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Annual Life Cycle of Adult Marbled Sole *Pleuronectes yokohamae* in the Coastal Waters around Kikonai Bay, Hokkaido, Japan

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Abstract: To clarify the annual life cycle of adult marbled sole *Pleuronectes yokohamae* in the coastal waters around Kikonai Bay, the seasonal changes in gonadosomatic index, hepatosomatic index, and condition factor of samples obtained with commercial set nets and research bottom trawl nets during May 1994 through October 1996 were examined. In addition, feeding intensity was investigated using a stomach contents index and noting the percentage of empty stomach. The annual life cycle of adult *P. yokohamae* was divided into the following three periods: spawning (February－April), active feeding and recovering (May－July), and inactive feeding (August－January). The inactive feeding period was further divided into two sub-periods: immature (August－October) and mature (November－January). It seems likely that *P. yokohamae* in this study area feeds intensively after spawning and matures under these high levels of nutrition.

Key words: *Pleuronectes yokohamae*; Annual life cycle; Spawning season; Feeding intensity

Marbled sole *Pleuronectes yokohamae* (Günther) is an important fishery resource in the coastal waters of Japan. The northernmost fishing grounds of this species are Kikonai Bay and the adjacent coastal waters of the Tsugaru Strait between Fukushima and Hakodate. Ishino and Sano (1996) reported that *P. yokohamae* around the Tsugaru Strait could be divided into four local stocks- Tsugaru Peninsula, Mutsu Bay, Shimokita Peninsula, and Kikonai Bay stock- based on the irrespective spawning periods and isozymes of the GPI antifreeze protein. Annual landings of *P. yokohamae* in the waters around Kikonai Bay varied between 173 and 349 metric tons from 1985 through 2002 (Ishino 1997; Nakaya 2004), which were higher than values obtained for the adjacent stocks in the Tsugaru Strait.

As for *P. yokohamae* in the waters around Kikonai Bay, food habits (Takagi et al. 1997) and growth of immature and adult fish (Shafieipour et al. 1999), food habits (Nakagami et al. 2000) and spatial distribution of larvae and juveniles (Nakagami et al. 2001), and predation by shrimp (Nakaya et al. 2004) have been reported. However, a detailed study on

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the annual life cycle has not been conducted. In this study we examined the annual life cycle of adult *P. yokohamae*, based on seasonal changes in gonadosomatic index, hepatosomatic index, condition factor, stomach contents index, and the percentage of empty stomach. In addition, we compared the annual life cycle of *P. yokohamae* in this study area with that of Mutsu Bay (Takahashi et al. 1983).

Materials and Methods

A total of 1936 specimens were used to determine the annual life cycle of adult *P. yokohamae* in the coastal waters around Kikonai Bay (Fig. 1, Table 1). Of these specimens, 36% were collected from monthly catches with commercial set nets during May 1994 through July 1995, 50% were collected during bottom-trawl survey conducted by the R/V "Ushio Maru", Faculty of Fisheries, Hokkaido University from May 1994 to October 1996 and the remaining fish (14%) were taken during bottom-trawl survey conducted by fishing boat in July and September 1995 and September 1996. In addition, the samples (185 specimens consisting of 1–6 year-old fish) collected during December 1994 and January 1995 were used to estimate the age at maturity. The bottom water temperature at each station during the survey conducted by the R/V "Ushio Maru" was measured with a CTD (Sea-Bird Electronics, Inc., SBE-19).

The specimens were measured to the nearest 1 mm in total length, weighed to the nearest 0.1 g, and ovaries, testis and livers were weighed to the nearest 1 mg. Stomachs were fixed in a 10% buffered formalin solution, thereafter the contents were weighed to the nearest 1 mg. The age of *P. yokohamae* specimens was determined by the surface reading method for otoliths.

