Cetacean Systematics

Approaches in genetics, morphology, & behavior

Symposium, 28–29 April 2004, La Jolla, CA

CETACEAN SYSTEMATICS:
APPROACHES IN GENETICS, MORPHOLOGY AND BEHAVIOR

28 - 29 April 2004
La Jolla, California

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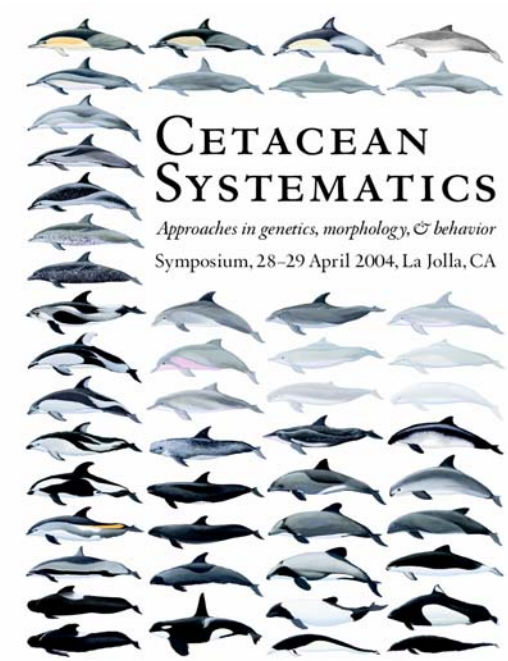

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CETACEAN SYSTEMATICS: APPROACHES IN GENETICS, MORPHOLOGY AND BEHAVIOR

Scripps Institution of Oceanography
28 – 29 April 2004

Wednesday, April 28, 2004

8:00-9:00 Registration – Sumner Auditorium Portico

9:00-9:15 WELCOME AND OPENING REMARKS: William F. Perrin (*Southwest Fisheries Science Center and Scripps Institution of Oceanography*) and Nancy Knowlton (*Scripps Institution of Oceanography and Smithsonian Tropical Research Institute*)

SESSION 1: The status of cetacean taxonomy and the interface with conservation
Session Chair: Phillip Clapham, *Northeast Fisheries Science Center*

9:20- 9:45 How the holes in cetacean taxonomy are affecting conservation and management
*(**Barbara L. Taylor**, Southwest Fisheries Science Center)*

9:45-10:10 A review of cetacean taxonomy at the species and subspecies level
*(**William F. Perrin**, Southwest Fisheries Science Center; Randall R. Reeves)*

10:10-10:40 BREAK

10:40-11:05 Species: hard to define, but we know it when we see it…or do we?
*(**Richard G. Le Duc**, Southwest Fisheries Science Center)*

11:05-11:30 www.DNA-surveillance: implementing a comprehensive and validated molecular taxonomy for cetaceans
*(**C. Scott Baker**, Auckland University, New Zealand)*

SESSION 2: Species-level questions
Session Chair: John Heyning, *Los Angeles County Museum of Natural History*

11:30-11:55 Some observations upon the systematics of Tursiops
*(**James G. Mead**, U.S. National Museum of Natural History, Smithsonian Institution)*

11:55-12:20 Does cranial morphology reflect adaptive evolutionary divergence of sympatric killer whale (orcinus orca) ecotypes?
*(**Charissa W. Fung**, University of British Colombia, Canada)*

LUNCH
12:20-1:45 Sumner Auditorium Portico (bring your own) and environs (Snackcropolis, CUPS)

SESSION 2 (continued)

1:45-2:10 Recognition of a new species *Balaenoptera omurai*
*(**Tadasu Yamada**, National Science Museum, Japan)*

2:10-2:35 Low world-wide genetic diversity in the killer whale (orcinus orca): implications for demographic history
*(**A. Rus Hoelzel**, Durham University, U.K; A. Natoli; M. E. Dahlheim, C. Olavarria, R. W. Baird; N. A. Black)*

2:35-3:00 Use of biomarkers as species-level systematic and forensic molecular tools in the detection of cetacean hybrids
*(**Deborah A. Duffield,** Portland State University; H. Zornetzer; E. O. Espinoza; N. B. Barros)*

3:00-3:25 Systematics of coastal tropical small cetaceans: a review
*(**Thomas A. Jefferson**, Southwest Fisheries Science Center)*

3:25-4:10 BREAK AND POSTER SESSION

4:10-4:35 Large inter-ocean genetic differences within *Kogia sima* indicate long-term isolation and possibly a new species
*(**Susan J. Chivers**, Southwest Fisheries Science Center; R. G. Le Duc; K. M. Robertson; N. B. Barros; A. E. Dizon)*

4:35-5:00 Color patterns of the dwarf minke whale and use of coloration in assessing taxonomic relationships of the baleen whales
*(**Peter W. Arnold**, Museum of Tropical Queensland, Australi; A. Birtles; A. Dunstan; V. Lukoschek; M. Matthews)*

5:00-5:25 *Tursiops* in Atlantic South America: is *Tursiops gephyreus* a valid species?
*(**A. S. Barreto**, Vale do Itajaí University)*

5:25-5:40 Geographic variation of the franciscana, *Pontoporia blainvillei* (Gervais & d'Orbigny, 1844) based on skull morphology
*(**Alessandra Higa**, University of São Paulo, Brazil)*

Thursday, April 29, 2004

8:00-8:30 Registration – Sumner Auditorium Portico

SESSION 3: Approaches in genetics and behavior

Session Chair: Lisa Ballance, *Southwest Fisheries Science Center*

8:30-8:55 Genetic distinctiveness, exclusivity and concordance: a molecular taxonomic approach to species identification and discovery
*(**Merel L. Dalebout**, Auckland University, New Zealand)*

8:55-9:20 New methods for measuring gene flow between closely related populations of species
*(**Jody Hey**, Rutgers University)*

9:20-9:45 Incorrect and unresolved phylogenies from mtDNA: cautions for relying on one genetic marker for species designations
*(**Phillip A. Morin**, Southwest Fisheries Science Center)*

9:45-10:10 Combined versus separate analyses of systematic data sets for cetaceans
*(**John E. Gatesy**, University of California, Riverside)*

10:10-10:40 BREAK

10:40-11:05 A rigorous framework for assessing molecular genetic characters as indicators of species level conservation units
*(**Howard C. Rosenbaum**, American Museum of Natural History, New York; G. Amato; R. DeSalle)*

11:05-11:30 Population forecasting and conservation unit definition in cetaceans
*(**J. McDonald**, University of New South Wales, Australia; W. B. Sherwin; J. Mann; M. Krüetzen; R. Connor)*

11:30-11:55 Why Y? Chromosome–specific markers and their use for cetacean systematics
*(**Leila Hatch**, Cornell University, New York; S. M. Bogdanowicz; R. G. Harrison)*

11:55-12:20 Contrasting patterns of mtDNA diversity and molecular taxonomy of cetaceans
*(**Shane Lavery**, Auckland University, New Zealand; M. Dalebout; H. Ross; C. S. Baker)*

LUNCH

12:20-1:45 Sumner Auditorium Portico (bring your own) and environs (Snackropolis,CUPS)

SESSION 3 (continued)

1:45-2:10 Does genetic differentiation imply speciation? Phylogenetic and paternity analyses show that this is not the case in West Australian bottlenose dolphins (*Tursiops sp.)*
*(**Michael Krüetzen**, University of New South Wales, Australia; W. B. Sherwin)*

2:10-2:35 Listen to my song: Bioacoustical insights into cetacean taxonomy
*(**Sarah L. Mesnick,** Southwest Fisheries Science Center and Scripps Institution of Oceanography; Jay P. Barlow)*

SESSION 4: Taxonomy at various hierarchal levels, and perspectives from other taxa
Session Chair: Annalisa Berta, *San Diego State University*

2:35-3:00 Evolutionary relationships and species identification within Delphininae: a comparison of single-locus sequence data and multi-locus anonymous nuclear markers
*(**Patricia E. Rosel**, Southeast Fisheries Science Center, Charleston, S. C.; S. Kingston; A. Westgate; L. Adams)*

3:00-3:25 Using polymorphic data and frequency analysis to infer the phylogenetic relationships and biogeography of porpoises (Cetacea: Phocoenidae)
*(**Liliana Fajardo**, San Diego State University; A. Berta; R. L. Brownell, Jr.; C. C. Boy)*

3:25-4:10 BREAK AND POSTER SESSION

4:10-4:35 Application of alternative ESU concepts to a common problem in applied conservation biology
*(**Robin Waples**, Northwest Fisheries Science Center, Seattle, Washington)*

4:35-5:00 Adaptive genetic variation and units for conservation
*(**Robert Wayne**, University of California, Los Angeles)*

5:00-5:25 The origins and evolutionary history of modern felid species
*(**Warren E. Johnson**, Laboratory of Genomic Diversity, National Cancer Institute; Eduardo Eizirik, William J. Murphy, Jill Pecon-Slattery, Agostinho Antunes, and Stephen J. O'Brien)*

5:25-5:50 Applications of ESU and subspecies concepts in birds
*(**Robert Fleischer**, U.S. Museum of Natural History, Smithsonian Institution)*

6:30-8:00 **PUBLIC FORUM – BIRCH AQUARIUM**
Are there new species of killer whales in the Antarctic?
(***Robert L. Pitman,*** *Southwest Fisheries Science Center)*

POSTER SESSION TITLES

MOLECULAR SYSTEMATICS OF THE SOUTH AMERICAN COASTAL AND RIVER DOLPHIN *SOTALIA sp*.

Susana Caballero, *Population Genetics and Evolution Research Group, The University of Auckland; C. S. Baker; L.M. González; F. Trujillo; J.A. Vianna; F.R. Santos; H. Barrios; M.G. Montiel; S. Beltrán*

DIFFERENTIATION OF GEOGRAPHIC STOCKS OF ROUGH-TOOTHED DOLPHINS, *STENO BREDANENSIS*, USING MOLECULAR TECHNIQUES

Palma M. Formica and Frank Cipriano, *Conservation Genetics Laboratory, San Francisco State University; H. Markowitz*

A MAXIMUM LIKELIHOOD TEST OF PHYLOGENETIC IDENTIFICATION OF SPECIES

Matthew Goode, *School of Biological Sciences and Institute for Bioinformatics, University of Auckland; H. Ross, S. Lavery, C.S. Baker and A.G. Rodrigo*

MALE GENES AND MALE SONGS: FIN WHALES AS A CASE STUDY FOR INTEGRATING GENETIC AND ACOUSTIC DATA IN DEFINING BALEEN WHALE STOCKS

Leila T. Hatch, *Department of Ecology and Evolutionary Biology, Cornell University; S. M. Bogdanowicz, R.G. Harrison; C.W. Clark*

PHYLOGEOGRAPHY OF THE ATLANTIC SPOTTED DOPLHIN (*STENELLA FRONTALIS*)

M. Martinez-Vergara, *Department of Biology, San Diego State University; A.J. Bohonak; S. Hildebrandt and J.M. Afonso-López*

ENFRANCHISING POPULATIONS UNDER THE ESA: THE LEGAL DEFINITION OF "SPECIES"

Brent Plater, *Staff Attorney, Center for Biological Diversity, Oakland, CA, USA*

COMPOSITE SPECIES-LEVEL CETACEAN PHYLOGENY

Samantha A. Price, *Department of Biology, University of Virginia*

GENETIC STRUCTURE AND DIFFERENTIATION OF *TURSIOPS TRUNCATUS* ECOTYPES IN THE GULF OF CALIFORNIA

Iris Segura García, *CICESE, Biological Oceanography Department;*
A. Rocha-Olivares; L. Rojas Bracho

TRUMPING THE NEED FOR TAXONOMIC RESOLUTION WITH EFFECTIVE CONSERVATION ACTIONS: THE CASE OF GREEN/BLACK SEA TURTLES IN THE EASTERN PACIFIC OCEAN

Jeffrey A. Seminoff, *NOAA – National Marine Fisheries Service, Southwest Fisheries Science Center; W.J. Nichols*

PRESENT CONSERVATION AND MANAGEMENT STATUS OF THE FRANCISCANA (PONTOPORIA BLAINVILLEI) ALONG THE BRAZILIAN COAST AND THE TAXONOMIC STATUS OF ITS DISCONTINOUS POPULATIONS

Salvatore Siciliano, *FIOCRUZ/Escola Nacional de Saúde Pública/Departamento de Endemias/Grupo de Estudos de Mamíferos Marinhos da Região dos Lagos & Universidade Estadual do Norte Fluminense-UENF, Centro de Biociências e Biotecnologia, Laboratório de Ciências Ambientais;* **Ana Paula M. Di Beneditto**

VARIATION IN CALLING BEHAVIOR IN NORTHEASTERN PACIFIC BLUE WHALES (BALAENOPTERA MUSCULUS)

Kathleen M. Stafford, *National Marine Mammal Laboratory; Sue E. Moore*

CONSERVATION STATUS OF THE BLACK SEA BOTTLENOSE DOLPHIN (TURSIOPS TRUNCATUS PONTICUS): AN ASSESSMENT USING MORPHOLOGY AND GENETIC VARIATION

K. Viaud, *Department of Biology, San Diego State University; A. J. Bohonak; R.L. Brownell*

MORPHOLOGICAL AND PHOTOMETRIC CHARACTERIZATION OF INTERGENERIC HYBRIDS BETWEEN TURSIOPS AND DELPHINUS, TURSIOPS AND PSEUDORCA

Heather R. Zornetzer, *Department of Biology, Portland State University, D.A. Duffield*

Abstracts

COLOR PATTERNS OF THE DWARF MINKE WHALE AND USE OF COLORATION IN ASSESSING TAXONOMIC RELATIONSHIPS OF THE BALEEN WHALES

Peter Arnold
Museum of Tropical Queensland

Alastair Birtles
Tourism Program, James Cook University of North Queensland

Vimoksalehi Lukoschek
School of Tropical Environment Studies & Geography
James Cook University of North Queensland

Andy Dunstan and Monique Matthews
Undersea Explorer

In this presentation we describe color patterns of the dwarf minke whale, based on underwater observations and documentation of over 200 animals from northeastern Australia and comparative material from throughout its range. We review color patterns in other mysticetes and present a phylogenetic analysis (13 color characters; 12 taxa, outgroup: sperm whale).

