identifies and prioritizes management activities for vernal pools on land owned by the City of San Diego.

2.0 METHODS

2.1 *Research on existing vernal pool inventories*

In fall 2002, City staff researched historical and recent reports and current expert knowledge to determine locations and existing survey data for vernal pools within the City's jurisdiction. Site names reflecting location, ownership, or project were assigned to series and/or complexes, and all areas were reviewed using aerial photographs to determine their current status. A Geographic Information System (GIS) was then utilized to document all sites containing extant vernal pools. Unnatural pools formed in tire tracks, called road ruts, were included when vernal pool indicator species were present. The following general geographic areas of interest were identified: Del Mar Mesa, Carmel Mountain, Mira Mesa, Nobel Drive, Kearny Mesa, Mission Trails Regional Park, urban San Diego areas, Otay Mesa, Otay Lakes, and Marron Valley (refer to Fig. 1). Vernal pool complexes within each area were identified, and data were compiled for each complex, including previous site designation and recent survey information, such as number of vernal pools, plants and/or animals present, and specific threats to the site.

2.2 *Field data collection*

Each site was visited following storm events with recorded rainfall of 0.33 inches or more from January through May 2003. At or near maximum inundation, individual basin boundaries were recorded using a differential sub-meter GPS, and the following attributes were documented: site name; Bauder identification number (if any); vernal pool identification number (assigned by series at time of survey); date; time; state of inundation; comments; presence/absence and percent cover for *Eryngium aristulatum* (San Diego button celery), *Navarretia fossalis* (spreading prostrate navarretia), *Orcuttia californica* (California Orcutt grass), *Myosurus minimus* (little mousetail), *Pogogyne abramsii* (San Diego mesa mint), and *Pogogyne nudiscula* (Otay mesa mint); presence/absence for *Branchinecta* spp. (unidentified fairy shrimp), *Branchinecta sandiegonensis* (San Diego fairy shrimp), *Branchinecta lindahli* (versatile fairy shrimp), *Streptocephalus woottoni* (Riverside fairy shrimp), tadpoles/metamorphs (unidentified), *Spea hammondi* (tadpole/metamorph/adult Western spadefoot toad), *Bufo boreas* (tadpole/metamorph/adult Western toad), and *Hyla regilla* (tadpole/metamorph/adult Pacific tree frog). Where possible, the spatial and/or biological data were obtained from current consultant survey information. In cases when such data were not available, information was collected during site visits by City staff. Dates and personnel information for data collection are available in Appendix A.

Two hand-held devices were used in spatial data collection: Trimble TSC1 datalogger using Asset Surveyor version 5.00, and Compaq iPaq running ESRI's ArcPad 6.0. Each collection device was linked to a Trimble Pro XR sub-meter GPS receiver. The Pro XR utilizes real-time differential GPS correction which results in increased field accuracy while eliminating the need for post-processing data correction. In the office, spatial data and attributes from each collection system were combined using Trimble's Pathfinder Office 3.00 and ESRI's ArcView 3.3 software, respectively.

Fairy shrimp were sampled from 84 basins as dormant cysts or live adults. Cysts were collected in soil samples and grown under laboratory conditions. Mature shrimp were collected from inundated basins and preserved in 95% ethanol. All sampling was conducted by members of the Branchiopod Research Group of USD appropriately permitted by USFWS under the direction of Dr. Simovich (Appendix B).

Fairy shrimp collection was based on a nested design: in most cases, ten individuals were sampled from each of three basins per complex (six at some larger complexes) across the species range within the City of San Diego. A matrix of the Eucladian distance between sampled basins was determined from the vernal pool inventory, and will be used to assess the possible correlation between distance and genetic similarity. Collection at 24 locations within the City resulted in 1000 *B. sandiegonensis* samples which will be analyzed as described below.

Extracted DNA was amplified for the mitochondrial gene Cytochrome Oxidase I, using novel primers designed by Bohonak for work on other arthropods. Amplifications were screened on a 2% agarose gel, and successful amplifications were cycle sequenced using ABI BigDye v.3 with manufacturer's suggested protocols. Sequences were visualized on an ABI 377 automated sequencer. Preliminary sequences were used to verify the accuracy of this protocol with *B. sandiegonensis*.

