International Journal of Comparative Psychology, 2010, 23, 644-663.

Recent Studies on Captive Cetaceans in Japan: Working in Tandem with Studies on Cetaceans in the Wild

Tadamichi Morisaka
Atmosphere and Ocean Research Institute, University of Tokyo, Japan
Wildlife Research Center of Kyoto University, Japan
Japan Society for the Promotion of Science (JSPS), Japan

Shiro Kohshima
Wildlife Research Center of Kyoto University, Japan

Motoi Yoshioka
Mie University, Japan

Miwa Suzuki
Nihon University, Japan

Fumio Nakahara
Tokiwa University, Japan

Recent technological advances have allowed researchers to acquire a vast amount of information on wild cetaceans, much of which had previously been inaccessible. However, despite these new technologies, existing studies on cetaceans in captivity remain valuable. In this article, we review the recent research conducted on captive cetaceans in Japan to show their importance. We indexed the existing studies regarding behavior (resting behavior, vocal development, social behavior, and behavior differences between species), comparative cognition (echolocation ability), stress reduction, and reproductive physiology. The resulting data, as well as an understanding of the techniques used to obtain these data, will help improve the condition of cetaceans (especially endangered species) kept in captivity and fill in the gaps of studies done on cetaceans in the wild.

Recent technological advances have allowed us to access a vast amount of information about cetaceans, which was previously inaccessible. For example, bio-logging science, which uses animal-borne recorders (data-loggers) that are able to record 3D movement, acoustic behavior, physiology, etc, has revealed the superb diving behavior and physical ability of cetaceans (see Naito, 2004; Rutz & Hays, 2009 for review). Animal-borne cameras such as the Crittercam have also revealed previously unknown behaviors of cetaceans in the depths of the ocean

Tadamichi Morisaka was partially supported by a grant from the Research Fellowships of Japan Society for the Promotion of Science for Young Scientists and the program "Bio-logging Science of The University of Tokyo (UTBLS)". The authors would like to express gratitude to the many aquariums described in this article for supporting these researches. The authors are grateful to Stan Kuczaj for inviting us to contribute to this special issue of “What We Have and Can Learn from Studies of Captive Marine Mammals.” The authors thank one anonymous reviewer for helpful comments on the manuscript. Correspondence concerning this article should be addressed to Tadamichi Morisaka, Bio-logging Project, Atmosphere and Ocean Research Institute, The University of Tokyo, Room 281a, 5-1-5, Kashiwa-no-ha, Kashiwa, Chiba 277-8564, Japan (morisaka@aori.u-tokyo.ac.jp).

(Dietz et al., 2007). We are even able to non-invasively monitor steroid hormone levels in wild whales by collecting their blow (Hogg et al., 2009). Such technological developments seemingly cover many areas within the field of cetacean research.

New technologies, however, require controlled settings to verify accuracy of these new methods. In other words, the behavioral data obtained from data-loggers would be of no use without a foundation of basic behavioral description. It is also important, for example, to calibrate the hormone levels measured from the blow of a cetacean in the wild by comparing them to previously obtained blood hormone levels of similar cetaceans in captivity. Applied technologies for wild animals and traditional studies on captive animals ought not to be an either-or choice; rather, they should develop and grow in tandem.

The Baiji (Yangtze River dolphin, *Lipotes vexillifer*) was recently declared functionally extinct, making it the first cetacean species to be driven to extinction by human activity (Turvey et al., 2007). The Vaquita (Gulf of California porpoise, *Phocoena sinus*) is now the most critically endangered cetacean species in the world; approximately 150 individuals remain (Jaramillo-Legorreta et al., 2007). Thus, it is high time that the preservation and artificial reproduction of these endangered cetaceans is considered seriously, following the example of the giant panda (*Ailuropoda melanoleuca*) (Chin, 1979), of which preservation and artificial reproduction were successfully conducted. To do so, it is clear that we need to have “basic” information about cetacean physiology, psychology, and behavior. It is important to learn in advance how these elements are measured and how the appropriate captive environments for preservation and artificial reproduction can be established in order to avoid species extinction. Our work here reviews several recent studies on captive cetaceans in Japan, focusing on cetacean behavior, comparative cognition, stress reduction, and reproductive physiology, and it also discusses the significance of applying these reports to the study and conservation of wild animals.

Behavioral Studies of Captive Cetaceans

Although strong restrictions exist in captive settings, many similar behaviors remain between wild and captive cetaceans (see Dudzinski, this issue). Studying cetacean behavior in captivity allows us to make long-term (e.g., several years), fine-scale, and “natural” observations. Long-term observations of captive cetaceans, such as studies on the development of behavioral patterns, can complement the fragmented information drawn from studies of wild cetaceans. Fine-scale observations on areas such as communication signal exchange can enable more precise explanations of signals recorded in the wild. Fine-scale behavioral analysis is also necessary to interpret the behavioral data obtained by data-loggers. It is especially important to observe behaviors in captivity such as the “natural” interaction between the mother and calf cetacean, because human observations in the wild, especially underwater, are likely to cause the animals more stress and or cause a change in behavior. In the following pages, we review the existing Japanese studies on the resting behavior, acoustic development, echolocation ability, social and acoustic communication, and species differences of cetaceans.

Resting behavior

Rest and sleep are 2 of the most basic and important behaviors for animal life. Without natural rest or sleep, animals cannot remain healthy. Since cetaceans must come to the surface in order to breathe, even during sleep, their resting and sleeping behaviors are significantly different from those of terrestrial mammals. Researchers, however, have very little information regarding how cetaceans rest and sleep in the sea. There exist almost no data on the resting behavior of wild dolphins, except on the Hawaiian spinner dolphin (*Stenella longirostris*) (Norris & Dohl, 1980) and the dusky dolphin (*Lagenorhynchus obscurus*) (Würsig & Würsig, 1980), which have been reported to swim slowly and quietly during periods of inactivity in tight formations near the bottom of the sea. Although several researchers, such as McCormick (1969), Flanigan (1974), and Gnone, Benoldi, Bonsignori, and Fognani (2001) previously reported on the resting and sleeping behaviors of captive dolphins, they simply assumed that the inactive behaviors observed at nighttime were rest or sleep, and subsequently described these behaviors without performing a quantitative analysis.

Behavioral patterns during rest and sleep. Sekiguchi and Kohshima (2003) quantitatively analyzed the behavior of 16 bottlenose dolphins (*Tursiops truncatus*) at three aquariums in Japan (Shinagawa Aquarium in Tokyo, Minami-chita Beach Land in Aichi, and Kamogawa Sea World in Chiba). The dolphins' resting behavior patterns during the low-activity time period (00:00 – 03:00) could be categorized into the following three types: ‘‘surface-rest,’’ characterized by a long immobile stay with the blowhole at the water's surface (> 5 s, mean = 12.75 min, Fig. 1a); ‘‘bottom-rest,’’ characterized by a long immobile stay at the tank bottom (> 0.5 min, mean = 2.23 min, Fig. 1b); and ‘‘swim-rest,’’ characterized by slow circle-swims near the bottom along a fixed trajectory, with a low-frequency sound emission (Fig. 1c). Breath frequency was significantly lower during these behavior patterns than the daily mean and at least 1 eye tended to be closed (93.2%, Fig. 1d). Since dolphin sleep is characterized by unihemispheric slow-wave patterns in electroencephalograms, which are invariably linked to closure of the contralateral eye (Lyamin, Mukhametov, & Siegel, 2004), these 3 resting behavior patterns were considered to be sleep. Since the eye on the inner side of the swimming circle tended to be closed and the animals periodically changed their circle-swim direction during swim-rest, a relationship between this behavior and unihemispheric sleep is suggested.

