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Measurement of the electroencephalogram of the bottlenose dolphin under different light conditions

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Abstract

The brain wave (Electroencephalogram, EEG) of bottlenose dolphin (*Tursiops truncatus*) was measured under different photic phases. It was verified if it was possible to record EEG from the surface of the head and to find a correlation of the external visual stimuli with the electrical brain activity. Our experiments showed that EEG can be detected if its sensitivity and artefact are processed properly. The power of EEG deflected to lower frequency band under dark condition in comparison to that under light phase, suggesting that brain activity of bottlenose dolphin changed in response to the external photic condition.

Introduction

Vision of cetaceans plays an important role in their natural habitat. While there are a lot of works concerning the ecological significance and the physiological characteristics of visual sense of dolphin (Nachtigall, 1986; Dawson, 1988; Madsen and Herman, 1988; Herman, 1990; Mobley and Helweg, 1990), the relation between the outer visual stimuli and the accompanying change of inner state of dolphin remains unclear in many ways. In discussing the function of cetacean's vision, it is important to investigate not only the anatomical and morphological features of the visual system and its sensibility to photic stimuli but also the physiological effects of light condition on the central nerve system (CNS) of the animal. In the present study, the physiological change in response to the surrounding light condition was examined in order to know how the photic stimulus influences dolphin physiology. We regarded the brain wave (Electroencephalogram, EEG) as an index of the response of the CNS and examined the change of EEG under the different light conditions.

To date, the studies on brain electrical activity were performed in some marine mammals. EEG during sleeping, which fluctuated spontaneously (Mukhametov, 1984, 1987; Mukhametov *et al.*, 1985), was analyzed, and evoked potentials (Ridgway *et al.*, 1981) and event-related potentials (Wood *et al.*, 1986) in response to auditory stimuli were reported. Except for some cases (reviewed in Morgane *et al.*, 1986), however, there are few registrations concerning the brain electrical activity reacted to the external photic stimuli.

The first purpose of the present study is to verify whether it is possible to measure EEG from the surface of skin of the dolphin as is done with humans. In measuring EEG, therefore, we did not implant an electrode into the skull but placed it on the surface of skin of the dolphin's head, that is, the animal was kept healthy and in a normal state.

The second purpose was to find a correlation of the surrounding photic environment with the electrophysiological activity in the brain. Since the change of EEG reflects the consciousness or mental conditions in human (Hassett, 1978), this activity may be one of keys that elucidates the mental characteristics of dolphins.

Materials and methods

Study animals

The experiment was performed in two adult bottlenose dolphins (*Tursiops truncatus*) (body length 214 cm and weighing 220 kg for the male, body length 269 cm and weighing 214 kg for the female) that were kept under natural conditions in a pool at Kamogawa Sea World, Chiba prefecture in Japan. Prior to the experiment, the animals were trained for the procedure of experiment. On the occasion of EEG measurement, the individual was taken out of the water and placed onto a mattress. Vaseline was applied over the body of the dolphin, except the areas around both eyes and the blowhole, so that the body might not be dried up. The experiment was started when the animal became

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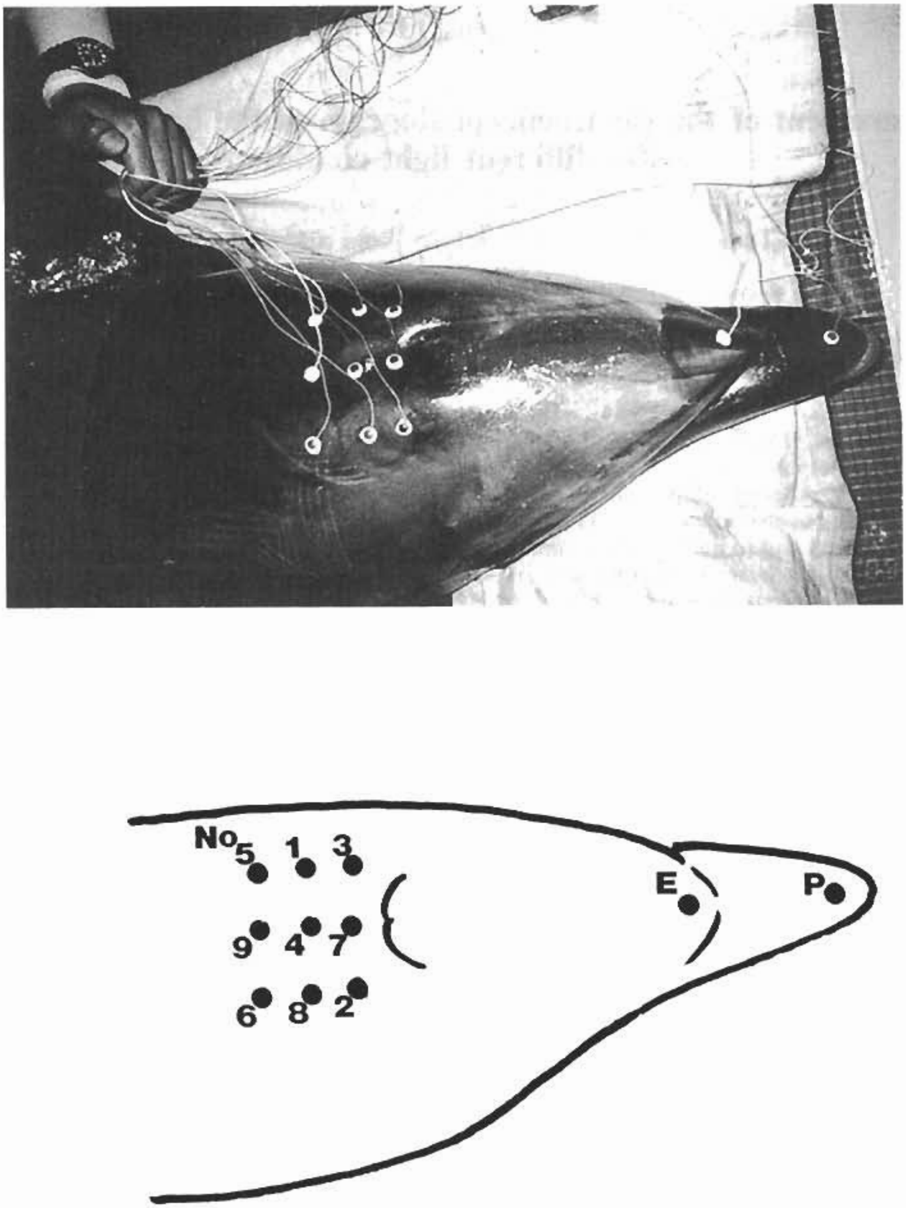


