

スズイルカとコビレゴンドウにおける血中プロゲステロン濃度 と性状態との関係

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Correlation of Serum Progesterone Levels with Reproductive Status in Female Striped Dolphins and Short-finned Pilot Whales

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Correlation between serum progesterone levels and female reproductive status was examined in 46 striped dolphins and 11 short-finned pilot whales taken by the driving fishery. Progesterone levels in immature, resting and lactating individuals were as low as approximately 1 ng/ml or below for both species. Pregnant striped dolphins maintained high progesterone levels of 30-40 ng/ml throughout the gestation period with fetus over 10 cm. The levels of pregnant short-finned pilot whales were also high ranging from 3 to 9 ng/ml, but they were lower than those of the pregnant striped dolphins. In striped dolphins, the diameter of corpus luteum of ovulation showed significant positive correlation to serum progesterone levels.

Progesterone is known as one of the important sex steroids produced in the ovaries and placenta of many mammalian species. In cetaceans, progesterone was identified in the corpora lutea (CL) of pregnant fin and blue whales by Prelog and Meister.¹⁾ Long-term hormonal studies were recently conducted in order to elucidate changes in circulating progesterone levels of female small cetaceans in captivity, and some cyclic and prolonged increases which were certainly different from basal levels were observed in bottlenosed dolphins and common dolphins.²⁻⁸⁾ Such high progesterone levels were apparently caused by the CL formation following ovulation; however, these studies did not describe the actual ovarian conditions while monitoring progesterone levels, and there was not enough evidence to establish a firm conclusion concerning the cause of high circulating levels. The present study was undertaken to elucidate the correlation of serum progesterone levels with reproductive status by collecting ovaries and blood samples from wild animals.

Materials and Methods

Blood samples were taken from 46 female striped dolphins *Stenella coeruleoalba* and 11 female short-finned pilot whales *Globicephala macro-rhynchus* taken by the driving fishery at Taiji, Wakayama Prefecture, Japan. These animals were driven on October 18 and 22, 1984, and were processed at the Taiji Fish Market on October 19,

23 and 24, 1984 after one or two day confinement within the enclosure.

After each animal was beheaded, approximately 20-30 ml of blood flowing out from the vessels running along the cervical vertebrae was taken in a disposable paper cup. Part of the blood was immediately transferred into a 10 ml-glass tube on ice, and brought to laboratory facilities. Within several hours after collection, blood was centrifuged (3,200 rpm, 5 min) and about 4-5 ml of serum were separated. Serum was stored in plastic tubes at -20°C until further hormonal analysis. Serum progesterone concentration was measured by applying simplified radioimmunoassay (RIA) omitting purification as described previously.⁹⁾

Left and right ovaries were collected where possible, and fixed with 10% formalin solution. The numbers of CL and CA (corpus albicans) were counted at laboratory. Three dimensions of all CLs were measured, and the diameter was calculated as cube root of their product.⁷⁾ Mammary glands were inspected at the field and classified into lactating and nonlactating. The presence of fetus was examined with naked eyes by slitting uteri in the field. This procedure left a possibility of overlooking an embryo of the very early stage of development, but regarding the relatively large size of embryonic membrane in these species the possibility would not be large.^{8,9)}

Female animals were classified into two categories by the presence/absence of CL in the ovaries.

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Females having no CL were then divided into the following three groups: 1) immature: possessing no CA, 2) resting: possessing CA or CAs and non-lactating, and 3) lactating: possessing CA or CAs and lactating. The females with CL were classified into the following two categories: 1) pregnant: with visible embryo or fetus, 2) ovulated: without visible embryo nor fetus. This last category would include females after recent ovulation that were not followed by conception, and after recent abortion in addition to females having embryo not yet visible.

Results

Striped Dolphins

Table 1 indicates serum progesterone levels of 46 striped dolphins according to reproductive status. For the immature group only one animal was examined, because small individuals were not processed at the market. Its serum level was so low as 0.82 ng/ml. The mean serum progesterone levels of resting and lactating groups were 1.03 ng/ml ($n=7$) and 0.79 ng/ml ($n=13$), respectively, which were nearly even with that of the immature animal. Individual hormone levels were less than 1 ng/ml in most of animals, but four females (2 resting and 2 lactating individuals) showed slightly higher levels ranging from 1.19 to 2.17 ng/ml.