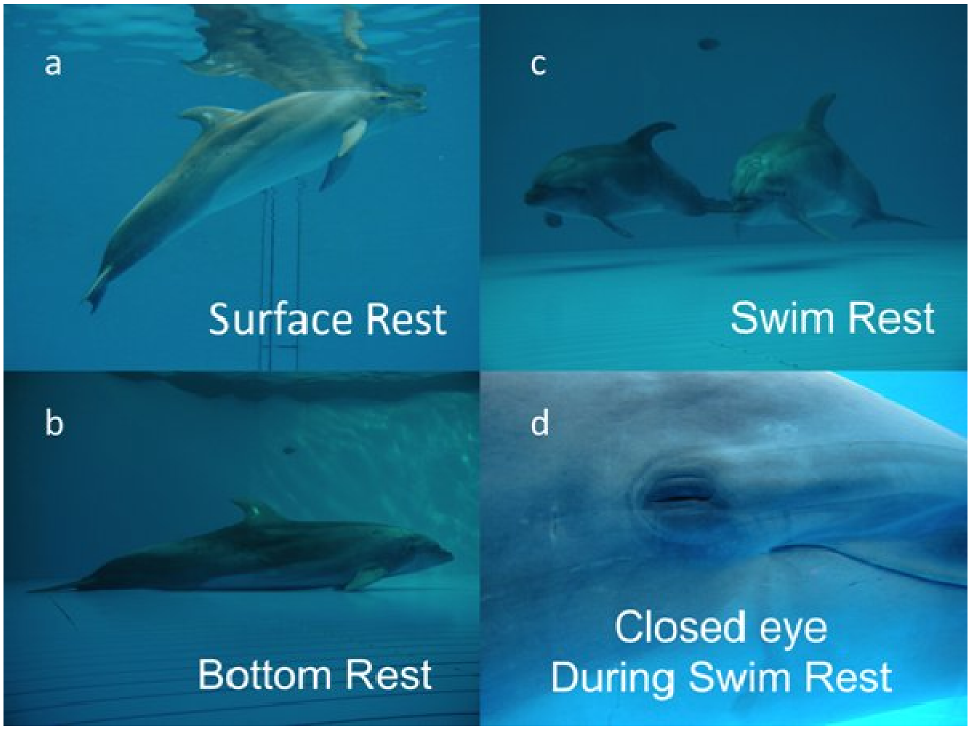

Figure 1. Three resting behavior patterns: a) surface-rest b) bottom-rest, c) swim-rest, d) during the exhibition of these behaviors, one eye tended to be closed.

Sekiguchi and Kohshima (2003) also studied the effects of introduction of novel tank partners (a mature female and an immature female) on resting behavior in a female. The bottom-rest behavior disappeared (35.5% to 0% of the observation time) and the surface-rest behavior greatly decreased (14.5% to 4.7%) in a female 1 day after the introduction of novel tank partners. The total resting time during the night also decreased from 84.3% to 43.1% of the observation time. However, the original levels of these behaviors as well as the total resting time recovered within 2 weeks after the introduction. Bottom-rest and surface-rest behaviors also disappeared (9.5% to 0% and 29.9% to 0%, respectively) in a pregnant female after childbirth. The female showed no bottom-rest or surface-rest behaviors and exhibited only swim-rest behavior even 5 weeks after childbirth. These changes in resting behavior suggest that dolphins flexibly change the type and quantity of resting behaviors according to the situation.

Do the mother and neonate dolphins sleep for the first month after birth? Sleep has been assumed to be a vital function and necessary factor in the development of mammals (Hoppenbrouwers & Sterman, 1975; Roffwarg, Muzio, & Dement, 1966) and other animals (Rechtschaffen, Gilliland, Bergmann, & Winter, 1983; Shaw, Tononi, Greenspan, & Robinson, 2002). However, Lyamin, Pryaslova, Lance, and Siegel (2005) claim that neonate and mother bottlenose dolphins and killer whales (*Orcinus orca*) show almost no sleep behavior for the first month after birth. This conclusion is based on their observations that the cetaceans continue to swim, avoid obstacles, and rarely close their eyes for 24 hours a day throughout that period.

Sekiguchi, Arai, and Kohshima (2006), however, have challenged these findings. Their analyses of the behavior and eye closures of 3 neonate-mother pairs of bottlenose dolphins at Kamogawa Sea World revealed that neonate and mother dolphins, particularly the neonates, tended to close their eyes underwater but open their eyes when they surfaced to breathe, even during swim-rest. For example, in the intensive observation of a neonate-mother pair, the first eye closure (unilateral) of the neonate was recorded 14 hours after birth, and the eye closures of its mother were recorded at 11 hours (unilateral) and 108 hours (bilateral) after birth. The mother almost always opened the eye facing the neonate (97.8%), indicating that she might be watching the neonate during swim-rest. A similar tendency was observed in other neonate-mother pairs. These results show that, although the animals tended to open both eyes when surfacing to breathe, one or both eyes were closed underwater during swim-rest, a sleeping behavior associated with continuous activity. This observation calls into question the conclusions of Lyamin and colleagues (2005), who overlooked this type of sleep by analyzing the animals' eye state only when they surfaced to breathe. It is likely that the bottlenose dolphins and killer whales described by Lyamin and colleagues also experienced periods of swim-rest sleep, a type of sleep activity that profoundly differs from the sleep behavior of terrestrial mammals. These insights could not have been found without access to captive animals, which could be closely observed continuously for the entire month both above and below the water surface.

Vocal development

Some cetaceans have been known to display vocal learning ability (Foote et al., 2006; Janik & Slater, 1997); thus, it is important to know how cetaceans develop sounds or acoustic communication throughout the course of their lives. The study of vocal development requires long-term, continuous recording within a behavioral context over several months or years, conditions that are difficult to meet in the wild.

Development and functions of neonatal bottlenose dolphin sounds. Morisaka, Shinohara, and Taki (2005a, b) recorded and quantitatively analyzed the sounds and behaviors of 2 captive bottlenose dolphin neonates of the same mother dolphin beginning from 1.5 hours after birth at Suma Aqualife Park in Hyogo. The sounds produced by dolphins are usually classified 2 broad categories: 1) pulsed sounds, including clicks and burst-pulse sounds; and 2) whistles or frequency-modulated narrow-band pure-tonal sounds (see Morisaka et al., 2005a). Neonates do not produce clicks (short-pulsed sounds that are mainly used for echolocation) during the first several days after birth, as these sounds require practice to produce (Reiss, 1988). In this context, neonatal sounds were classified as pure-tonal whistles (Fig. 2A) and pulsed sounds (Fig. 2B), which do not include clicks.

