CAN STRESS HORMONES BE USED AS AN INDICATION OF FOOD AVAILABILITY AND REPRODUCTIVE PERFORMANCE? AN EXPERIMENTAL APPROACH.

Project Number:

Restoration Category:	Research (new)
Proposed By:	DOI-USGS; University of Antwerp, Belgium
Lead Trustee Agency:	DOI-USGS
Cooperating Agencies:	N/A
Alaska SeaLife Center:	No
Duration:	1st year, 1-year project
Cost FY 01:	$18,900
Geographic Area:	Middleton Island
Injured Resource:	Black-legged Kittiwake, Pelagic Cormorant

ABSTRACT

This study will complement and enhance an existing *Exxon Valdez* Oil Spill Trustee Council (EVOSTC) project (99479) that investigates how stress hormone levels (i.e., corticosterone) in adult seabirds relate to local food conditions and indicate the future reproductive health of a colony. This study will (1) test for differences in corticosterone levels between supplementally fed and unfed black-legged kittiwakes that are nesting at one colony, thereby removing any inherent environmental differences present when birds from two colonies are compared, (2) measure changes in corticosterone levels in adults throughout the breeding season including birds recently arriving from the wintering grounds, (3) explore the effects of adult gender on corticosterone levels, and (4) evaluate how corticosterone levels relate to the an individual's reproductive success and survival, as well as overall productivity of the colony. Results from a pilot study conducted in 1999 indicate the relationships among stress hormones, food availability, and reproduction may be more complex than previously suggested. A second year of data collection planned for the summer of 2000 will help elucidate these relationships. We are asking for minimal financial support to analyze plasma samples collected in 2000, and salary to complete and publish manuscripts. This study will assist other efforts to evaluate the efficacy of using corticosterone levels to assess the effects of fluctuations in food supply on the reproductive performance and survival of seabirds.

INTRODUCTION

In the Gulf of Alaska, some seabird and marine mammal species have experienced reduced productivity, increased mortality and resultant population declines during the past few decades (Hatch 1987, Merrick et al. 1987, Pitcher 1990, Hatch et al. 1993a, Byrd et al. 1999). Black-legged kittiwakes (*Rissa tridactyla*), for example, have failed to reproduce at many colonies in Alaska (Murphy et al. 1991, Hatch et al. 1993a, Irons 1996). Productivity in Alaska kittiwake colonies has declined from an average of about 0.6 young per nest in the 1960s and 1970s to less than 0.2 young per nest in the 1980s (Hatch et al. 1993a). Circumstantial evidence points to a deficient food supply as the cause of poor productivity in Alaskan kittiwakes and other seabirds (Hatch 1987, Hatch and Hatch 1990, Hatch et al. 1993a, Roberts and Hatch 1993, Piatt and Anderson 1996, Anderson and Piatt 1999). Depressed food supplies or other natural perturbations (called 'modifying factors') are thought to increase circulating levels of stress hormones (e.g., corticosteroids) in free-living animals (Wingfield et al. 1997), and cause these individuals to redirect their behavior away from reproduction towards survival (Silverin 1998). This stress response is thought to be an emergency reaction that promotes survival (e.g., through increased foraging) while temporarily suspending other unessential activities (Sapolsky 1987, Wingfield 1994).

The level of stress in an individual can be determined through measurements of circulating plasma levels of corticosterone in the peripheral system of most vertebrates (Wingfield et al. 1997). Recently, researchers have suggested using this "field endocrinology" approach to monitor the health of seabird populations (Wingfield et al. 1997; A. Kitaysky and J. Piatt, unpubl. proposal to the EVOSTC 1999). Indeed, these investigators suggest stress hormone levels can be used to assess the current health of a population (i.e., likelihood of successful reproduction) and to predict the vulnerability of the population to other deleterious events. They cite additional advantages to measuring stress hormones, including ease of obtaining samples, subject animals not being harmed, and the ability to collect incisive information in a few days. Traditional field methods, in contrast, may require weeks to collect alternative information, and enable biologists to recognize problems only after they have had an effect on a large number of individuals.

Despite the appeal of using corticosterone levels as predictors of individual or colony health, stress hormones have the disadvantage of being responsive to a variety of physical, seasonal and environmental factors. For example the adrenocortical response, which produces stress hormones, can be affected by an individual's body weight, body condition, and gender (Astheimer et al. 1995, Fowler et al. 1994, Holberton et al. 1996, Wingfield et al. 1994, 1997, 1999; Duffy and Belthoff 1997; Newman and Zinkl 2000). Stress hormone levels also may change within a breeding season (Wingfield et al. 1997) or within a breeding stage if food availability changes or the level of parental care changes. In species like black-legged kittiwakes, which provide biparental care, males and females within a pair might exhibit different corticosterone levels if they contribute unequally to parental care (K. O'Reilly, pers. comm.). Additional environmental factors that may affect corticosterone levels include pollution, habitat conditions (Marra et al. 1998), intraspecific competition for nest sites or mates, and predation. Indeed, any

stimulus that is perceived as imposing a threat, either real or anticipated, may cause a physiological response that results in increased stress hormone levels (Newman and Zinkl 2000). Consequently, care must be taken when cause and effect relationships that include corticosterone levels and other biological factors are investigated. Such care should include experimental manipulations that alter only the factor thought to increase stress hormone levels (e.g. food) while standardizing as many other factors as possible (e.g., habitat, predation, parental care needs).

To date, the relationship between food supply and corticosterone levels have been poorly investigated. A study on black-legged kittiwakes and common murres (*Uria aalge*) found that adults sampled at a food-rich colony had lower baseline levels of corticosterone compared to adults sampled at a food-poor colony (Kitaysky et al. 1999). Because stress hormone levels in these birds may have been caused by factors other than food (e.g., differences in predation levels or habitat characteristics between colonies or in the gender of sampled birds), these results provide only qualitative support for this relationship. A more controlled laboratory study found that black-legged kittiwake chicks, raised without parents, had elevated baseline levels of corticosterone when their diets were qualitatively and quantitatively restricted compared to chicks fed ad libitum (Kitaysky et al. 1999). No study to date has provided food to free-ranging adult seabirds at natural breeding colonies and tested for changes in corticosterone levels.

The relationship between stress hormone levels and reproduction has also been poorly investigated. Silverin (1986) found that male and female pied flycatchers (*Ficedula hypoleuca*) experimentally implanted with corticosterone decreased the frequency with which they fed their young and consequently had lower reproductive success. Similarly, black-legged kittiwake adults implanted with corticosterone brooded their young less and had lower inter-year return rates, although food provisioning to chicks by parents and chick fledging success was not lowered (Kitaysky et al. MS). Such changes in parental care suggest a negative relationship between elevated corticosterone levels and reproductive success. Unfortunately, sample sizes in these studies were low, and additional studies are needed to verify the reproductive consequences of elevated corticosterone levels.

A long-term decline in black-legged kittiwake numbers on Middleton Island (north-central Gulf of Alaska) coupled with the development of a unique study colony on the island presents an exceptional opportunity to explore more fully the relationships between stress hormone levels, food availability, and reproduction. Formerly one of the largest aggregations of kittiwakes anywhere in the world, black-legged kittiwakes at Middleton have declined from 166,000 in 1981 to fewer than 16,000 today, an 85% decrease (Fig. 1). Mean productivity on Middleton from 1983 to 1999 has been 0.06 chicks per pair (S. Hatch and V. Gill, unpubl. data), with successful nests restricted to man-made structures such as an old radar tower on an abandoned Air Force base and portions of a World War II shipwreck. Nesting attempts on natural cliffs surrounding the island have decreased, and virtually no young have fledged from such sites since 1988 (S. Hatch and V. Gill, unpubl. data).

