 7.0 mm, indicating a smaller change between the latter two stages. The same pattern was retained after standardizing pectoral fin length by total length. In addition, the 95% confidence intervals (CI) for standardized pectoral fin length overlapped between Y2 (95% CI, 4.96–5.59) and S1 (5.16–5.80), whereas that of Y1 (4.37–4.67) did not overlap with those of the other stages. TL, BW, EV, and EH also increased progressively across stages. Because TL and BW increased across silvering stages, body size and silvering stage were partially confounded in the present dataset. This issue was considered in the interpretation of stage-associated comparisons, and exploratory body-size-adjusted analyses were conducted for the SW18 Y2–S1 comparison.

Table 2. Morphological parameters of Japanese eels for the different silvering stages. For each parameter, mean $\pm$ SD, range, and 95% CI are shown for the individuals used in this study.

Parameter	Stage	Mean $\pm$ SD	Range	95% CI [Lower, Upper]
TL (cm)	Y1	55.78 $\pm$ 2.34	53.4–58.9	[52.87, 58.69]
	Y2	72.43 $\pm$ 5.95	62.7–81.0	[68.64, 76.21]
	S1	82.25 $\pm$ 6.71	73.2–93.0	[76.64, 87.86]
BW (g)	Y1	219.20 $\pm$ 46.33	183–300	[161.67, 276.73]
	Y2	596.00 $\pm$ 192.35	310–862	[473.79, 718.21]
	S1	939.38 $\pm$ 206.31	720–1366	[766.90, 1111.85]
EH (mm)	Y1	4.48 $\pm$ 0.69	3.7–5.3	[3.63, 5.33]
	Y2	6.46 $\pm$ 0.31	5.9–7.0	[6.26, 6.66]
	S1	8.20 $\pm$ 0.65	7.5–9.2	[7.65, 8.75]
EV (mm)	Y1	4.28 $\pm$ 0.71	3.6–5.3	[3.40, 5.16]
	Y2	6.43 $\pm$ 0.79	5.1–7.4	[5.92, 6.93]
	S1	8.25 $\pm$ 0.61	7.5–9.1	[7.74, 8.76]
Fin (mm)	Y1	25.22 $\pm$ 1.56	24.0–27.8	[23.28, 27.16]
	Y2	38.09 $\pm$ 3.82	31.5–44.5	[35.67, 40.52]
	S1	45.09 $\pm$ 5.12	35.8–52.8	[40.81, 49.36]
EV/TL (%)	Y1	0.77 $\pm$ 0.14	0.64–0.99	[0.60, 0.94]
	Y2	0.89 $\pm$ 0.06	0.79–0.99	[0.85, 0.93]
	S1	1.01 $\pm$ 0.05	0.91–1.08	[0.96, 1.05]
EH/TL (%)	Y1	0.81 $\pm$ 0.14	0.66–0.99	[0.64, 0.97]
	Y2	0.90 $\pm$ 0.09	0.79–1.10	[0.84, 0.95]
	S1	1.00 $\pm$ 0.06	0.91–1.11	[0.95, 1.05]
Fin/TL (%)	Y1	4.52 $\pm$ 0.12	4.38–4.72	[4.37, 4.67]
	Y2	5.27 $\pm$ 0.50	4.69–6.17	[4.96, 5.59]
	S1	5.48 $\pm$ 0.38	4.62–5.81	[5.16, 5.80]

3.2. Speed-Specific Oxygen Consumption and Cost of Transport

The eels used in this study were tested at flow velocities ranging from 0.1 to 1.3 m s^{−1}, with 0.1 m s^{−1} used as an operational low-flow condition, although the number of individuals contributing measurements decreased at higher speeds. Across experimental conditions, MO_2 generally increased with increasing swimming speed (Figure 2). Because sample size differed among speed steps and became small at high speeds, speed-specific

MO_2 values were summarized descriptively rather than tested statistically. Detailed speed-step-level MO_2 values and the number of contributing trials at each speed are provided in Supplementary Table S2.