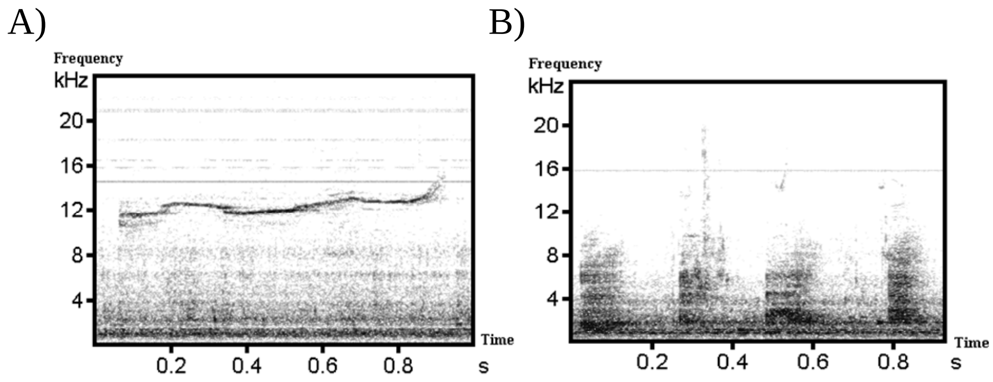

Figure 2. Spectrogram of a A) whistle and B) burst-pulses from a neonatal bottlenose dolphin.

Each neonate had already produced both whistles and pulsed sounds by the time the observation started (1.5 hr after birth). Whistle durations became longer by the hour and correlated positively with respiration intervals, which indicated that whistle duration could be limited by the development of the neonate's breath-holding ability. There were significant individual differences between neonates. One neonate (female, born in 1999) produced whistles more often than pulsed sounds, while the other (male, born in 2000) produced pulsed sounds more often than whistles. Moreover, the former neonate produced whistles with higher frequency than those of the mother, while the latter neonate produced them with lower frequency. In most mammals, olfactory cues play an important role in a mother's ability to recognize her offspring (Curley & Keverne, 2005). Dolphins, however, cannot use olfaction (Kishida, Kubota, Shirayama, & Fukami, 2007), so they are instead dependent on auditory communication and recognition between a mother and her neonate. It is possible that mother dolphins recognize their neonates through sound differences between neonates (Morisaka et al., 2005a). It is, however, also possible that this difference was actually gender related. Further research is needed to answer why sounds differ between neonates.

Morisaka and colleagues (2005b) analyzed the relationship between nursing behavior and sound production by neonatal dolphins (Fig. 3) and found that about 70% of all nursing sequences consist of the following: 1) a period before nursing in which neonates frequently produce sounds; 2) a period in which neonates do not produce any sounds; 3) a period in which neonates move below the mother; and 4) a suckling period. Whistles were produced at a higher rate than usual before suckling, which indicated that a higher rate of whistle production might function as care solicitation or as begging calls.

Figure 3. Nursing behavior of a bottlenose dolphin mother-calf pair at Suma Aqualife Park.

Vocal development of intergeneric hybrid dolphins. Published references of putative or confirmed hybridization events in the wild within the order Cetacea demonstrate that the potential for interspecific hybridization exists among a wide range of cetacean species (Zornetzer & Duffield, 2003). Since 1974, 29 hybridization events have been recorded in captivity in which a bottlenose dolphin mated with an individual of another species and produced offspring. There have been very few quantitative studies of the effect of hybridization on vocalization. If vocal traits are biparentally inherited, they would be expected to be intermediate to those of pure breeds. Alternatively, if vocal traits are learned and offspring are exposed only to a single parental species, then hybrid offspring should develop vocal traits similar to that of the parental species (Page, Goldsworthy, & Hindell, 2001). Nakahara (1998) studied the vocal development of a captive-born hybrid of a bottlenose dolphin (mother) and a Pacific white-sided dolphin (*Lagenorhynchus obliquidens*) at Shinagawa Aquarium. This individual produced pulsed sounds 2 days after birth but did not produce whistles until 3 months after birth (Fig. 4). Burst-pulsed sounds were used for communicating with the mother. To properly explain this result, further studies of vocal development in the Pacific white-sided dolphin are required.

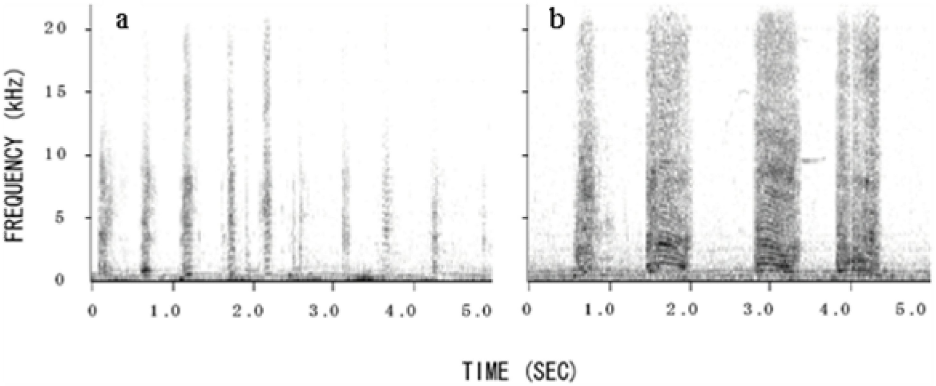

Figure 4. Spectrograms of burst-pulses from an interspecific hybrid dolphin infant a) 2 days and b) 5 days after birth.

Such fine-scale and consecutive studies of sound development and function in neonatal dolphins are difficult to conduct in the wild because neonates and their mothers may change their behavior in response to the presence of researchers who are recording sounds and behaviors (e.g., Stensland & Berggren, 2007); these signs of stress increase when researchers hold them temporally (Esch, Sayigh, Blum, & Wells, 2009; also see "Reproductive Physiology and Stress Reduction Studies on Captive Cetaceans" in this paper). Combining temporal studies performed in the wild with fine-scale captive studies will deepen our understanding of sound development and function in neonatal dolphins.

Social behavior

Cetaceans, especially odontocetes, have a complex society comparable to that of the great apes and even humans (Mann, Connor, Tyack, & Whitehead, 2000). Social behaviors play a key role in maintaining such a complex society. The study of social behavior requires continuous recording in order to identify behavioral sequences before and after the social behavior in question.

Vocal exchange and recognition in bottlenose dolphins. Some cetaceans, especially bottlenose dolphins, tend to produce individually distinctive vocalizations called signature whistles, which are thought to function in individual and conspecific recognitions and as cohesion calls that maintain consistent associations (Janik & Slater, 1998). Bottlenose dolphins use signature whistles to maintain contact with other dolphins from which they have been separated. Signature whistle exchanges may also permit dolphins to make contact with specific group members (Nakahara, 2002) (Fig. 5).

