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Relation	



1 **Diel feeding rhythms, daily ration, and seasonal changes thereof in marbled flounder**

2 *Pseudopleuronectes yokohamae*

3

4 Running title: Daily ration of marbled flounder

5

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The authors have no conflict of interest to declare.

14 **Summary**

15 This paper aims to assess the diel feeding pattern and seasonal variation in the daily ration of  
16 immature and mature marbled flounder (*Pseudopleuronectes yokohamae*). A day-night  
17 collection by bottom trawls was conducted in Sendai Bay in July 2014. Marbled flounder  
18 (131–493 mm total length; N of collected individuals = 1830, N of analyzed individuals =  
19 497) fed mainly on polychaetes during the day. At night, stomach content weight decreased  
20 with time, but the weight and proportion of bivalve siphons were consistently higher at night  
21 than during the day, suggesting nocturnal feeding by the flounder on bivalve siphons. Daily  
22 ration was greater in females (<300 mm: 2.6–3.2% body weight; ≥300 mm: 1.5–2.5%) than in  
23 males (<300 mm: 1.7–2.6%; ≥300 mm: 1.3–1.9%). Seasonal surveys were also carried out,  
24 and the greater ration in females than males were consistent throughout the year, suggesting  
25 that greater growth in females than males attributes to the greater food intake of females. The  
26 ration was highest in June, especially for large individuals, although water temperature in  
27 June was lower than that in September. These results indicate that the amount of food intake  
28 is related to the annual life cycle of the marbled flounder.

29

30 1 Introduction

31

32 Assessment of food consumption by predators is an important ecological issue for  
33 understanding energy flow and ecosystem structures. Daily food demand can be estimated in  
34 laboratory experiments, but daily food consumption in the field generally needs day-night  
35 field collections of the predator at small intervals (Elliott & Persson, 1978). Furthermore,  
36 seasonal dynamics of food consumption should be investigated to reveal annual life cycle of  
37 the predator.

38 Pleuronectid flatfishes are visual diurnal predators that use chemical and mechanical  
39 senses (de Groot, 1969, 1971). Diel feeding rhythms of pleuronectid flatfishes have been  
40 studied in juveniles (Thijssen, Lever, & Lever, 1974; Chen, Purser, & Blyth, 1999; Hurst,  
41 Ryer, Ramsey, & Haines, 2007), immature, and adults (Worobec, 1984; Yang & Livingston,  
42 1988). However, only limited studies have been performed regarding daily food intake and  
43 daily ration. The food consumption by flatfish is essential in understanding the role of  
44 predators in benthic ecosystems as they are often an important component.

45 The present study aimed to reveal diel feeding patterns and daily rations of immature and  
46 mature marbled flounder (*Pseudopleuronectes yokohamae*). This species is a common and  
47 important flatfish distributed in the coastal waters of Japan (Minami, 1981; Tomiyama, 2013).  
48 In Sendai Bay, northern Japan, the great earthquake and associated tsunami and resulting  
49 accident in the nuclear power plants in March 2011 considerably affected the coastal fisheries.  
50 Owing to the sudden decrease in fishing pressure, the stocks of commercially important fishes,  
51 such as Japanese flounder (*Paralichthys olivaceus*) and Pacific cod (*Gadus macrocephalus*),  
52 have greatly increased (Narimatsu, Shibata, Hattori, Yano, & Nagao, 2017; Shibata et al.,  
53 2017). Under such circumstances, studies on food web dynamics in these areas are of  
54 importance. We first conducted a survey cruise for day and night collections of marbled  
55 flounder to assess diel feeding patterns. We then conducted seasonal surveys to assess annual

56 changes in the daily ration of marbled flounder. Body size and sex were taken into  
57 consideration when assessing variations in daily ration.

58

## 59 2 Materials and methods

60

### 61 2.1 Study site and field surveys

62

63 Field collections of marbled flounder were conducted in Sendai Bay, northeastern Japan  
64 (Figure 1). Marbled flounder is a common flatfish in this area.

65 A day-night survey was carried out at a site (38° 12' N; 141° 13' E; 37 m deep; Figure 1)  
66 in July 2014. Otter trawls with 5.4-m wide mouth-openings, length of 44 m, and 50-mm mesh  
67 net covered with 8-mm mesh at cod-end were towed by RV Wakatakamaru (692 tons) at ca.  
68 3.0 knots for 30 min. This trawl sampling was repeated at 3–4 h intervals in day  
69 (06:00–19:00) and night (21:00–04:00) surveys. The day survey was conducted on two days,  
70 and nocturnal survey was conducted on one night (total number of tows = 11, Table 1). At  
71 each tow, 74–339 individuals of marbled flounder were collected. Of these samples, 65–120  
72 individuals/tow were randomly selected and measured (total length [TL], mm) onboard  
73 immediately after collection. Subsamples of 15–36 individuals/tow were used for stomach  
74 content analysis with sex determination from the gonads: stomachs of the flounder were  
75 removed onboard and preserved in 10% formalin for later observation to minimize a decrease  
76 in stomach contents weight by autolysis after collection. Other additional subsamples (30–41  
77 individuals/tow) were frozen onboard for later measurements. Bottom water temperature was  
78 measured at each tow by AAQ175 (JFE Advantech Co., Ltd., Nishinomiya, Japan).

79 To determine seasonal changes in the food intake of marbled flounder, daytime surveys  
80 were conducted at six stations at depths of 35–80 m in Sendai Bay (Figure 1) from February  
81 to November 2014. This survey was carried out approximately once every three months. The

82 otter trawls were towed at ca. 3.0 knots for 30 min at each station during 05:30–12:00 (total  
83 number of tows = 21). Marbled flounder were collected and brought to the laboratory under  
84 chilled conditions. Then, fish were frozen for later measurements. Bottom water temperature  
85 was measured at each tow by AAQ175.