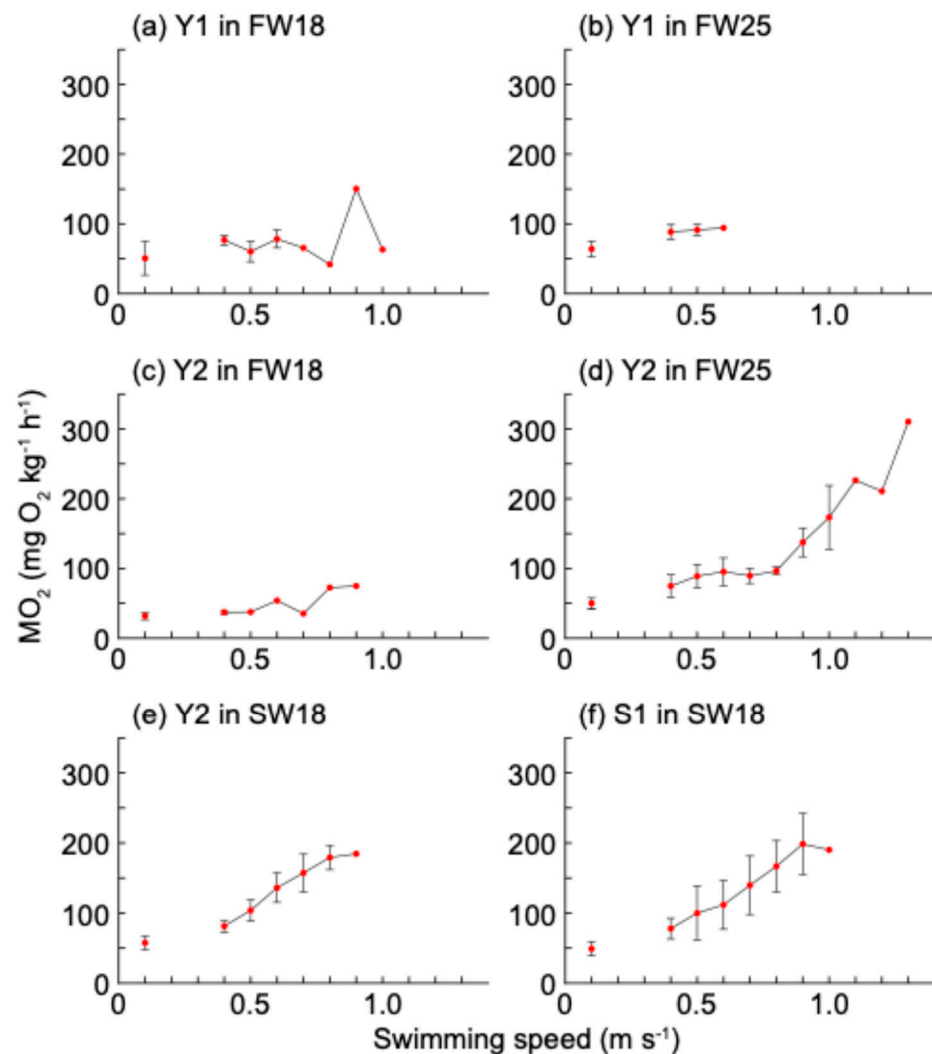


Figure 2. Mass-specific oxygen consumption rate (MO_2 ; $\text{mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$) at each swimming speed under each experimental condition. Points and error bars indicate mean $\pm$ SD. The number of contributing trials differed among speed steps and is provided in Supplementary Table S2. Lines are shown only as visual guides.

COT also varied with swimming speed and was used to determine U_{opt} and $minCOT$ from observed active swimming speeds $\geq 0.4 \text{ m s}^{-1}$ (Figure 3). The 0.1 m s^{-1} step was not used for U_{opt} or $minCOT$ estimation because it represented an operational low-flow condition. Detailed speed-step-level COT values, together with the speed-step sample sizes, are provided in Supplementary Table S3. Because the number of individuals decreased at higher speeds, the MO_2 –speed and COT –speed relationships were interpreted descriptively.