These data were analyzed using the standard summary statistics A (number of alleles in a sample), H_e (expected heterozygosity) and π (average pairwise sequence divergence) at the levels of the basin, complex and species. Genetic differences among populations were analyzed in terms of F_{ST}, patterns of isolation by distance, nested clade analysis, standard phylogeography and maximum likelihood and coalescent estimates of gene flow. These analyses provide inferences into historical and contemporary processes that have shaped population genetic structure in *B. sandiegonensis.*

When observed, the presence/absence of the following amphibians (tadpoles, metamorphs, and adults) were noted at each basin: *Bufo boreas, Hyla regilla*, and *Spea hammondii*. A total of 44 basins at 11 sites were randomly sampled with the assistance of USGS Biological Resources Division staff. Ten individuals were taken from each sampled basin, and a total of 440 individuals were collected and preserved in 95% ethanol. Samples were used to verify field identification and in future genetic testing not associated with this project. All samples are stored at the San Diego office of the USGS.

Helix Environmental has prepared a comprehensive list of vernal pool plant species present by series per Corps vernal pool indicator guidelines (U.S. Army Corps of Engineers, Los Angeles District, Regulatory Branch, November 1997) and vernal pool plant species listed in Tables 6a and 6b in Zedler's *Ecology of Southern California Vernal Pools: A Community Profile* (1987). Each site was surveyed twice by an experienced vernal pool biologist during the winter and spring of 2003. Surveys were timed to coincide with 1) the highest level of basin inundation, and 2) optimal flowering time later in the season for detection and identification of both early and late showing vernal pool species. Wetland and upland species present at each complex were also surveyed.

At sites where one or more of the following endangered, threatened, or MSCP covered species occur, City staff estimated percent cover for each species in each

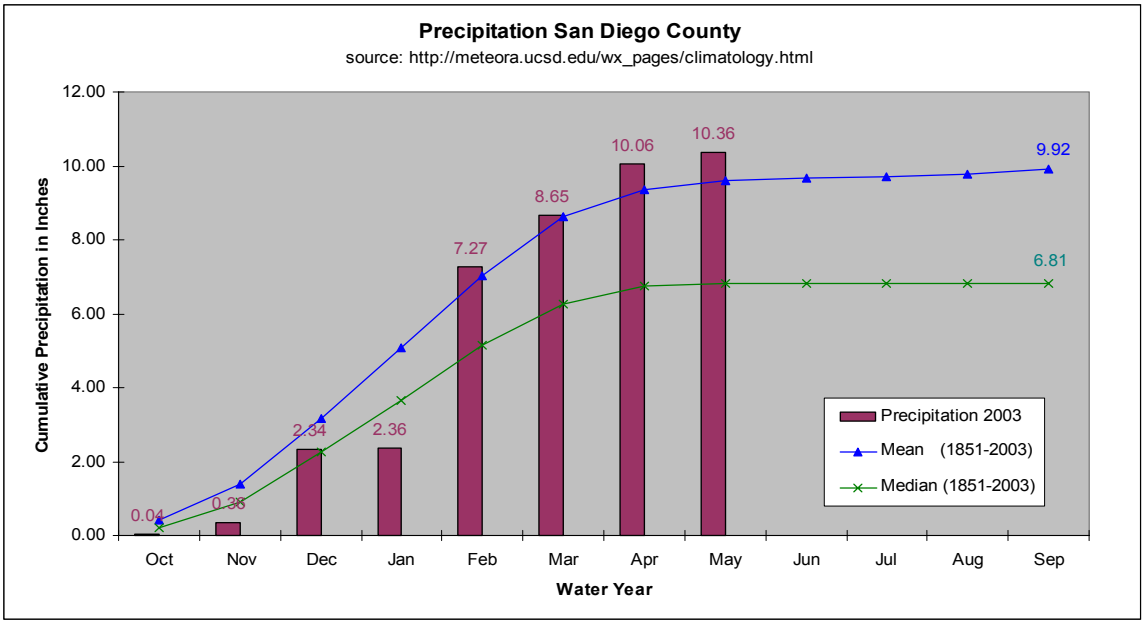
vernal pool: *Eryngium aristulatum* spp. *parishii*, *Navarretia fossalis*, *Orcuttia californica*, *Myosurus minimus*, *Pogogyne abramsii*, and/or *Pogogyne nudiscula*. Staff visits were scheduled to coincide with the maximum foliage of vegetation, and multiple visits were necessary in many cases. Estimates were derived from the California Native Plant Society percent cover worksheet, and were calculated based on vernal pool basin area. Estimated relative percent cover was grouped in the following classes to track major changes in population size over time: <1%, 1-5%, 5-10%, 10-25%, 25-50%, 50-75%, 75%+.

