

Research and Management Techniques for the Conservation of Sea Turtles

Prepared by IUCN/SSC Marine Turtle Specialist Group

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Development and publication of *Research and Management Techniques for the Conservation of Sea Turtles* was made possible through the generous support of the Center for Marine Conservation, Convention on Migratory Species, U.S. National Marine Fisheries Service, and the Worldwide Fund for Nature.

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ISBN 2-8317-0364-6

Printed by Consolidated Graphic Communications, Blanchard, Pennsylvania USA

Cover art: leatherback hatchling, *Dermochelys coriacea*, by Tom McFarland

This publication should be cited as follows: Eckert, K. L., K. A. Bjorndal, F. A. Abreu-Grobois, and M. Donnelly (Editors). 1999. *Research and Management Techniques for the Conservation of Sea Turtles*. IUCN/SSC Marine Turtle Specialist Group Publication No. 4.

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Preface

In 1995 the IUCN/SSC Marine Turtle Specialist Group (MTSG) published *A Global Strategy for the Conservation of Marine Turtles* to provide a blueprint for efforts to conserve and recover declining and depleted sea turtle populations around the world. As unique components of complex ecosystems, sea turtles serve important roles in coastal and marine habitats by contributing to the health and maintenance of coral reefs, seagrass meadows, estuaries, and sandy beaches. The *Strategy* supports integrated and focused programs to prevent the extinction of these species and promotes the restoration and survival of healthy sea turtle populations that fulfill their ecological roles.

Sea turtles and humans have been linked for as long as people have settled the coasts and plied the oceans. Coastal communities have depended upon sea turtles and their eggs for protein and other products for countless generations and, in many areas, continue to do so today. However, increased commercialization of sea turtle products over the course of the 20th century has decimated many populations. Because sea turtles have complex life cycles during which individuals move among many habitats and travel across ocean basins, conservation requires a cooperative, international approach to management planning that recognizes inter-connections among habitats, sea turtle populations, and human populations, while applying the best available scientific knowledge.

To date our success in achieving both of these tasks has been minimal. Sea turtle species are recognized as “Critically Endangered,” “Endangered” or “Vulnerable” by the World Conservation Union (IUCN). Most populations are depleted as a result of unsustainable harvest for meat, shell, oil, skins, and eggs. Tens of thousands of turtles die every year after

being accidentally captured in active or abandoned fishing gear. Oil spills, chemical waste, persistent plastic and other debris, high density coastal development, and an increase in ocean-based tourism have damaged or eliminated important nesting beaches and feeding areas.

To ensure the survival of sea turtles, it is important that standard and appropriate guidelines and criteria be employed by field workers in all range states. Standardized conservation and management techniques encourage the collection of comparable data and enable the sharing of results among nations and regions. This manual seeks to address the need for standard guidelines and criteria, while at the same time acknowledging a growing constituency of field workers and policy-makers seeking guidance with regard to when and why to invoke one management option over another, how to effectively implement the chosen option, and how to evaluate success.

The IUCN Marine Turtle Specialist Group believes that proper management cannot occur in the absence of supporting and high quality research, and that scientific research should focus, whenever possible, on critical conservation issues. We intend for this manual to serve a global audience involved in the protection and management of sea turtle resources. Recognizing that the most successful sea turtle protection and management programs combine traditional census techniques with computerized databases, genetic analyses and satellite-based telemetry techniques that practitioners a generation ago could only dream about, we dedicate this manual to the resource managers of the 21st century who will be facing increasingly complex resource management challenges, and for whom we hope this manual will provide both training and counsel.

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Forensic Aspects

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Forensic Science

Forensic science has been defined as the “application of the natural and physical sciences to the resolution of matters within a legal context” (Thornton, 1994). Therefore, forensic science is concerned with the crime and the suspects, followed by investigation and comparative/analytical analysis of the evidence, and can culminate in testimony regarding an interpretation of what the analyses mean in the context of the crime.

