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Reproductive biology of the blue marlin *Makaira nigricans* around Yonaguni Island, southwestern Japan

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***ABSTRACT*:** The spawning season and reproductive characteristics of the blue marlin *Makaira nigricans* were studied using 718 female and 384 male specimens landed on Yonaguni Island, southwestern Japan from February 2003 to February 2005. Lower jaw-fork length of females (234±24 cm, mean±SD) was greater than males (191±12 cm). The spawning season was estimated to be from May to September around Yonaguni Island by histological observation of ovaries and the presence of empty follicles. GSI of both sexes was a good indicator of spawning activity, although histologically-determined stages of males were not useful and contained a high proportion of spermatozoa all year round. Condition factors for both sexes decreased during the spawning period as a result of expending reproductive energy. Females slightly dominated, even during the spawning season, but mature females were rare. Lower jaw-fork length at first maturity was estimated to be 183 cm for females, and smaller immature fish were thought to inhabit other areas. Ripe egg diameter was about 1.2 mm, and females were thought to spawn at least 3 times per spawning season by fecundity and batch fecundity. The blue marlin was thought to leave Yonaguni Island after spawning in the autumn.

KEY WORDS: blue marlin, *Makaira mazara*, *Makaira nigricans*, northwestern Pacific, reproductive characteristics, spawning season, Yonaguni Island.

INTRODUCTION

The Indo-Pacific blue marlin *Makaira mazara* and the Atlantic blue marlin *Makaira nigricans* have thought to be different species.[1] However, these two are now considered to be same species by the recent molecular analyses.[2,3] In this paper, scientific name *M. nigricans* will be used for both Indo-Pacific and Atlantic population. In the previous works on the Indo-Pacific blue marlin, scientific name *M. mazara* was used for same species with the present study. Blue marlin is the largest istiophorid billfish and can reach a weight of over 906 kg.[1] The species is distributed in the tropical to temperate areas of the Indo-Pacific and the Atlantic Ocean and possibly migrates to all areas.

Blue marlin is an important commercial and recreational fish, and many studies have so far been reported. The reproductive biology of blue marlin has been studied in the eastern Pacific Ocean,[4] western Indian Ocean,[5,6] Hawaiian waters[7] and the Atlantic Ocean.[8,9] However, its biology in the northwestern Pacific, where blue marlin concentrate, is poorly known.[10,11] Blue marlins are thought to migrate latitudinally correlated with their spawning activity, and their spawning area and season are important information in terms of understanding their migration pattern. Maturation size and reproductive characteristics are also important information for fishery management. In this paper, seasonal abundance and reproductive biology of blue marlin in the northwestern Pacific were studied using samples landed at Yonaguni Island, southwestern Japan.

MATERIALS AND METHODS

Sampling method

Catch data and gonad samples of blue marlin were collected at the Kubura fishing port in Yonaguni Island (24˚ 27’ N, 122˚ 57’ E) from 20 February 2003 to 28 February 2005 (Fig. 1). In Yonaguni Island, there is a coastal bait-trolling fishery targeting blue marlin and they operate mostly throughout the year. Thirty-five fishing boats (1.5-5.5 t) were registered on Yonaguni Island during the study period, and all of them primarily target

blue marlin and land all their catch at the Kubura fishing port.

The number of fishing boats operated each day was recorded. Processed weights (PW: body weight without bill, caudal fin, gills and viscera) of 1,685 blue marlin (ca. 97% of all landed fish in the study period) were recorded to the nearest kg. Lower jaw-fork length (LJFL) and eye-fork length (EFL) of each blue marlin were recorded to the nearest cm for as many specimens as possible. The criterion of lower jaw-fork length was following the body length in Nakamura.[1] Lower jaw-fork length was primarily used in the present study, and additionally eye-fork length was sometimes shown in parentheses following lower jaw-fork length. Bills and caudal fins of 496 blue marlin were discarded before landing and could not be measured. In those cases, two lengths were calculated by the PW-LJFL and PW-EFL equations shown below.

$LJFL = 55.4 \times PW^{0.306}$; $n = 1{,}189$, $r^2 = 0.95$

$EFL = 46.5 \times PW^{0.315}$; $n = 1{,}189$, $r^2 = 0.95$

Determination of gonad condition

Gonads of each blue marlin were removed and sexed for as many specimens as possible. Gonads of 583 blue marlin were discarded before landing and could not be determined. Gonad weights (GW: weight of both lobules) were recorded to the nearest 10 g and parts of gonads were fixed in 10% formalin solution. Gonadosomatic indices (GSI = GW×100/PW) were calculated for each specimen. Histological staging was also conducted. Fixed gonads were dehydrated and embedded in paraffin wax, and sectioned transversely at 7-10 μm. They were stained with Mayer's haematoxylin and counterstained with eosin. The nomenclature of oocytes followed that of Yamamoto,[12] and ovarian stages were established by the most developed oocyte. Testicular stages were established by the ratio of spermatozoa area.

Determination of body condition

Body condition was estimated by the condition factor. Condition factor ($PW \times 10^6/LJFL^3$) was calculated for only measured and sexed samples. Processed weights were in proportion to almost the cube of lower jaw-fork length, and the condition factor was independent of lower jaw-fork length.

Fecundity estimation

Fecundity and batch fecundity were estimated. Oocyte diameters of one mature female (landed on 28 May 2005, 232 cm LJFL) were measured, and the mean diameter of tertiary yolk oocytes and maturation oocytes were estimated. Fecundity was defined as the number of tertiary yolk and more advanced oocytes. Batch fecundity was defined as the number of maturation oocytes. Pieces of gonad (ca. 0.1 g) were weighed, and oocytes of two stages were counted at least three times. Mean oocyte number per gram converted to whole gonad weight.

