TASK PROPOSAL FOR FISCAL YEAR 1992
NMFS MARINE ENTANGLEMENT RESEARCH PROGRAM

TASK TITLE: Effect of Persistent Marine Debris on Juvenile, Pelagic Sea Turtles

TASK NUMBER: 3d

ESTIMATED COST: $40.5K

OVERVIEW: Our proposed research program has three parts. First, we will initiate a new direction of research--based on our results from the last two years--to quantify the sub-lethal effects of ingestion of debris on rate of nutrient uptake in sea turtles. We will elucidate the levels of debris ingestion (or nutrient dilution) necessary to significantly affect growth and productivity.

Second, we will continue to quantify the effect of marine debris on the pelagic population of loggerheads in the eastern Atlantic. Continued studies of their diet, growth and movements are necessary to determine the extent and length of time to which this life stage is exposed to marine debris.

Third, through our sea turtle observation network, we will continue to collect records of sightings of sea turtles at sea. These records will be compiled to plot the distribution of sea turtle populations in pelagic habitats and to determine the extent to which, on a worldwide basis, entanglement in marine debris has a negative effect on the sea turtles within these habitats.

BACKGROUND: Our studies of pelagic-stage sea turtles and the interactions with persistent marine debris, which began in 1989, have been funded by the NMFS Marine Entanglement Research Program. Our project was first based on the theory, developed by the late Dr. Archie Carr, that the earliest developmental habitats of sea turtle hatchlings, once they leave the nesting beach, are oceanic driftlines (Carr 1986a, 1986b, 1987). Loggerheads *(Caretta caretta)*, green turtles *(Chelonia mydas)*, hawksbills *(Eretmochelys imbricata)*, and ridleys *(Lepidochelys kempi)* are often associated with rafts of sargassum in these driftlines that provide both a physical refuge and a food resource. The physical factors responsible for concentrating and establishing these driftline communities are also responsible for concentrating marine debris (primarily plastic flotsam, petroleum products, and discarded fishing gear) in these same driftlines (Galt 1985). Balazs (1985) has summarized the anecdotal accounts of pelagic sea turtles adversely affected by ingestion of or entanglement in this debris.

Part I: Sub-lethal Effects of Ingestion of Debris on Rate of Nutrient Uptake

Ingestion of marine debris can affect marine turtles through a number of mechanisms, which can be divided into three categories: mechanical, toxic, and nutrient dilution. Mechanical effects would include blockage of the gut, coating of absorptive surfaces (especially by tar), abrasion of gut epithelia, and blocking digesta from digestive processing. Toxic effects can act either directly on the turtle following absorption of toxic compounds from the gut or can indirectly affect the nutrition of turtles by having a toxic effect on gut microflora. Nutrient dilution occurs when the ingestion of debris dilutes the natural diet so that the net nutrient gain is significantly reduced.

To gain a better understanding of how debris negatively affects sea turtles, more information is needed for all three types of mechanisms. Studying mechanical and toxic effects is difficult because controlled experiments cannot be undertaken without sacrificing or threatening the lives of endangered sea turtles. However, feeding trials can be designed to study the effects of nutrient dilution on sea turtles using inert diluents so that experimental animals are not at risk.

Our research has demonstrated the extent to which sea turtles ingest marine debris in two populations (Bolten and Bjorndal 1991). Digestive tracts from eight pelagic-stage loggerheads from the Azores were examined, and all contained styrofoam, plastic and tar throughout their length. Of 52 sea turtles stranded in Florida, 50% had ingested debris. Sometimes, such ingestion will result in death from blockage of the digestive tract or severe toxic reaction. However, probably more commonly, ingestion of debris will have sub-lethal effects on the turtle through sub-lethal toxic effects or a decrease in nutrient gain. If an organism's rate of nutrient gain is significantly reduced, decreased growth rates and reproductive productivity will result. The Committee on Sea Turtle Conservation of the National Academy of Sciences stressed the importance of quantifying the effect of marine debris on sea turtle demography (Magnuson et al. 1990). At a recent workshop, it was agreed that determining sub-lethal effects, although difficult to quantify, are essential to understanding the long-term effects of human activities on sea turtle populations (MMS 1991).

