

# REINTRODUCING SAN JOAQUIN KIT FOX TO VACANT OR RESTORED LANDS: IDENTIFYING OPTIMAL SOURCE POPULATIONS AND CANDIDATE FOXES



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## EXECUTIVE SUMMARY

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Endangered San Joaquin kit foxes (*Vulpes macrotis mutica*) currently persist as a meta-population in central California consisting of 3 core and several satellite populations. Many of these populations are small and the meta-population is highly fragmented, which increases extinction risk. Translocation is a potential strategy for increasing the number of populations. Various factors would need to be considered when selecting source populations and candidate foxes for translocation. One such factor is behavioral attributes of individual foxes, as reflected by level of boldness, particularly in response to novel resources and potential threats.

We compared behavioral attributes between and within populations of kit foxes in urban and non-urban habitats, and also examined the relationship of these attributes to survival and fitness. The overall goal of this project was to identify optimal source populations and individual foxes for relocation efforts, and in particular to determine whether urban foxes could be used in such efforts.

Behavioral attributes were assessed among foxes in an urban (city of Bakersfield) and non-urban (Lokern Natural Area) environment. Assessments were conducted using three methods. In “Experiment One”, boldness levels of pups marked at natal dens were assessed by measuring baseline behaviors and responses to potentially beneficial (i.e., food) and potentially threatening (i.e., simulated predator) stimuli. Pups were then translocated and monitored to assess survival and reproductive success. In “Experiment Two”, boldness levels of translocated adult and juvenile foxes were assessed by measuring responses to a novel object that was neither beneficial nor threatening. Animals were then monitored to assess survival and reproductive success. In the third method, behavior of foxes in traps and handling bags was scored to determine level of boldness.

For all observations combined in Experiment One, mean boldness was significantly higher for Bakersfield foxes than for Lokern foxes. Among individual stimuli, boldness of urban foxes was higher when presented with a novel food, but were not different when exposed to the simulated predator. Thus, urban foxes might be more likely to investigate new foods, but might be equally wary of predators. Mean boldness also was significantly higher for urban foxes based on the results of Experiment Two and the Trap/Handling data. Furthermore, the range of boldness values was considerably high among urban foxes compared to non-urban foxes indicating greater behavioral variation. No relationships between boldness and survival were detected. However, non-urban foxes with higher boldness scores tended to have higher reproductive success and larger litters.

Urban kit foxes exhibited a higher mean and variation in boldness compared to non-urban foxes. This could be a result of different selective pressures between the two environments. Boldness levels of non-urban foxes may reflect adaptation to natural environments, and therefore might be “optimal” for reintroduction to similar environments. However, founders with a diversity of boldness levels may provide a greater capacity for adaptation to a novel environment, and therefore, urban foxes or a mix of urban and non-urban may be preferable. Finally, if removal of foxes might pose unacceptable risk to populations in natural lands, then relocating urban foxes conceivably would have a reasonable probability of success based on behavioral attributes.

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## INTRODUCTION

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San Joaquin kit fox (*Vulpes macrotis mutica*) populations have been significantly reduced throughout their historic range in central California, primarily due to profound habitat loss and degradation. Much of the habitat within their former range was displaced by agricultural, industrial, and urban development, facilitated by the completion of the Central Valley Project and the California Water Project in the early 1970's (U.S. Fish and Wildlife Service 1998).

As a result of this decline, San Joaquin kit foxes are listed as Federally Endangered and California Threatened. Kit foxes currently persist in a meta-population of 3 core populations and several satellite populations of varying size (U.S. Fish and Wildlife 1998). Population fragmentation of a species increases vulnerability to demographic and stochastic events, thereby increasing the risk of extinction in an already rare species such as the San Joaquin kit fox. Thus, it is desirable to increase the numbers of both individuals and populations. However, due to a lack of connectivity between existing populations and habitat restrictions, current populations are likely at carrying capacity. Therefore, a more effective conservation measure is to increase the number of kit fox populations. In addition, the establishment of new populations may facilitate movement from existing populations by reducing traveling distances, provided movement corridors are available.