One ripe female was landed at the Kubura fishing port on 4 July 2005, and it was also observed. Lower jaw-fork length, processed weight and gonad weight were measured, and GSI was calculated. Diameters of ripe eggs from the female were also measured.

RESULTS

Monthly landing number and sex ratio

Monthly landing number, sample sizes of gonads and sex ratios are shown in Table 1. Monthly landing number of blue marlin was constantly high during the study period, except for October, November 2003, June, September 2004 and February 2005, and CPUE (landing number /boat /day) showed almost the same trend (Fig. 2). Landing number peaked from March to May 2003 and January to May 2004. Peak seasons of two years differed, although the same tendency of a decrease in autumn was observed during both years. Landing number during January and February 2003 was too high, but in the same months in 2005 they were small. Landing number in June 2004 was affected by a typhoon, which interfered with the operation of the bait-trolling boats.

Females were more abundant than males in every month, and the number of females was almost double that of males. The sex ratio differed between the 2 years, and females dominated more in 2004 (71.0%) than in 2003 (60.0%).

Body size composition

Lower jaw-fork length of blue marlin was 217±28 cm (mean±SD) and ranged from 155 to 352 cm (134 to 320 cm EFL) (Fig. 3). The majority was in the range between 170 and 280 cm, and accounted for 97.0%. Two distinct modes were observed in the size composition of blue marlins, whose smaller mode was composed mainly of males and the larger one by females. Lower jaw-fork length of females was 234±24 cm and ranged from 155 to 352 cm (134 to 320 cm EFL). Lower jaw-fork length of males was 191±12 cm and ranged from 160 to 233 cm (140 to 203 cm EFL). Females were extremely larger than males, and the sex ratio was 50% at about 206 cm LJFL from the logistic curve (Fig. 4).

Histological stage

For females, 8 histological ovarian stages were established, and the GSI of each stage is shown in Table 2. The early peri-nucleolus stage was the most undeveloped stage and contained only small oocytes (Fig. 5A). The late peri-nucleolus stage was the second undeveloped stage and contained slightly larger sized oocytes than the earlier stage (Fig. 5B). The yolk vesicle stage contained oocytes with yolk vesicles (Fig. 5C). The primary yolk stage contained oocytes that possessed a small proportion of yolk globule, and it was considered to be the beginning of maturation (Fig. 5D). The secondary yolk stage contained oocytes in which the yolk globule had increased (Fig. 5E). The tertiary yolk stage contained oocytes filled with yolk globules and were actually mature (Fig. 5F). The maturation stage contained the largest and most mature oocytes (= hydrate oocyte), and was the final stage before ovulation (Fig. 5G). The atresia stage contained a large proportion of atretic oocytes compared to any other oocytes, and generally defined the end of spawning (Fig. 5H). Empty follicles, which are evidence of spawning, were also observed from three tertiary yolk stage ovaries in July, September 2003 and July 2004 (Fig. 5I).

In Males, 4 histological testicular stages were established, and the GSI of each stage is shown in Table 2. The first stage was an inactive stage, and contained less than 25% spermatozoa (Fig. 6A). The second stage was the active (or spent) stage, and it contained more than 25% and less than 50 % spermatozoa (Fig. 6B). The third stage was a

functional mature stage, and contained more than 50% and less than 75% spermatozoa (Fig. 6C). The fourth stage was the full sperm stage, and contained more than 75% of spermatozoa (Fig. 6D). All testes contained spermatozoa, however, active to full sperm stages were considered to be mature.

Monthly changes in gonad condition

The frequency of histological stages in females changed seasonally and mature females (tertiary yolk stage<) were observed from May to September 2003 and May to August 2004 (Fig. 7). Females that had reached the beginning stage of maturation (primary yolk stage<) were observed from April to October 2003 and March to August and October 2004. The frequency of male histological stages did not change with the season and mature males (active stage<) were observed all year round at rather a high proportion. Full sperm males occurred abundantly in November, December 2003 and October to December 2004.

Mean GSI of females was high (0.73~1.21) from April to September 2003 and May to October 2004, and were low during other months (Fig. 8). Mean GSI of males was high (0.67~1.25) from May to October 2003 and May to August 2004, and were low during other months. Obvious peaks in mean GSI value of males were observed in August 2003 and July 2004.

Monthly changes in LJFL and condition factor

There were no significant changes in mean LJFL of females, and smaller males (<190 cm LJFL) slightly dominated from March to October 2003 and May to August 2004 (Fig. 9). The condition factor of females decreased from April to October in both years, and the same trend was observed in males (Fig. 10). During other months, the condition factor increased or stabilized for both sexes.

Maturation size

The primary yolk stage and more advanced stages were observed as female attained mature size, and the smallest mature size of females was 183 cm LJFL (159 cm EFL) and

50 kg PW (Fig. 11). In contrast, all male specimens had spermatozoa in the testes and the smallest male of a functional mature stage measured 160 cm LJFL (140 cm EFL) and 34 kg PW.

Oocyte size and fecundity

The frequency distribution of oocyte diameter in one maturation stage ovary was trimodal, and it divided at 400 and 800 μm (Fig. 12A). The ovary mainly contained peri-nucleolus, tertiary yolk and maturation oocytes, and they matched three modes. In short, oocytes measured 400 to 800 μm (mid mode) were 634±65μm (mean±SD) in diameter and were considered to be tertiary yolk oocytes. Oocytes greater than 800 μm (larger mode) were 996±258μm in diameter and were considered to be maturation oocytes. Fecundity and batch fecundity were defined as the number of oocytes greater than 400 μm and 800 μm oocytes, respectively. Fecundity and batch fecundity of ten females are shown in Table 3. Fecundity was ca. 1.9~14.1 million for 204~246 cm LJFL, and batch fecundity was 0.6 and 1.7 million for specimens of 225 and 232 cm LJFL, respectively. Fecundity divided by batch fecundity were 3.0 and 4.7 for two specimens.