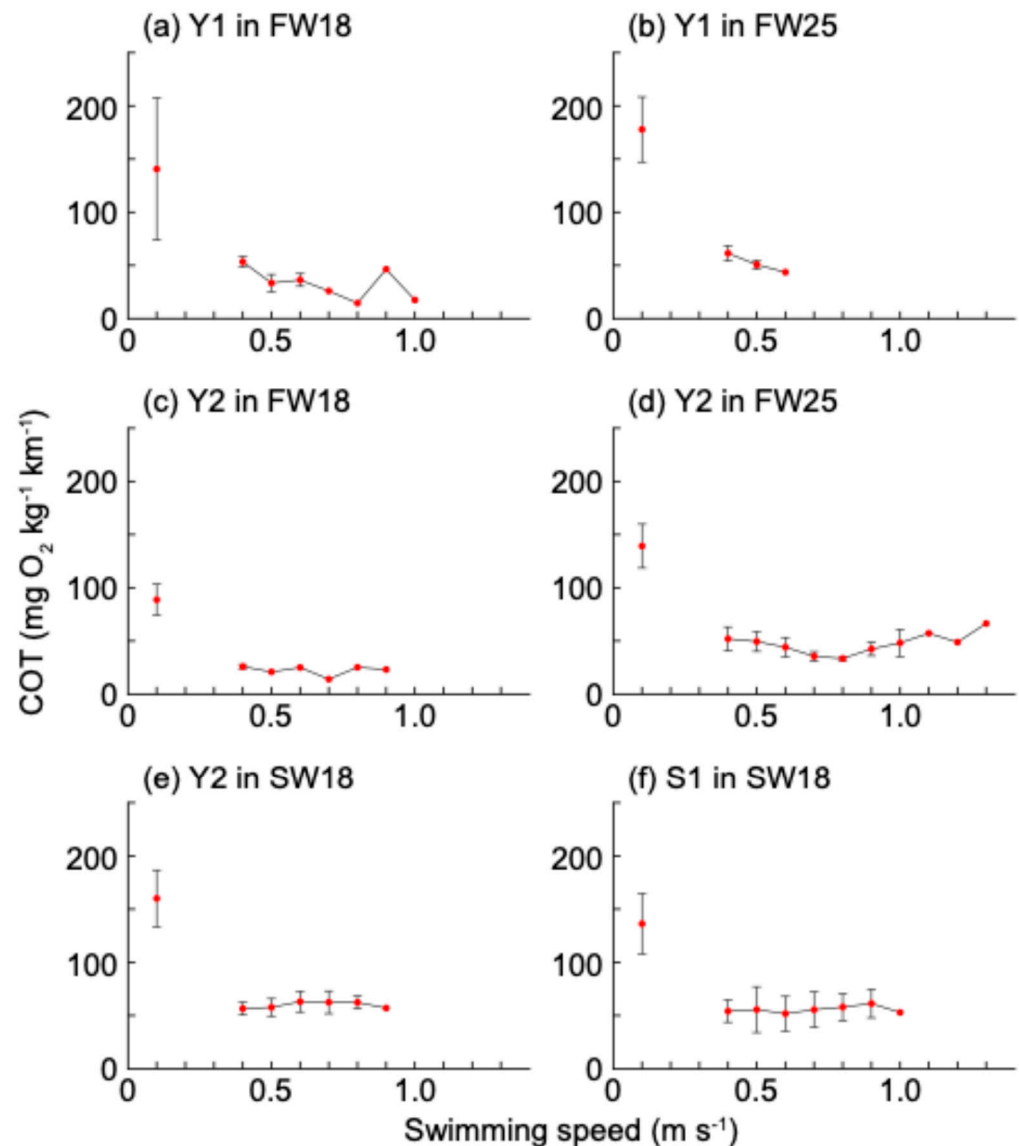


Figure 3. The cost of transport (COT; $\text{mg O}_2 \text{ kg}^{-1} \text{ km}^{-1}$) at each swimming speed under each experimental condition. Points and error bars indicate mean $\pm$ SD. COT values at active swimming speeds $\geq 0.4 \text{ m s}^{-1}$ were used to determine U_{opt} and minCOT . The number of contributing trials differed among speed steps and is provided in Supplementary Table S3. Lines are shown only as visual guides.

3.3. Sequential FW25 and FW18 Comparison in Yellow Eels

In the sequential freshwater trials, the same yellow eels were first tested under FW25 and then under FW18. Therefore, this comparison was interpreted as a temperature-associated contrast under a fixed trial order rather than as an isolated temperature effect.

U_{crit} was numerically higher under FW25 than under FW18, but the confidence intervals were wide and the differences were not supported after FDR adjustment. Absolute U_{crit} was $0.69 \pm 0.31 \text{ m s}^{-1}$ under FW25 and $0.52 \pm 0.27 \text{ m s}^{-1}$ under FW18, with a paired mean difference of 0.16 m s^{-1} (95% permutation CI, -0.09 to 0.45 ; exact sign-flip $p = 0.23$; FDR-adjusted $p = 0.26$). TL-standardized U_{crit} showed a similar pattern, with a paired mean difference of 0.23 TL s^{-1} (95% permutation CI, -0.17 to 0.66 ; exact sign-flip $p = 0.26$; FDR-adjusted $p = 0.26$).

