

How Do Backyard Habitats Compare to the Wild? Temperature and Vegetation in Sonoran Desert Tortoise Habitat

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Research Article

Keywords: Captive reptile husbandry, *Gopherus morafkai*, land surface temperature, NDVI, remote sensing, urbanization

Posted Date: October 7th, 2026

DOI: <https://doi.org/10.21203/rs.3.rs-11147273/v1>

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Additional Declarations: No competing interests reported.

Abstract

Sonoran Desert tortoises (*Gopherus morafkai*; "tortoise") are an iconic species and pose unique management challenges because a growing captive population is unable to be repatriated to the wild and instead lives in the backyards of thousands of Arizona residents. Wild tortoises rely on environmental features (e.g., burrows, rock shelters, shrubs) for thermoregulation, yet captive tortoises in urban backyards experience higher temperatures than natural habitats because of the urban heat island effect and the prevalence of impervious surfaces. We examine how thermal and vegetative conditions differ between wild and residential habitats to determine if captive tortoises in backyards are exposed to different conditions compared to wild populations. Using existing datasets on adopter zip codes from Arizona Game and Fish, Tortoise Adoption Program, and locations from telemetered wild tortoises from the McDowell Sonoran Conservancy, we compared land surface temperature and Normalized Difference Vegetation Index (NDVI) using remote sensing. From 2021 to 2023, land surface temperatures increased in both habitats, but were highest in residential areas. NDVI decreased in wild habitats over time but remained consistent in residential areas. Wild areas had higher NDVI than residential areas in 2021 and 2022, but both had similar values in 2023. Backyard tortoises may experience greater stability in vegetation cover but face higher thermal conditions over their lifetime compared to wild tortoises. Future research on how environmental factors connected to specific backyards and implications for the overall health of tortoises could inform tortoise caretakers on providing the best care.

INTRODUCTION

The urban human population now outnumbers those living in rural areas and towns; cities are projected to absorb two-thirds of the global population growth by 2050 (United Nations 2025). Rapid urbanization has expanded the geographic extent of metropolitan areas and increased opportunities for human-wildlife interactions (Soulsbury and White 2015; Schell et al. 2021). Large urban areas experience the urban heat island (UHI) effect, where temperatures are higher in built environments due to the heat retained by impervious surfaces (e.g., asphalt, concrete, etc.) that replace native vegetation (Archer and Predick 2008). As urban areas continue to expand into previously undeveloped landscapes, documenting how wildlife responds to these altered environments has become increasingly important to understand urban impacts on wildlife behavior, movement, habitat utilization, and disturbance effects, especially for species that are less resilient to change (Averill-Murray et al. 2020; Frazier et al. 2025).

The Sonoran Desert is a unique ecosystem that spans the southwestern United States and northwestern Mexico. It encompasses the greater Phoenix metropolitan area located within Maricopa County, Arizona. Maricopa County's population was 4.5 million in 2022 and continues to rise, making it the 4th largest county in the United States. The rapid urbanization has quickly altered the landscape in this region. Many studies have looked at the impact of urbanization and the UHI effect on wildlife in and around the Phoenix metropolitan area (Albuquerque et al. 2021; Bateman et al. 2023; Frazier et al. 2025). Although numerous studies have documented physiological, behavioral, and welfare differences between captive

and wild animals (Ardjima et al. 2020; Glass et al. 2024; Hanson et al. 2025; Jin et al. 2026), few investigators have quantified differences in environmental conditions experienced by captive reptiles (White et al. 2026). Our study helps address this gap by comparing the thermal and vegetative characteristics of captive backyard habitats with those of wild desert tortoise habitat, providing an ecological basis for evidence-based captive husbandry.

One of the Sonoran Desert's most iconic species inhabits both the natural desert landscapes surrounding the Phoenix metropolitan area and the backyards of thousands of Arizona residents. The Sonoran Desert tortoise (*Gopherus morafkai*), which is listed as a Species of Greatest Conservation Need in Arizona, is protected under Arizona state regulations. Despite their long lifespan, Sonoran Desert tortoise populations are declining due to disease, habitat loss and fragmentation, climate change, and low reproductive output (Averill-Murray et al. 2018, 2020; Sullivan et al. 2014). Desert tortoises are vulnerable to climate change due to their specific habitat and temperature preferences (Griffis-Kyle et al. 2018). It is predicted that Sonoran Desert tortoise population declines will increase by 3% between 2035 and 2060 due to climate-induced temperature increases and drought severity (Zylstra et al. 2013).