One ripe female landed on 4 July 2005 was 238 cm LJFL with the highest GSI (8.93). The mean diameter of eggs from the ripe female was 1,187±43μm (Fig. 12B).

DISCUSSION

Spawning season

The spawning season of blue marlin was estimated from May to September around Yonaguni Island by the presence of mature females and empty follicles in ovaries. The length of the spawning season may be shorter or longer from year to year by the influence of factors such as water temperature and the course of the Kuroshio Current. The spawning season of blue marlin has been reported from May to July around the east of Luzon[10] and from April to July around Taiwan,[11] and the appearance of larval blue marlin in October near Taiwan have been thought to due to the Kuroshio Current from the south.[13] However, the present results of spawning in September around Yonaguni

Island permitted the appearance of larvae near Taiwan in October. The results obtained from around Yonaguni Island do not fully represent the spawning season of the northwestern Pacific population, and further research in other regions is needed. Quite a few mature females were observed during the spawning season, however, gravid female marlin are thought to be difficult to collect by trolling methods,[9] and mature females are rarely collected in other regions.[5,8,9] Other sampling gear such as drift gill nets would be more suitable for collecting mature female blue marlin.

Mean GSI of both sexes was high during the spawning season, and GSI calculations were thought to be an easy method for estimating spawning activity. However, the results of histological stages in males were not linked with spawning season, and mature males were observed all year round like that observed with Atlantic billfishes.[9] Males were thought to be able to join the spawning events throughout the year, however, the testicular volume during the spawning season may be important for prolonged spawning. Spermatogonia, spermatocytes and spermatids were spent and spermatozoa proportionally increased at the end of the spawning season, and the histological criteria used in the present study were deemed to not be useful to determine the reproductive activity of males.

The condition factor for both sexes decreased during the spawning period as a result of expending reproductive energy, and blue marlin became thinner during the spawning season. In males, reproductive energy consumed for the production of gametes is thought to be smaller than for females and environmental conditions (food availability) during the spawning season are not too bad (Shimose et al., unpubl. data, 2004), and males may expend energy searching for females and courting.

Reproductive characteristics

Females dominated both during the spawning season (May to September) and outside the spawning season (October to April) around Yonaguni Island. The sex ratio is reported to become nearly equal during the spawning season around Puerto Rico and the Virgin Islands,[8] but males were shown to dominate during the spawning season in Hawaiian waters.[7] The sex ratio is thought to relate with spawning behavior, and the dominance of males implied that multiple males chase and court a single female, which have been observed for blue marlin around Hawaii.[7] The present result of total sex ratio during the

spawning season around Yonaguni Island differed with the case around Hawaii. However, few mature females (assuming that mature females are not difficult to collect) and abundant mature males may be more likely around Hawaii. On the other hand, quite low mean GSI values of male blue marlin even during the spawning season (0.67~1.25) implied that the species is a pair spawner,[14] like that speculated for the Atlantic sailfish *Istiophorus albicans*.[15] The sex ratio and spawning behavior may change due to the magnitude of spawning activity and the different seasonal distribution pattern by sex. Further research is needed to estimate their spawning strategies and spawning behavior by sex.

Length at first maturation of females was thought to be 183 cm LJFL or possibly smaller in the northwestern Pacific Ocean because of a lack of smaller sized specimens. Various authors have also shown the smallest size of mature females, but not mentioned the true size at sexual maturity because of a lack of smaller sized specimens.[4,5,7-9] The smallest sizes of mature females in other regions were larger than for the present results, except for the eastern Pacific with almost the same result (155 cm EFL).[4] Though smaller mature females possibly exist, the absence of smaller sized females during the spawning season and in the spawning area implied that smaller sizes are not able to join the spawning events and are thought to be immature. In males, the smallest specimens (160 cm LJFL) were already mature and the length at first maturation was thought to be smaller than 160 cm LJFL. The lack of smaller males was caused by size selectivity of the trolling bait fishery in Yonaguni Island, and length at sexual maturity of males was unknown.

The trimodal distribution of oocyte diameter indicated that blue marlin is a multiple spawner, and spawn at least 3 to 5 times per season. Maturation oocyte diameter was about 1.0 mm, but ripe egg diameter was about 1.2 mm. This ripe egg size was smaller than egg diameters of shortbill spearfish *Tetrapturus angustirostris* (1.4 mm) and Indo-Pacific sailfish *Istiophorus platypterus* (1.3 mm).[16] However, the ripe egg size was larger than in previous reports on blue marlin (0.8 to 1.0 mm).[1,8] An oocyte with a measured diameter larger than 0.4 mm indicated completion of yolk accumulation (tertiary yolk stage), and will be an easy and accurate indicator of female maturation.

Migration pattern

The landing number of blue marlin decreased after spawning in autumn and they were thought to have left Yonaguni Island. According to the summary of Nakamura,[1] blue marlin are thought to concentrate in equatorial waters or the southern hemisphere from November to March and these migrations are thought to be linked to water temperature and food availability. In the present study, migration was also thought to be linked with reproductive activity and may include water temperature or food availability of larvae and juvenile blue marlin.

Little size difference was observed between spawning and out of spawning season in males, and implied little size advantage for spawning may exist for male blue marlin. Further extensive research about spawning magnitude and sex ratios in other regions are needed for a complete understanding of the reproductive biology and migration pattern of blue marlin in the northwestern Pacific Ocean.