Between 1995 and 1999, biologists have enhanced the radar tower to promote nesting by kittiwakes, while simultaneously allowing experimentation and observation of birds. To determine if food was limiting the productivity of kittiwakes on Middleton, a supplemental feeding study was initiated on the tower in 1996. Over the next four years, reproductive data, collected on birds limited to natural food conditions and birds supplementally fed, confirmed that food was limiting the ability of birds to reproduce (see details below). In 1998, an effort was begun to understand the physiological aspects of food availability and reproduction. Adult and chick testosterone levels were measured and related to food availability, sibling aggression, and chick survival (Lanctot et al. in prep[a]). Corticosterone levels of supplementally fed and unfed adults were sampled in 1999 for the first time, providing the initial data on which we base this proposal. We propose conducting a second year of data collection in 2000.

Our goal is to investigate whether baseline levels of corticosterone in free-living black-legged kittiwakes reflect stress associated with a deficient food supply, and to determine how corticosterone levels correlate with parental care, reproductive effort and success, and survival of individual black-legged kittiwakes. These relationships will be explored at the radar tower colony on Middleton Island, where food can be experimentally provided, adults can be observed and captured with ease, and corticosterone levels, reproductive performance, and survival of nesting individuals can be monitored accurately with minimal disturbance.

NEED FOR THE PROJECT

A. Statement of Problem

Biologists have actively monitored the status of seabird colonies in the North Pacific for more than 30 years (Hatch et al. 1994). A principal goal of these studies has been to understand the factors that regulate seabird populations and their ability to recover from natural and man-induced environmental perturbations (USFWS 1992). Most studies station personnel at seabird colonies where they monitor seabird breeding effort and reproductive success, and relate these parameters to local environmental conditions (Cairns 1987, Monaghan 1996). Given the expense and personnel time required to conduct such studies, researchers have searched for alternative ways to monitor seabird colonies that are inexpensive, practical, and applicable over a large geographic area. One such alternative is the measurement of stress hormones that may reflect local food conditions and the likelihood of successful breeding. In 1999, the EVOSTC funded project 99479 to investigate how corticosterone levels in adult seabirds relate to local food conditions and whether corticosterone levels can be used as an indicator of colony health. Preliminary comparisons between a food-rich (Gull Island) and a food-deprived (Chisik Island) seabird colony suggest that corticosterone levels in breeding adults reflect local food availability. Although these results seem promising, corticosterone measurements must be interpreted carefully. Stress hormones have the disadvantage of being responsive to a variety of physical, seasonal and environmental factors. This study will aid in the interpretation of project 99479 in four important ways. First, this study

will test for differences in corticosterone levels between food limited (natural foraging conditions) and food unlimited (supplementally fed) black-legged kittiwakes nesting at one colony. In this way, any confounding environmental differences, such as predation levels and habitat characteristics, present when comparing birds from two colonies will be removed. Second, this study will measure changes in stress hormones in adults throughout the breeding season, including birds that have recently arrived from the wintering grounds. Relatively little is known about how food conditions on the wintering grounds influences a bird's ability to reproduce in the subsequent summer, and whether local food conditions on the breeding grounds can alter this ability. Third, this study will explore the effects of adult gender on baseline levels of corticosterone. Because male and female kittiwakes may contribute unequally to the reproductive effort (e.g., only males feed females during courtship), we predict stress hormone levels of males to be more strongly affected by food availability, especially before the breeding season. Fourth, the unique colony of kittiwakes nesting on the abandoned radar tower on Middleton Island will allow the relationships between corticosterone, food availability and colony reproductive success to be studied accurately and with little disturbance. Nest site characteristics are virtually identical throughout the tower, there is no predation of eggs or chicks, and adults can be sampled for corticosterone within 2 minutes of initiating a capture. Taken together, these factors make the tower colony at Middleton Island an ideal setting to explore the physiological aspects between food availability, reproduction and survival.

B. Rationale/Link to Restoration

Efficient and reliable methods are needed to determine how seabird populations respond to natural and human-induced environmental perturbations. Indeed, little is known about how seabirds respond to such stressful conditions and whether such conditions influence reproduction and survival of birds. Recently, researchers have suggested using physiological parameters indicative of an individual bird's stress level as a measure of a bird's potential to reproduce and survive. Traditional methods for assessing seabirds do not evaluate the direct relationships between environmental conditions, adult physiology and reproduction; they simply monitor whether individuals breed and survive. To evaluate the relationships between environmental conditions, adult physiology, and reproduction requires a unique research facility where environmental conditions can be experimentally modified, adult physiology can be measured, and the resulting reproduction can be monitored. Such a setting occurs at Middleton Island where a wild population of black-legged kittiwakes nests on the sides of an abandoned radar tower. This study will complement and enhance project 99479 by allowing a thorough assessment of the use of a stress hormones as a tool for assessing the effects of fluctuations in food supply on the survival and reproductive performance of seabirds.

C. Summary of Major Hypothesis

Ho: There are no significant differences in baseline levels of circulating corticosterone between black-legged kittiwakes that are supplementally fed and those relying solely on natural food conditions, after controlling for the effects of gender and breeding stage.

Ho: There is no significant relationship between natural forage fish conditions and baseline levels of circulating corticosterone in unfed black-legged kittiwakes.

Ho: There is no significant difference in parental care, breeding chronology, reproductive success, or over-winter survival between black-legged kittiwakes with elevated and depressed baseline levels of corticosterone, after controlling for the effects of gender, breeding stage, and food availability.

D. Location

The proposed field experiments will be conducted on Middleton Island (north-central Gulf of Alaska). Studies will be conducted at an abandoned radar tower that has been retrofitted to hold 210 nest sites equipped with supplemental feeding tubes and sliding glass windows. Among these sites, we have established two treatment groups to evaluate how stress hormone levels are affected by food provisioning, and to relate these levels to kittiwake reproduction and survival. Approximately 16,000 kittiwakes were counted on Middleton in 1999, of which 1,206 nested on the tower. An additional 100 nest sites are being prepared in April 2000 to support nesting of pelagic cormorants (*Phalacrocorax pelagicus*). See description of pelagic cormorant study below.

COMMUNITY INVOLVEMENT

Each year between four and six undergraduate and graduate students assist us in studying the seabirds on Middleton Island. These students learn not only about black-legged kittiwakes but also conduct studies on tufted puffins, rhinocerous auklets, glaucous-winged gulls and black oystercatchers. To date, two post-doctoral studies, three Master's theses, and one honor's thesis have been conducted on the seabirds at Middleton Island. These and other studies have involved collaboration among researchers from the University of Alaska Anchorage, Oregon State University, University of Antwerp (Belgium), Konrad Lorenz Institute for Comparative Ethology (Vienna), and the National Oceanic and Atmospheric Administration. In addition, numerous private groups visit the island each year to observe the large diversity of bird life that occurs on the island. For example, the Alaska state chapter of the Audubon Society is planning a field trip to Middleton in May. We typically invite these people to visit the tower and we explain our field research to them. The seabirds of Middleton Island were recently filmed by All Bird TV yielding a documentary entitled "Mysterious Middleton Island" that aired on the Animal Planet channel last fall.