Low-flow MO_2 was also numerically higher under FW25 than under FW18. Mean low-flow MO_2 was $57.8 \pm 12.5 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$ under FW25 and $42.5 \pm 21.7 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$ under FW18, with a paired mean difference of $15.3 \text{ mg O}_2 \text{ kg}^{-1} \text{ h}^{-1}$ (95% permutation CI,

−1.72 to 31.52; exact sign-flip $p = 0.07$; FDR-adjusted $p = 0.21$). Because the FW18 trial was conducted after the FW25 trial and fasting continued through the transition/acclimation period, these differences were interpreted cautiously.

Detailed results for all individual-level endpoints in this contrast are provided in Supplementary Table S4. U_{opt} , $minCOT$, and MO_2 at U_{opt} could be estimated for only three paired trials in the main analysis, and only one paired trial remained in the valid-only sensitivity analysis. Therefore, these active-speed-derived variables were treated as descriptive for this contrast.

3.4. Exploratory FW18–SW18 Comparison in Y2 Eels

In Y2-stage yellow eels, FW18 and SW18 were compared at 18 °C as an exploratory freshwater–seawater contrast. This comparison involved independent groups and was therefore interpreted as a salinity-associated comparison rather than as a definitive test of salinity effects.

Absolute U_{crit} ($m\ s^{-1}$) was numerically higher under SW18 than under FW18. Mean U_{crit} was $0.47 \pm 0.26\ m\ s^{-1}$ under FW18 and $0.80 \pm 0.16\ m\ s^{-1}$ under SW18, with a mean difference of $0.33\ m\ s^{-1}$ (95% permutation CI, 0.04 to 0.58; exact label-permutation $p = 0.04$; FDR-adjusted $p = 0.06$). TL-standardized U_{crit} showed a similar pattern, with a mean difference of $0.37\ TL\ s^{-1}$ (95% permutation CI, −0.04 to 0.73; exact label-permutation $p = 0.07$; FDR-adjusted $p = 0.07$).

The clearest difference in this contrast was observed for low-flow MO_2 . In this exploratory independent-group contrast, SW18 was associated with higher mean low-flow MO_2 than FW18 (SW18, $57.6 \pm 10.5\ mg\ O_2\ kg^{-1}\ h^{-1}$; FW18, $31.9 \pm 5.9\ mg\ O_2\ kg^{-1}\ h^{-1}$), with a mean difference of $25.7\ mg\ O_2\ kg^{-1}\ h^{-1}$ (95% permutation CI, 12.74 to 37.38; exact label-permutation $p < 0.01$; FDR-adjusted $p = 0.01$).

Detailed results for this exploratory contrast are provided in Supplementary Table S4. Because only one Y2 individual under FW18 contributed valid or boundary-estimate values for U_{opt} , $minCOT$, and MO_2 at U_{opt} , differences in these active-speed-derived variables were not interpreted as evidence for or against salinity-associated changes in active swimming energetics.

3.5. Stage-Associated Comparison Under SW18

Under SW18, Y2-stage yellow eels and S1-stage silver eels were compared to evaluate stage-associated differences under seawater conditions. No clear stage-associated differences were detected for U_{crit} . Mean absolute U_{crit} was nearly identical between Y2 and S1 eels (Y2, $0.795 \pm 0.155\ m\ s^{-1}$; S1, $0.795 \pm 0.164\ m\ s^{-1}$), with a mean difference of $-0.0003\ m\ s^{-1}$ (95% permutation CI, −0.19 to 0.19; exact label-permutation $p = 1.00$; FDR-adjusted $p = 1.00$). TL-standardized U_{crit} was also similar between stages, with a mean difference of $-0.08\ TL\ s^{-1}$ (95% permutation CI, −0.37 to 0.21; exact label-permutation $p = 0.56$; FDR-adjusted $p = 0.84$).