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Table 1

Year	Month	Landing number	Gonad		Female (%)
			Female	Male	
2003	February	62	25	21	54.3
	March	118	55	32	63.2
	April	139	43	36	54.4
	May	150	68	47	59.1
	June	52	26	15	63.4
	July	86	38	20	65.5
	August	49	25	8	75.8
	September	71	25	24	51.0
	October	8	3	3	50.0
	November	10	5	3	62.5
	December	41	18	11	62.1
2004	January	193	69	44	61.1
	February	187	78	35	69.0
	March	121	61	22	73.5
	April	91	29	12	70.7
	May	97	33	11	75.0
	June	7	3	1	75.0
	July	62	34	4	89.5
	August	32	14	6	70.0
	September	10	9	0	100.0
	October	21	15	5	75.0
	November	17	14	2	87.5
	December	23	11	9	55.0
2005	January	33	15	12	55.6
	February	5	2	1	66.7
Total		1,685	718	384	65.2

Table 2

				GSI	
	Histological stage	*n*=	%	Mean±SD	Range
Female					
	Early peri-nucleolus	241	33.6	0.52±0.25	0.03-2.83
	Late peri-nucleolus	299	41.6	0.64±0.22	0.07-1.45
	Yolk vesicle	116	16.2	0.72±0.27	0.06-1.45
	Primary yolk	14	1.9	1.08±0.48	0.11-2.23
	Secondary yolk	15	2.1	1.56±0.33	0.91-2.13
	Tertiary yolk	23	3.2	2.37±1.17	0.97-6.38
	Maturation	3	0.4	2.92±0.71	2.11-3.47
	Atresia	7	1.0	1.00±0.30	0.54-1.40
	Total	718	100.0	0.71±0.49	0.03-6.38
Male					
	Inactive	5	1.3	0.48±0.23	0.20-0.78
	Active (or spent)	45	11.7	0.48±0.38	0.11-2.05
	Functional mature	215	56.0	0.53±0.29	0.03-1.69
	Full sperm	119	30.1	0.48±0.27	0.09-1.30
	Total	384	100.0	0.51±0.30	0.03-2.05

Table 3

Month	BL (cm)	PW (kg)	GW (g)	GSI	Fecundity	Batch fecundity
March	244	120	1,950	1.62	4,579,957	
May	204	86	2,720	3.16	6,143,707	
May	232	75	2,380	3.17	5,055,751	1,686,968
May	246	106	3,050	2.88	8,951,525	
June	225	72	1,520	2.11	2,868,109	609,837
July	219	80	1,830	2.29	3,328,952	
July	225	94	6,000	6.38	14,119,962	
July	230	100	970	0.97	1,893,068	
July	233	114	4,600	4.04	10,501,984	
August	240	108	2,820	2.61	8,070,921	

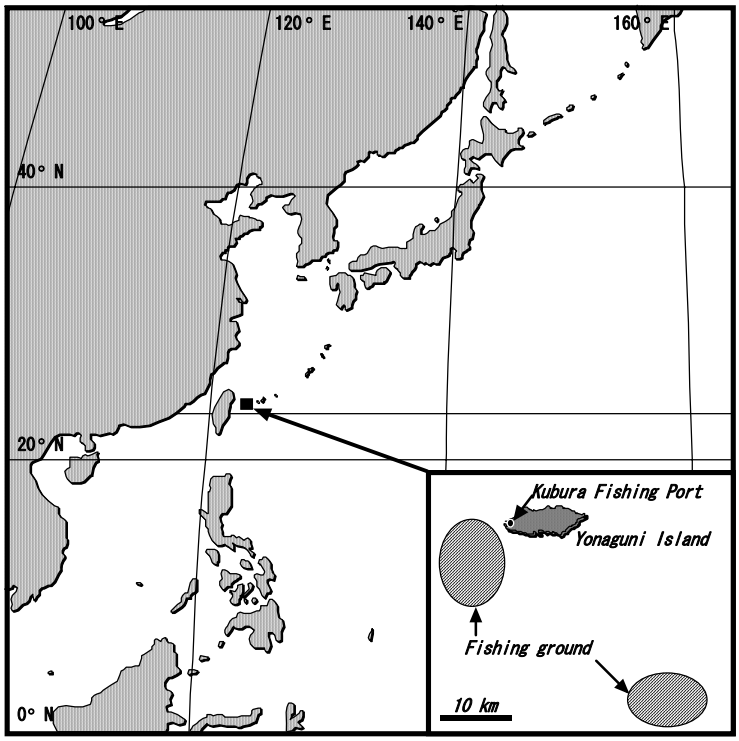

Fig. 1 Map showing Kubura Fishing Port and the fishing ground for blue marlin around Yonaguni Island.

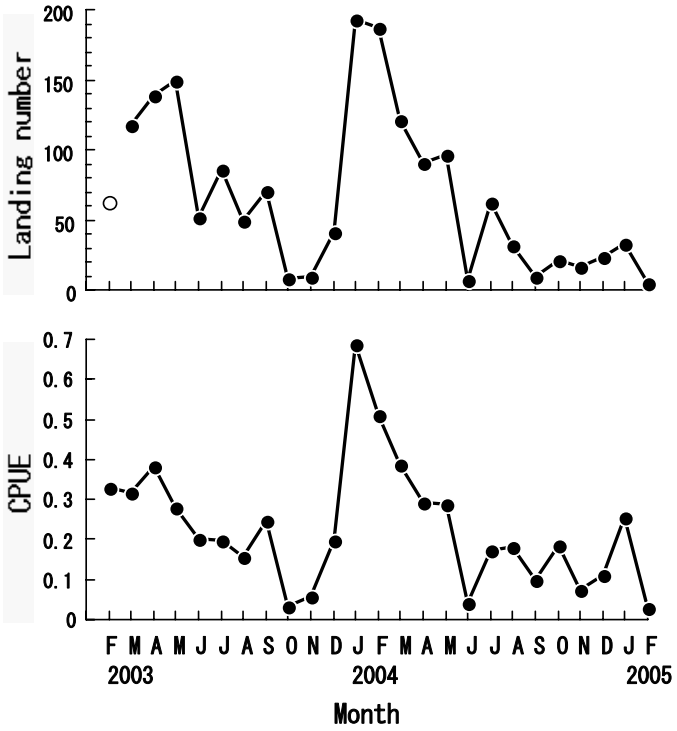

Fig. 2 Monthly changes in landing number and CPUE (catch number /boat /day) for blue marlin around Yonaguni Island. Full day sampling was not conducted in February 2003.