The principal investigator will be available to present highlights of the research program to interested parties; and will provide information, photographs and articles for the Trustee Council newsletter if requested.

PROJECT DESIGN

A. Background and Results of Pilot Studies

Establishment of Tower Colony

Groundwork for this study has been laid by capitalizing on the fact that kittiwakes nest on an artificial structure on Middleton Island - an abandoned radar tower - which, with various enhancements has resulted in a colony that is uniquely accessible for observation and experimentation (Fig. 2a). As the exterior corrugated siding of this building was removed by high winds over the years, a limited number of horizontal ledges (2x4 cross members in the wall frame) became available to kittiwakes as nest sites. The first pair of kittiwakes nesting on the tower was noted in 1986, and numbers have steadily increased to the present level of 1200 pairs. In 1994, the tower walls were outfitted with 216 wooden ledges and the deteriorating inner wallboard was replaced with plywood paneling. Between 1995 and 1998, one-way sliding glass windows and feeding holes were installed behind 210 of these ledges (Fig. 2b-d). The feeding holes are designed so that plywood plugs can be inserted when pairs are not being fed. The one-way glass is virtually 100% effective, allowing feeding and observations of the birds to be conducted unobtrusively from inside the tower while the birds nest on ledges surrounding the tower. In 1996, a permanent AC power source was installed at the tower, enabling us to store large amounts of frozen fish on site in three chest freezers.

Supplemental Food Provisioning

Commercially available bait herring and capelin have been used to supplementally feed kittiwakes on the tower between 1996 and 1999. A recent analysis of kittiwake prey obtained in Prince William Sound indicated herring and capelin were at the high end of prey quality (Anthony and Roby 1997). Roughly, 4000 pounds of supplemental food were provided to kittiwake nest sites each year of our studies. Thiamine (vitamin B1) will be added to the capelin diet as a food supplement because this nutrient is known to be deficient in frozen fish (Altman et al. 1997, Crissey 1998). Members of each pair are fed individual items of food ad libitum each day in the morning and evening (and at mid-day during chick-rearing) by observers positioned inside of the building.

Capture and Measurement of Birds

In addition to the sliding one-way glass windows, narrow grooves have been cut in the plywood panels beneath each window at the height of the bird's tibio-tarsus (Fig. 2d). This groove allows researchers to slip coat hangers, whose ends are fashioned into small hooks, underneath adults and hook them by their feet. This method works so well that adults can be captured repeatedly within and between field seasons. P. Jodice and D.

Roby (unpubl. data) used this technique at the tower to capture 73 individuals twice during a 24-hr period for a doubly-labeled water study in 1998. The technique has the added advantage of catching birds quickly with no pre-capture disturbance that could artificially inflate baseline levels of corticosterone. Indeed, an initial analysis investigating how time from capture to blood sampling effects baseline corticosterone levels indicated a significant increase in corticosterone levels within as little as 2 min (Fig. 3). Previous researchers have noted a ten-fold or greater increase in plasma corticosterone levels measured within a few minutes of capturing and handling of a bird (Beuving and Vonder 1978, Harvey et al. 1980). Capturing of adult kittiwakes at natural cliffs with noosepoles in under 2 min is virtually impossible given the approach time needed to reach birds. Adults have been captured since 1995 at the tower, and virtually all adults nesting on the ledges are individually marked with a unique combination of color bands and USFWS rings. Unique markings allow us to follow individual birds throughout and across breeding seasons so as to monitor their breeding chronology, reproductive success and over-winter survival. The sex of most adults on the tower has been determined through a combination of morphological measurements, behavior, and genetics (Jodice et al., in press). Adult measurements also have proven useful for generating adult body condition indices which can be related to baseline levels of corticosterone.

Effects of Food Provisioning on Kittiwake Breeding Chronology and Reproduction

The easy viewing of kittiwake nest sites through the windows also allows nests to be monitored in detail. We can record easily the date when (1) adults begin to construct nests, (2) eggs are laid, lost or hatched, and (3) chicks are lost or fledged. The eggs and chicks residing in each nest can also be easily measured by simply opening the window behind each nest. For example, eggs can be individually measured for size, and then marked so that the emerging chick can be identified. This level of detail allows us to determine how food supplementation and egg order is related to egg size and chick sex (Lanctot et al. In prep [b]). Similarly, chicks can be measured at specific ages to determine growth, and blood samples can be taken periodically to determine changes in hormone concentrations. The one-way glass windows also allows the behavior of adults and their young to be observed easily. Depending on the parameter in question, up to 10 nests can be monitored simultaneously. During the past four years, we have measured adult attendance at nests, male and female feeding rates of young, chick begging rates (by first and second hatched chicks), and sibling aggression rates.

Results to date indicate unequivocally that supplementally feeding kittiwakes alters a variety of breeding parameters relative to kittiwakes subjected to natural food supplies (Gill 1999; Gill and Hatch, in prep.; Gill et al., in prep). During pre-egg laying, fed females begged food from their mates at significantly greater rates and attended their nests at lower rates than unfed females. During egg-laying and incubation, fed females initiated nests 3-4 days earlier and laid significantly larger second eggs than unfed females. During chick-rearing, fed pairs hatched their young earlier, had faster growing young, and had young that attained significantly heavier peak weights than unfed pairs. Further, parents at fed sites had significantly higher nest attendance during the second

half of brood-rearing than parents at unfed sites. On average, unfed chicks survived at significantly lower rates than fed chicks, primarily because of the extremely low survival rate of second hatched chicks in the unfed nests. These results suggest a strong relationship between food availability and kittiwake breeding success. If stress hormones in adults are related to food availability, then we would predict kittiwake adults that are supplementally fed to have lower baseline levels of corticosterone relative to unfed kittiwake pairs.

Pilot Study of Corticosterone Levels at the Middleton Kittiwake Colony

In 1999, we initiated a pilot study to investigate how baseline levels of corticosterone in adult kittiwakes relate to food supplementation and reproduction. We collected blood samples from 10 males and 10 females that were and were not supplementally fed during three reproductive stages (prior to egg-laying, incubation and brood-rearing). In addition, blood was sampled from 20 birds one day after their arrival at Middleton in early April—about 1 ½ to 2 months prior to egg laying. Supplemental feeding began on 15 May in 1999, and pairs received food for about 26 days prior to the first eggs being laid. An analysis of corticosterone levels indicated kittiwakes that were supplementally fed had similar base-line levels of corticosterone as kittiwakes limited to natural food conditions during the pre-egg laying and incubation stages of reproduction (Fig. 4). This was not true during the chick-rearing stage, however, when supplementally fed kittiwakes had significantly lower levels of corticosterone (Fig. 4). Because we could not control the amount of food unfed kittiwakes obtained through natural foraging, these results appeared to indicate that unfed kittiwakes were able to obtain adequate amounts of natural food during pre-egg laying and incubation stages but not during brood-rearing (under the assumption that food availability is related to corticosterone levels). Our independent measures of food availability (quantity and type of food brought to nest sites by kittiwakes, rhinocerous auklets and tufted puffins) and reproductive success, however, did not support this supposition. Indeed, 1999 was a year with lower than normal food availability early in the season followed by an increase in food availability later in the season (S. Hatch and V. Gill, unpubl. data). Kittiwakes limited to natural food conditions also had poorer reproductive success than fed kittiwakes during pre-egg laying and incubation for most reproductive parameters (Table 1). During chick-rearing, there was no difference in fledging success (a key seabird monitoring variable) between fed and unfed kittiwakes despite significant differences in corticosterone levels (Table 1).