To clarify the spawning season and seasonal change in the nutritive conditions of *P. yokohamae*,

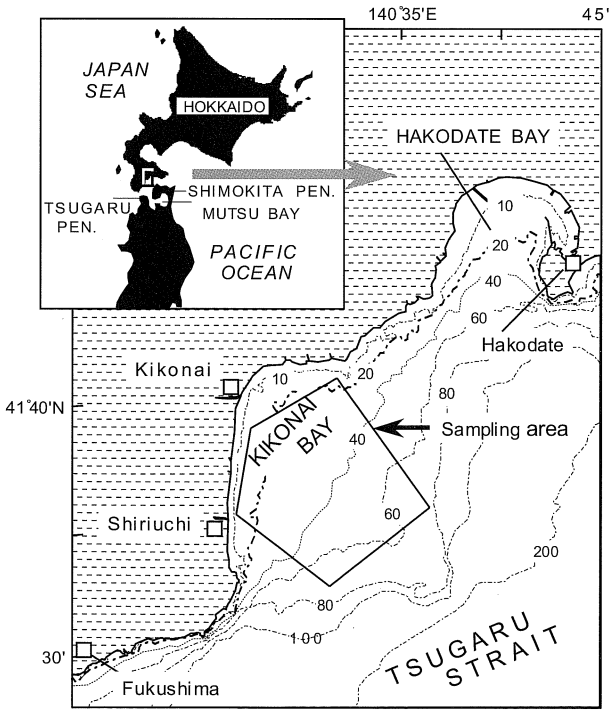

Fig. 1. Sampling area in the coastal waters around Kikonai Bay and other locations referred to in the text.

Table 1. Samples used for investigating seasonal changes in gonadosomatic index, hepatosomatic index, and condition factor of adult *Pleuronectes yokohamae* in the coastal waters around Kikonai Bay

Sampling date	Sampling gear	No. of fish	
		Male	Female
17 May 1994	Bottom set net	39	47
30 May 1994	Bottom set net	–	6
24–26 May 1994	Otter trawl net*	44	59
15 Jun. 1994	Bottom set net	7	31
30 Jun. 1994	Bottom set net	22	23
21 Jul. 1994	Bottom set net	12	25
8 Aug. 1994	Bottom set net	21	–
22–24 Aug. 1994	Otter trawl net*	51	46
21 Sep. 1994	Bottom set net	14	14
12 Oct. 1994	Bottom set net	10	17
25 Oct. 1994	Bottom set net	6	9
14 Nov. 1994	Bottom set net	19	16
28, 29 Nov. 1994	Otter trawl net*	31	22
7 Dec. 1994	Bottom set net	31	8
24, 25 Jan. 1995	Bottom set net	26	24
24 Feb. 1995	Bottom set net	25	25
1, 2 Mar. 1995	Otter trawl net*	6	19
6 Mar. 1995	Bottom set net	–	21
5 Apr. 1995	Bottom set net	32	22
26 Apr. 1995	Bottom set net	10	16
15 May 1995	Bottom set net	14	12
26 Jun. 1995	Bottom set net	25	16
17, 18 Jul. 1995	Otter trawl net**	53	32
19 Jul. 1995	Bottom set net	28	17
20 Sep. 1995	Otter trawl net**	68	20
6, 7 Nov. 1995	Otter trawl net*	75	63
13–15 Mar. 1996	Otter trawl net*	127	79
29, 30 May 1996	Otter trawl net*	37	86
26–28 Sep. 1996	Otter trawl net**	86	21
1–3 Oct. 1996	Otter trawl net*	122	99
Total		1041	895

*R/V "Ushio Maru", **Fishing boat.

the gonadosomatic index (GSI), hepatosomatic index (HSI), and condition factor (CF) were calculated as follows:

$$GSI = GW \times 10^2/BW$$
$$HSI = LW \times 10^2/BW$$
$$CF = (BW - GW) \times 10^5/TL^3$$

where GW, LW, and BW are the weight (g) of the gonads, livers and body of fish, and TL is the total length (mm). Seasonal changes in these indices were examined and outliers (above and below mean ±3SD) were excluded from the analysis.

Feeding intensity was estimated from the stomach contents index (SCI) and percentage of empty stomach, based on the samples collected by trawls during the daytime (10:45 – 14:45; Table 2).

$$SCI = SCW \times 10^2/BW$$

where SCW is the weight of the stomach contents (g). Given that the SCW of *P. yokohamae* can vary intensively during daytime (Takahashi et al. 1982), relationships between the SCI or percentage of empty stomach and sampling time (median time of towing) were examined every month.