The informal use of the term “forensics” is popular among the wildlife research community today; much of this research is typically for information use only and never intended to reach a court of law. Because forensic tests can have significant impact on the outcome of a trial or public decision process, the informal use of the term forensics should be avoided. Attempting to infer support for the legal process while not instituting formal forensic investigative procedures is likely to compromise the admissibility of the analysis as evidence in a court of law and prevent successful prosecution of law enforcement cases. It is imperative that researchers who agree to conduct biochemical identifications of wildlife evidence for legal proceedings be fully aware of the unique responsibilities that forensic scientists have when supporting law enforcement and the law.

When is Forensic Science Used for Sea Turtles?

In the United States, forensic techniques for species identification have been used in conservation efforts concerning sea turtles since the late 1970s by the marine Forensics Program at the Charleston Laboratory of the National Marine Fisheries Service. In 1997, the laboratory, with its Marine Forensics Pro-

gram, came under the jurisdiction of NOAA’s National Ocean Service (NOS). Though the majority of species identification analyses have been conducted in support of law enforcement activities for the Convention on International Trade in Endangered Species of Wild Fauna and Flora (CITES) or other violations of the U.S. Endangered Species Act, management and research questions can be addressed using the same technology. An example of a legal case involved the seizure of a chunk of red meat claimed to be venison (deer, *Odocoileus* sp.) during the boarding of a shrimp trawler. The meat was forensically analyzed using isoelectric focusing and positively identified as loggerhead sea turtle, *Caretta caretta*, (Colbert, 1993).

What Kind of Samples Can Be Used as Evidence?

The most common types of evidence from suspected illegal take or trade in sea turtles or turtle products are meat, eggs, shells, or cosmetics containing turtle oils (see Table 1). Unusual or difficult circumstances should not rule out forensic analysis, and discussion with marine forensic scientists can often lead to new approaches in evidence analyses. New questions in sea turtle conservation continually arise. The Marine Forensics Program attempts to initiate methods development and collaborative research to meet new needs. Such activities frequently require collection of new standard samples from some or all sea turtle species for comparison to evidence and for database development. If capabilities are not currently available to address a sea turtle forensic issue, efforts are made to locate researchers that may be able to provide technical support. In such cases, the Marine Forensics Program may refer an inquiry to another researcher, or may offer to act as liaison for the issue so that the evi-