Taken together, these results suggest the relationships among adult base-line levels of corticosterone, local food conditions, and the reproductive success of a colony may be more complicated than previously thought. One factor that has been neglected is how the gender of a bird influences those parameters, though previous studies have identified gender as an important predictor of corticosterone levels (Duffy and Belthoff 1997, Wingfield et al. 1999). Further, an energetics study on the tower colony indicated male kittiwakes expend more energy in raising offspring than do females (P. Jodice and D. Roby, unpubl. data), suggesting females may be less stressed by deficient food supplies. Because most of the kittiwakes breeding on the tower had been previously marked and sexed (Jodice et al., in press), we investigated the effects of sex and food supplementation

on corticosterone levels. Contrary to our expectations, we found no differences between males and females during pre-egg laying, incubation and chick-rearing stages (data not shown). There was, however, a tendency for females sampled during the pre-nesting stage to have higher levels of corticosterone if they did not subsequently breed at the tower colony (Fig. 5). No such pattern was found in males. Unfortunately, small sample sizes hamper a meaningful biological interpretation of the data from 1999.

We plan to explore these relationships further in 2000 by repeating this experimental design and supplementing our sample sizes during the pre-nesting stage.

B. Objectives

1. Determine whether male and female kittiwakes exhibit different baseline levels of corticosterone throughout the breeding season (i.e., pre-nesting, pre-egg laying, incubation, and brood-rearing) when they are and are not supplementally fed.
2. Determine whether baseline levels of corticosterone in unfed individual kittiwakes reflect local natural food conditions.
3. Determine whether parents with elevated levels of corticosterone provide lower levels of parental care (i.e., attendance during brood-rearing), have delayed breeding chronologies (i.e., laying, hatching and fledging dates), have lower reproductive success (i.e., egg laying success, hatching success, fledging success), and have lower over-winter survival.

C. Methods

The proposed research investigates wild birds nesting on the tower at Middleton Island. Field experiments will be conducted on birds marked in previous years to investigate the endocrinological characteristics of kittiwakes that have ad libitum food and those limited to natural forage conditions. Although we will focus on black-legged kittiwakes in this study, we also plan to begin testing methods for sampling pelagic cormorants that also breed on ledges of the tower (see below).

Corticosterone Sampling

To determine baseline levels of corticosterone, adult kittiwakes will be captured at the tower using hooked hangers (see above). Approximately 100-200 μL of blood will be drawn from the basilic vein of the wing of each adult within 2 minutes after capture is initiated (i.e., the time between inserting the hooked hanger into the window and when the blood is successfully extracted). After sampling, birds will be released at the tower and their nest will be monitored to determine when they return. Field sampling in 1998 and 1999 indicated adult kittiwakes typically returned to their nests within 1 to 10 minutes and successfully incubated eggs and raised broods afterwards (R. Lanctot, V. Gill, and S. Hatch, pers. obs.). Blood samples will be centrifuged within 1 hour of collection and the separated plasma will be stored below freezing until it can be removed from the field and sent to the Laboratory of Immuno-Neuroendocrinology at the University of Leuven in Belgium. Laboratory technicians will determine corticosterone

levels blindly for each sample using radioimmuno-assay techniques (as described in Wingfield et al. 1992). This is the same technique being used by A. Kitaysky and J. Piatt in their corticosterone research on black-legged kittiwakes. Each sample will be measured in duplicate and intra- and inter-assay coefficients of variation will be calculated.

We anticipate sampling 40 birds (10 males and 10 females each from fed and unfed pairs) during each breeding stage (i.e., pre-egg laying, incubation and brood-rearing) in 2000. A larger sample of pre-nesting birds will be sampled within a few days after being first observed at the tower on Middleton. Blood will be collected from each bird only once during the breeding season to ensure samples from each stage are independent. Hormone levels within the pre-nesting birds may reflect their physiological response to prior winter conditions because the birds may not have been present in the local area sufficiently long to reflect local food conditions. Pre-egg laying birds will be sampled within 10 days of their first egg being laid. Ten days is the time it takes black-legged kittiwakes to form eggs (Neuman et al. 1998). Incubating birds will be sampled 14-16 days after their first egg is laid, and brood-rearing birds will be sampled 19-21 days after their first chick has hatched.

Natural Food Supply

The composition and seasonal changes in the natural food supply of kittiwakes will be determined three ways. First, we will collect regurgitated food samples from adults and chicks throughout the season. Adults and chicks instinctively regurgitate when captured. Emphasis will be placed on kittiwake pairs that are not being fed. Prey composition and biomass of collected regurgitations will be determined by laboratory analysis in the fall of 2000 through a contract with Falco Inc. (Alan Springer, pers. comm.). Second, we will conduct beach seining in the waters surrounding Middleton to monitor seabird prey. Third, we will collect food deliveries of adult tufted puffins and rhinocerous auklets to their young during brood-rearing. Although these species may use different forage fish as food, our long-term database (back to 1978) on food deliveries by these species allows the food availability within each year to be evaluated. These three sources of information will provide a crude but quantitative view of the natural food supply available to kittiwakes nesting at the tower. Based on the composition and quantity of forage fish, we will determine whether food (especially high caloric food) is lacking during each stage of reproduction. Here we would predict elevated corticosterone levels to be present in unfed kittiwakes during reproductive stages when the quantity or quality of food is limited. In contrast, kittiwakes that are supplementally fed should have consistently low baseline levels of corticosterone.

Corticosterone, Reproductive Success and Survival

To determine whether birds with elevated levels of corticosterone exhibit different life history tactics, we will contrast birds with elevated and depressed baseline levels of corticosterones with the level of parental care, breeding chronology, reproductive effort and success, and over-winter survival of individuals within the fed and unfed treatment

groups. Food supply has been shown to affect these parameters in previous studies (Gill 1999; V. Gill and S. Hatch, unpubl. data).

Parental Care

As an indication of parental care, we will measure adult attendance at nests during brood-rearing. Previous research on Middleton indicated that adult attendance was highly sensitive to food availability and can have a strong effect on chick survival (in contrast to feeding rate which did not differ with food availability; see Gill 1999). Adult attendance will be quantified by recording the percentage of time each parent (male or female) is observed at its nest during incubation and chick-rearing.

Breeding Chronology

We will record two parameters that are indicative of breeding chronology. These will include laying date (date at which first egg of nest is laid) and hatching date (date at which first egg of nest hatches). These variables are extremely easy to record at the tower given our ability to approach nests without being seen.

Reproductive Effort

As a measure of reproductive effort, we will record the clutch size, egg size (egg length and width converted into egg volumes using Coulson's [1963] formulas), and egg weight (A and B eggs alone and together).

Reproductive Success

Measures of reproductive success will include egg laying success (whether pairs successfully laid at least one egg or not), hatching success (whether a pair successfully hatched at least one egg or not), fledging success (whether a pair successfully fledged at least one young or not). We will also determine the productivity (number of chicks fledged/nest) of fed and unfed treatment groups.