The dark throat patch, shoulder blaze and basal flipper blaze were consistently present in the dwarf minke whales examined and are confirmed as diagnostic characters. Cladistic analyses of other color elements suggested that the common (northern hemisphere) minke whale *Balaenoptera acutorostrata* and dwarf minke whales are most closely related. Shared derived features included a specialized flipper pigment (with distal dark flipper patch), a well developed peduncle blaze and a linear to diffuse nape streak. The monophyly of the Antarctic minke whale *Balaenoptera bonaerensis*, common and dwarf minke whales was supported by the occurrence only in those species of a double caudal chevron, as well as a dark cape-like thorax field separating the light gray lateral field into anterior thorax and posterior flank patches. A review of osteological, morphological and molecular data is needed, in addition to the color pattern data, to determine whether the dwarf minke whale is recognized as a subspecies of *acutorostrata* or as a distinct species. Pending such information we support recognition at subspecies level as the more conservative approach.

Our analyses also suggested a suite of color characters that may distinguish the genus *Balaenoptera,* including a light gray flipper with white leading edge, light gray transitional lateral field (possibly absent in sei whales), blowhole streaks, ear stripe, and a tendency for color asymmetry. Blue and humpback whales lack most of these color elements, although the blue whale does show the generalized flipper pattern. The absence of these color elements in humpback and blue whales could not be linked to their habitat or lifestyle, as has been suggested.

The relationships between generally recognized families of baleen whales were poorly reflected in the analyses based on color, with the pygmy right whale grouped with the rorquals.

Although there appears to be a phylogenetic basis for the color patterns of rorquals, other factors may be present. In particular, the sharply defined patterns of dark gray, light gray and white of dwarf minke whales, the most complex coloration of any mysticete, could be used in intraspecific signaling.

Documentation of color remains poor for several species, especially in the Bryde's whale complex. By highlighting that there are consistent color elements in balaenopterid whales, we hope that those with the opportunity will record such features. Such data are needed to check whether our interpretation of available information on color is correct and thus test the validity of the taxonomic hypotheses suggested by our preliminary analyses.

WWW.DNA-SURVEILLANCE: IMPLEMENTING A COMPREHENSIVE AND VALIDATED MOLECULAR TAXONOMY FOR CETACEANS

C. Scott Baker, Merel L. Dalebout, Shane Lavery, Howard A. Ross, Susana Caballero, Mathew Goode, Greg Ewing and Allen Rodrigo
School of Biological Sciences and Institute for Bioinformatics
University of Auckland

DNA Surveillance is a Web-based program to assist with the phylogenetic identification of species within a taxonomic group (e.g., Order or Family), particularly those threatened by exploitation or illegal trade. The program aligns a user-submitted DNA 'test' sequence with curated datasets of pre-aligned reference sequences and returns a phylogenetic tree (including bootstrap support) showing the relationship of the test sequence to the reference sequences. A table summarizing the evolutionary distances between the test sequence and each reference sequence in the dataset is also provided (Ross et al. 2003). The reliability and discriminatory power of *DNA Surveillance* is dependent on establishing a comprehensive dataset of reference sequences from validated 'voucher' specimens representing the maximum diversity observed at each taxonomic level, including phylogeographic diversity below the species level. In its current form, *DNA Surveillance* is implemented for the phylogenetic identification of whales, dolphins and porpoises (Cetacea). Version 3.1 consists of two reference databases for cetaceans: the hypervariable 5' end of the mitochondrial (mt) DNA control region (285 – 476 bp, depending on the dataset); and the more conservative cytochrome *b* gene (405 bp). These sequences were drawn from published articles, GenBank submissions, and the genetic archives of the NMFS Southwest Fisheries Science Center and the University of Auckland, New Zealand. A total of 87 species in 14 families of the 89 recognized cetaceans are represented in one or both databases (mtDNA control region, 285 sequences representing 78 species; mtDNA cytochrome *b*, 165 sequences representing 83 species). In most cases, each species is represented by several reference sequences obtained from different populations or 'stocks' to maximize representation of phylogeographic diversity, and allow potential assignment of the test sequence to geographic origin or source population.

Here, and in accompanying presentations (Dalebout et al. and Lavery et al.), we report on progress with validating the reference datasets and testing the reliability of *DNA Surveillance* for identification of cetacean species. We discuss the need for designating 'virtual curators' for species- or genera-specific phylogeographic datasets to improve identification of geographic origin for test sequences and help resolve problems of apparent paraphyly among the mtDNA lineages of some groups. Finally, we compare and contrast the taxon-specific, phylogenetic approach implemented in *DNA Surveillance* to that of the 'universal', distance matching or clustering approach of DNA 'barcoding' (Hebert et al. 2003).

TURSIOPS IN ATLANTIC SOUTH AMERICA: IS *TURSIOPS GEPHYREUS* A VALID SPECIES?

André S. Barreto
CTTMar – Vale do Itajai University

The number of species in the genus *Tursiops* has been in debate for a long time. In South America, the species *Tursiops gephyreus* was originally used for bottlenose dolphins from its southern Atlantic coast. However, since the original description was based on only two specimens and no taxonomic study was done to confirm the species validity, its use gradually decreased when the morphological plasticity of the genus was recognized and the scientific opinion changed to a single, cosmopolitan, species. In order to verify the existence of different bottlenose dolphin morphotypes in the area, skulls from 103 cranially mature specimens were analyzed for 53 metrical and 15 non-metrical characters. Multivariate exploratory analysis (multidimensional scaling) revealed the existence of two groups of bottlenose dolphins along the studied area. These morphotypes were latitudinally separated, and were thus initially named "southern" and "northern". They were compared using the ANOSIM routine of PRIMER 4.0 and the differences between the groups were found to be statistically significant. Differences in the overall size of the skulls (condylobasal lengths, CBL) were observed, with the northern morphotype being smaller, exhibiting CBLs up to 545mm, while the southern one had CBLs larger than 550mm. Also, statistically significant differences were found when measurements were expressed as a percentage of CBL, indicating differences in shape as well as in size. Considering that in the geographical zone were the two morphotypes occur in sympatry specimens with intermediate characteristics weren't observed, it can be assumed that interbreeding between the types is, at least, restricted. Comparing the morphotypes with the original description of *T. gephyreus* it was found that most of the characteristics described for it applied to the southern morphotype. Thus, it is possible that *T. gephyreus* should be considered a valid species, due to its diagnosable characteristics and the absence of identifiable hybrids in the zone of sympatry with *T. truncatus*.

MOLECULAR SYSTEMATICS OF THE SOUTH AMERICAN COASTAL AND RIVER DOLPHIN *SOTALIA sp.*

Susana Caballero and C. Scott Baker
Population Genetics and Evolution Research Group, The University of Auckland

Luis Medrano González
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Fernando Trujillo
Fundación Omacha

Julianna A. Vianna and Fabricio R. Santos
Departamento de Biologia Geral, ICB, Universidad Federal de Minas Gerais

Héctor Barrios and María Gabriela Montiel
Departamento de Biología, Universidad del Zulia

Sandra Beltrán
Colecao Zoologica Paulo Burheim, Centro Universitario Luterano de Manaus

The genus *Sotalia*, the tucuxi dolphin, is currently considered a member of the Stenoninae sub-family, along with the genuses *Steno* and *Sousa*. Currently only one species is recognized (Rice, 1998) but two ecotypes or subspecies have been described. The coastal ecotype (*S.fluviatilis guianensis*) is found in estuaries and bays from Nicaragua to Southern Brazil and the riverine (*S.f.fluviatilis*) is found throughout the Amazon River and most of its tributaries. Here we evaluate this current classification using mitochondrial and nuclear genes, the Control Region and a portion of the Cytochrome *b* at the mitochondrial level, the first intron of the Actin muscle gene and two additional nuclear introns of the Glucocerebrosidase gene (GBA) and Interferon (IFN) genes. Four Y chromosome introns have been sequenced and are now under analysis. *Sotalia* samples from six different geographic locations were analyzed, including two tissue samples from the Colombian and Brazilian Amazon. Additional DNA samples extracted from bones and tooth from these regions were successfully amplified and sequenced for the mitochondrial genes.

Preliminary results show *Sotalia* being basal to other delphinids and separated from other members of the Stenonidae. These results are not consistent with current subfamily classification. Also, clear differentiation between the two ecotypes was found at the mitochondrial and nuclear genes, although sample size was small for the latter. This suggests that the two ecotypes should be recognized as Evolutionarily Significant Units and confirmed as subspecies, requiring further considerations for elevation to species. Regional and ecological differences of *Sotalia* populations have special significance for conservation management programs.

LARGE INTER-OCEAN GENETIC DIFFERENCES WITHIN *KOGIA SIMA* INDICATE LONG-TERM ISOLATION AND POSSIBLY A SECOND SPECIES

Susan J. Chivers, Richard G. Le Duc and Kelly M. Robertson
NMFS, Southwest Fisheries Science Center

Nelio B. Barros
Hubbs-Seaworld Research Institute

Andrew E. Dizon
NMFS, Southwest Fisheries Science Center

We reconstructed a phylogeny for the two extant species in the Family Kogiidae using mitochondrial DNA genes. For our analyses, we sequenced 406 base pairs of the mitochondrial control region and 398 base pairs of the mitochondrial cytochrome *B* gene from 108 *K. breviceps* and 47 *K. sima* samples collected throughout their range. Haplotypic diversity was high, and 74 haplotypes were identified for *K. breviceps* and 27 for *K. sima*. As expected, the two extant sister species: *K. breviceps* and *K. sima*, were reciprocally monophyletic in phylogenetic reconstructions rooted with the closest living relative: *Physeter macrocephalus*. However, within *K. sima* we observed another reciprocally monophyletic relationship, which separated haplotypes between the Atlantic and Indo-Pacific Oceans. Descriptive statistics summarizing haplotype characteristics revealed approximately equivalent genetic differentiation within *K. sima* between the Atlantic and Indo-Pacific Oceans (e.g. $\Phi_{st} = 0.91$) as between *K. breviceps* and *K. sima* ($\Phi_{st} = 0.86$). In contrast, the genetic differentiation between Atlantic and Indo-Pacific *K. breviceps* was small ($\Phi_{st} = 0.10$). One explanation for the apparent long-term isolation of *K. sima* between the Atlantic and Indo-Pacific Oceans may be due to differences in distribution of the two species. *K. sima* appears to be more restricted to tropical waters than *K. breviceps*, which may have limited the movement of *K. sima* around South Africa during the Pleistocene's glacial maxima events but not those of *K. breviceps*. Although our results are inarguably consistent with species-level differences between the two *K. sima* clades, recognition of a third *Kogia* species awaits supporting evidence of reproductive isolation.

GENETIC DISTINCTIVENESS, EXCLUSIVITY AND CONCORDANCE: A MOLECULAR TAXONOMIC APPROACH TO SPECIES IDENTIFICATION AND DISCOVERY

Merel L. Dalebout
Biology Department, Dalhousie University

C. Scott Baker
School of Biological Sciences, University of Auckland

James G. Mead
National Museum of Natural History, Smithsonian Institution

Genetic distinctiveness, exclusivity and concordance are important components of a molecular taxonomy. Sequences from orthologous loci from specimens assumed to represent a given taxon are expected to form monophyletic lineages, excluding lineages representing other recognized species. Synapomorphic nucleotide substitutions should distinguish these lineages from one another. The robustness for such species-specific clades can be assessed statistically (e.g., non-parametric bootstrap or maximum likelihood). Phylogenetic analyses of multiple loci, together with assessment of morphological features, geographic distribution and behavior, should yield concordant results. Within this framework, a molecular taxonomy can aid in the discovery and formal taxonomic description of new species. We will review the application of this approach using the family Ziphiidae (beaked whales) and discovery of a new species, *Mesoplodon perrini,* as a case study. The distinctiveness of mtDNA lineages among recognized species of beaked whales is contrasted with patterns observed among members of another highly speciose family of cetaceans, the Delphinidae (Lavery et al. this meeting). Finally, the ability of a molecular taxonomy to function in the absence of a morphological holotype is considered. This would be undesirable in most cases, but may sometimes be the only option. This issue is of particular importance given the rapid loss or decline of the earth's biological diversity.

