NEONATE DEVELOPMENT
OF THE
ATLANTIC BOTTLENOSE DOLPHIN
(*TURSIOPS TRUNCATUS*)

DRAFT

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Abstract

Atlantic Bottlenose Dolphins (*Tursiops truncatus)* are the most common occurring species of the order *Cetacea* in zoos, aquariums, and marine life parks around the world. However, scientific knowledge about cetaceans is limited. In addition, there is a poor understanding of the factors influencing reproductive success and neonate development. My study, conducted at Dolphin Quest in Waikoloa, Hawai'i, involved collecting and compiling data on the development of four Atlantic Bottlenose Dolphin neonates. I observed one neonate daily from October through November of 1996, and I documented data on nursing bout durations, use of right and left teats, and linear growth. Changes in neonate length reveal a rapid growth increase over the two month study period. The data on hourly nursing bout durations show a decrease over time for all of the calves. Results indicate that neonates exhibited no preference for right or left teats. Effective nursing of dolphin neonates is vital to their survival. Therefore, although nursing patterns may not be conclusive in determining calf needs, any deviations from normal development patterns may be very useful in assessing the health of neonates. The study of animals in captivity can benefit cetaceans in the wild. There is much to be learned from cetaceans in marine life parks to help keep wild populations healthy and safe.

Introduction

The world's human population is approximately 5.6 billion and is increasing exponentially. More than 50 percent of the U.S. human population inhabits an area within 80 km of the coast (OECD, 1991). As coastal populations grow more stress is being place on already strained coastal environments. Among those affected are the Atlantic Bottlenose dolphin (*Tursiops truncatus*), which are coastal water inhabitants. As degradation of marine habitats continues, Bottlenose dolphins in the wild are affected in many ways (Grace, 1996). Human disturbances have negatively affected the spawning of fish species that dolphins prey on. In addition, man-made toxins are thought to be responsible for mass die-offs of Atlantic Bottlenose dolphins (Darling *et al*, 1995). Human encroachment not only leads to degradation of our coastal waters, but also disrupts cetaceans natural feeding, migration, and resting patterns.

Bottlenose dolphins are the most common occurring species, of the order *Cetacea*, found in zoos, aquariums, and marine life parks around the world. We have acquired a mass amount of knowledge from dolphins, and other cetaceans, by studying their mental capabilities, use of echolocation, and even their anatomy. This includes modeling cetaceans form and functions, and the use of sonar to help adapt humans to aquatic environments. Cetaceans also have important ecological roles, not only as top predators in their environment, but as a major food source for deep sea eco-systems. For example, the remains of one whale may fuel mini eco-systems in the deep sea for over 15 years (Morlan, 1994). But now in a time of habit degradation, incidental deaths (like drowning in fishing nets), direct cetacean takes (hunting or intended kills), and with increased marine mammal strandings, the need for cetacean conservation is essential. Moreover, the scientific knowledge we have acquired about cetaceans is limited. This extends to a poor understanding of the factors influencing reproductive success and calf development. This deficiency makes determining effective avenues for conservation difficult. If science is going to aid in the conservation of cetaceans, fundamental, applied, and monitoring research must be employed (Hutchinson and Simmonds, 1996). The data with reference to nursing bout durations and the growth of cetaceans are inadequate. Accordingly, the main goals of this study were to establish baseline data for one captive Atlantic Bottlenose dolphin neonate, compare those data with those of other neonates, and then to establish norms to calf development.