Over-winter Survival

Over-winter survival is relatively easy to determine in black-legged kittiwakes because adults exhibit strong nest site and mate fidelity across years (Coulson and Thomas 1985, Hatch et al. 1993b). For our purposes, over-winter survival will be the percentage of marked adults known to breed on the tower in one year that return to Middleton in a subsequent year. We conduct searches at all the Air Force buildings and natural cliffs to locate marked birds from previous years.

Statistical Analyses

For most analyzes, corticosterone levels will be compared and contrasted across food treatment groups (i.e., fed and unfed birds). We will attempt to reject the null hypothesis of no difference in baseline corticosterone levels (over all nest sites within a treatment group) between kittiwakes that are supplementally fed and those that are not. Similarly, we will test for differences in reproduction and survival between birds with high and low levels of corticosterone. Data will be normalized, if necessary and possible, and differences will be tested with parametric methods such as a one-way and multi-variate analysis of variance. Where data cannot be normalized, we will rank-transform data and/or use non-parametric methods such as the Kruskal-Wallis ANOVA to test for differences in results.

Pelagic Cormorant Pilot Study

Measuring baseline levels of corticosterone in pelagic cormorants has the potential to offer new insights into the physiological relationship between food availability and reproductive success. Cormorants, unlike black-legged kittiwakes, have the ability to dive for their food and may therefore have access to different (and additional) food resources. Several researchers have suggested that diving seabirds are able to buffer decreases in food availability better than surface feeders because they are not restricted to a two-dimensional feeding zone and have more flexible time budgets (i.e., time available to forage; Burger and Piatt 1990, Monaghan 1996). These differences might result in decreased corticosterone levels for these species. The development of a pelagic cormorant colony at the Middleton tower, along with the existing black-legged kittiwake colony, will facilitate an inter-species comparison that can investigate how foraging strategies interacts with food supply and reproductive performance.

In April 2000, we will begin testing protocols for capturing, banding, and sampling blood from pelagic cormorants using newly installed window sites on the tower. Adult pelagic cormorants are less tolerant to handling (R. Lanctot, pers. obs.) and frequently abandon nest sites after being captured. Consequently, we plan on using anesthetics (e.g., isoflurane) during capture to reduce abandonment. This technique has proven to be successful with other intolerant bird species (P. Flint, pers. comm.). If successful, we will sample up to five adults during each breeding stage in 2000. Cormorant chicks, in contrast, can be easily captured and sampled with little disturbance. We plan on measuring base-line levels of corticosterone from 10 nests (1 chick/nest) in 2000.

V. Cooperating Agencies, Contracts, and Other Agency Assistance

Seabird biologists Ms. Verena Gill and Dr. Scott Hatch of the US Geological Survey, with the assistance of five field assistants, will carry out the field component of this study. All field expenses associated with this project are borne by the Biological Resources Division of the US Geological Survey. Support is requested to provide salary to Dr. Richard Lanctot to analyze and write the reports and publications associated with this study. Dr. Marcel Eens of the University of Antwerp in Belgium will provide

laboratory space at the Laboratory of Immuno-Neuroendocrinology in Leuven (through a collaboration with the University of Antwerp) and will coordinate the radioimmuno-assay analyses of plasma samples.

Permission to work on the tower and camp on Middleton Island has been received from Midico Corporation - a private consortium of business people who retained title of the land in the mid-1960s (Andy Milner, pers. comm.). The Federal Aviation Administration has also given us permission to land on the island runway and have access to water and electricity from their local facility. We have authorization from the Alaska Biological Science Center, Biological Resources Division of the U.S. Geological Survey and the State of Alaska to capture, band and sample blood from kittiwake adults and young. We have operated under similar permits since 1978 when field research began on the island.

SCHEDULE

A. Measurable Project Tasks for FY 01 (October 1, 2000 – September 2001)

Field work will be conducted by us during the summer of 2000. Funds are being sought to analyze plasma samples from this summer, and to analyze and write manuscripts describing our results from the 1999 and 2000 field seasons.

October – December:	corticosterone analyses, regurgitated food analysis, conduct preliminary analyses of 1999 data.
January 16 – 26:	Attend Annual Restoration Workshop
February 11-14:	Attend Pacific Seabird Group Meeting
January – April 15:	Analyze 2000 data and prepare manuscript
April 15:	Submit final report.

B. Project Milestones and Endpoints

A determination of how baseline levels of corticosterone in male and female kittiwakes are related to natural and experimental changes in food availability will require at least two years of field research. The first year of data was collected in 1999 and the second year will be collected this summer. Consequently this project milestone will have been accomplished prior to receiving funds from the EVOSTC. Smaller, but more significant, milestones include analyses of plasma samples for corticosterone due December 2000, and analyses and manuscript preparation due April 15, 2001.

C. Completion Date

The study will be completed by April 15, 2001.

PUBLICATIONS AND REPORTS

A final report, which will be in manuscript format, will be presented to the EVOSTC by April 15, 2001. Presently, we plan on submitting two manuscripts from this study. The

principal manuscript resulting from this study will be submitted to Hormones and Behavior and would be entitled “Effects of food availability on corticosterone levels and breeding success in male and female black-legged kittiwakes: an experimental study”. A second paper, entitled “Effect of sampling time on the measurement of circulating levels of corticosterone in black-legged kittiwakes” will be submitted to Auk.

PROFESSIONAL CONFERENCES

We plan on presenting the results of this study at the Annual Restoration Workshop to be held in mid-January in Anchorage, and at the Pacific Seabird Group meeting to be held 11-14 February 2001 at Kauai, Hawaii. For both conferences, Dr. Lanctot, in collaboration with Ms. Gill, Dr. Hatch, and Dr. Eens, will present a talk describing how natural and experimental changes in food availability relate to corticosterone levels and reproductive success in male and female black-legged kittiwakes.

NORMAL AGENCY MANAGEMENT

The US Geological Survey would not normally conduct field endocrinology research. Blood sample collection equipment and laboratory analyses conducted to date have been funded by the Belgium Fund for Scientific Research (via a post-doctoral fellowship to Dr. Lanctot and a project grant to Dr. Marcel Eens). The post-doctoral fellowship ended in February 1999 and there is little money left in the project grant. Accordingly, no money is available to finance the plasma samples from 2000 or to pay Dr. Lanctot to complete the analysis and prepare manuscripts for this study.

COORDINATION AND INTEGRATION OF RESTORATION EFFORT

This study tests an alternative endocrinology method for assessing forage availability at seabird colonies throughout the state of Alaska. The study complements and improves upon project 99479 that investigates similar concepts using two breeding islands that differ in natural forage availability. This study eliminates potential confounding problems associated with the breeding islands mentioned above, and investigates in more detail how baseline levels of corticosterone vary with food availability and breeding stage, and whether corticosterone levels are predictive of future reproduction and over-winter survival. Overall, this very controlled study will test the efficacy of using corticosterone to measure seabird responses to other environmental disasters.

PROPOSED PRINCIPAL INVESTIGATOR

Dr. Richard Lanctot
U.S. Geological Survey
Alaska Biological Science Center
1011 East Tudor Road
Anchorage, Alaska 99503
Phone: 907-786-3609
Fax: 907-786-3636
E-mail: Richard_lanctot@usgs.gov

PRINCIPAL INVESTIGATORS

Principal Investigator and Project Leader – Dr. Richard Lanctot, Research Wildlife Biologist, Alaska Biological Science Center, Biological Resources Division, U.S. Geological Survey, Anchorage, Alaska. Obtained Ph.D. in Behavioral Ecology from Carleton University in Ottawa, Ontario in 1996. Since 1989, he has studied the breeding biology of seabirds and shorebirds throughout Alaska. His post-doctoral fellowship was on the effects of food supplementation on offspring sex ratio in black-legged kittiwakes, and how chick testosterone levels relate to sibling aggression and juvenile survival. He will be responsible for analyzing data and writing manuscripts.