City staff has completed nearly 3500 hours of research and field surveys for this project and have mapped 2516 individual vernal pool basins within the City of San Diego.

2.3 *Limitations*

Vernal pool ecosystems are extremely sensitive to inter- and intra-annual environmental variability. The size and number of inundated basins, and therefore their associated biota, are directly correlated to the amount and timing of precipitation. Rainfall during the 2003 water year has been consistent with average recorded precipitation in San Diego over the past 153 years (Figure 2), and the majority of basin areas were recorded during March when cumulative precipitation for 2003 equaled cumulative average rainfall. Based upon the amount and timing of rainfall, it is believed that the data gathered for 2003 represents a "typical" year for vernal pool biota.

Figure 2: Precipitation in San Diego County for the 2002-2003 rainfall year



The use of GPS technology greatly improved the spatial precision of this inventory over previous vernal pool research efforts. The system utilized for this survey represents the best available technology; however, the submeter resolution can become problematic in certain situations. Spatial inaccuracies of up to one meter are

considered precise for larger basins but may off-set the boundaries of small vernal pools (i.e., basins with diameters less than two meters). In addition, basins that are close together may appear to overlap due to changes in the angle of the receiver beacon and/or number of positional satellites available. Even with the described errors, the data are considered to be accurate to less than one meter, and provide the most comprehensive and spatially accurate information within the City to date.

Due to the lack of a federal recovery permit, City staff was unable to verify visual identification of fairy shrimp. The size of the inventoried areas was also prohibitive to the identification of all fairy shrimp species present in every basin. Because of these factors, it was impossible to conclusively determine presence of fairy shrimp. As such, the current inventory may under-represent the true number of vernal pools with fairy shrimp and should only be used for large scale planning purposes.

The size of the survey area and staff scheduling proved to be an obstacle to completing plant surveys and cover estimates. A comprehensive plant survey per vernal pool was not possible due to the high number of basins; therefore, these surveys were conducted at the series scale. In addition, vernal pool plants species tend to occur at different sites at different times throughout the spring. Cover estimates for the endangered, rare, or MSCP covered species at a given site were timed to coincide with maximum foliage of each species; however, due to rainfall, scheduling, etc., certain complexes may have shown greater cover of a given species before or after the estimate was completed. Because of the longevity of the species, it is felt that the cover estimate classes adequately represent the basins for the year surveyed.

Although the inventory represents an extensive and thorough effort, it does not provide a complete record of vernal pools and vernal pool species found within the City of San Diego. Military lands, including the Miramar Mounds National Natural Landmark at MCAS Miramar, are excluded. Efforts to work in cooperation with the military, including a formal written request, went unanswered. Private lands inaccessible to the City (e.g., posted "No Trespassing") were also not surveyed. A minimal number of additional vernal pools may have been overlooked due to their location in the midst of dense vegetation or other access issues. To help evaluate this limitation, the following project is underway to determine the usefulness of remote sensing to detect vernal pools in such areas.