Desert vegetation provides refuge for reptiles and other ectothermic species that depend on environmental elements to thermoregulate and maintain physiological functions (Huey and Stevenson 1979). Tortoises, in particular, seek specific microhabitats and shelter types (e.g., burrows, rock shelters, and shrubs) to facilitate thermoregulation (Grandmaison et al. 2010; Bridges et al. 2016), and reduced vegetation cover can limit access to these thermoregulatory resources, placing greater physiological stress on individuals (Kearney et al. 2009). Major reductions in vegetation cover can occur naturally from wildfires, and desert tortoises can acclimate to short-term changes in plant cover with changes in their basking, foraging, and shelter-seeking behavior (Esque et al. 2003; Drake et al. 2015). However, because captive Sonoran Desert tortoises remain in captivity for life, they cannot rely on behavioral flexibility to account for long-term habitat deficiencies in the same way wild tortoises respond to temporary stochastic disturbances. Comparing the environmental conditions of backyard tortoises to those of their wild counterparts is therefore critical for understanding potential long-term health consequences and determining whether backyard habitats adequately meet their ecological needs.

Before regulations were placed in 1988, people collected wild Sonoran Desert tortoises as pets, creating a substantial captive population that could not be returned to the wild due to disease and genetic concerns (Berry 1986; Dodd Jr. and Seigel 1991; Field et al. 2007). To manage the large captive population of Sonoran Desert tortoises, Arizona Game and Fish's Tortoise Adoption Program was created, inviting Arizona residents seeking a long-term, non-traditional pet to adopt a backyard tortoise. Between the program's inception in 1982 and 2005, over 4,600 Sonoran Desert tortoises were adopted through the Tortoise Adoption Program (Jones 2008). However, the true number of tortoises is likely much higher, as many owners were grandfathered into the regulation, and illegal activities (i.e., removal from the wild and backyard breeding) have persisted.

The objective of this study is to evaluate environmental conditions found in residential areas with large numbers of captive tortoises compared with conditions from telemetry locations of wild Sonoran Desert tortoises. Specifically, we assess whether captive tortoises experience different thermal and vegetative conditions than wild populations, using long-term land surface temperature and Normalized Difference Vegetation Index (NDVI) data for the Greater Phoenix area. Urbanization typically increases impervious surface cover, which is strongly and negatively associated with NDVI (Yuan and Bauer 2007; Myint et al. 2013). In turn, greater impervious surface cover and lower NDVI are associated with elevated land surface temperatures, a pattern especially pronounced in urban heat islands such as the Phoenix metropolitan area (Chow et al. 2012; Wang et al. 2016); thus, we predict that captive tortoise habitats will exhibit higher land surface temperatures and lower vegetation cover than wild habitats, reflecting the greater extent of built environment and urban heat island effects within city limits.

METHODS

Study Area

Phoenix, Maricopa County, Arizona, is one of the fastest-growing metropolitan areas in the United States (Fig. 1). The study area is located within the Sonoran Desert at an elevation of 331 m. The region is characterized by mild winters, hot and dry summers, and a summer monsoon season, with a mean annual precipitation of 16.2 cm (6.38 in; National Weather Service 2026). Summer temperatures can reach 48.3°C (119 °F), with mean daily highs exceeding 37.8 °C (100 °F) between April and October (National Weather Service 2026).

Wild Tortoise Location Data

On the northeastern edge of the Phoenix metropolitan area, the McDowell Sonoran Preserve is a region of 12,340 ha of protected desert habitat with 380 km of trails for public recreation. The McDowell Sonoran Conservancy, the official partner of the McDowell Sonoran Preserve, is a non-profit organization that contributes to stewardship, science, and education. With support from the Arizona Game and Fish Department and the City of Scottsdale, they monitor Sonoran Desert tortoise populations via surveys, radio telemetry, and GPS tracking.

Existing Sonoran Desert tortoise telemetry data collected by the McDowell Sonoran Conservancy between 2021 and 2023 under Arizona Game and Fish Department Scientific Activity License No. SP041594 were provided to the authors with permission for use in this study (McDowell Sonoran Conservancy 2025). We filtered all locations to only include those collected between 21 June and 21 September for 2021–2023 to match our spatial dataset for land surface temperature and NDVI.

We then separated locations by year. To ensure spatial independence, we performed a spatial thinning of tortoise locations for each year using a custom Python script with a k-dimensional tree spatial index, followed by a greedy thinning algorithm with a nearest-neighbor buffer distance of 185 m. The buffer

distance was obtained by averaging the maximum cross-sectional length of a single tortoise's home range (100% minimum convex polygon) following the procedure outlined in Bridges et al. (2016). We then selected a single random record per unique tortoise ID to avoid repeat locations from the same individual within the same season. We repeated this process for each survey year, averaging 19 wild tortoise locations per year. Elevation of wild tortoise locations varied from 623–1009 m (mean = 843.5, SE = 10.7)