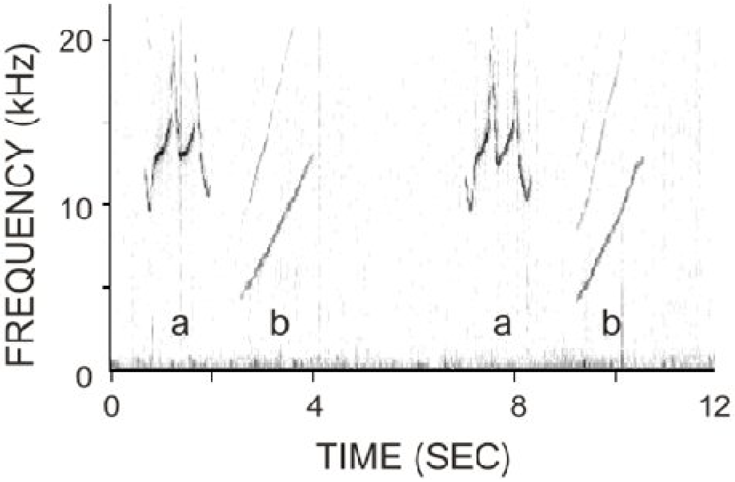

Figure 5. Signature whistle exchanges in bottlenose dolphins: sequence of 2 consecutive whistles in which the second whistle (b) was uttered by a caller different from the first (a).

Observations and playback experiments were conducted at Minami-chita Beach Land and Kamogawa Sea World to study the vocal exchange and mutual recognition of whistles in bottlenose dolphins (Nakahara & Miyazaki, 1997; Nakahara, 2002; Nakahara, 2006). The temporal patterns of the vocalizations during vocal interactions were studied by analyzing inter-call intervals between 2 consecutive whistles. Identification of vocalizing individuals was accomplished by visually associating the position of the animals in a frame of the video tapes with the sound sources as determined by arrival time differences of the same sound on 2 channels. Inter-whistle intervals, defined as the time from the end of a whistle to the beginning of the next, were measured and used in the data analysis. The results indicate that when a second call is uttered by an individual that is different from the first, the second call may occur in response to the first within 1 second, and when 2 consecutive calls have been uttered by the same individual, the dolphin remains silent for a short interval. When no response occurs, the individual is likely to give further whistles to address the group members. The results indicate that a temporal rule exists in the vocal exchange of bottlenose dolphins. The dolphins mainly use the individually distinct signature whistle during vocal exchanges.

Single-trial playbacks of the whistles of either a familiar or an unfamiliar group member were used to test for individual discrimination. Subjects responded to the stimuli whistle of the familiar animal within the temporal rule of vocal exchange. These results indicated that dolphins are able to discriminate between whistles of familiar animals and those of unfamiliar animals. A recovery-from-habituation paradigm was then used to test for individual recognition. Subjects habituated to successive presentations of different exemplars of the whistles of a familiar individual, then showed a significant rebound in response to the subsequent presentation of a whistle from a second familiar animal. These results indicate that bottlenose dolphins recognize each other on the basis of their signature whistles.

Reconciliation behavior of bottlenose dolphins. Tamaki, Morisaka, and Taki (2006) studied the temporal relationships of aggressive behaviors (including chasing, biting, hitting, head butting, and flipper rubbing, in which one dolphin rubs or touches another dolphin with its flipper) among 3 captive bottlenose dolphins (1 juvenile male (3 years old) and 2 adult females) at Suma Aqualife Park. In 2 sets of a juvenile and an adult female dolphin, flipper rubbings between former opponents who had participated in aggressive interactions occurred more frequently during the post-aggressive period (10 min after an aggressive interaction) than during the control period. Furthermore, flipper rubbing delayed successive aggressive interactions in all pairs, which implies that flipper rubbing may reduce the probability of conflict recurrence. Flipper rubbing between an opponent and a third party was also significantly more frequent in the post-aggressive period than in the control period, which implies that the flipper rubbing may have other functions, such as tension-easing by the third party or reconciliation between opponents. These results show that flipper rubbing may serve to maintain and repair social relationships. Such a fine-scale and consecutive analysis of temporal relationships among dolphins would be difficult in the wild; captive studies allow us to complement the long-term, accumulative studies of the social networks (Lusseau, 2003) and affiliative social relationships among dolphins (Connor, Smolker, & Bejder, 2006; Sakai, Hishii, Takeda, & Kohshima, 2006) that contribute to a more complete understanding of dolphin societies.

Behavioral differences among species

Numerous studies exist on the behavior of odontocetes. The behaviors of 11 odontocete species in captivity in Japanese aquariums (Commerson's dolphins, *Cephalorhynchus commersonii*; belugas, *Delphinapterus leucas*; Risso's dolphins, *Grampus griseus*; short-finned pilot whales, *Globicephala macrorhynchus*; Pacific white-sided dolphins; finless porpoises, *Neophocaena phocaenoides*; killer whales; false killer whales, *Pseudorca crassidens*; harbor porpoises, *Phocoena phocoena*; bottlenose dolphins; and Indo-Pacific bottlenose dolphins, *T. aduncus*) were surveyed (Nakahara & Takemura, 1997). Behavior questionnaires were sent to participants holding odontocetes at 16 aquariums in Japan. Respondents were asked to estimate the frequency of 57 observed behaviors in 7 behavioral categories through use of a scale of (1) to (4): (1) never observed, (2) rarely observed, (3) occasionally observed, and (4) frequently observed. To clarify similarities in the behavioral characteristics of each of these species, a cluster analysis was conducted using the mean ratings of species within each of the behavioral categories. Behavioral rating differences among species were then correlated with social structure (the group size of wild animals) and taxonomic relationships. Species with close taxonomic relationships were found to have similar ratings and were closely clustered. Socialized members of the *Delphinidae* family were rated high in terms of affiliative/social/contact behavior, aggressiveness, and curiosity/manipulation/play. The *Phocoenidae* species were rated low in most behavioral categories, except for sexual behavior, compared to the *Delphinidae* and *Monodontidae* species, which were rated high in most categories.

Cognitive Studies on Captive Cetaceans

Several cognitive studies have been conducted in captivity in Japan that would have been impossible to conduct in the wild. The reviews of Morisaka (2009) and Tomonaga, Uwano, Ogura, and Saito (2010) should be referred to for additional information on this topic.

Echolocation ability of the finless porpoise

The properties and abilities of dolphin sonar systems have been the subject of several recent investigations (Au, 2000). Most studies have been conducted on bottlenose dolphins, false killer whales, and belugas. The echolocation ability of very few other cetacean species, such as harbor porpoises, have been analyzed and described in detail. We believe that it is necessary to extend such research to the various species of odontocetes.