U_{opt} was numerically higher in S1 than in Y2 eels under SW18, but the confidence interval overlapped zero and the result was not robust after FDR adjustment. Mean U_{opt} was $0.48 \pm 0.08\ m\ s^{-1}$ in Y2 eels and $0.58 \pm 0.14\ m\ s^{-1}$ in S1 eels, with a mean difference of $0.09\ m\ s^{-1}$ (95% permutation CI, −0.04 to 0.23; exact label-permutation $p = 0.20$; FDR-adjusted $p = 0.75$). $MinCOT$ and MO_2 at U_{opt} also showed no clear stage-associated differences. Mean $minCOT$ was $51.8 \pm 5.22\ mg\ O_2\ kg^{-1}\ km^{-1}$ in Y2 eels and $48.0 \pm 13.8\ mg\ O_2\ kg^{-1}\ km^{-1}$ in S1 eels, with a mean difference of $-3.79\ mg\ O_2\ kg^{-1}\ km^{-1}$ (95% permutation CI, −15.70 to 9.19; exact label-permutation $p = 0.56$; FDR-adjusted $p = 0.75$).

Low-flow MO_2 was numerically lower in S1 than in Y2 eels, but this difference was not clearly supported. Mean low-flow MO_2 was $57.6 \pm 10.5\ mg\ O_2\ kg^{-1}\ h^{-1}$ in Y2 eels and $49.0 \pm 10.9\ mg\ O_2\ kg^{-1}\ h^{-1}$ in S1 eels, with a mean difference of $-8.57\ mg\ O_2\ kg^{-1}\ h^{-1}$ (95% permutation CI, −20.31 to 4.62; exact label-permutation $p = 0.16$; FDR-adjusted $p = 0.47$). Overall, no clear stage-associated differences were detected under SW18, but this comparison should

be interpreted cautiously because silvering stage and body size were partially confounded. Detailed results for this contrast are provided in Supplementary Table S4.

3.6. Body-Size Sensitivity Analyses for the SW18 Stage Comparison

Exploratory body-size-adjusted analyses were conducted for the SW18 Y2–S1 comparison to assess whether stage-associated differences were sensitive to TL or BW adjustment, and the results are summarized in Supplementary Table S5. These analyses did not provide clear evidence for stage-associated differences after accounting for body size. For absolute U_{crit} , the estimated S1–Y2 difference was 0.108 m s^{-1} after TL adjustment (95% CI, -0.09 to 0.30 ; $p = 0.24$) and 0.11 m s^{-1} after BW adjustment (95% CI, -0.11 to 0.32 ; $p = 0.31$). Low-flow MO_2 also showed no clear stage-associated difference after TL or BW adjustment. Because body size and silvering stage were partially confounded and sample sizes were small, these analyses were treated as exploratory sensitivity analyses rather than definitive tests of silvering-stage effects. U_{opt} tended to be numerically higher in S1 eels in some body-size-adjusted models, but the confidence intervals were wide and the results were not robust after multiple-testing adjustment.

4. Discussion

4.1. Swimming Performance of Japanese Eel Across Growth and Migratory Phases

The present study provides swim-tunnel respirometry data on Japanese eels across selected environmental and developmental contrasts. Because the experimental design was not fully factorial, the results should be interpreted within the specific contrasts tested rather than as general effects of temperature, salinity, or silvering stage. Within this framework, three main patterns emerged. First, in the sequential freshwater trials, U_{crit} and low-flow MO_2 were numerically higher under FW25 than under FW18, although these differences were not robust after FDR adjustment and were confounded with fixed trial order and fasting history. Second, in the exploratory Y2 freshwater–seawater comparison at 18°C , SW18 was associated with higher low-flow MO_2 than FW18, whereas active-speed-derived variables were difficult to interpret because of limited sample size. Third, under SW18, no clear stage-associated differences between Y2 and S1 eels were detected, and body-size-adjusted analyses did not resolve the partial confounding between silvering stage and body size. These results should be considered alongside previous anguillid studies that reported mixed stage-related patterns in swimming or metabolic traits [20,21].

4.2. Temperature-Associated Differences Under a Fixed Trial Order

In the sequential freshwater trials, U_{crit} and low-flow MO_2 were numerically higher under FW25 than under FW18, although these differences were not robust after FDR adjustment. Because blank-chamber respiration was not measured, the FW25–FW18 difference in low-flow MO_2 cannot be attributed solely to fish oxygen consumption. Background respiration may itself vary with temperature; therefore, part of the observed contrast may reflect temperature-dependent background respiration in the chamber rather than fish metabolism alone. Subject to this limitation, the direction of the observed low-flow MO_2 difference is consistent with the general expectation that higher temperature can increase metabolic demand in ectothermic fishes [22].