Recent years have seen the restoration of areas of agricultural land to suitable kit fox habitat. Also, kit foxes occasionally are extirpated from smaller patches of fragmented habitat due to annual variations in environmental conditions (e.g., drought) or random changes in survival and reproductive rates. However, it may be difficult for kit foxes to recolonize these areas due to their distance from existing populations, and/or a lack of adequate dispersal corridors.

### ***BEHAVIORAL VARIATION AND SUITABILITY***

In addition to following protocols established by The World Conservation Union (IUCN) for planning a translocation (see Bremner-Harrison & Cypher 2007 and IUCN 1995 for details), it is essential to improve the chance of success by selecting the most suitable kit fox release candidates. In many reintroduction or translocation programs the selection of release candidates is often based solely on age, sex, genetic and health criteria. However, another factor affecting the success of reintroduction is intraspecific behavioral variation, such as seen in levels of boldness.

In recent years variation between individuals in types of behavior associated with 'personality' has been highlighted (Caro & Bateson 1986, Mather & Anderson 1993, Hansen 1996, Coleman & Wilson 1998). The International Academy of Animal Welfare Sciences recognizes the importance of individual variation and includes this under the heading 'character' when listing factors for selection of animals for reintroduction (IAAWS 1992). The existence of different personalities indicates adaptive strategies within a species (Wilson & Richards 2000), which are likely subject to natural selection

(Wilson *et al.* 1994). Variation in levels of boldness and shyness to unfamiliar objects or events within humans, and individuals of other species, are consistent and are displayed early in development (Kagan *et al.* 1988). Individual variation in boldness/shyness is widespread amongst different species, and may have important implications for survival and reproduction (Buirski *et al.* 1973). It has been shown that levels of boldness are subject to natural selection (Huntingford & Giles 1987) and therefore that inappropriate levels of boldness will have deleterious effects on fitness. This is important for any release program, as the object of release is to provide behaviorally adept individuals for successful reproduction in the wild. If there is a substantial variation in boldness of individuals destined for release some are likely to suffer reduced survival and reproduction as a consequence. This was demonstrated in a recent study of reintroduced swift fox (*Vulpes velox*), where animals that died following release were those with overly high levels of boldness (Bremner-Harrison *et al.* 2004). In a translocation program, individuals that are too bold for a particular habitat may be less likely to avoid potential predators, conspecifics or anthropogenic stimuli that may pose a risk. However individuals that are too shy may not explore their new habitat, and thus may have difficulties in locating suitable dens, food, or mates.

With levels of boldness subject to natural selection, it is possible that release candidates with optimal levels of boldness for a source-habitat similar to the release site are more likely to be successful following translocation than individuals from a source population with differing selection pressures than the release site. Little work has been conducted on levels of boldness in animals in differing habitats, both between and within populations. Kit foxes in urban and rural environments are likely to be subject to differing selection pressures so it is expected that populations as a whole would show diverse levels of boldness according to habitat. Urban foxes tend to have abundant food sources that are not subject to environmental fluctuations. However, in order to access many of these food sources urban foxes often frequent areas where humans or other anthropogenic stimuli might be present (Cypher & Frost, 1999). Natural predators such as coyotes (*Canis latrans*) are not present in the urban environment, however foxes do suffer high levels of mortality due to vehicle strikes, human activities such as construction and canal maintenance, and rodenticide poisoning. In addition, den availability is limited compared to the natural habitat. Foxes in the natural lands tend to have less abundant food sources and focus on resources that also show nocturnal activity patterns. Reproductive output mirrors food availability and trends. The main source of mortality in natural habitat is predation from coyotes. Denning patterns and home ranges cover a larger area than those of urban foxes (Nelson, 2007). Each of these factors is likely to lead to selection for optimal levels of boldness within the specific habitat type. In addition, it is likely that variation also exists within each population, demonstrating individual ability to cope with environmental change. A further possibility is that variation may be higher in the urban habitat where selection pressures may not be as strong due to limited predation and abundant food sources.