USE OF BIOMARKERS AS SPECIES-LEVEL SYSTEMATIC AND FORENSIC MOLECULAR TOOLS IN THE DETECTION OF CETACEAN HYBRIDS

Deborah A. Duffield and Heather R. Zornetzer
Portland State University

Edgard O. Espinoza
National Fish and Wildlife Forensics Laboratory

Nélio B. Barros
Center for Marine Mammal and Sea Turtle Research, Mote Marine Laboratory

Evolutionarily-conservative protein molecules, such as hemoglobin, myoglobin and serum albumin, are informative biomarkers at the species and sub-species/stock levels. Currently, hemoglobin and myoglobin biomarkers have been applied successfully to the identification of 144 species of mammals, 108 species of birds, and 2 of reptiles. These two biomarkers, evaluated in concert, differentiate over 40 marine mammal species to date and are an integral part of the National Fish and Wildlife Forensics Laboratory (Ashland, OR) database for the diagnostic determination of species falling under the protection of CITES treaties. Current research shows that hemoglobin and myoglobin biomarkers confirm species identification of *Kogia breviceps vs. K. sima*, useful for stranding and rehabilitation efforts along the east and west U.S. coasts. Hemoglobin is also useful in distinguishing coastal *vs*. offshore *Tursiops truncatus* ecotypes in the Atlantic and Gulf of Mexico, as well as in identifying mixing between these stocks. Myoglobin and albumin have been useful in confirming and detecting hybridization involving *Tursiops*, *Delphinus*, *Pseudorca* and *Stenella* species and have the potential to be diagnostic across many more cetacean species. Distinctions between these biomarkers can be made based on molecular weight differences detected using mass spectrometry (either ESI MS or MALDI-TOFMS) and by protein electrophoresis. Both mass spectrometry and electrophoretic methods are rapid procedures, robust to molecular degradation (especially mass spectrometry) and only require very small samples which can be stored frozen or dried. We present a comprehensive species-specific and stock-specific catalogue of these three biomarkers, including examples of confirmed and potential hybrid detection. Emphasis is placed on the application of these biomarkers in identifying species particularly in situations where field identification of individuals is doubtful or confusing (i.e., strandings), to add information when investigating species and sub-species differentiation, and in the exploration of potential species hybridization and introgression.

USING POLYMORPHIC DATA AND FREQUENCY ANALYSIS TO INFER THE PHYLOGENETIC RELATIONSHIPS AND BIOGEOGRAPHY OF PORPOISES (CETACEA: PHOCOENIDAE)

Liliana Fajardo and Annalisa Berta
San Diego State University, Department of Biology

Robert L. Brownell Jr.
Southwest Fisheries Science Center

Claudia C. Boy
Museo Acathusun, Tierra del Fuego, Argentina

The inclusion or exclusion of variable characters or polymorphic data poses a number of challenges for systematists. Several studies (Mabee, 1993; Martins, 1997; Wiens, 1999) have shown that polymorphic data can be highly informative and increase accuracy; thus the exclusion of such characters is unjustified. Among the different methods described to code polymorphic characters frequency analysis has been identified as the most accurate (Wiens, 1999). The purpose of this study is to infer the phylogeny of the family Phocoenidae (true porpoises) using polymorphic data and frequency analysis.

The Phocoenidae includes six extant species that have a worldwide anti-tropical distribution. Two prior phylogenetic hypotheses have been proposed based on morphology and molecular sequence data. The results of these studies conflict, resulting in unresolved phylogenetic relationships among members of this group. This study evaluates a comprehensive set of morphological characters from the cranial, post-cranial and soft-anatomical regions to infer the relationships among phocoenids. All extant species and well known fossil taxa are included. Outgroup taxa include several delphinids and monodontids, as well as Inia and Pontoporia, sister taxa to delphinids and delphinoids, respectively. A second part of the study examines the origin and distribution of porpoises using physical and ecological information (i.e. past geologic events related to opening and closing of seaways, paleo-oceanic models, changes in global temperature) within the context of a phylogenetic framework.

Preliminary phylogenetic results of 20 cranial, 20 post-cranial and 19 soft-anatomical characters strongly support the monophyly of Phocoenidae using maximum parsimony and Bayesian analyses. Morphological characters were coded as polymorphic (using McClade 4.0) and frequency data (using Manhatan distances, Berlocher & Swofford, 1997). Both analyses result in the same tree topology, however, the frequency data provided higher support for all internal nodes, as well as higher resolution when the fossil taxa are included. Results support the position of *Neophocaena phocaenoides* to be basal to all extant phocoenids as proposed by previous molecular analysis. The extinct genus *Piscolithax†* appears to be basal to all phocoenids, however relationships among members of this group are weakly supported. The oldest fossil phocoenid from the late Miocene (10-11 mya), *Salumophocanea stocktoni†* is closely related to *Phocoena spinipinnis* and *Phocoena sinus.* A previous morphological study (Barnes, 1985) proposed the division of phocoenids into two subfamilies: Phocoeninae and Phocoenoidinae; however, this division was not supported by this study. Based on these preliminary phylogenetic results, support for a North Pacific origin of phocoenids was found. An increase in ocean temperatures during the mid-Pliocene probably formed a barrier in tropical regions, thus forcing a restricted distribution of phocoenids in the northern and southern hemisphere, resulting in the current anti-tropical distribution of extant species.

APPLICATIONS OF ESU AND SUBSPECIES CONCEPTS IN BIRDS

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Genetic methods can be applied to issues of avian conservation in a variety of ways. One of the more useful applications has been to apply neutral genetic markers to determine population structure and phylogenetic or conservation units of birds. Non-neutral genetic systems can be useful for assessing the potential adaptability of a population to environmental change or translocation, however, birds are thought to be more coarse-grained in associations with their environment than most other organisms, and so local adaptation may not be considered as much of a factor as with fish, amphibians, invertebrates or plants. Endangered bird populations can become highly fragmented and decrease to very low levels, and ensuing genetic bottlenecks could impact fitness. However, flight provides most birds with greater powers of dispersal and thus gene flow is relatively high and may mitigate these impacts. DNA markers are very powerful for determining the natural boundaries between populations of species that may indicate separate evolutionary pathways (often defined as evolutionarily significant units or ESUs). This information is important for defining an endangered species or subspecies under the U.S. Endangered Species Act, and also for determining natural levels of gene flow for future management. I will illustrate some of the inherent problems of subspecies and ESU methods with a number of recent studies of birds, including examples from Pacific (e.g., Hawaiian and Marianas species) and North American birds (e.g., Yellow-billed Cuckoo, Common Raven and Spotted Owl).

DIFFERENTIATION OF GEOGRAPHIC STOCKS OF ROUGH-TOOTHED DOLPHINS, *STENO BREDANENSIS*, USING MOLECULAR TECHNIQUES

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Little is known about the life history of the rough-toothed dolphin, *Steno bredanensis*, except that they are typically found within deep tropical, subtropical, and warm temperate waters, usually above 25º C. Genetics may prove a useful tool in distinguishing the extent of genetically separate stocks of rough-toothed dolphins and aiding in management decision-making.

Using full control region sequences from 31 specimens from the eastern Pacific, western Atlantic and Gulf Coast of Florida, we found clear separation between ocean basins and several geographically restricted clades within the Pacific. Three beachcast specimens from central California were unrelated to other populations included in the current sample. Molecular analysis partitioned all but one of the remaining samples into two geographic regions, three clades each off the Central American coast and in the tropical central Pacific.

Genetic distances between Atlantic/Gulf of Mexico and Pacific Steno clades averaged about 2% sequence divergence. This is comparable with control region genetic distances between closely related delphinid congeners. Surprisingly, genetic divergence between some distinct Pacific clades was just as high as that found between the Atlantic/Gulf of Mexico and the Pacific, ranging from 0.5% to 2.2%.

Three distinctive clades (colored pink, purple and blue) were found only in the Eastern Tropical Pacific region, off the coast of Central America. Another three clades (green, aqua, and brown) are found in the Central Pacific, with one vagrant biopsy-sampled specimen in the ETP (1317ETP). Three animals stranded along the central California coast (orange) are unrelated to other groups.

DOES CRANIAL MORPHOLOGY REFLECT ADAPTIVE EVOLUTIONARY DIVERGENCE OF SYMPATRIC KILLER WHALE (*ORCINUS ORCA)* ECOTYPES?

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In the northeast Pacific, two sympatric killer whale (*Orcinus orca*) ecotypes exhibit dramatic resource polymorphisms: residents feed exclusively on fish, and transients hunt marine mammals. Recent genetic studies have corroborated behavioural evidence that the resident and transient forms are reproductively isolated despite the absence of any geographic or temporal barrier. Segregation of the sympatric ecotypes is apparently maintained by cultural mechanisms alone, which is extremely unusual among non-human mammalian species. Subtle morphological differences in pigmentation and dorsal fin shape have been described and are useful for distinguishing residents and transients in the field. However, these differences have no known adaptive significance and likely resulted from genetic drift. Members of the two ecotypes use fundamentally different foraging techniques that almost certainly exert different selective pressures on morphology. We therefore asked whether residents and transients are evolving along divergent morphological trajectories, or whether the present dietary dichotomy is too recent or ephemeral for adaptive evolution to be observed. To test the hypothesis that adaptive evolutionary divergence has in fact occurred, we are characterizing and comparing the cranial skeleton of residents and transients both qualitatively and quantitatively. Linear, outline, and landmark data is being collected from the skulls and mandibles of northeast Pacific killer whales. The ecotype of each specimen will be determined by analyzing mitochondrial DNA sequences. Results will be interpreted in the context of the biomechanical advantages of, or constraints on, cranial structure in relation to ecotype-specific differences in diet and foraging behaviour. We predict that transient killer whales that bite and tear apart large mammalian prey have more robust skulls, mandibles, and dentition than the piscivorous residents that handle smaller prey items.

COMBINED VERSUS SEPARATE ANALYSES OF SYSTEMATIC DATA SETS FOR CETACEANS

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In order to incorporate as much data as possible into phylogenetic hypotheses, systematists have taken two approaches: "total evidence" and "supertree" analysis. The total evidence framework calls for the concatenation of all characters into a single comprehensive "supermatrix," followed by a single analysis of the combined data. In the supertree approach, separate analyses of different systematic data sets are executed first, followed by a reconciliation of trees supported by the different individual analyses. We show how these two very different approaches to phylogenetic analysis can lead to very different conclusions, and discuss the implications of these methods for cetacean systematics at the species level. A nearly comprehensive total evidence analysis of extant Cetartiodactyla (whales and even-toed hoofed mammals) and supermatrix/supertree analyses of Mysticeti, based on mitochondrial and nuclear characters, will be presented.

A MAXIMUM LIKELIHOOD TEST OF PHYLOGENETIC IDENTIFICATION OF SPECIES

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The web-based program, DNA-Surveillance, aims to provide a robust system for phylogenetic identification of species using a database of reference sequences under curation of species specialists. Such a method allows an indication of the uncertainty in the match, or the potential of a novel classification, by its position within a phylogenetic tree, using Neighbor Joining reconstruction, and the support of that positioning by non-parametric boostrap resampling. Trees are constructed based on the combination of query sequences and verified cetacean sequences stored in a database. The flagship database from the DNA-Surveillance effort is the 'Witness for the Whales' website (http://www.dna-surveillance.auckland.ac.nz), an identification system for cetaceans. The database allows taxonomic queries at different hierarchical levels of resolution (e.g. family, genus, species), as well as plans for geographic assignment by phylogeographic analysis.

A new set of approaches is being investigated that derive from a likelihood-based phylogenetic model. In this approach the database would not only include sequence data but also a predefined reference tree. The position of a query sequence may then be estimated, for example, by attaching the query sequence at all possible positions in the reference tree, and applying a maximum likelihood analysis. Various methods for estimating the support of possible positioning within tree are possible but perhaps the most promising is a Monte-Carlo Markov-Chain approach that would estimate posterior distributions of query sequence location. This provides a general statistical framework for assessing alternative hypotheses of species identity.

WHY Y? Y CHROMOSOME-SPECIFIC MARKERS AND THEIR USE FOR CETACEAN SYSTEMATICS

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Parallel analysis of maternally-inherited mitochondrial (mtDNA) and bi-parentally-inherited autosomal nuclear DNA (aDNA) is commonly used to provide evidence of cetacean population structure, and to address hypothesized gender biases in dispersal rates. To assess differences between male and female dispersal rates, however, the ideal comparison is between markers transmitted exclusively by each sex, i.e. between maternally-inherited mitochondrial DNA and paternally-inherited DNA from the long arm of the Y chromosome (yDNA). In addition to documenting sex differences in admixture coefficients, comparing mtDNA, yDNA and aDNA can be used to illuminate differences in the demographic and selective histories of genomes whose evolutionary paths are mediated by male versus female versus bi-parental reproductive success. Finally, comparing maternally and paternally-derived phylogenetic relationships and/or species'-level assignment test results has the potential to elucidate questions of directionality in cases of known or suspected hybridization among cetacean species.

The broader utility of Y chromosome markers for cetacean systematics is presented in light of results from a case study of fin whales (*Balaenoptera physalus*). The distribution of variation at Y chromosome-specific anonymous loci, Y-specific microsatellite loci, control region mtDNA, and autosomal microsatellite loci is examined among sampled fin whale populations in the North Atlantic and North Pacific Oceans. Comparison among multiple estimates of genetic differentiation derived from Y chromosome, mitochondrial and autosomal loci supports the hypothesis that, on an evolutionary timescale, male fin whales disperse more widely than females. Eight additional baleen whale species and the sperm whale were also sequenced for Y chromosome and mitochondrial loci. Phylogenetic signal present in paternally-inherited DNA is compared to signal present in maternally-inherited DNA, and tree congruence analyses are briefly discussed.