86

## 87 2.2 Measurements

88

89 In the day-night survey, individuals frozen onboard were measured in the laboratory. After  
90 being defrosted, TL and the body wet weight (BW, g) of each fish were measured, their sex  
91 was identified from the gonadal observation, and their stomachs were removed and preserved  
92 in 10% formalin. BW of flounder of which stomachs were preserved onboard was estimated  
93 based on the relationship between BW and TL estimated from flounder frozen onboard:  $BW =$   
94  $7.055 \times 10^{-6} \times TL^{3.102}$  ( $n = 173$ ,  $r^2 = 0.99$ ). Stomachs of a total of 497 individuals were used.  
95 Stomach contents were blotted with paper towels to remove excess water. Effects of  
96 preservation in 10% formalin were regarded as negligible. Preliminarily, the weight of  
97 stomachs was measured for 10 individuals before and 1 mo after the formalin preservation,  
98 and it did not change significantly (paired  $t$ -test,  $t = 0.88$ ,  $p = .40$ ). The stomach content wet  
99 weight (SCW) was determined to the nearest 0.01 g. Stomach contents were sorted into the  
100 following categories and weighed: polychaetes, bivalve siphons, bivalves, opisthobranchs,  
101 actinarians, crustaceans, and others.

102 For the seasonal survey samples, fish were defrosted and their TL and BW were  
103 measured similarly. Sex was identified from the gonads. Total SCW was determined  
104 similarly.

105

## 106 2.3 Analyses

107

108 Prey composition was assessed on the basis of the percentage weight of each prey category.  
109 To test whether diet composition varied between day and night, between sexes, and between  
110 the size classes of marbled flounder, a multivariate analysis of variance (MANOVA) was  
111 carried out. The percentage weight of each prey category was determined for each individual  
112 flounder. The data were arcsine square root transformed. Series of net tows (tows #1–3 [night],  
113 4–7 [day], or 8–11 [day]), sex (male or female), and TL were used as explanatory variables.

114 Diel feeding rhythms were assessed based on changes in empty stomach rates, stomach  
115 content weight, and prey composition. To analyze empty stomach rates, we constructed a  
116 generalized linear mixed model (GLMM) with binomial family and logit-link function. The  
117 response variable was presence/absence of stomach contents; initial explanatory variables  
118 were day/night time, sex (male/female), methods (measured onboard/frozen onboard), and TL  
119 of fish; the random variable was the tow number.

120 The stomach content weight index (SCI) was used to standardize the stomach contents  
121 data:  $SCI = SCW \times BW^{-1} \times 100$ . To analyze variation in the SCI, we constructed a GLMM  
122 with Gaussian family and log-link function, following Hattori, Okuda, Narimatsu, Ueda, &  
123 Ito (2009) with some modifications: the response variable was  $[SCW + 0.00001]$ , initial  
124 explanatory variables and random variable were same as those in the GLMM for empty  
125 stomach rates, and  $\log(BW)$  was used as an offset term. Because the SCW involves zero data,  
126 we added a negligible value of 0.00001 to the SCW to allow logarithm transformation. The  
127 final model was selected based on the Akaike information criterion (AIC). Similarly, we  
128 constructed a linear mixed model (LMM) for the weight of bivalve siphons in the stomachs of  
129 marbled flounder, using the same initial explanatory variables except methods and the random  
130 variable.

131 To estimate the daily ration (*DR*: percentage to the body weight), gastric evacuation rate  
132 (*GER*) should be determined. It was calculated from the water temperature (*WT*, °C),  
133 following Durbin, Durbin, Langton, & Bowman (1983):



134 
$$GER = 0.0406 \times e^{0.111 \times WT} \quad (1)$$

135 To test the validity of estimated GER, changes in the SCI from the dusk (tows # 7 and  
 136 11) to dawn during the night (tows # 1–3) in the day-night survey were analyzed.

137 Preliminarily, weight of the bivalve siphons were subtracted from stomach contents weight  
 138 because the flounder seemed to consume bivalve siphons during the night (see results). The  
 139 SCI that does not include the weight of bivalve siphons was defined as SCI'. The linear model  
 140 for the SCI' was constructed; the [SCI' + 0.1] was natural-logarithm transformed because  
 141 SCI' involves zero data. The constant of 0.1 was used because the regression coefficient was  
 142 the highest between using 1, 0.5, 0.1, 0.05, and 0.01. Initial explanatory variables were time  
 143 from 18:00 (h), sex, and body size class ( $\geq 300$  and  $< 300$  mm TL). The final model was  
 144 selected based on the AIC. The coefficient of the time from 18:00 was regarded as the GER.

145 In the day-night survey, we determined the *DR* of marbled flounder of two size groups  
 146 ( $\geq 300$  mm TL and  $< 300$  mm TL) across sexes using the Elliott & Persson model (Elliott &  
 147 Persson, 1978), as follows:

148 
$$DR = \sum C_t \quad (2)$$

149 
$$C_t = (S_t - S_0 \times e^{-GER \times t}) \times GER \times t \times (1 - e^{-GER \times t})^{-1} \quad (3)$$

150 where  $C_t$  is the ration from time 0 to time  $t$  (h),  $S_t$  and  $S_0$  are the mean SCI at times  $t$  and 0,  
 151 respectively. Empty stomachs were included for calculating both  $S_t$  and  $S_0$ . The SCI data from  
 152 both specimens measured with their stomachs preserved in formalin onboard and specimens  
 153 frozen onboard and measured in the laboratory were pooled for analysis, because the  
 154 “methods” were excluded from the GLMM explaining the SCW (see results). The  $C_t$  was  
 155 estimated for the time between each successive two tows, e.g. average SCIs of samples from  
 156 tows # 2 and 1 were used as  $S_t$  and  $S_0$ , respectively. The daily ration was determined by  
 157 summing  $C_t$  regardless of whether  $C_t$  was positive or negative. Since the day survey was  
 158 conducted twice (Table 1), the daily ration was determined from two combinations of diurnal

159 and nocturnal datasets, i.e. tows # 1–3 + 4–7 and tows # 1–3 + 8–11 (Table S1, supporting  
160 information).