In these studies, the ability of a finless porpoise to discriminate the material and size of a target via echolocation was investigated at Marine World Uminonakamichi (Nakahara 2003; Nakahara, Takemura, Koido, & Hiruda, 1997). The porpoise was required to choose a standard target of a 15-mm diameter solid steel cylinder from 2 simultaneously presented stimuli, a standard and a comparison target. The porpoise was able to distinguish a standard cylinder from acrylic resin and brass targets but had difficulty distinguishing the steel from the aluminum cylinder. In terms of size discrimination, the porpoise could distinguish the standard cylinder from the 12-, 18-, and 20-mm diameter cylinders but had difficulty distinguishing the 15-mm from the 14-mm diameter cylinder. Echo measurements suggest that the porpoise could detect: 1) the time difference between the two echo highlights to within approximately 1 μs, 2) frequency shifts of approximately 7 kHz in a broadband echo with a peak frequency of nearly 140 kHz, 3) a time-separation pitch of approximately 30 kHz, and 4) target strength differences of approximately 1 dB.

Stress Reduction Studies and Reproductive Physiology on Captive Cetaceans

As previously mentioned, some cetacean species are currently endangered. Thus, it is high time that the preservation and artificial reproduction of these endangered cetaceans in captivity is considered seriously. From this perspective, it is clear that the reproductive health of captive cetaceans is crucial. It is also important to reduce stressors from the captive environment to ensure that captive cetaceans remain healthy and stress-free.

Physiology of stress

A wide spectrum of marine mammals exists in aquariums all over the world, and their keepers continuously strive to care for them and improve their captive conditions. To ensure that these animals are in the best of health, it is important to keep their stress levels as low as possible.

Basics of stress. Biologically, the term *stress* has been defined as a situation in which the homeostasis of a creature is disrupted by endogenous or exogenous stimuli (Chrousos, 1998). The typical stress response was found and named by Selye (1936) as the general adaptation syndrome, which is identified by activation of the hypothalamus-pituitary-adrenal cortex (HPA) axis and sympathetic nerves and immunosuppression. Adrenal corticosteroids, the representative of which in mammals is cortisol, are released from the adrenal cortex via the HPA axis, and adrenaline is simultaneously secreted from the adrenal medulla, which is stimulated by the sympathetic nerves. These hormones function to overcome stressful conditions that affect various organs and to recover homeostasis. As a result, the stress levels of animals may be monitored by evaluating the changes in these properties, especially in blood cortisol concentration levels (Morton, Anderson, Foggin, Kock, & Tiran, 1995).

Applied studies on stress in captive cetaceans. Studies on the stress levels of cetaceans have been performed using both captive and wild animals (St. Aubin & Dierauf, 2001; Thomson & Geraci, 1986) in an attempt to determine the stressors for captive dolphins. Cortisol concentrations have been used as the central indicator of stress.

Contrary to a report by Carballeira, Brown, Fishman, Trujillo, and Odell (1987), Suzuki et al. (2002) confirmed the biosynthesis of cortisol in the adrenal cortex of wild minke whales (*Balaenoptera acutorostrata*). Using the same method, cortisol biosynthesis was verified in the organ of bottlenose dolphins and false killer whales that died in aquariums (data unpublished). Blood cortisol concentrations have been measured in several cetacean species, and Suzuki and colleagues (2002) reported that a tendency of inverse correlation between body size and serum cortisol concentration was observed in healthy captive species under normal conditions. It also has been revealed that serum cortisol concentrations showed basal diurnal changes in several species, with higher levels seen in the early morning and the lowest levels seen in the evening at both Kamogawa Sea World and Okinawa Churaumi Aquarium (Suzuki et al., 2003). This data is indispensable in the evaluation of cortisol concentrations as a stress indicator, and the collection of such fundamental data on cortisol can only be achieved using captive animals who are accustomed to being handled by humans.

On the basis of this data, applied studies have been performed with the aim of determining the stress-inducing stimuli for captive cetaceans. As reported in some previous studies (Koopman, Westgate, Read, & Gaskin, 1995; St. Aubin & Geraci, 1989; Thomson & Geraci, 1986), the acts of human handling, capturing, and taking cetaceans into captivity were found to provoke considerable elevations in blood cortisol concentrations of cetaceans. The same was found to be true in domestic animals.

Transport is a clear stressor for dolphins, even for individuals who have been kept in captivity over a long period of time (Noda, Akiyoshi, Aoki, Shimada, & Ohashi, 2007; Suzuki et al., 2008). Suzuki and colleagues (2008) have explored new ways to improve methods of transport for Indo-Pacific bottlenose dolphins at Okinawa Churaumi Aquarium. They reported that a combination of high-performance mattresses [EV-17 (soft foam) on EE-20 (hard foam), Bridgestone Corp.] brought about a reduction in maximum and average pressures, a wider support area, and slower pressure gradients when used for the dolphins. On the mattress system, lower breathing rates, lower heart rates, and higher exhaled CO_2 concentrations were observed compared to those obtained with standard mattresses, suggesting better performances of the system in terms of dolphin cardiopulmonary function.

However, room for improvement remains regarding the thermoregulatory and psychological effects of transportation. If completely safe transportation methods for dolphins are established, animals can be moved between facilities frequently and more easily. This ability will contribute to breeding control for dolphins, which now heavily relies on sperm transportation, which will be discussed below.

Reproductive physiology and its application in captive breeding and conservation of wild populations

Reproduction is one of the most important features that we need to understand to manage cetacean species and their populations (Atkinson & Yoshioka, 2007). In aquariums that keep cetaceans for public display and education, captive breeding is given the highest priority for sustaining the population. An understanding of reproductive physiology and its application in captive breeding programs, including artificial insemination techniques, will no doubt contribute to achieving the above 2 goals both in wild and in captive cetacean populations.

In Japan, monitoring of sex steroids (estrogens and progesterone) began in the early 1980s with the goal of better understanding the estrous cycles of common bottlenose dolphins in captivity. Yoshioka, Mohri, Tobayama, Aida, and Hanyu (1986) showed that: 1) the estrous cycle of the species is about 1 month long; 2) seasonal (spring to fall), multiple, and spontaneous ovulations occurred; and 3) pseudo-pregnancy occurred during 1- to 3-year profiles of estradiol, progesterone, and luteinizing hormone in 3 females who were kept separate from adult males at Kamogawa Sea World. This was the first reproductive physiology report of cetaceans in Japan that described long-term profiles of circulating hormone levels. The aquarium’s research group continued the endocrinological and reproductive studies on their dolphins in captivity and reported hormonal diagnoses of early pregnancy by measuring serum progesterone levels (Mohri, Tobayama, & Yoshioka, 1988). They also determined sexual maturity and seasonal testicular activity of male common bottlenose dolphins and Pacific white-sided dolphins by monitoring serum testosterone levels (Atkinson & Yoshioka, 2007; Katsumata, Tobayama, Yoshioka, & Aida, 1994). Serum samples taken for the study in the late 1980s to early 1990s were collected from tail fluke veins of an animal that was physically immobilized on a stretcher at the side or bottom of the tank.