In studies with fish (reviewed in Pandian 1987) and mammals (reviewed in Weston and Poppi 1987), it has been shown that animals feed to meet a set energy or protein requirement. If the diet is diluted with indigestible material (e.g., kaolin),

animals will increase their food intake of the dilute diet so that intake of nutrients is held constant. However, there is a point beyond which the animal cannot increase its intake to correct for nutrient dilution ("critical point" in Figure 1); that is, there is a limit to the amount of food the animal can physically process in one day. If the diet is diluted beyond this point, the intake of the animal will remain constant, but the amount of nutrients the animal absorbs each day will decrease (Figure 1). The effect of a given percent of diet dilution will vary with the nutrient density of the original diet. A diet that has a low nutrient density will require less dilution to push the animal past the critical point than would a diet with high nutrient density.

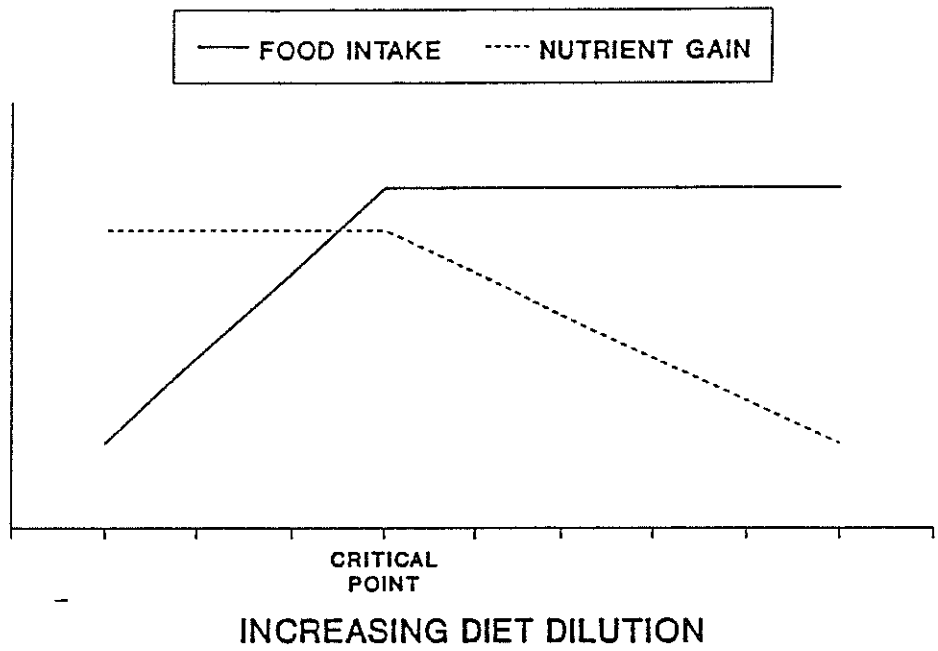

Figure 1. Predicted response in both food intake and nutrient gain in turtles fed increasingly diluted diets. As the diet is diluted, the turtle will increase intake to compensate for the dilution. However, at the "critical point" the turtle can no longer increase intake sufficiently to compensate for dilution. There is a limit to the amount of digesta an animal can physically process. As the diet is diluted beyond this critical point, intake level will remain the same (as high as is physically possible for the turtle). Over the range of dilutions for which the animal can compensate for the dilution by increasing intake, nutrient gain will remain constant. Beyond the critical point, nutrient gain will decrease, and productivity in terms of growth or reproduction will be reduced.

Pelagic-stage loggerheads in the eastern Atlantic may be particularly at risk to such nutrient dilution. Their diet is primarily composed of jellyfish (Bolten, unpubl. data), which has a low nutrient density (Franc et al. 1976; Kimura et al. 1983; Larson 1986). Thus, it may take very little nutrient dilution, or ingestion of debris, to place the turtles beyond the critical point. If this is true, ingestion of only moderate amounts of debris could decrease nutrient uptake, and thus decrease growth rates and productivity of the turtles. Knowledge of the critical point will allow us to assess whether rates of debris ingestion in natural populations are significantly affecting nutrient gain, and in turn, their demography.