161 We also assessed seasonal changes in *DR* for the two size groups ( $\geq 300$  mm TL and  $< 300$   
162 mm TL) across sexes of marbled flounder, using the equation following Eggers (1979):

$$163 \quad DR = 24 \times \bar{S} \times GER \quad (4)$$

164 where  $\bar{S}$  is the average SCI over 1 day and GER is the value estimated from the equation (1).

165 The  $\bar{S}$  cannot be determined directly because our seasonal samplings were carried out only in  
166 the morning (from 06:00 to 11:00). Therefore,  $\bar{S}$  was estimated from the average SCI during  
167 the morning. A relationship between  $\bar{S}$  and the average SCI during the morning (tows # 4–5  
168 and 8–9) was obtained from two size groups across sexes of marbled flounder in the day-night  
169 survey:  $\bar{S} = 1.057 \times (\text{SCI during the morning})$  ( $n = 8$ ,  $r = 0.69$ ). We also confirmed that the  
170 *DR* by Eggers was similar to the *DR* estimated by Elliot & Persson model from the day-night  
171 survey ( $[DR \text{ by Elliott \& Persson}] = 1.037 \times [DR \text{ by Eggers}]$ ,  $n = 8$ ,  $r = 0.68$ ). All calculation  
172 for SCI included individuals with empty stomachs.

173

## 174 3 Results

175

### 176 3.1 Feeding habits of marbled flounder

177

178 The size of marbled flounder ranged from 131 to 485 mm TL, with a single peak around  
179 250–300 mm TL (Figure 2). Empty stomach rates were the greatest at the tow conducted at  
180 around 03:00 (Table 1).

181 Marbled flounder fed mainly on polychaetes, bivalves and their siphons, opisthobranchs,  
182 and actinarians (Figure 3). Dietary composition differed significantly between series of net  
183 tows and body size, but not between males and females (MANOVA; series of net tows,  $F_{2,361}$   
184  $= 16.87$ ,  $p < .001$ ; TL,  $F_{1,361} = 6.08$ ,  $p < .001$ ; sex,  $F_{1,361} = 1.82$ ,  $p = .082$ ). The proportion of

185 bivalve siphons was consistently higher during the night than that during the day and this  
186 tendency was clear in flounder of small size classes ( $\geq 300$  mm: 0.2–4.9% and 5.9–48.1%;  
187  $< 300$  mm: 0.8–12.1% and 34.6–57.6% during the day and night, respectively). The weight of  
188 bivalve siphons per individual marbled flounder was also greater during the night than in the  
189 day (Figure 4). In the LMM for the bivalve siphon weight, “day/night” and TL were adopted  
190 as the explanatory variables (“day/night”:  $p < .001$ ; TL:  $p = .08$ ), and sex was eliminated from  
191 the selected model. The proportion of bivalves was also greater at night than in the day ( $\geq 300$   
192 mm: 1.3–12.3% and 3.0–24.8%;  $< 300$  mm: 3.2–10.4% and 1.3–14.2% during the day and  
193 night, respectively). The proportion of opisthobranchs was greater in large individuals  $\geq 300$   
194 mm (2.4–59.3%) than in small individuals (0.3–17.9%).

195

### 196 3.2 Diel feeding rhythm and daily ration

197

198 In the GLMM for empty stomach rates, only “day/night” was adopted as an explanatory  
199 variable. The empty stomach rates were significantly higher during the night than during the  
200 day (GLMM,  $p < .001$ ). In the GLMM for SCW, sex and TL were adopted as explanatory  
201 variables (Table 2). The day/night and methods were excluded from the model (Table S2,  
202 supporting information). From the selected model, the SCI of females and smaller fish was  
203 greater than males and larger fish.

204 Stomach content weight decreased during the night (Figure 5). Average SCI changed  
205 from 0.69 to 0.08, 0.71 to 0.16, and 0.90 to 0.14 in large female, large male, and small male  
206 flounder at night, respectively, although it was relatively high at night ( $> 0.5$ ) in small females.  
207 During the day, it increased as the time passed with a single peak before sunset.

208 Daily ration, as calculated from two datasets, was 1.54 and 2.47% BW in females  $\geq 300$   
209 mm, 1.27 and 1.89% BW in males  $\geq 300$  mm, 2.63 and 3.17% BW in females  $< 300$  mm, and  
210 1.73 and 2.56% BW in males  $< 300$  mm.

211 Average bottom water temperature was 10.5 °C (Table 1). The GER at 10.5 °C, estimated  
212 from the equation (1), was 0.130. On the other hand, the estimated GER from the changes in  
213 SCI' was 0.169 (Figure 6). Preliminarily, the time from 18:00 was only adopted in the  
214 selected linear model for SCI', and sex and the size class was removed from the model. The  
215 estimated daily ration would increase by 30% when the GER of 0.169 was used, compared  
216 with the above data.

217

### 218 3.3 Seasonal variations in daily ration

219

220 Although individuals with empty stomachs were frequently observed from samples, greater  
221 rations in smaller (<300 mm TL) than in larger individuals were generally common, except  
222 for samples collected in February (Table 3). Large individuals, both male and female,  
223 consumed larger amounts of food (>1% BW of daily ration) during the lower temperature  
224 conditions of February–June than during the higher temperatures of September–November  
225 (>16°C, Table 3). However, small females consumed considerable amounts of food (>2%  
226 BW) from June to November, and small males consumed greater amounts in June–September,  
227 even when the temperature in June was low (<9°C). Empty stomachs were observed more  
228 frequently in males than in females. Females generally consumed more prey than males  
229 throughout the year.