Figure 1. Measurement of EEG from the surface of the head. Each numeral means the location of electrode. E: earth electrode, P: reference electrode.

relaxed, and breathing and heart beat were checked regularly during the experiment.

Experimental procedures

Nine electrodes (Ag/AgCl, 5 mm in diameter) were located on the surface of the skin of the dolphin's head (Fig. 1). In order to reduce the electrical noise, the floor and the other things around the animal were electrically sealed. A lighting fixture was set up about 2 meters in front of the head of the individual examined, and a translucent shield was placed between the lighting fixture and the animal so that the dolphin could not look at it directly. Illuminance was changed with a variable resister which was connected with the lighting fixture.

The experiment took place in the air, and the measurement was performed under following photic conditions, that is, a light phase that illuminance (photon quantities) about the eyes of animal was 58 [μ ein] and a dark phase that illuminance was 0.1 [μ ein]. EEG was detected via EEG-7314 (NIHON KODEN Co., Japan) twice in the male individual and once in the female for 3 minutes at each phase and was recorded on a tape recorder.

Analysis of EEG

By Fourier transform, a non-periodic function such as a brain wave is replaced into sine-curves containing the various frequency and amplitude. The power spectrum, which is obtained through the Fourier transform, is useful for realizing the frequency-composition of a function (Hino, 1977), and the energy distribution of the wave can be seen by surveying the power spectrum. The EEG was then analyzed in terms of the power spectrum at first. Furthermore, we calculated the power ratio in order to know not only the frequency characteristics of the wave but also to compare the degree of concentration of powers among different waves measured under different photic conditions.

According to the sampling-theorem, we sampled the wave of the EEG at the frequency of 50 Hz, and after analogue-to-digital-conversion, Fast Fourier Transforms (FFT) (Wave Master II, CANOPUS Electronics Co.) were performed. After spectral analyses were made in *ca.* 2.7 min of EEG, the power spectrums were obtained on each brain wave (resolving power 0.012 Hz). Then, the power ratio was calculated by the following equation:

$$\mathrm{PR} = \frac{\text{Power composed at every 0.1 Hz}}{\text{Total power}} \times 100\ [\%]$$

Results and discussion

During the experiment, both dolphins showed an overall quiet and normal state. Figure 2 shows a part of EEG recorded in one individual. The amplitude of the wave, as a whole, was very small in comparison to that of a human. Perhaps electrophysiological signals generated in the dolphin's brain were enfeebled through the thick blubber of the dolphin. In the present experiment, although abnormal waves such as spike-wave, sharp-wave and spike-and-slow-wave-complex were not observed, many artefacts that originated especially from breathing and heart beat of the animal were intermixed in the EEG. We analyzed the EEG after elimination or exclusion of such artefacts according to the procedures listed below.

On the other hand, the EEG did not show a great difference among the positions of electrodes (see Fig. 2). Then, as the object of EEG analysis, we chose the wave which was detected through the No. 4 electrodes (see Figs 1, 2) that were positioned near the visual projection areas in the dolphin cortex (Morgane *et al.*, 1986).

Elimination of Electromyogram

Figure 3 includes Electromyogram (EMG) originated from the breathing or other body movement of the dolphins during the measurement. Since EMG assumes high amplitude, fast wave and overranged fluctuation and it is tailing for a while, the brain electrical activity is thought to be masked under the EMGs. We did an FFT excluding the sections that included the EMG, or we performed a lineal-interpolation on such sections where the EMG was recorded.

Mixture of ECG and the selection of analyzing frequency band

Since the Electrocardiogram (ECG) is originated from the heart beat of the animal, it generally occurs in the EEG with a specific waveform and large amplitude. In the present experiment, the ECG was recorded regularly in the EEG (Fig. 2). The ECG has a regular period (or frequency), therefore it is necessary in analyzing the EEG to select such a frequency band that ECG does not influence.