Results

Age at maturity

Figure 2 shows the relationship between the GSI value and total length for *P. yokohamae* collected during December 1994 and January 1995 (ripened and spent fish were not included). Male fish could easily be divided into two groups with GSI ≥3.0 and GSI ≤0.2. On the other hand, female fish could be divided into two groups of GSI ≥4.0 and GSI ≤0.6, except for two individuals. Although the GSI of these two individuals was 1.5 and 2.0 respectively, the gonads were large (13.3 g, 5.8 g) as compared with those of the group with GSI ≤0.6 (0.3 – 1.8 g). For this reason, the individuals with GSI ≥3.0 for males and GSI ≥1.5 for females were treated as mature fish. The percentage of

Table 2. Samples used for investigating seasonal changes in stomach contents index and percentage of empty stomach of adult *Pleuronectes yokohamae* in the coastal waters around Kikonai Bay

Sampling date	Towing time	Bottom depth (m)	Bottom water temperature (℃)	No. of fish	
				Male	Female
1 Mar. 1995	14:14 – 14:45	52 – 63	6.4	–	9
13 Mar. 1996	11:27 – 12:00	49 – 65	7.1	28	16
14 Mar. 1996	11:04 – 11:39	31 – 33	7.0	–	16
14 Mar. 1996	13:08 – 13:39	51 – 61	6.9	–	8
15 Mar. 1996	12:12 – 12:45	66 – 75	6.9	16	9
25 May 1994	11:45 – 12:07	16 – 16	11.0	6	10
25 May 1994	13:47 – 14:29	28 – 35	9.5	9	17
26 May 1994	10:50 – 11:21	35 – 35	10.0	–	9
29 May 1996	13:32 – 14:03	26 – 29	9.8	14	40
30 May 1996	11:08 – 11:32	25 – 27	9.2	23	44
18 Jul. 1995	10:51 – 11:22	26 – 26	14.3	10	24
23 Aug. 1994	11:34 – 12:20	24 – 28	18.3	6	18
23 Aug. 1994	13:57 – 14:25	26 – 28	18.3	7	17
24 Aug. 1994	11:45 – 12:15	17 – 18	23.8	13	–
26 Sep. 1996	12:15 – 12:40	14 – 16	–	9	–
26 Sep. 1996	14:07 – 14:34	14 – 15	–	14	–
1 Oct. 1996	11:05 – 11:35	25 – 27	18.0	9	17
1 Oct. 1996	12:10 – 12:42	24 – 26	18.0	23	28
2 Oct. 1996	10:45 – 11:15	25 – 27	17.9	32	19
3 Oct. 1996	12:48 – 13:18	27 – 28	17.7	46	34
6 Nov. 1995	10:50 – 11:11	12 – 14	15.0	7	–
6 Nov. 1995	12:33 – 13:10	26 – 32	15.3	13	24
7 Nov. 1995	10:57 – 11:30	50 – 52	15.4	19	–
7 Nov. 1995	13:05 – 13:37	53 – 76	15.4	6	7
28 Nov. 1994	13:18 – 13:49	49 – 56	13.5	22	9
Total				332	375

mature fish in each age group was 94% at age 2 and 100% at age ≥3 in males, while 92% at age 3 and 100% at age ≥4 in females (Table 3). From these results, as for the April – November samples which were difficult to distinguish adult fish from immature fish based on physical observations or GSI values, the males of age ≥2 and the females of age ≥3 were used for examining the annual life cycle of adult fish.