MALE GENES AND MALE SONGS: FIN WHALES AS A CASE STUDY FOR INTEGRATING GENETIC AND ACOUSTIC DATA IN DEFINING BALEEN WHALE STOCKS

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Understanding genetic relationships among whales from different geographic areas is essential to effectively managing these endangered species. Here, we focus on defining relatedness and gene flow among fin whales (*Balaenoptera physalus*) in the North Pacific and North Atlantic to better understand the utility of genetic and acoustic variation for identifying "stocks" of vocally active baleen whales. To directly estimate male-mediated gene flow, Y chromosome-specific markers were isolated from fin whales. Gene flow estimated from Y-specific markers is compared directly to female-mediated gene flow estimated from mitochondrial DNA to determine whether gene flow is "gender-biased". Geographic variation in songs produced by male fin whales in the North Pacific and North Atlantic are quantified for comparison with estimates of genetic distance based on Y chromosome, mitochondrial DNA, and autosomal DNA to identify particular signal features that are concordant with patterns of gene flow mediated by male fin whales, female fin whales or both. Acoustic features useful for detecting shorter-term discontinuities in fin whale movement and/or social behavior are also identified. Integrating acoustic and genetic data will help define management units for fin whales and inform conservation efforts for other baleen whale species whose pelagic life histories have made monitoring recovery status difficult.

NEW METHODS FOR MEASURING GENE FLOW BETWEEN CLOSELY RELATED POPULATIONS OF SPECIES

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Gene flow between populations causes genetic variation to be shared. However if two populations have recently separated, or if two new species have recently diverged, both new entities are expected to share variation that was present in the ancestral population, regardless of whether or not there has been gene flow. Thus assessing the role of gene flow, which will also cause shared variation, in the early stages of species formation is very difficult. A new analytical protocol and a new empirical protocol, that together should overcome this difficulty, are described.

The analytical protocol is a multi-locus extension of a Likelihood/Bayesian approach that provides posterior probability densities for parameters of population size, divergence time, and gene flow rates. The empirical protocol relies upon loci that have two very different mutation rates. Most DNA sequence data come from loci with low mutation rates and these will not provide sufficient resolution for distinguishing different kinds of recent events. High mutation rate loci (e.g. short tandem repeat -STR or microsatellite loci) could provide this resolution, in principal, however the recurrent mutation at these loci can obscure allele sharing due to ancestry and allele sharing due to recurrent mutation.

In our procedure, haplotype data are collected for loci that include a region of unique DNA sequence linked to a simple STR region. The two different components offset each other's shortcomings. We have included this two-mutation-rate model into the program for fitting the isolation with gene flow model. An example is described using genes from closely related species of cichlid from Lake Malawi.

GEOGRAPHIC VARIATION OF FRANCISCANA, *PONTOPORIA BLAINVILLEI* (GERVAIS & D'ORBIGNY, 1844) BASED ON SKULL MORPHOLOGY

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In order to evaluate the geographic variation of the Franciscana, *Pontoporia blainvillei*, I studied 228 adult specimens from 117 localities along the genus distribution in the Western South Atlantic. These localities were grouped into 15 geographical samples. I analyzed 30 morphometric cranial characters along a latitudinal transect, throughout uni and multivariate statistical analyses. Dice-Leeras diagrams, Principal Components Analysis and Discriminant Canonical Analysis indicated the existence of significant geographic variation. Skull size was the main factor responsible for sample differentiation. Sample means from the northernmost portion of the distribution (Itaúnas, ES, 18°25'S 39°43'S) decrease southwards until the Brazilian states of São Paulo and Paraná (~25°52'S). Continuing southwards, averages began to increase. Between the island of Florianópolis (SC, ~27°36'S) and the south of the Buenos Aires Province (~41°01'S), samples presented smaller variation, with mean size larger than those from the northern samples. The exception was the sample from the La Plata River estuary (~36°00'S), with smaller averages than those from adjacent samples. Besides this variation, I could not detect any discontinuities that could be taxonomically related. This result provides further evidence that the geographical samples represents a single species, meanwhile, the groups should be consider separately for management and conservation purposes. Considering that water depth may be an important limiting factor in *Pontoporia* distribution - restricted to shallow waters - the pattern of geographic morphological variation that I observed is consistent with cyclical Quaternary sea-level oscillations along the eastern coast of South America. During the last glacial maximum, the coastlines reached up to 100 meters below the present level resulting in a narrowing or disappearance of the continental shelf in some areas of the species range. This possibly acted as geographic barrier, creating the geographic variation pattern described above after a secondary contact of previously isolated populations.

LOW WORLD-WIDE GENETIC DIVERSITY IN THE KILLER WHALE (*ORCINUS ORCA*); IMPLICATIONS FOR DEMOGRAPHIC HISTORY

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A low level of genetic variation in mammalian populations where the census population size is relatively large has been attributed to various factors, such as a naturally small effective population size, historical bottlenecks, and social behaviour. The killer whale is an abundant, highly social species with reduced genetic variation. We find no consistent geographic pattern of global matrilineal diversity, and no mtDNA variation within some regional populations. The regional lack of variation is likely due to the strict matrilineal expansion of local populations. World-wide mtDNA diversity is also low (π= 0.0052 ± SD 0.0031 for the control region, π= 0.0039 ± 0.0026, for the combined ATPase6 and ATPase8 loci). A level of diversity this low is most common among species thought to have undergone a population bottleneck, and various other analyses, including mismatch distributions and tests for neutrality, are consistent with this interpretation for the killer whale. Microsatellite DNA diversity is also comparatively low. We discuss these data in the context of current considerations about taxonomy in the genus *Orcinus*.

SYSTEMATICS OF COASTAL TROPICAL SMALL CETACEANS: A REVIEW

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This paper reviews the taxonomy and systematics of six genera of coastal small cetaceans found in tropical waters. The genus *Orcaella* currently contains a single species (*Orcaella brevirostris*, Irrawaddy dolphin) that occurs from the northern coasts of Australia and Vietnam to central East India. Recent work on skull morphometrics and genetics strongly suggests that the genus should be split into two, an Asian and an Australian species. Such a proposal has recently been made. The genus *Sotalia* also consists of a single species (*Sotalia fluviatilis*, tucuxi). The tucuxi occurs in South American coastal waters from Nicaragua/Honduras in the north to southern Brazil, and in the Amazon Basin. The relationship between the coastal and riverine forms is controversial. Some have considered them to be two subspecies: *S. f. fluviatilis* (Amazon) and *S. f. guianensis* (marine), but some recent work has suggested that they should be considered separate species. Clearly further work is needed. *Sousa* contains the humpback dolphins of the Indo-Pacific and eastern Atlantic oceans. Most recent authors consider that there are two species (*Sousa teuszii* off the coast of West Africa and *Sousa chinensis* in the Indo-Pacific). This genus has been particularly controversial, with various recent authors listing from one to five species. A large-scale study of skull morphometrics suggests that 2-3 species are valid, and there is an ongoing study of global genetic variation, which should shed light on the issue in the near future. The globally-distributed genus *Delphinus* is considered by most recent authors to contain two species (*Delphinus delphis*, the short-beaked common dolphin and *Delphinus capensis*, the long-beaked common dolphin). Some consider a third species, *D. tropicalis*, valid, but recent work on morphometrics has suggested that it is probably a subspecies of *D. capensis*. Various ongoing studies are examining the taxonomy and systematics of this widely-distributed group of dolphins. *Pontoporia* is a genus of platanistoid dolphin found in coastal waters of South America. Only one species is recognized (*Pontoporia blainvillei*, fransiscana). Again, recent work has suggested the existence of various geographical forms along its north/south range. However, there is some controversy as to whether these forms represent separate species, subspecies, or simply different populations. The relationships are being examined with both morphometric and genetic data. In northern Asia, the genus *Neophocaena* is found from Japan and Indonesia to the Persian Gulf, and most recent authors consider there to be a single species (*Neophocaena phocaenoides*, finless porpoise) with three subspecies. There appear to be two very distinct morphological forms, and preliminary evidence suggests that they do not interbreed, despite being sympatric over a large area in central China. Further work, using both morphological and molecular characters, is underway to examine the situation. In general, while there is undoubtedly great geographic variation within each of these coastal, tropical genera, the exact patterns of variation and the relationships of the various forms are not know with certainty. This largely results from a lack of adequate samples, and to a lesser extent, a paucity of marine mammal biologists working on these animals.

THE ORIGINS AND EVOLUTIONARY HISTORY OF MODERN FELID SPECIES

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A comprehensive understanding of the evolutionary history of modern taxa involves not only a more precise description of the phylogenetic relationships among species, but also an assessment of the uniqueness among groups of the same species and the study of historic patterns of gene flow. The application of genetic markers and tools to these questions often results in the establishment of objective criteria for recognizing species and subspecies, to the interpretation of their natural history, and to improved conservation efforts. Resolution of the relationships among the 37 Felidae species using molecular genetic techniques has been challenging because of the numerous rapid and recent speciations in the family. Phylogenetic and chronometric analyses of 22,789 bp from 38 autosomal, X-linked, Y-linked, and mitochondrial gene segments of all species of the Felidae family suggest that the eight major modern felid lineages diverged in rapid succession between the late Miocene (10.2 MYA) and early Pliocene (5.5 MYA) and most living species emerged 3.1 to 0.7 MYA. These analyses provide insights into the difficulties that will be encountered reconstructing the evolutionary history of all living taxa, especially radiations with bifurcations spanning less than 500,000 years, and demonstrate that Y-linked markers may be particularly informative for these time frames in mammals.

DOES GENETIC DIFFERENTIATION IMPLY SPECIATION? PHYLOGENETIC AND PATERNITY ANALYSES SHOW THAT THIS IS NOT THE CASE IN WEST-AUSTRALIAN BOTTLENOSE DOLPHINS (*TURSIOPS* SP.)

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The taxonomic status of the genus *Tursiops* is currently unclear. Two questions predominate in the current literature. The first is whether the genus *Tursiops* actually comprises a single species (*T. truncatus*), or whether there are several species or subspecies, dependent on their geographic occurrence. The second question is whether the genus *Tursiops*, as it is currently recognized, forms a single clade. According to Rice (1998), there are currently two species recognized in this genus: *T. truncatus* and *T. aduncus*. Previous phylogenetic analyses, based either on cytochrome *b* (LeDuc *et al.* 1999) or mitochondrial DNA (mtDNA) control region sequences (d-loop) (Curry 1997), as well analysis of morphological characters and control region (Wang *et al.* 1999), suggest that *T. truncatus* and *T. aduncus* are two separate species.

We used a combination of both phylogenetic and behavioural approaches to determine the species status of Indian Ocean inshore bottlenose dolphins from Shark Bay in Western Australia by sampling 217 individuals and sequencing 351 base pairs of the hypervariable part I of d-loop. Nucleotide diversity between clusters ranged from 0.045 to 0.051. Four different methods of phylogenetic reconstruction showed that there are two distinct clusters of haplotypes: six of the eight haplotypes found in Shark Bay unambiguously formed a strongly supported clade within previously published *T. aduncus*. In contrast, the other two haplotypes formed a strongly supported group among previously published *T. truncatus* when maximum likelihood and Bayesian analyses was used, but lacked resolution using maximum parsimony and neighbour joining.

This raised the question whether Shark Bay dolphins form two sympatric, but reproductively isolated populations. To test this hypothesis, we partitioned the data set into two groups. In the first group we pooled all Shark Bay dolphins carrying a haplotype from the *truncatus* clade, and in the other group we pooled the animals from the *aduncus* clade. Using microsatellite allele frequency data from 10 loci, we performed two analyses of molecular variance. We investigated two different hypotheses: firstly, that the major subdivision of microsatellite genotype frequencies is between members of the two mtDNA groupings, which may be sympatric populations with isolated gene pools; and secondly, that the major subdivision is between localities, with members of the two mtDNA groups exchanging nuclear genes freely. For both hypotheses, most of the variation was found within each sampling locality (88.36% and 88.42%, respectively, both $p < 0.01$). For the first hypothesis, 0.00 % of the variation was explained by grouping the samples according to mtDNA clades ($p < 0.01$), and 11.63 % of the variation was found among sampling localities within mtDNA clades ($p < 0.01$). For the second hypothesis, 6.61% of the variation was explained by placing the samples into seven sampling localities ($p < 0.01$), and only a small but significant amount of variation (4.97%) was explained by stratifying the samples into the two mtDNA clades within each sampling locality ($p < 0.01$). These two analyses give no, or very weak support to the

notion that there is reproductive isolation between members of the two major clades of mtDNA in Shark Bay. In addition, comprehensive paternity analyses including more than 305 individuals and behavioural observations suggest that 25% of successful reproduction was between mtDNA clades, which is not significantly different from the expected ($p = 0.52$).

Overall, our analyses overwhelmingly suggest that Shark Bay dolphins with both *aduncus* and *truncatus* haplotypes form part of a large, panmictic population. This contradicts previous findings stating that *T. aduncus* and *truncatus* form two separate species. Our results could be explained by incomplete lineage sorting between these two species at least in Western Australia, but they also emphasize that a collaborative effort in the Indo-Pacific region and worldwide is needed to resolve taxonomic issues in this family.