Urban and natural populations of kit fox were investigated from a behavioral standpoint, in order to determine whether optimal boldness levels of foxes within a particular environment type differed significantly. Identifying foxes of suitable behavioral types for relocation will assist with selecting foxes for reintroduction that are i) from a population that has overall levels of boldness most suited to release into a new environment, or ii) of

a behavioral type similar to foxes who are successful in an environment similar to the release habitat. Specific individuals would be identified as being of a successful behavioral type to ensure that the reintroduced population was made up of individual foxes with the highest likelihood of surviving. These individuals would be those that show optimal responses to stimuli such as predators and novel food items, indicating a propensity towards survival. Knowledge of the behavioral profile of release candidates would allow for the compilation of a release group representative of boldness scores around the optimal score for the specific release site, but with sufficient levels of variability to allow for adaptation to environmental change.

In addition to determining an overall population level and mix of boldness types to aim for, levels of boldness has been shown to affect individual post-release factors such as dispersal from specific release sites (Bremner-Harrison *et al.* 2004). Therefore identifying foxes of particular boldness levels would be beneficial in deciding fox placement during releases, for example foxes that show higher levels of boldness would be placed further away from roads or other risk factors.

Obtaining behavioral data on potential reintroduction candidates would allow the potential for assessing the likely contribution and potential survival of each specific kit fox prior to selection. This would be of great benefit as only those foxes that have a high chance of survival, potential site fidelity, and likelihood of reproductive output would be considered candidates for reintroduction, thus not risking foxes that may be more suited to remaining in their existing environment. This more conservative approach also would reduce the potential number of foxes needed for successful reintroduction thereby minimizing impacts to the source population. Attempts to maximize survival, and limit the number of reintroduction-related mortalities are especially desirable given the status of the San Joaquin kit fox.

## ***OBJECTIVES***

The overall objective of this study was to enhance kit fox recovery efforts by devising a means of selecting translocation candidates most suited to the destination environment. Successful habitat colonization is likely to be achieved through a combination of (i) selecting founders who are able to survive and reproduce in their new habitat, and (ii) relocating individuals representing maximum genetic variability. Specific objectives were:

- Assess whether intraspecific behavioral variation in terms of boldness exists **between** populations of San Joaquin kits foxes occupying different habitats (urban/natural).
- Assess whether intraspecific behavioral variation in terms of boldness exists **within** populations of San Joaquin kit foxes.
- Assess local survival and fitness of individuals within natural and urban environments, and determine whether a relationship exists between these parameters and boldness/shyness.
- Utilize fitness and behavioral data to produce a measure of the most suitable release candidates based on behavioral assessment.

## METHODS

Two experiments were conducted to collect behavioral profiling data on urban and natural land San Joaquin kit foxes. These are referred to as Experiment One and Experiment Two. The protocol for Experiment One has been used successfully for the collection of behavioral data in the past with both wild and captive swift fox and captive channel island fox (*Urocyon littoralis*). However, several aspects of the experimental methods were unsuited for studying San Joaquin foxes in the natural lands habitat, and therefore the project was broadened to include Experiment Two. In addition, a further behavioral measure was developed from trap and handling data to provide a third measure of boldness. A detailed description of the study sites and three boldness measures is provided below.

### *STUDY SITES*

Both study sites occurred within the boundaries of Kern County, California, and are considered core populations of San Joaquin kit foxes (Nelson 2007). Figure 1 shows the location of each of the study sites within Kern County.