Table 1. Collection and Analysis Methods for Sea Turtle Body Parts

Evidence Type	Collection/Preservation	Analysis/ Methodology	Limitations of Methodology
<i>Edible Quality Raw Meat</i>	Chain of Custody and Documentation Place Tissue in plastic bag, ice immediately upon collection, then freeze as soon as possible *For DNA Analysis, meat may also be dried, salted or placed in Ethyl Alcohol, If refrigeration is not available	Isoelectric Focusing (IEF) DNA – RFLP or Sequencing	<ul style="list-style-type: none"> • Availability of standards from certain geographical locations • Availability of standards • Database incomplete
<i>Eggs – Raw</i>	Chain of Custody and Documentation Place Eggs in plastic bags, ice, refrigerate or freeze *If above not immediately available, eggs may be kept in damp sand for up to 48 hours	Lipid Analysis DNA RFLP or Sequencing	<ul style="list-style-type: none"> • Loggerhead, Kemp’s and Olive Ridley are indistinguishable from one another • Standards lacking from certain geographical locations • Availability of standards • Database incomplete
<i>Eggs – Cooked</i>	Chain of Custody and Documentation Place Eggs in plastic container and refrigerate or freeze	Lipid Analysis DNA – RFLP or Sequencing	<ul style="list-style-type: none"> • Same as for raw eggs • Method under development • Availability of standards across geographic range • Characterization of additional markers needed
<i>Blood (Taken from Animal)</i>	Chain of Custody and Documentation Refrigerate up to 48 hours Freeze at -20°C – -80°C Dry on cotton cloth, cotton swab or filter paper Place in Lysis solution (provided by forensic lab)	DNA – RFLP or Sequencing	<ul style="list-style-type: none"> • Availability of standards across geographic range • Characteristics of additional markers needed
<i>Blood Stains</i>	Chain of Custody and Documentation Dry, bag material containing stain Scrape dried blood into plastic bag or tube, keep dry	DNA – RFLP or Sequencing	<ul style="list-style-type: none"> • Availability of standards across geographic range • Characteristics of additional markers needed
<i>Other Internal Tissues</i>	Chain of Custody and Documentation Only small piece of tissue required, ~.2-2cm ³ Refrigerate or freeze Place in salt to dry tissue Place in Ethyl Alcohol	DNA Analysis	<ul style="list-style-type: none"> • Availability of standards • Database incomplete
<i>Carapace – Fresh with Tissue Attached</i>	Chain of Custody and Documentation Refrigerate See above for additional tissue preservation for DNA analysis	Visual/Morphological Analysis IEF DNA Analysis	<ul style="list-style-type: none"> • Subjective, based on expert interpretation • Tissue must not be decomposed • Availability of standards • Database incomplete
<i>Carapace – Dried, No Attached Flesh</i>	Chain of Custody and Documentation Bag material and document with chain of custody	Visual/Morphological Analysis	<ul style="list-style-type: none"> • Subjective, based on expert interpretation
<i>Skeletal Remains</i>	Chain of Custody and Documentation Bag material and document with chain of custody Keep dry	Visual/Morphological Analysis DNA Analysis	<ul style="list-style-type: none"> • Subjective, based on expert interpretation • Availability of standards • Database incomplete
<i>Dried Tissue</i>	Chain of Custody and Documentation Bag material. Keep dry.	DNA Analysis	<ul style="list-style-type: none"> • Availability of standards • Database incomplete
<i>Cooked Meats</i>	Chain of Custody and Documentation Bag material. Refrigerate up to 1 week and/or freeze	DNA Analysis	<ul style="list-style-type: none"> • Method under development • Availability of standards • Database incomplete
<i>Canned Meats</i>	Chain of Custody and Documentation Bag material AS IS	DNA Analysis	<ul style="list-style-type: none"> • Method under development • Availability of standards • Database incomplete
<i>Oils and Cosmetics</i>	Chain of Custody and Documentation Bag material AS IS, Protect from air and sunlight	Lipid Analysis	<ul style="list-style-type: none"> • Results may be confounded by additives • Concentration of Turtle Oil in cosmetics may be below level of assay detectability
<i>Jewelry/Skin Products/Dried Turtle Penis</i>	Chain of Custody and Documentation Bag material AS IS	Visual DNA	<ul style="list-style-type: none"> • Subjective, based on expert interpretation • Extent of processing tissue/skin has undergone

dence is handled in a manner that would most likely be acceptable in court and in the scientific community.

What Kind of Documentation is Needed for a Forensic Analysis?

Permits are the first requirement for handling or possessing sea turtles, or their parts or products. In the field of marine forensics, seized property (evidence) or morphologically unidentifiable samples are compared to special samples from an archive. These special samples, called standards, have been collected from carefully identified whole animals by authorized experts who also provide signed documents verifying the species. Standards and evidence alike are accompanied by a “chain of custody” or a traceable audit trail that originates with the collector and accompanies the sample at all times. Every person in possession of an evidence sample or a standard sample signs and dates the chain of custody when they receive and release the sample.

Chain of custody is maintained by keeping the sample under secure conditions with limited access, shipping or transferring the sample in a secure manner so that tampering is detectable, and using a laboratory facility where chain of custody procedures are followed during analysis. Any person who signs a chain of custody can potentially be called into court to testify about his/her possession of a sample, as can a person who verifies the species of a standard sample. The analyst is the most likely person to be asked to testify regarding forensic activities, and should be able to testify that the procedure was conducted accurately and that no individual could have tampered with the samples during analysis. In addition to chain of custody documentation, the analyst must produce a case report and be able to provide case related notes and other laboratory information, if requested.

How are the evidence samples identified? It is often difficult in the field to definitively determine the species of origin of sea turtle eggs or other tissues when the whole animal is not available. When the species of a sample cannot be conclusively determined by observation with the naked eye, chemical or biochemical analysis can often be used to reliably and definitively identify the species. For example, egg morphology, in conjunction with beach or origin, are sometimes useful in identifying eggs to species. Egg size ranges do, however, overlap between many species, and often more than one species of turtle nests on a particular beach. In such circumstances, chemi-

cal analyses such as lipid chemistry or DNA analyses are usually necessary.

