



Environmental DNA Pilot Monitoring Program for Invasive Species and Biodiversity Assessments on Santa Cruz Island

Interim Report, September 2025



UC San Diego graduate student collecting an environmental DNA sample on Santa Cruz Island.
USGS / A. SEPULVEDA

Environmental DNA Pilot Monitoring Program for Invasive Species and Biodiversity Assessments on Santa Cruz Island: Interim Report, September 2025

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Abstract

The U.S. Geological Survey (USGS) and Southern California Coastal Water Research Project supported Channel Islands National Park, The Nature Conservancy's (TNC) Santa Cruz Island Preserve, and University of California San Diego (UCSD) researchers in using environmental DNA sampling to monitor for invasive Argentine ant (*Linepithema humile*) and to describe spatial biodiversity patterns in a freshwater stream on Santa Cruz Island that is colloquially known as Cañada del Puerto Creek (hydrologic unit code 12 number 180600140201). An eDNA autosampler was deployed May 27–June 02, 2025, which filtered and preserved four 2-L water samples every 12 hours (n = 48 samples). These samples were extracted by USGS and shipped to UCSD for targeted eDNA analyses of Argentine ant. Environmental DNA sampling kits were used to filter and preserve water samples at four sites along the creek spanning ~5 km of stream distance. Six 1-L samples were collected at each site May 27–28 and samples were analyzed for eDNA metabarcoding using a comprehensive panel, which encompasses species across the tree of life. In these samples, eDNA from 2,134 unique taxa spanning 30 taxonomic groups (e.g., amoebas, insects, plants) were found. These sampling efforts demonstrated how eDNA autonomous and manual sampling approaches can be applied by National Park Service, TNC, and partners at scale to provide otherwise difficult and expensive to acquire information about biological threats and biodiversity.

List of Terms

NPS: National Park Service

SCCWRP: Southern California Coastal Water Research Project

TNC: The Nature Conservancy

UCSD: University of California San Diego

USGS: U.S. Geological Survey

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Introduction

The Channel Islands, a chain of eight islands located along the coast of southern California, is a biodiversity hotspot and includes over 2,000 plant and animal species, with 145 of these species being endemic (Rick et al. 2014). Five of the islands are managed by the National Park Service (NPS) and one island is partially owned and managed by The Nature Conservancy (TNC). Shared management goals of NPS and TNC include conservation and preservation of the islands' endemic species by (a) reversing the extensive ecological effects of the ranching era (1800s and 1900s), which introduced livestock and other nonnative animals and plants to the islands; (b) prevention and control of any additional introductions associated with island visitation and operations; and (c) tracking island biodiversity change over time and space to assess management action impacts.

Environmental DNA (eDNA) sampling detects genetic material released by organisms into environments such as water using easy-to-collect samples and provides key capabilities for monitoring biodiversity and alerting to new species' introductions in both aquatic and terrestrial habitats (Sepulveda, Nelson, et al. 2020). Here, the U.S. Geological Survey (USGS) worked with NPS, TNC, Southern California Coastal Water Research Project (SCCWRP), and University of California San Diego (UCSD) scientists to demonstrate how eDNA autonomous samplers (autosamplers) and user-friendly eDNA sampling kits can be easy and potentially scalable approaches to help achieve management goals.

Unmanned, eDNA autosamplers can be programmed to collect and preserve an eDNA water sample any time of day or night at high frequencies (e.g., hourly or daily). Autosamplers that can collect many samples at appropriate temporal scales should result in improved monitoring performance and overcome many monitoring constraints associated with the Channel Islands, such as travel and work schedules, safety concerns, and adverse weather (Sepulveda, Birch, et al. 2020). However, autosamplers can be expensive, heavy, and difficult to use at remote sites or when expanding spatial survey coverage. User-friendly eDNA sampling kits are portable and inexpensive but do require personnel to be on site to manually filter and preserve water samples.

The Smith-Root eDNA autosampler (Vancouver, Washington) was used to survey for the invasive Argentine ant (*Linepithema humile*) eDNA at a focal site downstream of a recent eradication effort on the Santa Cruz Island's colloquially named Cañada del Puerto Creek (hydrologic unit code 12 number 180600140201) watershed. We also demonstrated how eDNA sampling kits can be used to expand spatial coverage for biodiversity surveys and Argentine ant surveillance in this watershed.