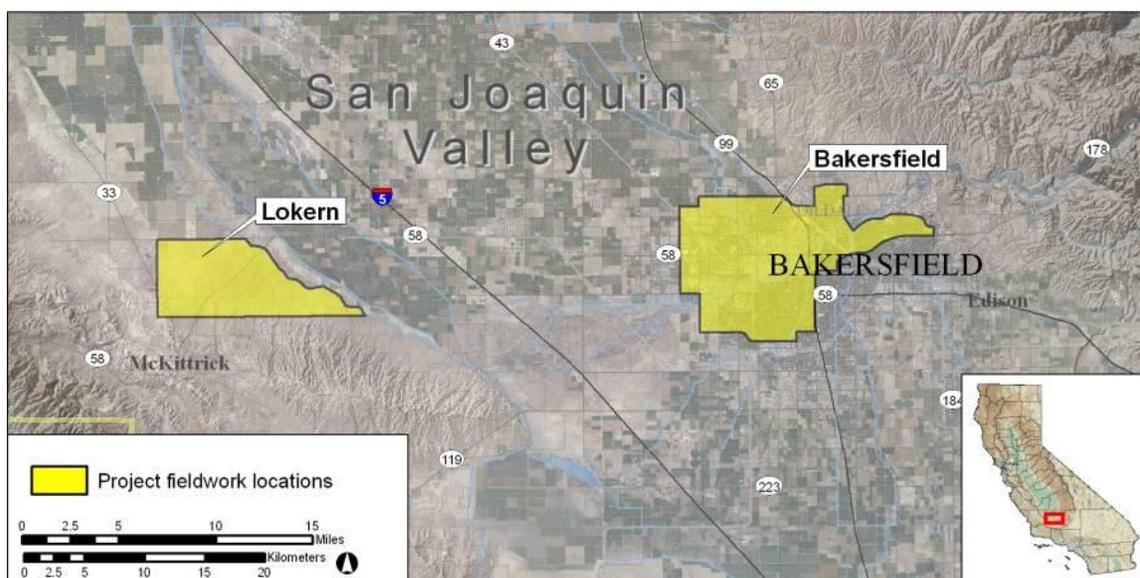


Figure 1 - Location of study sites for behavioral of San Joaquin kit fox.

### Urban study site – Bakersfield, Kern County

The city of Bakersfield is located in Kern County at the southern end of the San Joaquin Valley, CA and covers approximately 300 km<sup>2</sup> (115 mi<sup>2</sup>) with the full metropolitan area covering 580 km<sup>2</sup> (224 mi<sup>2</sup>). The city currently has a population of approximately 328,692, which is projected to continue to grow (Bakersfield Chamber of Commerce 2010). In 2008, the population of the metropolitan Bakersfield area was listed as 800,458, which was up 20.98% from the 2000 Census (United States Census 2008).

Bakersfield has a large self-sustaining population of kit foxes which appear to have successfully adapted to the urban environment, modifying their diet to include anthropogenic food items and reproducing within short distances of human businesses and residences (Cypher *et al.*, 2003, Cypher 2010). Foxes are distributed throughout the city in areas with open ground such as college and university campuses, school campuses, business districts, water collection sumps, water movement canals, undeveloped lots and golf courses. Due to an abundance of food availability, reproductive levels are likely high enough to sustain the removal of surplus animals, which may be from areas of the city that are reaching carrying capacity, or genetically surplus.

### **Natural lands population – Lokern Natural Area**

The Lokern Natural Area is located on the western side of Kern County, approximately 60 km west of Bakersfield, and is considered part of the core area for San Joaquin kit fox recovery (U.S. Fish and Wildlife Service 1998). The area comprises both native and non-native vegetation, consisting of grasses, forbs and shrubland areas. The Lokern Natural Area is owned and managed by a number of entities. Some measure of management coordination is achieved via recommendations and information-sharing through the Lokern Co-ordination Group.

Lokern contains a number of species of importance to kit foxes, both in terms of prey and predators. Known prey species found throughout the study site include three species of kangaroo rats (*Dipodomys nitratooides*, *D. heermanni*, and *D. ingens*), along with grasshopper mice (*Onychomys torridus*), deer mice (*Peromyscus maniculatus*), pocket gophers (*Thomomys bottae*), antelope squirrels (*Ammospermophilus nelsoni*) leporids, and insects (Nelson 2005). Coyotes, the primary predator of San Joaquin kit foxes, are found mainly in shrubland areas of Lokern but are also sighted on a regular basis in grassland areas where the majority of kit fox dens are located. The demographics and ecology of the San Joaquin kit fox population at Lokern have been investigated by ESRP through several studies. In most years, this population is sufficiently demographically robust that some individuals could be relocated to new sites.

### **Kit fox capture and handling**

Kit fox capture and handling techniques were identical in both the urban and natural lands environment, and for Experiment One and Two. A detailed description is provided here and applies to the remainder of the methods section. Trapping and handling was conducted in accordance with permits TE023496-1 and TE825573-2 from the U.S. Fish and Wildlife Service and a Memorandum of Understanding from the Californian Department of Fish and Game. Trapping took place each year between May 1 and January 15 as outlined in permit protocols. No trapping was conducted between January 16 and April 30 during the pupping season due to potential detrimental effects on young litters.