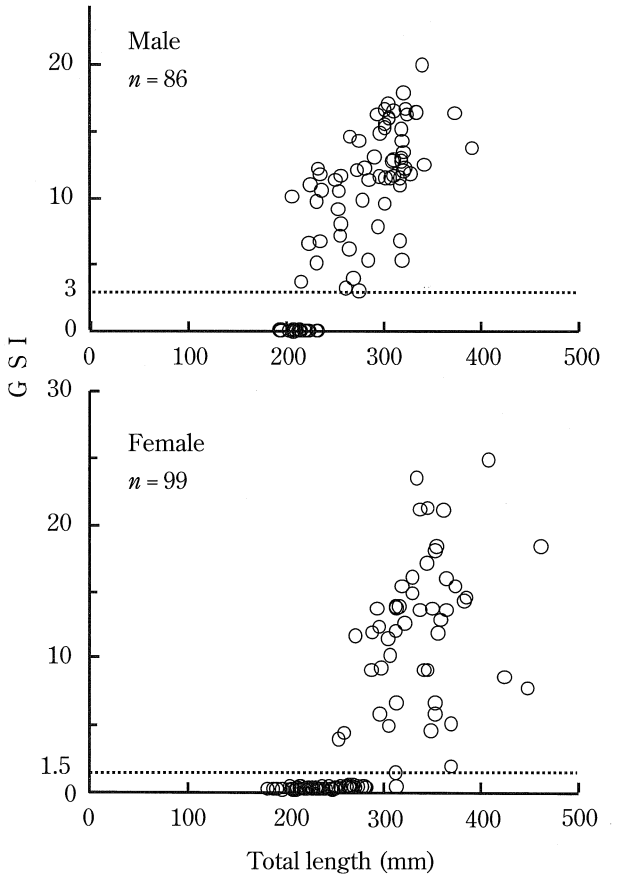

Fig. 2. Relationship between the gonadosomatic index (GSI) and total length for male and female *Pleuronectes yokohamae* collected in December 1994 and January 1995. *n*: number of fish examined.

Table 3. Percentages of male *Pleuronectes yokohamae* with gonadosomatic index (GSI) ≥3.0 and females with GSI ≥1.5 in the coastal waters around Kikonai Bay

Age	Male		Female	
	GSI≥3.0 (%)	No. of fish	GSI≥1.5 (%)	No. of fish
1+	32.1	28	0	33
2+	94.1	17	50.0	32
3+	100	34	92.0	25
4+	100	5	100	1
5+	100	1	100	4
6+	100	1	100	4

Values were calculated from samples collected in December 1994 and January 1995.

Seasonal changes in gonadosomatic index, hepatosomatic index, and condition factor

Seasonal changes in GSI, HSI, and CF of adult males and females are shown in Figs. 3 and 4, respectively. The mean GSI for both sexes was observed to increase in November and reached a maximum value between January and February in males and in February in females, and thereafter decreased remarkably. The mean GSI in April to October remained at approximately 0.5 – 1.0 in both sexes.

The mean HSI for both sexes were lower from February to April, thereafter increased to peaks in July, and gradually decreased during the period from August to March. The CF exhibited a similar seasonal pattern as HSI.

Seasonal change in feeding intensity

The relationship between sampling time and the mean SCI for adult fish are shown in Fig. 5. No correlation could be discerned in these regressions because of the limited

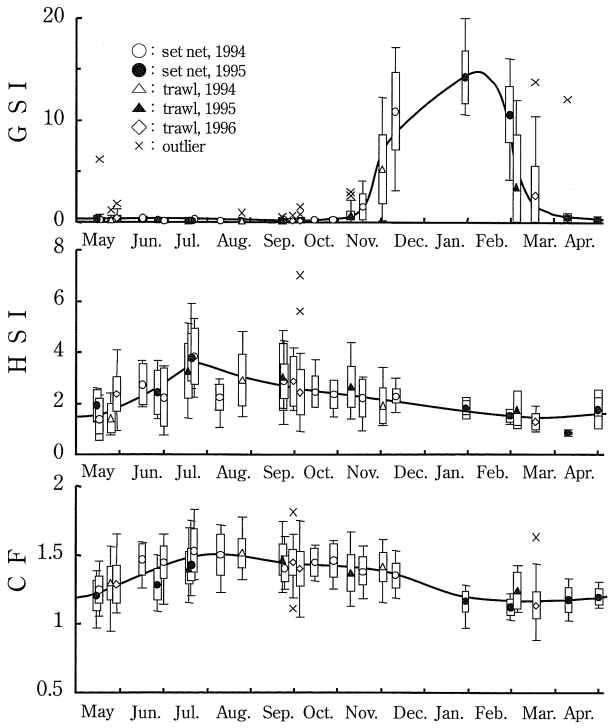

Fig. 3. Seasonal changes in gonadosomatic index (GSI), hepatosomatic index (HSI), and condition factor (CF) of male *Pleuronectes yokohamae*. Upper and lower limits of open boxes represent standard deviations (SD), and vertical lines represent the range in specimens. Mean values were calculated except for outliers (cross: above and below mean ±3SD).

