CALIFORNIA
ENERGY COMMISSION

Energy Research and Development Division

FINAL PROJECT REPORT

Habitat Influences on Desert Tortoise Translocation Success

May 2023 | CEC-500-2023-033

PREPARED BY:

Primary Authors:
Melissa J. Merrick, Ph.D.[1]
Talisin T. Hammond, Ph.D.[1]
Ronald R. Swaisgood, Ph.D.[1]

[1]San Diego Zoo Wildlife Alliance
15600 San Pasqual Valley Road
Escondido, CA 92027-7000 USA
www.sdzwa.org

Contract Number: EPC-16-053

PREPARED FOR:
California Energy Commission

David Stoms, Ph.D.
Project Manager

Kevin Uy
Branch Manager
Energy Generation Research Branch

Jonah Steinbuck, Ph.D.
Director
ENERGY RESEARCH AND DEVELOPMENT DIVISION

Drew Bohan
Executive Director

ACKNOWLEDGEMENTS

The authors thank Kemp Anderson, Kelsi Black, Corey Chan, Daniel Essary, Carrie Gonzalez, Sarah Greely, Susanne Marczak, Reed Newman, and Brian Sandstrom for field support, husbandry, and data collection. The authors express thanks to Drs. Kristin Berry, James Danoff-Burg, Michael Tuma, Melia Nafus, and Lisa Nordstrom for advising on project protocols, ecological and methodological expertise, and partnership. The authors thank Bajada Ecology and Ironwood Consulting for help with tracking, searching for, and radiographing tortoises. The authors thank Drs. Nadine Lamberski and Josephine Braun for oversight of animal care, welfare, and advising on tortoise health and disease investigations. All animal husbandry and research were conducted in accordance with Institutional Animal Care and Use Committee approvals (San Diego Zoo Wildlife Alliance Institutional Animal Care and Use Protocol #18-006).

PREFACE

The California Energy Commission's (CEC) Energy Research and Development Division supports energy research and development programs to spur innovation in energy efficiency, renewable energy and advanced clean generation, energy-related environmental protection, energy transmission and distribution and transportation.

In 2012, the Electric Program Investment Charge (EPIC) was established by the California Public Utilities Commission to fund public investments in research to create and advance new energy solutions, foster regional innovation and bring ideas from the lab to the marketplace. The CEC and the state's three largest investor-owned utilities—Pacific Gas and Electric Company, San Diego Gas & Electric Company and Southern California Edison Company—were selected to administer the EPIC funds and advance novel technologies, tools, and strategies that provide benefits to their electric ratepayers.

The CEC is committed to ensuring public participation in its research and development programs that promote greater reliability, lower costs, and increase safety for the California electric ratepayer and include:

- Providing societal benefits.
- Reducing greenhouse gas emission in the electricity sector at the lowest possible cost.
- Supporting California's loading order to meet energy needs first with energy efficiency and demand response, next with renewable energy (distributed generation and utility scale), and finally with clean, conventional electricity supply.
- Supporting low-emission vehicles and transportation.
- Providing economic development.
- Using ratepayer funds efficiently.

Habitat Influences on Desert Tortoise Translocation Success is the final report for the Habitat Influences on Desert Tortoise Translocation Success project (Grant Number: EPC-16-053) conducted by the San Diego Zoo Wildlife Alliance. The information from this project contributes to the Energy Research and Development Division's EPIC Program.

For more information about the Energy Research and Development Division, please visit the CEC's research website (www.energy.ca.gov/research/) or contact the CEC at ERDD@energy.ca.gov.

ABSTRACT

The solar energy industry and the Mojave desert tortoise (*Gopherus agassizii*) share a preference for the sunny desert regions of southeastern California. The tortoise is listed as threatened by the U.S. Fish and Wildlife Service and the state of California, which impose a rigid permitting process on the solar industry and other development. The release of immature tortoises following captive rearing, or headstarting, is recommended as a recovery tool to mitigate population declines, although this can be costly and require four to ten years in captivity. Therefore, identifying optimal headstart methods is key for species recovery.

This research aimed to understand the role of headstarting methods and careful selection of habitat characteristics at the release site on post-release growth, movement, and survival of juvenile tortoises in hopes of shortening the captive rearing phase. This research identified key microhabitat characteristics that are important for juvenile tortoise resource selection and began to identify relationships between habitat variables and post-release movement and survival in translocated juvenile tortoises.

Findings suggest that tortoises preferentially select habitat with a greater density of burrows, shrubs, and the small sand mounds that form under shrubs. The researchers do not yet have evidence for any significant effects on survival of these habitat features or of age at release (one versus two years). Severe drought following release inhibited juvenile tortoise growth as no forage was available and may have contributed to mortalities. Due to the slow life history of the Mojave desert tortoise, impacts of age at release and habitat on demographic variables may take more time to manifest. The findings do suggest that home-range sizes and travel distances are larger for two-year-olds in comparison to one-year-olds. These findings could have important implications for carrying capacity, release site selection, target densities of translocations, and other factors pertinent to tortoise conservation.

Keywords: Desert tortoise, *Gopherus agassizii*, headstarting, translocation, habitat selection, resource selection, optimal habitat, survival, movement

Please use the following citation for this report:

Merrick, Melissa J., Talisin T. Hammond, and Ronald R. Swaisgood. 2021. *Habitat Influences on Desert Tortoise Translocation Success.* California Energy Commission. Publication Number: CEC-500-2022-033.

TABLE OF CONTENTS

Page

LIST OF FIGURES

Page

LIST OF TABLES

Page

EXECUTIVE SUMMARY

Background

Construction of solar and other renewable energy facilities enables California to meet electricity demand while reducing effects on the global environment from climate change. Because Southern California contains a great deal of land that is highly suited towards the production of solar and wind energy, the region has been a major target for developing large-scale renewable energy infrastructure. Because large amounts of land are affected by these development projects, industry and public regulators have acknowledged the need to reduce or minimize the impacts of development on sensitive species or habitats.

One such species is the Mojave Desert tortoise (*Gopherus agassizii*) that lives exclusively in the Mojave Desert. The U.S. Fish and Wildlife Service listed the Mojave Desert tortoise as threatened under the Endangered Species Act in 1990, and the state of California also protects it as a threatened species. Renewable energy projects potentially represent a major contributing factor in the continued decline of the desert tortoise. Therefore, wildlife agencies impose requirements in the permitting of solar energy facilities to minimize impacts and help the tortoise population recover, such as moving all tortoises from a site to other suitable habitat prior to development. One promising method is called headstarting, in which juvenile tortoises are raised in captivity and then released into the wild when they are past their most vulnerable stage. Current headstart methods rely on at least four years in captivity, which is expensive and therefore may be impractical. Research into ways to decrease the time spent in captivity by individuals is necessary to reduce program expenditures. Shifting attention to more careful and science-based selection of release habitat, rather than multi-year headstarting programs, may provide a more cost-effective method of ensuring survival of relocated tortoises.

Project Purpose and Description

Little is known about the juvenile age class in desert tortoises, including what constitutes ideal habitat for juveniles and the effects of habitat in which they are released on post-release growth, movement, and survival. Previous work in Nevada by the research team identified four major habitat characteristics that appear likely to influence desert tortoise survival and productivity: abundance, availability, or quality of burrows; vegetation community and a diversity of forbs and grasses, substrate composition; and precipitation patterns. These resources or habitat characteristics may vary depending on broad-scale perennial vegetative communities. In particular, the presence or absence of species such as Joshua tree or Mojave yucca may provide key indicators of the relative productivity or value of habitat parcels for maintaining positive desert tortoise population growth. This project sought to improve understanding of the relationships among broad-scale and fine-scale habitat features and juvenile desert tortoise space use, habitat selection, and survival to identify the most suitable sites for releasing headstarted tortoises.

The primary objective was to characterize which environmental factors boost the success of juvenile headstarted desert tortoises after they are released from captivity. To increase generalizability of the findings, parallel studies were conducted at Edwards Air Force Base in the western Mojave Desert and in Ward Valley, near Chambless, California, in the northern Colorado Desert. Such information can be used to guide desert tortoise headstart programs implemented or regulated by federal and state agencies as mitigation for renewable energy development and for desert tortoise recovery more broadly.

Project Approach

Existing captive rearing pens at Edwards Air Force Base were improved, whereas new pens were constructed by Cadiz, Inc. on their property in Ward Valley. In 2018 and 2019, researchers searched for and located adult female desert tortoises at the two study areas. Once adult females were located, they were fit with a radio transmitter for tracking, and x-rayed every 7 to 10 days until the presence of calcified eggs was detected. At that point, they were brought into the closest headstart pens. Once females laid their eggs in artificial burrows, each female was returned to where she was captured. The hatchlings were reared in outdoor rearing pens until fall of 2020, resulting in groups of tortoises that were one and two years of age.

Headstarted juvenile tortoises from both age groups were released in fall of 2020 at sites distinguished by vegetation community (yucca woodland versus creosote scrubland), elevation, and substrate type (gravel versus sand). At Edwards Air Force Base and Ward Valley, tortoises were released at one yucca woodland site and one creosote scrub site (four total release sites). Each juvenile was fitted with a radio transmitter enabling researchers to track their movements, habitat selection, and survival, which were compared among release sites. These individuals will continue to be tracked and monitored as part of an ongoing long-term study, using other sources of funding.

This project was a collaborative effort among the San Diego Zoo Wildlife Alliance, the United States Geological Survey (USGS), and Edwards Air Force Base (EAFB). Ecologists from the United States Fish and Wildlife Service (USFWS), California Department of Fish and Wildlife (CDFW), and private consulting organizations assisted with permitting, design, logistics, and implementation.

A technical advisory committee (TAC) was formed with representatives from United States Fish and Wildlife Service, United States Air Force, local agencies, scientists, and the energy industry. The TAC met annually in 2017, 2018, and 2019 to provide project guidance.

Project Results

Tortoises from both age groups remained healthy and showed significant growth over time in captivity with similar growth rates. Post-release growth appeared to be affected by unusually low precipitation and resulting lack of foraging resources. They did not display significant increases in growth post-release and no changes in the rate of growth across age classes. While slow growth rates are expected as individuals were inactive and remained below ground for a significant portion of the post-release monitoring period, post-release growth was much lower than observed in other studies. Low magnitude decreases in weight post-release were documented as a result of an extreme drought year, with no significant differences in the rate of change across age classes.

Researchers found significant differences in habitat characteristics at locations occupied by tortoises in comparison to randomly selected locations. Juvenile tortoises appeared to select habitat with a significantly larger number of burrows, lower proportion of bare ground cover, and higher proportion of perennial and shrub cover. Plots used by tortoises also tended to have significantly more sand on the surface in comparison to rockier types and were significantly more likely to contain coppice mounds, small sand dunes that typically form around vegetation like creosote bushes due to wind. Coppice mounds contain friable soil that facilitates digging and have a high abundance of small mammal burrows available to juvenile tortoises for shelter.

Post-release movement by tortoises varied by age class, with two-year-olds moving more than one-year-olds. Home range sizes tended to be slightly larger in the creosote scrub sites compared to yucca woodland sites and for tortoises hatched in 2018 compared to 2019. Animal home range size is determined in part by body size and resource availability, and these results suggest that yucca woodland may provide more resources, such as burrows and forage, compared to creosote scrublands.

In the 13 months of post-release monitoring, 14 confirmed mortalities (out of 144 released tortoises) were documented. In many cases, it was not possible to determine a cause of death, but in six of the 14 cases, predation was suspected. Survival rates did not significantly differ between yucca and creosote sites, or between age classes. Of tortoises with known status (alive or carcass found), the study reported about a 78 percent survival rate. Of the 144 tortoises, nine had detached transmitters and their status is currently unknown, and 74 remain missing (in part due to transmitter failure). However, long-term monitoring may be required to reveal patterns of survival as influenced by habitat types.

The complete nest failure at our Cadiz site in Ward Valley in 2018, while unfortunate, sheds light on potential thermal limits for nesting tortoises and informs future headstart efforts. Average burrow temperatures at the Cadiz site approached 104 degrees Fahrenheit (°F) during June and July, while average burrow temperatures at two study sites at Edwards Air Force Base remained below 95 ºF, just below temperatures known to cause tortoise egg mortality. Especially with a warming climate, site selection for headstart facilities should be guided by expected temperatures and may preclude areas formerly in the desert tortoise range experiencing higher temperature profiles.

Knowledge Transfer

Throughout this work, the research team coordinated with regulatory agencies, land managers, developers, and other project partners for all major project activities. Annual status reports were provided to project partners summarizing activities and findings. The final results of this project will be made publicly available and will be provided to local, state, and federal agencies. These agencies will be able to directly use the research results to improve California's mitigation guidelines.

This research was featured through the San Diego Zoo Wildlife Alliance's online media and the Community Engagement department as an educational activity involving tortoise tracking and conservation. Research activities were also shared with a broader audience through a number of presentations that include the United States Fish & Wildlife Service Mojave Desert Tortoise

Coordination Meeting, the American Association of Zookeepers, the San Diego Zoo Wildlife Alliance's Advanced Inquiry Program's Master's students, several university and professional meetings, and a widely attended conference on *Extinction: Solutions for Species on the Brink* at the University of California, Irvine.

Benefits to California

This research aimed to make headstart mitigation more cost-effective for the solar energy industry and more successful in recovering a threatened species. This double benefit could help facilitate achieving California's clean energy and climate goals through future renewable energy deployment. It is important to determine the best practice methods for headstarting juvenile tortoises, including reducing time spent in captivity and increasing survival in the wild after release. By expanding the understanding of the effects of tortoise size at release, and the effects of careful site selection and post-release habitat selection on tortoise movements and survival, mitigation costs could be reduced. Implementation of more effective mitigation practices could increase the probability of removing the desert tortoise from the threatened species list in the future.

CHAPTER 1: Introduction

Current Status of the Mojave Desert Tortoise

The southwestern United States, with large expanses of undeveloped government-owned land, ample wind, high solar radiation, and cloud-free days, has tremendous potential for renewable energy development (Lovich & Ennen 2011; Agha et al. 2020). California and many western states have adopted ambitious renewable energy targets that will support efforts to reduce carbon emissions and decrease dependence on fossil fuels. However, these energy targets require extensive development and infrastructure in areas that often overlap sensitive habitats and species, creating a conflict between biodiversity conservation and renewable energy goals (Lovich & Ennen 2011; Agha et al. 2020). The iconic Mojave desert tortoise (*Gopherus agassizii*, referred to in this report as desert tortoise or tortoise) is a State- and Federally-listed threatened species with designated critical habitat in portions of California, Nevada, Arizona, and Utah (U.S. Fish and Wildlife Service [USFWS], 2011; California Code of Regulations, 2013). Wild populations of tortoises have been declining for decades due to threats from urbanization (U.S. Fish and Wildlife Service 2011), habitat loss and degradation (USFWS 2010), heightened predation by subsidized predators (for example [Esque et al. 2010]) and disease (Brown et al. 1994, 2004; Homer et al. 1998; Jacobson et al. 2012).

The 2016 Desert Renewable Energy Conservation Plan allocated 388,000 acres of public land in the Mojave Desert of California for potential development of renewable energy projects (Bureau of Land Management 2016). Given the extensive loss of habitat projected for this species, it is critical to identify what constitutes high quality habitat to guide long-term preservation, translocation, and habitat restoration efforts. Furthermore, lack of recruitment or addition of young tortoises into reproductive populations has been and continues to be a major barrier to recovery, leading the USFWS to identify augmenting depleted populations through headstarting and translocation efforts as a strategic recovery action (U.S. Fish and Wildlife Service 2011).

Predation and Development Are Major Threats

Two primary threats impacting recovery of the desert tortoise are predators and human development (Esque et al. 2010). Subsidized predators are predator species that thrive in urbanized or human-associated habitats, thanks to their ability to take advantage of resources that are associated with humans. In the Mojave Desert, common ravens (*Corvus corax*) are a subsidized predator that consume desert tortoises with potentially devastating impacts on survival rates (Kristan & Boarman 2003; Boarman et al. 2006). Tortoises are also depredated opportunistically by other predator species, including coyotes and kit foxes (Kelly et al. 2021).

Previous studies have noted unusually high predation on tortoises, particularly near human population centers. Increased predator densities are often associated with human development due to abundant resources like water, road kill and trash, and perching and nesting sites. Predation on tortoises by human-subsidized predators may be even higher in drought years when populations of other prey species like small mammals are low. In drought years, the smallest, youngest age classes suffer the highest mortality (Esque et al. 2010; Nagy et al. 2015).

Development also directly threatens tortoise populations by reducing habitat quality, e.g. increasing fragmentation, increasing risk of road-related mortality, and altering vegetation communities by enabling invasive species proliferation or allowing off-road vehicle or agricultural land use; (Averill-Murray et al. 2012, 2021; Berry et al. 2020).

Development projects on public lands that affect threatened and endangered species, such as the Mojave Desert tortoise, are required to mitigate for habitat loss or degradation under various federal policies and practices. However, methods of calculating these mitigation credits often do not appropriately estimate the actual value of habitat (Searcy & Shaffer 2008). Typical habitat suitability assessments, including those used for the desert tortoise, often rely on broad-scale habitat features (e.g., vegetative cover, substrate type) associated with presence-absence of a species (Goertz 1964; Power 1984; Nussear et al. 2009). However, presence or density of individuals is driven by a number of factors, not all of which reflect habitat quality (Fretwell & Lucas 1970; Van Horne 1983). Consequently, reliance on presence only metrics may lead to the conservation of "ecological traps"–areas attractive to tortoises but associated with poor survival and/or reproduction (Schlaepfer et al. 2002; Battin 2004). This concern is especially worrisome for the desert tortoise, which, because of its long life and ability to survive in suboptimal habitats, may continue to persist in degraded habitats for many years. Ultimately, for species like the desert tortoise that are already in decline, assessing what features represent quality of habitat is critical for improving models of population persistence (Robles & Ciudad 2012), and for evaluating both the current and future value of a habitat patch.