The Bottlenose dolphin only produces one offspring every two to three years. Neonates are subjected to many biological and physical stresses. Coupled with the dependency of the calf on its mother, survival for neonates born in the wild is very low, roughly 15-20% (Kirtland, 1993). Dolphins born in captive environments have a survival rate of about 50%; of which, the majority die within the first month of life. Although there are many reasons for the high mortality rate, poor suckling habits and/or the absence of milk are high contributing factors (Krames and Krames, 1994). Effective nursing of dolphin calves is vital to their survival. For a number of reasons, including the inexperience of the mother to nurse, young mothers often lose their first and second calves (Sweeney, 1986). Unlike some marine mammals that are thought to be born with immunoglobulin, cetaceans are not (Sweeney, *pers. com*). In the first few days of nursing, a neonate will receive antibodies from the mother's milk. These antibodies are needed to protect the neonate from micro-organisms, which co-exist in their environment (Krames and Krames, 1994). If a neonate is not nursing effectively, it may not receive ample antibodies, thereby running the risk of illness or death. The prosperity of cetacean populations is largely dependent on the birth and survival rates of neonates (Darling, 1995). For these reasons it is vital that we are not only able to determine what affects survival, but detect abnormalities in these factors as well. With this study, I sought to answer three main questions:

1) Do neonates exhibit a preference for left or right teats and could absence of preference be an indicator of health?

2) How does the duration of nursing bouts change as the neonate gets older and is that nursing pattern consistent with other neonates?

3) What does the growth pattern look like?

There have been two previous studies investigating neonate development; however, both projects collected data for only one calf. Therefore, with my research I hope to fill some of the gaps in our understanding of cetacean needs.

Background

Dolphin Quest, located in the Hilton Waikoloa Village of Hawai'i, was opened by veterinarians Dr. Jay Sweeney and Dr. Rae Stone in 1988 and is home to eleven Atlantic Bottlenose dolphins. Dolphin Quest is a public display facility that has made a commitment towards education and conservation of marine mammals and their environment. Dolphin Quest offers interactive programs, educational programs for children, and research support. Fifteen percent of the proceeds produced by the dolphin encounters and retail goods sold are given to the Waikoloa Marine Life Fund, a non-profit organization that funds research projects and educational programs.

The pregnancy of two dolphins at Dolphin Quest led to an internship and research opportunity. The first neonate was stillborn due to complications during birth; however, the second neonate was born healthy on October 3, 1996. The mother, Leilani, was a twelve-year old female dolphin that has resided at Dolphin Quest since its beginning in the spring of 1988. Her neonate, named Kainalu, was the focus of my study. Leilani gave birth to one calf previously, Lokahi, on July 8, 1994. Lokahi was an unhealthy calf; however, medical intervention from the Dolphin Quest staff brought him back to health. Data from Lokahi (Calf 4) and two other calves, of mothers Pele (Calf 3) and Kona (Calf 2), born at Dolphin Quest in Fall of 1997 were compiled to use in comparison with Kainalu (Calf 1).

Female cetaceans have two nipples, called teats, retracted into slits along either side of the genital slit. It is worthy to note that neonates do not suckle. When a neonate sets up to nurse, the mother will push the teat out of the slit, the neonate will grasp it with his tongue, and the mother uses her muscles to expel the milk into the calf's mouth. This mode of transfer for nursing is advantageous for the calf, because the milk intake is accelerated (Harrison and Bryden, 1988).

The two previous studies on calf development looked at use of right and left teats, nursing frequencies, and growth patterns for one calf (Cockcroft and Ross, 1990; Peddemors *et al.*, 1992). Both of these studies analyzed teat preference by using a student's t-test. The use of a t-test may be inadequate for the question of neonate teat preference. Teat preference is likely not only a question of the time the calf spends on the teat, but also the frequency of teat use. In addition, because the mother is using her muscles during nursing events, it is probable that the mother is the one to terminate a nursing bout; therefore the t-test alone would not be a good estimation of preference for the calf.

Materials and methods

This research was conducted in Hilton Waikoloa Village, on the Big Island of Hawai'i, where Dolphin Quest is located. A *quantitative behavior sampling technique* was used for this study. Focal animal sampling, where an individual animal is the focus of each observation session, was conducted, having the neonate as the focal animal (Altmann, 1974). While the mother and calf were in the main lagoon, observations took place underwater using a mask and snorkel. The man-made lagoon holds 2,000,000 gallons of sea water, which is continuously replaced by 8,333 gallons a minute. The sea water is pumped in at the bottom of the lagoon from the ocean. When the mother and calf were in the nursing pen, which is 24.3 m by 6.1 m and 2.4 m in depth, observations were conducted above water from a dock.