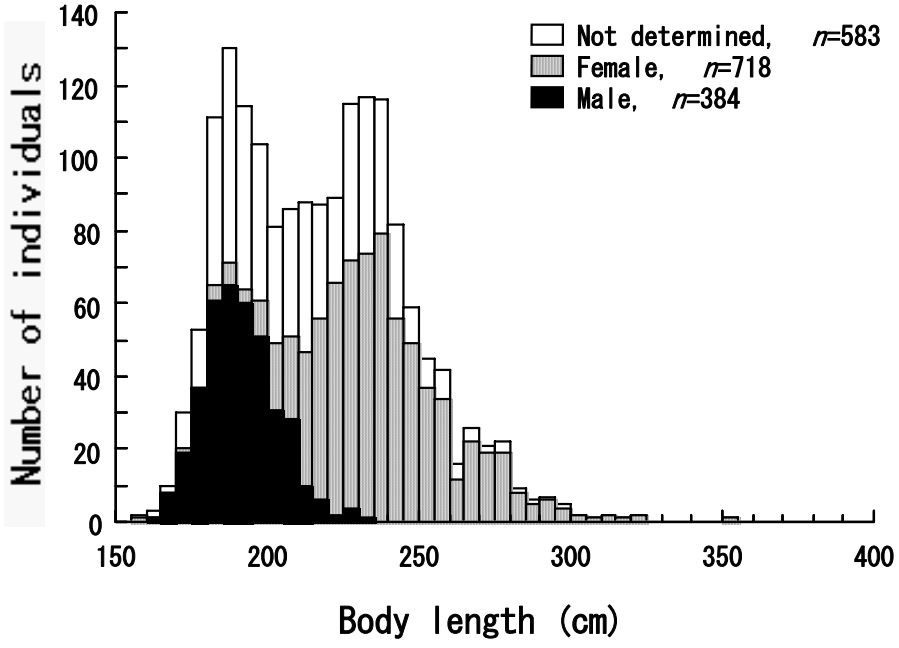

Fig. 3 Size frequency distribution of blue marlin around Yonaguni Island.

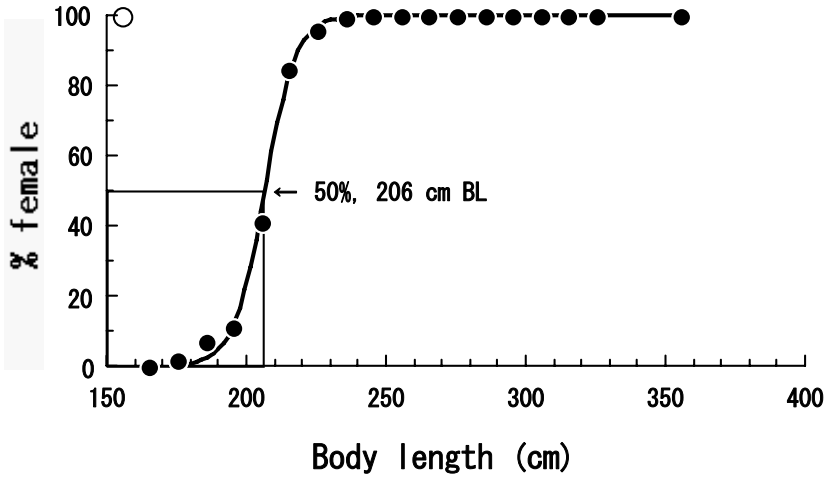

Fig. 4 Logistic curve of the frequency (%) of females to lower jaw-fork length for blue marlin around Yonaguni Island. A plot in the 150-160 cm class was excluded during fitting of the logistic curve.