According to recently published reports on captive cetaceans, sexual cycle lengths depend on the species, being 36 days for bottlenose dolphins (Robeck, Steinman, et al., 2005), 42 days for killer whales (Robeck et al., 2004), 48 days for belugas (Robeck, Monfort, et al., 2005), and 31 days for Pacific white-sided dolphins (Robeck et al., 2009). In parallel with the study of circulating hormonal changes, rectal temperatures of belugas and killer whales were intensively and regularly measured, and new methods of detecting ovarian cycles and parturition timing were also discovered (Katsumata, Furuta, Katsumata, Watanabe, & Taya, 2006a; Katsumata et al., 2006b). In the 1990s, fresh semen could be easily collected with the hand manipulation method (Schroeder & Keller, 1989) from bottlenose and Pacific white-sided dolphins (Fig. 6), and differences in general semen characteristics and seasonal changes between the 2 species were reported (Yoshioka, 2008). During the long-term monitoring of semen characteristics, Yoshioka and colleagues found that the dolphin could ejaculate up to 10 times during one 15–20 min sampling session (Yoshioka, 2008). Kita, Yoshioka, Kashiwagi, Ogawa, and Tobayama (2001) also found differences in sperm morphology among 10 cetacean species by using sperm samples collected from captive animals and from other stranded or captured animals.

Yoshioka (2008) also successfully cryopreserved sperms from 2 species using the simplified pellet method (a modification of Schroeder & Keller, 1990) and later using the straw method for artificial insemination (a modification of Robeck, Atkinson, & Brook, 2001; Robeck & O'Brien, 2004; Yoshioka, 2008). In 2003, 3 bottlenose dolphins were artificially inseminated by fresh and/or frozen-thawed semen, and 1 female (inseminated by fresh semen) gave birth to a female calf in July 2004 (Katsumata, 2006; Robeck, Steinman, et al., 2005). In 2004, the successful birth of a male calf occurred after insemination of only frozen-thawed sperm to another female (Katsumata, 2006). Frozen sperm was exported to the US, and the first-ever artificially inseminated Pacific white-sided dolphin gave birth to a calf in 2003 (Robeck et al., 2009). Artificial insemination research thus can only be accomplished through captive studies. The importance of this method is significant in ensuring the genetic diversity of populations currently in captivity to maintain endangered species, with much less stress for the animals than transport an entire animal.

Hormonal data obtained from the samples (e.g., feces, blubber, and blow) of wild whales, dolphins, and porpoises should be evaluated and validated with reliable data from captive animals with known reproductive and other physiological conditions, before applying the results to wild populations. A basic understanding of cetacean endocrinology and reproduction gained by studying captive dolphins will no doubt greatly help in evaluating the health of wild cetaceans.

Figure 6. Semen collection from a Pacific white-sided dolphin using the hand manipulation method (Kamogawa Sea World).

Conclusion

As our reviews here show, studies of cetaceans in captivity in Japan are a vital complement to studies performed in the wild, where conditions and effects cannot be continuously monitored. Newly developed technologies including data-logging science, now commonly used in Japan, have allowed for a previously unknown understanding of cetacean behavior (Akamatsu, Wang, Wang, Naito, 2005; Sato et al., 2007). We believe that technological developments in the study of wild animals will never eliminate the necessity for basic studies on captive animals, and that a cooperative relationship between wild and captive animal research may best contribute to science and conservation.

References

Akamatsu, T., Wang, D., Wang, K., & Naito, Y. (2005). Biosonar behaviour of free-ranging porpoises. *Proceedings of the Royal Society of London, Series B, 272*, 797-801.

Atkinson, S., & Yoshioka, M., (2007). Endocrinology of reproduction. In D. Miller (Ed.), *Reproductive biology and phylogeny of cetacea: Whales, porpoises and dolphins* (pp. 171-192). Enfield, NH: Science Publishers, Inc.

Au, W. W. L. (2000). Hearing in whales and dolphins: An overview. In W. W. L. Au, A. N. Popper, & R. R. Fay (Eds.), *Hearing by whales and dolphins* (pp. 1-42). New York: Springer-Verlag.

Carballeira, A., Brown, J. W., Fishman, L. M., Trujillo, D., & Odell, D. K. (1987). The adrenal gland of stranded whale (*Kogia breviceps* and *Mesoplodon europaeus*): Morphology, hormonal contents, and biosynthesis of corticoid. *General and Comparative Endocrinology*, *68*, 293-303.

Chin, H. (1979). China's first baby giant panda reproduced by artificial insemination. *International Zoo News*, *26*, 8-9.

Chrousos, G. P. (1998). Stressors, stress, and neuroendocrine integration of the adaptive response. *Annals of New York Academy of Science*, *851*, 311-335.

Connor, R. C., Smolker, R. A., & Bejder, L. (2006). Synchrony, social behavior and alliance affiliations in Indian Ocean bottlenose dolphins (*Tursiops aduncus*). *Animal Behaviour*, *72*, 1371-1378.

Curley, J. P., & Keverne, E. B. (2005). Genes, brains and mammalian social bonds. *Trends in Ecology and Evolution*, *20*, 561-567.

Dietz, R., Shapiro, A. D., Bakhtiari, M., Orr, J., Tyack, P. L., Richard, P., et al. (2007). Upside-down swimming behaviour of free-ranging narwhals. *BMC Ecology*, *7*, 14.

Esch, H. C., Sayigh, L. S., Blum, J. E., & Wells, R. S. (2009). Whistles as potential indicators of stress in bottlenose dolphins (*Tursiops truncatus*). *Journal of Mammalogy*, *90*, 638-650.

Flanigan, W. F., Jr. (1974). Nocturnal behavior of captive small cetaceans II. The beluga whale, *Delphinapterus leucas*. *Sleep Research*, *3*, 85.

Foote, A. D., Griffin, R. M., Howitt, D., Larsson, L., Miller, P. J. O., & Hoelzel, A. R. (2006). Killer whales are capable of vocal learning. *Biology Letters*, *2*, 509-512.

Gnone, G., Benoldi, C., Bonsignori, B., & Fognani, P. (2001). Observations of rest behaviors in captive bottlenose dolphins (*Tursiops truncatus*). *Aquatic Mammals*, *27*, 29-33.

Hogg, C. J., Rogers, T. L., Shorter, A., Barton, K., Miller, P. J. O, & Nowacek, D. (2009). Determination of steroid hormones in whale blow: It is possible. *Marine Mammal Science*, *25*, 605-618.

Hoppenbrouwers, T., & Sterman, M.B. (1975). Development of state patterns in the kitten. *Experimental Neurology*, *49*, 822-838.

Janik, V. M., & Slater, P. J. B. (1997). Vocal learning in mammals. *Advances in the Study of Behavior*, *26*, 59-99.

Janik, V. M., & Slater, P. J. B. (1998). Context-specific use suggests that bottlenose dolphin signature whistles are cohesion calls. *Animal Behaviour*, *56*, 829-838.