For land and restoration targets to accurately reflect true measures of habitat quality, two critical, and often overlooked, data pieces are availability and quality of forage and refugia. Due to cost limitations, restoration efforts are unlikely to seed degraded habitat with the full variety of species that were historically present on the landscape. These omissions may be particularly important for smaller tortoises, which typically have limited ability to move across the landscape into better foraging areas. These limitations may combine with the availability and quality of burrows for shelter from heat, cold, and predators. The identification of high-quality habitat types, and important microhabitat features therein, should guide preservation and restoration and translocation efforts.

To best conserve and protect desert tortoises, it is critical to characterize how these two sources of risk (predators and human development) can be mitigated. In the context of headstarting programs, the most viable options for mitigating these risks may pertain to careful selection of release sites, and to headstarting tortoises to older ages to make them less vulnerable to predation.

Species Management Through Wildlife Headstarting and Translocation

Description of Headstarting Programs

Headstarting is a process wherein neonatal (juvenile) animals are maintained under human care and reared to a certain age before release into the wild, usually with the goal of getting animals past young life stages that are associated with high mortality. Headstarting is a specialized subset of wildlife translocations that has been employed for a variety of species (Cunninghame et al., 2015; Nagy et al., 2015) after being pioneered with chelonians (turtles and tortoises) (Shaver & Wibbels 2007). For the Agassiz's desert tortoise, headstarting is listed as a specific recovery strategy under the USFWS Revised Recovery Plan for the Mojave Population of the Desert Tortoise (Recovery action 3.1; USFWS 2011). Nevertheless, the efficacy of headstart programs has been insufficiently validated, and, therefore, the technique remains controversial in chelonians and other species (Allen 2001; Germano & Bishop 2009; Bubac et al. 2019). Very little, however, is known about the ecology of juvenile tortoises or the efficacy of headstarting for improving survival outcomes. Key questions about headstarting include the optimal release age at which to translocate individuals, methods for assessing trade-offs between age-related boosts in survival versus increases in cost for headstarting, methods for best preparing animals for translocation into the wild, and strategies for optimizing release site selection.

Factors Predicting Translocation Success

Research on translocation efforts, failures, and successes across diverse taxa provides a framework upon which best practices in the translocation toolkit can be assembled. Successful translocations often consider one or more of the following:

- Natural history–Understanding the natural history of a species is key for translocation efforts. The timing of reproduction, activity patterns, and the relative importance and timing of extrinsic drivers such as resource availability can help determine when and where translocation efforts will be most effective.
- Habitat–Understanding what variables constitute high quality habitat for a species or age class is essential for selecting where to release translocated animals. Anchoring animals to the release site and limiting post-release dispersal is one of the greatest challenges to translocation success (Berger-Tal et al. 2020). For Mojave Desert tortoises, availability of burrows, substrate consistency and texture, and cover are known to reduce detection by predators, and post-release dispersal (Nafus et al. 2015a, 2017a). Limiting detection means that tortoises are more challenging to locate in their environment, which may make them less vulnerable to predation. Across taxonomic groups, higher post-release dispersal distances tend to be associated with lower survival rates; thus, better understanding factors that reduce post-release dispersal may be advantageous in improving translocation success.
- Mitigating the cause of decline–Ultimately, across 554 translocation studies, the most important factor determining the success or failure of a translocation effort was whether the cause of the species decline had been mitigated (Bubac et al. 2019). Important

drivers of decline across species include habitat loss, invasive species, overexploitation, and predation from native species (Bubac et al. 2019).

Measures of Translocation Success: Site Fidelity, Growth Rate, and Survival

The value, and hence quality, of a habitat patch should be reflected in demographic parameters that affect probability of population persistence [i.e., vital rates, (Todd & Rothermel 2006)]. Specifically, habitat can be considered higher quality if individuals exhibit rates of growth, survival, and reproductive success that reduce likelihood of population extinction (Schooley & Branch 2009; Kennedy et al. 2011; Robles & Ciudad 2012). There should be a demonstrable relationship between habitat characteristics and individual fitness to establish a cause-effect relationship between population vital rates and habitat components (Morrison 2001). Forage and refuge availability and quality is likely an important driver of post-release growth and survival for the desert tortoise as well as post-release site fidelity, measured via the magnitude of movements away from the release site. A particularly important aspect of preserving and restoring the desert landscape is knowledge of which plant species should be considered critical resources necessary for tortoise growth, survival, and ultimately reproduction. Because desert tortoises are a fossorial or burrowing species that are exposed to extreme thermal fluctuations, a second important limitation may be refuge or burrow quality and quantity (Zimmerman et al. 1994). Thus, documenting movements, growth, survival relative to macro- and micro-habitat features like vegetation community types, forage species, and burrow availability following translocation is essential for understanding translocation success.

Recent desert tortoise research has made significant inroads into successful headstarting techniques that increase survival. Researchers have identified morphometric targets that may improve survival outcomes, such that older and larger tortoises can have greater survival, with the highest survivorship demonstrated in juveniles aged 9 years or more with a midline carapace length (MCL) of greater than 100mm (Nagy et al., 2015). Whether the age or the size was the stronger contributing factor remains to be determined. Larger body size may convey additional protection from predators such as the common raven (*Corvus corax*). Todd et al. (2021) found size at release to be the best predictor of post-release survival, but Daly et al., (2019) found no differences in survivorship between larger sized indoor reared headstart tortoises and smaller outdoor reared tortoises or controls, with avian predation being the leading cause of mortality. In addition, coyotes (*Canis latrans*) can and do take tortoises of this size, and do so with greater frequency in drought years (Nagy et al. 2015). Therefore, the criterion that juveniles greater than 100 mm MCL be released to improve survival is a presumption that must be better investigated.

Previous EPIC-funded work by Todd et al. (2021) tested one strategy for enhancing post-release survival of juvenile tortoises: by rearing individuals indoors, which allows for young tortoises to reach a larger body size in a shorter amount of time, thereby making them less vulnerable to predation. The work described here tests another strategy for enhancing post-release survival of juvenile tortoises: by testing whether carefully selected release sites mitigate predation risk, allowing tortoises to survive even at small body sizes through use of crypsis, or concealment by cover, burrow refuges, and matching the substrate.

Increasing Translocation Success by Selecting Release Sites That Address Specific Ecological Needs

Testing Effects of Age at Release in California

Across taxonomic groups, there is significant uncertainty about the optimal age at which to translocate headstarted animals (Lloyd et al. 2019; DeGregorio et al. 2020; Resende et al. 2021). It is more financially costly to rear animals to older ages, and some studies have suggested that animals that spend longer periods in captivity may habituate to captive settings and may lose critical survival skills (Bloxam & Tonge 1995; Hellstedt & Kallio 2005; Mason et al. 2013). However, in general, younger, smaller animals are more susceptible to predation and have higher overall mortality rates (Chaparro-Pedraza & de Roos 2020). This may be true for desert tortoises, for which certain studies have documented significantly improved survival after reaching at least 100 mm MCL (Daly et al. 2018; Tuberville et al. 2019; McGovern et al. 2020a), but see Nafus et al. (2017a), largely due to predation risk.

To address this issue, an outdoor headstart rearing program was implemented to examine whether ecological factors at the release site influence desert tortoise growth, behavior, and survival. Habitat may be used to improve survival of smaller sized juveniles in the wild to equivalent rates observed in larger size classes in a more cost-effective manner if age or size at release can be reduced. The aim was to support the development of cost-effective release methods for headstarting and translocation programs for this threatened species. Further, knowledge gained on how specific habitat characteristics influence tortoise growth, survival, and post-release movements will aid in management decisions regarding selection of the most suitable areas for preservation and provide targets for restoration efforts, thus furthering tortoise recovery and reducing the burden of mitigation for tortoises by energy developers. Because factors influencing the outcomes of headstarting may vary with geographic location (e.g., predation pressure), headstarted tortoises were released in two distinct geographic regions to increase the generalizability of the findings and develop location-specific release strategies.

Habitat Selection as a Tool for Mediating Predation Risk

Other traits besides age and body size affect predation risk for desert tortoises. For instance, habitat features may impact the ability of tortoises to actively or passively hide in their environments. Previous work in the eastern to northeastern Mojave found that habitat can significantly mediate relationships between body size and survival for juvenile desert tortoises. As a part of this previous work 140 juvenile tortoises were released and monitored (ranging in size from 50–150 mm MCL and aged from 6 months to > 10 years) across five different release sites in Nevada (Germano et al. 2017; Nafus et al. 2017a, 2017b). Notably, survival for juveniles between 100–150 mm MCL experienced a first-year post-release survival rate of 85 percent at the Nevada National Security Site, which was slightly less than the 88 percent survival documented here of animals ranging from 60–80 mm MCL released into yucca woodland habitat across four sites. Smaller juveniles (less than 100 mm MCL) had variable survival rates depending on release-site habitat characteristics. Two-year post-release survival in yucca woodland with a high degree of rock camouflage and high density of small mammal burrows was a remarkable 82 percent. But when released in creosote scrub, with low burrow density and low camouflage potential, survival fell to 0 percent. Tortoises released into intermediate habitats had intermediate survival rates: tortoises in creosote scrub with high burrow density and high camouflage potential experienced 67 percent survival, while 50 percent survived when released into yucca woodland with low burrow density and low camouflage potential (Nafus et al. unpubl. data).

Research therefore suggests that habitat and specific resources within the environment, such as rodent burrows, can be carefully selected to enhance juvenile survival and protect them from predators in the wild (Nafus et al. 2015; Todd et al. 2016; Nafus et al. 2017a) perhaps without costly long-term captive husbandry. Empirical data indicate that headstarting tortoises for more than 4 years may be unnecessary, whereas paying close attention to the details of habitat at the release site may produce greater gains for headstarting efficacy than methods and duration of headstarting.

Goals

Evaluate Best Practice Husbandry for Effective Desert Tortoise Headstarting

The first goal was to develop and evaluate best practices for headstarting desert tortoises in ex situ environments and implement cost-effective tortoise headstart programs for mitigation, translocations, and species recovery. The main objectives under this goal were to:

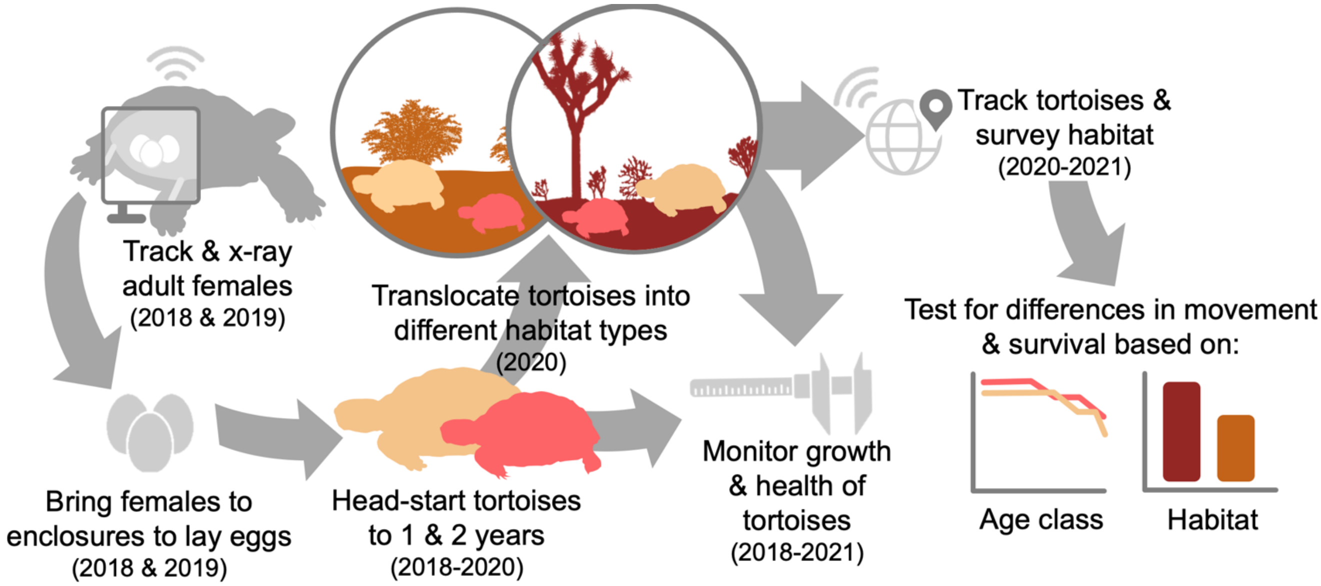
- Rear juvenile desert tortoises to different ages (1 and 2 years of age).
- Evaluate effects of husbandry conditions on growth pre-release.
- Release and monitor juvenile tortoises headstarted for 12 and 24 months in habitats with variable ecological conditions.

Enhance Understanding of Environmental and Habitat Factors to Improve Post-Release Success

The second goal was to characterize which environmental factors boost the post-translocation success of juvenile headstarted desert tortoises. Specifically, the effects of burrow availability, substrate characteristics, and vegetation community on survival of tortoises was assessed. The main objectives under this goal were to:

- Measure relationships among growth, microhabitat selection, movements, and survival of juvenile tortoises post-release to better estimate the effects of size and ecological factors in the release habitat.
- Measure foraging and refuge resources, substrate, and other habitat variables at release sites and correlate with juvenile growth, survival, and settlement decisions.

Figure 1: Flow Chart of Project



Flow chart illustrating project objectives for juvenile desert tortoise headstarting and translocation work.

Source: Talisin Hammond, San Diego Zoo Wildlife Alliance

CHAPTER 2: Project Approach

Study Area

Headstarting Facilities

Two headstart facilities were established in 2018: one at Edwards Air Force Base (EAFB) for the western Mojave Desert and one at Chambless, California, on property privately owned by Cadiz Inc. for Ward Valley in the northern Colorado Desert. At EAFB, the pens already existed from a previous headstart program (Juvenile Hatchery at the Edwards Tortoise Study Site) begun in 2002 (Figure 2). Visits to the existing EAFB headstart rearing facility were made, and it was determined to be suitable, subject to modest improvements. Work was conducted to modify the EAFB pens in preparation for the holding of adult females for egg laying and their hatchlings. San Diego Zoo Wildlife Alliance (SDZWA) and U.S. Geological Survey also visited lands owned by Cadiz Inc. to determine suitability for construction of a headstart rearing facility and evaluate potential release sites (Figure 3). A suitable site was found in a secure location near existing infrastructure with access to water and electricity. The Cadiz operations manager agreed to build the tortoise enclosures as an in-kind contribution to the project. Construction of the facility was completed in April 2018. Both facilities were inspected by the SDZWA Institutional Animal Care and Use Committee to ensure that they met with animal care standards and were suitable for adult females to lay their eggs and the rearing of juvenile tortoises.

Figure 2: Headstart Facilities at Edwards Air Force Base

Overview of outdoor rearing pens at Edwards Air Force Base used to house adult females and pre-release hatchlings 2017-2020.

Source: Lisa Nordstrom, San Diego Zoo Wildlife Alliance

Figure 3: Headstart Facilities at Cadiz

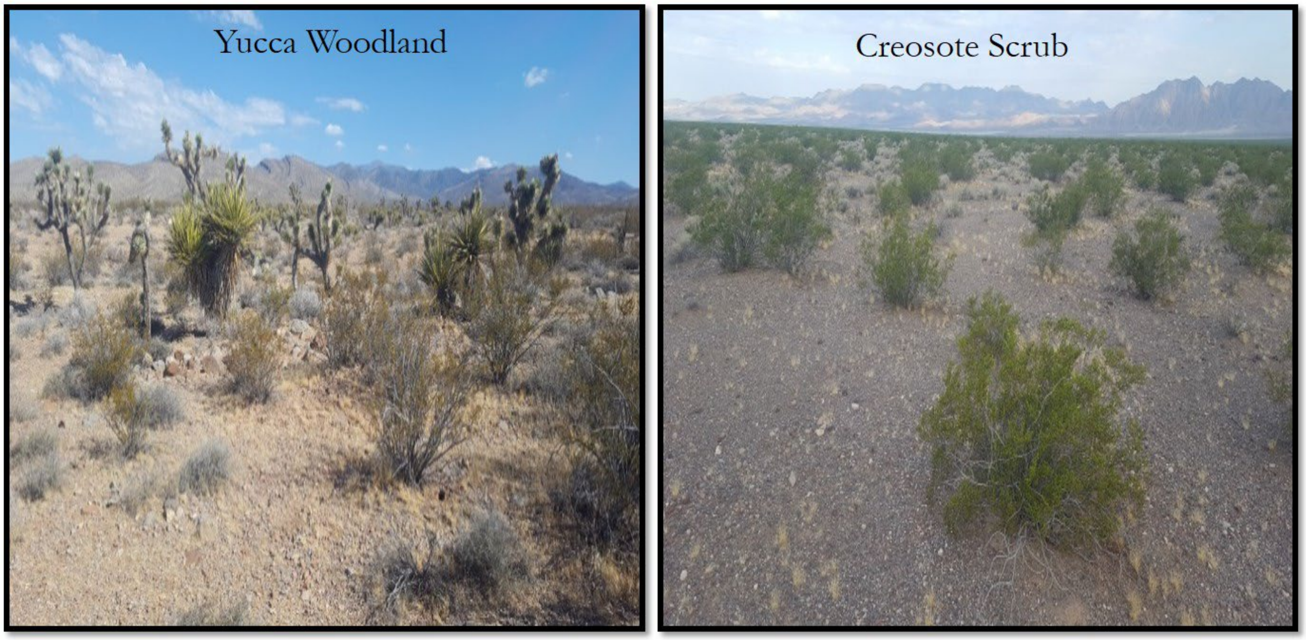
Overview of outdoor rearing pens at Cadiz used to house adult females and pre-release hatchlings 2018-2019, and incubator-raised hatchlings in 2020.