CONTRASTING PATTERNS OF MTDNA DIVERSITY AND MOLECULAR TAXONOMY OF CETACEANS

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The problems of species identification in cetaceans have been assisted enormously in recent years through the rapid advances in molecular taxonomy. One of these approaches has been the implementation of DNA-SURVEILLANCE, a web-based application that assists in the identification of species of unknown specimens of cetacean by aligning user-submitted DNA sequences with a validated and data set of reference sequences under curation by species-specialists. This program takes a phylogenetic approach to species identification, using as a criterion high bootstrap support for a monophyletic grouping with reference sequences from one species. Within the cetaceans, certain groups (notably the baleen and beaked whales) provide clear and strongly supported species identification, because of the relatively low diversity within species, and the relatively deep phylogenetic divergences between species. However, other groups, such as some Delphinidae, have proven more difficult, due to high within-species diversity, and relatively shallow divergences between species (e.g., Dizon et al 1999). This pattern leads to apparent paraphyly among some species for mtDNA lineages (i.e., mtDNA sequences do not form strongly supported, reciprocally monophyletic clades, concordant with accepted species-level organismal taxonomy). To evaluate the extent of this problem in cetaceans, a validation exercise for DNA-SURVEILLANCE was undertaken by submitting an unaligned copy of each reference sequence as a query to each data set in which it occurs. This exercise has highlighted the species for which paraphyly is a problem, given the current reference database. Does this paraphyly truly exist, or is it due to inadequately resolving genetic markers, or even mistaken morphological identification? How does under-represented geographical sampling of populations complicate these issues? We attempt to clarify the apparent paraphyly in the framework of "congeneric phylogeography" (Funk & Omland 2003) and consider ways to improve identification of these species.

SPECIES: HARD TO DEFINE, BUT WE KNOW IT WHEN WE SEE IT…. OR DO WE?

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Evolutionary biologists have long debated and discussed the relative merits of different species concepts, such as biological, phylogenetic or evolutionary species. Considerations of their relative applicability, objectivity, and relevance to the evolutionary process all (should) enter into serious evaluations of the concepts. However, these issues are only infrequently considered in studies where species boundaries are actually delimited and defined, especially in genetic studies of cetaceans. There have been numerous studies that have presented protocols for diagnosing species, but none are applicable for all situations or scenarios. In this presentation, an attempt will be made to consider some of the relationships among species concepts, data characteristics, and evolutionary processes. Some guidelines for interpreting data for decisions about species status will be presented.

PHYLOGEOGRAPHY OF THE ATLANTIC SPOTTED DOPLHIN (*STENELLA FRONTALIS)*

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The Atlantic spotted dolphin, *Stenella frontalis* is a small delphinid that is endemic to the tropical and warm temperate Atlantic Ocean. Morphological variation has been documented for this species over its entire range but assessments of genetic variation have been restricted to dolphins from the Gulf of Mexico and the Western coast of United States. Other researchers have previously found significant genetic differentiation among dolphins from the Gulf of Mexico and two North Western Atlantic regions for mitochondrial DNA (mtDNA) control region sequences and microsatellites. Differences between these markers suggest that further investigation is needed. The goal of our study is to analyze historical phylogeographical patterns and population structure across the entire species distribution using control region variation. Sequences (390 bp) from 60 Atlantic Spotted dolphins were compared and analyzed among the following putative populations: Gulf of Mexico (18), North Western Atlantic (12), the Caribbean (5), and West Africa (Canary Islands) (25). Results show 29 unique haplotypes; 9 haplotypes were found in the Gulf of Mexico samples, 7 in the North Western Atlantic, 4 in the Caribbean and 18 in West Africa. Gene diversity was high in each of these regions. Overall F-st values (based on haplotype frequencies) were low but significantly greater than zero. Analyses of population pairs showed that this was primarily due to the West African population, which had the largest sample size. Analyses with Phi-st (based on haplotype frequencies and genetic distances between haplotypes) only found significant differentiation between the Gulf of Mexico and West Africa. Similarly, an exact test showed significant genetic differences among populations ($p<0.0001$), due to divergence of the Gulf of Mexico from the North Western Atlantic ($p<0.05$) and West African populations ($p<0.0001$). Patterns of subtle but statistically significant genetic differentiation are concordant with previously published morphological analyses in the same regions. The absence of obvious genetic structure among populations suggests recent mixing (contemporary gene flow) and/or moderate to high levels of historical gene flow. Definitive conclusions about the historical and contemporary status of Atlantic Spotted Dolphin populations will require additional studies with genealogically informative nuclear markers.

POPULATION FORECASTING AND CONSERVATION UNIT DEFINITION IN CETACEANS

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The aim of conservation science is to forecast the likely effect of different threats and management scenarios in a usually patchy environment. As part of this effort, debate has raged about the best way to define conservation units. The common themes sought by all proponents are the definition of separate contributions to biodiversity (species, ESUs etc), and demographically independent populations (MUs, etc). Separate contributions to biodiversity can be discerned by phylogenetic analyses, although cutoff criteria are debated hotly. Demographic independence can also be analysed by genetic methods, and again it is possible to use cutoff criteria such as significant differentiation of genetic markers. However, depending upon the number of loci and individuals analysed, significant differentiation can be identified even when there is considerable gene flow between areas, as we found in *Tursiops* sp in Shark Bay, WA. In this case, application of the criterion of significant genetic differentiation could lead to erroneous definition of separate MUs. Thus it is more meaningful to use genetic studies to establish levels of exchange between areas, then use these levels in a metapopulation model for forecasting the likely outcome of different threats and management scenarios. In cetaceans, often such models rely on partial data, or even data from elephants or humans. There is no substitute for large observational data sets, but genetic analyses can assist with determining sex and variation of reproductive output. A model of the Shark Bay dolphin metapopulation was constructed using genetic data and long-term observational data. This model was then used to assess the likely effect of different threatening scenarios in this World Heritage Area.

SOME OBSERVATIONS UPON THE SYSTEMATICS OF TURSIOPS

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There has been a growing feeling amongst systematists that the genus Tursiops in the North Atlantic exhibits too much diversity for a single species, and initially an offshore form, Tursiops truncatus (Montagu 1821) has been defined (Mead and Potter 1995). The earlier synonym, Tursiops nesarnak was suppressed by the International Commission on Zoological Nomenclature in Opinion 1413 (1986). Delphinus tursio obtusus, Schlegel 1862 and Tursiops parvimanus, Van Beneden 1886 are considered junior synonyms of Tursiops truncatus (Montagu 1821). Tursiops truncatus ponticus, Barabash-Nikiforov 1940 is considered a valid subspecies of Tursiops truncatus (Montagu 1821).

This leaves Tursiops erebennus, (Cope 1865) and Tursiops subridens, True 1884 as available for the coastal form. Tursiops erebennus has no skull and is currently indistinguishable from Tursiops truncatus (Montagu 1821).

Tursiops aduncus (Ehrenberg 1832) has received considerable usage for some forms of Tursiops in the Indian Ocean, particularly animals with dark spots on their belly. Hershkovitz (1966:51) recognizes T. aduncus as a subspecies of Tursiops truncatus (Montagu 1821) and says the name was based on a stranded animal, that was not recovered, from Belhosse Island, Red Sea. Ehrenberg (1832) gives Belhosse Island as the type locality, but does not say in which part of the Red Sea it was. Ehrenberg does not say whether the type was recovered and most researches, following Hershkovitz, have operated as though the species did not have a holotype. We did extensive historical research and came up with Stresemann (1954) who did a narrative of Hemprich and Ehrenberg's expedition to the Red Sea which enable us to pinpoint Belhosse Island in the Dahlak Archipelago, Eritrea. That locality is a maze of islands and probably the kind of habitat that supports differentiated regional populations of Tursiops that can be diagnosed from the forms that occur elsewhere in the Indian Ocean. We think that Tursiops has differentiated in the open Indian Ocean into a variety of forms, all of which are different from Tursiops aduncus.

LISTEN TO MY SONG:
BIOACOUSTICAL INSIGHTS INTO CETACEAN TAXONOMY

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In many terrestrial species, vocalizations are the predominant means by which individuals communicate; species-level and regional variation is well known and it is not uncommon for song to be a powerful isolating mechanism among taxa. Due to recent advances in bioacoustics, vocal differentiation has become increasingly important in the taxonomy of terrestrial species and the structure of vocalizations is becoming common in the descriptions of new species of birds, mammals, amphibians and insects. Bioacoustics may be particularly useful in cetacean taxonomy because the recording of vocalizations offers a relatively cost-effective means of obtaining samples of sufficient size for analysis. In addition, traditional markers, such as those from morphology or genetics, may fail to detect taxonomic structure because they may evolve too slowly or because there are insufficient data. Vocalizations, which are social signals, may evolve more rapidly and may be particularly useful in detecting cryptic, insipient and sibling cetacean species. To assess the utility of bioacoustics in cetacean taxonomy, we summarize species in which acoustic characters are diagnostic at the species and subspecific level and examine concordance with known genetic and morphological data. We illustrate how distinct regional differences in baleen whale song, with examples from minke and blue whales, can provide taxonomic insights. We suggest that integrating data from multiple data sets, including bioacoustical markers, will make the strongest inferences about cetacean taxonomy. Furthermore, we recommend that when there are a lack of other data, or lack of clarity in other data sets, evidence of stable, diagnostic acoustical differences be examined for its utility as a provisional hypothesis of taxonomic structure. Regional differences in vocalizations can also be used to generate testable phylogenetic and phylogeographic hypotheses, thus focusing attention on geographic areas requiring additional data to resolve uncertainties in structure. When taxonomic decisions are important in conservation, it is prudent to listen to what the whales are saying.

INCORRECT AND UNRESOLVED PHYLOGENIES FROM MTDNA: CAUTIONS FOR RELYING ON ONE GENETIC MARKER FOR SPECIES DESIGNATIONS

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Mitochondrial DNA has been used extensively for phylogenetic and phylogeographic inference. Although it has often been hailed as the ideal marker for such studies because of maternal inheritance, small effective population size, and high mutation rates, problems with resolution, homoplasy, and lineage sorting can still result in incorrect inferences of taxonomic relationships. A recent review has shown that species level mitochondrial paraphyly and polyphyly are commonly reported, resulting primarily from undersampling of congeneric species variation. Even when species are adequately sampled, however, incorrect inferences can be drawn from a single locus phylogeny. Although technically more challenging, advances in methods for screening multiple nuclear loci are beginning to show that very close relationships can be resolved when >20-30 variable loci are used to study intraspecific and interspecific variation.

A REVIEW OF CETACEAN TAXONOMY AT THE SPECIES AND SUBSPECIES LEVELS

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The list of nine cetacean species described by Linnaeus in 1758 expanded greatly during Victorian times to several hundred. 20th-Century taxonomy added only a few species and pared the total to about 84. About 40 subspecies have been recognized in recent lists. Many unresolved problems remain, at both the species and subspecies levels. Some conservation-related reasons for ensuring that cetacean taxonomy is accurate and up-to-date are 1) management of exploitation for sustainability requires valid definition of population units, usually infraspecific stocks but sometimes species, 2) effective management of international trade requires codification of the distinctiveness of species and subspecific entities, 3) nomenclature affects conservation priorities, e.g., formally naming a subspecies focuses legal attention. Cetacean taxonomy has suffered from the piece-meal approach that is a natural consequence of variation in scientific interest in and economic importance of different taxa; an example is the attention given to subspecies in dolphins involved in the economically important tuna fishery of the eastern tropical Pacific, which has produced thousands of specimens for study. Another relevant factor is the debate over species concepts, exemplified in the current controversy about whether there are three species of right whales or only a single species with three subspecies. There is no consensus on necessary or sufficient criteria for subspecific status; many potential subspecies remain undescribed. Species groups for which unsettled but important issues remain include the right whales (Balaenidae), several of the rorquals (blue, Bryde's, minke; Balaenopteridae), the dwarf sperm whale (Kogiidae), the Amazon river dolphins (Iniidae), the South Asian river dolphins (Platanistidae), Cuvier's beaked whale (Ziphiidae), the finless and harbor porpoises (Phocoenidae), a number of dolphins (Hector's, common, dusky, Irrawaddy, tucuxi, humpbacked, spinner, bottlenose; Delphinidae), and pilot and killer whales (Delphinidae). Unresolved but less important issues also exist for a long list of other taxa. Of particular importance internationally are questions relating to minke whales, bottlenose dolphins, and humpbacked dolphins; and killer whales, of course, are currently of great interest in the US. A number of things could be done to increase the rate of progress in cetacean taxonomy, most relating to logistical issues surrounding examination of specimens and molecular approaches. Attempts should be made to reach consensus on the preferred species concept and on morphological and molecular criteria for subspecific status.

ENFRANCHISING POPULATIONS UNDER THE ESA: THE LEGAL DEFINITION OF "SPECIES"

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Congress modified the Endangered Species Act in 1978 allowing distinct population segments of vertebrate species to be listed as threatened or endangered "species." The term "distinct population segment" is a legal term created by Congress that has no direct counterpart in the scientific literature. A legal analysis of the ESA and its legislative history indicates that Congress used this term to create a new protected class of "species" based on both scientific principles and policy objectives. These principles and objectives are designed to protect imperiled populations where they are biologically important to the fitness of the taxon to which the population belongs; to protect populations that are important to the ecosystem to which the population belongs; and to protect populations that are important to the Nation and its people. However, policies published by the National Marine Fisheries Service and the United States Fish and Wildlife Service have attempted to define the term "distinct population segment" as coextensive with existing biological concepts such as the "evolutionary significant unit." These definitions essentially provide for the protection of populations only where it can be shown that the population is biologically important to its taxon. We present information indicating that these policies are inconsistent with the statute and Congressional intent because they effectively prohibit the protection of distinct, imperiled, vertebrate populations where the protection of those populations would advance Congress' other policy objectives. This poster explains the limitations of these policies, and suggests provisions for a policy more closely matching Congressional intent.