Dr. Scott Hatch, Seabird Project Leader, Alaska Biological Science Center, Biological Resources Division, U.S. Geological Survey, Anchorage, Alaska. Obtained Ph.D. in Zoology from University of California, Berkeley in 1985. Since 1978, he has funded and led research studies on seabirds at Middleton Island. He has published extensively on seabird population trends in the North Pacific and is in charge of the North Pacific Seabird Monitoring Database. He will continue to supervise the research on Middleton in 2000.

Ms. Verena Gill, Seabird Wildlife Biologist, Alaska Biological Science Center, Biological Resources Division, U.S. Geological Survey, Anchorage, Alaska. Obtained M.S. in Biology from the University of Alaska Anchorage in 1999 (thesis topic was "Breeding performance of black-legged kittiwakes in relation to food availability: a controlled feeding experiment"). She has coordinated all aspects of field work at Middleton since 1993 and her thesis work laid the ground work for this study. She will continue to coordinate the field work on Middleton in 2000, and will be primarily responsible for the field collection of blood samples.

Dr. Marcel Eens, Professor in Biology at the University of Antwerp in Belgium. Obtained Ph.D. in Behavioral Ecology at the University of Antwerp in 1992, and has published extensively on hormone effects on behavior in a variety of species. He supervised Dr. Lanctot during his post-doctoral fellowship studies on hormones in kittiwakes. He will supervise the analysis of plasma samples at his laboratory in Belgium.

OTHER KEY PERSONNEL

Research Assistants – at least 4 volunteer biotechnicians and 1 biologist will be employed from May through August 2000 on Middleton Island.

LITERATURE CITED

Altman, R.B., S.L. Clubb, G.M. Dorrestein, and K. Quesenberry. 1997. Avian Medicine and Surgery. W.B. Saunders Co., Philadelphia, PA.

Anderson, P.J. and J.F. Piatt. 1999. Community reorganization in the Gulf of Alaska following ocean climate regime shift. Marine Ecology Progress Series, 189:117-123.

Anthony, J.A. and D.D. Roby. 1997. Variation in lipid content of forage fishes and its effect on energy provisioning rates to seabird nestlings. Pages 725-729 in Forage Fishes in Marine Ecosystems. Proceedings of the International Symposium on the Role of Forage Fish in Marine Ecosystems. Alaska Sea Grant College Program Report No. 97-01. University of Alaska Fairbanks.

Astheimer, L.B., W.A. Buttemer, and J.C. Wingfield. 1995. Seasonal and acute changes in adrenocortical responsiveness in an Arctic-breeding bird. Hormones and Behavior 29:442-457.

Beuving, G. and G.M.A. Vonder. 1978. Effect of stressing factors on corticosterone levels in the plasma of laying hens. General and Comparative Endocrinology 35:153-159.

Burger, A.E. and J.F. Piatt. 1990. Flexible time budgets in common murres: buffers against variable prey abundance. Studies in Avian Biology 14:71-83.

Byrd, G.V., D.E. Dragoo, and D.B. Irons. 1999. Breeding status and population trends of seabirds in Alaska in 1998. U.S. Fish and Wildl. Serv. Report Alaska Maritime National Wildlife Refuge 99/02.

Cairns, D.K. 1987. Seabirds as indicators of marine food supplies. Biological Oceanography 5:261-267.

Coulson, J.C. 1963. Egg size and shape in the kittiwake *Rissa tridactyla* and their use in estimating age composition of populations. Proceedings of the Zoological Society of London 140:211-227.

Coulson, J.C. and C.S. Thomas. 1985. Changes in the biology of the kittiwake *Rissa tridactyla*: a 31-year study of a breeding colony. Journal of Animal Ecology 54:9-26.

Crissey, S.D. 1998. Handling fish fed to fish-eating animals: a manual of standard operating procedures. U.S. Dept. of Agriculture, Agriculture Research Service, National Agriculture Library.

Duffy, A.M., Jr., and J.R. Belthoff. 1997. Corticosterone and the stress response in young western screech-owls: effects of captivity, gender, and activity period. Physiological Zoology 70:143-149.

Fowler, G.S., J.C. Wingfield, P.D. Boersma, and R.A. Sosa. 1994. Reproductive endocrinology and weight change in relation to reproductive success in the Magellanic penguin (*Spheniscus magellanicus*). General and Comparative Endocrinology 94:305-315.

Gill, V.A. 1999. Breeding performance of black-legged kittiwakes (*Rissa tridactyla*) in relation to food availability: a controlled feeding experiment. M.S. thesis, University of Alaska, Anchorage.

Gill, V.A. and S.A. Hatch. In prep. Components of productivity in black-legged kittiwakes (*Rissa tridactyla*): response to supplemental feeding. Journal of Animal Ecology.

Gill, V.A., S.A. Hatch, and R.B. Lanctot. In prep. Sensitivity of breeding parameters to food supply in black-legged kittiwakes (*Rissa tridactyla*). Journal of Avian Biology.

Harvey, S., B.J. Merry, and J.G. Phillips. 1980. Influence of stress on the secretion of corticosterone in the duck (*Anas platyrhynchos*). Journal of Endocrinology 87:161-171.

Hatch, S.A. 1987. Did the 1982-1983 El Niño-Southern Oscillation affect seabirds in Alaska? Wilson Bulletin 99:468-474.

Hatch, S.A., G.V. Byrd, D.B. Irons, and G.L. Hunt, Jr. 1993a. Status and ecology of kittiwakes (*Rissa tridactyla* and *R. brevirostris*) in the North Pacific. Pages 140-153 *in* K. Vermeer, K.T. Briggs, K.H. Morgan, and D. Siegal-Causey (eds.), The status, ecology and conservation of marine birds in the North Pacific. Can. Wildl. Serv. Spec. Publ., Ottawa.

Hatch, S.A. and M.A. Hatch 1990. Components of breeding productivity in a marine bird community: key factors and concordance. Canadian Journal of Zoology 68:1680-1690.

Hatch, S.A., G.W. Kaiser, A.Y. Kondratyev, and G.V. Byrd. 1994. A seabird monitoring program for the North Pacific. Transactions of the 59th North American Wildlife Natural Resources Conference (1994):121-131.

Hatch, S.A., B.D. Roberts, and B.S. Fadely. 1993b. Adult survival of black-legged kittiwakes *Rissa tridactyla* in a Pacific colony. Ibis 135:247-254.

Holberton, R.L., B. Helmuth, and J.C. Wingfield. 1996. The corticosterone stress response in gentoo and king penguins during the non-fasting period. Condor 98:850-854.

Irons, D.B. 1996. Size and productivity of black-legged kittiwake colonies in Prince William Sound before and after the *Exxon Valdez* oil spill. Pages 720-737 in: Exxon Valdez Oil Spill Symposium Proceedings. Rice, S.D., R.B. Spies, D.A. Wolf, and B.A. Wright (Eds.). American Fisheries Society Symposium 18, Bethesda, Maryland.