Source: Lisa Nordstrom, San Diego Zoo Wildlife Alliance

Field Sites

Two sites for releasing the juvenile tortoises were identified for each headstarting facility—one in yucca woodland and one in creosote Scrub (Figure 4). The sites on EAFB included one located just north of Leuhman Ridge (yucca woodland dominated by *Yucca brevifolia*) and the other located approximately 20 km to the west, designated as Baker-Nunn (creosote scrub; Figure 5). These sites were selected for previous release of juvenile tortoises 2013-2014. The Baker-Nunn site was selected by EAFB staff using a habitat suitability model (Hailstone 2015) followed by site visits to verify appropriate habitat conditions. The Leuhman Ridge site was selected by USGS and SDZWA based on site visits to verify habitat conditions. The upper Ward Valley release sites were selected by USGS and SDZWA in spring 2017 following visitation to a number of potential sites in the northern Colorado Desert. The Ward-High site represented the yucca woodland (dominated by *Yucca schidigera*), and the Ward-Low site was creosote-scrub.

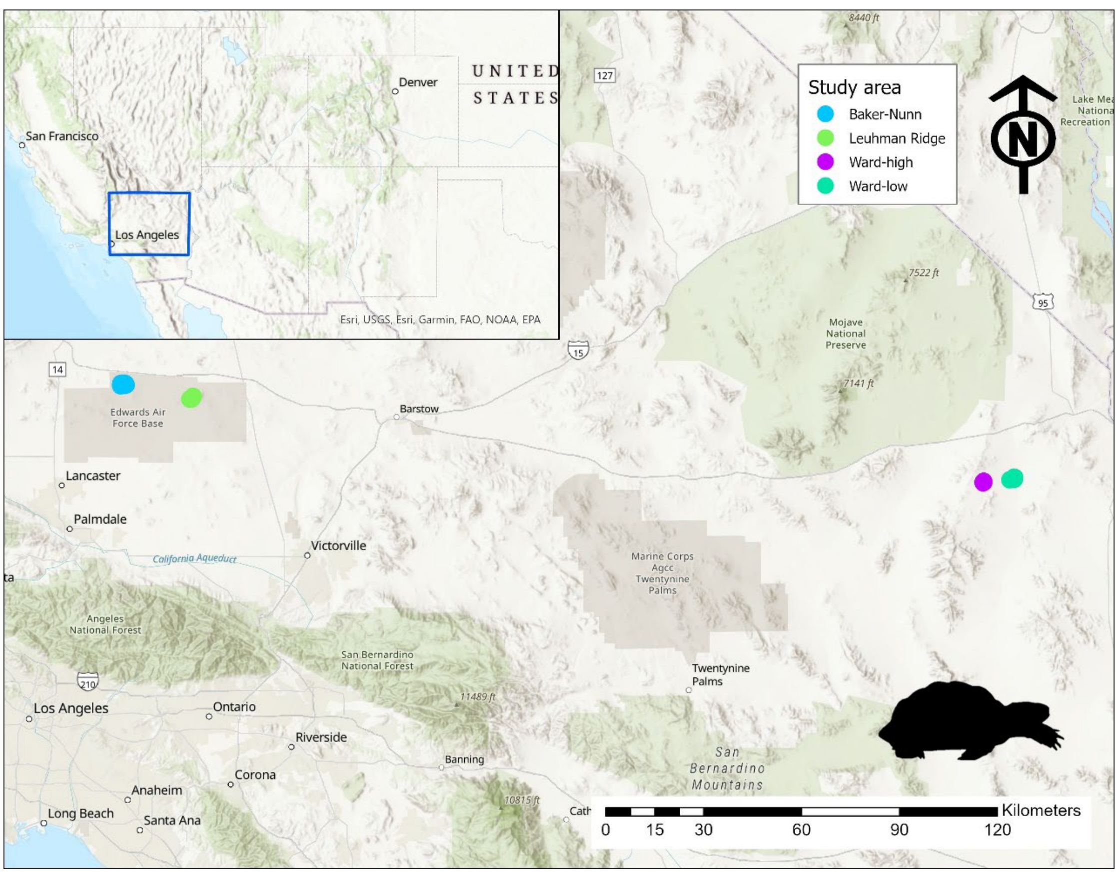
Figure 4: Comparison of Vegetation Community Types



Comparison of yucca woodland and creosote scrub vegetation community types selected for headstarted juvenile tortoise release, Mojave Desert, CA, USA .

Source: Lisa Nordstrom, San Diego Zoo Wildlife Alliance

Figure 5: Overview of Release Sites Selected for Juvenile Headstarted Tortoises



Overview of study areas and release sites selected for headstarted juvenile Mojave Desert tortoises hatched in 2018 and 2019, released in October 2020, Mojave Desert, California, USA.

Source: Melissa Merrick, San Diego Zoo Wildlife Alliance

Adult Female Collection

Adult female tortoises over 200 mm MCL were captured through visual surveys (Figure 6). Captured females were weighed, measured, and a health evaluation (including nasal, oral, and cloacal swabs to evaluate disease status) was conducted for each individual. A radio transmitter was attached to the first right or left caudal scute using 5-minute epoxy putty (tan or grey in color) with the antenna extending down the carapace. The Very High Frequency (VHF) transmitters themselves weigh 10 grams and last 24 months. Coupled with the epoxy adhesive and plastic tubes used for antenna guides, the transmitter package in total weighs approximately 20 grams, representing approximately 1 percent of the weight of the tortoise. The typical guideline for use of transmitter packages on chelonians is that they be limited to less than 5 percent of the body weight of the study animal. USFWS requires that packages not exceed 10 percent of the weight of the tortoise.

Radio tagged females were tracked to X-ray them each 7-10 days to determine if shelled eggs were present (Figure 7). Because resource availability can strongly influence fecundity, egg production by female tortoises during the breeding season (March-July) was monitored for two years (2018 and 2019). Females were radiographed (Digital Imaging Systems, Poskum Model PDX-B10, 60 kvp, 0.6 mAS, 48 cm focal length) in the field (and after transport to the pens) every 14 days from March 14–July 15 following permit guidelines issued by USFWS and previous research studying desert tortoise fecundity (Mueller et al. 1998). When calcified eggs were detected, egg counts from radiographs were used to estimate clutch size, clutch frequency, and annual fecundity. Handling for the purposes of radiography was not longer than 2-5 minutes per animal which is the duration of time it takes to put the animal beneath the x-ray, complete the x-ray, and then return the animal to its original location. Handling only occurred in temperatures below 35C. Females were rehydrated by allowing them to soak in a shallow tub of water if they voided during handling. Subcontractors Bajada Ecology (2018) and Ironwood Consulting (2019) assisted with tortoise searches, tracking, and radiography at both sites (EAFB and Ward Valley).

When a female was determined to have shelled eggs, she was collected from the field and transported to the headstart facility on EAFB (for females collected at EAFB) or at Cadiz, Inc. in Ward Valley (for females collected at Ward Valley). Females were housed in the pens individually until eggs were laid. Each female was closely monitored while in the pens, using camera traps, and observations during husbandry, to determine her health/behavior and nesting location. Each female was x-rayed weekly until confirming that eggs had been laid.

Figure 6: Adult Female Tortoise

An adult female tortoise in situ.

Source: Lisa Nordstrom, San Diego Zoo Wildlife Alliance

Figure 7: X-ray Scan Showing Female Tortoise With Eggs

An x-ray of an adult female desert tortoise (MT-5005 in June 2018) with five eggs (the opaque circle at the top right of the tortoise is the radio-transmitter).

Source: San Diego Zoo Wildlife Alliance

Unfortunately, in 2018 there was complete nest failure at Cadiz. A combination of high temperatures and rocky soils leading to shallow nests were likely responsible for the widespread egg loss. Temperature and humidity data loggers (iButtons) were placed approximately 0.5 m inside burrows at each site to measure temperature and humidity of the habitat as experienced by tortoises during the nesting season. The iButtons were deployed from May 15 through Aug 15, 2019 and documented temperature differences experienced by nesting females across sites.

Because of nest failures at the Cadiz headstart facility in 2018, a different strategy was employed in 2019.

In 2019, gravid females with fully shelled eggs were transported from Ward Valley to The Living Desert Zoo and Gardens (TLD) for hormonal egg induction. Tortoises were evaluated by a TLD vet, weighed, and soaked in a water bath prior to egg induction. Ultrasound was also used to confirm that the eggs were ready for oviposition if x-rays were not taken on the same day as transport to TLD. Oxytocin was administered intramuscularly based on the body weight of the tortoise to induce labor. Eggs were monitored daily for hatching. Hatchlings were then transported to the Cadiz headstart facility and were outdoor-reared until release in October 2020.

After laying eggs, females were re-released at their last known burrow location and monitored once per month via radio tracking for 12-18 months. After completion of the 2019 nesting season, radio transmitters were removed from the adult female tortoises at EAFB and Ward Valley and each was given a final health assessment, including the collection of blood and oral swabs for genetic and disease testing.

Headstarting

Husbandry Practices

Nests laid by females were left to incubate naturally in the nest cavities when possible. For the eggs in incubators, the temperature and humidity were maintained at a constant level to maximize hatching success. Because sex determination for tortoises is temperature dependent, a pivotal temperature of 31.3 degrees Centigrade was used produce an equal sex ratio (Rostal & Jones 2002). In August, pen/incubator checks were conducted at least weekly to locate emerging neonates. Immediately upon detection (after the yolk sac had been absorbed), hatchlings were weighed, measured, marked (for example paper tag, shell notching via nail clippers), and subjected to a health evaluation. Hatchlings were then water soaked and placed into a pen. Juveniles kept in pens were offered water and fed a mixed diet of native plants supplemented with tortoise pellets at least weekly during their active periods in the fall prior to overwintering and in the spring after emergence from their nests. This diet was a standard normal diet, following the diet protocols developed at the Desert Tortoise Conservation Center in Nevada and other headstart facilities, and was not modified to accelerate growth. To entice juveniles to emerge for feeding, water was placed into a water dish first and then liberally poured at the entrance of burrows. After watering and evidence of juvenile activity, a mix of native annuals (when available) and moistened ZooMed Grassland Tortoise diet pellets was placed in a food dish in the shade. During weekly feedings, a census was collected of present individuals. Every individual was weighed, measured (Figure 8), evaluated for health, and offered a water bath quarterly while in headstart pens. Where necessary, ant abatement efforts were employed to minimize mortality of juvenile desert tortoises due to ants.

Figure 8: Measuring a Juvenile Tortoise

A researcher collecting measurement data for a juvenile tortoise using calipers.

Source: Ron Swaisgood, San Diego Zoo Wildlife Alliance

Release Site Selection and Habitat Data Collection

Potential release points were randomly generated at both EAFB and Ward Valley. Random points were spaced 20 m apart for each study site and vegetation community type: EAFB Baker Nunn (creosote scrub), EAFB Leuhman Ridge (yucca woodland dominated by *Yucca brevifolia*), Ward Valley low (creosote scrub), Ward Valley high (yucca woodland dominated by *Yucca schidigera*) (Figure 4). A 200 m buffer around known tortoise locations at EAFB was generated and these areas were excluded as potential release sites. At each of the randomly generated points, a pre-release screening of micro-habitat variables was conducted within a 2 m x 2 m plot, centered at the nearest potential burrow where a tortoise could be released. If no release burrow could be identified within 5 m of the random point, the site was designated as unsuitable, and a different random point was chosen. At each potential release point, habitat features were recorded, including rodent burrow abundance, wash presence, substrate texture, presence of coppice mounds, coppice mound area, and vegetation cover type. Coppice mounds are small sand dunes that typically form around vegetation like creosote bushes due to wind.

For post-release habitat surveys, habitat data were also collected in a 2 m x 2 m quadrat around each tortoise detection location (see Radio-Telemetry Tracking in the following section) and at a randomly selected set of points at each release site. This procedure allows for characterization of both tortoise-inhabited (used) and overall available habitat, which can then be used to calculate tortoise habitat preference metrics.

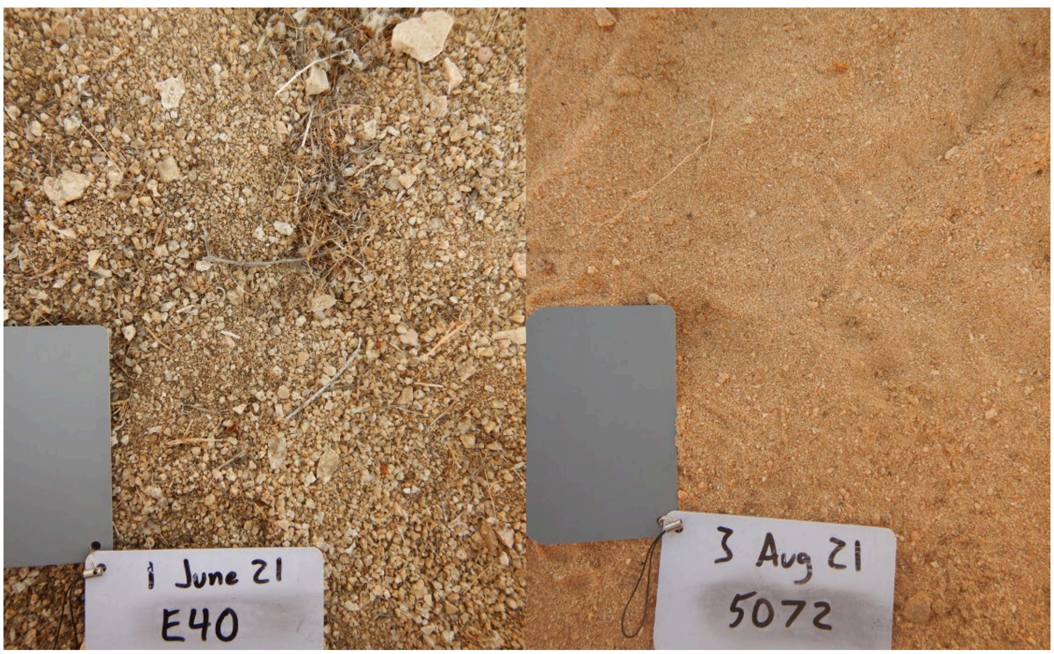
Vegetation Community

For each habitat survey 2m quadrat, visual estimates of coverage by shrubs or perennial plant species, annual forage plant species, and invasive plant species were recorded, as were the dominant shrub or perennial species, annual forage species, and invasive plant species.

Substrate

For each habitat survey 2m quadrat, visual estimates of rock and bare ground coverage were recorded, as were substrate texture/rock size (sand, small pebbles, or large cobbles), and data regarding whether a coppice mound or a wash was present within the quadrat. Finally, data were collected on substrate color, either by using a Munsell soil color chart, or by taking a photograph of substrate near the tortoise location using a Nikon Coolpix camera with a gray card standard, and thereafter quantifying color using ImageJ (Figure 9).

Figure 9: Substrate Differences at Desert Tortoise Sites



Example photos showing differences in substrate at two tortoise locations. The photo on the left shows substrate with sand and pebble while the right photo shows sand.

Source: Daniel Essary, San Diego Zoo Wildlife Alliance

Burrow Availability

For each habitat survey, the number of rodent burrows within each 2 m^2 quadrat was counted. Whenever tortoises were located, it was recorded whether they were in a tortoise burrow (dome-shaped), a rodent burrow (round), or in a different microhabitat (such as in the open, under vegetation, in rocks).

Figure 10: Desert Tortoise Burrows

Photograph showing dome-shaped tortoise burrow.

Source: Ron Swaisgood, San Diego Zoo Wildlife Alliance

Radio-Telemetry Tracking

Prior to release, tortoises were fitted with a VHF radio transmitter (Holohil, Inc., model BD-2 or PD-2) attached to the 4^{th} or 5^{th} vertebral scute using epoxy (Figure 11). All juvenile tortoises were released into the wild at EAFB and Ward Valley between 3-6 October, 2020.

Figure 11: Desert Tortoise With VHF Radio-Transmitter

Photo showing transmitter placement on 5th vertebral scute for a juvenile desert tortoise.

Source: Ron Swaisgood, San Diego Zoo Wildlife Alliance

After being translocated into the wild, tortoises were monitored at least once per month. During the active period (October; March through May) tortoises were monitored twice per month. When tortoises were initially translocated and whenever transmitters were subsequently replaced, tortoises were checked within 24 hours. On each visit, a receiver (Telonics TR-8) and a Yagi 3-element antennae were used to locate each released tortoise. After confirming the tortoise's location, geographic coordinates were collected using a sub-meter accuracy geolocation unit (Juniper Systems, Geode). In cases where visual confirmation of the tortoise was possible, data on tortoise behavior was collected (for example basking, resting, eating). Habitat data were then collected at the tortoise location, including visual estimates of habitat cover categories, counts of burrows, description of substrate, and more (Table 1). Habitat data was also collected at points that were not occupied by tortoises to characterize habitat available to tortoises in each study area. These available habitat points were randomly generated within a minimum convex polygon around all tortoise locations at each site, and available habitat plots were surveyed in late spring and summer 2021.

Table 1: Habitat Data Collected

Variable Name	Description
# Burrows	Count of rodent or tortoise burrows in quadrat
Coverage by invasive plants	Visual estimate of proportion of quadrat covered by invasive plants
Coverage by perennials/shrubs	Visual estimate of proportion of quadrat covered by perennial/shrub plants
Coverage by annual forage plants	Visual estimate of proportion of quadrat covered by annual forage plants
Coverage by bare ground	Visual estimate of proportion of quadrat covered by bare ground
Coverage by rocks	Visual estimate of proportion of quadrat covered by bedrock/large rocks larger than a juvenile tortoise
Substrate category	Presence of each of the following substrate categories within the quadrat: sand, small pebbles (less than 65 mm), large cobbles (more than 65 mm)
Coppice Mound	Presence of a coppice mound within the quadrat

Above habitat data was collected for a 2m^2 quadrat centered around each tortoise location or each random point.