Kit foxes were captured in wire-mesh box live traps (measuring 38 x 38 x 107 cm) that were set up at dusk, covered with a tarpaulin to protect foxes from inclement weather and sun, and baited with a variety of food items. To reduce tooth injuries, each trap contained two rope chew toys, with one attached to each end of the trap. Traps were checked at

dawn; any trap not containing a fox was collapsed and removed to prevent entry by any other animals during daylight hours.

Captured foxes were coaxed from the trap into a handling bag measuring approximately 75 x 75 cm. Using this method, the animal was manually restrained, precluding the need for chemical immobilization and associated risks. The handling bag not only restrains the fox, but also covers its eyes and affords it a sense of security, and most foxes are generally calm while in the bag. During processing, various parts of the fox were exposed for data collection and handling purposes. Foxes were weighed, sexed, fitted with a unique numbered ear-tag, aged, checked for injuries, and genetically sampled. Genetic samples comprised a 2-mm tissue sample collected with a biopsy punch (Miltex Inc., Pennsylvania, USA) from a pinna and stored in alcohol, and 25-50 hairs with roots stored in a coin envelope. Pups captured for Experiment One were also given a dye mark for visual identification using a non-toxic hair dye (Nyanzol-D). Foxes of an appropriate weight were fitted with a radio-telemetry collar weighing 40 g (Advanced Telemetry Systems, Isanti, Minnesota). Once handling was completed, foxes were released at the capture site.

## ***EXPERIMENT ONE***

Experiment One focused on assessing boldness levels of pups at natal dens within each study site and then collecting survival, movement and reproductive data of individuals following dispersal from the natal den. The aim of this process was to provide a behavioral profile for each individual fox, assess individual fitness, and then determine whether the boldness variable was playing a role in success rates. Boldness at both the population and individual level could then be compared between the urban and natural lands to assess whether boldness played a role in survival, distances moved, and reproductive output. The protocol for this experiment comprised trapping and collaring adults within each study sites and tracking them through the breeding season to locate natal dens. Pups were then trapped and individually marked using a non-toxic dye (Nyanzol-D; Figure 2), and behaviorally assessed using both novel stimulus and non-stimulus observations. Observed pups were then trapped and collared to assess survival, dispersal and reproduction.



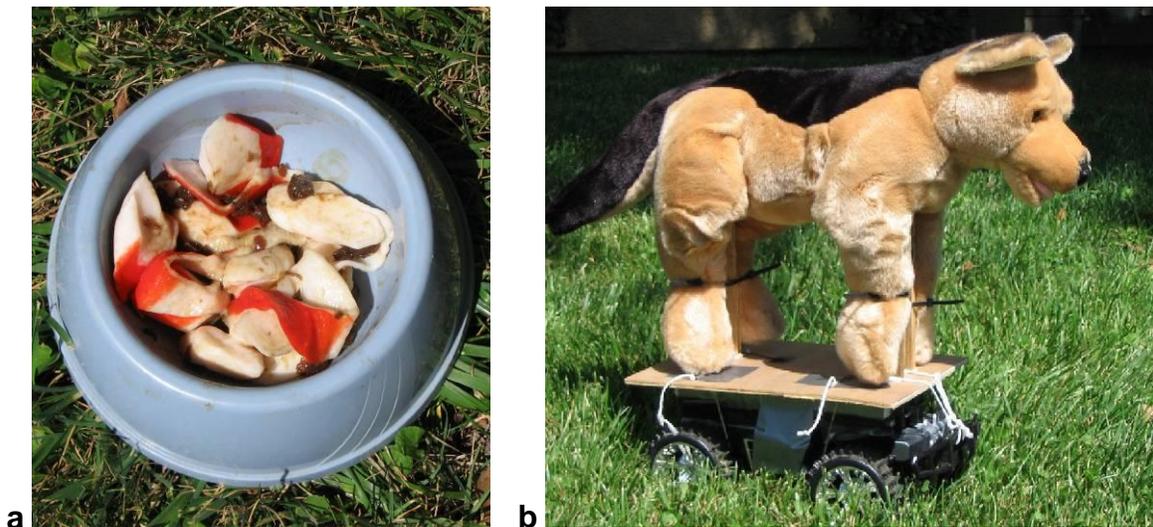
**Figure 2 - San Joaquin kit fox pup dye-marked with a vertical stripe for observations.**