At first, we calculated the amplitude spectrum of the EEG (Fig. 4). There were three obvious peaks at the frequency of about 1.8–2.0, 3.7–4.3 and 5.5–6.0 Hz in each spectrum. On the other hand, a period of heart beat which was obtained by measuring the interval of ECG mixed in EEG (Fig. 2) was approximately 0.53 sec, and it was converted into the value in frequency as 1.89 Hz. Since this value corresponded to the frequency of the first peak in Figure 4 (also above mentioned), it is thought that the first peak in the amplitude spectrum was originated from heart beat and that its fundamental-frequency was 1.89 Hz. Then, the second and third peaks were thought to be harmonics of the fundamental-vibration because each frequency was twofold and threefold values of the fundamental-frequency, respectively. The same results were obtained also in the case of other EEG data of the present experiments. It was indicated that it was proper to analyze EEG in the lower frequency band that the ECG did not influence since the brain wave was masked under those factors in the higher frequency band. Considering the frequency bandwidth of the ECG and its harmonics, we examined the EEG in the section of 0–1.0 Hz.

From the results above mentioned, the EEG measurement from the surface of the head of

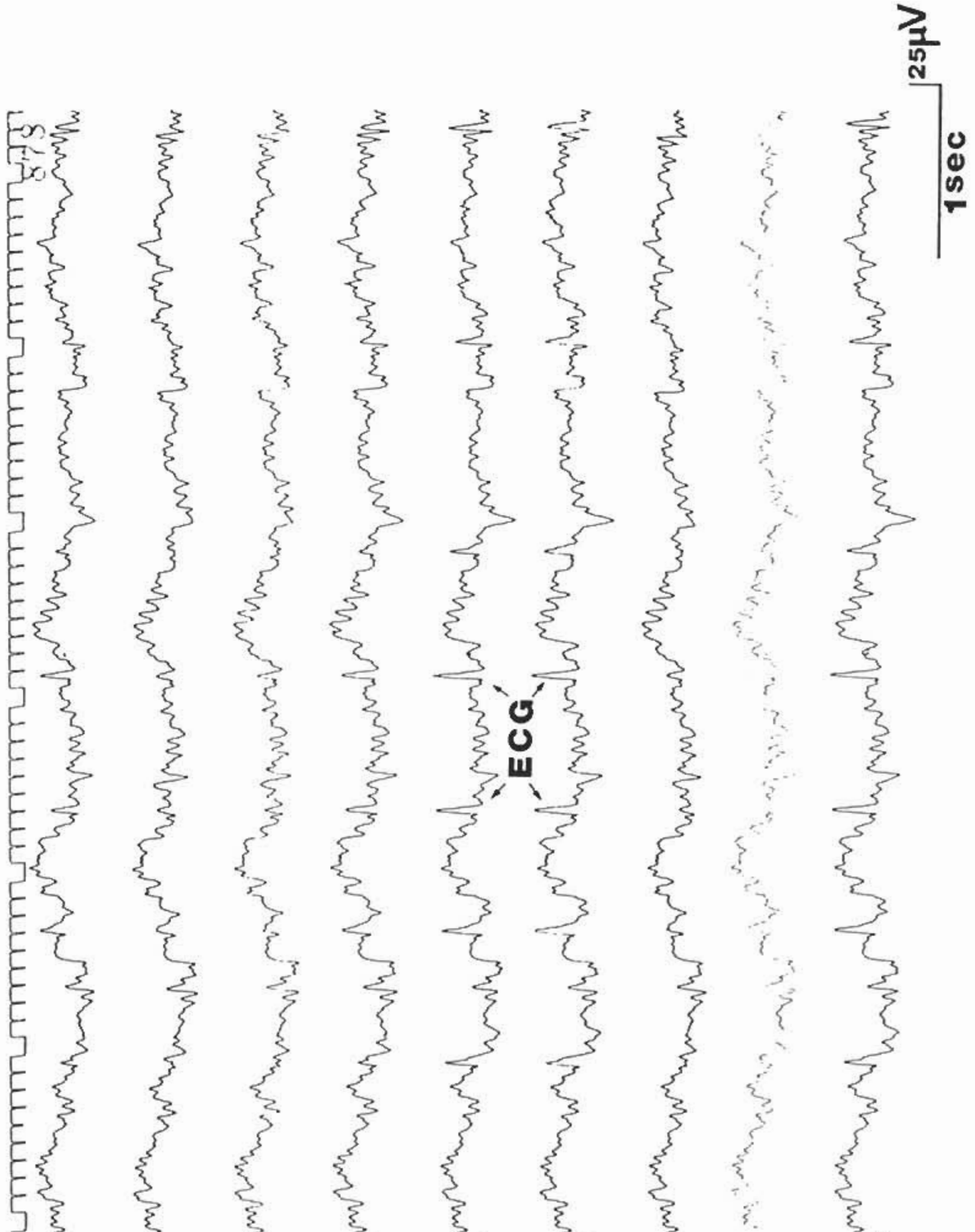


Figure 2. EEG of bottlenose dolphin. Each numeral coincides with the location number of electrode in Figure 1. ECG: Electrocardiogram.