Source: San Diego Zoo Wildlife Alliance

To avoid disturbing animals during winter dormancy, target transmitter replacement dates were set approximately five months after release at the start of the spring active period, in early March 2021. During transmitter change-outs, a full health assessment was conducted, including weighing and measuring tortoises, monitoring them for any signs of injury or health issues, and taking multiple photographs of each individual.

In cases where tortoise transmitters were missing or no signal was detected, an intensive search protocol was initiated at the tortoises' last known location. During intensive searches, water was sprayed around all tortoise-shaped burrows within a 30 m radius, and each burrow was checked with a flashlight. Intensive searches were targeted to follow 1-3 days of optimal weather with temperatures between 75-95 degrees F. Missing radio-frequencies were regularly scanned for from vantage points within each study area.

Data Analysis

All analyses were conducted in R (version 4.0.3, R Core Team 2020) and ArcGIS Pro (version 2.8.0 ESRI 2021). Across study questions, generalized linear mixed models (GLMM) were implemented in the 'lme4' package (Bates et al. 2014), using the 'lmerTest' package to assess significance of individual fixed effects (Kuznetsova et al. 2017). For all GLMMs, continuous variables were rescaled prior to analysis to allow for comparison of beta estimates. The 'DHARMa' (Hartig 2017) and 'performance' (Lüdecke et al. 2021) packages were used to confirm that models met assumptions (such as normality and uniformity of residuals, no multicollinearity of fixed effects). For home range estimation, the 'adehabitatHR' package was used (Calenge 2006).

Growth in Captivity and Post-release—To describe pre-release growth, measures of MCL and mass were plotted as a function of age/date, and summary statistics on tortoise growth were calculated for each cohort. GLMMs were also fit with either MCL or mass as the response variable that included cohort (hatched 2018 or hatched 2019), age (#days since emergence from natal burrow), and an interaction between cohort and age as fixed effects, and maternal identity and tortoise identity as random effects to test for differences between cohorts. To describe post-release growth, additional GLMMs were fit with either MCL or mass as the response variable that included age, time point (pre or post release; only one post-release measurement was used), and their interaction as fixed effects, and maternal identity, tortoise identity, and site as random effects. In these models average habitat features used by each individual were included, including information on substrate type, shrub cover, and number of burrows (the full set of habitat features could not be included in these models due to limited sample size and collinearity issues; post-release measurement data was only available for a subset of the study individuals).

Post-release Movement—To describe post-release space use, 95 percent minimum convex polygons (MCP) and 95 and 50 percent kernel density home ranges were calculated for all individuals with more than 5 telemetry locations. Linear distance from release location to each individual's 50 percent fixed kernel home range centroid was calculated to assess site fidelity, the tendency of animals to maintain a home range or remain close to their release location. Home range size (hectares [ha]) was examined for each estimator as a function of hatch year, release site, vegetation community type, and important habitat components including mean coverage by coppice mounds, shrubs, forbs, and invasive vegetation averaged across observations for each tortoise. GLMMs were fit with Mother ID and site as random effects to account for variation in the data.

To describe movement, cumulative distance travelled from the release location as well as final settlement distance (defined as the distance from the release site to the last known location) were calculated. The 'trajr' package (McLean & Skowron Volponi 2018) in R was used to describe speed and straightness/sinuosity of movement. Specifically, straightness (total cumulative distance travelled divided by displacement distance from first to last point) and sinuosity scores were calculated for each individual. Three GLMMs were fit that included as the response variable either cumulative distance moved, straightness, or sinuosity of movement trajectories for each individual. For each individual average used habitat traits were calculated (substrate type; number of burrows; coverage by shrubs) and included as fixed effects in models (the full set of habitat features could not be included in these models due to limited sample size and collinearity issues). Maternal identity and release site were included as random effects. For the model of cumulative distance travelled date of last detection was also included as a fixed effect to control for differences in available number of days of data across individuals. To meet GLMM assumptions, the cumulative distance moved measure was square-root transformed prior to analysis.

Habitat Selection—To describe habitat selection in juvenile desert tortoises, GLMMs were used to compare the features of used (at tortoise detection sites) versus available (random points) habitat survey points. A binomial GLMM was fit with use category (used versus available) as a binary response variable and habitat features (number of burrows; coverage by bare ground, invasive plants, shrubs, and annuals; substrate category; and presence/absence of a coppice mound) as fixed effects, and site as a random effect.

Survival Analysis—To test for the effects of age class and habitat features on tortoise survival, the (Therneau 2015) 'survival' package in R was used to fit Cox proportional hazards models. Data was used from tortoises that were either confirmed dead or were known to be alive at the time of analysis (and did not include tortoises that were missing due to malfunctioning transmitters, for which statuses were unknown). Due to a limited number of documented mortalities and remaining "known status" individuals, it was not possible to include a large number of fixed effects in a single model without violating assumptions. Thus, separate models were fit containing the following fixed effects that were most expected to impact survival: age class, number of burrows (more than two per plot or two and fewer per plot), and habitat type (creosote vs. yucca woodlands), and all pairwise combinations of these variables. For all models it was confirmed that data met the proportional hazards assumption.

CHAPTER 3: Project Results

Female Collection

In 2018, tortoise searches and tracking began at the end of March and continued through the beginning of July. Radio transmitters were attached to 16 adult female tortoises at EAFB and 16 at Ward Valley. Each tortoise was tracked and radiographed weekly to determine egg development. Once they were determined to be gravid with shelled eggs, they were transported to the headstart facility at either Edwards AFB or Cadiz. Egg production is summarized in Table 2.

Table 2: Desert Tortoise Headstarting Outcomes by Year and Site

	EAFB	EAFB	Ward	Ward
	2018	2019	2018	2019
Adult females tracked	16	16	16	23
Females transported to facility to lay eggs	15	13	11	15
Eggs laid or induced	73	72	40	50
Hatch rate (percent)	89	88	0	67
Juveniles translocated	59	57	0	28

Table summarizing outcomes of female monitoring and headstarting activities.

Source: Data compiled by Talisin Hammond, San Diego Zoo Wildlife Alliance

In 2019, searches for additional adult females in Ward Valley were conducted on March 23-24 and resulted in 6 adult females being added to the study. A total of 21 adult females in Ward Valley were marked and tracked through the nesting (oviposition) season. In April 2019 monitoring of adult female tortoises at EAFB and Ward Valley began once per week for egg development. Two additional adult females in Ward Valley were marked and added to the study. A total of 23 adult females in Ward Valley and 16 adult females at EAFB were tracked through the nesting (oviposition) season. One of the adult female tortoises at Ward Valley was found dead on June 19.

In 2019 a total of 15 adult females from Ward Valley were transported to TLD for egg induction; induction of oviposition was successful in 14 of the 15 females, resulting in 50 eggs. Out of the 50 eggs induced, four eggs broke during oviposition and one was an extremely small, non-viable egg, leaving 45 eggs incubating in the indoor incubators at TLD. While most females responded to the hormonal induction, one female only partially responded to the treatment (laying only one of her three eggs) and another female did not respond at all and held onto her five eggs. Eggs were extracted post-mortem from a gravid female that was found dead, but none of the eggs were viable and are not included in the total. At EAFB, 13 of the 16 adult females were brought to the headstart pens to lay their eggs. A total of 72 eggs were laid by these 13 females.

Headstarting

2018 cohort-At the EAFB headstart facility, a total of 73 eggs were laid by 15 of the 16 adult females in 2018. At the Cadiz headstart facility, there were a total of 40 eggs (including one broken egg after hormonal induction) laid by 11 of the 16 females. Unfortunately, there was complete nest failure at Cadiz in 2018, and no juveniles were produced for the older cohort. The nest failure was attributed to an extreme heat event with temperatures in excess of 110 degrees Fahrenheit (°F) for multiple consecutive days. Temperatures at Cadiz were significantly higher than those at EAFB, with maximum temperatures in June and July exceeding 114.8 °F at Cadiz. It has been shown that multiple days of sustained high temperatures (greater than 98.6 °F) is associated with nest failure (Brian Todd, U.C. Davis, personal communication). Spotila et. al. (1994) also found that incubation at 95.5 °F was lethal for 72 percent of the eggs. Although it was proposed that rocky soils may have led to shallow nests and contributed to the egg loss, no significant difference was found in nest depth between EAFB and Cadiz. Based on these findings, high temperatures were likely the major factor for the complete nest failure at Cadiz.

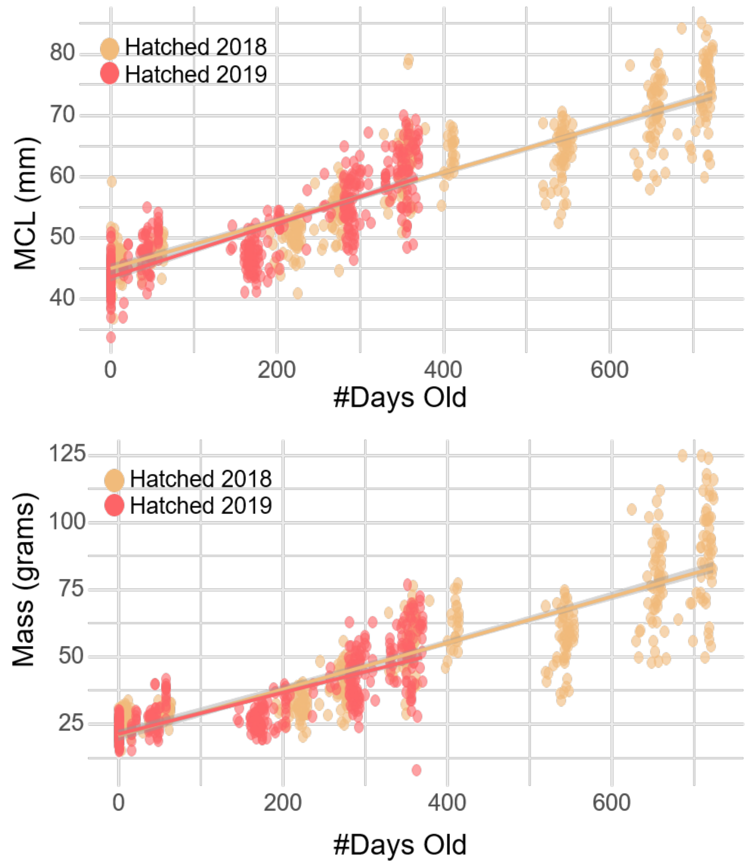
This setback precipitated a programmatic change to incubate 2019 eggs collected at Ward Valley indoors at TLD. At EAFB, 65 of the 73 eggs hatched, with one of the eggs containing twins. Ultimately, 59 individuals from this group survived to be translocated into the wild at EAFB.

2019 cohort-In 2019, the first hatchlings at EAFB emerged on September 11. By mid-September, a total of 63 hatchlings had emerged; eight eggs were confirmed not to have hatched, one hatchling died within the nest chamber, and one hatchling died shortly after emergence due to unknown causes. Ultimately, 57 animals from this group survived to be translocated into the wild at EAFB.

The first eggs from Ward Valley hatched on July 22 after 70 days of incubation. This is within the range of normal incubation time, which can range from 68 to 125 days (Rostal & Jones 2002). Only 30 of the 45 incubated eggs hatched, resulting in a 67 percent hatching success. A number of the remaining eggs appeared to have been unfertilized and exhibited no development, while some showed only early development before they failed. Unfortunately, one of the eggs that hatched contained twins that were nonviable. Twenty-nine hatchlings were transported from TLD to the Cadiz headstart facility on October 8, 2019. Ultimately, 28 animals from this group survived to be translocated into the wild at Ward Valley.

Tortoises from all cohorts showed significant growth over time in captivity (Table A1, A2; Figure 12). There were not significant differences in the rate of change in mass across the two cohorts, though the 2019 cohort did tend to be slightly smaller than the 2018 cohort in their first year of life (Table A1). In approximately the first year of life, tortoises hatched in 2018 grew from an average initial post-emergence measurement of 46.5 ± 3.13 mm (mean ± S.D.) to 63.0 ± 5.17 mm after the first year, while initial post-emergence measurements were 43.9± 2.79 mm compared to 60.9 ± 4.9 mm pre-release for tortoises hatched in 2019, when individuals were one year of age. These rates of growth (about 17 mm/year) are comparable to previous outdoor headstarting projects with this species (Tuberville et al. 2019; McGovern et al. 2020). The 2018 cohort exhibited slightly lower growth rates in their second year of life, and prior to release this cohort was on average 74.0 ± 6.0 mm in length.

Figure 12: Pre-release Growth Curves for Juvenile Tortoises



Top panel shows changes in MCL and bottom panel shows changes in mass over time for juvenile tortoises hatched in 2018 (gold) and 2019 (pink) prior to translocation.

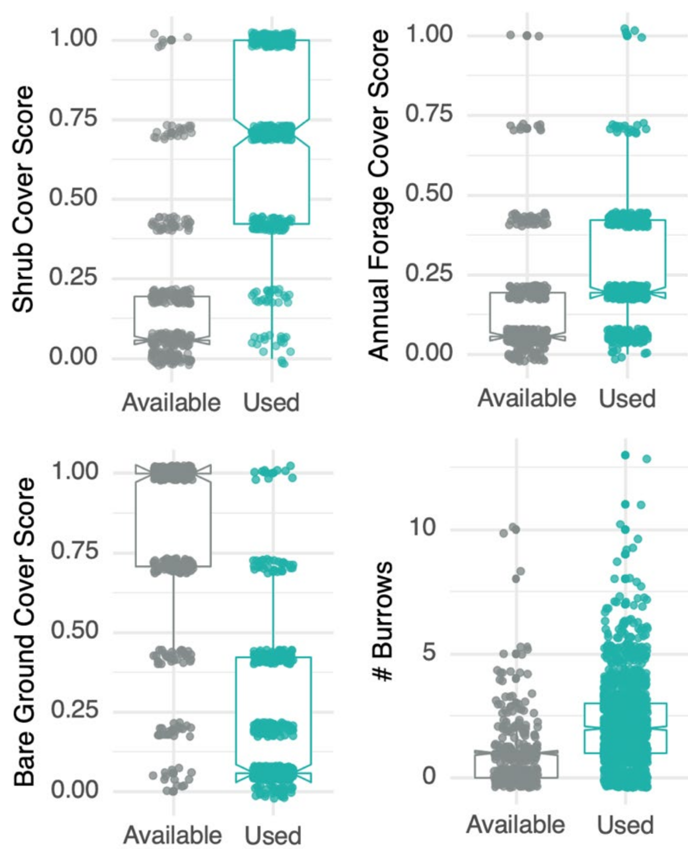
Source: Talisin Hammond, San Diego Zoo Wildlife Alliance

While it was planned to release tortoises in April 2020, releases were delayed until fall 2020 due to the COVID-19 pandemic and resulting closures at EAFB. Between 3-6 October 2020, 116 individuals (59 hatched in 2018 and 57 hatched in 2019) were released into the wild at two release sites (Baker Nunn and Leuhman Ridge) on EAFB, and 28 individuals (all hatched in 2019) were translocated into the wild at two release sites (Ward High and Ward Low) at Ward Valley.

Habitat Selection by Tortoises

There were significant differences in habitat traits at locations occupied by released tortoises in comparison to randomly selected locations (available habitat at the site; Table A-3). Specifically, tortoises appeared to select habitat with a significantly larger number of burrows, lower proportion of bare ground cover, and higher proportion of perennial/shrub cover (Figure 13). Plots used by tortoises also tended to have significantly more sand substrate in comparison to other substrate types and were significantly more likely to contain coppice mounds (Figure 14). There was no significant difference in invasive plant coverage at used versus available sites (Table A-3; Figure 15).

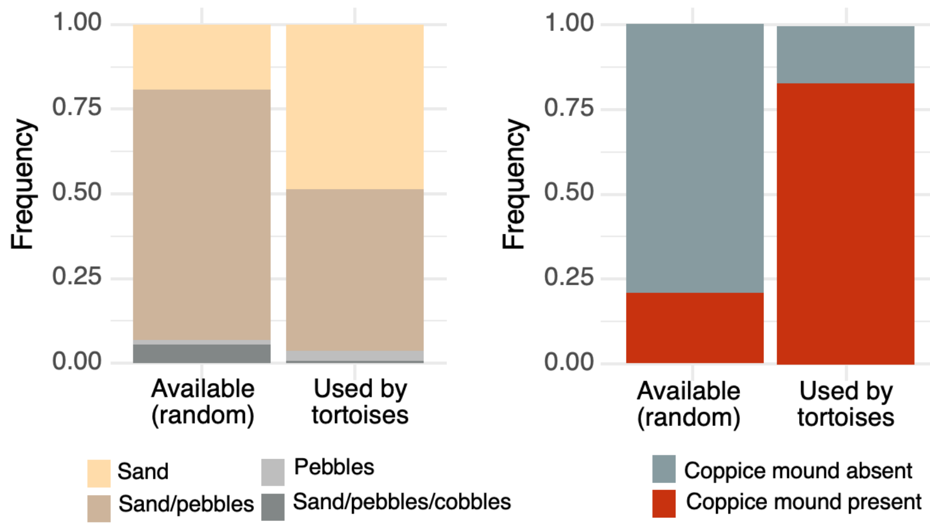
Figure 13: Boxplots Showing Habitat in Available Versus Used Survey Quadrats



Boxplots showing proportion of shrub cover (top left), annual forage cover (top right), and bare ground cover (bottom left), and number of burrows (bottom right) in available (random, grey) and used (tortoise inhabited, teal) survey quadrats.