Methods

Autosampler Description

As described in George et al. (2024), the eDNA autosampler functions by suctioning water through self-preserving (desiccating) filter housings on a user-defined schedule. The system components include (in flow order): an intake strainer, an intake hose, an upper manifold, a lower manifold, a pressure sensor, a diaphragm water pump in suction orientation, a flow sensor, and an outlet hose. Filters are placed between the upper and lower manifolds, creating independent sealed chambers for each filter with a pair of inlet and outlet valves that direct flow in and out of each chamber. The suction pump produces up to 12 pounds per square inch (psi) for filtration in the sealed chambers. The eight filter locations are indexed and can be scheduled independently to filter at specified times, using the onboard software interface. All samples taken by the autosampler were collected on single-use Smith-Root self-preserving 5-micrometer (μm) polyethersulfone (PES) filters.

Filter Kit Description

Each Wilderlab eDNA kit (Wellington, New Zealand) contains six encapsulated 30 millimeter (mm) diameter, 1.2 μm cellulose acetate syringe filters with luer-lock inlet and outlet fittings, a 60 milliliter (ml) luer-lock syringe, 400 microliter (μl) DNA/RNA Shield preservation buffer (Zymo Research, Tustin, California) pre-loaded in six 3 ml luer-lock syringes, and a pair of sterile nitrile gloves. For each sample, the 60 ml syringe is used to repeatedly push water through the encapsulated filter until a target volume is reached (typically 1 liter [L]) or until the filter is clogged. Residual water in the filter is then purged by forcing 60 ml of air through the filter, using the emptied 60 ml syringe. The material collected on the filter is then preserved by injecting the DNA/RNA Shield solution from the pre-loaded 3 ml syringe into the filter inlet with the outlet capped using the luer stopper provided with the preservative syringe. Optimal results are typically obtained by collecting six replicate samples from each site (Melchior and Baker 2023; Smith et al. 2024). Samples can be stored at room temperature for up to six months following collection.

Study Site and Field Methods

The USGS worked with TNC, SCCWRP, and UCSD to deploy the eDNA autosampler in Cañada del Puerto Creek (33.998830°, -119.715099°), ~300 m downstream of TNC's main ranch house complex, from 27 May to 02 June 2025 (Figure 1). Immediately prior to inserting the intake tubing into the creek, we collected a 2-L field negative control by placing the autosampler intake tubing into a sterile bottle of Milli-Q ultrapure water. We then secured the intake tubing and strainer in the creek's thalweg, keeping the strainer fully submerged but making sure it was not in direct contact with the creek bottom. We programmed the autosampler to collect four, 2-L water samples every 12 hours at ~10:00 and 22:00, which resulted in 48 samples. A UCSD scientist visited the autosampler daily to retrieve spent filters and reload with new filters. All filters were shipped to the USGS Northern Rocky Mountain Science Center (USGS NOROCK, Bozeman, MT).