We are well-qualified to undertake this nutritional study. We have conducted a number of studies on the nutritional ecology of sea turtles, freshwater turtles and tortoises, both in the field and under laboratory conditions. These studies include both quantification of digestive parameters (Bjorndal 1979, 1986, 1987, 1990; Bjorndal and Bolten 1990; Bjorndal et al. 1991), and experimentation designed to test theories of digestive strategies (Bjorndal 1980, 1982, 1985, 1989, in press; Bjorndal et al. 1990). The effect of nutrient dilution on intake and nutrient gain in sea turtles is of interest as it applies both to the effect of ingestion of marine debris and to the field of theoretical nutritional ecology.

Part II: Effects of Marine Debris on Loggerheads in the Pelagic Habitat of the Eastern Atlantic.

In this part of the project we will continue to focus on the biology of, and the impacts of marine debris on, a pelagic loggerhead population in the Azores Archipelago. Macronesian waters (Azores, Madeira, and Canary Islands) is the only known area in the Atlantic where small, pelagic loggerheads of this size range can be found. Our work in the Azores is conducted in close collaboration with the Department of Oceanography and Fisheries, University of the Azores, Horta (Bolten and Martins 1990; Bolten et al. 1990). Loggerheads in the Azores population range in size from 8.5 to 65 cm straight carapace length. We have already captured, tagged, and released 704 loggerheads in the Azores. This study is the first to document the size distribution of a pelagic population. These loggerheads, which occupy the pelagic habitat, are believed to be a developmental colony of the southeastern U.S. breeding population. The Azores loggerheads represent a size range that is not found in southeastern U.S. waters.

To estimate the extent to which exposure to marine debris in the pelagic zone affects the demography of U.S. loggerheads, we must

know how long they spend in this habitat. The early model developed by Dr. Archie Carr suggested that post-hatchlings spend from one to several years in the pelagic zone until, at approximately 40 cm carapace length, they settle in benthic habitats in the western Atlantic. This early model now appears to be over simplified. From our research, we know that turtles well over 40 cm remain in the pelagic habitat, and that turtles larger than 40 cm will leave the benthic zone in the western Atlantic, re-enter the pelagic habitat and return to the eastern Atlantic (Eckert and Martins 1989). Turtles much larger than those proposed by Archie Carr's theory are making trans-Atlantic crossings. The length of time loggerheads spend in the pelagic habitat is critical information. If we find that large juvenile loggerheads commonly inhabit the pelagic zone, then we must be even more concerned about the effect of marine debris in the pelagic habitat on the demography of loggerheads. In the most thorough analysis of loggerhead demography to date, Crouse et al. (1987) found that the life stage that is the most important to protect to ensure the recovery of the species is the large juveniles.

Duration of the pelagic life stage can be assessed through movements of tagged individuals and through growth rates of turtles, if the size at which they leave the pelagic zone is known. Growth rates can only be measured by recapturing and measuring tagged turtles. Growth rates of captive sea turtles cannot be used to estimate growth rates of free-ranging turtles (Bjorndal 1985). In addition, we are interested in the diet and foraging behavior of sea turtles in the pelagic zone, because how turtles feed will affect the degree to which they are exposed to ingestion of marine debris.

To answer these and other questions, we have established a cooperative sea turtle tagging program with the commercial fishing fleet that is centered in the Azores. This is an ideal commercial fishing fleet with which to collaborate because turtles are not taken incidental to the fishery, but rather are harmlessly captured in dip nets from the surface of the ocean as the vessels cruise the region in search of tuna. In 1990, crews from 30 vessels participated in our program and 298 turtles were caught, tagged, measured, and released. The captain of each boat was given instructions for tagging and measuring the turtles as well as for documenting observations of sea turtles impacted by marine debris. An employee of the Department of Oceanography and Fisheries of the University of the Azores (Horta) has been assigned to this project and has the responsibility of regularly interacting with the cooperating fishing boats to oversee data collection and tagging records.

Our working hypothesis is that pelagic loggerheads are carried by the Gulf Stream Current and north Atlantic gyre system from the nesting beaches in the southeastern U.S. to the Azores and then to Madeira, the Canary Islands, and the Cape Verde Islands before being returned to the western Atlantic. We are expanding tagging projects to areas outside of the Azores to increase the probability of recaptures of tagged turtles and to study movements and growth of turtles. During meetings in March in the Azores, one of us (AB) met with representatives from the Ministry of Fisheries from Madeira and the Cape Verde Islands. They were very interested in establishing a collaborative program with us, similar to the one we have with the tuna fishing fleet in the Azores. We already have established a collaboration in the Canary Islands.