COMPOSITE SPECIES-LEVEL CETACEAN PHYLOGENY

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I present a species-level phylogeny of all 78 extant cetaceans (as listed in Mammal Species of the World. Wilson and Reeder, 1993). It was built by combining 51 cetacean phylogenies from both morphological and molecular data published since 1960. To combine such disparate data a supertree construction technique known as Matrix Representation using Parsimony (MRP) was used. This combination of previously published trees effectively represents the weight of the phylogenetic evidence in the literature. The resulting topology does not support the claim that the Odontoceti are paraphyletic; the Physeteridae are placed at the base of the odontocete clade. Two traditional families are not resolved within the supertree: the Balaenopteridae is found to be paraphyletic and the Platanistidae is found to be polyphyletic. The Platanistidae is split in two with the Platanista distinct from the clade containing Lipotes, Pontoporia and Inia, while Eschrichtiidae is nested within Balaenopteridae. Where there is little, or contradictory, phylogenetic information a polytomy results. Hence the presence of polytomies strongly indicates the need for further phylogenetic study. For example, 65% of all cetaceans are either ziphiids or delphinids and yet only 15 source trees included more than 3 species of either family. It is therefore not surprising that the Ziphidae appear as a single polytomy within the supertree and that the Delphinidae is poorly resolved. The distribution of polytomies within the tree parallels the distribution of uncategorised and data deficient species within the IUCN RedList of threatened species. It is these taxa that need to be the focus for future systematic study.

EVOLUTIONARY RELATIONSHIPS AND SPECIES IDENTIFICATION WITHIN THE DELPHININAE: A COMPARISON OF SINGLE LOCUS SEQUENCE DATA AND MULTILOCUS ANONYMOUS NUCLEAR MARKERS

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The Family Delphinidae contains approximately 36 species of small cetaceans. The subfamily Delphininae continues to be a common subgroup within the Delphinidae, although its members are variant across different morphological and genetic studies. However, all studies to date have included within the Delphininae all members of the genera *Tursiops*, *Delphinus*, and *Stenella*. Species from all three of these genera are present along the US east coast and Gulf of Mexico. In a variety of cases, identification of beachcast and/or fishery caught animals to species can be difficult - color patterns fade, or degradation is so severe that identifying features are missing - and certain species pose more of a problem than others. Proper identification of individual strandings, bycatch and even of individuals sampled at sea is critical for effective management. Thus, we sought to improve molecular identification of these taxa, quantify the within species variability and re-examine phylogenetic relationships among the taxa through the use of mitochondrial DNA control region sequence data and anonymous polymorphic nuclear markers. We obtained 360 bp of sequence from the hypervariable 5' end of the control region from approximately 350 individuals of 7 species (5 *Stenella*, *Tursiops truncatus*, *Delphinus delphis*) from the western North Atlantic and collected data for anonymous polymorphic nuclear markers from 144 individuals of the same species using the AFLP technique. Unlike other groups of Cetacea, the control region sequences do not appear to provide a robust method for phylogenetic species identifications or reconstructions within the Delphininae. The gene region exhibits a high degree of genetic variability within all species coupled with very low variability among species. Phylogenetic reconstructions maintained most species in monophyletic groups. *S. coeruleoalba* proved to be most problematic, never forming a monophyletic group in any tree. Proper taxon sampling played an important role in the maintenance of species' monophyly. However, bootstrap analyses rendered resolution of relationships among taxa as well as monophyly of most taxa nearly nonexistent. Combining phylogenetic identification with other methods, near exact match or fixed-site comparisons, helped with identification of some unknowns but not all. In contrast, an analysis of 410 polymorphic anonymous nuclear (AFLP) markers yielded a topology in which species were monophyletic with strong bootstrap support. In addition, relationships among some species were also well supported. The ability of the AFLP technique to provide hundreds of loci, compared to a single mtDNA locus, suggests that this method may provide both more robust phylogenies for this difficult set of taxa and also provide a better method of species identification than mitochondrial DNA sequences.

A RIGOROUS FRAMEWORK FOR ASSESSING MOLECULAR GENETIC CHARACTERS AS INDICATORS OF SPECIES LEVEL CONSERVATION UNITS

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The analysis of genetic data for the purpose of identifying conservation priorities may draw from one or more biological sub-disciplines depending on the specific questions for management. The fields of molecular systematics and population genetics have been instrumental in contributing relevant data to conservation biology. Yet the interpretations of character data are not always straightforward, particularly related to questions concerning taxonomy at the species boundary. For many studies on cetaceans (and other taxa), analyses of molecular character distributions occur using some combination of tree-based reconstruction analyses with reciprocal monophyly being accepted for defining units of conservation. These tree-based approaches and the principal of reciprocal monophyly have also been used for determining species and population origin from unknown samples (i.e. forensic analysis). The ability to clearly discriminate species from population level differences for cetacean conservation and management requires a complimentary approach integrating concepts and approaches from systematics and population genetics. Using a diagnostic character approach combined with population genetic and phylogenetic methods applied to molecular data provides the most objective approach to evaluating differentiation at the species level, particularly in the absence of obvious morphological differences. While character fixation may be an objective way for delimiting species boundaries under the operational approach of the Phylogenetic Species Concept, it is essential that analyses consider multiple sources of character data (i.e. multiple gene regions, morphology, and behavior) and have adequate sampling in order to have the maximum confidence for management decision-making. Our approach is demonstrated using several case studies of cetaceans, including a group of large migratory whales (genus *Eubalaena*), a group of coastally restricted small cetaceans (genus *Sousa*), as well as other cetaceans where species designations have been controversial or complex.

GENETIC STRUCTURE AND DIFFERENTIATION OF *TURSIOPS TRUNCATUS* ECOTYPES IN THE GULF OF CALIFORNIA

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A high variability has been found among bottlenose dolphin Tursiops truncatus population worldwide. Historically, even though morphological polymorphisms have lead to the description of 20 nominal species of Tursiops, only two are considered valid. In the Gulf of California, studies on the genus Tursiops are still scarce, however, it is evident that morphological differences do exist; for instance the existence of two ecotypes (coastal and oceanic) has been suggested based on phenotypic characters. However, the extent to which this phenotypic differentiation is genetically encoded is unknown. In this study we hypothesize that T. truncatus within the GC is genetically structured to reflect the patterns of morphological variation. The genetic differentiation of ecotypes of T. truncatus in the Gulf of California is imperative due to the increasing interest in the management and conservation of the species. In this study, 58 individuals from different regions of the gulf were genotyped using sequences of mitochondrial DNA control region. Here we present levels of genetic diversity and analysis of genetic structure and phylogeographic patterns found in the mtDNA haplotypes. The results suggest that individuals of T. truncatus from the upper Gulf of California conform a distinct group from the rest and significant differences were found between coastal and oceanic ecotypes.

TRUMPING THE NEED FOR TAXONOMIC RESOLUTION WITH EFFECTIVE CONSERVATION ACTIONS: THE CASE OF GREEN/BLACK SEA TURTLES IN THE EASTERN PACIFIC OCEAN

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Solving current taxonomic questions about endangered wildlife populations can guide management decisions and help determine the relevance of national laws and international treaties that mandate conservation action. However, the need for such information is less urgent when effective conservation measures are already in place and threats are known. In the case of green turtles in the Eastern Pacific Ocean (a.k.a., east Pacific green turtles, black turtles), tremendous declines have occurred due to overexploitation of eggs and turtles as a food resource and, to a lesser extent, incidental mortality relating to marine fisheries and degradation of marine and nesting habitats. Apart from these conservation issues, there has been considerable debate regarding taxonomic status of green turtles in the Eastern Pacific: various arguments defend full species, subspecies, as well as unique population designations. Interestingly, many researchers believe that full species status will focus more international attention on this imperiled green turtle stock. Although this may be true, we believe that taxonomic resolution will do little to augment existing on-the-ground conservation programs. For years, such efforts have been underway at both nesting beaches and foraging areas. This multifaceted approach enlists scientists, resource managers, policy makers, fishers, and volunteers and has contributed, at least in part, to the early signs of recovery for this stock. Therefore, while reaching consensus on taxonomic status may be a worthwhile scientific exercise, the importance of such information will have comparatively less conservation relevance. A more efficacious outlet for scientific inquiry may be the ecological role of these animals, and the changes resulting from their declines.

PRESENT CONSERVATION AND MANAGEMENT STATUS OF THE FRANCISCANA (PONTOPORIA BLAINVILLEI) ALONG THE BRAZILIAN COAST AND THE TAXONOMIC STATUS OF ITS DISCONTINOUS POPULATIONS

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The franciscana (Pontoporia blainvillei) is restricted to coastal waters of the Western South Atlantic Ocean. The species presents two ecotypes separated by morphological and genetic characteristics: the northern one, between 20ºS and 27ºS, and the southern one, between 30ºS and 40ºS. The former presents lower genetic diversity and is particularly susceptible to decline, in relation to the latter. Isolated populations of this species along southeastern Brazil causes special concern about its taxonomic status and conservation needs. The possible ecological factors that could explain the occurrence of isolated populations of this species along Southeastern Brazil include: river discharge/food resources; protection against predators; maintenance of the water temperature; ocean floor morphology,-especially depth; presence of predators and trophic competitors. The crude separation of franciscana into northern and southern genetic populations is unlikely to be sufficient for effective conservation of this species. Because the threats to the species are likely to occur at different levels along its distribution, it is important to determine stock discreteness in order to guide conservation and management procedures on a local level. The Brazilian Government and Fiocruz are elaborating a National Management Plan for the franciscana. In the near future, the plan will be implemented as a National Recovery plan for the franciscana in order to save the fransciscana from extinction.

VARIATION IN CALLING BEHAVIOR IN NORTHEASTERN PACIFIC BLUE WHALES (*BALAENOPTERA MUSCULUS*)

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Blue whales produce geographically distinct stereotypic call types that may be used to provisionally identify different stocks and/or subspecies. In the North Pacific, blue whales produce two call types, one each from the eastern and western sides of the basin. These two call types sometimes overlap in time and space in the central North Pacific and the Gulf of Alaska. The western call type is recorded from the Gulf of Alaska westward along the Aleutian Islands. This call type is comprised of units whose fundamental frequency is 21 Hz (max) to 18 Hz (min), each lasting ~23 s. The eastern call type is recorded from the Gulf of Alaska southwards along North and Central America to the equator. This call type is comprised of combinations of two units, commonly referred to as 'A' and 'B' calls, whose fundamental frequencies are 18 Hz (max) to 15 Hz (min), each lasting ~17 s. Close visual examination of spectrograms from hydrophones in the Gulf of Alaska and the eastern tropical Pacific (ETP) resulted in the detection of anomalies in these call types. First, calls that are truncated in time and/or expanded in frequency were noted in data from the ETP. Second, series of a previously undescribed call type were regularly detected. This call type's characteristics are intermediate of the eastern and western call types in both time and frequency. Lastly, on one occasion a single animal produced a call series that included units from both eastern and western call types. In this instance, a calling whale substituted a western unit for the eastern A-unit which was followed by B-units that had intermediate characteristics. This suggests that at least one individual is capable of producing both call types and that blue whale calling behavior may be somewhat plastic. In areas where different call types overlap, acoustic data alone may not be a foolproof indicator of stock identification. Despite these anomalies, however, the vast majority of blue whale calls detected in the Gulf of Alaska and the ETP were distinct, stereotyped geographic variants.

HOW THE HOLES IN CETACEAN TAXONOMY ARE AFFECTING CONSERVATION AND MANAGEMENT

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Understanding the structure of cetaceans at the species, sub-species and evolutionarily significant unit (ESU) levels (all subsumed in the discipline of systematics) is pivotal to global conservation efforts. Cetaceans present special difficulties for attaining data needed for taxonomy: species live in remote waters, many skulls are too large for routine museum collecting, most animals sink when dead and cannot for the most part be killed for collections. Consequently, taxonomic uncertainty is greater than for most terrestrial species and much uncertainty can be expected to remain in the foreseeable future. Conservation risks are high and increasing, particularly in coastal waters or rivers adjacent to high human densities. For cetaceans to receive conservation protections equivalent to terrestrial species, taxonomists must devise appropriate methods to function despite uncertainty. I illustrate how conservation efforts are stymied by uncertainties in taxonomy using examples: killer whales, river dolphins and Hector's dolphins. I focus on the importance of incorporating measures of uncertainty into taxonomy to allow managers to shift the burden of proof and to choose levels of precaution commensurate with other species that have different quantities of data available. I suggest that taxonomists can benefit from methods developed in other fields of conservation that utilize default criteria when data are lacking and use lower standards of proof when ignorance is greater.