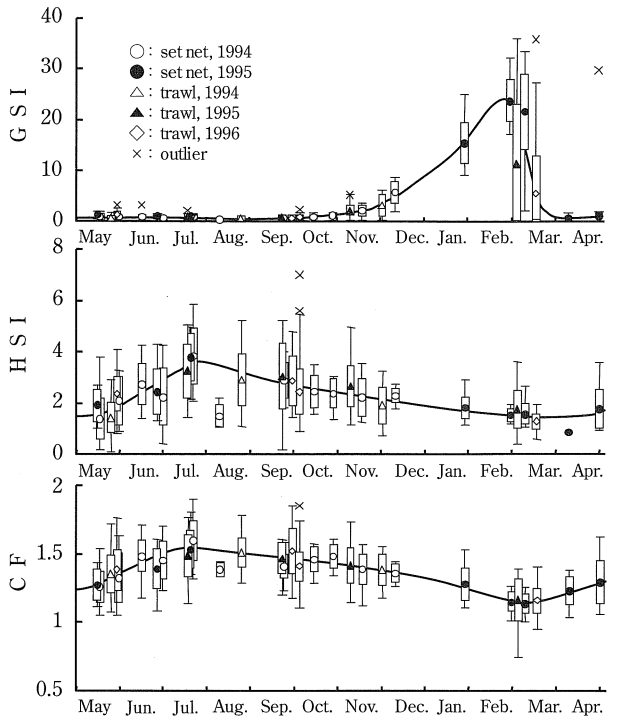

Fig. 4. Seasonal changes in gonadosomatic index (GSI), hepatosomatic index (HSI), and condition factor (CF) of female *Pleuronectes yokohamae* (partly, Ishino and Sano 1996). Upper and lower limits of open boxes represent standard deviations (SD), and vertical lines represent the range in specimens. Mean values were calculated except for outliers (cross: above and below mean ± 3SD).

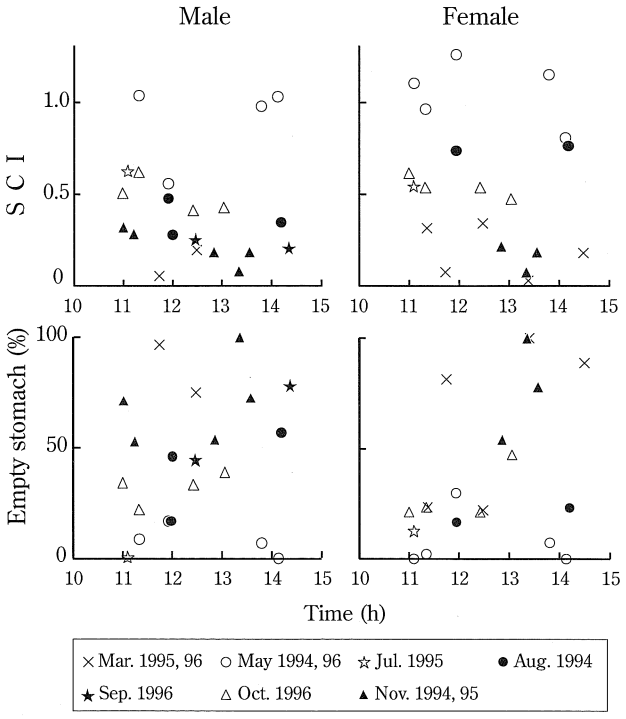

Fig. 5. Relationship between median time of towing and mean stomach contents index (SCI, upper) and percentage of empty stomach (lower) of male (left) and female (right) *Pleuronectes yokohamae*. Mean values of SCI were calculated except for outliers (above and below mean ± 3SD). Samples ≤5 individuals are omitted.

number of samples (≤5). However, the mean SCI tended to decrease from 11:00 to 15:00 on the whole, indicating that samples were not actively feeding at this time of day. In both sexes, the mean SCI was high in May (males: 0.55 − 1.04, females: 0.80 − 1.26), and low in March (males: 0.05 − 0.20, females: 0.02 − 0.35) and November (males: 0.07 − 0.32, females: 0.07 − 0.22). The percentage of empty stomachs tended to increase from 11:00 to 15:00 in March, October, and November (Fig. 5). It was low in May (males: 0 − 16.7%, females: 0 − 30.0%) and July (males: 0%, females: 12.5%), and high in November (males: 52.6 − 100%, females: 54.2 − 100%) and March (males: 75.0 − 96.4%, females: 22.2 − 100%).