Source: Talisin Hammond, San Diego Zoo Wildlife Alliance

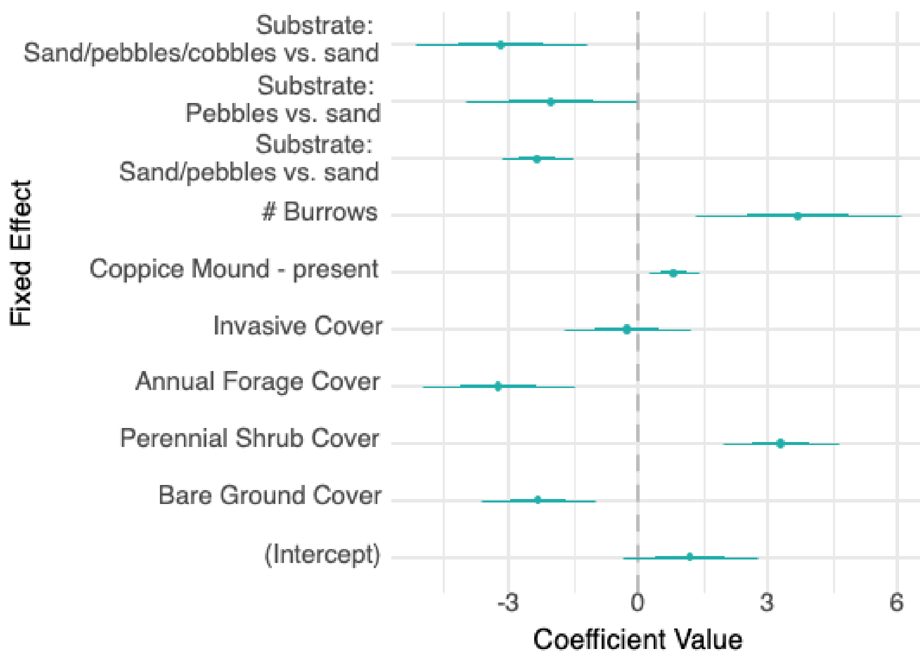
Figure 14: Stacked Bar Plot Showing Substrate and Coppice Mound Categories In Available (Random) Versus Tortoise-Occupied Locations



Stacked bar-plot showing proportion of substrate categories (left) and coppice mound categories (right) at available (random) habitat survey points on the left of each panel, and in habitat surveys at known tortoise locations on the right of each panel.

Source: Talisin Hammond, San Diego Zoo Wildlife Alliance

Figure 15: Model Results Characterizing Used Versus Available Habitat



Coefficient plot showing generalized linear mixed model (GLMM) results indicating the relationship among habitat variables and sites tortoises used relative to what is available to them. Coefficient values that do not cross the dotted zero line are considered to have significant effects on tortoise habitat selection; values greater than 0 are positively associated with tortoise use in comparison to random sites, and values less than 0 are negatively associated with tortoise use.

Source: Talisin Hammond, San Diego Zoo Wildlife Alliance

Post-Release Growth and Movement

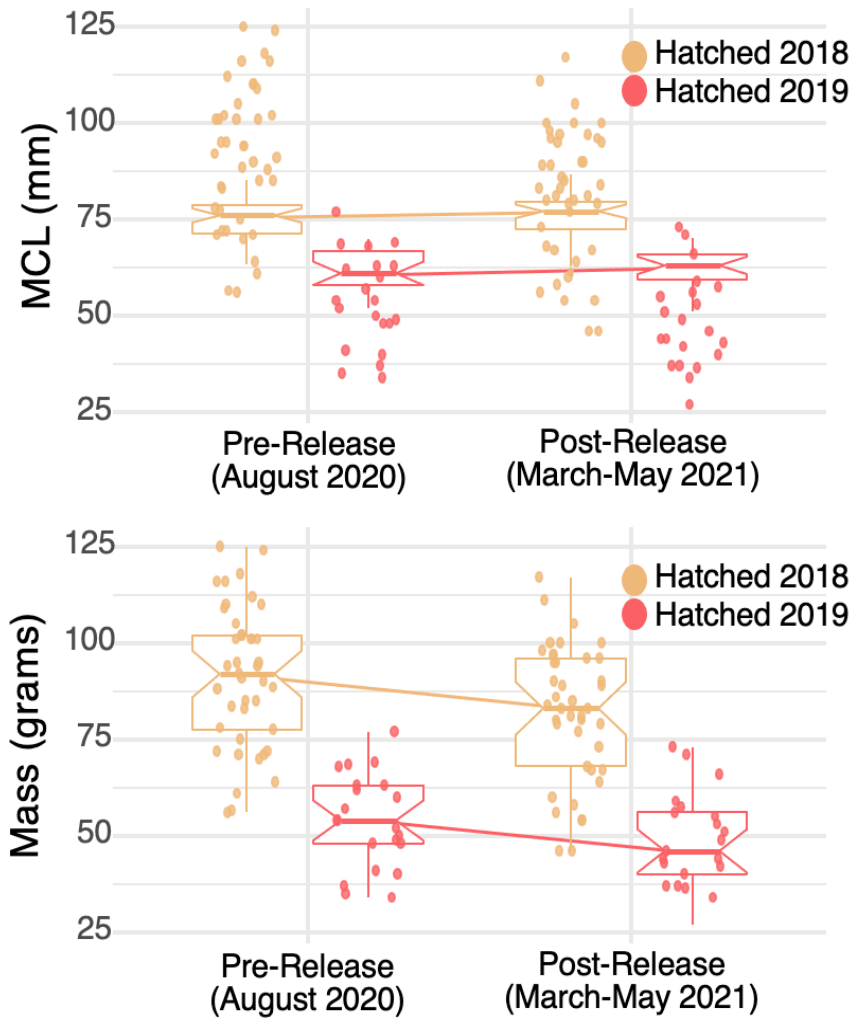
Post-Release Growth

Tortoises of both age classes showed limited changes in MCL and mass after release (Figure 16). There were no significant increases in MCL post-release documented and no changes in the rate of growth across age classes (Table A4). Low magnitude but significant decreases in mass were documented post-release, with no significant differences in the rate of change across age classes. Because tortoises were inactive (overwintering underground in burrows) for a large portion of the monitoring period, low growth rates are expected. However, others (for example Tuberville et al. 2019b) have documented an MCL growth rate of 10.7 mm/year for juvenile tortoises released to the field as hatchlings in the Mojave National Preserve, California. Across age classes and within and across sites, tortoises with lower MCLs and masses tended to inhabit sandier areas (Tables A-4, A-5). This observed association could be due to the fact that sandy substrates are easier to dig or travel in, but the data do not allow these hypotheses to be directly tested.

There were no significant impacts of burrow availability or shrub cover on post-release growth. Due to the extreme drought conditions in the Mojave Desert throughout late 2020-2021

(Figures 17 and 18), it is likely that tortoises had limited foraging opportunities and thus have not yet exhibited substantial growth post-release.

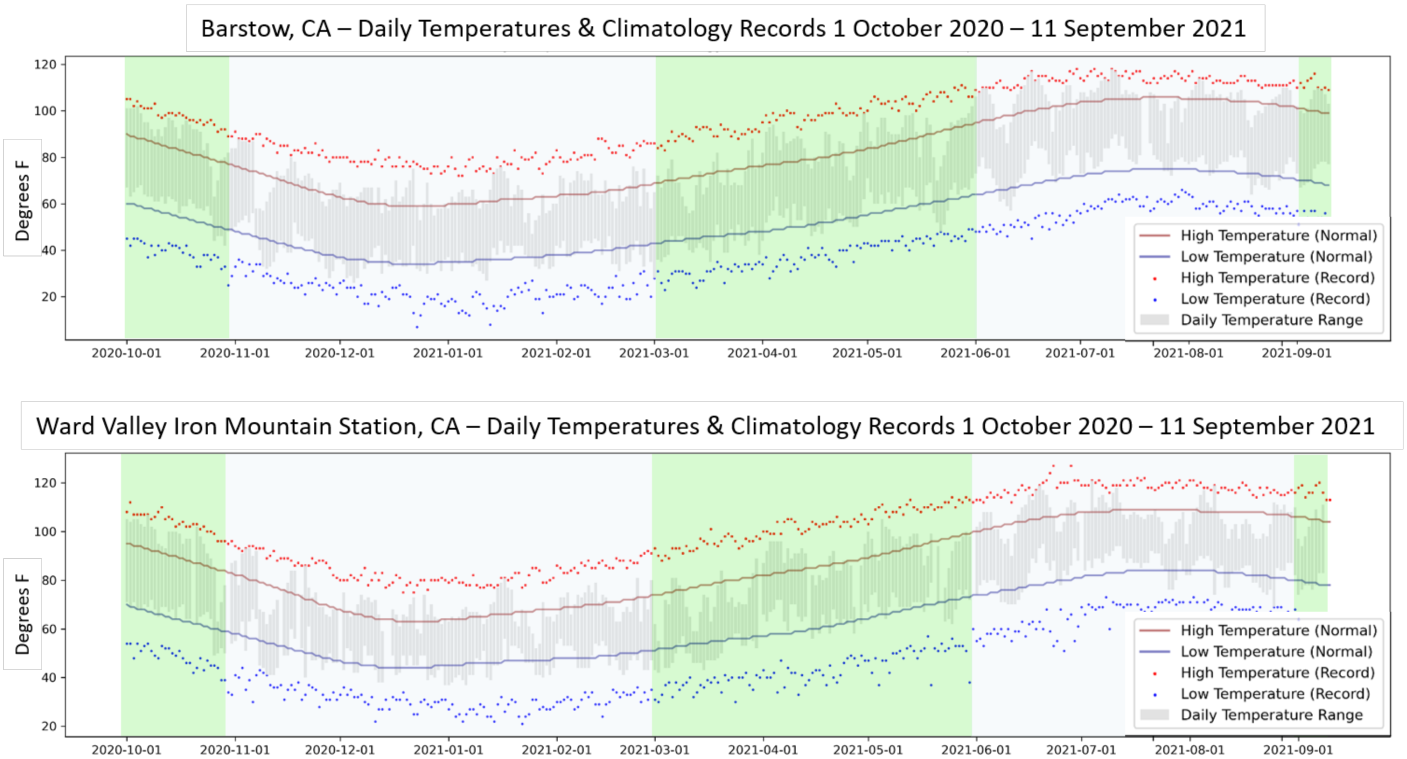
Figure 16: Juvenile Tortoise Pre-Versus Post-Release MCL and Mass



Boxplots showing MCL (top panel) and mass (bottom panel) for juvenile desert tortoises hatched in 2018 (gold) and 2019 (pink). The final pre-release measurement is shown in the left side of each panel, with post-release data collected approximately 7-9 months later on the right.

Source: Talisin Hammond, San Diego Zoo Wildlife Alliance

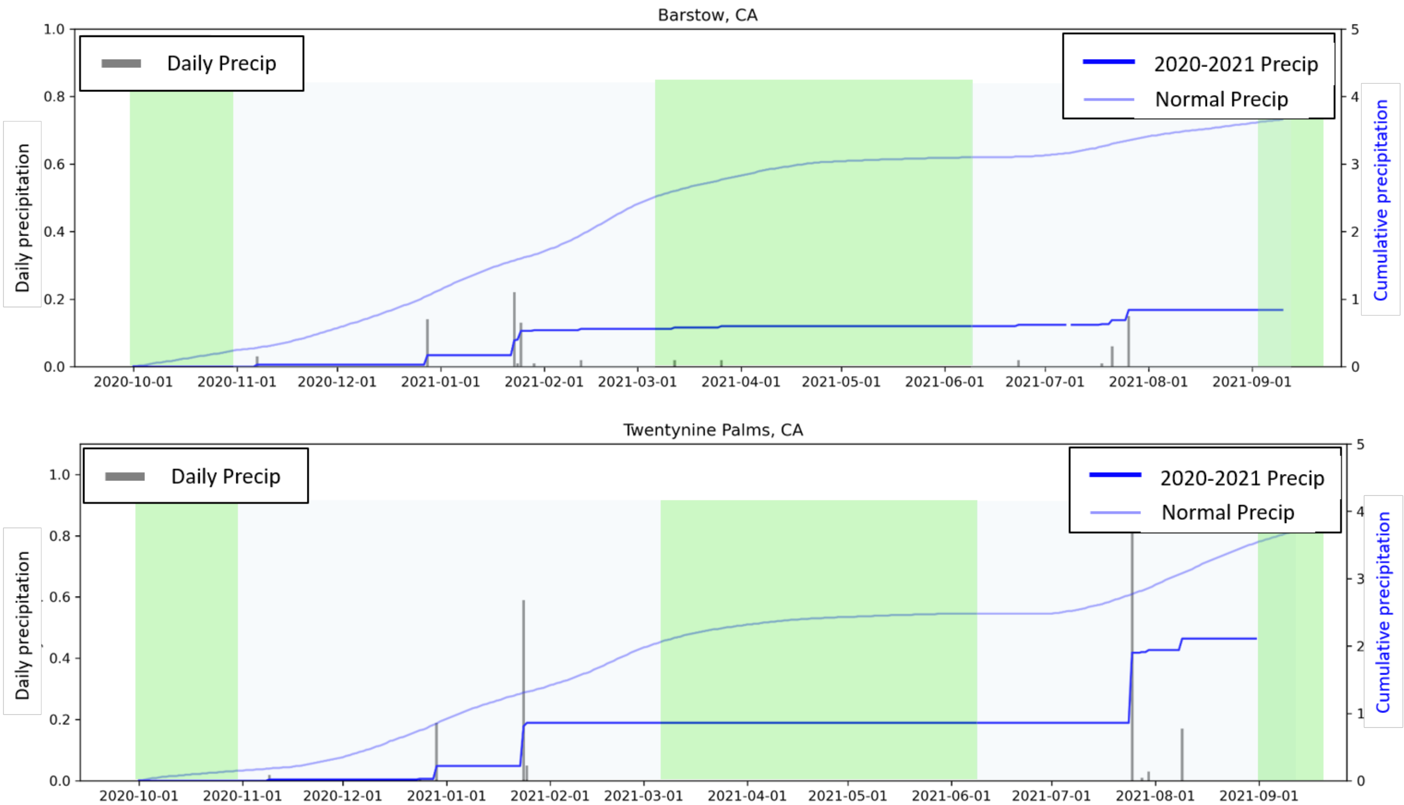
Figure 17: Post Release Temperature Data



Temperature plots showing observed daily minimum and maximum temperatures post tortoise release (gray bars) relative to normal mean daily high (red line) and low temperature (blue line), and record high (red dot) and low (blue dot) temperatures across the period of record (1943- present) for Barstow, California (top panel) and Ward Valley, California (bottom panel). Green shading represents tortoise active periods.

Source: Melissa Merrick, San Diego Zoo Wildlife Alliance

Figure 18: Post Release Precipitation Data



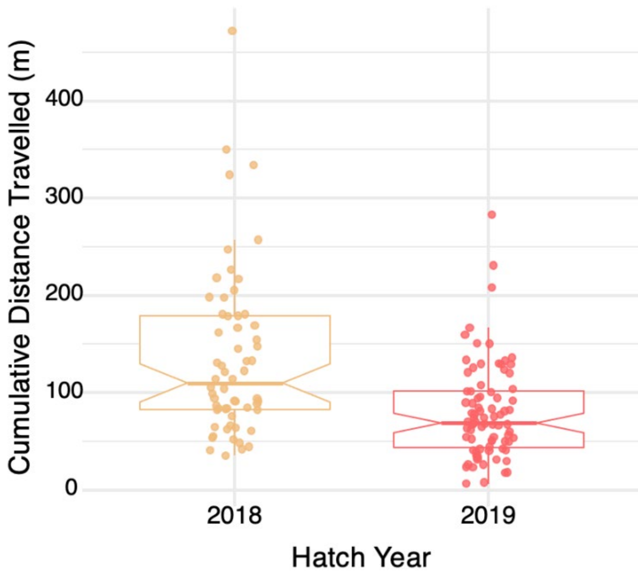
Precipitation plots showing daily (gray bars; inches), cumulative precipitation post tortoise release (dark blue curve; inches) relative to normal cumulative precipitation during the period of record (1943- present) for Barstow, California (top panel) and Twentynine Palms, California (bottom panel). Green shading represents tortoise active periods.

Source: Melissa Merrick, San Diego Zoo Wildlife Alliance

Post-Release Movement and Home Range Analysis

Post-release movement by tortoises depended on age class. The 2019 cohort travelled significantly shorter distances than the (older) 2018 cohort (Table A-6; Figure 19), but the 2019 cohort had significantly more sinuous paths than the 2018 cohort (Table A-7). Individual movement was highest in October immediately following release prior to winter dormancy in November 2020; the 2019 cohort moved 45.5 ± 38.7 m (mean ± S.D.) during this period while the 2018 cohort moved cohort moved 66.8 ± 50.6 m. During the overwinter period (November through February) cumulative movement was generally low (on average 18.7 ± 26.6 m). During the spring active period (March through June), tortoise movement was slightly higher (on average 30.0 ± 32.0 m). Movement distances were not significantly predicted by habitat features (Table A6). On average, settlement distances (defined as the straight-line distance between tortoise release locations and their last known locations) were 40.9 ± 40.5 m (mean ± S.D.; range: 0.66-233 m).