CONSERVATION STATUS OF THE BLACK SEA BOTTLENOSE DOLPHIN (TURSIOPS TRUNCATUS PONTICUS): AN ASSESSMENT USING MORPHOLOGY AND GENETIC VARIATION

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The bottlenose dolphin (*Tursiops truncatus*) is one of three species of cetaceans living in the Azov-Black Sea basin. Until 1966, Black Sea cetaceans were mainly threatened by dolphin fisheries. Since then, anthropogenic impacts from pollution, diminishing food resources, live catches, diseases and physical injuries have killed more than 5 million Black Sea cetaceans. Despite many studies of bottlenose dolphins elsewhere, data on Black Sea populations are scarce. Thus, the overall status of Black Sea bottlenose dolphins is unclear and previous attempts to protect them have failed. In order to adopt international agreements and implement specific management plans that will protect Black Sea cetaceans, the first steps are to scientifically demonstrate that they are isolated from Mediterranean populations, and establish whether they are evolutionarily unique. The aim of this study is to estimate the degree of morphological distinctiveness and genetic isolation of Black Sea bottlenose dolphins from Mediterranean populations for conservation purposes. Cooperation with researchers from countries surrounding the Black Sea facilitated access to genetic material and skulls for morphological analysis. Seventy-four bottlenose dolphin skulls from the Black Sea (27), the Mediterranean Sea (27) and the Atlantic Ocean (20) were sampled for 31 cranial measurements. Mitochondrial DNA (mtDNA) variation in 109 bottlenose dolphins was compared among the same localities, which were analyzed as putative populations. Results from 442 base pairs of control region show low haplotype diversity for the Black Sea population: although 33 unique haplotypes were identified, only six were found in 43 Black Sea individuals. Five haplotypes were shared among regions among which 2 Black Sea haplotypes were shared with Mediterranean Samples. Φst (which quantify differences among populations) was significantly greater than zero for all Black Sea pairwise comparisons. An exact test showed significant differences among populations, and pairwise exact tests showed that the Black Sea is different from all other populations. The haplotype network demonstrates the isolation of Black Sea haplotypes around a widespread ancestral allele. Gene flow is low enough to facilitate genetic differentiation, although there is evidence for at least one recent migrant. A principal component analysis performed on 31 skull variables indicates that size is the major discriminant component between populations. Black Sea bottlenose dolphins are significantly smaller than the contiguous Mediterranean populations. There is sufficient evidence to designate the Black Sea *Tursiops truncatus* a genetically and evolutionarily unique unit worthy of separate conservation.

APPLICATION OF ALTERNATIVE ESU CONCEPTS TO A COMMON PROBLEM IN APPLIED CONSERVATION BIOLOGY

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This talk considers the question, "How similar (or different) would the outcomes be if a variety of published methods were used to define conservation units in the same species?" In this case, the common problem is how to define ESUs of anadromous Pacific salmonids (*Oncorhynchus* spp). These seven species have already been the subject of a large-scale effort in applied conservation biology that has resulted in identification of over 50 ESUs in Northwestern North America, based on an approach developed by NMFS. This body of work provides a context for comparing the likely results of applying ESU approaches proposed by Dizon, Moritz, Vogler & DeSalle, Bowen, and Crandall. Each species of Pacific salmon exists as a hierarchically-structured complex of a large number of populations or stocks, and it appears that the diverse approaches for defining ESUs would likely result in an equally diverse range of outcomes. The NMFS approach in general has focused on units that are midway along the continuum of intraspecific biodiversity—that is, striking a balance between lumping and splitting. As a result, each salmon ESU typically contains 20-40 separate populations or stocks, with populations within ESUs showing more similarity in genetic, ecological, and life history features than populations in different ESUs. In contrast, it appears that the other published approaches would be more likely to recognize ESUs at one of the extremes of the continuum—either at or near the level of the local population or stock, or at or near the level of the taxonomic species. This exercise should provide some insights into the likely results of applying the different approaches to other taxa. In evaluating which approach is preferable in any given application, it is important to consider the likely outcomes in the context of the goals one is trying to achieve.

ADAPTIVE GENETIC VARIATION AND UNITS FOR CONSERVATION

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Evolutionarily significant units (ESUs) and management units (MUs) are discrete classifications that emphasize reproductive isolation over long and short evolutionary time periods, respectively. Previously, we have argued that this dichotomous summary poorly represents patterns of population differentiation and is not adequate for determining appropriate management actions. We have suggested an alternative approach to categorizing biodiversity based on evolutionary processes indicated by ecological and genetic exchangeability and described a management and classification scheme based on this approach. Here, I describe several real-life examples of the approach using case studies of several species. I outline possible methodologies for surveying adaptive variation in nature made possible by new molecular techniques and discuss examples from large carnivores that demonstrate how ecology rather than genetic isolation gives rise to genetic patterns.

RECOGNITION OF A NEW SPECIES *BALAENOPTERA OMURAI*

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A new balaenopterid species *Balaenoptera omurai* was recognized based on both morphological and molecular biological evidences. Existence of an undescribed balaenopterid species has been suspected since the early 1990's when Wada and Numachi (1991) drew attention on the 'small form Bryde's whales' . A medium-sized baleen whale carcass was collected on a coastal island in the Sea of Japan in September 1998. While examining this individual we realized several unique characters. Comparison of external morphology, osteology and mitochondrial DNA data grouped this specimen with the eight specimens collected in the late 70's in Solomon sea and the sea around Cocos island as a single species but separated them from all known baleen whale species. Description of a newly defined species *Balaenoptera omurai* was summarized in Wada, et al. (2003). We will introduce this new species with some additional data, for the character state for the diagnostic characters. Our morphological findings on the type specimen of *B. edeni* mainly on the bone configuration at the vertex in Indian Museum Colcata are described. We would also like to propose the necessity of defining a neotype of B. brydei, after relevant discussion will be made in the future. Finally, our conclusion includes the recognition of a new species *B. omurai* and separating *B. brydei* from *B. edeni* to make Bryde's whale complex more reasonably comprehensive, otherwise leaving this group as a confusing systematic entity.

Cetacea Brisson, 1762
Mysticeti Flower, 1864
Balaenopteridae Gray, 1864
Balaenoptera Lacépède, 1804
Balaenoptera omurai sp. nov.

Etymology. The specific name is in honour of the late H. Omura, a Japanese cetologist, for his contribution to the knowledge of Cetacea.
Holotype. Adult female, NSMT-M32505, National Science Museum, Tokyo. A complete skeleton, both complete baleen rows and frozen pieces of muscle, blubber and kidney were collected at Tsunoshima Island (34˚ 210˙ N, 130˚ 53˙) by T.K.Y., T. Kuramochi, M.O., E. Jibiki and S. Fujioka. The collection was undertaken three days after the accidental death of the animal.

Paratypes. Five females and three males, NRIFSF1–8, National Research Institute of Far Seas Fisheries, Fisheries Research Agency, Shizuoka. The longest baleen plate, an earplug and a piece of the sixth thoracic vertebra with epiphysis were collected by trained staff from each animal. NRIFSF6 includes 18 more baleen plates.
Locality. The Sea of Japan (type locality), the Solomon Sea and the eastern Indian Ocean near the Cocos Islands.
Diagnosis. *Balaenoptera omurai* differs from all of its congeners by having the following unique characters: medially expanded posterior portion of ascending process of maxilla, which conceals posterior end of premaxilla along the adjacent nasal; approximately 200 baleen plates on one si de, which is the smallest number among all of its congeners except for *B. edeni* (the baleen plate number of which is still unknown); 21 diagnostic sites in the complete mitochondrial (mt)DNA control region sequence.

MORPHOLOGICAL AND PHOTOMETRIC CHARACTERIZATION OF INTERGENERIC HYBRIDS BETWEEN TURSIOPS AND DELPHINUS, TURSIOPS AND PSEUDORCA

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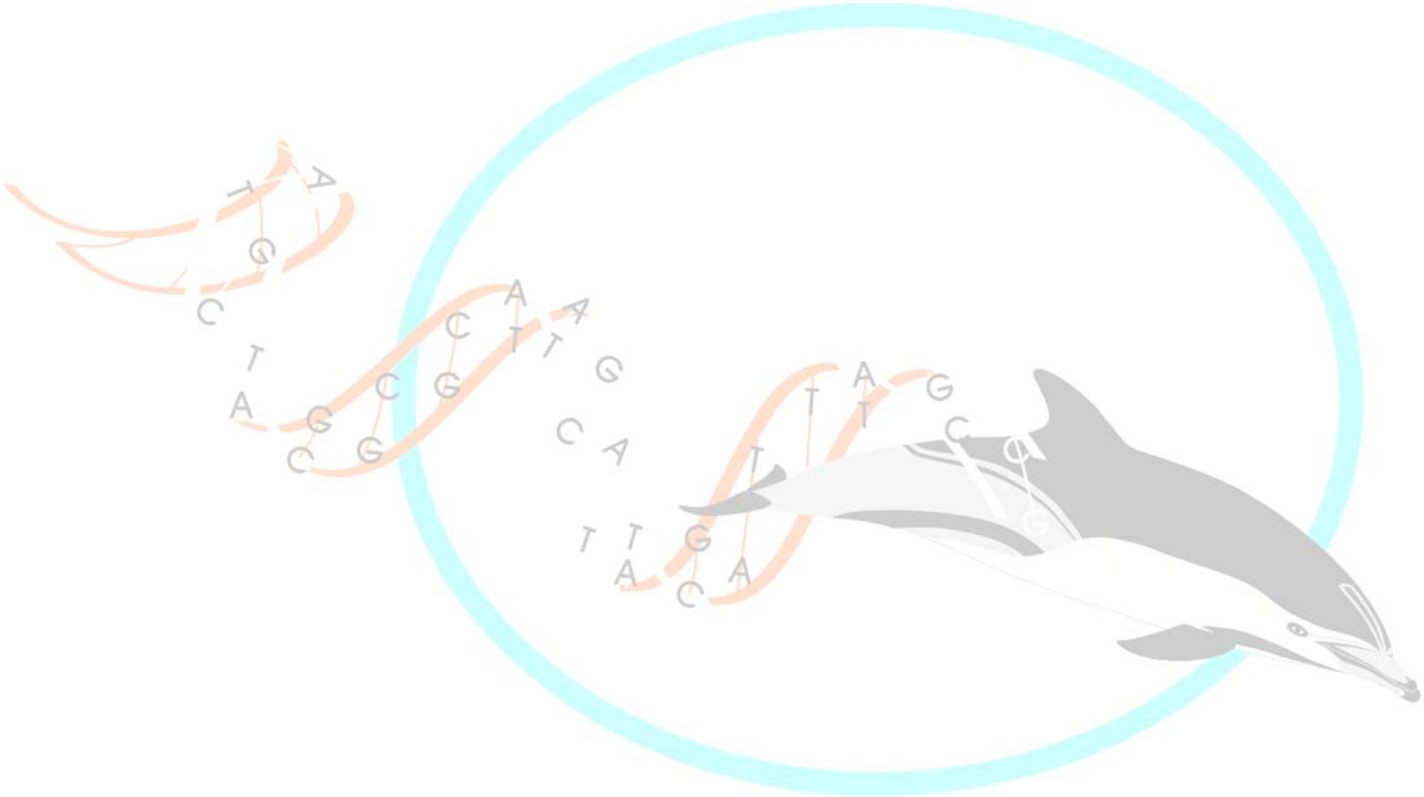
Putative and confirmed hybridization among wild or captive cetaceans has been reported involving a wide range of variously related genera within both the Odontoceti and the Mysticeti. There have been 26 captive intergeneric F_1 hybrids produced between *Tursiops* and either *Grampus*, *Pseudorca*, *Delphinus*, *Steno* or *Globicephala*. The hybridizations that have occurred in captivity make it possible to document growth and development, morphological and color pattern characteristics, vocalizations and fertility for the respective crosses. These events draw attention to the possibility of hybridization and introgression between these species in the wild. The existence of intergeneric hybrids in cetaceans also suggests that interspecific (within genera) introgression is possible and should be considered in regions of geographical overlap. We present the morphological and color pattern characteristics of two different intergeneric crosses, *Tursiops truncatus* x *Delphinus capensis* (one male F_1, one female F_1 and one female second generation backcross) and *Tursiops truncatus* x *Pseudorca crassidens* (two female F_1s and two female second generation backcrosses). As apparent from the backcrosses, F_1 females from both intergeneric hybridizations are fertile. Documenting the appearance and characteristics of these individual hybrids may help in identifying putative hybrids in the field, making it possible to begin an assessment of the frequency and distribution of natural hybridization among cetaceans in the wild.