Vernal pool preserves at Del Mar Mesa are characterized by thick upland vegetation of chaparral and scrub oak that hindered a comprehensive survey. Research has shown high resolution, multi-spectral imagery, such as Airborne Data Acquisition and Registration (ADAR), to be the preferred remote sensing option in high detail land cover detection and classification over relatively small areas (Stow, et al., 1998.) ADAR is a visible spectrum and near-infrared aerial digital imaging system which has been used successfully to detect recently created vernal pools (surrounded by bare ground) through visual and software-directed image classification (Hope, et al., 2002.) ADAR images of the study sites within the City were taken shortly after a major rain event late in the rainy season in an attempt to image the inundated basins. The contrast in spectral reflectance between the soil, vegetation, and water (vernal pools) has been used to detect nearly 40 vernal pools located in the inaccessible uplands. This basin detection effort will be compared to the vernal pool inventory to determine the accuracy

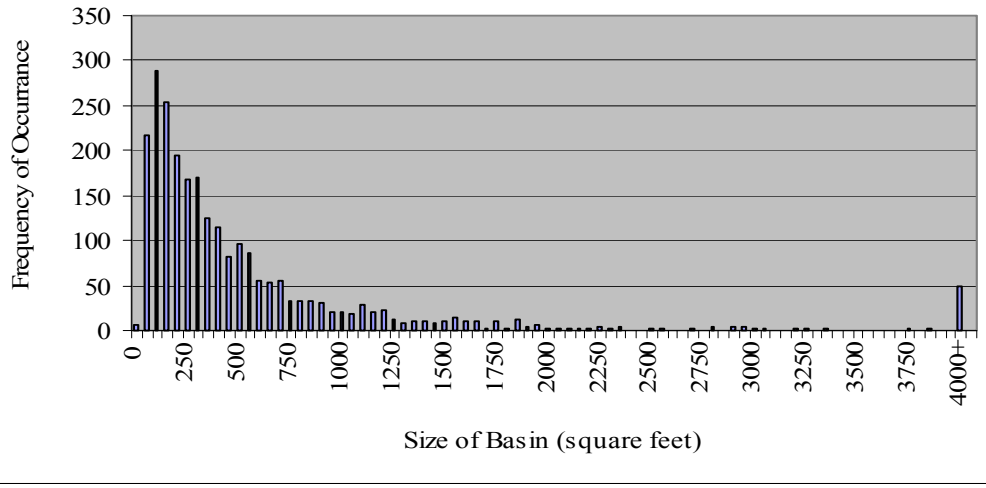
of the procedure and assess the utility of remote sensing to detect or monitor vernal pools in the future.

3.0 RESULTS

3.1 *Inventory summary*

A total of 2516 vernal pool basins at 62 sites were mapped during the 2002-2003 rainy season. For this study, a vernal pool basin was defined as the extent of land surface covered by ponding water following a large rain event during an average rainfall year. Watershed area is not included in the basin area. Basin areas were highly variable—from 2.13 to 68,364 square feet—with a standard deviation of 2,138 around the mean of 668. The frequency distribution of basin areas is given in Figure 3.

Figure 3
Histogram of Vernal Pool Basin Area



The sites and associated Bauder identification numbers are listed with the number of mapped basins in Table 1. Sites in this inventory that were not recognized by the Bauder survey have been assigned a Bauder-type identification number in the following manner: Sites located adjacent to Bauder complexes were included in the existing designation, while isolated sites were assigned to the nearest complex and given a series number subsequent to those of historical designations. These revised Bauder identification numbers are shown in bold in Table 1.

Sites are designated as conserved according to the following criteria: vernal pool basin area occurring on land covered by a conservation easement, dedicated in fee title to the City for mitigation purposes, or designated City open space. A total of 1,369 vernal pools within the City of San Diego are conserved; this represents 54% of the basins mapped in this inventory. Of the remaining 1,193 vernal pools, 419 (17% of total basins) are located on publicly owned parcels but are not considered conserved according to the stated criteria; these sites are noted by an asterisk in Table 1. Note that site acreage for unconserved sites is based on parcel boundaries and is not indicative of natural habitat and/or vernal pool watershed. Site acreage for conserved sites corresponds to the preserved area surrounding the vernal pool basins.