Part III: International Network for At-sea Observations of Sea Turtles and Impact of Marine Debris

The primary objectives are to define the sea turtle populations (species, size distribution, and density) in pelagic habitats, and to determine the extent to which marine debris has a negative effect on the sea turtles within these habitats. The most cost-effective approach is to make use of the many people who see marine turtles at sea. Turtle-sighting report forms have been developed, translated into Spanish and Portuguese, and sent to government agencies, research institutions and individuals. This approach has been very successful. There are now over 800 individuals and institutions in our network, and the network has provided valuable data. Over 60% of all observations are of turtles less than 50 cm in length, thus the network is locating small, pelagic turtles about which we had little previous information. Of 1119 observations of sea turtles at sea, 4.3% were reported to be negatively affected by marine debris, based on external evidence. Now that we have the basis of a productive network, it is cost-effective to continue the work.

SCOPE OF WORK:

Researchers will have all necessary permits to allow them to conduct these studies.

Part I: Sub-lethal Effects of Ingestion of Debris on Rate of Nutrient Uptake

An artificial diet will be developed that has the nutrient composition (energy, protein, dry matter, organic matter and mineral content, as well as amino acid balance) of jellyfish in the diet of loggerheads. The diet will be cubes jelled with

either agar or gelatin so that the diets can be easily diluted in a stepwise fashion with an inert, indigestible substance. Dilution of the experimental diet will model the dilution of a natural diet with marine debris.

Feeding trials will be conducted with diets of different nutrient dilutions. For each diet dilution, food intake and digestibility will be measured, and nutrient gain will be calculated as the product of intake and digestibility. From these trials, the "critical point" (Figure 1) will be determined. This critical point will represent the minimum proportion of debris that, if ingested by loggerheads, will reduce nutrient gain in the turtles.

To ensure that there will be no risk to loggerheads during the feeding trials, we will run an initial series of trials with the freshwater turtle *Trachemys scripta scripta*, a common species with which we have worked for several years. Once the protocol for the trials has been developed, we will conduct another series of trials with post-hatchling loggerheads. One would predict that the "critical point" will vary among species, among size classes within a species and among diets.

Once the critical point for post-hatchling loggerheads has been determined, groups of 10 turtles will be maintained on diets at various levels of dilution, both above and below the critical point. Growth will be monitored and compared among the groups to test the effect of lowered nutrient gain on growth rate. Blood parameters (i.e., chemical, morphological and physiological) will be compared among the treatment groups to identify those parameters that can be used to evaluate natural populations for the extent of ingestion of marine debris.

Part II: Effects of Marine Debris on Loggerheads in the Pelagic Habitat of the Eastern Atlantic

Because of the tremendous success of the cooperative tagging program with the commercial tuna fleet in the Azores, we plan to continue this aspect of the project. In addition, we will expand this collaborative tagging effort to Madeira, Canary Islands and Cape Verde Islands.

During our 6 to 8 week field season in the Azores, we will continue to focus our efforts on determining the adverse effects of marine debris on juvenile turtles in pelagic communities by conducting the following work.

1. The presence and density of sea turtles will be confirmed by visual observations. Turtles will be caught by dip nets. The turtles will be identified, measured, weighed, and inspected for

marine debris, epibiotics and evidence of predation attempts.

2. The density and diversity of marine debris will be described for the vicinity (within 100 meter radius) for each sea turtle observed. For statistical comparisons, control areas of the same size will be surveyed for marine debris in areas without turtles. Debris will be characterized by the potential for a negative impact on sea turtles either through entanglement or ingestion.

3. Blood samples will be collected from the dorsal cervical sinus using a non-injurious technique. Blood profiles (chemical and morphological) from these turtles will be compared with those from other populations (Lutz and Dunbar-Cooper 1987; Bolten and Bjorndal, unpubl. data) and with results from the feeding trials described above in Part I. Samples will also be used for DNA analyses to test the hypothesis that the Azores loggerheads are a juvenile population of the southeastern U.S. loggerhead nesting population.

4. Stomachs will be gently flushed to determine diet and extent of ingestion of marine debris. We will experiment with endoscopy to determine whether this may be a more successful technique for examining stomach contents. Some turtles will be held in small buckets to collect feces to determine presence of marine debris.