Once the neonate was born, the study period began and then continued for eight weeks. Daily, for the first two weeks, Dolphin Quest staff and interns assisted in data collection. In that time frame, observations were recorded for eight to ten hour periods. From two to four weeks, there was a single observer, and observations were conducted for a minimum of four hours per day. From four to eight weeks, observations were continued every two to three days, for four hours daily.

Within the observation time, every successful nursing attempt was recorded, including the duration of the event (also called period or bout) and which teat was used. The suckling period was recorded once contact of the neonate and teat had occurred, by counting the number of seconds the neonate spent on the teat. Initial hook up time was not counted into the bout time, and the bout count was terminated once the calf separated from the teat.

Every two to three days throughout the study period, photographs of the mother and calf were taken, using underwater disposable cameras. This method was used to track the linear growth of the neonate. Photographs were taken when the calf was directly beneath the mother; therefore, errors of parallax could be avoided or lessened (Cockcroft and Ross, 1990; Peddemors *et al*, 1992). The known length of the mother was used as a standard to assess the linear growth of the neonate. From these assessments, weekly averages were determined for the linear growth. Direct measurements of the calf could not be taken because the stress to the calf due to handling could result in injury or even death (Sweeney, 1986; Cornell *et al*, 1987). Moreover, there is a bond and a level of trust between the animals and their caretakers, and handling the calf may jeopardize that relationship.

The use of right and left teats was examined in Kainalu and Lokahi using a Chi-Square Analysis and a two-sample t-test. The hourly nursing durations were calculated for all four neonates and were then plotted and examined for trends; however, the number of days for data collection varied for each neonate. Their slopes were compared using a regression analysis, and the mean nursing durations were compared using a one-way analysis of covariance. Reliable photographs were taken for only neonate, Kainalu; therefore, only his weekly averages were plotted and examined for trends.

Results

To address the first question on teat preference, data from two calves were analyzed using a Chi-Square Analysis to test the null hypothesis that the neonates displayed no preference for right or left teats (Table 1). There were a total observed 1052 bouts for Kainalu, yielding a test statistic of 2.57. That is less than the critical value of 3.841 ($\alpha = 0.05$, $v = 1$); therefore, the null hypothesis was accepted and Kainalu showed no preference. For Lokahi, the unhealthy calf, there were 761 observations, which yielded a test statistic of 1.431. This is much less than the critical value of 3.841($\alpha = 0.05$, $v = 1$); hence, this calf showed no teat preference.

Table 1: Results of Chi-Square Analysis for teat preference of two Atlantic Bottlenose dolphin (*Tursiops truncatus*) neonates.

	Left Teat	Right Teat	Test Statistic	P-Value
Kainalu	552	500	2.57	>0.05
Lokahi	397	364	1.431	>0.05

The second part of teat preference was analyzed by using a t-test to test the null hypotheses that the mean nursing durations were not different for left or right teats. The test statistics for both Kainalu and Lokahi were below the critical values of 1.962 ($\alpha(2)$ = 0.05, v=1000) and 1.963 ($\alpha(2)$ = 0.05, v=700) respectively; therefore, there was no difference between mean durations on each teat (Table 2). The strength of this design and the likelihood of making a type II error was tested using a power analysis on the data from both calves. The minimum detectable difference was set at 10%. The power of both tests were 0.99; therefore, β=1%.

Table 2: Results of student's t-test for teat preference of two Atlantic Bottlenose dolphin (*Tursiops truncatus*) neonates.

	Test Statistic	Critical Value	P-value	Power analysis
Kainalu	-1.12	1.962	0.26	0.99
Lokahi	0.55	1.963	0.58	0.99

To address the second question, the average nursing bout durations were calculated for each day. Kainalu's bout durations started out very high, spending well over one minute of every hour nursing (Fig. 1). Over time, there was a decrease in hourly bout durations.