## Behavioral observations

Behavioral profiling comprised four observation periods per natal den. Two of the observation periods recorded behavior with a novel stimulus present at the den; the remaining two observation periods had no novel stimulus present and thus collected baseline boldness data. Each observation period lasted for 60 minutes, and began 30 minutes before sunset. Prior to the start of the observation the observer would go to the natal den area, situate themselves and the observation equipment in a position where they could see the den but not influence the behavior of the foxes, and if necessary place the novel stimulus at the den site. Observations were conducted using 12 x 50 binoculars (Ranger Edition, Eagle Optics, Middleton, Wisconsin, USA), and the digital voice recorder on a Mio Digi Walker hand-held computer (Mio Technology). Observations were also filmed using a digital video recorder (Sony Handycam DCR-HC46) to provide a data back-up.

Novel stimuli consisted of one potentially beneficial stimulus (PBS) and one potentially threatening stimulus (PTS). The PBS was a novel food source, presented in a small pet food bowl (Figure 3a). The food source was a mixture of imitation Krab meat bought from a local supermarket, Mouse-Special Bait – a commercially available trapping bait (R & M Lures, Iowa, USA) and Canine Call - a commercially available trapping lure (The Snare Shop, Iowa, USA). The PTS was designed to simulate a possible predator (Figure 3b). A large plush toy dog (Toys R Us) was mounted onto the base of a modified remote-controlled toy vehicle. A cd player and speakers were inserted into a slit in the back of the toy dog and played a series of coyote howls and a coyote-grey fox (*Urocyon cinereoargenteus*) fight interaction. In addition, the toy dog was doused in coyote urine (The Snare Shop, Iowa, USA).

Behavioral observations were conducted using instantaneous scan sampling at 1-minute intervals (Martin and Bateson 1983). The behavior of each pup was recorded using an adapted version of an ethogram previously developed for swift fox (Table 1; Bremner-Harrison *et al.* 2004).



**Figure 3 - Potentially beneficial stimulus (a) and threatening stimulus (b) for testing boldness response of San Joaquin kit foxes.**

**Table 1: Behavioral ethogram used in Experiment One observation.**

<b>Behavior</b>	<b>Definition</b>
1. In den	below ground in the den
2. Resting relaxed	lying or sitting in relaxed posture/asleep, ears lowered, eyes may be closed
3. Resting alert	lying, sitting or standing with ears erect and eyes open
4. Stretching	elongating limbs with a bout of yawning
5. Rolling	rubbing face and body on ground or object
6. Walking	slowest gait of locomotion
7. Trotting	steady pace faster than walk, lift diagonal pairs of legs
8. Loping	slow bouncy run
9. Running	fastest pace of locomotion
10. Jumping	leaping either into the air or on an object
11. Climbing	prolonged effort to climb up an object
12. Sniffing	sniffing at the air, nose up
13. Investigating	walking, running or standing sniffing at ground or object in enclosure
14. Bold approach - object	direct approach towards novel stimulus, ears erect
15. Bold approach - conspecific	direct approach towards conspecific, ears erect
16. Hesitant approach - object	slow approach towards novel stimuli with frequent retreats and advances, ears and body usually lowered
17. Hesitant approach-conspecific	slow nervous approach towards conspecific with frequent retreats and advances, ears and body usually lowered
18. Chasing conspecific	chasing a conspecific not in play, often away from a novel stimulus or food item
19. Fleeing	run towards den or away from object, often in response to a warning bark
20. Fleeing conspecific	moving quickly away from conspecific
21. Following conspecific	moving slowly behind a conspecific, not chasing
22. Stalking	approaching an object/prey item in a crouched position
23. Pouncing on object	leaping onto an object using forelegs to land, often occurs after stalking
24. Pouncing on conspecific	leaping onto conspecific, often occurs during an existing play bout, or as an invitation to play
25. Fighting	aggressive interaction between conspecifics
26. Fighting over object	aggressive interaction as a result of competition over object
27. Discipline	snapping or growling at a conspecific, may knock them to the ground and stand over them. Usually performed by an adult, directed towards pups
28. Submission	directed towards conspecific, lowered posture, ears flattened, often wagging tail
29. Play chase	running, chasing alone or with other conspecifics, often alternate role of pursuer
30. Play flee	running away from a conspecific or object, ears more erect than Fleeing. When with another conspecific, often alternate who is chasing and fleeing
31. Play fight	wrestling, tumbling, biting and jumping with a conspecific
32. Play stalk	slow approach to conspecific with body held low to ground, occurs within a play bout
33. Playing with object	biting, tossing in the air, or jumping with an object
34. Digging	using front paws to make holes
35. Eating	all masticatory behaviors associated with food
36. Drinking	intake of liquid
37. Food gathering	collecting and carrying items of food in the mouth
38. Food offering	presenting a food item to a conspecific
39. Food beg	position of mouth and nose close to mouth of conspecific whilst wagging tail
40. Caching	storing food item, usually by placing it in a small hole, and covering with it debris
41. Unearthing food	retrieving a previously cached food item
42. Hunting	predatory behavior towards prey item, including stalking and jumping on/catching
43. Defecating	discharge feces or urine from body
44. Scent marking	scent marking, either by depositing minimal amount of urine, scat, or rubbing body on a prominent object in enclosure
45. Grooming (self)	biting, licking, nibbling or scratching at own body
46. Grooming (conspecific)	biting, licking, nibbling or scratching at a conspecific
47. Greeting conspecific	ears back, head low, tail wagging
48. Watching (conspecific)	observing another fox within the enclosure