5. A necessary part of the research on the diet of sea turtles will be the identification of the fauna (particularly the invertebrates) and flora of the pelagic community. Voucher specimens of all organisms will be collected for identification by appropriate taxonomists. Diet items (based on #4 above) will be collected and dried for nutrient analyses.

6. Small tags with an identification number and return address will be applied to the front flippers of all turtles captured. It is hoped that future tag recoveries will elucidate the growth rates of juvenile turtles and their movements both within and out of the oceanic gyre systems.

7. Before release, any turtles observed to be affected by debris will be freed from the debris to the extent possible.

Part III: International Network for At-sea Observations of Sea Turtles and Impact of Marine Debris

We will continue to expand our network of collaborators and to enter the turtle sightings in our database for analysis. The network approach requires time to develop an effective group of collaborators and a meaningful database from reports of anecdotal sightings. Results from the compilation of these reports will enable us to develop distribution maps for pelagic sea turtles.

These maps, when combined with data on ocean currents will enable us to evaluate movements of sea turtles in the pelagic habitat. In addition, knowledge of the distribution of sea turtles in the pelagic habitat will enable us to predict potential adverse effects from known distributions and sources of pollutants.

PRINCIPAL INVESTIGATORS

Dr. Karen Bjorndal
Center for Sea Turtle Research and Department of Zoology and Range Sciences
University of Florida
Gainesville, FL 32611

Dr. Alan Bolten
Center for Sea Turtle Research and Department of Wildlife
University of Florida
Gainesville, FL 32611

Telephone: (904) 392-5194
FAX: (904) 392-9166

VI. LITERATURE CITED

Balazs, G.H. 1985. Impact of ocean debris on marine turtles: entanglement and ingestion. *In* R.S. Shomura and H.O. Yoshida (editors). Proceedings of the Workshop on the Fate and Impact of Marine Debris, 27-29 November 1984, Honolulu, Hawaii, pp 387-429. U.S. Dept. Commerce, NOAA Technical Memorandum NOAA-TM-NMFS-SWFS-54.

Bjorndal, K.A. 1979. Cellulose digestion and volatile fatty acid production in the green turtle, *Chelonia mydas*. Comparative Biochemistry and Physiology 63A:127-133.

Bjorndal, K.A. 1980. Nutrition and grazing behavior of the green turtle, *Chelonia mydas*. Marine Biology 56:147-154.

Bjorndal, K.A. 1982. The consequences of herbivory for the life history pattern of the Caribbean green turtle. *In* K.A. Bjorndal (editor). Biology and conservation of sea turtles, pages 111-116. Smithsonian Institution Press, Washington, D.C.

Bjorndal, K.A. 1985. Nutritional ecology of sea turtles. Copeia 1985:736-751.

Bjorndal, K.A. 1986. Effect of solitary versus group feeding on intake in *Pseudemys nelsoni*. Copeia 1986:234-235.

Bjorndal, K.A. 1987. Digestive efficiency in a temperate herbivorous reptile *Gopherus polyphemus*. Copeia 1987:714-720.

Bjorndal, K.A. 1989. Flexibility of digestive responses in two generalist herbivores, the tortoises *Geochelone carbonaria* and *Geochelone denticulata*. Oecologia 78:317-321.

Bjorndal, K.A. 1990. Digestibility of the sponge *Chondrilla nucula* in the green turtle, *Chelonia mydas*. Bulletin of Marine Science 47:567-570.

Bjorndal, K.A. In press. Diet mixing: non-additive interactions of diet items in an omnivorous freshwater turtle. Ecology.

Bjorndal, K.A. and A.B. Bolten. 1990. Digestive processing in a herbivorous freshwater turtle: consequences of small intestine fermentation. Physiological Zoology 63:1232-1247.

Bjorndal, K.A., A.B. Bolten and J.E. Moore. 1990. Digestive fermentation in herbivores: effect of food particle size. Physiological Zoology 63:710-721.

Bjorndal, K.A., H. Suganuma and A.B. Bolten. 1991. Digestive fermentation in green turtles, *Chelonia mydas*, feeding on algae. Bulletin of Marine Science 48:166-171.

Bolten, A.B. and K.A. Bjorndal. 1991. Effect of marine debris on juvenile, pelagic sea turtles. Interim Project Report to the National Marine Fisheries Service Marine Entanglement Research Program. 53 pages.

