Primary stressors to the American badger in southern California include:

- 1. Road mortality
- 2. Habitat loss
- 3. Habitat fragmentation: Lack of open habitat and/or corridors for movement and dispersal.
- 4. Hunting and trapping: Predator control/ sport shooting/ fur trapping
- 5. Consumption of pesticides through small mammal prey

Methods

Defining Target Survey Locations

We obtained historical badger records as a map (pdf) from the San Diego Natural History Museum with verified historical locations and from museum records accessed from the Global Diversity Information Facility Data Portal (*http://data.gbif.org*) and the California Natural Diversity Database (CNDDB; California DFG http://www.dfg.ca.gov/biogeodata/cnddb/). To obtain more information, we collaborated with San Diego Monitoring and Management Program (SDMMP: Yvonne Moore) to send out a general call for any San Diego County badger sighting information to wildlife agencies, consultants, and land managers. Many individuals responded by sending locations and descriptions of badger sightings or sign to USGS. After individual interviews and reviews of evidence, we assigned confidence levels to each record. We created a target survey map in ArcGIS by overlaying historical and recent badger records (high and medium-high confidence only), San Diego conserved lands (2010; SANDAG) and Vegetation (1995; SANDAG).

We then created a target survey site list (Appendix 1) based upon

- 1. Areas with historical and/or recent badger records (2 points);
- 2. Conserved lands with priority given to MSCP and MHCP lands (2 points); and
- 3. Areas containing moderate to abundant grassland habitat (1 point).

Access permits were acquired from the County of San Diego, City of San Diego, City of Escondido, California Department of Fish and Game (San Diego and Riverside), Bureau of Land Management, Sweetwater Authority, U.S. Fish and Wildlife Service, San Diego City Water Authority, Vista Irrigation District, Marine Corps Base (MCB), Camp Pendleton, Fallbrook Naval Weapons Station (NWS), Army Corps of Engineers, and California State Parks.

Preliminary Sign Surveys

It is optimal to have local areas of known occupied badger habitat to reinforce the canine scent detection training, both prior to and during the overall canine scent survey period. Therefore, we contracted Barry Martin of the Western Tracking Institute (WTI) to conduct preliminary surveys for badger sign (i.e. tracks, scat, digs, burrows, trails) in historically occupied areas to identify potential training areas in or near San Diego County. Mr. Martin focused surveys on the Santa Rosa Plateau, which historically had abundant badger sign (C. Brehme pers. observation, Carol Bell pers. comm.). and also conducted rapid surveys of the Ramona grasslands, Mission Trails Regional Park, and Crestridge Ecological Reserve. The only badger sign found during the preliminary surveys were some tracks and digs on Burro Mesa in the Santa Rosa Plateau Ecological Reserve. No badger sign was found in other areas. Mr. Martin also provided training in badger sign identification.

Canine Scent Surveys

The Center for Conservation Biology (CCB, University of Washington) obtained American badger scat from the Washington Zoo. With this scat, CCB initially trained one detection dog "Pips" at their training facility in Eatonville, Washington, following the methods outlined in Wasser et al 2004.

"Dogs selected for the program were initially introduced to target species odor (scat) utilizing a scent box. The scent box is a $2 \text{ m} \times 30 \text{ cm} \times 30 \text{ cm}$ hinged rectangle with five compartments open to the outside by a 5-cm hole. Scat is placed in one of the five compartments. The search is initiated by the verbal command "find it". The dog is guided to investigate each compartment of the scent box and encouraged to smell at the hole openings. Initially, the "find it" command is verbalized between each hole. Upon sniffing the hole containing the sample, the dog is immediately rewarded with a well-timed toss of a tennis ball across its visual field followed by verbal praise and ~90 s of play. The dog quickly learns to associate sample detection with the reinforcement of the reward. This maintains a strong motivation level for these high play drive dogs to locate the source of target odors throughout the day. Samples are next hidden at multiple indoor locations, varying height, and degrees of detection difficulty. After 1–2 days, the scent box is again briefly used to teach the dog to sit at the sample prior to receiving the reward. This keeps the dog focused on the scat until the handler can confirm its presence. Scat samples are then gradually hidden over a progressively larger, defined area in the field. Samples are set out in the training area at least several hours prior to any given training session. This allows the scat scent to percolate into the environment and any human scent trail to dissipate. Dogs are introduced to scat from many different individuals of each target species".

After the initial training in Eatonville, the handler (Heath Smith) travelled to the Snake River Birds of Prey area, outside of Kuna, Idaho to reinforce the scat scent training with Pips in natural conditions of high density badger habitat (10 animals per square mile). This training was done for three consecutive days (November 10-12, 2011). Once in San Diego, the canine scent detection team surveyed targeted sites from November 14 to December 14, 2011. The schedule was typically 3 days on with one day off as recommended by Conservation Canines. USGS biologists assisted as orienteers and for data collection. The schedule was based upon the priority level of the site (Appendix 1), ability to obtain access permits, and proximity of locations to each other. On several days, we would survey more than one site in a day, so nearby priority locations were chosen. All routes and detection locations were recorded using a GPS unit attached to the dog. GPS coordinates were taken and pin flags were placed at locations where the dog indicated a scent detection (behavior change, "hit"). After a dog "hit", the handler would state the confidence level in the dog's response as well as the handler's confidence in the dog's response. All scat was collected with gloved hands, placed into a plastic bag, and stored frozen until DNA testing. The orienteer also recorded information on the condition of the scat (color, freshness, and contents), vegetation type, dominant soil type, and took photos of the scat and representative habitat.

Scat DNA Testing

The goal was to identify if scat samples collected in the field were from the American badger. CCB developed a badger specific identification assay that amplifies two American badger specific DNA markers. Four extractions were performed from each scat sample using a modified version of Qiagen DNeasy Tissue kit and performed PCR reactions using the designed primers. The amplified strands were analyzed using a highly sensitive ABI DNA fragment analyzer that measures the alleles precisely using a laser to read fluorescently labeled product against a known DNA standard. Because of the specificity of the assay, all positive results can be interpreted as DNA from the American badger. Negative results should be interpreted as either being from another species or from the scat of an American badger where the DNA was too degraded to amplify in the PCR's.

7