Jodice, P.G.R., R.B. Lanctot, V.A. Gill, D.D. Roby, and S.A. Hatch. In press. Differentiating Male and Female Black-legged Kittiwakes by Behavior, Morphology, and DNA. Waterbirds.

Kitaysky, A.S., J.C. Wingfield, and J.F. Piatt. 1999. Dynamics of food availability, body condition and physiological stress response in breeding black-legged kittiwakes. Functional Ecology 13: 577-584.

Kitaysky, A.S., J.F. Piatt, and J.C. Wingfield. MS. Parent-offspring feeding interactions during food shortages. II. How does a parent black-legged kittiwake respond to a stressful event?

Kitaysky, A.S., J.F. Piatt, J.C. Wingfield, and M. Romano. 1999. The adrenocortical stress-response of black-legged kittiwake chicks in relation to dietary restrictions. Journal of Comparative Physiology B. 169:303-310.

Lanctot, R.B., M. Eens, V.A. Gill, B. Kempenaers, & S.A. Hatch. In prep(a). Effects of testosterone on sibling aggression in black-legged kittiwake chicks under food-rich and food-poor conditions. Journal of Animal Ecology.

Lanctot, R.B., V.A. Gill, S.A. Hatch, M. Eens, V. Friesen, & S. Talbot. In prep(b). Effects of food provisioning on offspring sex ratio in black-legged kittiwakes. Proceedings of the Royal Society of London B.

Marra, P.P and R.L. Holberton. 1998. Corticosterone levels as indicators of habitat quality: effects of habitat segregation in a migratory bird during the non-breeding season. Oecologia 116: 284-292.

Merrick, R.L., T.R. Loughlin, and D.G. Calkins. 1987. Decline in abundance of the northern sea lion, Eumetopias jubatus, in Alaska, 1956-86. Fishery Bulletin 85: 351-365.

Monaghan, P. 1996. Relevance of the behavior of seabirds to the conservation of marine environments. Oikos 77:227-237.

Murphy, E.C., A.M. Springer, and D.G. Roseneau. 1991. High annual variability in reproductive success of kittiwakes (*Rissa tridactyla*) at a colony in western Alaska. Journal of Animal Ecology 60: 515-534.

Neuman, J., J.W. Chardine, and J.M. Porter. 1998. Courtship feeding and reproductive success in black-legged kittiwakes. Colonial Waterbirds 21:73-80.

Newman, S.H. and J.G. Zinkl. 2000. Examining avian stress parameters associated with rehabilitation methodologies: Part I & II. Final Report. Oiled Wildlife Care Network, Wildlife Health Center, School of Veterinary Medicine, University of California, Davis, CA.

Piatt J.F., and P.J. Anderson. 1996. Response of Common Murres to the Exxon Valdez oil spill and long-term changes on the Gulf of Alaska marine ecosystem. Pp. 720-737 in: Exxon Valdez Oil Spill Symposium Proceedings. Rice, S.D., R.B. Spies, D.A. Wolfe and B.A. Wright (Eds.). American Fisheries Society Symposium 18, Bethesda, Maryland.

Pitcher, K.W. 1990. Major decline in number of harbor seals, Phoca vitulina richardsi, on Tugidak Island, Gulf of Alaska. Mar. Mamm. Sci. 6: 121-134.

Roberts, B.D. and S.A. Hatch. 1993. Behavioral ecology of black-legged kittiwakes during chick rearing in a failing colony. Condor 95: 330-342.

Sapolsky, H. 1987. Stress, social status and reproductive physiology in free-living baboons. Pp. 291-322 in: Psychobiology of reproductive behaviour: an evolutionary perspective. Crews, D. (Ed.). Prentice Hall, Englewood Cliffs, New Jersey.

Silvern, B. 1986. Corticosterone-binding proteins and behavioral effects of high plasma levels of corticosterone during the breeding period in the pied flycatcher. General Comparative Endocrinology 64:67-74.

Silverin, B. 1998. Stress response in birds. Poultry and Avian Biology Reviews 9:153-168.

United States Fish and Wildlife Service (USFWS). 1992. Alaska Seabird Management Plan. Unpublished report by U.S. Fish and Wildlife Service, 102 pp.

Wingfield, J.C., C.M. Vleck, and M.C. Moore. 1992. Seasonal changes of the adrenocortical response to stress in birds of the Sonoran Desert. Journal of Experimental Zoology 264:419-428.

Wingfield, J.C. 1994. Modulation of the adrenocortical response to stress in birds. Pp. 520-528 in: Perspectives in Comparative Endocrinology. Davey, K.G., R.E. Peter, S.S. Tobe, (Eds.). National Research Council of Canada, Ottawa.

Wingfield, J.C., R. Suydam, and K. Hunt. 1994. The adrenocortical responses to stress in snow buntings (*Plectrophenax nivalis*) and lapland longspurs (*Calcarius lapponicus*) at Barrow, Alaska. Comp. Biochem. Physiol. 108C:299-306.

Wingfield, J.C., K. Hunt, C. Bruener, K. Dunlap0, G.S. Fowler, L. Freed, and J. Lapson. 1997. Environmental stress, field endocrinology, and conservation biology. Pp. 95-

131 in: Behavioral Approaches to Conservation in the Wild. Clemmons, J.R. and R. Buchhols (Eds.). Cambridge University Press, Cambridge.

Wingfield, J.C., G. Ramos-Fernandez, A. Nunez-de la Mora, and H. Drummond. 1999. The effects of an El Niño southern oscillation event on reproduction of male and female blue-footed boobies, *Sula nebouxii*. General Comparative Endocrinology 114:163-172.

Table 1. Corticosterone levels and breeding parameters in black-legged kittiwakes (BLKI) in relation to supplemental feeding treatment, Middleton Island 1999. See methods for definition of parameters. Values are listed as mean ± standard error where applicable, and samples sizes are shown in parentheses below each variable.

Breeding stage	Parameter	Fed BLKI		Unfed BLKI
Pre-egg laying	Corticosterone (ng/ml)	4.42±1.11 (21)	NS	4.98±0.99 (20)
	Laying success	92% (48)	P < 0.001	37% (178)
	Laying date	10 June ± 1.0 (44)	P < 0.001	18 June ± 0.6 (65)
	Clutch size	1.68±0.08 (44)	P < 0.001	1.14±0.04 (65)
Incubation	Corticosterone (ng/ml)	4.07±0.47 (21)	NS	5.36±0.87 (22)
	Hatching success	43% (76)	NS	56% (75)
	Shift length (hrs)	4.26 (29)	P < 0.001	11.95 (27)
	Egg volume (cc)	46.33±0.40 (73)	P = 0.033	45.00±0.50 (74)
Chick-rearing	Corticosterone (ng/ml)	1.75±0.14 (20)	P < 0.001	4.67±0.63 (22)
	Fledging success	82% (33)	NS	79% (42)
	A-Chick growth (g/day)	16.37±0.34 (22)	P = 0.004	14.87±0.34 (30)
	Adult attendance (% of both adults)	56.15±0.99 (21)	P < 0.001	41.83±0.81 (26)

2001 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 2000 - September 30, 2001

Budget Category:	Authorized FY 2000	Proposed FY 2001
Personnel		$12,000.0
Travel		$1,500.0
Contractual		$2,100.0
Commodities		$1,400.0
Equipment		$0.0
Subtotal	$0.0	$17,000.0
General Administration		$1,854.5
Project Total	$0.0	$18,854.5
Full-time Equivalents (FTE)		0.2
Other Resources	$57,800.0	$57,800.0

LONG RANGE FUNDING REQUIREMENTS					
			Estimated FY 2002		
			$0.0		

Dollar amounts are shown in thousands of dollars.