Relationship between bottom depth and stomach contents index in summer and early autumn

The relationship between sea bottom depth and the mean SCI of adult males and females collected from late August to early October are shown in Fig. 6. In males, the mean SCI in the shallower areas (14 − 18 m depth) was significantly lower than that observed in the deeper areas (24 − 28 m depth; Mann-Whitney *U*-test, $P<0.05$).

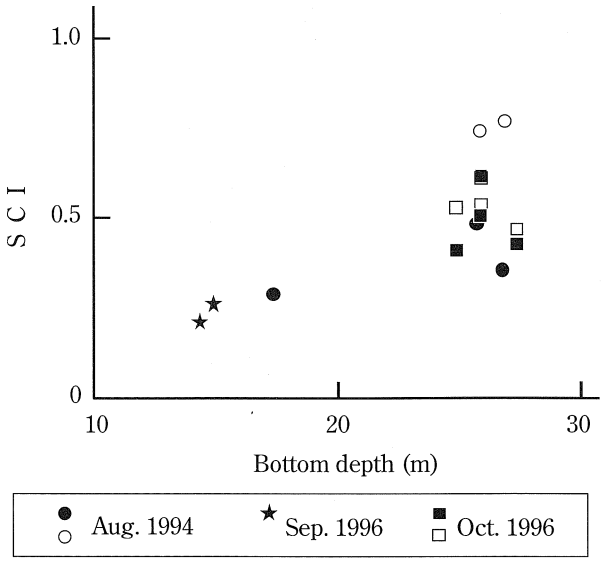

Fig. 6. Relationship between sea bottom depth and mean stomach contents index (SCI) of *Pleuronectes yokohamae* from late August to early October. Closed and open symbols represent males and females, respectively. Samples ≤5 individuals are omitted.

Discussion

The mean GSI of female *P. yokohamae* reached their highest levels in February and decreased markedly in March, although pre-spawning fish was found until late April (Fig. 4). Spent females of *P. yokohamae* can be found in the coastal waters around Kikonai Bay from February to April (Ishino 1995). It would thus seem that the spawning of *P. yokohamae* in this study area took place during February through April with a peak in March.

The HSI reached its lowest levels during the spawning season and increased during May through July in both sexes (Figs. 3, 4). A similar seasonal pattern was found in CF, although it began to increase in females in April. Consequently, by July, the nutritive conditions of *P. yokohamae* were observed to have recovered. HSI and CF for both sexes gradually observed to decrease after August. Such a seasonal change in nutritive conditions was closely related to the feeding intensity. The feeding activity seemed to be very active in May and inactive during October through March, judging from the SCI and percentage of empty stomach (Fig. 5). Otherwise, in late August and late September the mean SCI in the shallower areas (14－18 m depth) was lower than that of the deeper areas (24－28 m depth; Fig. 6). Although the main habitat of adult fish is between 10－40 m during summer and autumn (Shafieipour 1999), the bottom water temperature in late August differed largely between areas (shallow areas: 23.4－23.9℃ and deep areas: 18.3－18.4℃). According to Takahashi et al. (1987), the daily feeding rate of captive *P. yokohamae* began to decrease remarkably at about 25℃. The highest bottom water temperature in this study area was recorded between early September and early October (Takahashi et al. unpubl.). Consequently, it can be presumed that the low SCI in the shallower areas during summer and early autumn is related to high water temperatures. Bottom water temperatures in this study area do not exceed 20℃ in July (Takahashi et al. unpubl.) and the mean SCI in this period was higher (both sexes combined, 0.57).