A numerically higher U_{crit} under FW25 may also be consistent with temperature-dependent changes in sustained swimming capacity, because warmer conditions can increase physiological rates and potentially support higher aerobic activity within a tolerable thermal range. However, higher temperature can also increase the energetic cost of maintaining a given swimming speed. Therefore, the numerically higher U_{crit} under FW25

is compatible with temperature-associated variation in sustained swimming performance, but the present data do not allow the underlying mechanisms to be separated clearly.

This interpretation should be cautious because the freshwater trials were conducted in a fixed order, with FW25 always preceding FW18. In addition, fasting began before the FW25 trial and continued through the transition/acclimation period before the FW18 trial. Thus, the observed FW25–FW18 differences may reflect a combination of temperature, trial order, recovery from the first swimming trial, and fasting history. The active-speed-derived variables, including U_{opt} , $minCOT$, and MO_2 at U_{opt} , were especially limited because only a small number of paired estimates were available. Consequently, the present results should be viewed as evidence of temperature-associated differences under the tested sequential protocol, rather than as a definitive test of temperature effects. Future experiments with randomized or counterbalanced temperature order, standardized fasting duration, and larger sample sizes will be needed to isolate the effects of water temperature on swimming energetics.

4.3. Freshwater–Seawater Comparison in Y2 Eels

In the exploratory independent-group comparison of Y2 eels at 18 °C, SW18 was associated with higher low-flow MO_2 than FW18. Because this operational measurement was not blank-corrected, the contrast could reflect greater fish oxygen demand, condition-dependent chamber respiration, or both. Background respiration may have differed between freshwater and seawater, and its direction and magnitude cannot be quantified from the present data. If part of the observed association was attributable to the fish, its direction would be consistent with possible physiological costs of seawater holding, including osmoregulatory processes [23].

However, this interpretation should be cautious. The FW18 and SW18 groups were independent groups rather than the same individuals measured repeatedly, and they differed in experimental history, cohort, and fasting duration. In addition, plasma osmolality, ion concentrations, and other osmoregulatory indicators were not measured. Therefore, the present data cannot distinguish stable seawater-acclimated physiology from residual effects of prior holding history or transitional physiological state. Moreover, low-flow MO_2 was measured at 0.1 m s^{-1} and was not assumed to represent true resting or standard metabolic rate.

Active-speed-derived variables in the freshwater–seawater comparison were particularly limited because only one Y2 eel under FW18 contributed valid or boundary-estimate values for U_{opt} , $minCOT$, and MO_2 at U_{opt} . Accordingly, the absence of clear differences in these variables should not be interpreted as evidence that salinity has little effect on active swimming energetics. Instead, the freshwater–seawater comparison should be regarded as exploratory and as a basis for future fully factorial experiments using matched individuals or larger independent groups.

4.4. Stage-Associated Comparison and Body-Size Confounding

Under SW18, no clear stage-associated differences were detected between Y2 and S1 eels in U_{crit} , $minCOT$, MO_2 at U_{opt} , or low-flow MO_2 . U_{opt} was numerically higher in S1 eels in some analyses, but the confidence intervals were wide and the result was not robust after FDR adjustment. Exploratory body-size-adjusted analyses also did not provide clear evidence for stage-associated differences.

These results should not be interpreted as evidence that silvering has no effect on swimming physiology. In the present dataset, silvering stage and body size were partially confounded, and Y2 and S1 eels may also have differed in physiological maturity [24]. Therefore, the comparison cannot fully separate stage-related changes from size-related

effects. In addition, the S1 individuals represented an early silvering stage, and later migratory stages may show different physiological patterns.

Previous studies on anguillid eels have reported mixed stage-related patterns in swimming or metabolic traits. For example, Holmberg and Saunders [20] reported no clear difference in oxygen consumption between yellow- and silver-stage American eels, whereas Quintella et al. [21] reported higher U_{crit} in silver European eels than in yellow eels. The absence of clear Y2–S1 differences in the present study may reflect species differences, the early silvering stage examined here, or methodological differences in swim-test protocols. In particular, the present protocol used 2-h velocity steps without external stimulation and therefore emphasized prolonged aerobic swimming performance rather than short-term maximal performance.