49. Watching (observer)	looking at the observer who is collecting data
50. Pup carrying	adult moving pup to another area by scruff of the neck
51. Warning bark	short loud bark, usually emitted by an adult at perceived danger
52. Suckling	pup feeding by sucking at mother's teat
53. Entering den	moving out of sight into a den hole
54. Leaving den	coming into view from a den hole
55. Vomiting	regurgitation of food
56. Left den site	is not present or is leaving den site area (away when fox can no longer see the den)
57. Arriving den site	returning to den site area after a period away
58. Out of sight	fox is not visible
59. Location unknown	unknown whether fox is in den or away from den
60. Food carrying	bringing food into the den site or moving it around within den site
61. Play bow	front elbows on ground, head lowered, rear in air, tail wagging

### **Survival, reproductive and movement data**

Following completion of the behavioral observations, pups were re-trapped and fitted with radio-collars (for capture and handling see section on Kit fox capture and handling). Foxes were tracked to their daytime resting location a minimum of once per week to determine their survival status and movements. If a fox could not be located during the day initially, it was then searched for at night during the usual periods of activity for San Joaquin kit foxes. If individuals were not located for several consecutive weeks an aerial search was conducted. Location data were entered into ArcView GIS (Version 3.2, ESRI).

During the pup season any instances of denning with another individual were recorded to ascertain whether pups might have paired. During the pup season, den watches were conducted during early evening until darkness to determine whether individuals had produced pups.

### ***EXPERIMENT TWO***

Experiment Two consisted of focal observations conducted on collared adult and juvenile foxes. Foxes in both the urban and natural lands environment were tracked to their daytime resting location and a novel object placed close to the den entrance. The novel object was neither potentially beneficial nor threatening (Figure 4). When the fox emerged from the den its behavior was recorded using continuous sampling (Martin & Bateson 1983) using an ethogram specific to this experiment (Table 2). Observations ran for one hour, starting 30 minutes before sunset and ending 30 minutes after sunset. If a fox did not emerge during the observation period the novel stimulus was removed and the observation repeated on a different day. Observations were again conducted using 12 x 50 binoculars (Ranger Edition, Eagle Optics, Middleton, Wisconsin, USA), and the digital voice recorder on a Mio Digi Walker hand-held computer (Mio Technology). Observations were also filmed using a digital video recorder (Sony Handycam DCR-HC46) to provide a data back-up.