$Y = 100/(1+\exp(37.3-(0.181\times X)))$

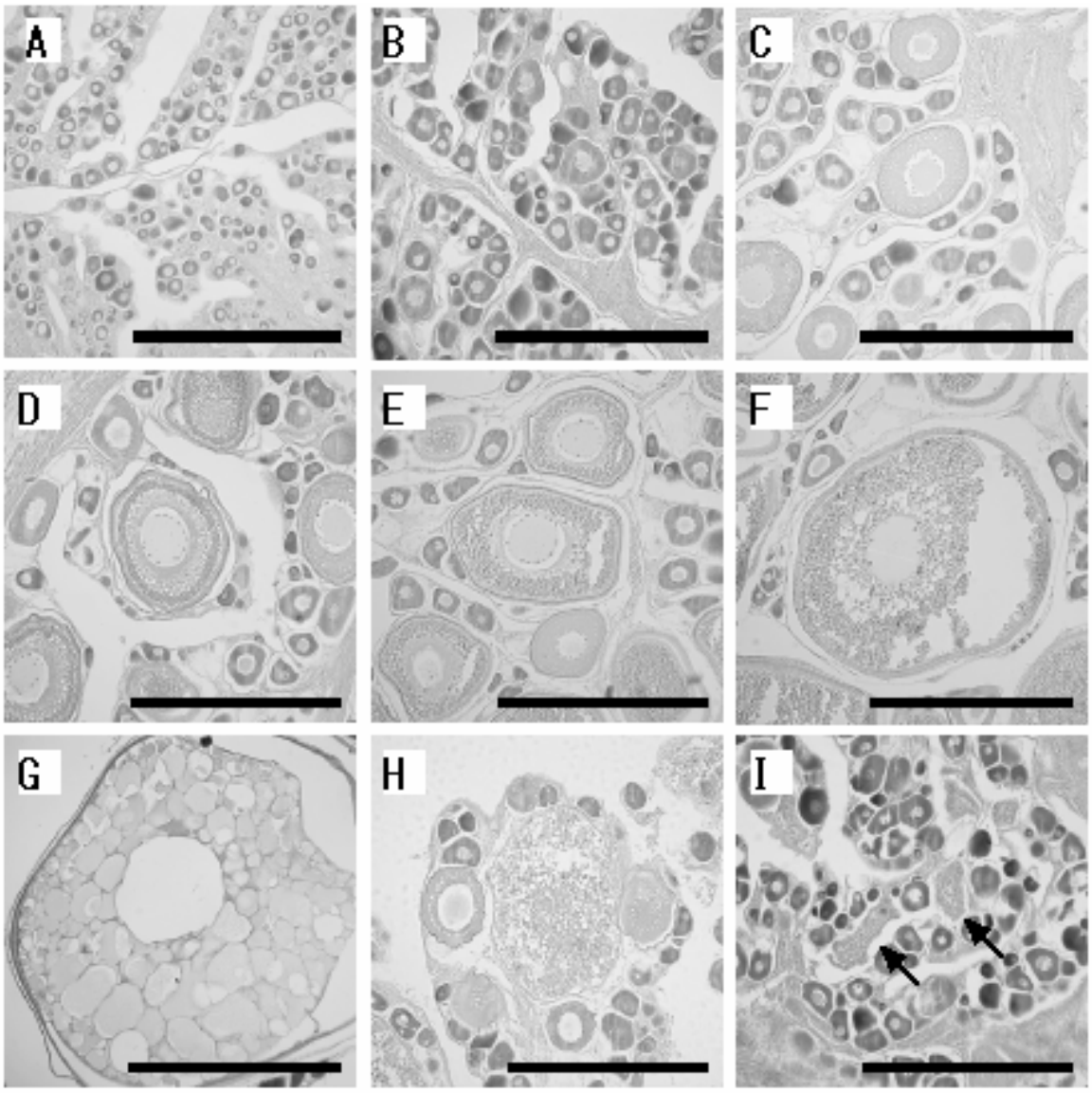

Fig. 5 Photomicrographs of 8 ovarian histological stages and empty follicles for blue marlin around Yonaguni Island. (A) Early peri-nucleolus stage; (B) late peri-nucleolus stage; (C) yolk vesicle stage; (D) primary yolk stage; (E) secondary yolk stage; (F) tertiary yolk stage; (G)

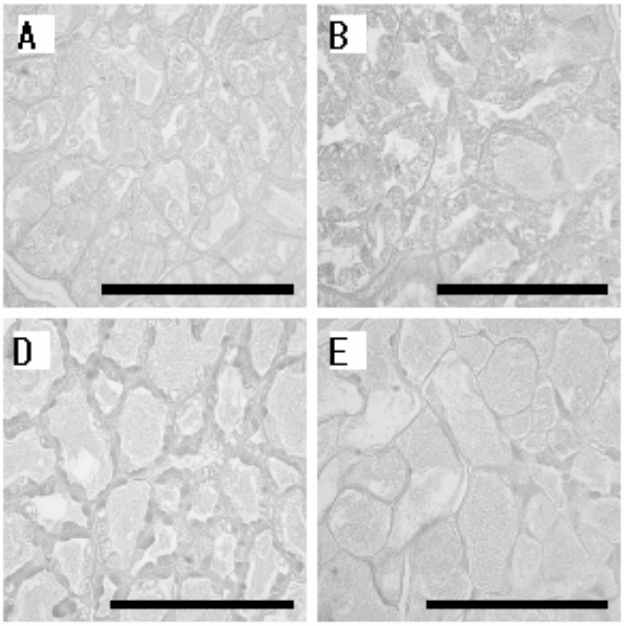

Fig. 6 Photomicrographs of 4 testicular histological stages for blue marlin around Yonaguni Island. (A) Inactive stage; (B) active (or spent) stage; (C) functional mature stage; (D) full sperm stage. Each scale bar indicates 500 μm.