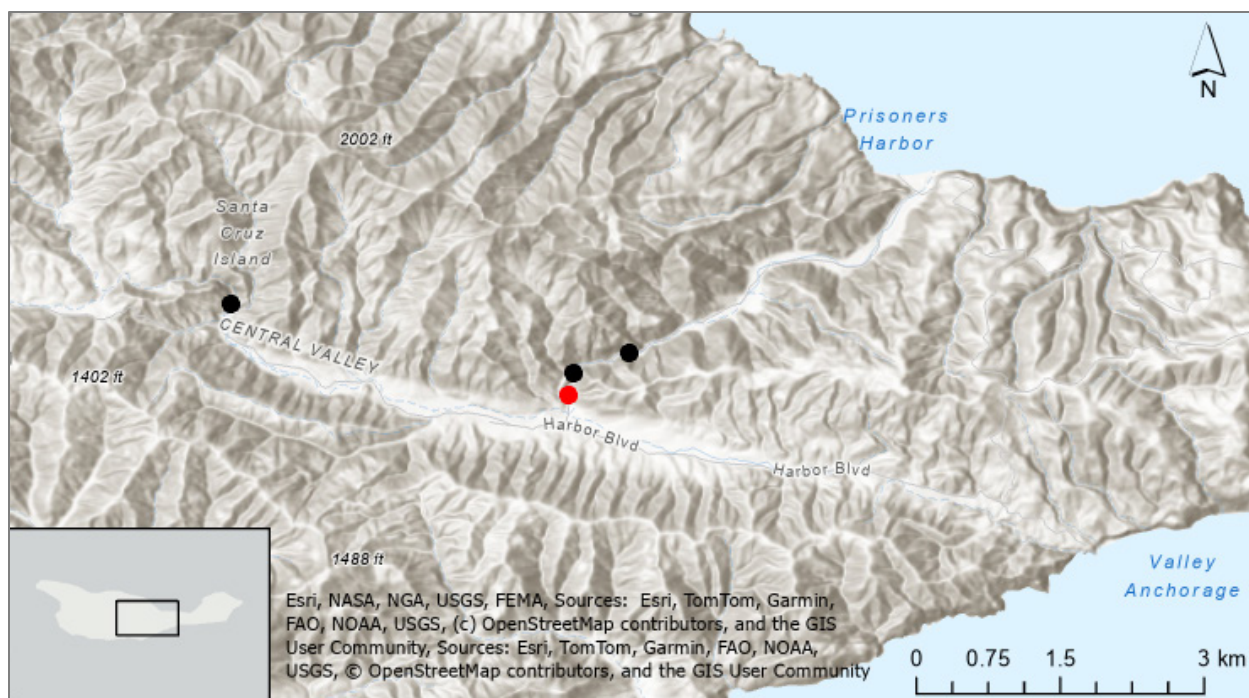


Figure 1. Sampling sites on Santa Cruz Island, California. The red point indicates where the eDNA autosampler was deployed; all sites were sampled using the eDNA kits.

USGS

The USGS worked with TNC, SCCWRP, and UCSD to collect eDNA samples using the Wilderlab kits at four sites that spanned ~5 kilometers (km) of stream distance in Cañada del Puerto Creek in May 2025. The most upstream site was a pool associated with a check-dam structure (34.007448°, -119.746703°) and the three remaining sites were downstream of the TNC’s main ranch house complex where the road intersects the creek, including the site where the autosampler was deployed (upstream to downstream: 33.998830°, -119.715099°; 34.000963°, -119.714723°; and 34.002805°, -119.709403°). Six replicate samples, each consisting of ~1 L of filtered water, were collected at each site. All filters were shipped to Wilderlab (North Bend, WA).

Bioinformatic Methods

Personnel at the USGS NOROCK extracted autosampler filters using the Investigator Lyse & Spin Basket Kit (Qiagen catalog #19598) in concert with the DNeasy Blood & Tissue Kit (Qiagen catalog #69506) and eluted in 400 µl of AE buffer. The procedure was carried out according to the manufacturer’s instructions with no significant modification. Aliquots of this DNA extract were then shipped to UCSD for analysis of Argentine ant eDNA. Methods will be provided by UCSD when analyses are complete.

Wilderlab used their standard operating protocol to extract and analyze samples using their “comprehensive” freshwater panel of assays that target at least 10 distinct species groups ([Wilderlab SOP](#), [Wilderlab assays](#)). Metabarcoding results, which included exact sequence variants (ESVs), their taxonomic assignments found in each sample using each assay, and Taxon-Independent Community

Index (TICI) scores for eDNA-based ecological health assessment—analogueous to indices of biological integrity derived from observational data (Wilkinson et al. 2024)—were emailed to USGS NOROCK. Because we lacked data to inform false-positive error rates, we used a conservative approach and subset this dataset to only include ESVs with at least five reads in a sample and taxa that were detected in more than one sample replicate at each site.

Results

We collected 48 field samples and one field negative control with the autosampler. Analyses are underway at UCSD and final results are pending.

We collected six 1-L samples at each of four sites using the Wilderlab kits. No Argentine ant eDNA was detected in these samples, but we did detect 2,134 unique taxa spanning 30 taxonomic groups (Figure 2). This included 1 amphibian species, 2 bird species, 59 insect species, 22 plant species, and 3 spider species. Full eDNA results, which included DNA sequence barcodes and taxonomic assignments, are reported in Sepulveda (2025)—[Environmental DNA sample results from Santa Cruz Island, California, May 2025: U.S. Geological Survey data release](#).