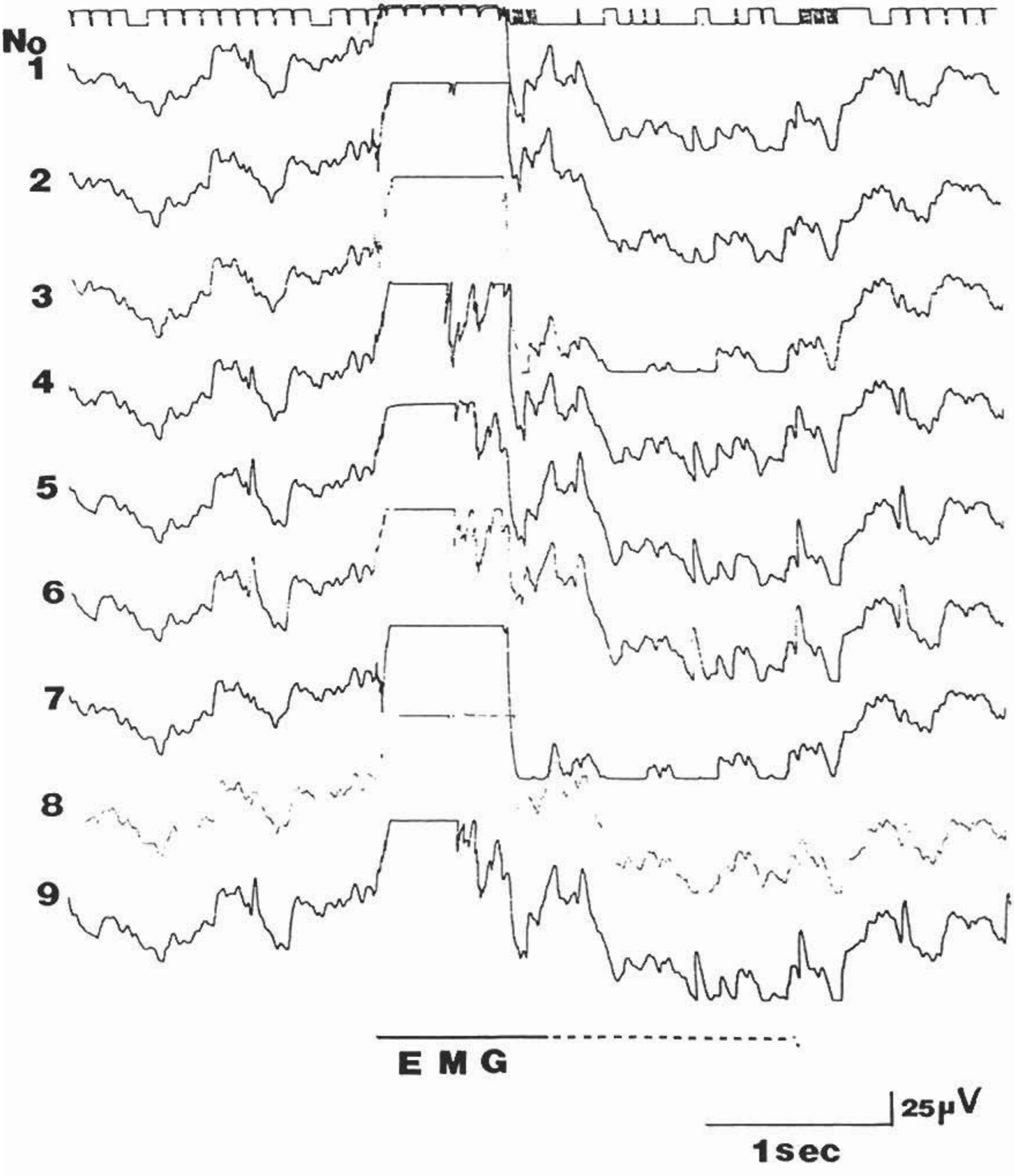


Figure 3. Electromyogram (EMG) mixed in EEG.

dolphin is thought to be possible if the recording sensitivity of EEG and the influence of EMG and ECG are processed properly. From only our experiment, however, the dominant frequency of EEG on dolphin was not clear.

EEG under different photic conditions

Figure 5 includes power spectrums of the EEG recorded in light and dark conditions. The power at every frequency, being normalized, represents the relative value.