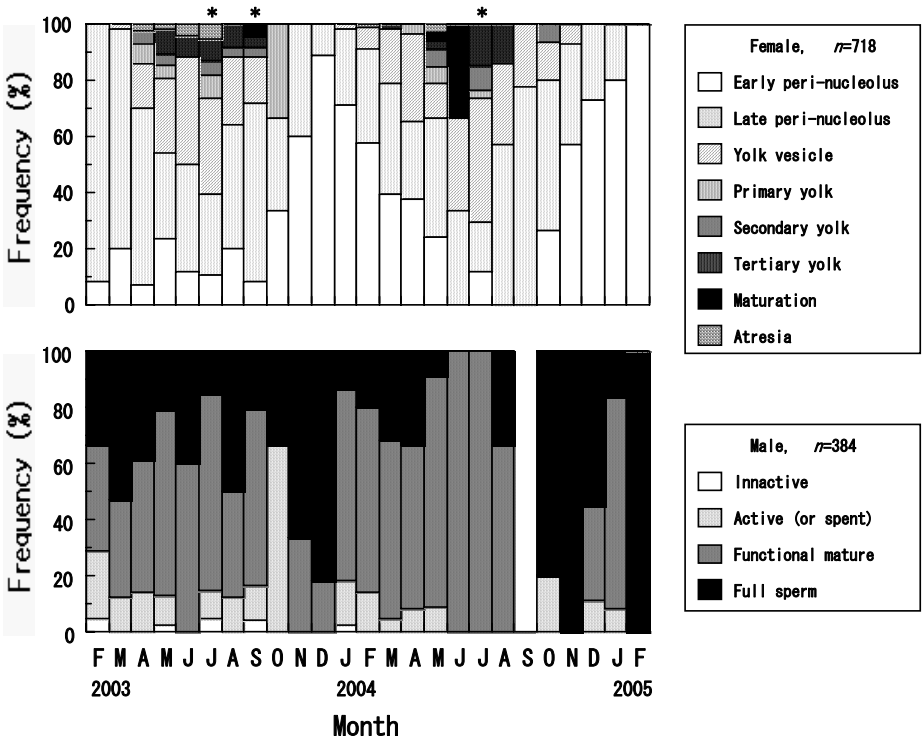

Fig. 7 Monthly changes in the frequency of histological stages for blue marlin around Yonaguni Island. Asterisks indicate the month when empty follicles were observed.

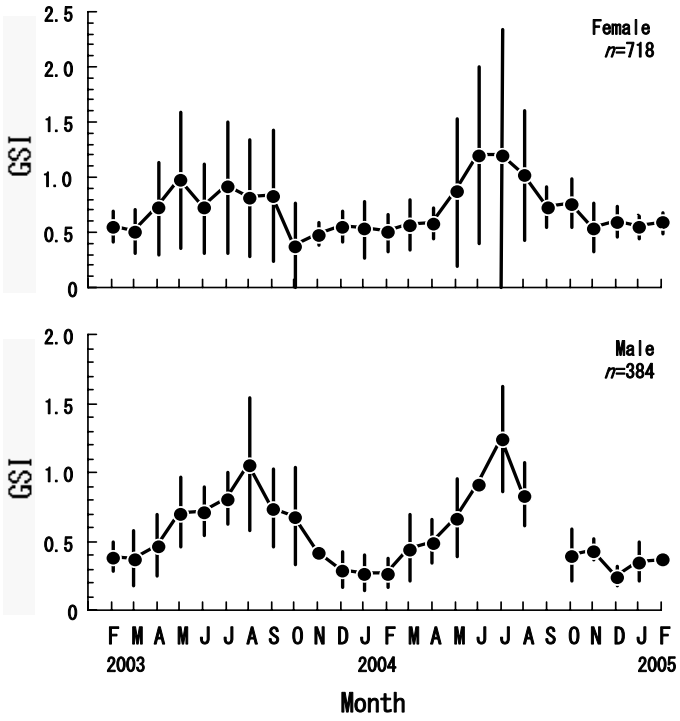

Fig. 8 Monthly changes in mean GSI for blue marlin around Yonaguni Island. Vertical bars indicate standard deviation.

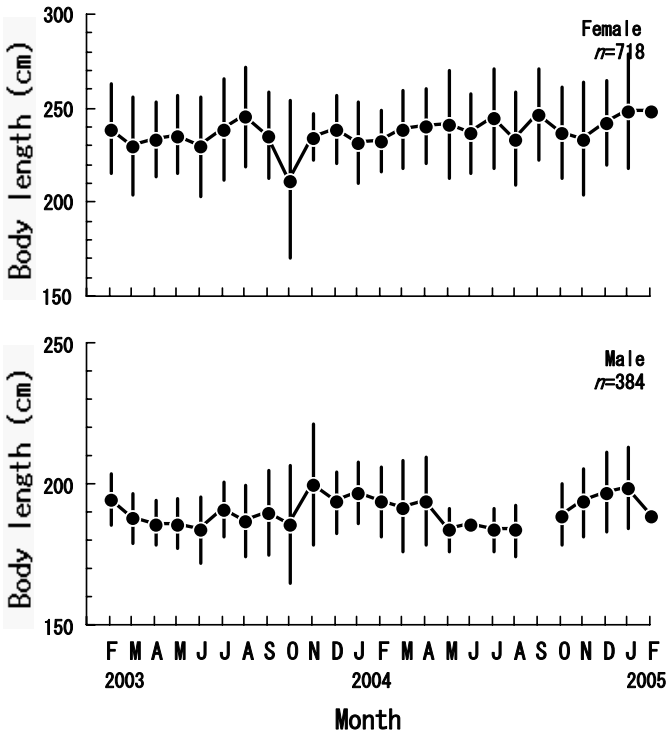

Fig. 9 Monthly changes in mean lower jaw-fork length for blue marlin around Yonaguni Island. Vertical bars indicate standard deviation.