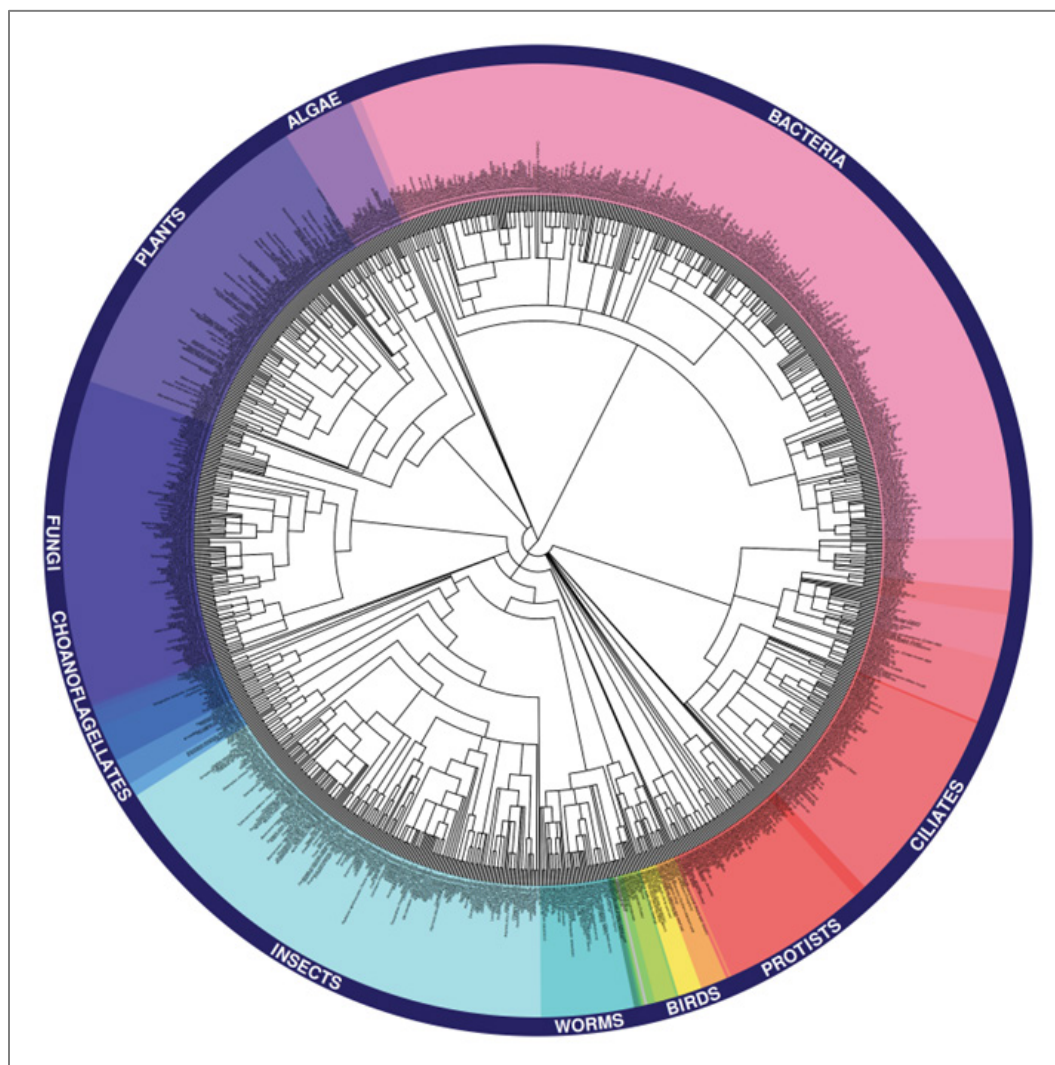


Figure 2. Wheel of life to visualize the biodiversity detected in all eDNA kit samples collected on Santa Cruz Island, California, in May 2025, according to their taxonomic grouping. This wheel of life was generated using Wilderlab ShinyWheels.

USGS

TICI scores were “good” (101–103) for the three sites downstream of the TNC’s main ranch complex and “average” (97–99) for the most upstream site, indicating that the ecological condition at the upstream site is more likely to be affected by human disturbance than communities in the downstream sites (Wilkinson et al. 2024).