Residential Tortoise Location Data

The Arizona Game and Fish Department's Tortoise Adoption Program, created in 1982, adopts out hundreds of captive tortoises per year. The majority of tortoises adopted are surrendered pets that people could no longer care for. Arizona Game and Fish provided existing adoption records, including dates and zip codes of adoptions between 2021 and 2023 (published with permission). As a proxy for backyard tortoise locations, we selected the 15 zip codes within Maricopa County with the highest adoption frequencies ("zip codes of interest"). Using active parcel data (excluding State and Federal lands) from the Maricopa County Enterprise GIS Database (Maricopa County Assessor's Office 2025), we filtered for residential parcels using property use codes (PUCs) defined by the Arizona Department of Revenue Property Tax Division Property Use Code Manual (Arizona Department of Revenue 2000) in the software QGIS (QGIS Development Team 2026). Property use codes included single-family residential sites of 5 acres or less (PUC prefix 01) and residential improvement on more than 5 acres (PUC prefix 87). We generated a set of 19 random points across all residential parcels within the zip codes of interest to represent residential tortoise locations (for a detailed example, see Fig. S1). These points were buffered at 185 m, which is the same spatial extent as the wild tortoise locations. Elevation of residential locations varied from 380–634 m (mean = 507.8, SE = 18.8).

Spatial Data

We obtained publicly available datasets provided by Central Arizona Phoenix Long-Term Ecological Research (CAP LTER) from remote-sensed surface reflectance imagery from Landsat 5–9 at a 30x30 m pixel resolution for land surface temperature (Haight et al. 2025a) and NDVI (Haight et al. 2025b). Both datasets contained seasonally aggregated imagery to create median composite images for 2021, 2022, and 2023, with 'summer' defined as 21 June to 21 September. Using QGIS, we extracted summer land surface temperatures and NDVI values for all residential ($N_{2021-2023} = 19$) and wild tortoise locations for each year.

Data Analyses

We performed all statistical analyses in R (R Core Team 2026) using the tidyverse (Wickham et al. 2019), FSA (Ogle et al. 2026), and PMCMRplus (Pohlert 2024) packages. First, to account for our small sample

size and any violations of normality, we performed a Kruskal-Wallis test to evaluate whether land surface temperatures and NDVI values differed across years for wild tortoise locations. We then determined which years statistically differed using a Bonferroni-adjusted Dunn post-hoc test. Next, to compare land surface temperatures and NDVI values across years for residential tortoises, we used a Friedman test with a Bonferroni-adjusted Conover post-hoc test, as locations for each year were resampled and thus not independent of each other. Lastly, we compared land surface temperature and NDVI values between study areas within the same year using a Mann-Whitney U test to account for violations in normality and a small sample size and repeated this for each year.

RESULTS

Differences in temperature between wild and residential tortoise habitats

After spatially thinning McDowell Sonoran Preserve tortoise GPS locations for each year, we analyzed single summer locations from 17 tortoises in 2021, 19 tortoises in 2022, and 22 tortoises in 2023. Summer land surface temperatures in wild tortoise habitats differed across all three study years ($H[2] = 30.66$, $P < 0.001$). Wild habitats in 2021 had the coolest land surface temperature (mean = 46.7 °C, SE = 0.65). Land surface temperatures increased between 2021 and 2022 (mean = 50.3 °C, SE = 0.36, $P < 0.01$) and again between 2022 and 2023 (mean = 52.2 °C, SE = 0.51, $P = 0.046$).

Mean residential parcel size in our zip codes of interest was 1,337.2 m² (SD = 2,890.8 m²). We analyzed 19 randomly selected points within all residential parcels and resampled each location every year for land surface temperature and NDVI. Land surface temperatures in residential areas varied between study years ($Fr[2] = 20.21$, $P < 0.001$). Land surface temperature was warmer from 2021 (mean = 50.3 °C, SE = 0.68) to 2022 (54.0 °C, SE = 0.47, $P < 0.001$), but not from 2022 to 2023 (mean = 54.2 °C, SE = 0.49, $P = 1.00$).

Summer land surface temperatures were consistently warmer in residential areas than in wild habitats across all three years (Fig. 2). In 2021, residential areas exhibited land surface temperatures that were 3.5 °C warmer than those of wild habitats ($U = 260$, $P < 0.01$). We saw similar trends in 2022, when residential land surface temperatures exceeded those of wild habitats by 3.7 °C ($U = 331$, $P < 0.001$), and in 2023, when residential areas remained 2.0 °C warmer than wild habitats ($U = 311$, $P < 0.01$).