Thirteen females were pregnant, including one simultaneously lactating animal. The mean progesterone level of this group (=32.9 ng/ml) was significantly higher than those of immature, resting and lactating groups (Duncan's new multiple range test, $p<0.01$). Fig. 1 indicates the scatter plot of maternal serum progesterone levels on fetal length. With the exception of a near-term female at 9.8 ng/ml, progesterone levels were maintained within a limited range of about

Table 1. Serum progesterone levels according to reproductive status in striped dolphins taken by the driving fishery

Reproductive status*1	CL-Absent			CL-Present	
	Im	R	L	P	Ov
Mean (ng/ml)	0.82	1.03	0.79	32.9	26.7
S. D.	—	0.50	0.25	7.9	24.8
N	1	7	13	13*2	12
Maximum	—	2.17	1.32	43.6	92.7
Minimum	—	0.54	0.62	9.8	1.4

*1 CL=corpus luteum, Im=immature, R=resting, L=lactating, P=pregnant, Ov=recent ovulation with no visible embryo.

*2 Includes one pregnant female simultaneously lactating.

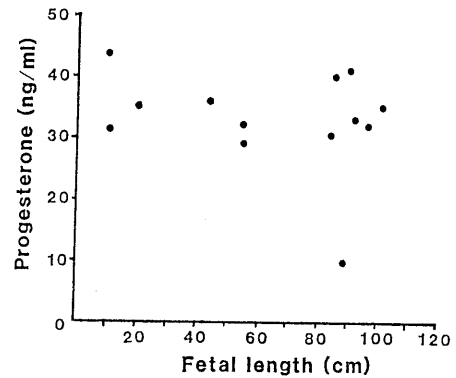


Fig. 1. Scatter plot of maternal serum progesterone levels on fetal length in the striped dolphins.

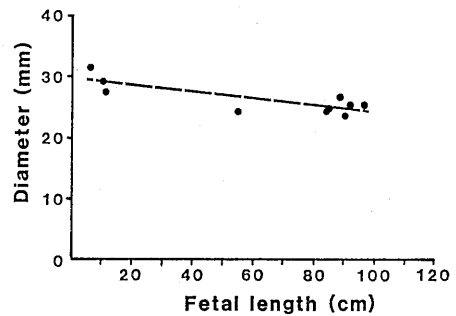


Fig. 2. Relationship between diameter of corpus luteum and fetal length in the pregnant striped dolphins. Line was obtained by the least squares.

30–40 ng/ml for the fetal growth stage of 10 to 101 cm, showing no definite inclination towards either rising or falling. Fig. 2 shows the relationship between CL diameter and fetal length of 10 pregnant females from which ovaries were collected. The size of CL appears to decrease, though only slightly, with fetal growth as reported previously.¹⁰ The regression line was represented by the least squares as follows ($p<0.01$):

$$Y = 29.686 - 0.056X, \quad r = -0.844$$

where X indicates the fetal length in cm, Y the diameter of CL of pregnancy in mm, and r the correlation coefficient. No significant correlation was observed between maternal serum progesterone levels and CL diameter in the pregnant females.

Progesterone levels of ovulated females were also higher than those of adult females without CL. Fig. 3 indicates the relation between serum progesterone levels and diameter of CL of ovulation. The following significant correlation was obtained by the least squares ($p<0.001$):

$$\ln Y = 0.155X - 0.573, \quad r = 0.871$$

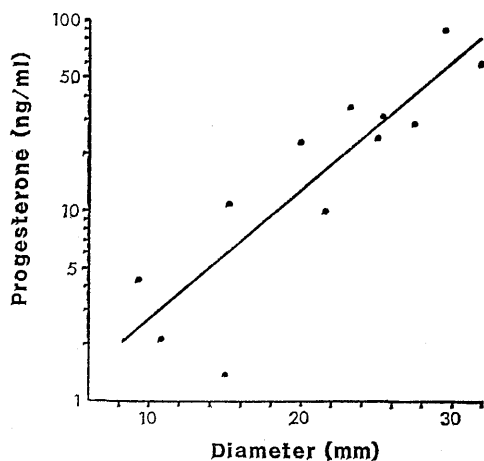


Fig. 3. Relationship between serum progesterone levels and diameter of corpus luteum of striped dolphins having no visible embryo. Line was obtained by the least squares.