Silvering may also involve traits not directly captured by the present swim-tunnel endpoints, such as visual function, buoyancy regulation, endocrine state, orientation behaviour, and motivation to migrate. Previous studies have reported silvering-related changes such as development of the rete mirabile in the swim bladder wall [7] and increased activity associated with endocrine changes, including elevated 11-ketotestosterone [25]. Thus, the present results only indicate that clear differences in the measured aerobic swimming endpoints were not detected between Y2 and S1 eels under the SW18 conditions tested.

4.5. Implications for Spawning Migration in Japanese Eels

The comparison between laboratory-derived swimming parameters and field-estimated swimming speeds provides useful ecological context for interpreting spawning migration in Japanese eels. Under SW18, the laboratory-derived TL-standardized U_{opt} and U_{crit} values were higher than active horizontal swimming speeds reported for migrating silver Japanese eels in the field [14]. This pattern suggests that migrating eels may often move at horizontal speeds lower than laboratory-estimated U_{opt} . However, field estimates reflect movement under complex oceanic conditions and may be influenced by ocean currents, vertical migration, orientation behaviour, and thermal gradients, as indicated by previous tracking studies [9–12,14,26]. Therefore, the laboratory–field comparison should be viewed as a plausible ecological interpretation rather than direct evidence that migrating eels swim below their physiological optimum.

Moving at horizontal speeds lower than laboratory-estimated U_{opt} could be consistent with an energy-saving migration strategy if part of the horizontal displacement is provided by ocean currents or if diel vertical migration allows eels to exploit different current or thermal environments. Because Japanese eels are thought not to feed during the spawning migration [17], conserving endogenous energy reserves is likely important for successful migration and reproduction. Nevertheless, the present swim-tunnel experiments were conducted under controlled, unidirectional flow and cannot reproduce the full behavioural and physical complexity of oceanic migration. Future studies combining swim-tunnel physiology, field tracking, ocean-current reconstruction, and bioenergetic modelling will be needed to evaluate how Japanese eels balance swimming effort, current assistance, and energy conservation during spawning migration.

4.6. Limitations and Future Directions

Several limitations should be considered when interpreting the present results. First, the experiment was not fully factorial. Temperature-associated differences were evaluated only in freshwater Y1 and Y2 eels, the freshwater–seawater comparison was limited to Y2 eels at 18 °C, and the stage-associated comparison was limited to Y2 and S1 eels under SW18. Therefore, the present design cannot disentangle the independent effects of temperature, salinity, silvering stage, body size, cohort, and experimental history. In

addition, the freshwater trials were conducted in a fixed order, with FW25 always preceding FW18. Consequently, the FW25–FW18 comparison cannot separate temperature-associated differences from possible effects of trial order, recovery from the first swimming trial, and fasting history.

Second, sample sizes were small, and the number of individuals contributing active-speed-derived variables decreased in several comparisons. This limitation was particularly important for U_{opt} , $minCOT$, and MO_2 at U_{opt} , because these variables depended on the availability of multiple active-speed COT values. U_{opt} was determined using a discrete-minimum approach, and estimates located at the lowest or highest available active swimming speed were treated as boundary estimates. Only valid sensitivity analyses therefore resulted in very small sample sizes in some comparison. In addition, 0.4 m s^{-1} was treated as the first active swimming speed in the experimental protocol, but this velocity may already represent a relatively high sustained swimming speed for smaller Y1 individuals. Thus, a COT minimum at speeds below 0.4 m s^{-1} could have been missed in some fish.

Third, aspects of holding history and physiological state could not be fully controlled or reconstructed. The 2011 and 2012 eels came from different cohorts, and the exact capture month and pre-acquisition holding conditions of the 2011 cohort were unavailable. These cohort- and history-related differences may have influenced baseline physiological state and swimming performance. For the freshwater trials, fasting began before the FW25 trial and continued through the subsequent FW18 transition/acclimation period. Therefore, fasting duration differed between the two freshwater trials. In addition, osmoregulatory indicators such as plasma osmolality, ion concentrations, or gill ionoregulatory markers were not measured. Thus, the freshwater–seawater comparison cannot distinguish stable seawater-acclimated physiology from transitional or history-dependent physiological states.