Meat with attached skin or flippers may be identifiable from claw counts and/or morphology of skin and scales, if a sufficiently large piece is available. However, if only a small piece of meat or skin is available, forensic methods such as DNA and protein techniques will be needed to make identifications. The potential now exists, through the application of DNA technologies, to ascertain the parentage of eggs or hybrid individuals resulting from crosses between two species, and to determine the species identity of bone and scute fragments, as well as make identifications from trace evidence such as blood spatters. Details for all of the methods described in this section can be found in the “Charleston Laboratory Marine Forensics Manual,” an internal standard operating protocol. Additional information is available upon request from the NOAA/NOS Charleston Laboratory.

How are Forensic Samples Collected and Stored?

The effectiveness of forensic activities in law enforcement cases, management, and research issues is largely dependent upon the handling of samples prior to analysis. Errors can be made in sample documentation, collection, storage, packaging, and shipping that can diminish the value of or even exclude the samples for forensic analysis. Samples should be separated to prevent the contamination of one sample by another sample. Therefore, when multiple samples are collected, they should be individually bagged and labeled and a chain of custody started at the point the evidence is seized. It is recognized that some researchers and enforcement personnel working in remote areas may not have access to ideal sampling tools or shipping supplies such as dry ice, gel-packs, and Styrofoam containers. Should this situation arise, select an appropriate storage or preservation method from Table 1 until shipping or transport to the laboratory can be arranged. Though most types of samples can be safely frozen and then shipped by over night courier on dry ice or frozen gel-packs, it is preferred than anyone seeking forensic assistance call the NOAA/NOS Charleston Laboratory at (843) 762-8500; Fax: (843) 762-8700; email: marine.forensic@noaa.gov. Ask for Forensics Program personnel and discuss the issue prior to storing or shipping samples.

What is the Transport or Shipping Protocol?

Once notice has been given to the appropriate analyst at the laboratory that a shipment is being sent, the samples may be sent to the scientist at the NOAA/NOS Charleston Laboratory. The phone number of the laboratory, (843) 762-8500, and the correct zip code, 29412, must be used on the shipping label. Use of the wrong zip code can cause delays in delivery and may result in loss of the samples for forensic analysis. The services of the Marine Forensics Program are available to scientists and law enforcement personnel from around the world.

Who conducts forensic analyses? Again using the United States as an example, there are currently two federal wildlife and marine agencies with active forensic programs: the Fish and Wildlife Service (National Fish and Wildlife Forensics Laboratory in Ashland, Oregon) and the National Ocean Service (Marine Forensics Program, Charleston Laboratory in South Carolina). The goal of the U.S. Marine Forensics Program is to provide forensic support in matters of marine resources enforcement, management, utilization, safety, and conservation.

Federal U.S. forensic services have been made available to anyone without charge if the request falls generally within the realm of the agency mandates. However, if the requested sample analyses are not strictly for law enforcement purposes and result in detection of illegal activities, the information must be made available to the proper authorities. The program interacts freely with federal and state wildlife law enforcement agents, fisheries managers, state marine resource agencies and university personnel engaged in marine research. When a request exceeds the program's normal capabilities, arrangements can often be made to accommodate the need on a col-

laborative or contractual basis, or on the basis of some other mutually acceptable arrangement.

Who is Qualified to Conduct Forensic Analyses?

Forensic analyses should be conducted by scientists who are familiar with and practice forensic procedures, who are experienced with the methods used for species identification and understand issues of cross-contamination, and who are well aware that they may be subpoenaed to testify in support of the evidence or opinion they produce. Previous experience or training in expert witness testimony is very beneficial. Such criteria for the conduct of forensic analyses should increase the likelihood that a scientist will be qualified as an "expert" in a court of law.

When compiling a national database, researchers are advised to follow forensic guidelines and to identify the species from which their standard samples are collected. Interested researchers should be aware that species identification by DNA requires an extensive database from numerous turtle individuals within each species category. A sound database, with a thorough statistical examination of data derived from well-documented and securely maintained samples, is likely to withstand legal and scientific scrutiny in law enforcement action regarding the conservation of marine turtles.

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