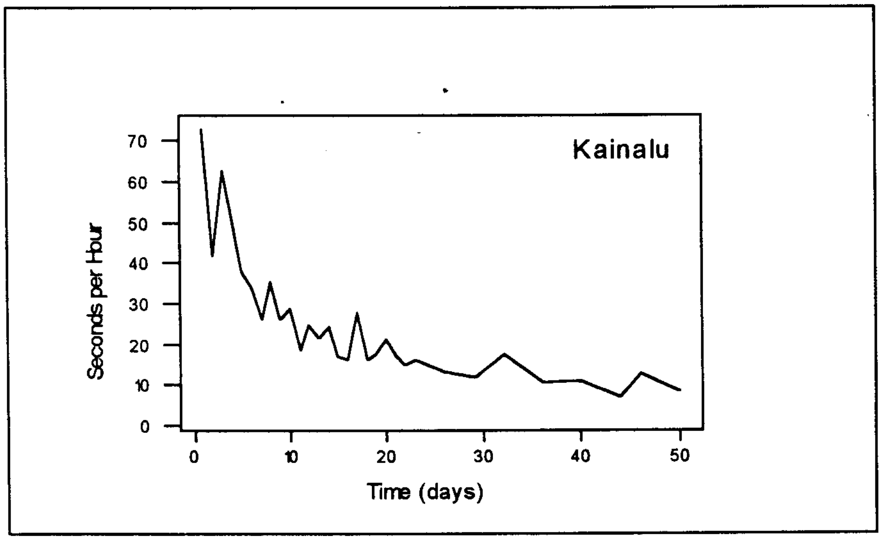

Figure 1: Hourly bout durations for a healthy Atlantic Bottlenose dolphin (*Tursiops truncatus*) neonate, shown in seconds per hour, from parturition to two months of age.

For comparison of data from all four calves, the plots of nursing durations are shown in Figures 2-5. Only the first ten days of calculations are represented for Kainalu (Fig. 2) to allow for a more valuable visual comparison.

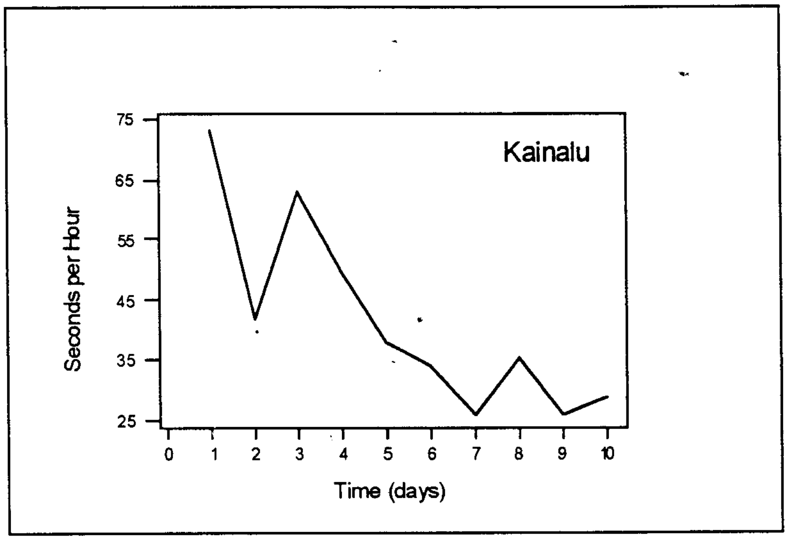

Figure 2: Hourly bout durations for a healthy Atlantic Bottlenose dolphin (*Tursiops truncatus*) neonate, shown in seconds per hour, from parturition to ten days old.