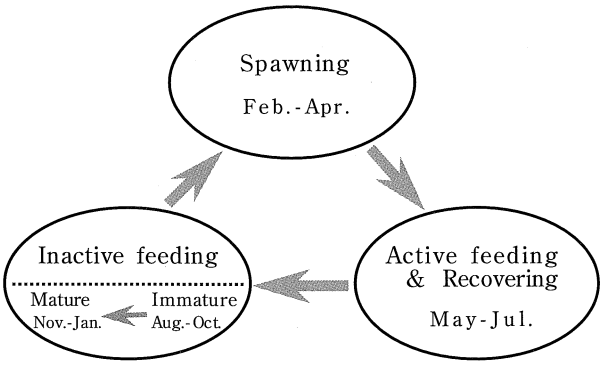

Fig. 7. Schematic diagram of annual life cycle of adult *Pleuronectes yokohamae* in the coastal waters around Kikonai Bay.

From these results, the annual life cycle of adult *P. yokohamae* in this study area was roughly divided into the following three periods (Fig. 7): spawning (February－April), active feeding and recovering (May－July), and inactive feeding (August－January). In addition, the inactive feeding period could be divided into the following two sub-periods: immature (August－October) and mature (November－January).

Pleuronectes yokohamae in Mutsu Bay near this study area spawn during December and thereafter spend wintering season (Takahashi et al. 1983). Because this population hardly feeds during the winter and resumes active feeding in May, it takes about 8 months for the fish to recover their body mass after spawning. Conversely because *P. yokohamae* in the coastal waters around Kikonai Bay commence feeding actively immediately after spawning, the fish recover their body mass within 4 months after spawning. In both areas, the minimum values of HSI and CF were found in March. In this period, the mean CF did not differ between the areas, but a large difference was found in the mean HSI (both sexes combined, 1.63 in this study area; 1.03 in Mutsu Bay). Similarly, the maximum values of HSI and CF in the annual cycle in this study area (both sexes combined, HSI: 3.64 and CF: 1.58) were higher than those in Mutsu Bay (HSI: 3.00 and CF: 1.38). These two areas are influenced by the Tsugaru Warm Current originating in the

Kuroshio throughout a year, but Mutsu Bay has a narrow bay mouth and the exchange of water is considerably less (Ohtani and Terao 1973; Ohtani and Nakamura 1985). Consequently, bottom water temperatures in the western part of the Mutsu Bay that constitute the main habitat of *P. yokohamae* in winter fall to 3–5℃ (Takahashi and Maeda 1989). In contrast, bottom water temperatures in the coastal waters around Kikonai Bay may measure 6℃ or more during winter (Shafieipour et al. 1999). It therefore seems likely that the annual life cycle of *P. yokohamae* around the Tsugaru Strait is affected largely by the topography of habitat.

Pleuronectes yokohamae in the coastal waters around Kikonai Bay feed immediately after spawning under suitable temperature conditions, and commence maturation at higher levels of nutrition, compared with the population in Mutsu Bay. Consequently, an investigation of whether there is a difference in the quality and size of eggs between these two geographic locations would be of interest and form part of future studies in the region.

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北海道木古内湾におけるマコガレイ成魚の生活年周期

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木古内湾に生息するマコガレイ成魚の生活年周期を明らかにするため，1994年５月から1996年10月までの期間，底建網と底曳網で得られた標本の生殖腺体指数（GSI），肝臓重量指数（HSI），肥満度（CF），胃内容物重量指数（SCI），空胃率の季節変化を検討した。GSIは雌雄ともに11月から増加して２月から４月にかけて著しく低下した。HSIとCFは雌雄ともに７月に最も高く，３月に最も低かった。SCIは５月に高く，11月と３月に低かった。また，空胃率は５月と７月に低く，11月と３月に高かった。以上から，本種成魚の生活年周期は産卵期（２－４月），摂餌盛期・回復期（５－７月），摂餌緩慢期（８－１月）の３期に区分され，さらに摂餌緩慢期は未成熟期（８－10月）と成熟期（11－１月）に分けられた。当海域はマコガレイのほぼ最北の漁場であるが，津軽暖流水の流入により冬季間も温暖なため産卵後活発に摂餌を行い，良好な栄養状態のもとで成熟を開始すると判断された。