230

## 231 4 Discussion

232

233 The diel feeding patterns of immature and mature marbled flounder revealed in the present  
234 study were similar to those reported for immature fish in previous studies (Takahashi,  
235 Tominaga, Maeda, & Ueno, 1982; Tokai & Ito, 1991): their feeding intensity was highest  
236 after sunrise and before sunset. Additionally, the present study constitutes the first report that

237 the food intake of marbled flounder is greater in females than in males throughout the year.  
238 Since their spawning season is around December–January (Hatanaka & Iwahashi, 1953;  
239 Takahashi, Saito, Maeda, & Kimura, 1983; Tanda, Nakamura, & Okamoto, 2008), the sexual  
240 differences in food intake, as observed in the day-night survey in July, would be hardly  
241 related to their reproductive investment. The dietary composition did not differ significantly  
242 between males and females, but food intake of females was greater than males, suggesting  
243 that female fish consume more food than males without changing their prey.

244 The dietary composition differed between day and night, with the most considerable  
245 difference being the greater proportion of bivalve siphons at night. One possible explanation  
246 is that bivalves extend their siphons up to the sediment surface to feed at night, to avoid  
247 siphon predation by visual day feeders (Levinton, 1971). Pleuronectid flatfish are visual day  
248 feeders but they can use sensory organs to detect prey, as well as visual searches (de Groot,  
249 1969, 1971). In fact, juvenile stone flounder (*Platichthys bicoloratus*) usually forage for food  
250 during the day, but can capture prey, such as bivalve siphons, in the dark (Tomiyama,  
251 Katayama, Yamamoto, & Shoji, 2016). Marbled flounder could possibly detect bivalve  
252 siphons that actively feed in the dark. Another possible explanation is the difference in gastric  
253 evacuation rates between prey items, as observed in the field (Rindorf, 2004). However, it is  
254 unlikely that bivalve siphons are difficult to digest by marbled flounder. A greater siphon  
255 weight per individual flounder stomach was observed at night than during the day (Figure 4),  
256 which supports nocturnal feeding by marbled flounder on bivalve siphons. Additionally,  
257 newly bitten siphons were observed in the stomachs of flounder collected at tow #3 (around  
258 03:30), strongly indicating nocturnal feeding on bivalve siphons.

259 The greater daily ration of females than males was consistent throughout the year (Table  
260 3), which could be related to the faster growth and greater maximum size of females than  
261 males (Solomon, Sano, Shimizu, & Nose, 1987; Tanda, Gorie, Nakamura, & Okamoto, 2008).  
262 The greater food intake of females than males was also observed for *Limanda limanda* in the

263 laboratory (Pandian, 1970; Lozán, 1992). The ration of males was extremely low in  
264 November. Gonad weight of male and female fish peaks in November and December,  
265 respectively (Hatanaka & Iwahashi, 1953; Kume, Horiguchi, Goto, Shiraishi, Shibata, Morita,  
266 & Shimizu, 2006; Tanda, Nakamura, & Okamoto, 2008), indicating that male fish could  
267 reduce their feeding activity and prepare for reproduction earlier than females. High food  
268 consumption under preferable temperature conditions has been observed in pleuronectid  
269 flatfishes (Pandian, 1970; Worobec, 1984; Kusakabe, Hata, Shoji, Hori, & Tomiyama, 2017),  
270 and food consumption of marbled flounder of 200 mm increases as temperature increases  
271 from 2 to 24°C under laboratory conditions (Takahashi, Tominaga, & Maeda, 1987).

272 The daily ration was greatly affected by the GER. This study consistently used the GER  
273 estimated from the literature (Durbin et al., 1983) to apply it to the seasonal data, because the  
274 GER is temperature-dependent and the GER estimated from our day-night survey in July is  
275 not applicable to estimate the daily ration of fish in other seasons with different temperatures.  
276 However, the literature-derived value was lower than that estimated from the day-night  
277 survey on the assumption that flounder consume no food except bivalve siphons during the  
278 night. As the GER would also be affected by the food type or size (Durbin et al., 1983; He &  
279 Wurtsbaugh, 1993), the estimation of GER should be developed to increase the accuracy of  
280 the daily ration.

281 Daily ration estimated in the present study was <3% BW, which was lower than that in  
282 the laboratory experiment, 4–10% BW at 10–25°C (Takahashi et al., 1987). The lower values  
283 in the present study may be related to the high stock level. In another locality,  
284 density-dependent effects in the growth of marbled flounder have been observed (Lee et al.,  
285 2009). The stock of marbled flounder has increased considerably in the study area because of  
286 the temporal termination and subsequent reduction of fishing efforts of coastal fisheries after  
287 the environmental disaster in 2011. Therefore, their food intake and growth could be similarly  
288 reduced, although the main prey for marbled flounder were polychaetes and opisthobranchs

289 and were similar to the past studies (Omori, 1974; Takahashi et al., 1982). It is necessary to  
290 determine whether the ration estimated in this study changes with various stock levels.  
291 Furthermore, the impact of predation by increased demersal fishes on benthic ecosystems  
292 should also be studied.

293 This study revealed the food intake of marbled flounder in relation to season, body size,  
294 and sex. This information is expected to contribute to future studies revealing the flow of  
295 energy and radioactive materials in ecosystems of the study area.