Jaramillo-Legorreta, A., Rojas-Bracho, L., Brownell R. L., Jr., Read, A. J., Reeves R. R., Ralls, K., et al. (2007). Saving the vaquita: Immediate action, not more data. *Conservation Biology*, *21*, 1653-1655.

Katsumata, E. (2006). *Studies on reproduction of captive marine mammals*. Doctoral dissertation, The United Graduate School of Veterinary Science, Gifu University, Gifu, Japan.

Katsumata, E., Furuta, C., Katsumata, H., Watanabe G., & Taya, K. (2006a). Basal body temperature method for detecting ovarian cycle in the captive beluga (*Delphinapterus leucas*). *Journal of Reproduction and Development*, *52*, 59-63.

Katsumata, E., Jaroenporn, S., Katsumata, H., Konno, S., Maeda, Y., Watanabe, G., et al. (2006b). Body temperature and circulating progesterone levels before and after parturition in killer whales (*Orcinus orca*). *Journal of Reproduction and Development*, *52*, 65-71.

Katsumata, E., Tobayama, T., Yoshioka, M., & Aida, K. (1994). Seasonal changes in serum testosterone levels in a male bottlenose dolphin (*Tursiops truncatus*) in captivity. *Journal of the Japanese Association of Zoological Gardens and Aquariums*, *35*, 75-80.

Kishida, T., Kubota, S., Shirayama, Y., & Fukami, H. (2007). The olfactory receptor gene repertoires in secondary-adapted marine vertebrates: Evidence for reduction of the functional proportions in cetaceans. *Biology Letters*, *3*, 428-430.

Kita, S., Yoshioka, M., Kashiwagi, M., Ogawa, S., & Tobayama, T. (2001). Comparative external morphology of cetacean spermatozoa. *Fisheries Science*, *67*, 482-492.

Koopman, H. N., Westgate, A. J., Read, A. J., & Gaskin, D. E. (1995). Blood chemistry of wild harbor porpoises *Phocoena phocoena* (L.). *Marine Mammal Science*, *11*, 123-135.

Lusseau, D. (2003). The emergent properties of a dolphin social network. *Proceedings of the Royal Society of London. Series B, 270 (Supplement 2),* S186-S188.

Lyamin, O. I., Mukhametov, L. M., & Siegel, J. M. (2004). Relationship between sleep and eye state in cetaceans and pinnipeds. *Archives of Italian Biology*, *142*, 557-568.

Lyamin, O. Pryaslova, J., Lance, V., & Siegel, J. (2005). Animal behaviour: Continuous activity in cetaceans after birth. *Nature, 435*, 1177.

McCormick, J. G. (1969). Relationship of sleep, respiration, and anesthesia in the porpoise: A preliminary report. *Proceedings of the National Academy of Sciences of the United States of America*, *62*, 697-703.

Mann, J., Connor, R. C., Tyack, P. L., & Whitehead, H. (2000). *Cetacean societies: Field studies of dolphins and whales.* Chicago: University of Chicago Press.

Mohri, E., Tobayama, T. & Yoshioka, M. (1988). Hormonal diagnosis of pregnancy in captive cetaceans. *Journal of the Japanese Association of Zoological Gardens and Aquariums*, *30*, 67-70.

Morisaka, T. (2009). Overview of comparative cognitive studies of dolphins in Japan. *Japanese Psychological Research*, *51*, 168-176.

Morisaka, T., Shinohara, M., & Taki, M. (2005a). Underwater sounds produced by neonatal bottlenose dolphins (*Tursiops truncatus*): I. Acoustic characteristics. *Aquatic Mammals*, *31*, 248-257.

Morisaka, T., Shinohara, M., & Taki, M. (2005b). Underwater sounds produced by neonatal bottlenose dolphins (*Tursiops truncatus*): II. Potential function. *Aquatic Mammals*, *31*, 258-265.

Morton, D. J., Anderson, E., Foggin, C. M., Kock, M. D., & Tiran, E. P. (1995). Plasma cortisol as an indicator of stress due to capture and translocation in wildlife species. *Veterinary Record*, *136*, 60-63.

Naito, Y. (2004). Bio-logging science and new tools for marine bio-science. *Proceedings of the International Symposium on SEASTAR2000 and Bio-logging Science (The 5th SEASTAR2000 Workshop)*, Bangkok, Thailand, 72-75.

Nakahara, F. (1998). Vocal development and vocal behavior of dolphins. *IBI Report*, *8*, 43-51.

Nakahara, F. (2002). Social functions of cetacean acoustic communication. *Fisheries Science*, *68 (Supplement I),* 298-301.

Nakahara, F. (2003). Sound characteristics and echolocation ability of finless porpoises. *Kaiyo Monthly*, *35*, 571-574.

Nakahara, F. (2006). Dolphin acoustic research in aquarium. *Aquabiology*, *28*, 355-361.

Nakahara, F., & Miyazaki, N. (1997). Vocal exchange and recognition in bottlenose dolphins, *Tursiops truncatus*. *Abstract of the 25th International Ethological Conference*, Vienna, Austria.

Nakahara, F., & Takemura, A. (1997). A survey on the behavior of captive odontocetes in Japan. *Aquatic Mammals*, *23*, 135-143.

Nakahara, F., Takemura, A., Koido, T., & Hiruda, H. (1997). Target discrimination by an echolocating finless porpoise, *Neophocaena phocaenoides*. *Marine Mammal Science*, *13*, 639-649.

Noda, K., Akiyoshi, H., Aoki, M., Shimada, T., & Ohashi, F. (2007). Relationship between transportation stress and polymorphonuclear cell functions of bottlenose dolphins, *Tursiops truncatus*. *Journal of Veterinary Medical Science*, *69*, 379-383.

Norris, K. S., & Dohl, T. P. (1980). Behavior of the Hawaiian spinner dolphin, *Stenella longirostris*. *Fisheries Bulletin (US)*, *77*, 812-849.

Page, B., Goldsworthy, S. D., & Hindell, M. A. (2001). Vocal traits of hybrid fur seals: intermediate to their parental species. *Animal Behaviour*, *61*, 959-967.

Rechtschaffen, A., Gilliland, M. A., Bergmann, B. M., & Winter, J. B. (1983). Physiological correlates of prolonged sleep deprivation in rats. *Science*, *221*, 182-184.

Reiss, D. (1988). Observations on the development of echolocation in young bottlenose dolphins. In P. E. Nachtigall & P. W. B. Moore (Eds.), *Animal sonar: Processes and performance* (pp. 121-127). New York: Plenum Press.

Robeck, T. R., Atkinson, S. K.C., & Brook, F. (2001). Reproduction. In L. A. Dierauf & F. M. D. Gulland (Eds.), *CRC handbook of marine mammal medicine (2nd ed.)*. (pp. 193-236). Boca Raton, FL: CRC Press.