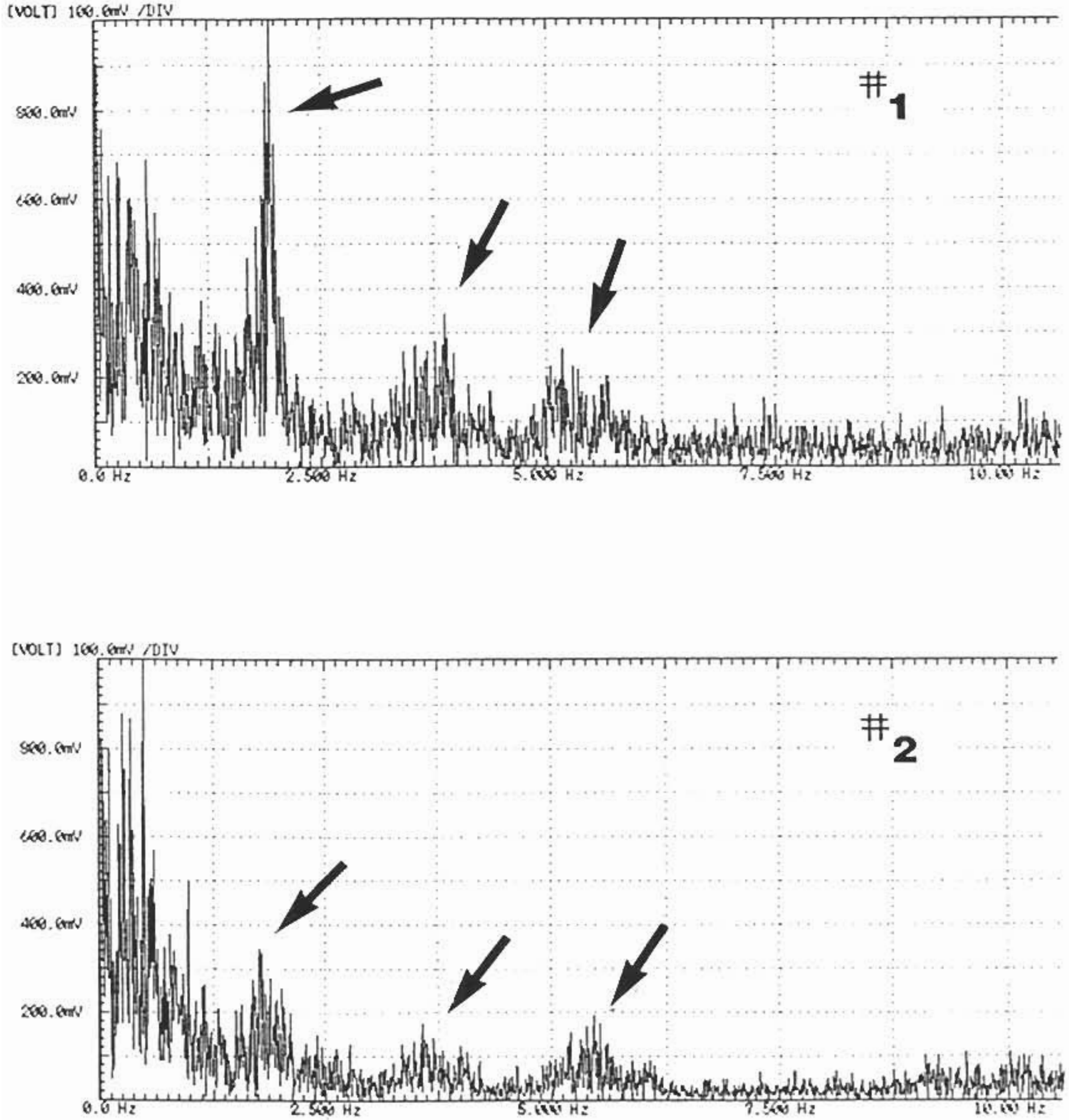


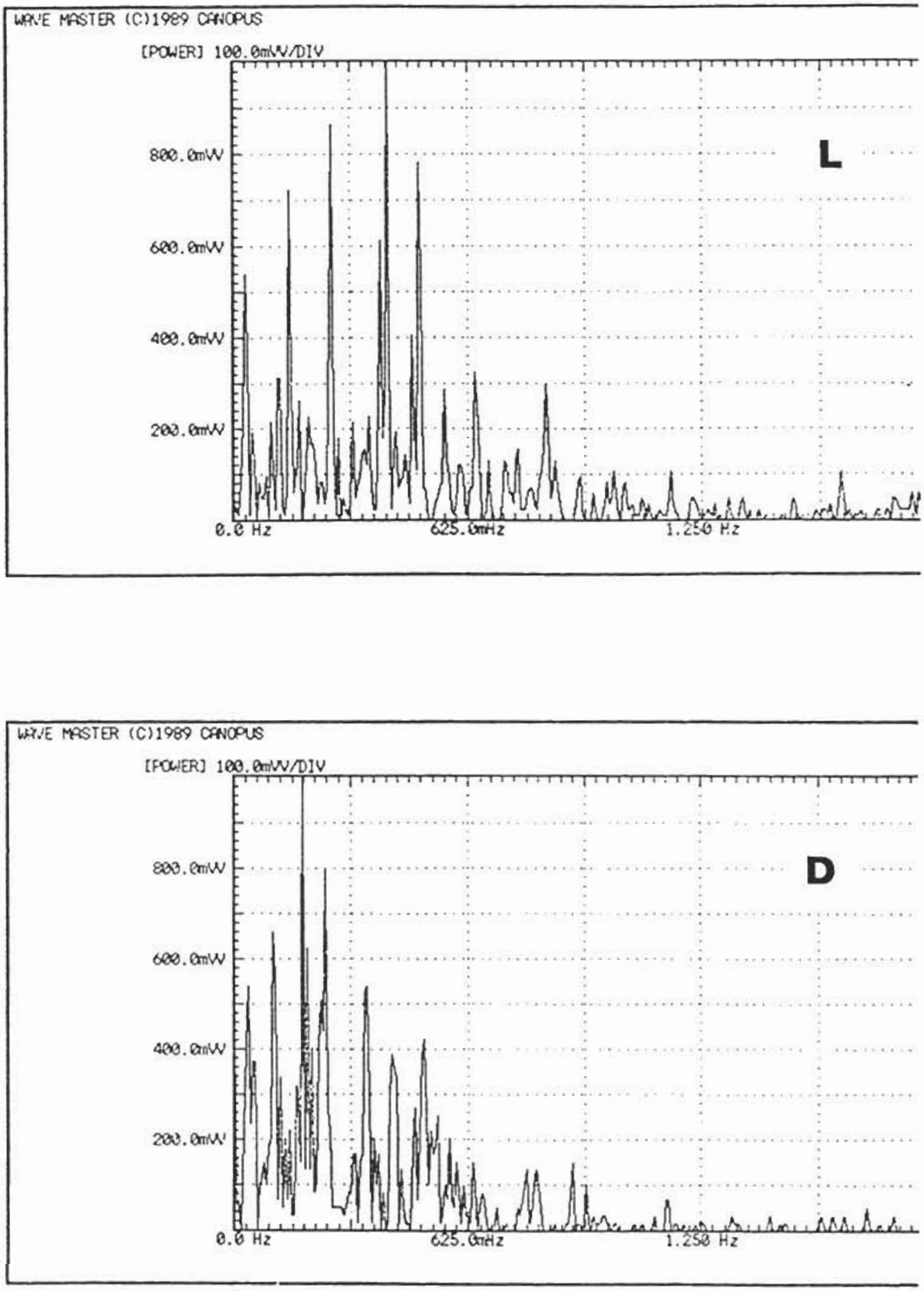
Figure 4. Amplitude spectrum of EEG of the two individuals. There are three obvious peaks (arrows) in both spectrums. Each numeral means the individual number.