296

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415

416 **TABLE 1** Cruise details for the day-night surveys in 2014

Tow #	Date	Time of net tows	BWT (°C)	N of collected fish	N of fish analyzed			
					Female $\geq 300$ mm TL	Female $< 300$ mm TL	Male $\geq 300$ mm TL	Male $< 300$ mm TL
1	July 6	21:13–21:45	10.7	183	5 (0)	23 (3)	9 (1)	17 (1)
2	July 7	0:38–1:09	10.3	339	6 (1)	2 (0)	1 (0)	6 (2)
3	July 7	3:15–3:45	10.5	243	14 (9)	17 (5)	9 (4)	30 (18)
4	July 8	6:59–7:29	10.2	154	10 (3)	17 (0)	9 (0)	27 (2)
5	July 8	10:08–10:39	10.5	129	9 (1)	2 (0)	4 (1)	18 (1)
6	July 8	14:34–15:04	10.7	74	14 (0)	22 (3)	2 (1)	36 (4)
7	July 8	18:06–18:36	10.5	144	14 (1)	17 (4)	8 (0)	28 (4)
8	July 9	7:03–7:35	10.5	123	4 (1)	7 (0)	3 (1)	17 (0)
9	July 9	10:07–10:47	10.2	177	5 (0)	5 (0)	6 (0)	14 (0)
10	July 9	14:25–14:54	10.5	130	3 (0)	6 (0)	7 (0)	14 (0)
11	July 9	18:23–18:52	10.6	134	2 (0)	6 (0)	9 (0)	13 (0)

417 Numerals in parentheses show the number of individuals with empty stomachs. Sunrise and  
 418 sunset were at 04:20 and 19:03, respectively, on July 8. N = the number of individuals of  
 419 marbled flounder (*Pseudopleuronectes yokohamae*); BWT = bottom water temperature; TL =  
 420 total length.

421

422 **TABLE 2** Results of the generalized linear mixed model for stomach content weight index  
 423 (SCI) of marbled flounder (*Pseudopleuronectes yokohamae*) in the day-night surveys in 2014

Analysis of deviance				Summary			
Error source	df	Chisq	<i>p</i>	Parameter	Estimate	SE	<i>p</i>
				Intercept	-4.20	0.25	< .001
Sex	1	12.30	< .001	Sex (male)	-0.23	0.07	< .001
TL	1	26.17	< .001	TL	-0.0027	0.0005	< .001

424 Analysis of deviance was operated by the Type II Wald chi-square tests. Response variable  
 425 was stomach contents weight (SCW) + 0.00001. Initial explanatory variables were day/night,  
 426 sex, preservation method, and total length (TL). Tow number was included as a random  
 427 variable. Day/night and the preservation method were excluded after model selection based on  
 428 the Akaike information criterion. Effect of males was assessed on the basis of females.

429

430

431 **TABLE 3** Seasonal surveys to determine the daily ration of marbled flounder  
 432 (*Pseudopleuronectes yokohamae*) in 2014

Season	N of fish analyzed	SCI (%BW)	Daily ration (%BW)
$\geq 300$ mm TL, female			
Feb	27 (5)	$0.67 \pm 0.66$	1.71
June	51 (8)	$0.75 \pm 0.69$	2.03
Sep	23 (13)	$0.15 \pm 0.21$	0.98
Nov	30 (8)	$0.19 \pm 0.19$	1.22
$\geq 300$ mm TL, male			
Feb	7 (3)	$0.40 \pm 0.58$	1.03
June	15 (2)	$0.66 \pm 0.75$	1.79
Sep	7 (5)	$0.06 \pm 0.11$	0.41
Nov	15 (14)	$0.01 \pm 0.05$	0.09
$< 300$ mm TL, female			
Feb	23 (8)	$0.36 \pm 0.43$	0.91
June	25 (6)	$0.83 \pm 1.04$	2.26
Sep	23 (4)	$0.43 \pm 0.43$	2.90
Nov	9 (0)	$0.42 \pm 0.24$	2.73
$< 300$ mm TL, male			
Feb	24 (13)	$0.14 \pm 0.18$	0.35
June	34 (6)	$0.72 \pm 0.52$	1.97
Sep	30 (7)	$0.29 \pm 0.26$	1.94
Nov	17 (11)	$0.11 \pm 0.19$	0.70

433 Numerals in parentheses show the number of individuals with empty stomachs. Stomach  
 434 content weight index (SCI) is presented as mean  $\pm$  standard deviation, calculated including  
 435 individuals with empty stomachs. Both SCI and daily ration are shown by percentage to body  
 436 weight (%BW). Survey dates were February 2 and 22 for “Feb”, June 15 and 21 for “June”,  
 437 August 30 and September 13 for “Sep”, and November 11 for “Nov”. Average bottom water  
 438 temperatures was 8.2, 8.8, 16.9, and 16.5°C for Feb, June, Sep, and Nov, respectively.

439

440 Figure captions

441

442 FIGURE 1. Map of the study area in Sendai Bay, Japan. An open triangle indicates the station  
443 for the day-night survey in July 2014. Solid circles indicate stations for seasonal surveys  
444 in 2014.

445

446 FIGURE 2. Length-frequency distribution of marbled flounder (*Pseudopleuronectes*  
447 *yokohamae*) collected in the day-night survey in July 2014. Open and shaded bars denote  
448 female and male fish, respectively.

449

450 FIGURE 3. Diet composition (proportion in wet weight) of marbled flounder  
451 (*Pseudopleuronectes yokohamae*) (a)  $\geq 300$  mm TL and (b)  $< 300$  mm TL in the day-night  
452 survey. Solid and open horizontal bars indicate night and day, respectively. Numerals  
453 above bars indicate sample sizes without empty stomachs. Abbreviations are as follows:  
454 Po, polychaetes; Si, bivalve siphons; Bi, bivalves; Op, opisthobranchs; Ac, actinarians;  
455 Cr, crustaceans; Ot, others.

456

457 FIGURE 4. Weight of bivalve siphons in the stomachs of (a) female and (b) male marbled  
458 flounder (*Pseudopleuronectes yokohamae*). Large and small indicate flounder  $\geq 300$  and  
459  $< 300$  mm TL, respectively. Numerals indicate sample sizes including individuals with  
460 empty stomachs.