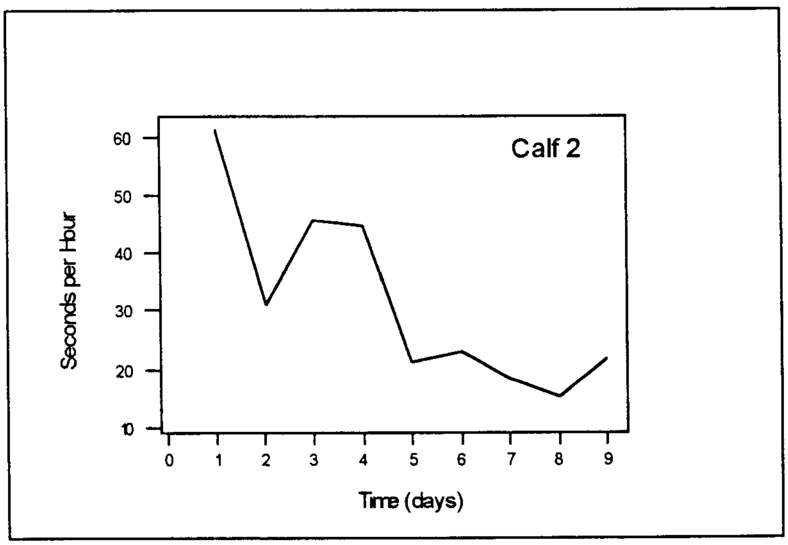

Figure 3: Hourly bout durations for a healthy Atlantic Bottlenose dolphin (*Tursiops truncatus*) neonate, shown in seconds per hour, from parturition to nine days old.

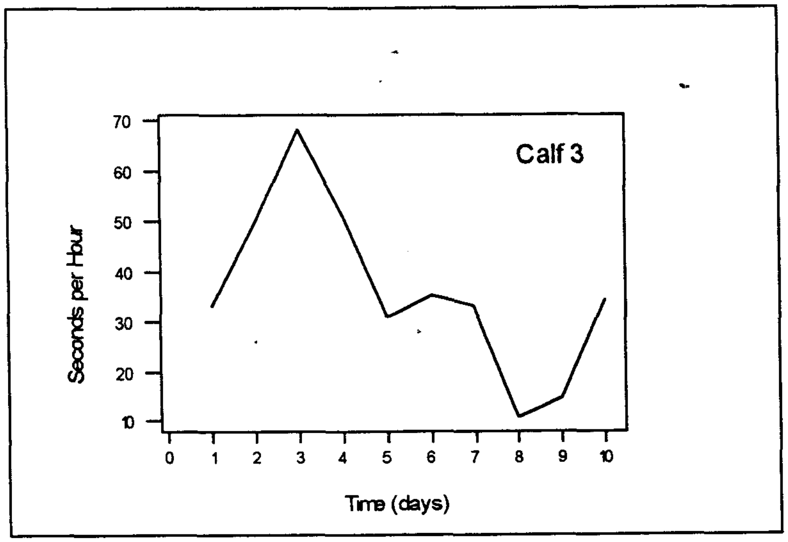

Figure 4: Hourly bout durations for a healthy Atlantic Bottlenose dolphin (*Tursiops truncatus*) neonate, shown in seconds per hour, from parturition to ten days old.

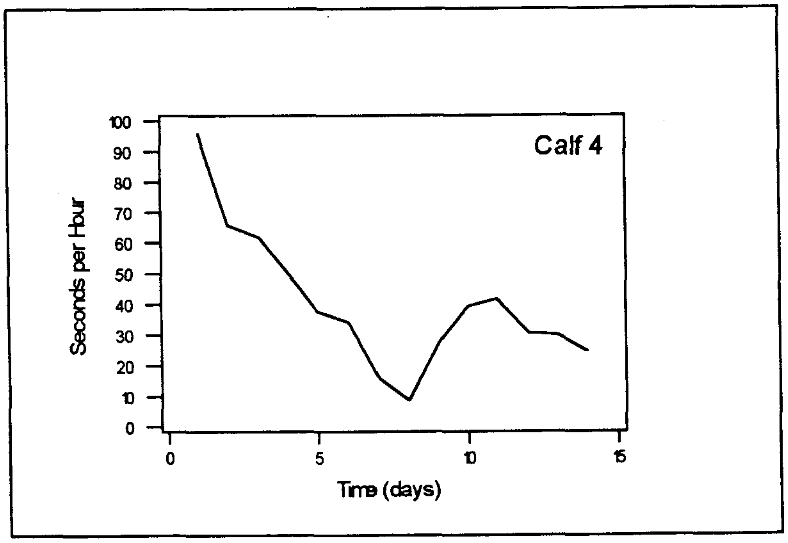

Figure 5: Hourly bout durations for an unhealthy Atlantic Bottlenose dolphin (*Tursiops truncatus*) neonate, shown in seconds per hour, from parturition to fourteen days of age.