As those power spectrums indicate, the EEG measured in both individuals showed different characteristics between the two photic phases. While the powers in the light condition covered a relatively wider range of frequency, those in the dark condition tended to distribute in a narrower bandwidth. For statistical analysis, average frequency and variance of the power-distribution were calculated as follows (Shimizu, *personal communication*);

$$\bar{f} = \frac{\sum_{i=1}^{N} f_n Y_n}{Y}$$

$$V = \frac{1}{Y^2} \times \sum_{i=1}^{N} \left\{ (f_n - \bar{f})^2 \cdot \left(\frac{Y_n^2}{2} \right) \right\}$$

where $\bar{f}$ is average frequency, f_n is frequency, Y_n is power at frequency f_n, Y is total power, N is


WAVE MASTER (C)1989 CANOPUS
[POWER] 100.0mVV/DIV
800.0mVV
600.0mVV
400.0mVV
200.0mVV
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625.0mHz
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WAVE MASTER (C)1989 CANOPUS
[POWER] 100.0mVV/DIV
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Figure 5A.

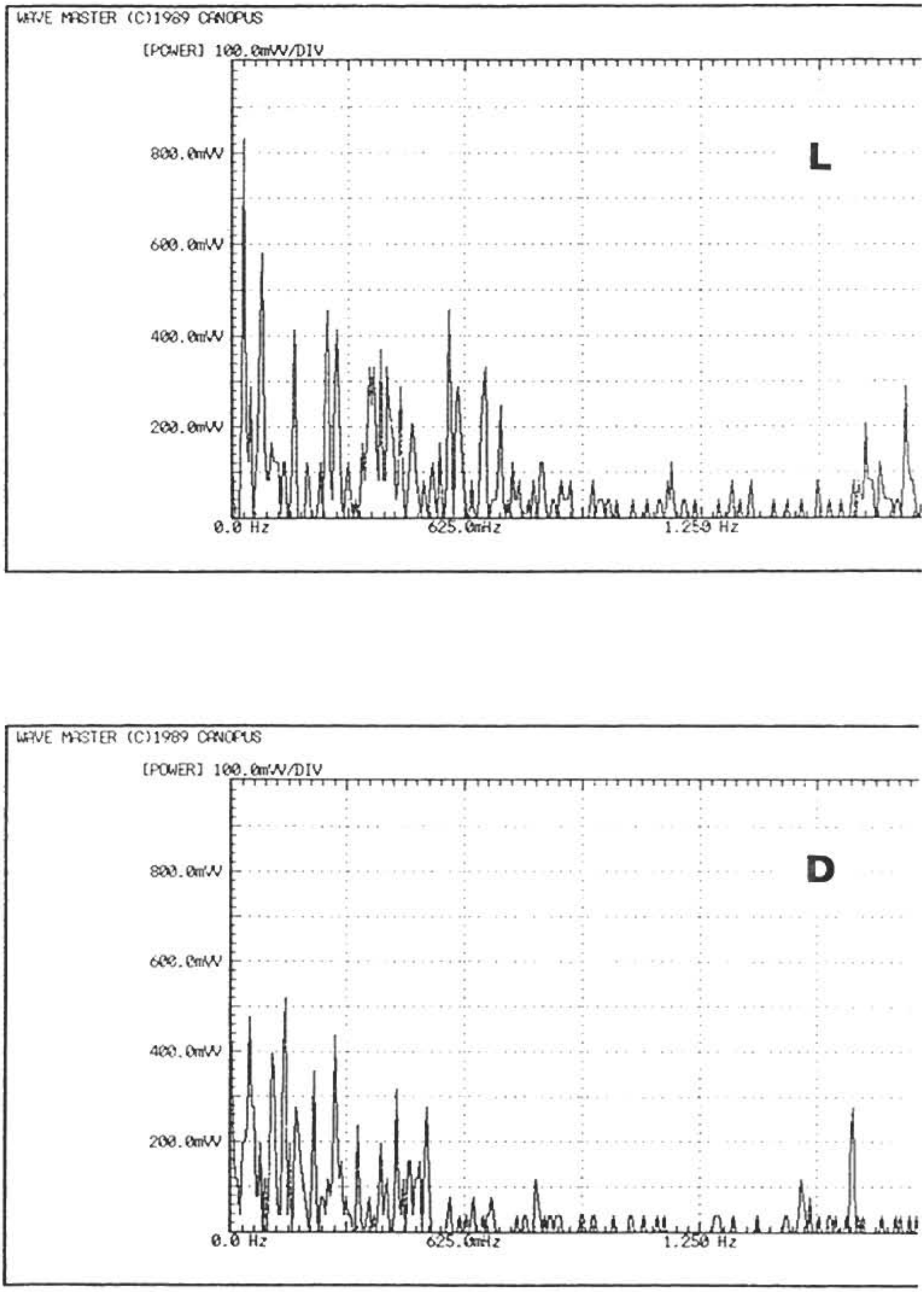
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Figure 5B.