Table 2. Serum progesterone levels according to reproductive status in short-finned pilot whales taken by the driving fishery

Reproductive status*	CL-Absent			CL-Present	
	Im	R	L	P	Ov
Mean (ng/ml)	0.77	0.74	0.39	5.9	20.4
S. D.	0.32	—	0.07	2.8	9.6
N	4	1	2	2	2
Maximum	1.09		0.46	8.7	30.0
Minimum	0.37		0.32	3.1	10.9

* For abbreviations see Table 1.

where X indicates the diameter of CL in mm, Y the serum progesterone concentration in ng/ml, and r the correlation coefficient. The larger the size of CLs of ovulation, the higher was the activity of progesterone synthesis.

Short-finned Pilot Whales

Table 2 shows serum progesterone levels of 11 short-finned pilot whales according to reproductive status. As in the striped dolphins, the progesterone levels of immature (4 individuals), resting (1) and lactating (2) females were lower than 1 ng/ml, while the levels of two pregnant and two ovulated females were markedly higher (over 3 ng/ml). The pregnant females had fetuses in body size of 66 and 77 cm, respectively. The mean progesterone level of pregnant females was noted to be significantly lower than that of pregnant striped dolphins (Duncan's test, $p < 0.01$). We do not have sufficient data to analyze the progesterone levels in relation to fetal size or size of CL.

Discussion

The presence of CL in the ovary was closely related to the high circulating progesterone levels in the striped dolphins and short-finned pilot whales as well as many other mammalian species studied.¹¹⁾ Temte and Spielvogel¹²⁾ reported similar results to the present ones regarding a correlation between serum progesterone levels and reproductive status of female Dall's porpoise *Phocoenoides dalli* incidentally taken by the Japanese salmon gillnet fishery. Results obtained from the three different species of *Stenella*, *Globicephala* and *Phocoenoides* suggest that CL plays an important role in progesterone synthesis during pregnancy also in cetaceans.

In captive bottlenosed dolphins, Kirby and Ridgway⁵⁾ have considered animals with serum progesterone concentrations below 1 ng/ml as neither ovulated nor pregnant. Present results support their interpretation, and indicated that concentrations below 1 ng/ml can be considered as basal circulating progesterone levels in small cetaceans. However, there were some females which had no CL in the ovaries and yet showed progesterone levels above 1 ng/ml (maximum = 2.2 ng/ml). Since no luteinized follicle was found in the ovaries of these females, this may have been caused by progesterone secretion from adrenal glands in the stressed animals.^{3,4)}

It is known that some mammalian species depend on the placenta for progesterone synthesis during pregnancy,¹¹⁾ and Cornell¹³⁾ has suggested for the captive bottlenosed dolphins that a shift will occur from CL progesterone synthesis in the early part of pregnancy to placental progesterone synthesis as pregnancy proceeds. We confirmed high concentration of progesterone in striped dolphins throughout the middle and later part of gestation and the slight decline of CL diameter during the period. However, our study could not confirm whether such a shift occurs in striped dolphins. Further study is needed on progesterone synthesis in cetacean placenta.

The fetus of one pregnant striped dolphin with progesterone levels of 9.8 ng/ml was 89 cm in length. This level of progesterone was remarkably low compared with the levels of other pregnant individuals (30–40 ng/ml). This may be due to the decrease of progesterone prior to parturition since the fetus size was near the body length range at birth of the species.^{10,14)} Temte and Spielvogel¹²⁾ reported that maternal pro-

gesterone levels in Dall's porpoise decreased significantly in cases where the length of fetus grew to exceed 100 cm, which is approximately equal to the mean length at birth.¹⁵⁾ However, it seems difficult to consider that lower progesterone levels of pregnant short-finned pilot whales show a decrease of the hormonal level prior to delivery. The fetuses of these two females were much shorter than the mean birth size of the stock of the species (=140 cm).¹⁶⁾ The progesterone levels at middle and late pregnancy in a captive short-finned pilot whale, which had been pregnant at capture and aborted 124 cm fetus after 8 months in captivity, ranged from 4.3 to 12.0 ng/ml with a mean of 7.81 ng/ml (s.d.=2.52, n=7) (Mohri, pers. commn.). On the contrary, recently ovulated short-finned pilot whales having no visible embryo exhibited higher progesterone levels than did pregnant ones (Table 2). These data suggest that the circulating progesterone levels and accompanying changes during pregnancy are different between cetacean species.

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