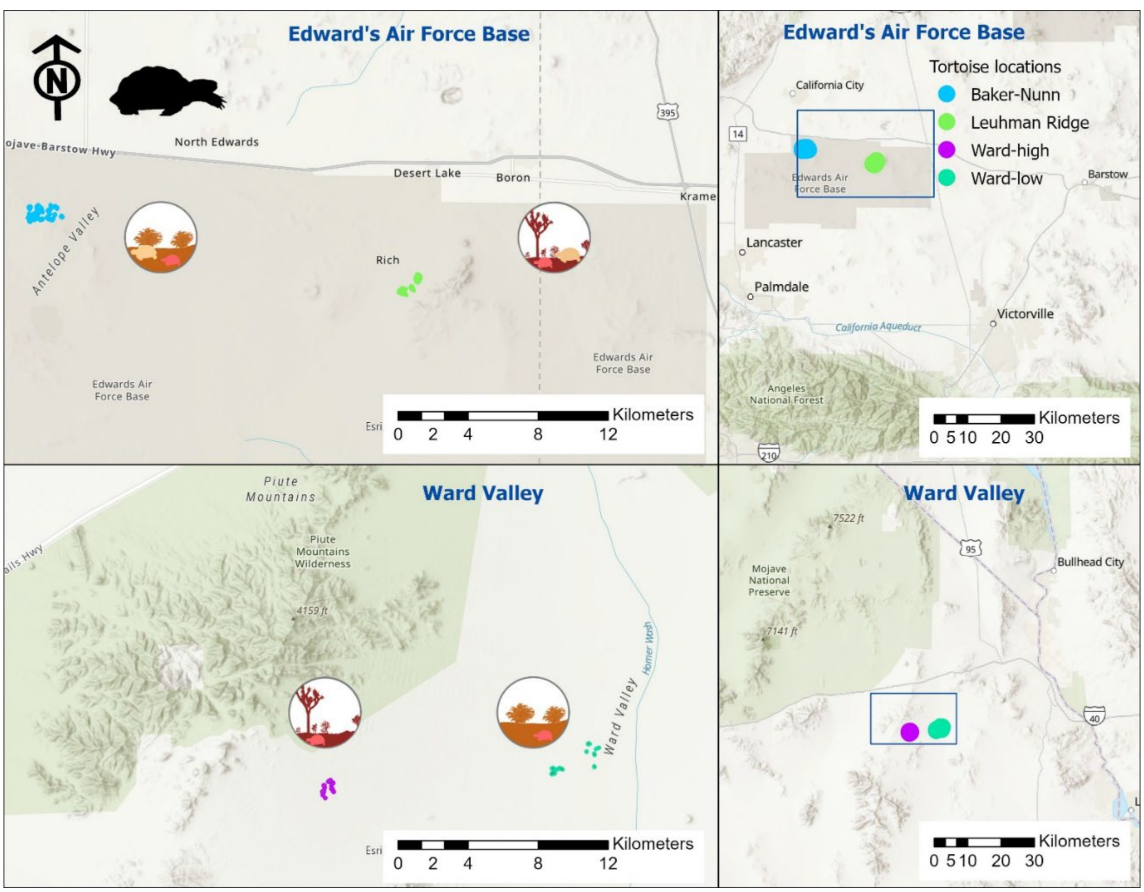
Figure 19: Juvenile Tortoise Post-Release Movement Distances



Cumulative distances travelled (October 2020 through August 2021) post-release by tortoises hatched in 2018 (left, gold) and 2019 (right, pink).

Source: Talisin Hammond, San Diego Zoo Wildlife Alliance

Figure 20: Overview of Juvenile Locations



Map showing overview of juvenile desert tortoise post-release telemetry locations.

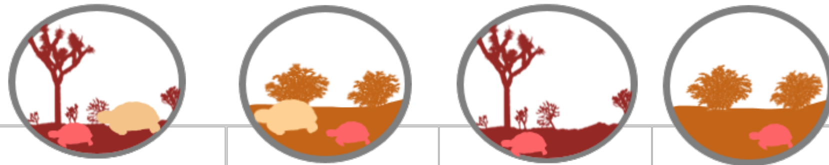
Source: Melissa Merrick, San Diego Zoo Wildlife Alliance

Home Range and Site Fidelity or Displacement

Best practice for estimating home range size requires a minimum of five observed locations. Of 144 released juvenile tortoises, 137 individuals met this threshold. Across home range estimators (kernel density and minimum convex polygon) home range sizes tended to be slightly larger in the creosote scrub sites (Baker-Nunn and Ward Low) compared to the yucca woodland sites (Leuhman Ridge and Ward High) and for tortoises hatched in 2018 compared to 2019 (released as 2-year-olds versus 1-year olds) (Table 3; Figures 21-24). The effect of hatch year was only significant for 95 percent minimum convex polygon home ranges (Table 3). Tortoises released as 2-year-olds established home range centers farther (35.2 m) from their original release site compared to tortoises released as 1-year olds (23.4 m). Across age classes, tortoises released in creosote scrub sites established home range centers farther from their release sites (Baker-Nunn: 37.2 m; Ward Low: 18.8 m) compared to tortoises released in yucca woodlands (Leuhman Ridge: 25.3 m; Ward High: 12.3 m). Note that only 1-year-old tortoises were released at Ward Valley sites.

Habitat variables collected within each individual tortoise's home range did not explain variation in home range size for any estimator (Table A-9; Figure 25).

Table 3: Desert Tortoise Home Range Estimates by Year and Site

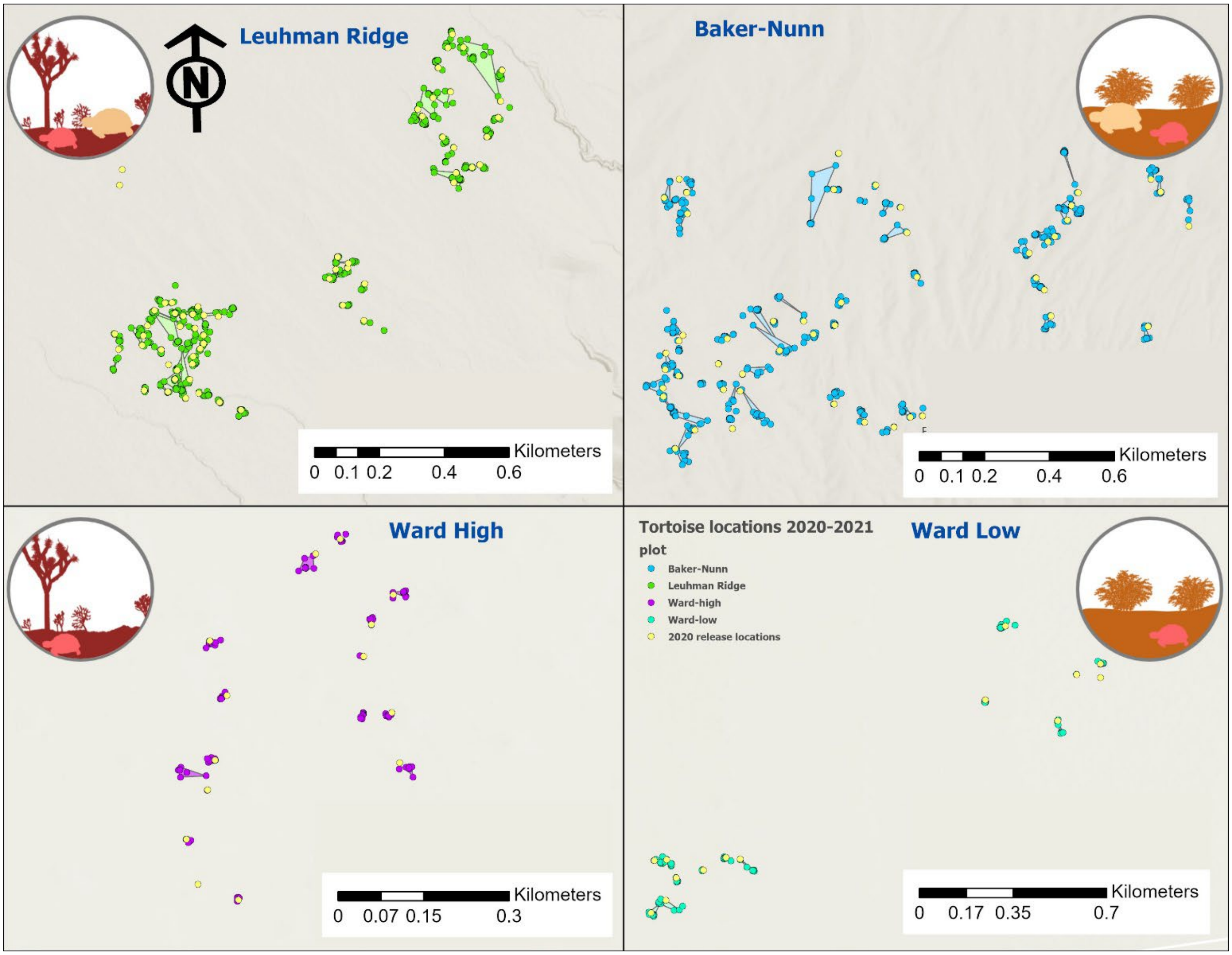


Estimator	Leuhman Ridge	Baker-Nunn	Ward High	Ward Low	$F_{3,133}$	*p*
50% KDE	0.09 ± 0.15 (0.004 - 0.62)	0.13 ± 0.29 (0.002-2.08)	0.02 ± 0.03 (0.002 - 0.10)	0.07 ± 0.08 (0.002 - 0.26)	1.67	0.33
95% KDE	0.45 ± 0.78 (0.02 - 3.60)	0.63 ± 1.21 (0.01 - 8.19)	0.09 ± 0.14 (0.01 - 0.54)	0.32 ± 0.36 (0.01 - 1.12)	1.40	0.25
95% MCP	0.05 ± 0.11 (0.001 - 0.78)	0.06 ± 0.09 (0.001 - 0.59)	0.01 ± 0.01 (0.001 - 0.04)	0.03 ± 0.04 (0.001 - 0.12)	1.18	0.32
					KW H_3	***p***
Displacement distance (m)	25.3 ± 30.7 (0.0 - 158.05)	37.2 ± 43.4 (0.0 - 192.22)	12.3 ± 15.3 (0.0 - 57.30)	18.8 ± 19.9 (0.0 - 63.86)	9.12	0.03
	Hatch year					
Estimator	**2018**	**2019**	**$F_{1,135}$**	***p***		
50% KDE	0.11 ± 0.15 (0.004 - 0.65)	0.09 ± 0.25 (0.002 - 2.08)	0.35	0.55		
95% KDE	0.58 ± 0.85 (0.027 - 3.66)	0.41 ± 1.00 (0.007 - 8.19)	1.16	0.28		
95% MCP	0.07 ± 0.11 (0.001 - 0.78)	0.03 ± 0.07 (0.001 - 0.59)	5.88	0.02*		
			KW H_1	***p***		
Displacement distance (m)	35.2 ± 41.1 (0.0 - 182.75)	23.4 ± 30.1 (0.0 - 192.22)	4.58	0.03		

Table summarizing mean home range area in hectares by estimator (50 percent, 95 percent kernel density estimator (KDE), 95 percent minimum convex polygon), by site and hatch year (mean (ha) ± standard deviation; range). Displacement distance is the mean distance (m) between individual tortoise release sites and the centroid of their 50 percent kernel density home range (mean (m) ± standard deviation; range). Because of the difference in sample size among sites, a Kruskal Wallis test was considered the most suitable method to compare data given the distributions of the data.

Source: Data compiled by Melissa Merrick, San Diego Zoo Wildlife Alliance

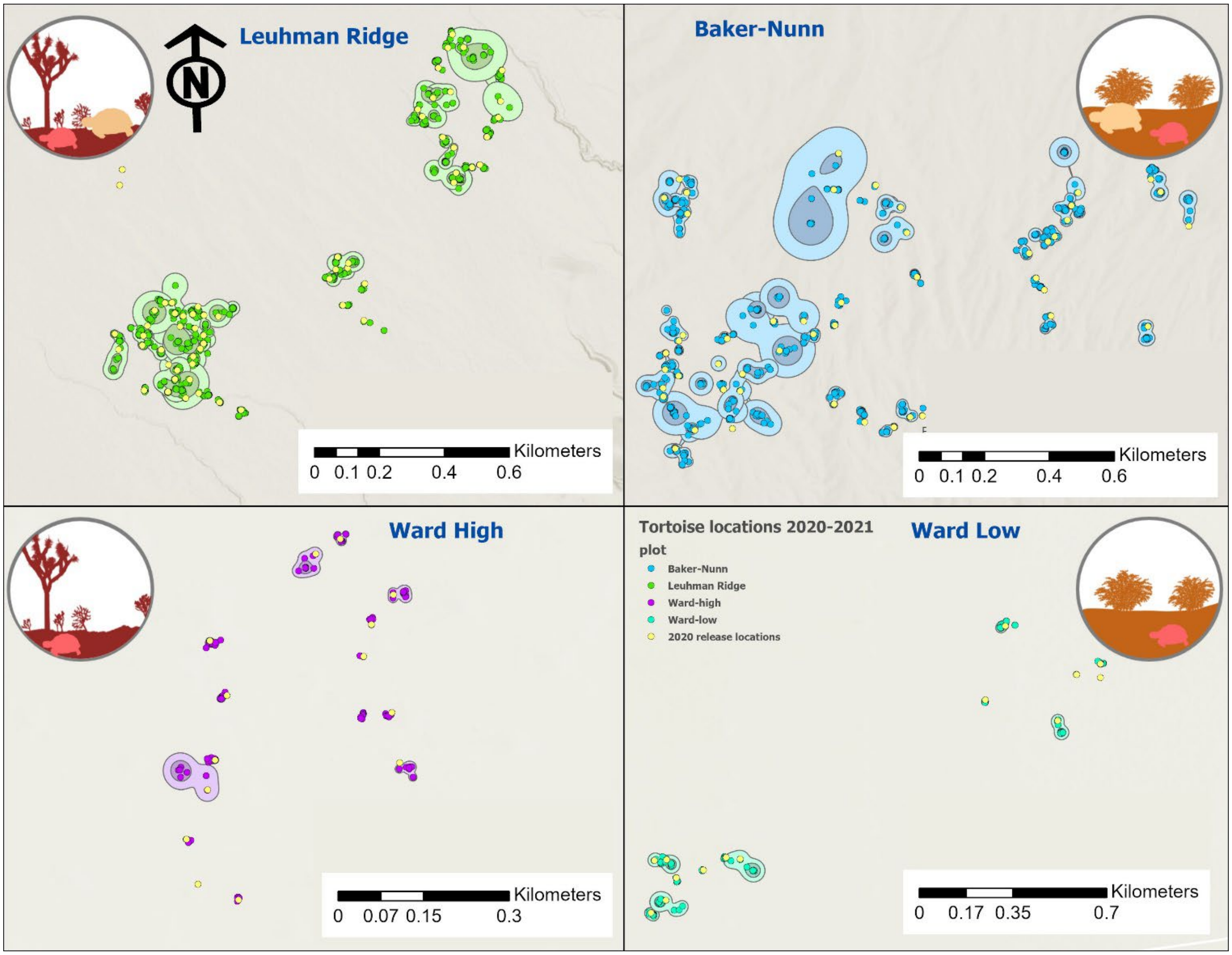
Figure 21: Overview of Juvenile Minimum Convex Polygons



Map showing overview of juvenile desert tortoise post-release telemetry locations and 95 percent minimum convex polygons. Individual release locations are shown as yellow circles.

Source: Melissa Merrick, San Diego Zoo Wildlife Alliance

Figure 22: Overview of Juvenile 50 Percent and 95 Percent Fixed Kernel Home Ranges



Map showing overview of juvenile desert tortoise post-release telemetry locations with 50 percent and 95 percent fixed kernel density home range estimates. Individual release locations are shown as yellow circles.

Source: Melissa Merrick, San Diego Zoo Wildlife Alliance

Figure 23: Comparison of Juvenile 50 Percent and 95 Percent Fixed Kernel Home Range Size

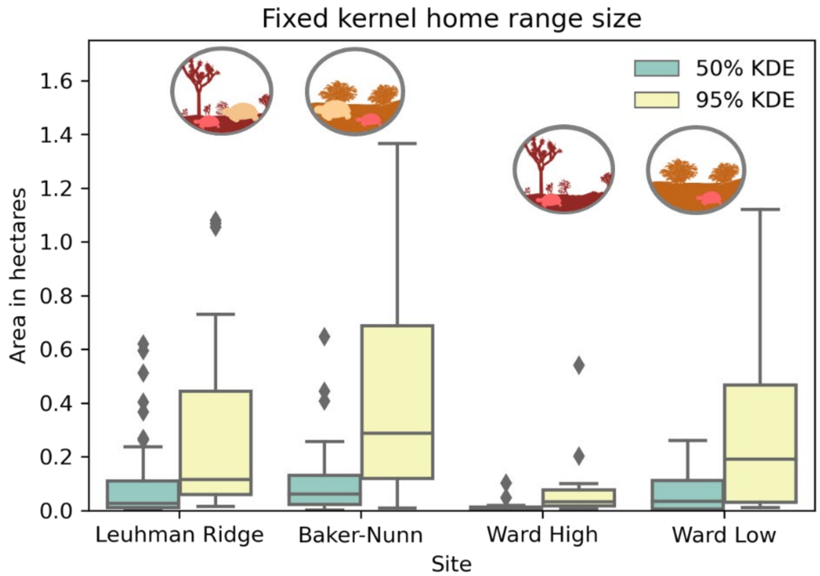


Figure showing size differences in juvenile desert tortoise 50 percent and 95 percent fixed kernel density home ranges by site.