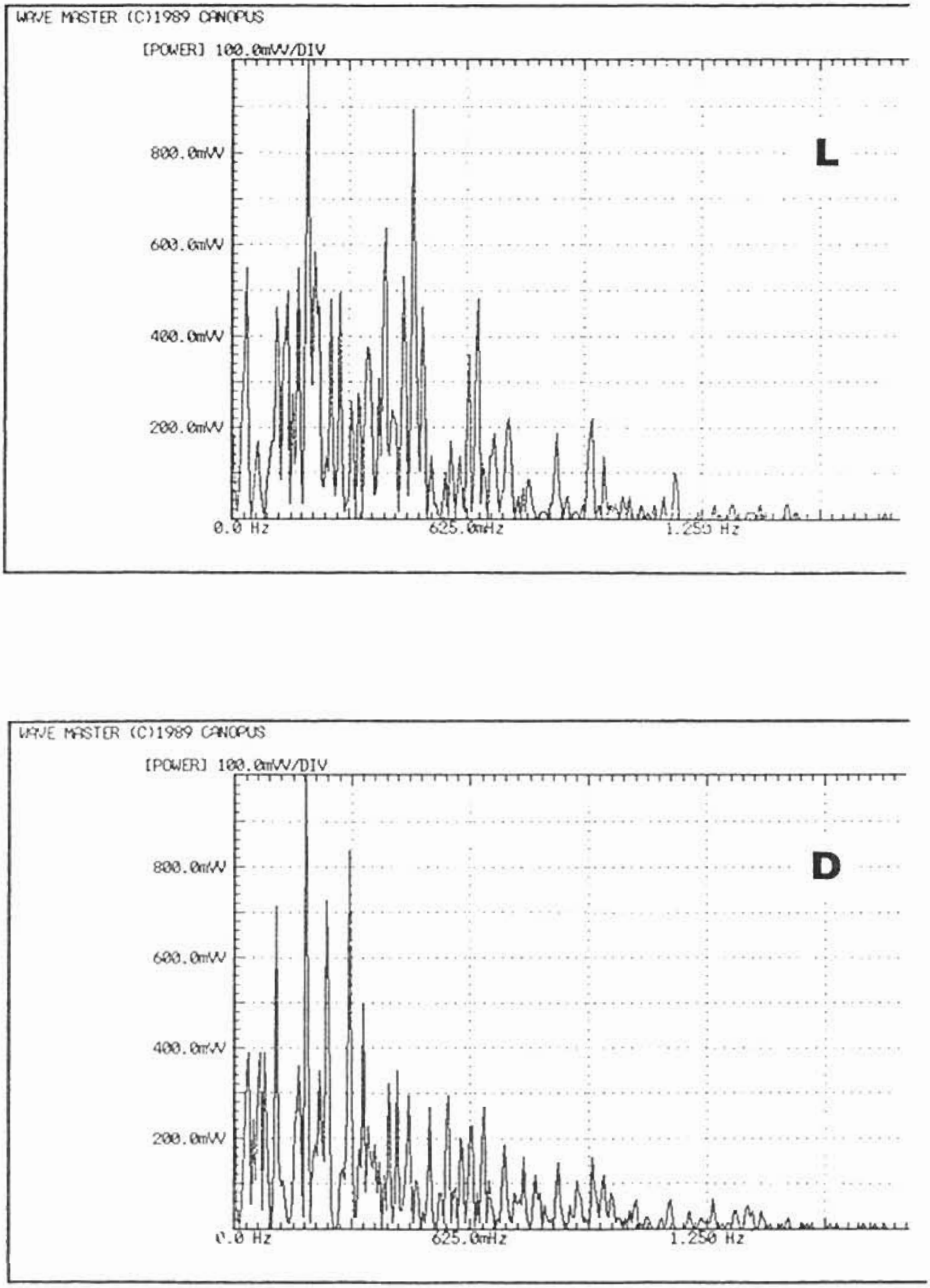

Figure 5C.

Figure 5. (**A**) Power spectrum of each EEG under two photic conditions (the first data from the female examined). L: light phase. D: dark phase. (**B**) (The second data from the female examined). (**C**) (The data from the male examined).