Bolten, A.B. and H.R. Martins. 1990. Kemp's ridley captured in the Azores. Marine Turtle Newsletter 48:23.

Bolten, A.B., H.R. Martins, M.L. Natali, J.C. Thome and M.A. Marcovaldi. 1990. Loggerhead released in Brazil recaptured in Azores. Marine Turtle Newsletter 48:24-25.

Carr, A. 1986a. Rips, FADS, and little loggerheads. BioScience 36:92-100.

Carr, A. 1986b. New perspectives on the pelagic stage of sea turtle development. NOAA Technical Memorandum NMFS-SEFC-190.

Carr, A. 1987. The impact of non-degradable marine debris on the ecology and survival outlook of sea turtles. Marine Pollution Bulletin 18:352-356.

Crouse, D.T., L.B. Crowder and H. Caswell. 1987. A stage-based population model for loggerhead sea turtles and implications for conservation. Ecology 68:1412-1423.

Eckert, S.A. and H.R. Martins. 1989. Transatlantic travel by juvenile loggerhead turtle. Marine Turtle Newsletter 45:15.

Franc, S., J.M. Franc and R. Garrone. 1976. Fine structure and cellular origin of collagenous matrices in primitive animals: Porifera, Cnidaria and Ctenophora. Front. Matrix Biol. 3:143-156.

Galt, J.A. 1985. Oceanographic factors affecting the predictability of drifting objects at sea. *In* R.S. Shomura and H.O. Yoshida (editors). Proceedings of the Workshop on the Fate and Impact of Marine Debris, 27-29 November 1984, Honolulu, Hawaii, pp 497-518. U.S. Dept. Commerce, NOAA Technical Memorandum NOAA-TM-NMFS-SWFS-54.

Kimura, S., S. Miura and Y.H. Park. 1983. Collagen as the major edible component of jellyfish (*Stomolophus nomural*). Journal of Food Science 48:1758-1760.

Larson, R.J. 1986. Water content, organic content and carbon and nitrogen composition of medusae from the Northeast Pacific. J. Exp. Mar. Biol. Ecol. 99:107-120.

Lutz, P.L. and A. Dunbar-Cooper. 1987. Variations in the blood chemistry of the loggerhead sea turtle, *Caretta caretta*. Fishery Bulletin 85:37-43.

Magnuson, J.J., K.A. Bjorndal, W.D. DuPaul, G.L. Graham, D.W. Owens, C.H. Peterson, P.C.H. Pritchard, J.I. Richardson, G.E. Saul and C.W. West. 1990. Decline of the sea turtles:

causes and prevention. National Academy Press, Washington, D.C.

MMS. 1991. Sea turtles and marine mammals of the Gulf of Mexico. Proceedings of a workshop held in New Orleans, August 1-3, 1989. Minerals Management Service, New Orleans, Louisiana. 211 pages.

Pandian, T.J. 1987. Fish. *In* T.J. Pandian and F.J. Vernberg (editors). Animal energetics, Vol. 2 Bivalvia through Reptilia, pages 357-465. Academic Press, San Diego.

Weston, R.H. and D.P. Poppi. 1987. Comparative aspects of food intake. *In* J.B. Hacker and J.H. Ternouth (editors). The nutrition of herbivores, pages 133-161. Academic Press, Sydney.

VII. REPORTING REQUIREMENTS

Reports	Date Due
Task Report 1	31 July 1992
Task Report 2	31 October 1992
Task Report 3	31 January 1993
Annual Report	30 April 1993

VIII. PERIOD OF PERFORMANCE

The period of performance is from 30 April 1992 to 1 May 1993.

IX. COST ESTIMATE

Salary	
Principal Investigators	$ 11,750
Technical Assistants	4,129
Expenses	
Travel	
Two roundtrip airfares Gainesville-Azores-Madeira-Canary Islands	2,500
Per diem (official Azores rate)	4,200
Boat rental	3,700
Supplies	
Tanks, diets and laboratory analyses for feeding trials; turtle tags and applicators; film and processing; dip nets; stomach sampling apparatus; blood analyses; and miscellaneous supplies	5,944
Subtotal	32,223
Overhead	8,277
Total	$ 40,500