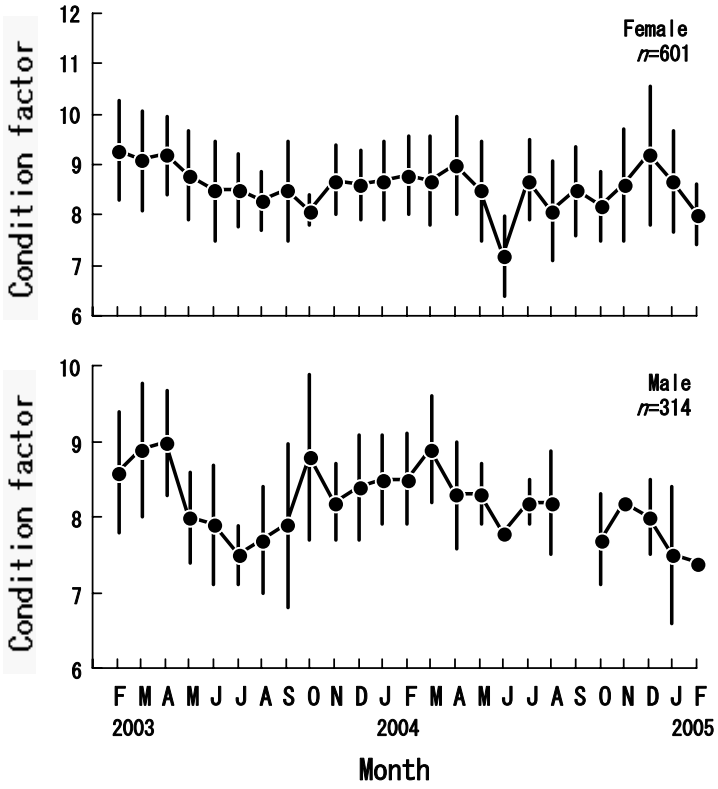

Fig. 10 Monthly changes in mean condition factor for blue marlin around Yonaguni Island. Vertical bars indicate standard deviation.

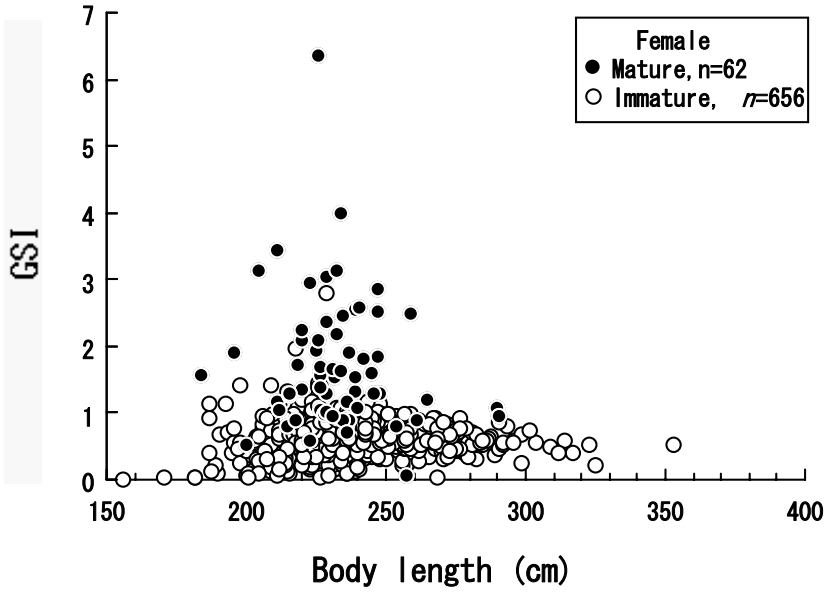

Fig. 11 The relationship between lower jaw-fork length and GSI for mature and immature female blue marlin around Yonaguni Island.

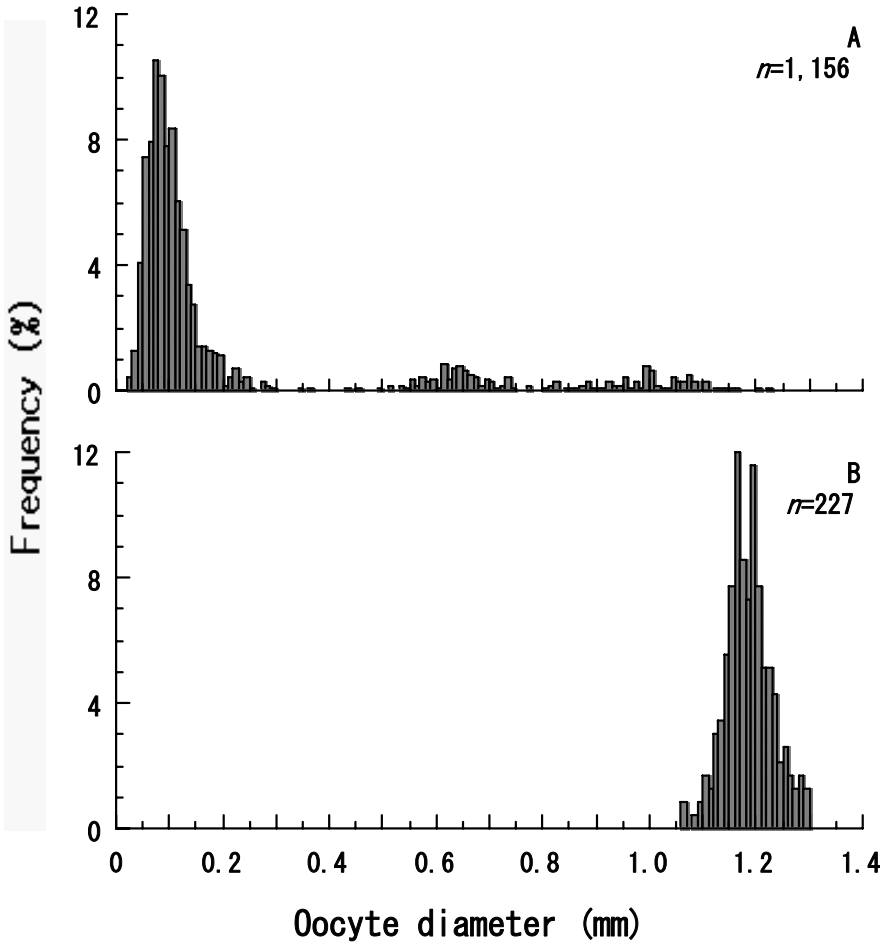

Fig. 12 Size frequency distribution of (A) oocyte diameter in mature female blue marlin (28 May 2004, 232 cm LJFL, 75 kg PW, GSI = 3.17) and (B) egg diameter in ripe female blue marlin (4 July 2005, 238 cm LJFL, 112 kg PW, GSI = 8.93) landed at Yonaguni Island.