Differences in vegetation between wild and residential tortoise habitats

Wild tortoise habitat NDVI values also differed across study years ($H[2] = 26.12$, $P < 0.001$). NDVI decreased over time, with 2021 exhibiting the highest NDVI (mean = 0.30, SE = 0.01). We saw no difference in NDVI between 2021 and 2022 (mean = 0.26, SE = 0.01, $P = 0.27$) but did see a significant

decrease between 2022 and 2023 (mean = 0.21, SE = 0.01, $P < 0.01$). There was no difference in NDVI of residential tortoise habitats across all three years ($F_{r[2]} = 0.11$, $P = 0.96$), and mean NDVI for all three years was 0.20 (SE = 0.01).

Mean NDVI differed significantly between residential areas and wild habitats at the beginning of our study period but converged over time (Fig. 3). In 2021, mean NDVI was 0.10 higher in wild habitats than in residential areas ($U = 27$, $P < 0.001$). The difference in 2022 was higher than in 2021, but decreased to 0.06 ($U = 64$, $P < 0.001$). By 2023, there was no longer a difference between wild and residential habitats ($U = 161$, $P = 0.22$), suggesting that vegetation greenness became comparable between the two habitat types.

DISCUSSION

Consistent with our predictions, vegetation cover was lower in residential areas compared to wild habitats. Whereas vegetation cover remained relatively constant in residential areas, as estimated using zip code-level data, vegetation cover measured at wild tortoise locations fluctuated among years, reflecting natural variation in the landscape. We also observed significantly warmer land surface temperatures in backyard tortoise habitats compared to wild habitats, with temperatures increasing over time in both habitat types. Land surface temperature and vegetation cover are key components of desert tortoise habitat because tortoises rely on their surrounding environment to regulate body temperature (Huey and Stevenson 1979), and vegetation provides both thermal refuge and forage. Consequently, differences in these habitat characteristics have important implications for tortoise ecology and husbandry. The consistently warmer conditions observed in residential habitats are consistent with the urban heat island effect, suggesting that captive tortoises in backyard environments are chronically exposed to higher thermal conditions than wild tortoises occupying natural habitats.

The magnitude of the temperature difference between residential and wild habitats is consistent with previous studies documenting the urban heat island effect in metropolitan Phoenix (Brazel et al. 2007; Chow et al. 2012; Connors et al. 2013; Li et al. 2016; Zhang et al. 2017). One of the major contributors to urban warming between 1990 and 2004 in Phoenix was the rapid expansion of residential development (Wang et al. 2016). One of the major contributors to urban heat between 1990 and 2004 in Phoenix was the rapid increase in residential development (Wang et al. 2016). Consistent with this pattern, land surface temperatures in residential tortoise habitat were nearly 3°C higher in every study year, confirming that backyard tortoises in the Phoenix-metropolitan area are experiencing warmer thermal conditions compared to wild tortoises in the McDowell Sonoran Preserve.

Prolonged exposure to elevated temperatures has documented physiological and ecological consequences across a wide range of taxa. For example, pocket mice and kangaroo rats exhibited lower body fat percentages, an important indicator of overall health, in areas with warmer temperatures and higher urbanization levels compared to individuals living in areas with cooler temperatures, greater vegetation cover, and low urbanization (Bateman et al. 2023). Similarly, studies investigating the long-

term impacts of warming on reptiles and amphibians found that prolonged heat exposure increases metabolic rates, leading to greater losses of body fat and water (Rutschmann et al. 2024; Kurnaz 2026) and reducing the overall probability of survival (Nishad et al. 2026). This can be exacerbated indirectly, as increased droughts reduce vegetation cover, thus decreasing food availability and shelter opportunities (Zylstra et al. 2013). Long-term, elevated temperatures influence the phenological patterns of many reptiles, typically shifting emergence from hibernation earlier and impacting incubation success (Kurnaz, 2026). Elevated temperatures in the short term have shifted diurnal activity in reptiles, which are utilizing nocturnal refuge to cope with unsuitable daytime temperatures (Rutschmann et al. 2024).

Patterns of vegetation in residential and wild areas exhibited different patterns than those of temperature. Vegetation cover remained relatively constant across the three study years in backyard habitats but was consistently lower than in the McDowell Sonoran Preserve. Dense urbanized areas are typically characterized by extensive impervious surfaces and correspondingly low NDVI values, whereas agricultural and residential land uses represent greener exceptions within the urban matrix; nevertheless, vegetation throughout metropolitan Phoenix has declined from the mid-20th century to the present (Stabler et al. 2005; Buyantuyev and Wu 2012). The temporal stability of vegetation in residential habitats likely reflects water supplementation from irrigation and landscape maintenance. In arid cities such as Phoenix, irrigation and landscape maintenance can decouple urban vegetation from natural precipitation patterns, allowing vegetation to remain relatively stable despite seasonal and interannual climatic variability (Grimm et al. 2008; Zhang et al. 2012). This suggests that although backyard tortoises may occupy areas with less green space, overall vegetation cover in residential areas is likely buffered by human care, whereas wild tortoises remain more directly affected by year-to-year variability in vegetation coverage.