Source: Melissa Merrick, San Diego Zoo Wildlife Alliance

Figure 24. Comparison of Juvenile 95 Percent Minimum Convex Polygon Home Range Size

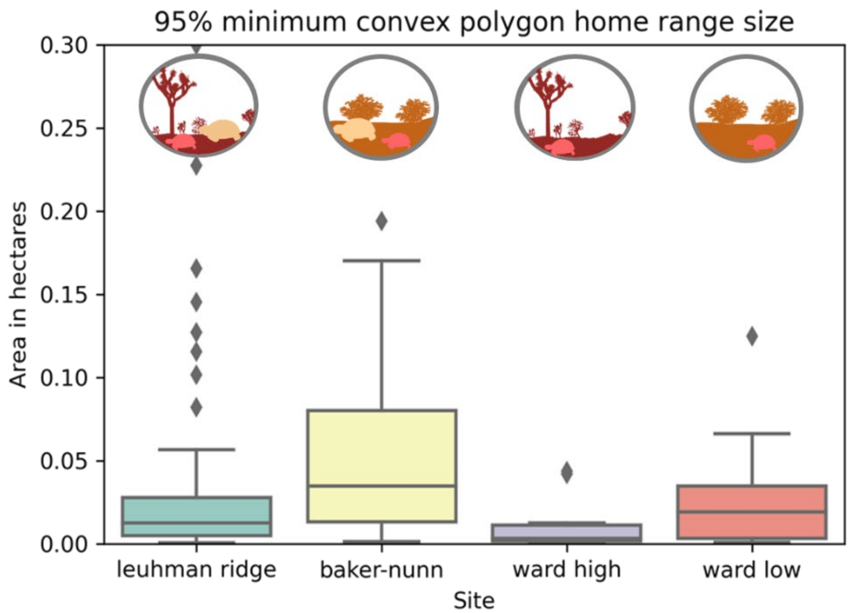
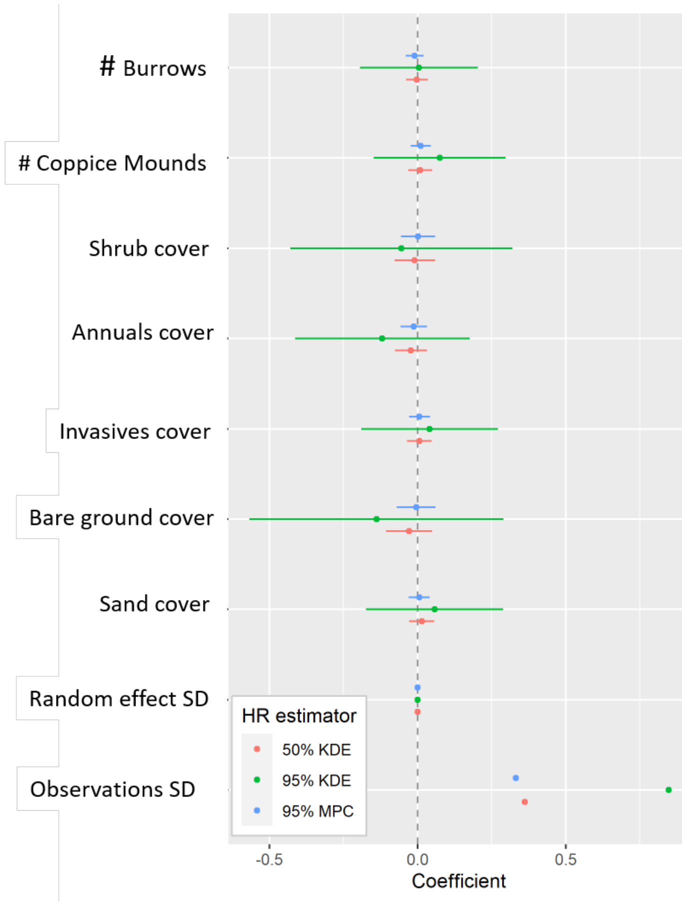


Figure showing size differences in juvenile desert tortoise 95 percent minimum convex polygon home ranges by site.

Source: Melissa Merrick, San Diego Zoo Wildlife Alliance

Figure 25. Model Results Characterizing Home Range Size as a Function of Habitat Variables



Coefficient plot showing generalized linear mixed model (GLMM) results characterizing home range size as a function of habitat variables within an individual's home range.

Source: Melissa Merrick, San Diego Zoo Wildlife Alliance

Survival Analysis

Unfortunately, by early March 2021, most of the smaller Holohil BD-2 transmitters (applied to the yearling cohort hatched in 2019) experienced suspected premature battery failure. Given the specifications indicated by Holohil, lifespan of the transmitters should have exceeded six or even seven months. Missing frequencies were searched for at least once per month and intensive searches were conducted for these individuals. Support for the hypothesis that transmitter battery failure was at play and documentation of attempts to search for and recover missing tortoises is presented in Appendix B.

While search results suggest that premature transmitter failure may account for some of the missing tortoises, numerous predation events were documented, and it is possible that some missing individuals have been depredated and transmitters moved out of range due to the large home ranges of many tortoise predators (such as coyotes, ravens). There were 14 confirmed mortalities documented during tortoise tracking activities (found carcasses or parts of carcass; Table 4). In many cases it was not possible to determine a cause of death, but in some cases (6/14) predation was suspected, most often due to canine predators like kit foxes.

Of the 144 tortoises released, 49 are confirmed alive as of the beginning of September 2021, nine had detached transmitters and their status is currently unknown, and 74 remain missing. Of tortoises with known status (alive or carcass found), a roughly 78 percent survival rate was documented (49/63). This rate is comparable to other one year post-release survival rates for this species (for example, McGovern et al. 2020).

Table 4: List of Documented Mortalities

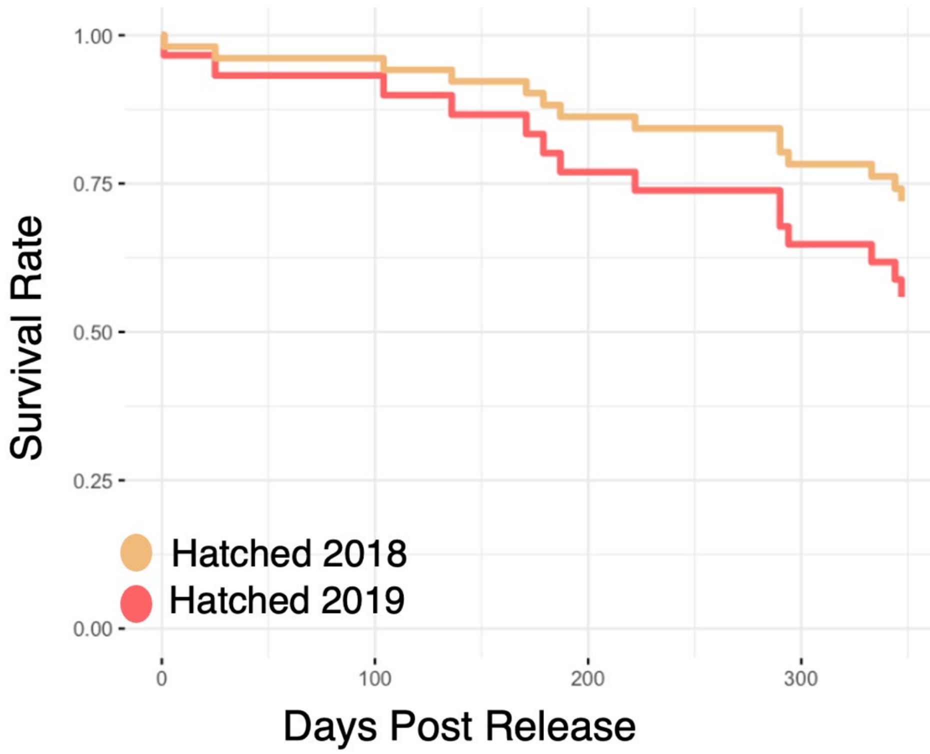
ID	Date Found Dead	Description
E55	2020-10-05	possible kit fox predation
5037	2020-10-28	A cause of death could not be determined.
5049	2021-01-15	A cause of death could not be determined. Adequate adipose tissue was seen histologically. Small numbers of nematodes present.
E58	2021-02-16	No signs of predation. A cause of death could not be determined. Extensive fungal colonization of cranial coelomic viscera appeared to be postmortem. Adequate adipose stores were seen histologically.
5045	2021-03-24	desiccated, no signs of predation.
5065	2020-04-01	no signs of predation.
5074	2020-04-08	puncture wounds, likely canine predation.
5071	2021-05-13	possible kit fox predation.
E19	2021-07-21	nearly complete carapace with broken edges and transmitter still attached.
5015	2021-07-21	partial carapace and pieces of plastron (with chewed edges), and detached transmitter.
W17	2021-07-27	radio transmitter and one scute were recovered on the ground, and fresh digging outside of the tortoise burrow entrance was observed, suggesting that a predator likely dug the individual out of the burrow.
5072	2021-09-01	no signs of predation.
E38	2021-09-13	Entire shell and transmitter found on ground - body appears mummified/completely desiccated. No apparent sign of predation.
5057	2021-09-15	Transmitter found glued a single scute, no other remains found.

Dates and accounts of mortalities of translocated juvenile tortoises.

Source: Data compiled by Talisin Hammond and Melissa Merrick, San Diego Zoo Wildlife Alliance

With the limited number of mortalities documented to date, it is still early to conduct survival analyses. Preliminary model comparison results are presented (Table A-10). Likely due to the limited statistical power, no significant predictors of survival were found in any models, and there were low delta Akaike Information Criteria (AIC) values across models, indicating uncertainty about meaningful predictors of survival (Table A-10). There were no significant differences in survival based on age class, though there was a non-significant pattern ($p = 0.31$) of older animals exhibiting higher survival (Figure 26). Data on tortoises and habitat will continue to be collected to test for other predictors of survival in the future.

Figure 26: Survival Curves Showing Survival Rate for Juvenile Tortoises



Survival curves estimated from a Cox proportional hazards models for tortoise survival over time. Curves show survival rate as a function of age class (2018 hatch year in gold, 2019 hatch year in pink). Age was not significantly predictive of survival in this model.

Source: Talisin Hammond, San Diego Zoo Wildlife Alliance

CHAPTER 4: Knowledge Transfer Activities

Knowledge Gained

This research identified key microhabitat characteristics that are important for juvenile tortoise resource selection and began to identify relationships between habitat variables and post-release movement and survival in translocated juvenile tortoises. Findings suggest that tortoises may preferentially select habitat with higher burrow, shrub, and coppice mound availability, and that they may have preferences for specific substrate types. While there is not yet evidence for any significant effects of these habitat features or of age at release (one versus two years) on survival, further data on long-term fitness of these cohorts will be collected in the future (continued work on these cohorts has been funded by other grants). Due to the longevity and slow life history of the Mojave Desert tortoise, impacts of age at release and habitat on demographic variables may take more time to manifest. Findings do suggest that home-range sizes and travel distances are higher for two-year-olds in comparison to one-year olds, which could have important implications for calculations of carrying capacity, release site selection, target densities of translocations, and other factors pertinent to tortoise conservation.

Target Audiences

These results could be pertinent to several audiences, including:

- Wildlife agencies including the USFWS and CDFW that regulate mitigation and monitoring of wildlife at renewable energy facilities.
- Organizations including zoos involved in tortoise recovery, headstarting, and translocation projects.
- Public and tribal land managers and military base environmental and engineering staff leading habitat restoration or protection efforts and seeking guidance on habitat variables that are pertinent to desert tortoise recovery.
- Solar energy developers and operators and their environmental consultants involved in mitigating the impacts of renewable energy and other sources of development.
- Scientists researching strategies for conservation and recovery of the Mojave desert tortoise.
- Interested members of the public who are invested in conservation of local wildlife species like the Mojave Desert tortoise.

Transfer Activities

Incorporating key stakeholders into project implementation–Throughout this work, coordination has taken place with regulatory agencies, land managers, developers, and other project partners for all major project activities. CDFW, USFWS, US Air Force, local agencies, scientists, and energy industry were part of the technical advisory committee (TAC). TAC committee meetings were held on October 11, 2017, October 2, 2018, October 24, 2019 to seek advice on methods, overcoming challenges such as the total nest failure at Cadiz in 2019, and disseminating new knowledge generated.

Status reports and presentations–Annual status reports have been provided to project partners. The results of this project will be made publicly available and will be provided to local, state, and federal agencies (for example CDFW, U.S. Bureau of Land Management, USFWS). These agencies will be able to directly use these research results to improve California's mitigation guidelines.

Public outreach–This research was highlighted in a recent article in the San Diego Zoo Wildlife Alliance's Journal, a popular science publication. The tortoise program was featured by the San Diego Zoo Wildlife Alliance's Community Engagement department as an educational activity involving tortoise tracking and conservation. Several presentations have been given to local schoolchildren. For example, in April 2021, an outreach activity was conducted for Escondido, California middle schools as part of their conservation science learning module. Desert tortoise research was discussed in relation to species conservation.

Presentations–Activities were shared with a broader audience through a number of presentations. These include presentations at the United States Fish & Wildlife Service Mojave Desert Tortoise Coordination Meeting, the American Association of Zookeepers, the San Diego Zoo Wildlife Alliance's Advanced Inquiry Program's Master's students, several university and professional meetings, and the University of California Irvine's widely attended conference on *Extinction: Solutions for Species on the Brink*. In January 2022 results from this study were shared with a large group of policy makers and land managers at the Mojave Desert Tortoise Habitat Restoration Workshop, convened by Clark County, Nevada, the U.S. Fish & Wildlife Service, and Natural Resource Conservation, LLC. Findings will be presented at the Desert Tortoise Council Symposium in February 2022.

Manuscripts–At least one scientific manuscript is currently in preparation covering findings from this project, testing the relationships between tortoise age and habitat and post-release movement and survival. This manuscript will likely be submitted for publication in late 2022 after additional data are collected. Any resulting publications will be shared with stakeholders.

CHAPTER 5: Conclusions/Recommendations

Summary of Major Findings

The objectives of this work were to headstart juvenile desert tortoises to different ages (one and two years of age) while evaluating health and growth in captivity pre-release, to translocate tortoises into the wild into different vegetation community types with careful attention to release site microhabitat characteristics, and to conduct post-release monitoring to test for influences of age class and release site on post-release growth, movement, survival.

This project set out to determine whether husbandry conditions cause behavioral or health aberrations that negatively affect survival. The tortoises reared in captivity (ex situ) appeared to have good health. Pre-release growth and survival rates and post-release survival of these individuals were comparable to other studies of ex situ reared tortoises.

Work with adult females showed that tracking and regularly x-ray scanning adult females is a viable strategy for finding gravid individuals and collecting eggs to incubate and headstart.

Field work documented significant habitat selection by tortoises. Habitat surveys suggested that tortoises prefer environments with less bare ground, more shrub cover, and higher availability of burrows and coppice mounds. Tortoises also appeared to prefer sandier substrates over substrates with pebbles and cobbles. Coppice mounds contain friable soil that facilitates digging, and are thought to be maintained in part by digging activities of small mammals and tortoises (Lee 1986; Soulard et al. 2013). Mounds are associated with increased vegetative cover which can provide favorable microclimates for tortoises as well provide nutrients and maintain seed banks of forage species important for tortoises (Soulard et al. 2013) and have a high abundance of small mammal burrows available to juvenile tortoises for shelter.

Tortoises released at two-years-old exhibited greater post-release dispersal and larger home range sizes than those released at one-year-old, with greater movement occurring immediately following release in comparison to other times of year. Home range sizes tended to be slightly larger in the creosote scrub sites compared to yucca woodland sites and for tortoises hatched in 2018 compared to 2019. Animal home range size is determined in part by body size and resource availability (McLoughlen & Ferguson 2000; Jetz et al. 2004), and these results could suggest that yucca woodland provides more resources, such as burrows and forage, compared to creosote scrublands. Juvenile tortoise home ranges are small. Compared to adult tortoises, whose home range size can range from 1-53 hectares (O'Connor et al. 1994; Dutcher et al. 2020), juvenile tortoise home ranges are two orders of magnitude smaller. The fact that juvenile tortoises established small home ranges in close proximity to their release site could indicate that careful selection of optimal habitat components (shrubs, coppice mounds, and rodent burrows) was important in anchoring individuals to a site, potentially reducing vulnerability to predation. That mean displacement distance between release site and home range center was slightly larger for tortoises released in creosote scrub compared to yucca woodland vegetation communities may suggest that among vegetation community types, yucca woodland results in stronger site fidelity. Because post-release dispersal is one of the biggest challenges for conservation translocations (Berger-Tal et al. 2020), monitoring the influence of release site habitat characteristics on the magnitude of post-release movements warrants further study.

Larger tortoises have the ability to move about more, and this could confer competitive advantages (such as greater foraging and burrow options). Previous work has suggested that more exploratory tortoises are better able to find burrows and survive in the wild (Germano et al. 2017). If distances travelled correlate with older age classes or exploratory individuals, it could be that two-year-olds have a higher chance of finding burrows and surviving. Results do not yet support higher survival of two-year-old versus one-year-old tortoises, however, these cohorts will continue to be monitored in the future. In the first-year post-release there were no statistically significant differences in survival across age classes or habitat types. However, given the longevity of tortoises, such patterns may appear in later years, and these individuals will continue to be monitored as part of a long-term research program.

Applications and Recommendations

Based on this research, recommendations can be made regarding selecting suitable habitat for desert tortoise. When development from renewable energy or other source impacts desert tortoise habitat, it is important that there is like-for-like mitigation to conserve or restore equally suitable habitat. If mitigation sites contain poorer habitat, then development will have a net negative outcome for the species and contribute to its increasing endangerment, making future development more difficult and costly. As part of the mitigation process, it is recommended that presence and volume of coppice mounds, rodent and tortoise burrow density, substrate, bare ground and shrub cover be included as habitat assessment metrics at both the development and release sites. This will allow the assessment of the quality of habitat that will be impacted. In some cases, this information could be used to decide not to develop a potential site (i.e., when the habitat is highly suitable). In other cases, these metrics set the criteria for the quality of habitat that needs to be conserved or restored to ensure that mitigation fully offsets impacts to the species.

New insights into relative habitat quality for desert tortoise can also be used to improve translocation outcomes. Releasing tortoises into areas that include higher levels of the resources identified in this study will have a higher probability of success. Although survival data from this study have not yet yielded statistically significant results, habitat preferences typically correlate with survival and population vital rates, so it is plausible that tortoises released into areas with preferred habitat (more burrows, coppice mounds, sandier soil and shrub cover and less bare ground) will be more likely to establish and contribute to population growth.