Fourth, several methodological limitations of the respirometry should be acknowledged. Low-flow MO_2 measured at 0.1 m s^{-1} was an operational measurement required for oxygen-sensor function and was not assumed to represent true resting or standard metabolic rate. Background microbial respiration was not measured directly, and no blank-chamber correction was applied. The absence of blank-chamber correction may have affected not only absolute MO_2 estimates but also between-condition contrasts if background respiration differed with temperature or water medium. Accordingly, some portion of the observed FW25–FW18 and FW18–SW18 differences in low-flow MO_2 may reflect condition-dependent chamber respiration rather than fish metabolism alone. The direction and magnitude of this potential bias cannot be determined retrospectively. Because COT was calculated from uncorrected MO_2 , background respiration may also have affected $minCOT$ and U_{opt} estimates, particularly at the lower active swimming speeds. Solid-blocking correction was also not applied and may have introduced additional uncertainty in effective flow speed. Sensor calibration, inspection of oxygen-decline traces, repeated water exchange, and temperature and salinity compensation were used to improve measurement consistency; however, these procedures cannot substitute for direct blank-chamber measurements.

Finally, the comparison between laboratory-derived swimming parameters and field-estimated swimming speeds should be interpreted as ecological context rather than a direct test of migratory swimming strategy. Field estimates of active horizontal movement may be influenced by ocean currents, vertical migration, orientation behaviour, and thermal gradients, whereas the present swim-tunnel experiments were conducted under controlled, unidirectional flow. Future studies should use fully crossed experimental designs, larger sample sizes, randomized or counterbalanced trial order, standardized fasting duration, direct osmoregulatory measurements, and blank-corrected respirometry. Integrating swim-tunnel physiology with field tracking, ocean-current reconstruction, and bioenergetic

modelling will also be important for testing the mechanisms suggested by the present exploratory contrasts.

5. Conclusions

This study provides baseline swim-tunnel respirometry data for Japanese eels across selected environmental and early migratory-stage contrasts within the constraints of the experimental design. In the sequential freshwater trials, U_{crit} and low-flow MO_2 were numerically higher under FW25 than under FW18, but these differences could not be attributed solely to temperature because the trial order and fasting history were not independent of condition. In the exploratory Y2 freshwater–seawater comparison at 18 °C, SW18 was associated with higher low-flow MO_2 than FW18, whereas active-speed-derived variables were limited by small sample size. Under SW18, no clear stage-associated differences were detected between Y2 and S1 eels, and body-size-adjusted analyses did not provide clear evidence for stage-associated differences.

These findings suggest that measured oxygen consumption and swimming performance may vary across environmental and developmental contexts in Japanese eels, but they do not provide definitive tests of independent temperature, salinity, or silvering-stage effects. The comparison with field-estimated swimming speeds provides useful ecological context and suggests that migrating Japanese eels may often move at horizontal speeds lower than laboratory-estimated U_{opt} . However, this interpretation should be regarded as plausible rather than definitive, because field speeds are influenced by ocean currents, vertical movements, orientation behaviour, and thermal gradients. Future studies using fully crossed designs, larger sample sizes, counterbalanced trial order, standardized fasting, direct osmoregulatory measurements, and integrated field-tracking and bioenergetic modelling will be needed to clarify how Japanese eels balance swimming performance, energetic cost, and energy conservation during spawning migration.

Supplementary Materials: The following are available online at <https://www.mdpi.com/article/10.3390/fishes11080437/s1>, Figure S1: Blazka-type swim-tunnel respirometry system used in the experiments; Table S1: Individual-level swimming and metabolic endpoints for each trial; Table S2: Speed-step-level MO_2 data for each trial; Table S3: Speed-step-level COT data for each trial; Table S4: Statistical summary of predefined individual-level contrasts; Table S5: Exploratory body-size sensitivity analyses for the SW18 Y2–S1 comparison.

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Institutional Review Board Statement: The experiments were conducted in 2011–2012, when fish were not subject to mandatory animal-experiment plan review under the applicable Japanese national guidelines. Nevertheless, all fish sampling, handling, and experimental procedures were conducted in accordance with the University of Tokyo guidelines for animal care and use, including the “Principles of morality in animal experiments” of the University of Tokyo (ethics protocol 4-3-3 and 4-3-4).

Informed Consent Statement: Not applicable.

Data Availability Statement: The processed individual-level and speed-step-level datasets used for the statistical analyses are provided in the Supplementary Information. The Python scripts used to generate the statistical summaries and figure data are available in the GitHub repository at https://github.com/TakatoshiHiguchi/swim-tunnel_respirometry_experiments_in_Japanese_eels (accessed on 21 July 2026). Raw oxygen time-series data are available from the corresponding author upon reasonable request.

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