Despite observing no difference in vegetation cover between wild and residential habitats in 2023, the negative trend in NDVI in wild habitats is concerning. Sonoran Desert tortoises are income breeders, only producing clutches in years when resources are plentiful, making them particularly susceptible to drought or invasive plant species outcompeting native food sources (Averill-Murray et al. 2018). Although our analyses focused on the primary active season, future work examining spring vegetation dynamics may improve understanding of reproductive investment, whereas fall conditions may provide insight into hatchling survival.

Collectively, our findings indicate that backyard tortoises could experience a combination of high land surface temperatures and less vegetation cover relative to wild tortoises. Regardless of the underlying cause, tortoises in the Phoenix metropolitan area are experiencing consistently higher land surface temperatures year over year compared to those in the McDowell Sonoran Preserve, making it imperative for backyard caretakers to provide thermal refuge for their tortoises.

Although the spatial resolution of this study was relatively coarse (30 m per pixel), the trends we observed provide a strong rationale for future investigations using higher-resolution imagery, such as the 1 m imagery available through the National Agriculture Imagery Program (NAIP). Our analyses also relied

on coarse spatial data, using residential zip codes filtered for residential parcels as a proxy for backyard tortoise habitat. A limitation of this study was that we did not investigate the exact addresses of adopters because doing so would breach the confidentiality agreement between agencies (e.g., the Arizona Game and Fish Department) and adopters. Furthermore, the 30 m resolution of Landsat imagery cannot reliably distinguish building infrastructure from yard space within a single residential parcel where tortoises are kept. These spatial limitations also prevented evaluation of other environmental variables that influence tortoise health, including rainfall, humidity, and fine-scale habitat structure.

Differences in summer land surface temperature between sites may be attributed to multiple factors, including urbanization and elevation. The McDowell Sonoran Preserve locations sampled were approximately 300 m higher in elevation on average. This study is specific to the Phoenix metropolitan area and a single nearby wild population; as such, findings may not be representative of tortoise habitat conditions across the species' range.

Expanding this analysis to additional metropolitan and wild populations across the Sonoran Desert, including both higher- and lower-elevation regions, would help determine whether hotter land surface temperatures are primarily driven by urbanization rather than local climatic differences. Integrating long-term body condition monitoring with habitat variables may also improve understanding of how chronic exposure to elevated temperatures and altered vegetation influences captive and wild tortoise health. Future research should therefore pair higher-resolution remote sensing with community-engaged approaches, such as partnering with Arizona residents to participate in backyard burrow and health monitoring, to capture environmental conditions at the spatial scale experienced by individual tortoises.

CONCLUSION

Overall, findings of this study are novel and offer insights into urban environmental conditions in an area that supports a large number of captive tortoises. Reptiles, such as tortoises, are often misunderstood in terms of interpreting behaviors related to poor husbandry, which can lead to inferior captive animal welfare conditions (Hoehfurtner et al. 2025). This project highlights important management considerations for the long-term welfare and persistence of captive Sonoran Desert tortoises. Prolonged exposure to higher thermal conditions affects many species native to the Sonoran Desert, and warmer temperatures may impose cumulative physiological stress over decades, particularly in captive settings where animals cannot easily seek refuge unless provided by caretakers. Because residential habitats consistently exhibited higher land surface temperatures and lower vegetation cover than wild habitats, our findings support husbandry practices designed to mitigate these differences. Providing ample shade, well-insulated burrows or other thermal refuges, and structurally diverse enclosures that more closely mimic natural habitat conditions may help reduce heat exposure experienced by captive tortoises. Additionally, applying knowledge of wild tortoise ecology to caretaking practices may strengthen species-wide conservation goals.

Declarations

ACKNOWLEDGEMENTS

We thank Arizona Game and Fish and the McDowell Sonoran Conservancy for providing us with the datasets to make this research happen. This material is based upon work supported by the National Science Foundation under grant number DEB-2224662, Central Arizona-Phoenix Long-Term Ecological Research Program (CAP LTER).

Funding

This material is based upon work supported by the National Science Foundation under grant number DEB-2224662, Central Arizona-Phoenix Long-Term Ecological Research Program (CAP LTER).

Competing Interests

The authors have no relevant financial or non-financial interests to disclose.

Author Contributions

All authors contributed to the study conception and design. Material preparation, data collection, and analysis were performed by Margaret Davis and Fábio Albuquerque. The first draft of the manuscript was written by Margaret Davis, and all authors commented on previous versions of the manuscript. All authors read and approved the final manuscript.