Comments: The Alaska Biological Science Center, Biological Resources Division, DOI-USGS, is responsible for coordinating all aspects of this project. Responsibilities include field logistics, collection of data and blood samples, hormone and data analyses, report writing, and presentation and publication of results. One other non-trustee ageny, the University of Antwerp in Belgium, will coordinate hormone analyses.

We anticipate no expenses for NEPA compliance (assuming a categorical exclusion), annual restoration workshop attendance, or community involvement. $1,700 has been budgeted to attend the Pacific Seabird Group Meeting in Hawaii, and $1,000 has been budgeted for manuscript preparation and publication costs.

Accounting, personnel hiring, purchasing, secretarial assisstance, computer equipment, and other office supplies will be provided by DOI-USGS. Other costs associated with project will be provided by DOI-USGS. These include $13,300 for air charters to Middleton Island, $2,800 for fish for feeding experiment, $1,600 for tower expansion for pelagic cormorant work, $200 for blood sampling supplies, $4,000 for fish regurgitation identification, $3,300 for four volunteers (travel and per diem), $5,100 for food for volunteers and field leaders, $24,500 for salary (1 month for Scott Hatch, 2 months for Verena

FY01

Prepared:10 April 2000

Project Number:
Project Title: Can stress hormones be used as an indication of food availability and reproductive performance? An experimental approach.
Agency: DOI-USGS

2001 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 2000 - September 30, 2001

Personnel Costs:		GS/Range/ Step	Months Budgeted	Monthly Costs	Overtime	
Name	Position Description					
Dr. Richard Lanctot	Principal Investigator - manages project, analyzes data, presents information at meetings, and writes manuscripts	GS/11/2	2.5	4800.0		
	Subtotal		2.5	4800.0	0.0	
					Personnel Total	

Travel Costs:	Ticket Price	Round Trips	Total Days	Daily Per Diem	
Description					
Pacific Seabird Group Meeting in Hawaii	600.0	1			
Housing			5	110.0	
Food			5	50.0	
				Travel Total	

FY01

Prepared: 10 April 2000

Project Number:
Project Title: Can stress hormones be used as an indication of food availability and reproductive performance? An experimental approach.
Agency: DOI-USGS

2001 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET
October 1, 2000 - September 30, 2001

Contractual Costs:	
Description	
4A Linkage	
When a non-trustee organization is used, the form 4A is required. **Contractual Total**	
Commodities Costs:	
Description	
Manuscript preparation and page charges Miscellaneous (federal express of plasma samples and other packages) Registration cost for Pacific Seabird Group Meeting	
Commodities Total	

FY01

Prepared: 10 April 2000

Project Number:
Project Title: Can stress hormones be used as an indication of food availability and reproductive performance? An experimental approach.
Agency: DOI - USGS

2001 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 2000 - September 30, 2001

New Equipment Purchases:	Number	Unit	
Description	of Units	Price	
None			
Those purchases associated with replacement equipment should be indicated by placement of an R.		**New Equipment Total**	

Existing Equipment Usage:	Number	
Description	of Units	
Camp gear (tents, sleeping bags, cook kits, etc.)	1	
Electric freezers	4	
Propane stove and heaters	2	
Inflatable boat and outboard motor for collecting fish samples at sea	1	
Float coats and basic survival gear	3	
Tower supplies (cleaning solutions, gloves, buckets, electric cords, etc.)	1	
Banding supplies (bands, measuring equipment, etc.)	1	
4-wheel ATVs and trailer	2	
Computers - pentium 1 minimum	2	
Office supplies, copying, telephone, fax, postage, electronic mail	1	

FY01

Prepared: 10 April 2000

Project Number:
Project Title: Can stress hormones be used as an indication of food availability and reproductive performance? An experimental approach.
Agency: DOI-USGS

2001 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 2000 - September 30, 2001

Budget Category:	Authorized FY 2000	Proposed FY 2001						
Personnel		$0.0						
Travel		$0.0						
Contractual		$0.0						
Commodities		$2,100.0						
Equipment		$0.0	LONG RANGE FUNDING REQUIREMENTS					
Subtotal	$0.0	$2,100.0				Estimated FY 2002		
Indirect								
Project Total	$0.0	$2,100.0				$0.0		
Full-time Equivalents (FTE)		0.0						
	Dollar amounts are shown in thousands of dollars.							
Other Resources	$3,800.0	$3,800.0						

Comments: Laboratory equipment, office supplies, computer equipment, and secretarial assistance will be provided through the University of Antwerp and the University of Leuven in Belgium.

Corticosterone analyses will be conducted at the Laboratory of Immuno-Neuroendocrinbology at the University of Leuven in Belgium, under the direction of Dr. Marcel Eens who resides at the University of Antwerp.

Salary support of $3,800 was provided in FY2000 and will be provided in FY2001 by a grant from the Fund for Scientific Research-Flanders in Belgium to Dr. Marcel Eens. This consists of $1,800 for a laboratory assistant and $2000 for Dr. Eens salary.

No money is requested for report writing, NEPA compliance, annual restoration workshop attendance, community involvement, or indirect costs.

Program management will be conducted through the DOI-USGS lead agency.

FY01

Prepared: 10 April 2000

Project Number:
Project Title: Can stress hormones be used as an indication of food availability and reproductive performance? An experimental approach.
Name: University of Antwerp, Belgium

2001 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET

October 1, 2000 - September 30, 2001

Personnel Costs:				Months	Monthly		
	Name	Position Description		Budgeted	Costs	Overtime	
	None						
			Subtotal	0.0	0.0	0.0	
						Personnel Total	

Travel Costs:		Ticket	Round	Total	Daily	
	Description	Price	Trips	Days	Per Diem	
	None					
					Travel Total	

FY01

Prepared: 10 April 2000

Project Number:
Project Title: Can stress hormones be used as an indication of food availability and reproductive performance? An experimental approach.
Name: University of Antwerp, Belgium

2001 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET
October 1, 2000 - September 30, 2001

Contractual Costs:	
Description	
None	
Contractual Total	
Commodities Costs:	
Description	
Laboratory costs for corticosterone analyses ($11.50 x 180 samples)	
Commodities Total	

FY01

Prepared: 10 April 2000

Project Number:
Project Title: Can stress hormones be used as an indication of food availability and reproductive performance? An experimental approach.
Name: University of Antwerp, Belgium

2001 EXXON VALDEZ TRUSTEE COUNCIL PROJECT BUDGET
October 1, 2000 - September 30, 2001

New Equipment Purchases:	Number of Units	Unit Price	
Description			
none			
Those purchases associated with replacement equipment should be indicated by placement of an R.		**New Equipment Total**	

Existing Equipment Usage:	Number of Units	
Description		
Laboratory space and equipment to conduct corticosterone analyses	1	
Office supplies, copying, telephone, fax, postage, electronic mail	1	

FY01

Prepared: 10 April 2000

Project Number:
Project Title: Can stress hormones be used as an indication of food availability and reproductive performance? An experimental approach.
Name: University of Antwerp, Belgium