For three of the neonates data was limited; therefore, a regression analysis was done on only the first nine days. A regression plot of all four neonates (Fig. 6) shows all calves having inversely negative slopes.

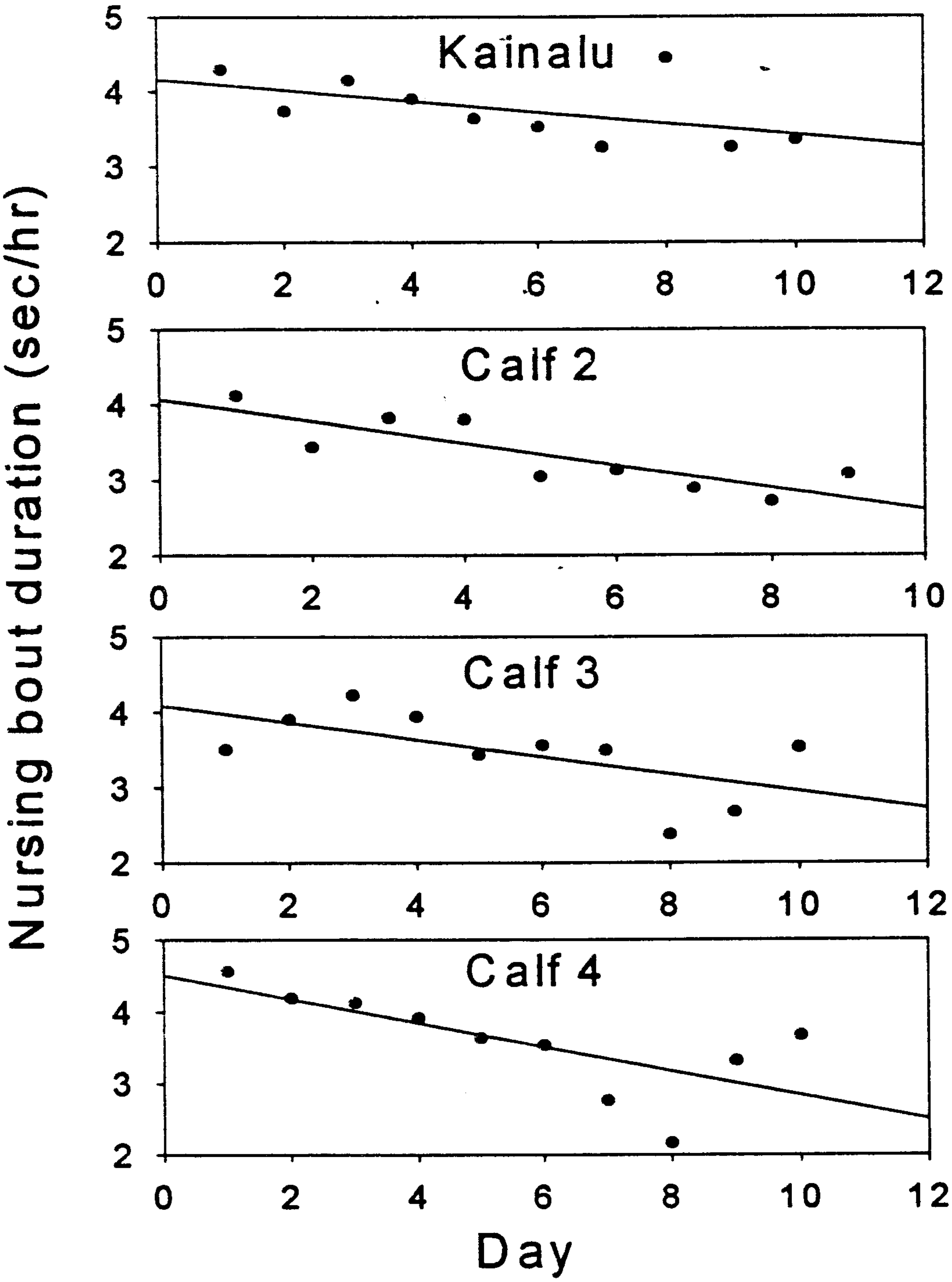

Figure 6: Regression plot for four Atlantic Bottlenose dolphin (*Tursiops trucatus*) neonates. All have been log transformed to achieve higher R-values for normal distribution and to achieve linearity. The top three were normal healthy calves; however, Calf 4 had many health complications.

The results for the analysis of covariance show that for the covariate, which is day, the p-value is less than 0.05 (Table 3); therefore there is a significant difference among days. For the adjusted means of the calves, the p-value is well above 0.05 (Table 3); therefore, there was not a significant difference for nursing durations among all calves.

Table 3: Analysis of Co-variance testing the mean hourly nursing bout durations for four Atlantic Bottlenose dolphin (*Tursiops trucatus*) neonates.

	P-value
Covariate	0.000*
Nursing Durations	0.113

* indicates significance at α= 0.05

* critical assumption of equality of slopes was not tested