Data Availability

Environmental data on land surface temperature and Normalized Difference Vegetation Index (NDVI) are available on the Environmental Data Initiative Data Repository under DOIs <https://doi.org/10.6073/pasta/ff8ddd9260cda83700d7875790163d4> and <https://doi.org/10.6073/pasta/9d8da777830bc90c452bac41ea2106f0>, respectively.

The locations of tortoises and ZIP codes of adopters that support the findings of this study are available from the McDowell Sonoran Conservancy (MSC) and the Arizona Game & Fish Department's Tortoise Adoption Center (AZGFD-TAP), but restrictions apply to the availability of these data, which were used under license for the current study and so are not publicly available. The data are, however, available upon request and with the permission of both agencies.

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Figures

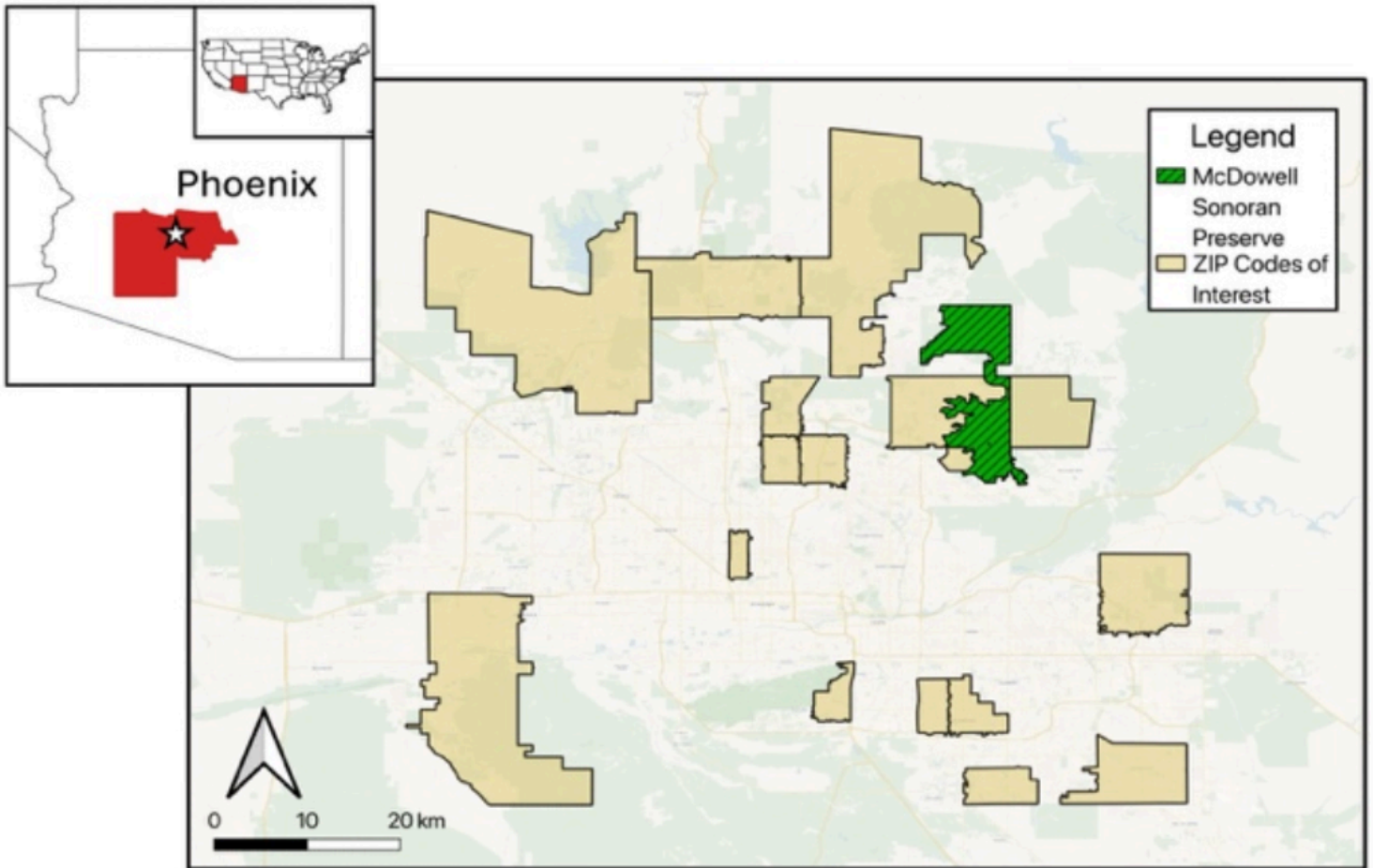


Figure 1

Wild and captive tortoise habitat study area in the greater Phoenix metropolitan area, Arizona, USA, for comparison of summer land surface temperature and Normalized Difference Vegetation Index from remote-sensed satellite imagery. Wild tortoise locations were provided by the McDowell Sonoran Conservancy (with permission) within the McDowell Sonoran Preserve. Nineteen random points within residential parcels of the 15 zip codes in Maricopa County with the highest adoption frequencies served as a proxy for backyard tortoise locations

