Identifying the Origin of the Marbled Murrelets Recovered from the Cosco Busan Oil Spill

Final Report

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Project Summary

Mortality from oil spills presents a major threat to Marbled Murrelet populations, especially in California where population declines have reduced the number of birds to less than 5,000 (McShane et al., 2004). To better assess the impact of oil spills and coordinate an effective response, it is important to understand the distribution and dispersal of Marbled Murrelets in California. We used genetic data from 13 microsatellites to assign three Marbled Murrelets, oiled from the Cosco Busan Oil Spill in the San Francisco Bay area, to their population of origin. All three murrelets were assigned to populations north of central California with assignment probabilities greater than 55%. This information may be useful for current and future oil spill mitigation and management of Marbled Murrelets in California.

Introduction

The Marbled Murrelet (*Brachyramphus marmoratus*) is a small, diving seabird that forages in the near-shore marine environment and lays a single egg in a nest in old-growth forests along the Pacific coast of North America. These birds are listed as threatened by the U.S. and Canada and are endangered in the state of California (USFWS, 1997, Piatt et al., 2007). Threats to Marbled Murrelet populations include harvesting of old-growth forests, gillnetting, declines in prey availability, increases in nest predators, and oil spills (Peery et al. 2004, Becker and Beissinger 2006, Peery et al. 2006, Becker et al. 2007). Like many alcids, Marbled Murrelets are particularly susceptible to oiling. Mortality from oil spills presents a major threat to murrelet populations, especially in California where population declines have reduced the number of birds to less than 5,000 (McShane et al., 2004). To better assess the impact of oil spills and coordinate an effective response, it is important to understand the distribution and dispersal of Marbled Murrelets in California.

The movements of Marbled Murrelets among populations are poorly understood due to difficulties in recapturing individuals and the limited attachment duration of radio-transmitters. Much of the information available on Marbled Murrelet dispersal is from observations of banded birds moving among populations. Marbled Murrelets in the southern portion of the range disperse during the non-breeding season (Peery et al., In press). Peery et al. (In press) observed radio-marked birds from central California traveled as far north as Mendocino County and as far south as San Luis Obispo County.

These long dispersal distances indicate that Marbled Murrelets from the northern and central California populations mix during the non-breeding season, making it difficult to create population specific impact assessments for oil spills and complicating mitigation efforts.

Genetic data may be useful for understanding distribution and dispersal of Marbled Murrelets in California. Range-wide genetic studies by Friesen et al. (2005) and Piatt et al. (2007) revealed significant genetic differentiation between central California and the northern populations of Marbled Murrelets. This genetic structure can be used to examine movement of individuals among regions. Because Marbled Murrelets in central California are genetically distinct from birds in the northern parts of the range, it is possible to assign individuals of unknown origin to either (1) central California or (2) the group of populations from Alaska to northern California based on their genotypes. The use of genetic data to assign individuals to their population of origin could help scientists better understand the movements of Marbled Murrelets in California and may allow scientists to separately assess the impact of oil spills on northern and central California Marbled Murrelet populations. Here, we report population assignments for three Marbled Murrelets that were recovered from the Cosco Busan Oil Spill, which occurred in San Francisco Bay in November 2007.

Methods

Three Marbled Murrelets were recovered dead and oiled near Point Reyes, California during the month of November 2007 (Table 1). The birds were frozen and stored at the Oiled Wildlife Care Network facility in Cordelia prior to sample collection. Liver tissue samples were collected from two of the murrelets. Toe skin was collected from the third murrelet because the carcass was heavily scavenged and mummified.

DNA was extracted from the liver tissue using a DNEasy Extraction Kit (Qiagen, Valencia, CA) following the manufacturer's protocol for animal tissues. The toe skin sample was soaked in 90% ethanol for 24 hours prior to DNA extraction. DNA was extracted from the toe skin using a phenol/chloroform extraction. DNA from each bird was sent to Zoogen (Davis, CA) for sexing.