Growth for Kainalu was rapid over the entire 8 week study period (Fig. 7). At birth he was about 96 cm. After one month, he grew to 123 cm; therefore, growth was about one foot for the first month.

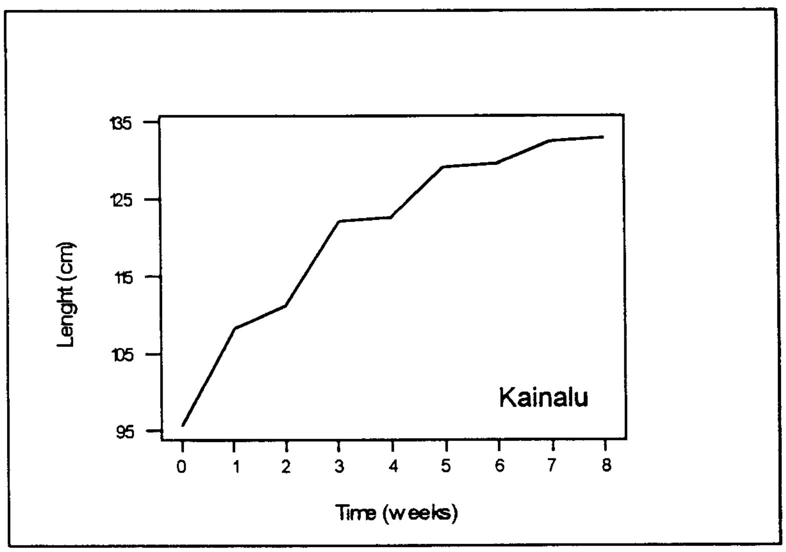

Figure 7: Linear growth of an Atlantic Bottlenose dolphin (*Tursiops truncatus*) neonate from partrution to eight weeks of age. Growth measurements were calculated using photographs taken of calf and mother.

Discussion

Although both tests for teat preference determined there to be no preference for the calves, some trends are evident. Considering the difference between the Chi-square test statistics for the two calves, Lokahi's test statistic is about half of that of Kainalu's (Table 1). Moreover, on examination of the test statistics from the t-test (Table 2), Kainalu's statistic is much closer to the critical value than Lokahi. Albeit there was not a statistical significance, this may suggest that a healthy calf may favor one teat over another; whereas an unhealthy calf may be less choosy. If the mother were to have an infection, as in the case of Lokahi's mother, it would likely effect the milk of both mammary glands.