❧ Notes ☙



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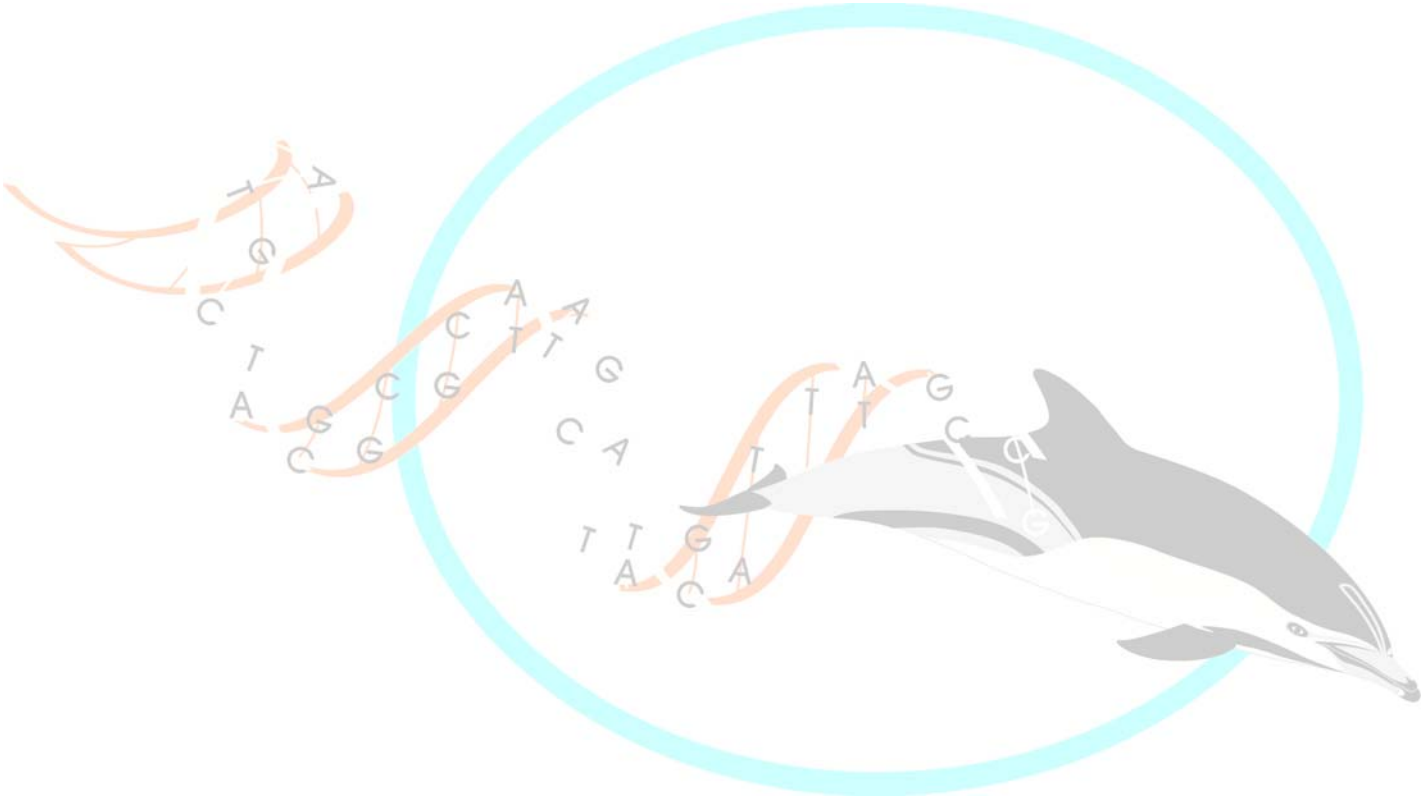

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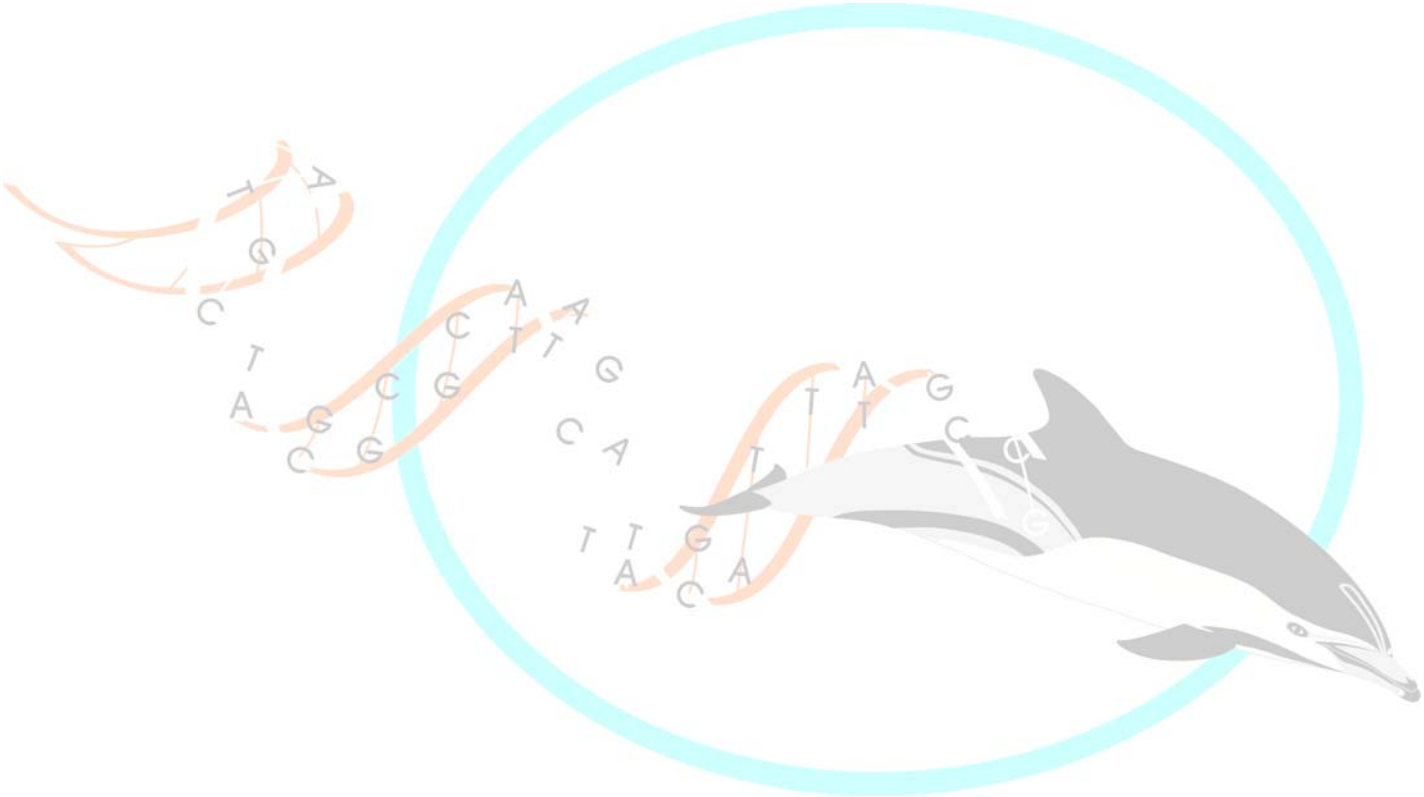

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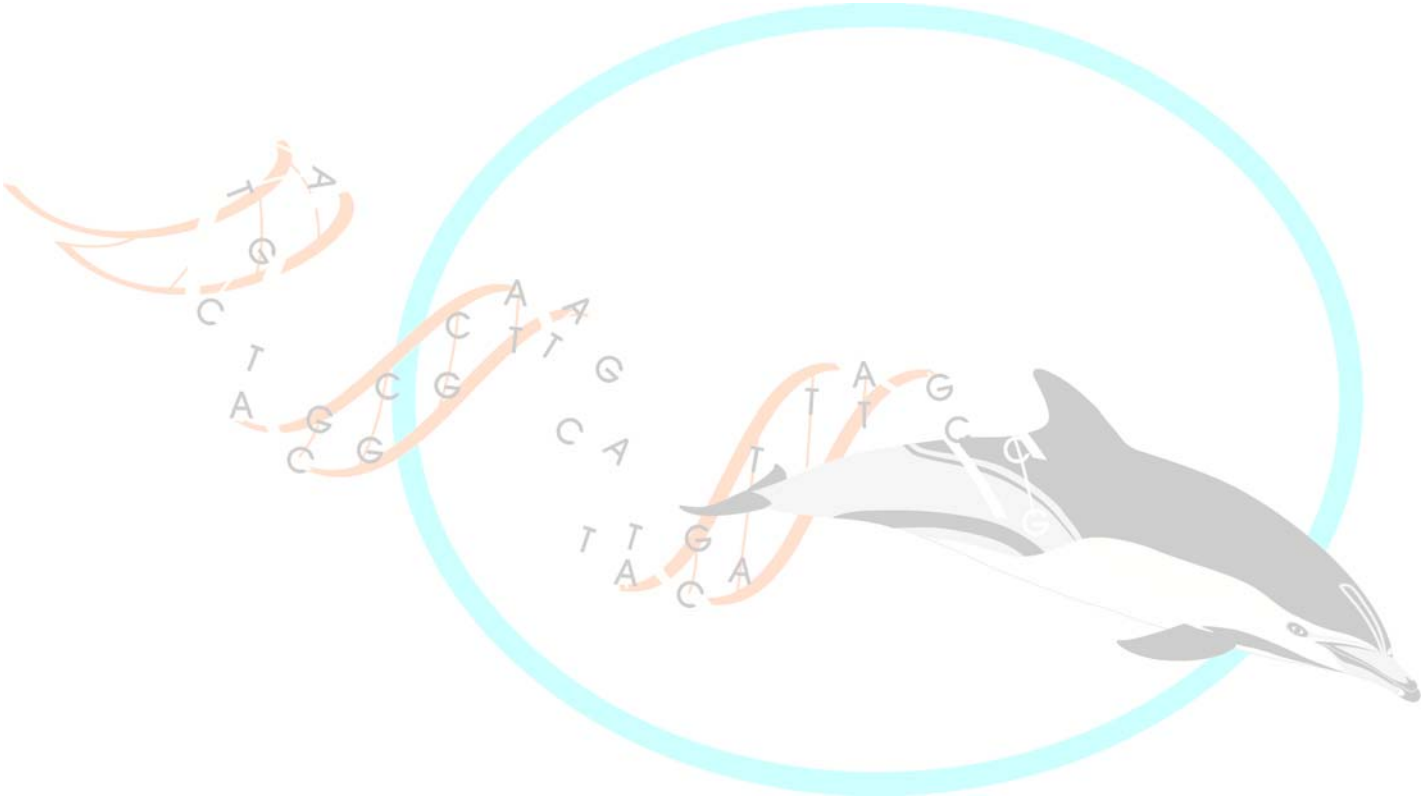
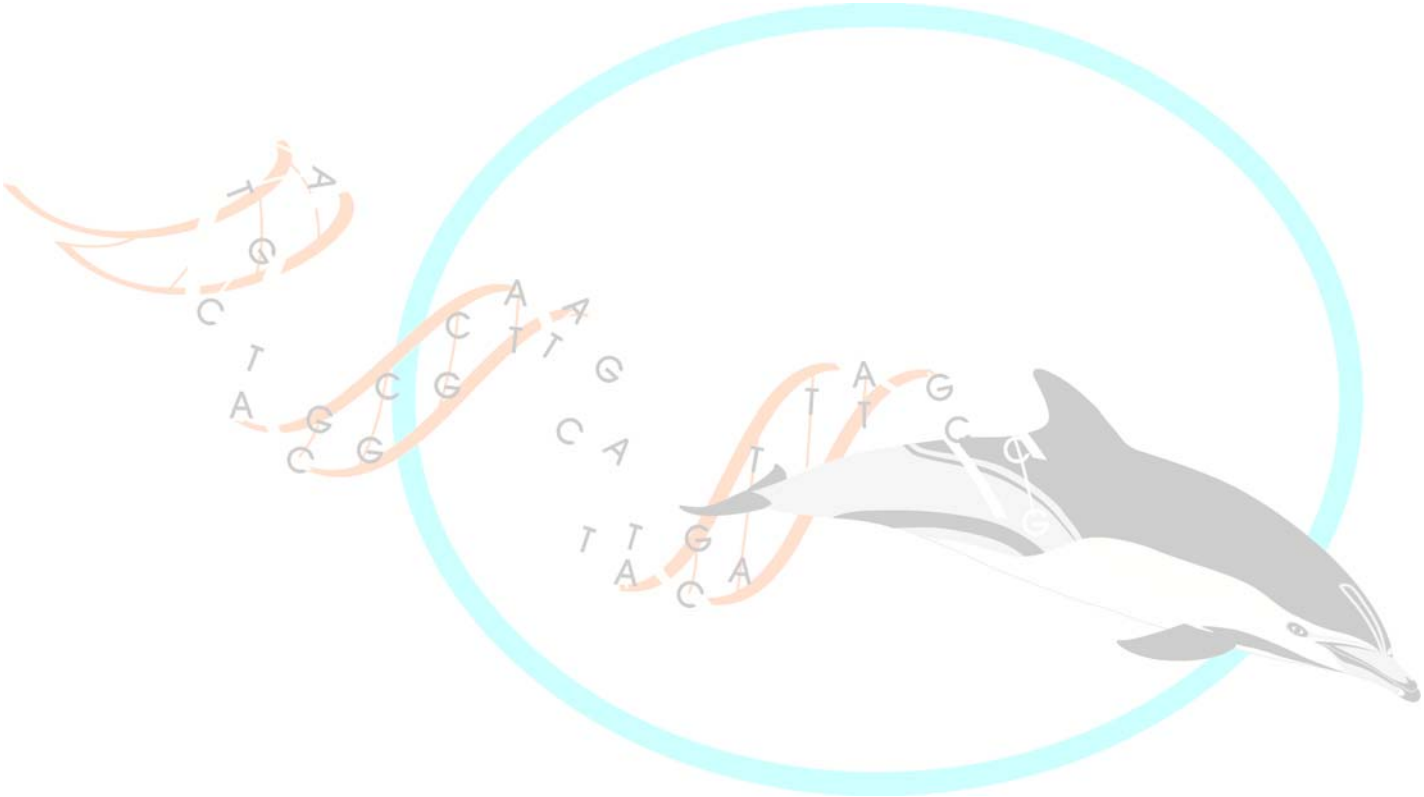
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