For each bird, polymerase chain reaction (PCR) was used to amplify 13 microsatellite markers (BmaACCT555, BmaGACA456, BmaTATC371, BmaAGGT503,

BmaGGAT313, BmaCCAT301, BmaGGAT368, BmaCCAT443, BmaTATC356, GATA365, BmaGATA439, BmaTGAA523, BmaGATA553), developed by Rew et al. (2006). The forward or reverse primer was labeled with one of the following fluorescent dyes: FAM, HEX, PET, or NED (Applied Biosystems, Foster City, CA) for subsequent electrophoresis and scoring. PCRs were conducted in a 10 µl volume, in a GeneAmp 9700 thermalcycler (Applied Biosystems, Foster City, CA), using Qiagen's Core PCR kit, under the following conditions: approximately 10 ng of genomic DNA in a cocktail that included 1 µL of 10x buffer, 0.2 mM of each dNTP, 0.25 U *Taq* DNA polymerase, 0.5 µM of reverse primer, and 0.5 µM of forward primer. Cycling profiles for all markers except BmaGGAT313 were composed of: (1) a 3 minute initial denaturing step at 94°C; (2) 24-40 cycles of a 30 second denaturing step at 94°C, a 30 second annealing step at 59-62°C, and a 30 second extension step at 72°C; and (3) a 10 minute final extension step at 72 °C. Cycling for BmaGGAT313 was composed of: (1) a 3 minute initial denaturing step at 94°C; (2) 19-22 cycles of a 30 second denaturing step at 94°C, a 30 second annealing step at 59°C, and a 30 second extension step at 72°C; (3) 7-10 cycles of a 30 second denaturing step at 94°C, a 30 second annealing step at 53°C, and a 30 second extension step at 72°C; and (4) a 10 minute final extension step at 72 °C. Microsatellites were run with LIZ 500 size standard on an ABI3730 sequencer (Applied Biosystems, Foster City, CA) and scored using Genemapper software (Applied Biosystems, Foster City, CA).

Microsatellite data was used to calculate population assignments for each bird using Geneclass2 software (Piry et al. 2004). Geneclass2 uses a Bayesian statistical method to calculate genotype likelihoods generated from multi-locus genetic data to assign individuals to genetically distinct populations. Genotype likelihoods were calculated for the northern populations and central California using approximately 300 Marbled Murrelets from each region. The three oiled Marbled Murrelets were compared to the genotype likelihoods calculated for each region and assigned to either central California or a population north of central California based on their assignment probabilities. If birds had a less than 5% probability (α=0.05) of originating from the northern populations, they were classified as central California birds.

Results

All three Marbled Murrelets recovered from the Cosco Busan Oil Spill were assigned to a population north of central California (Table 2). Because Marbled Murrelets from southern Alaska through northern California represent one genetic stock, we were unable to identify which of the northern populations these birds originated from. However, we were able to exclude central California as the population of origin for the three Marbled Murrelets. The assignment probabilities for these birds from the northern populations were greater than 55% (Table 2), which is much greater than the 5% probability threshold for excluding them from the northern populations. Assignment probabilities ranging from 50% to 60% are typical for Marbled Murrelets that are residents of the population in which they were captured.

Because the carcass of bird D993 was heavily scavenged and mummified, the only tissue available for DNA extraction was toe skin. Although this tissue is suitable for DNA extraction, the DNA which is extracted from this tissue type is often highly degraded and difficult to amplify in PCR. We were able to successfully amplify 8 of the 13 microsatellite loci from this sample using PCR. Therefore, the population assignment for this individual is only based on 8 microsatellite loci. We do not believe using fewer loci affected the outcome of the population assignment analysis for this individual because the assignment probability for this bird was high (59.1%, Table 2).

Understanding dispersal patterns and identifying the population of origin for individuals of a highly mobile species can assist policy makers in creating population specific impact assessments and coordinating an effective response for oil spills. Like other alcids, Marbled Murrelets are threatened by oil pollution. We found that all three Marbled Murrelets oiled by the Cosco Busan Oil Spill originated from populations north of central California. This information may be useful for current and future oil spill mitigation and management of Marbled Murrelets in California.

Table 1. Collection information and morphological data for Marbled Murrelets (*Brachyramphus marmoratus*) collected dead and oiled from the Cosco Busan Oil Spill in November 2007.

Bird #	Collection Date	Collection Location	Sex	Age	% Oil	Scavenged	Culmen (mm)	Bill Depth (mm)	Wing (mm)	Tarsus (mm)
D060	11/17/2007	Point Reyes	Female	Unknown	100	N	14.6	6.2	126	18.5
D140	11/13/2007	Point Reyes	Male	AHY	30	N	14	5.8	134	18.2
D993	11/21/2007	Point Reyes	Female	AHY	10	Y	n/a	n/a	130	16.5

Table 2. Genetic population assignments for three Marbled Murrelets (*Brachyramphus marmoratus*) collected dead and oiled from the Cosco Busan Oil Spill in November 2007.

Bird #	Tissue Type	Population Assignment	Assignment Probability	# Loci Used
D060	Liver	Northern Populations	0.557	13
D140	Liver	Northern Populations	0.556	13
D993	Toe skin	Northern Populations	0.591	8

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