With the exception of Calf 3, all neonates spent a very high number of seconds per hour nursing and then durations significantly drop over the first few days. This dramatic decrease in durations may be due to a variety of factors; however, it is most likely that the neonate and mother are getting adapted to one another and becoming more efficient in nursing. In the case of Calf 3, the neonate was born late in the day and attempted to nurse soon after parturition. Because observations started prematurely and only a few hours were observed, the calculations of hourly durations are underestimated. Calf 3 also had a drop in nursing bout durations around day eight. This deviation was not the result of poor health. Observation records indicate that due to the birth of Calf 2, many nursing attempt were disrupted. As the neonates get older they have higher nutritional requirements; however, the nursing durations decrease over time. The mother's milk is likely going through compositional changes that enable greater amounts of milk and protein to the calf with less time spent nursing. Studies examining milk composition have found that there are fluctuations in milk content throughout lactation of many species and among individuals within species (Oftedal, 1984; Pervaiz and Brew, 1986; Ridgway *et al,* 1995).

This unhealthy Calf 4, Lokahi showed no apparent deviations in nursing durations. His slope on the regression plot (Fig. 6) was not different from the other three neonates. However, because the plot only shows the first 10 days, his health problems would not be reflected in the slope unless it was continued out for a longer time interval. The analysis of co-variance also indicated that there was not a significant difference for nursing durations among all calves. As a result of the Calf 4's poor health, he was given an injection of pure immunogobulin from the father's blood and administered 100 ml of stored milk, from another other, that was rich in antibodies (Krames and Krames, 1994). If more data was incorporated from the sick calf and/or there was no medical intervention, there may have resulted in a significant difference.

A definite nursing pattern can be established; however, there is a possibility of error due to the untested critical assumption of equality of slopes. Deviations from normal patterns could be an indicator of poor nursing habits, or illness of either the calf or mother. Ineffective nursing can lead to pneumonia, which is a common cause of death in neonates (Cornell *et al*, 1987). Neonates are born with little blubber, and need to put on fat quickly to aid in regulation of their body temperature.

This rapid growth increase is similar to the results of two previous studies (Cockcroft and Ross, 1990; Peddemors *et al*, 1992). The high growth rate for cetaceans is attributed to the high fat and protein content found in cetacean milk (Harrison and Bryden, 1988). Milk fat in cetaceans has been reported as high as 50%, but with the Bottlenose dolphin, the levels seemingly peak in the range of 20-30% fat and 11% protein (Oftedal, 1984; Pervaiz and Brew, 1986; Ridgway *et al,* 1995). For the first few months of life, nursing is the calf's only source of nutrition (Shane, 1988). Therefore, the decrease in nursing bout durations is not a result of weaning or consumption of fish. Between the third and fifth month of age, calves will begin to consume fish. Around the ninth to twelfth month, fish become a large portion of their diet (Cornell, 1987). Calves may continue to nurse anywhere from one and a half to three years after parturition.

There are many questions still left unanswered by this study. More neonates need to be studied to get an accurate picture of nursing and growth trends. It is hard to determine the relationship between nursing bout durations and growth, because the nursing duration alone will not suggest the amount of nutrients that are actually transferred to the neonate. A calf may nurse for other reasons, aside from obtaining nutrition, which include bonding with the mother. Therefore, future studies could calculate the amount of milk that is transferred to the calf while nursing. Establishing the nursing patterns and milk transfer will enable caretakers to not only monitor the health of calves but also allow for hand-rearing of the neonate if the mother is ill or dies.

By studying the development of neonates, baseline data is available for future analysis. Klinowska (1991) states that management and husbandry are crucial to the well-being of dolphins in captivity. This data will enhance our understanding of marine mammal care in captive environments, provide details of bioenergetics, and ensure effective husbandry.

This also applies to wild dolphins. There is a lot to be learned from marine mammals in marine life parks to help keep wild populations healthy and safe. Data on nursing allows marine mammal veterinarians to interpret the needs of stranded animals. In the case of a stranding where a cetacean calf is involved, rescuers could properly care for the neonate. The more we know about their needs, the better we can plan to minimize human impacts. The Atlantic Bottlenose dolphin is just one of many species that inhabits coastal waters and is affected by pollution and habitat degradation. Our collective goal should be to strive every day to educate our children to the delicate balance between humans and wildlife and to teach them their responsibility to protect the environment.

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