Robeck, T. R., Monfort, S. L., Calle, P. P., Dunn, J. L., Jensen, E., Boehm, J. R., et al. (2005). Reproduction, growth and development in captive beluga (*Delphinapterus leucas*). *Zoo Biology*, *24*, 29-49.

Robeck, T. R., & O'Brien, J. K. (2004). Effect of cryopreservation methods and precryopreservation storage on bottlenose dolphin (*Tursiops truncatus*) spermatozoa. *Biology of Reproduction*, *70*, 1340-1348.

Robeck, T. R., Steinman, K. J., Gearhart, S., Reidarson, T. R., McBain, J. F., & Monfort, S. L. (2004). Reproductive physiology and development of artificial insemination technology in killer whales (*Orcinus orca*). *Biology of Reproduction*, *71*, 650-660.

Robeck, T. R., Steinman, K. J., Ramirez, K., Greenwell, M., Van Bonn, W., Yoshioka, M., et al. (2009). Seasonality, estrous cycle characterization, estrus synchronization, semen cryopreservation and artificial insemination in the Pacific white-sided dolphin (*Lagenorhynchus obliquidens*). *Reproduction*, *138*, 391-405.

Robeck, T. R., Steinman, K. J., Yoshioka, M., Jensen, E., O'Brien, J. K, Katsumata, E., et al. (2005). Estrous cycle characterization and artificial insemination using frozen-thawed spermatozoa in the bottlenose dolphin (*Tursiops truncatus*). *Reproduction*, *129*, 659-674.

Roffwarg, H. P., Muzio, J. N., & Dement, W. C. (1966). Ontogenetic development of the human sleep-dream cycle. *Science*, *152*, 604-619.

Rutz, C., & Hays, G. C. (2009). New frontiers in biologging science. *Biology Letters*, *5*, 289-292.

Sakai, M., Hishii, T., Takeda, S., & Kohshima, S. (2006). Flipper rubbing behaviours in wild bottlenose dolphins (*Tursiops aduncus*). *Marine Mammal Science*, *22*, 966-978.

Sato, K., Watanuki, Y., Takahashi, A., Miller, P. J. O., Tanaka, H., Kawabe, R., et al. (2007). Stroke frequency, but not swimming speed, is related to body size in free-ranging seabirds, pinnipeds and cetaceans. *Proceedings of the Royal Society of London B*, *274*, 471-477.

Schroeder, J. P., & Keller, K.V. (1989). Seasonality of serum testosterone levels and sperm density in *Tursiops truncatus*. *Journal of Experimental Biology*, *249*, 316-321.

Schroeder, J. P., & Keller, K. V. (1990). Artificial insemination of bottlenose dolphins. In S. Leatherwood & R. R. Reeves (Eds.), *Bottlenose dolphin* (pp. 447-460). San Diego, CA: Academic Press.

Sekiguchi, Y., Arai, K., & Kohshima, S. (2006). Sleep in continuously active dolphins. *Nature*, *441*, E9-E10.

Sekiguchi, Y., & Kohshima, S. (2003). Resting behaviors of captive bottlenose dolphins (*Tursiops truncatus*). *Physiology & Behavior, 79,* 643-653.

Selye, H. A. (1936). A syndrome produced by diverse nocuous agents. *Nature*, *138*, 32.

Shaw, P. J., Tononi, G., Greenspan, R. J., & Robinson, D. F. (2002). Stress response genes protect against t the lethal effects of sleep deprivation in *Drosophila melanogaster*. *Nature, 417*, 287-291.

St. Aubin, D. J., & Dierauf, L. A. (2001). Stress and marine mammals. In L. A. Dierauf & F. M. D. Gulland (Eds.), *Marine mammal medicine, 2nd ed.* (pp. 253-270). New York: CRC Press.

St. Aubin, D. J., & Geraci, J. R. (1989). Adaptive changes in hematologic and plasma chemical constitutions in captive beluga whales, *Delphinapterus leucas*. *Canadian Journal of Fisheries and Aquatic Science, 46*, 796-803.

Stensland, E., & Berggren, P. (2007). Behavioural changes in female Indo-Pacific bottlenose dolphins in response to boat-based tourism. *Marine Ecology Progress Series, 332*, 225-234.

Suzuki, M., Hirako, K., Saito, S., Suzuki, C., Kashiwabara, T., & Koie, H. (2008). Usage of high-performance mattresses for transport of Indo-Pacific bottlenose dolphin. *Zoo Biology, 27*, 331-340.

Suzuki, M., Ishikawa, H., Otani, S., Tobayama, T., Katsumata, E., Ueda, K., et al. (2002). The characteristics of adrenal glands and its hormones in cetaceans. *Fisheries Science*, *68 (Supplement I)*, 272-275.

Suzuki, M., Uchida, S., Ueda, K., Tobayama, T., Katsumata, E., Yoshioka, M., et al. (2003). Diurnal and annual changes in serum cortisol concentrations in Indo-Pacific bottlenose dolphins *Tursiops aduncus* and killer whales *Orcinus orca*. *General and Comparative Endocrinology*, *132*, 427-433.

Tamaki, N., Morisaka, T., & Taki, M. (2006). Does body contact contribute towards repairing relationships? The association between flipper-rubbing and aggressive behavior in captive bottlenose dolphins. *Behavioural Processes*, *73*, 209-215.

Thomson, C. A., & Geraci, J. R. (1986). Cortisol, aldosterone, and leucocytes in the stress response of bottlenose dolphins, *Tursiops truncatus*. *Canadian Journal of Fisheries and Aquatic Science*, *43*, 1010-1016.

Tomonaga, M., Uwano, Y., Ogura, S., & Saito, T. (2010). Bottlenose dolphins’ (*Tursiops truncatus*) theory of mind as demonstrated by responses to their trainers’ attentional states. *International Journal of Comparative Psychology*, *23,* 386-400.

Turvey, S. T., Pitman, R. L., Taylor, B. L., Barlow, J., Akamatsu, T., Barrett, L. A., et al. (2007). First human-caused extinction of a cetacean species? *Biology Letters, 3,* 537-540.

Würsig, B., & Würsig, M. (1980). Behavior and ecology of dusky dolphin, *Lagenorhynchus obscurus*, in the South Atlantic. *Fisheries Bulletin (US), 77,* 871-890.

Yoshioka, M. (2008). Present status and future of artificial breeding of small cetaceans in captivity. In H. Kato (Ed.), *Mammalogy in Japan Vol. 3* (pp. 123-146), Tokyo: University of Tokyo Press.

Yoshioka, M., Mohri, E., Tobayama, T., Aida, K., & Hanyu, I. (1986). Annual changes in serum reproductive hormone levels in the captive bottlenosed dolphins. *Bulletin of the Japanese Society of Scientific Fisheries*, *52*, 1939-1946.

Zornetzer, H. R., & Duffield, D. A. (2003). Captive-born bottlenose dolphin x common dolphin (*Tursiops truncatus* x *Delphinus capensis*) intergeneric hybrids. *Canadian Journal of Zoology*, *81*, 1755-1762.