461

462 FIGURE 5. Stomach content weight index (SCI) of (a) female and (b) male marbled flounder  
463 (*Pseudopleuronectes yokohamae*) in the day-night survey. Solid triangles with dotted  
464 lines and open circles with solid lines indicate flounder  $\geq 300$  mm TL and  $< 300$  mm TL,  
465 respectively. Solid and open horizontal bars indicate night and day, respectively. Vertical



466 bars denote standard deviation. The data include fish with empty stomachs. Sample sizes  
467 are shown in Table 1.

468

469 FIGURE 6. Changes in the stomach contents weight index determined without bivalve  
470 siphons (SCI') in marbled flounder (*Pseudopleuronectes yokohamae*) collected from  
471 18:00 to 04:00. Solid and open horizontal bars indicate night and day, respectively. The  
472 data were natural logarithm transformed with adding 0.1. The regression was fitted to the  
473 data:  $\log(\text{SCI}' + 0.1) = -0.169 \times \text{Time} - 0.0356$  ( $r^2 = 0.38$ ).

474

Fig. 1

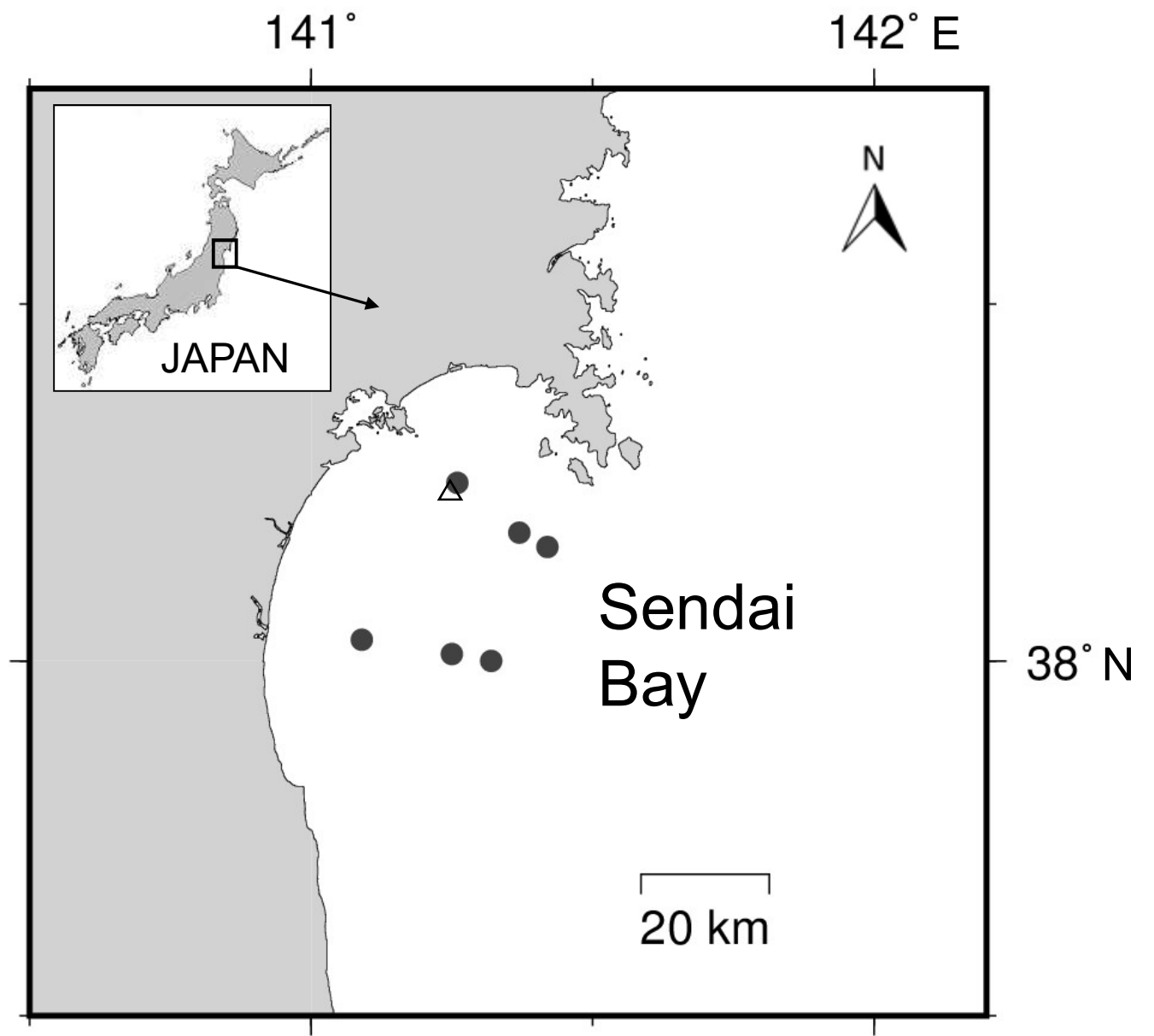


Fig. 2

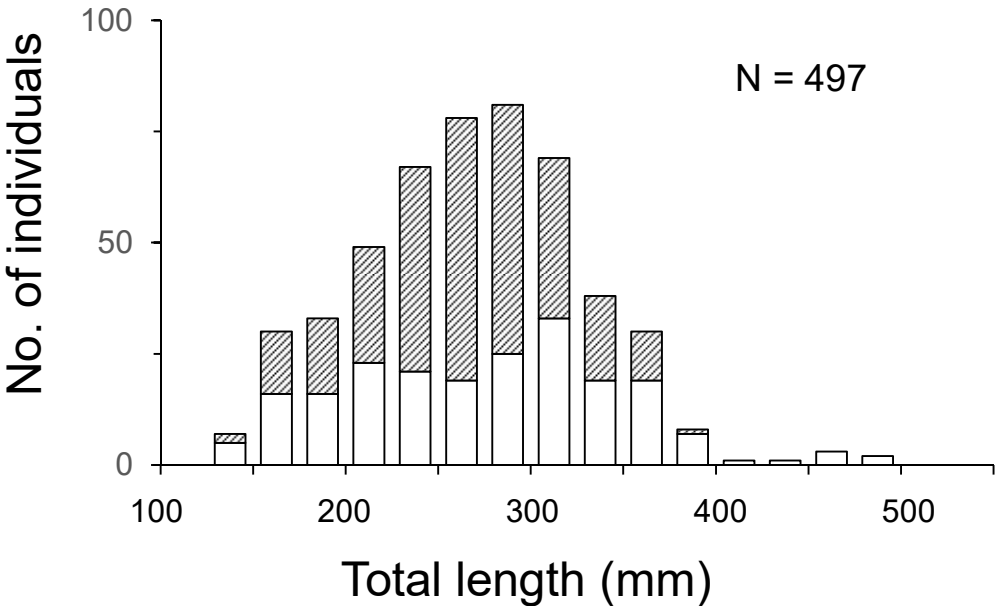


Fig. 3

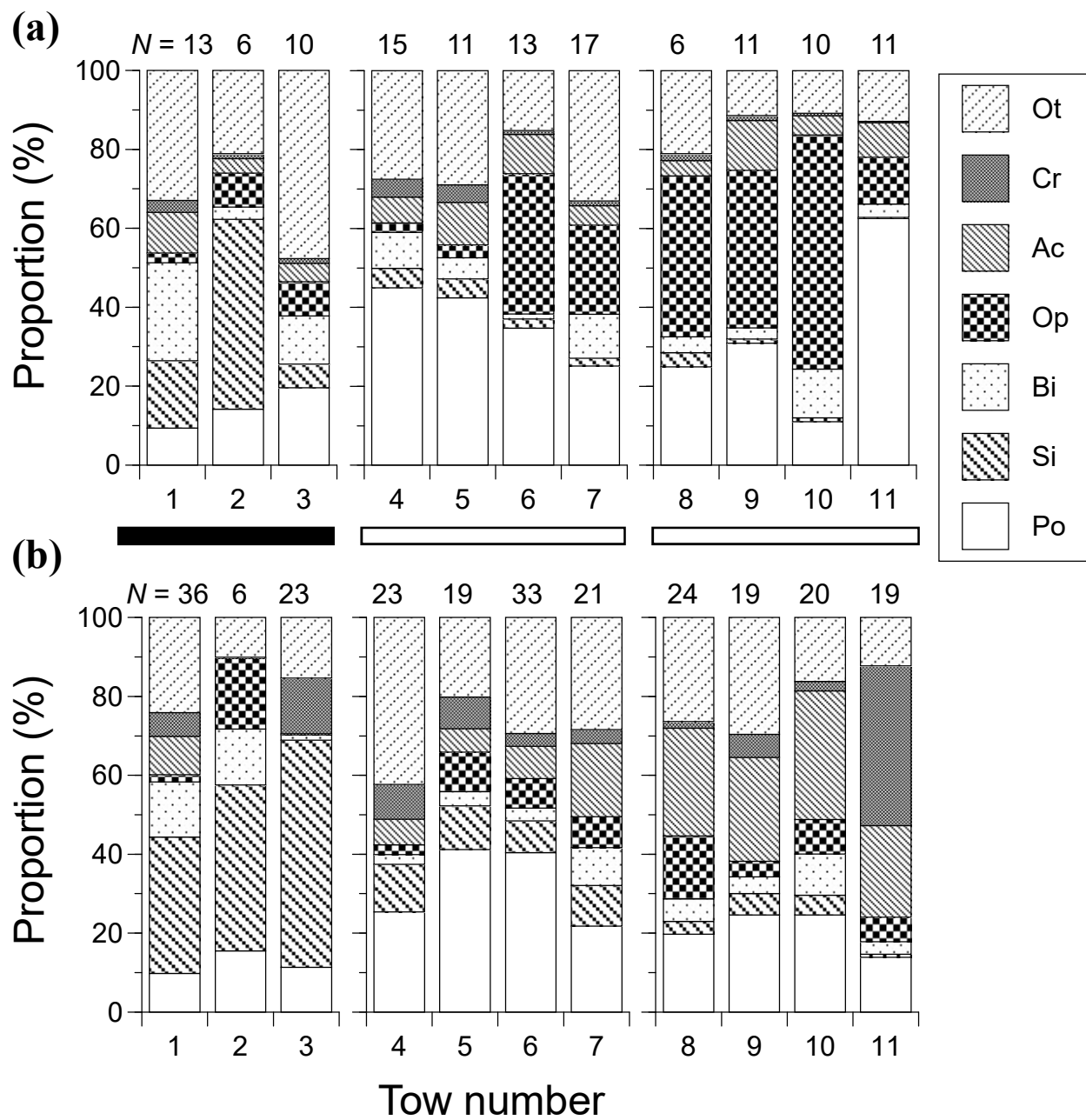


Fig. 4

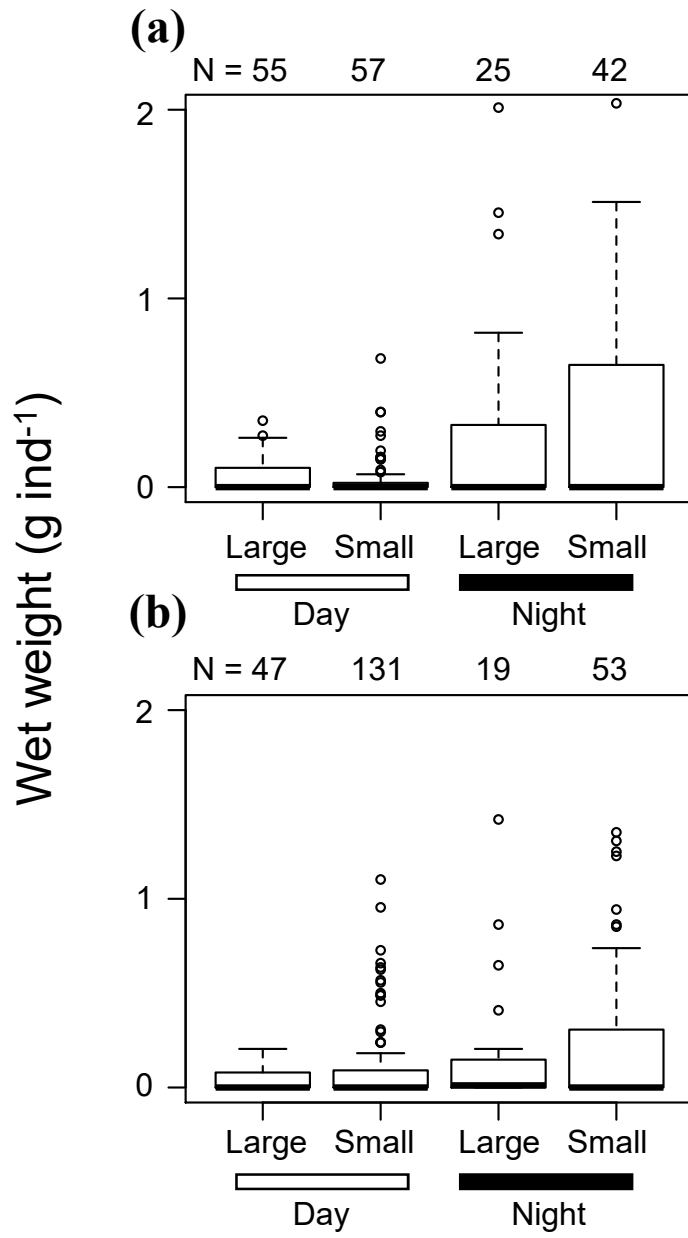


Fig. 5

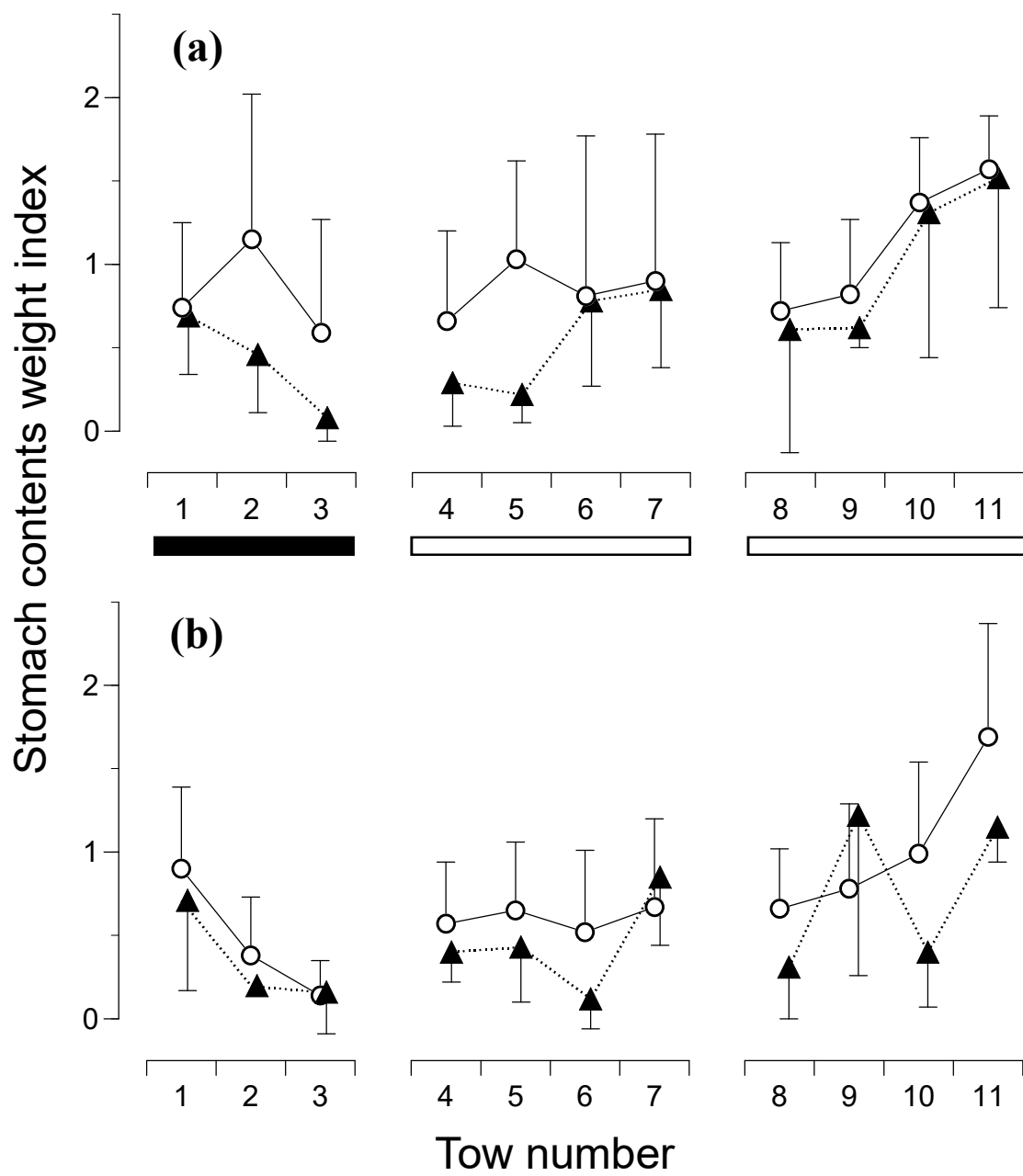


Fig. 6