Lessons Learned, Challenges, and Next Steps

- TNC and UCSD personnel were quickly trained on the eDNA autosampler, were able to operate it independently of USGS onsite supervision, and produced high-quality samples with no evidence of contamination. This success underscores the potential for USGS to support groups like TNC with an eDNA autosampler library program as a cost-effective, enterprise-level solution that allows TNC to leverage enhanced monitoring capabilities of autosamplers without being constrained by the technology's downsides.
- The autosampler enabled many eDNA samples to be collected during the workday, while TNC and UCSD personnel were doing other work and research tasks, and at less convenient times late at night. This demonstration underscores the potential for autosamplers to have a rapid return on investment by improving work efficiency.
- The Smith-Root eDNA autosampler used in this project is heavy (~65 kg/144 lb) and requires transport to/from Santa Cruz Island using port and dockside marine cranes, which are not readily available. It is otherwise impossible to load the autosampler on a boat or to unload onto the dock. Once on the island, the autosampler must be transported by vehicle and then carried by at least two people to the deployment site—this makes many desirable sites on the island inaccessible. Smaller, lighter autosamplers may be more fit-for-purpose for future work in the Channel Islands.
- Spatial coverage of biodiversity and biothreat monitoring was expanded with easy-to-use eDNA sampling kits. We were able to gain insight into a broad diversity of taxa and the ecological health of sites that typically require subject-matter experts to sample and identify. These types of broad, population-level screening approaches can help identify novel threats or evaluate biodiversity trends over time.
- The Wilderlab analysis workflow was fast, with results returned to USGS within 10–15 days of sample submission. Their workflow is optimized for New Zealand and Australian taxa but improvements in taxonomic assignments for North American taxonomic groups are ongoing. Nevertheless, these rapid workflows show how eDNA data across a broad range of taxonomic groups can be used to inform adaptive management decisions about NPS and TNC restoration efforts.

Literature Cited

- George, S.D., A.J. Sepulveda, P.R. Hutchins, D.S. Pilliod, K.E. Klymus, A.C. Thomas, B.C. Augustine, C.C. Huddleston Adrianza, D.N. Jones, et al. 2024. Field trials of an autonomous eDNA sampler in lotic waters. *Environmental Science & Technology* 58(47):20942–53. <https://doi.org/10.1021/acs.est.4c04970>
- Melchior, M., and C. Baker. 2023. Environmental DNA guidelines and field protocols for lotic systems. https://s3.ap-southeast-2.amazonaws.com/wilderlab/resources/literature/eDNA_guidelines_report_2023279HN.pdf
- Rick, T.C., T.S. Sillett, C.K. Ghalambor, C.A. Hofman, K. Ralls, R.S. Anderson, C.L. Boser, T.J. Braje, D.R. Cayan, et al. 2014. Ecological change on California’s Channel Islands from the Pleistocene to the Anthropocene. *BioScience* 64(8):680–92. <https://doi.org/10.1093/biosci/biu094>
- Sepulveda, A.J. 2025. Environmental DNA sample results from Santa Cruz Island, California, May 2025: U.S. Geological Survey data release. <https://doi.org/10.5066/P13K4WVB>
- Sepulveda, A.J., J.M. Birch, E.P. Barnhart, C.M. Merkes, K.M. Yamahara, R. Marin III, S.M. Kinsey, P.R. Wright, and C. Schmidt. 2020. Robotic environmental DNA bio-surveillance of freshwater health. *Scientific Reports* 10(1):1–8. <https://doi.org/10.1038/s41598-020-71304-3>
- Sepulveda, A.J., N.M. Nelson, C.L. Jerde, and G. Luikart. 2020. Are environmental DNA methods ready for aquatic invasive species management? *Trends in Ecology & Evolution* 35(8):668–678. <https://doi.org/10.1016/j.tree.2020.03.011>
- Smith, J., B. David, A. Hicks, S. Wilkinson, N. Ling, D. Fake, A. Suren, and A. Gault. 2024. Optimizing eDNA replication for standardized application in lotic systems in Aotearoa, New Zealand. *Environmental DNA* 6:e70017.
- Wilkinson, S.P., A.A. Gault, S.A. Welsh, J.P. Smith, B.O. David, A.S. Hicks, D.R. Fake, A.M. Suren, M.R. Shaffer, et al. 2024. TICI: a taxon-independent community index for eDNA-based ecological health assessment. *PeerJ* 12:e16963. <https://doi.org/10.7717/peerj.16963>

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