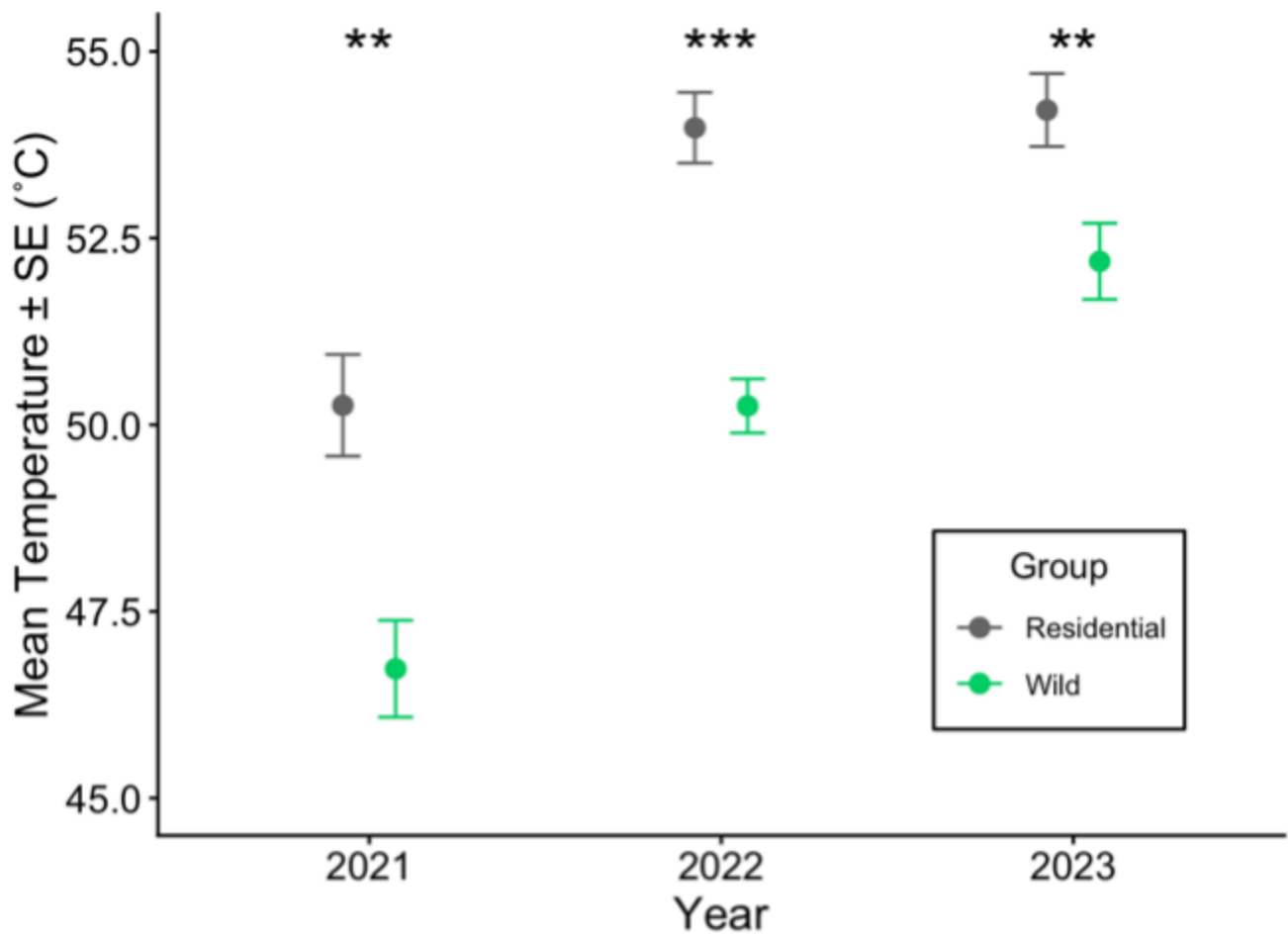


Figure 2

Mean summer land surface temperature of residential and wild tortoises from 2021 to 2023 via Landsat imagery. Data were provided by Central Arizona-Phoenix Long-Term Ecological Research by J. Haight et al. (2025a). Landsat data were filtered from June 21st to September 21st for each year to create a composite image with the median land surface temperature. For all three study years, residential areas ($N_{2021-2023} = 19$) had higher land surface temperatures than wild areas (2021: $U = 260$, $P < 0.01$; 2022: $U = 331$, $P < 0.001$; 2023: $U = 311$, $P < 0.01$). Significance codes are indicated as * = $P < 0.05$, ** = $P < 0.01$, and *** = $P < 0.001$