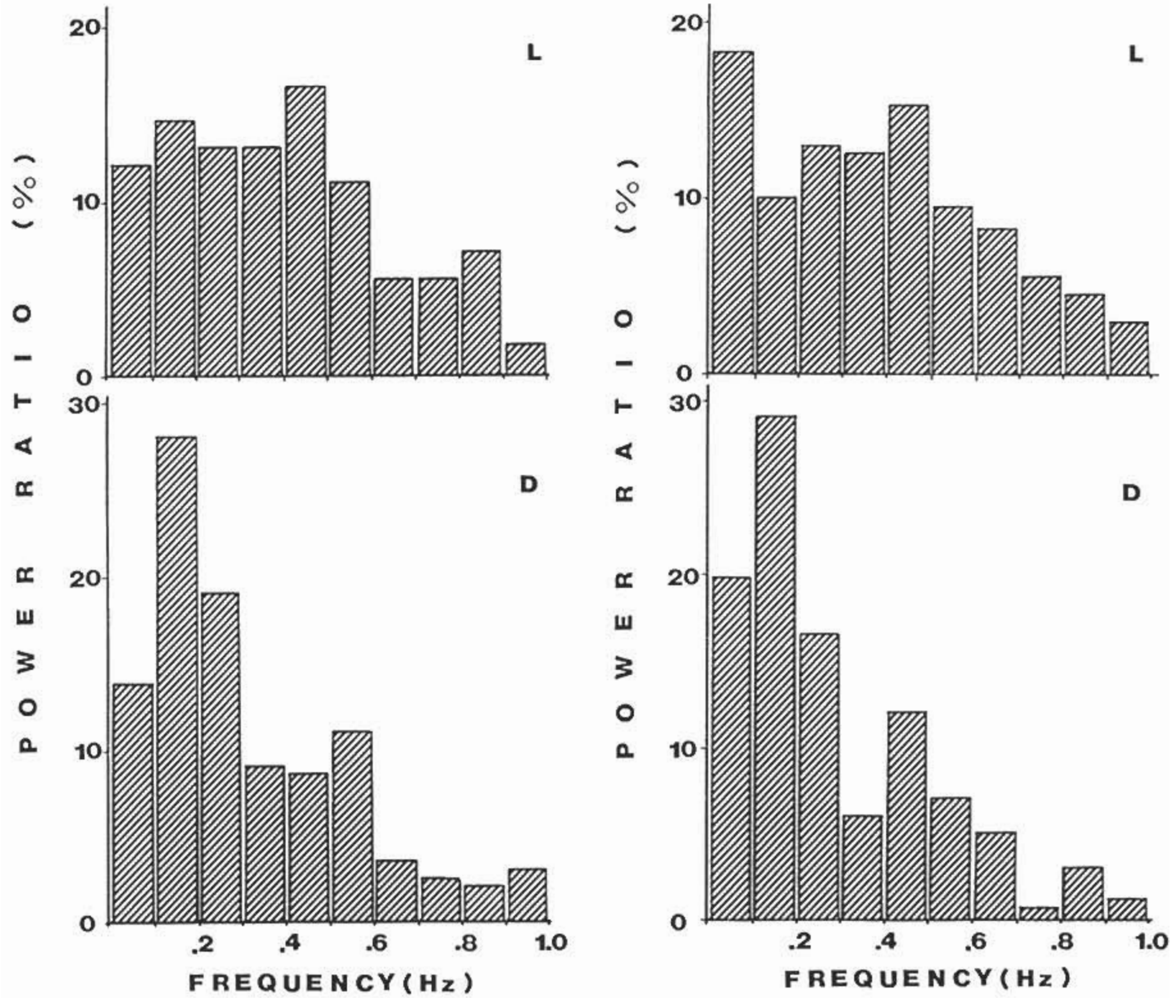

Figure 6A and B.

Figure 6A. Power ratio of each EEG (the first data from the female examined). L: light phase, D: dark phase.

Figure 6B. (The second data from the female examined)

number of samples and V is variance. Average frequency under the light phase of each data in Figure 5 was 399.2, 395.0 and 371.1 mHz and that under dark phase was 315.8, 285.8 and 362.7 mHz, respectively. Namely, when the illuminance was reduced, the average frequency became low significantly (t-test, $P<0.05$).

Figure 6 shows the power ratio of each EEG. The ratio of each data point in light phase was approximately 10% up to the range of 0.5–0.7 Hz, indicating that the power under that condition was even. In the dark phase, on the other hand, the lower the frequency was, the higher the power ratio was, suggesting that the power ratio in that condition tended to concentrate at the lower frequency band. These results show that the power of the EEG may deflect to the lower frequency area in proportion to the decline of the surrounding illuminance. When low frequency is dominant in a wave, its power spectrum shows the deflective features onto the low frequency zone. Therefore, it can be assumed from the characteristics of the power spectrum obtained in both photic conditions that the brain wave fluctuated with low frequency or slow periods during the dark condition in comparison to that in the light condition.

The present study proved that the electrical activity of dolphin's brain changed in response to the photic condition surrounding the animal. In the case of human, the dominant frequency of the EEG

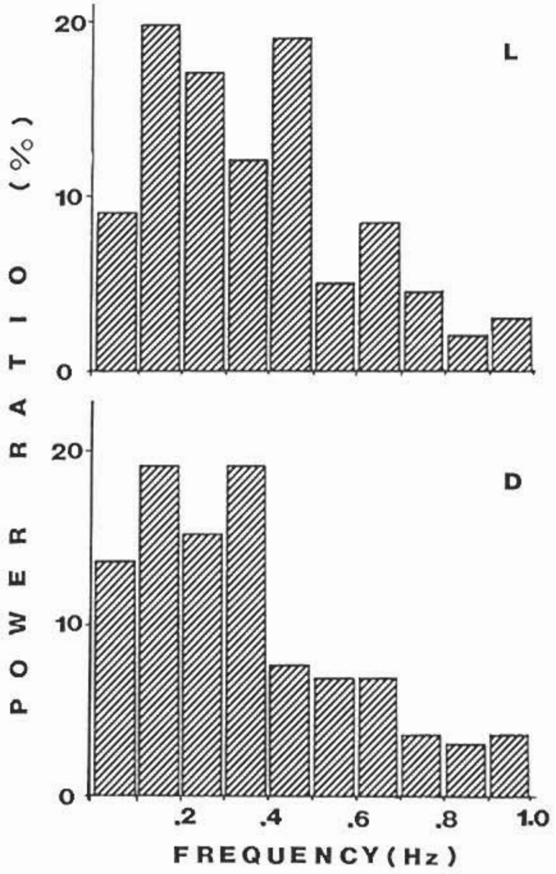

Figure 6C.

Figure 6C. (The data from the male examined)

differs at the stage of consciousness, and the feature of the EEG is relative to the mental state (reviewed by Grillon & Buchsbaum, 1987). Although the changes to the EEG pattern may describe the mental state in the dolphin, the significance of the phenomenon observed in the present study is not clear. Further research is needed on dolphins in order to know whether these EEG patterns are related to some mental factor and to discuss what category of psychology they belong to.

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