This work also informs guidelines for site selection for headstart facilities. Although the Cadiz facility falls within desert tortoise range, it experienced multiple consecutive days of excessive heat, which increased temperatures experienced by eggs in underground nests, leading to 100 percent egg mortality. Under climate change scenarios forecasting increasing summer temperatures throughout the desert tortoise range, marginal areas at lower elevations may be expected to experience such heat waves with increasing frequency. This may lead to loss of suitable habitat for tortoises. In addition, managers and policy makers selecting sites for preservation or for headstart facilities should avoid these areas forecast to experience excessive heat, favoring areas in cooler locations and higher elevations that will remain more resilient in the face of climate change. When a headstart facility is located in an area likely to experience excessive heat, remedial actions will need to be taken to reduce soil temperatures, such as shade provision.

Headstarting can be an important tool to offset renewable energy development impacts to desert tortoise if appropriate release site characteristics are selected.

CHAPTER 6: Benefits to Ratepayers

This study is expected to provide two potential benefits to California's electricity ratepayers: lowering costs of electricity and environmental benefits. Minimization of impacts of solar energy development on species through mitigation actions can be very expensive, particularly when listed species, such as the Mojave Desert tortoise, are involved. Identifying areas of lower conservation value for the placement of renewable energy facilities should reduce risk of costly project delays, less probability of environmental or conservation-based lawsuits directed at energy developers, and greater success of state and federally mandated mitigation strategies. The knowledge gained through this project may lower costs by making headstart mitigation more cost-effective and by reducing obstacles to future renewable energy deployment. Best management practices for headstarting juvenile tortoises, including releasing them in higher quality habitat, can reduce their time spent in captivity. This saves in the cost of rearing each tortoise but can also increase the number of tortoises that can be headstarted. In addition, being able to determine the conservation value of potential solar energy sites will encourage development on lower value sites that will require less mitigation than high value sites.

New scientific knowledge about the habitat features that allow tortoises to thrive can be applied to guidance for wildlife agencies about which land parcels to protect and set more meaningful restoration targets. As such, a better understanding of what constitutes ideal habitat, particularly for juvenile desert tortoises, not only supports responsible land management, but it also allows for increased specificity with management targets for conservation, restoration, and identifying sites that will optimize translocation success. Implementation of more effective recovery practices may increase the probability of de-listing the desert tortoise from the endangered species list in the future. Facilitating development of solar energy in the Mojave Desert will also help California achieve its goals for 100 percent clean energy and reducing greenhouse gas emissions caused by burning fossil fuels.

Research on the environmental effects or damages associated with energy infrastructure development is rarely funded by the competitive and regulated markets. In the case of how to optimize mitigation of listed species across an entire industry, the research required would typically be beyond the means or responsibility of a single solar energy project. Therefore, this study called for public-interest energy research funding through the EPIC program.

GLOSSARY OR LIST OF ACRONYMS

Term	Definition
°F	Degrees Fahrenheit
AIC	Akaike Information Criteria
CDFW	California Department of Fish and Wildlife
CEC	California Energy Commission
CI	Confidence interval
EAFB	Edwards Air Force Base
EPIC	Electric Program Investment Charge
GLMM	Generalized linear mixed model
Ha	Hectares
KDE	Kernel density estimate
Km	Kilometer
M, m^2	Meter, meters squared
Mm	Millimeter
MCL	Midline carapace length
MCP	Minimum convex polygon
SD	Standard deviation
SDZWA	San Diego Zoo Wildlife Alliance
TAC	Technical Advisory Committee
TLD	The Living Desert Zoo and Gardens
USFWS	United States Fish and Wildlife Service
USGS	United States Geological Survey
VHF	Very High Frequency

REFERENCES

Agha M, Lovich JE, Ennen JR, Todd BD. 2020. Wind, sun, and wildlife: Do wind and solar energy development "short-circuit" conservation in the western United States? Environmental Research Letters **15**.

Allen CH. 2001. It's time to give Kemp's ridley head-starting a fair and scientific evaluation. Available from http://www.seaturtle.org/mtn/archives/mtn56/mtn56p21.shtml (accessed January 9, 2021).

Averill-Murray RC, Darst CR, Field KJ, Allison LJ. 2012. A new approach to conservation of the mojave desert tortoise. BioScience **62**:893–899.

Averill-Murray RC, Esque TC, Allison LJ, Bassett S, Carter SK, Dutcher KE, Hromada SJ, Nussear KE, Shoemaker KT. 2021. Connectivity of Mojave desert tortoise populations - Management implications for maintaining a viable recovery network: U.S. Geological Survey Open-File Report 2021–1033.

Battin J. 2004. When good animals love bad habitats: ecological traps and the conservation of animal populations. Conservation Biology **18**:1482–1491. Available from http://doi.wiley.com/10.1111/j.1523-1739.2004.00417.x.

Berger-Tal O, Blumstein DT, Swaisgood RR. 2020. Conservation translocations: a review of common difficulties and promising directions. Animal Conservation **23**:121–131.

Berry KH, Yee JL, Lyren L, Mack JS. 2020. An uncertain future for a population of desert tortoises experiencing human impacts. Herpetologica **76**:1–11.

Bloxam QMC, Tonge SJ. 1995. Amphibians: suitable candidates for breeding-release programmes. Biodiversity and Conservation **4**:636–644.

Boarman WI, Patten MA, Camp RJ, Collis SJ. 2006. Ecology of a population of subsidized predators: Common ravens in the central Mojave Desert, California. Journal of Arid Environments **67**:248–261.

Brown DR, Merritt JL, Jacobson ER, Klein PA, Tully JG, Brown MB. 2004. Mycoplasma testudineum sp. nov., from a desert tortoise (*Gopherus agassizii*) with upper respiratory tract disease. International Journal of Systematic and Evolutionary Microbiology **54**:1527–1529.

Brown MB, Schumacher IM, Klein PA, Harris K, Correll T, Jacobson ER. 1994. Mycoplasma agassizii causes upper respiratory tract disease in the desert tortoise. Infection and Immunity **62**:4580–4586.

Bubac CM, Johnson AC, Fox JA, Cullingham CI. 2019. Conservation translocations and post-release monitoring: Identifying trends in failures, biases, and challenges from around the world. Biological Conservation **238**:108239. Elsevier. Available from https://doi.org/10.1016/j.biocon.2019.108239.

Bureau of Land Management US. 2016. Executive summary for the record of decision for the land use plan amendment to the California Desert Conservation Plan, Bishop Resource Management Plan, and Bakersfield Resource Management Plan.

Calenge C. 2006. The package “adehabitat” for the R software: A tool for the analysis of space and habitat use by animals. Ecological Modelling **197**:516–519.

Chaparro-Pedraza PC, de Roos AM. 2020. Density-dependent effects of mortality on the optimal body size to shift habitat: Why smaller is better despite increased mortality risk. Evolution **74**:831–841.

Cunninghame F et al. 2015. Conserving the critically endangered mangrove finch : Head-starting to increase population size. Page Galapagos Report 2013-2014. GNPD, GCREG, CDF and GC. Puerto Ayora, Galapagos, Ecuador.

Daly JA, Buhlmann KA, Todd BD, Moore CT, Peaden JM, Tuberville TD. 2018. Comparing growth and body condition of indoor - reared , outdoor - reared , and direct-released juvenile Mojave desert tortoises. Herpetological Conservation and Biology **13**:622–633.

DeGregorio B, Moody R, Myers H. 2020. Soft release translocation of Texas horned lizards. Animals **10**.

Dutcher KE, Vandergast AG, Esque TC, Mitelberg A, Matocq MD, Heaton JS, Nussear KE. 2020. Genes in space: what Mojave desert tortoise genetics can tell us about landscape connectivity. Conservation Genetics **21**:289–303. Springer Netherlands. Available from https://doi.org/10.1007/s10592-020-01251-z.

Esque TC et al. 2010. Effects of subsidized predators, resource variability, and human population density on desert tortoise populations in the Mojave Desert, USA. Endangered Species Research **12**:167–177.

Fretwell SD, Lucas HL. 1970. On territorial behavior and other factors influlencing habitat distribution in birds. Acta Biotheoretica **19**:16–36.

Germano JM, Bishop PJ. 2009. Suitability of amphibians and reptiles for translocation. Conservation Biology **23**:7–15.

Germano JM, Nafus MG, Perry JA, Hall DB, Swaisgood RR. 2017. Predicting translocation outcomes with personality for desert tortoises. Behavioral Ecology **28**:1075–1084.

Goertz JW. 1964. The influence of habitat quality upon density of cotton rat populations. Ecological Monographs **34**:359–381.

Hartig F. 2017. DHARMa: Residual diagnostics for hierarchical (multi-level/ mixed) regression models. R package version 0.1.5. Available from https://cran.r-project.org/package=DHARMa.

Hellstedt P, Kallio ER. 2005. Survival and behaviour of captive-born weasels (*Mustela nivalis nivalis*) released in nature. Journal of Zoology **266**:37–44.

Homer BL, Berry KH, Brown MB, Ellis G, Jacobson ER. 1998. Pathology of diseases in wild desert tortoises from California. Journal of Wildlife Diseases **34**:508–523.

Jacobson ER, Berry KH, Wellehan JFX, Origgi F, Childress AL, Braun J, Schrenze M, Yee J, Rideout B. 2012. Serologic and molecular evidence for testudinid herpesvirus 2 infection in wild agassiz’s desert tortoises, *Gopherus agassizii*. Journal of Wildlife Diseases **48**:747–757.

Jetz W, Carbone C, Fulford J, Brown JH. 2004. The scaling of animal space use. Science **306**:266–268.

Kelly EC, Cypher BL, Westall TL. 2021. Predation on desert tortoises (*Gopherus agassizii*) by desert canids. Journal of Arid Environments **189**:104476. Elsevier Ltd. Available from https://doi.org/10.1016/j.jaridenv.2021.104476.

Kennedy CM, Grant EHC, Neel MC, Fagan WF, Marra PP. 2011. Landscape matrix mediates occupancy dynamics of Neotropical avian insectivores. Ecological Applications **21**:1837–1850.

Kristan WB, Boarman WI. 2003. Spatial pattern of risk of common raven predation on desert tortoises. Ecology **84**:2432–2443.

Kuznetsova A, Brockhoff PB, Christensen RHB. 2017. lmerTest Package: Tests in linear mixed effects models. Journal of Statistical Software **82**.

Lee JA. 1986. Origin of mounds under creosote bush (*Larrea tridentata*) on terraces of the Salt River, Arizona. Journal of the Arizona-Nevada Academy of Science **21**:23–28.

Lloyd NA, Hostetter NJ, Jackson CL, Converse SJ, Moehrenschlager A. 2019. Optimizing release strategies: a stepping-stone approach to reintroduction. Animal Conservation **22**:105–115.

Lovich JE, Ennen JR. 2011. Wildlife Conservation and Solar Energy Development in the Desert Southwest , United States. BioScience **61**:982–992.

Lüdecke D, Ben-Shachar M, Patil I, Waggoner P, Makowski D. 2021. performance: An R package for assessment, comparison and testing of statistical models. Journal of Open Source Software **6**:3139.

Mason G, Burn CC, Dallaire JA, Kroshko J, McDonald Kinkaid H, Jeschke JM. 2013. Plastic animals in cages: Behavioural flexibility and responses to captivity. Animal Behaviour **85**:1113–1126. Elsevier Ltd. Available from http://dx.doi.org/10.1016/j.anbehav.2013.02.002.

McGovern PA, Buhlmann KA, Todd BD, Moore CT, Mark Peaden J, Hepinstall-Cymerman J, Daly JA, Tuberville TD. 2020a. Comparing husbandry techniques for optimal head-starting of the Mojave desert tortoise (*Gopherus agassizii*). Herpetological Conservation and Biology **15**:626–641.

McGovern PA, Buhlmann KA, Todd BD, Moore CT, Peaden JM, Hepinstall-Cymerman J, Daly JA, Tuberville TD. 2020b. The effect of size on post-release survival of head-started mojave desert tortoises. Journal of Fish and Wildlife Management **11**:494–506.

McLean DJ, Skowron Volponi MA. 2018. trajr: An R package for characterisation of animal trajectories. Ethology **124**:440–448.

McLoughlen PD, Ferguson SH. 2000. A hierarchical pattern of limiting factors helps explain variation in home range size. Ecoscience **7**:123–130.

Morrison ML. 2001. A proposed research emphasis to overcome the limits of wildlife-habitat relationship studies. Journal of Wildlife Management **65**:613–623.

Mueller JM et al. 1998. Size-specific fecundity of the desert tortoise (*Gopherus agassizii*). Journal of Herpetology **32**:313–319.

Nafus MG, Esque TC, Averill-Murray RC, Nussear KE, Swaisgood RR. 2017a. Habitat drives dispersal and survival of translocated juvenile desert tortoises. Journal of Applied Ecology **54**:430–438.

Nafus MG, Germano JM, Perry JA, Todd BD, Walsh A, Swaisgood RR. 2015. Hiding in plain sight: A study on camouflage and habitat selection in a slow-moving desert herbivore. Behavioral Ecology **26**:1389–1394.

Nafus MG, Germano JM, Swaisgood RR. 2017b. Cues from a common predator cause survival-linked behavioral adjustments in Mojave Desert tortoises (*Gopherus agassizii*). Behavioral Ecology and Sociobiology **71**. Behavioral Ecology and Sociobiology.

Nafus MG, Tuberville TD, Buhlmann KA, Todd BD. 2017c. Precipitation quantity and timing affect native plant production and growth of a key herbivore, the desert tortoise, in the Mojave Desert. Climate Change Responses **4**:1–10. Climate Change Responses.

Nagy KA, Hillard SL, Tuma MW, Morafka DJ. 2015. Head-started desert tortoises (*Gopherus agassizii*): Movements, survivorship and mortality causes following their release. Herpetological Conservation and Biology **10**:203–215.

Nussear KE, Esque TC, Inman RD, Gass L, Thomas K a, Wallace CS a, Blainey JB, Miller DM, Webb RH. 2009. Modeling habitat of the desert tortoise (*Gopherus agassizii*) in the Mojave and parts of the Sonoran Deserts of California, Nevada, Utah, and Arizona. Page US Geological Survey open-file report.

O'Connor MP, Zimmerman LC, Ruby DE, Bulova SJ, Connor MPO, Zimmerman LC, Ruby DE, Bulova SJ, Spotila JR. 1994. Home range size and movements by desert tortoises, *Gopherus agassizii*, in the eastern Mojave Desert. Herpetological Conservation and Biology **8**:60–71.

Power ME. 1984. Habitat quality and the distribution of algae-grazing catfish in a Panamanian stream. Journal of Animal Ecology **53**:357–374.

Resende PS, Viana-Junior AB, Young RJ, Azevedo CS. 2021. What is better for animal conservation translocation programmes: Soft- or hard-release? A phylogenetic meta-analytical approach. Journal of Applied Ecology **58**:1122–1132.

Robles H, Ciudad C. 2012. Influence of habitat quality, population size, patch size, and connectivity on patch-occupancy dynamics of the middle spotted woodpecker. Conservation Biology **26**:284–293.

Rostal DC, Jones DN. 2002. Population biology of the gopher tortoise (*Gopherus polyphemus*) in Southeast Georgia. Chelonian Conservation and Biology **4**:1–9. Available from https://www.researchgate.net/publication/283461282.

Schlaepfer MA, Runge MC, Sherman PW. 2002. Ecological and evolutionary traps. Trends in Ecology & Evolution **17**:474–480. Available from http://linkinghub.elsevier.com/retrieve/pii/S0169534702025806.

Schooley RL, Branch LC. 2009. Enhancing the area-isolation paradigm: Habitat heterogeneity and metapopulation dynamics of a rare wetland mammal. Ecological Applications **19**:1708–1722.

Searcy CA, Shaffer HB. 2008. Calculating biologically accurate mitigation credits: Insights from the California tiger salamander. Conservation Biology **22**:997–1005.

Shaver DJ, Wibbels T. 2007. Head-starting the Kemp's ridley sea turtle. Pages 297–323 Biology and conservation of ridley sea turtles. Johns Hopkins University Press, Baltimore, Maryland, 297-323.

Soulard CE, Esque TC, Bedford DR, Bond S. 2013. The role of fire on soil mounds and surface roughness in the Mojave Desert **121**:111–121.

Spotila JR et al. 1994. Effects of incubation conditions on sex determination, hatching success, and growth of hatchling desert tortoises, *Gopherus agassizii*. Herpetological Monographs **8**:103–116.

Therneau T. 2015. A package for survival analysis in S. version 2.38.

Todd BD, Halstead BJ, Chiquoine LP, Peaden JM, Buhlmann KA, Tuberville TD, Nafus MG. 2016. Habitat selection by juvenile Mojave Desert tortoises. Journal of Wildlife Management **80**:720–728.

Todd BD, Rothermel BB. 2006. Assessing quality of clearcut habitats for amphibians: Effects on abundances versus vital rates in the southern toad (*Bufo terrestris*). Biological Conservation **133**:178–185.

Todd BD, Tuberville TD, Buhlmann KA. 2021. Mitigating impacts of solar energy development on desert tortoises: Indoor rearing and release of head started desert tortoises.

Tuberville TD, Buhlmann KA, Sollmann R, Nafus MG, Peaden JM, Daly JA, Todd BD. 2019. Effects of short-term, outdoor head-starting on growth and survival in the mojave desert tortoise (*Gopherus agassizii*). Herpetological Conservation and Biology **14**:171–184.

U.S. Fish and Wildlife Service. 2011. Draft revised recovery plan for the Mojave population of the desert tortoise. Page Region 8, Pacific Southwest Region. Sacramento, CA.

Van Horne B. 1983. Density as a misleading indicator of habitat quality. The Journal of Wildlife Management **47**:893–901.

Zimmerman LC, Connor MPO, Bulova SJ, Spotila JR, Kemp SJ, Salice CJ. 1994. Thermal ecology of desert tortoises in the eastern Mojave Desert : seasonal patterns of operative and body temperatures , and microhabitat utilization. Herpetological Monographs **8**:45–59. Available from http://www.jstor.org/stable/1467069.

For information on Appendices, please contact Kevin Uy at Kevin.Uy@energy.ca.gov for EPC-16-053.