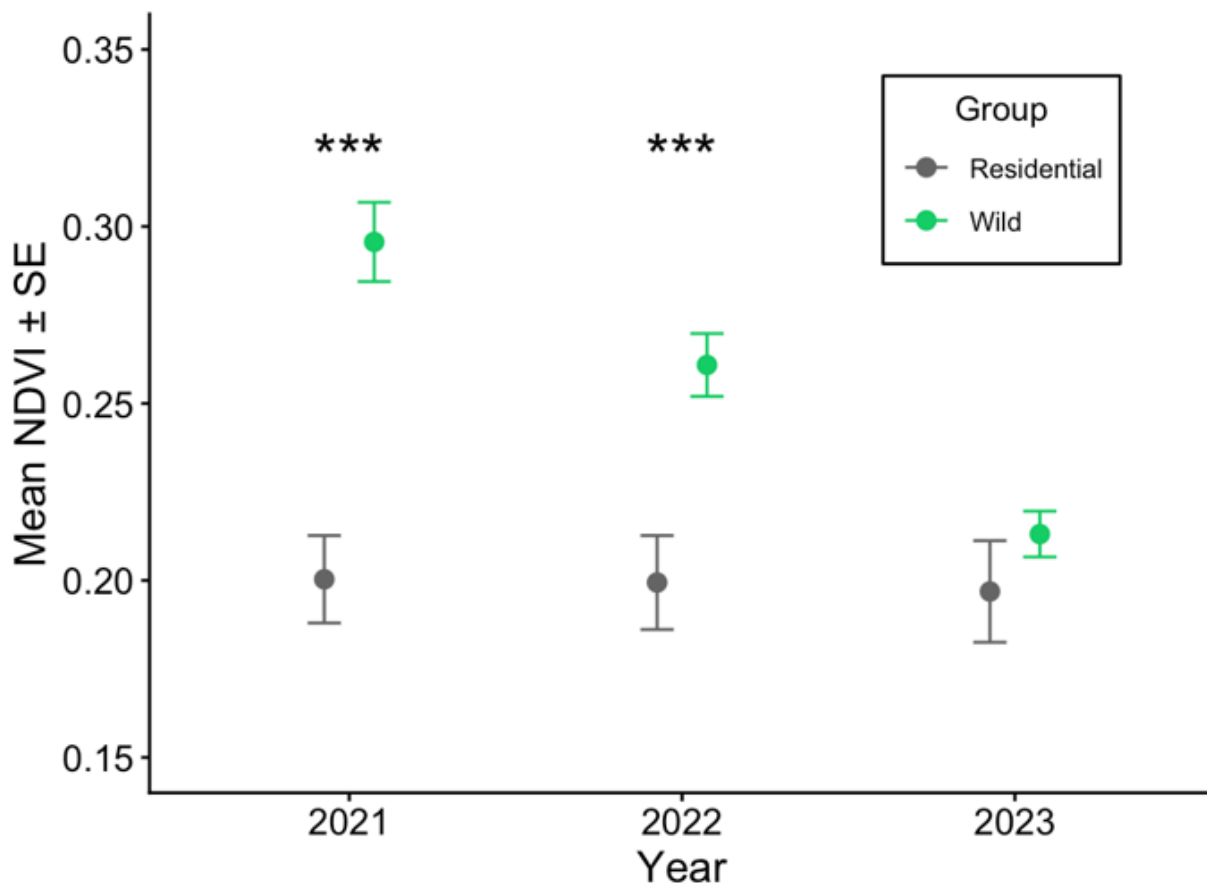


Figure 3

Mean summer Normalized Difference Vegetation Index (NDVI) values of residential and wild tortoise habitats from 2021 to 2023 via Landsat imagery. Datasets used for analysis were from Central Arizona-Phoenix Long-Term Ecological Research by J. Haight et al. (2025b). Landsat 5-9 data were filtered from June 21st to September 21st for each year to create a composite image with median NDVI. In 2021 and 2022, wild habitats ($N_{2021} = 17$, $N_{2022} = 19$) had higher NDVI than residential areas (2021: $N = 17$; $U = 27$, $P < 0.001$; 2022: $N = 19$; $U = 64$, $P < 0.001$). There was no difference in NDVI between the two habitats for 2023 ($N_{Wild} = 22$, $N_{Residential} = 19$; $U =$, $P = 0.22$). Significance codes are indicated as $*$ = $P < 0.05$, $**$ = $P < 0.01$, and $***$ = $P < 0.001$

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