**Figure 4 - Novel stimulus used to assess boldness response in Experiment Two.**

**Table 2: Behavioral ethogram used for Experiment Two focal observations**

<b>Behavior</b>	<b>Behavioral definition</b>
Not emerged from den	Below ground in the daytime resting den
Observe novel object	Watching the novel object
Investigating novel object	Sniffing or pawing at the novel stimulus
Investigating general	Sniffing or pawing at an item or area other than the novel stimulus
Vigilant	Lying, sitting or standing with ears erect and eyes open
Resting relaxed	Lying or sitting in relaxed posture/asleep, ears lowered, eyes may be closed
Approach	Moving towards the novel stimulus
Retreat	Moving away from the novel stimulus
Out of sight	Fox is above ground but cannot be seen
Grooming	Biting, licking, nibbling or scratching at fur
Back In Den	Fox has gone back below ground
Locomotion	Moving around the den area at either a walk, trot, lope or run
Left Den Site	No longer present at the den site

**Survival, reproductive and movement data**

Foxes were fitted with radio-collars prior to the collection of behavioral data. Survival, reproductive and dispersal data were collected using methods consistent with Experiment One.

***TRAPPING AND HANDLING DATA***

The behavior of all foxes trapped was recorded using a data collection form that recorded aspects of behavior during both time in the trap and during handling. Data were collected in binary format according to whether a behavior was observed or not during the capture process. The data collection form is shown in Appendix A.

## ***DATA ORGANIZATION AND STATISTICAL ANALYSES***

Spatial data was maintained and analyzed using ArcView version 3.2 (ESRI 1996). Statistical analyses were performed using StatView version 5.0 (SAS Institute Inc. 1992), the Excel Data Analyses pack, Microsoft Office 2000, SR-1 Professional and SPSS Statistics Version 17.0. Results were considered significant at  $P < 0.05$ , trends were reported for  $P$  values  $< 0.1$ .

## **RESULTS**

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### ***TRAPPING AND RADIO-COLLARING***

#### **Experiment One**

Adult kit foxes were trapped and collared between 1<sup>st</sup> October 2005 and 15<sup>th</sup> January 2006 in both Bakersfield and Lokern. Fifteen adults were collared in Bakersfield (6M : 9F) and 16 in Lokern (7M : 9F). Radio collars were fitted with both live and mortality signals. Live signals were collected weekly for each fox to determine their continuing survival during the breeding season. The date and location of live signals were recorded and maintained in a central database. Live signals were collected by day in the first instance, but if a fox was not detected during the day then the signal was then searched for at night when foxes are active in order to ascertain that the fox was alive and present on the study site. Beginning in February 2006, remaining living foxes were tracked via their telemetry signals to day-time resting locations in order to determine the presence of potential natal dens.

Evening observations confirmed 9 natal den sites, five in Bakersfield and four in Lokern. The parents moved pups from one of the four dens located in Lokern to a new location in early April. This den could not be located again as the parents continued to den away from the pups. A second litter was moved in May and could not be located, but extensive spotlighting found a replacement litter, giving a total of three litters in Lokern.

Between May and July 2006, there was a total of 28 new captures over 320 trap nights in Bakersfield and Lokern. Of these new captures, 21 pups (11M : 10F) and 2 adults (1M : 1F) were captured over 176 trap nights in Bakersfield. In addition, there were 15 recaptures for pups (7M : 8F) and 10 recaptured adults (6M : 4F). In Lokern, there were 5 (4M : 1F) new captures for pups over 144 trap nights, 0 new captures for adults, 1 recaptured pup (1M : 0F) and 7 (4M : 3F) recaptures for adults. None of the pups at one of the four dens in Lokern could be captured for dye-marking, but given the limited sample size at that study site a decision was made to include them in the behavioral assessment using mean data from all pups combined.

A final breakdown of pups caught at each den site and included in the study is shown in Table 3. The locations of natal den observation sites within each study site are shown in Figure 5 and Figure 6.

**Table 3: San Joaquin kit fox pups trapped at natal dens in the Bakersfield and Lokern study sites for Experiment One observations.**

<b>Study Site</b>	<b>Fox ID</b>	<b>Sex</b>	<b>Den Location</b>	<b>Dye Mark</b>
<i>Bakersfield</i>	6294	Male	Bakersfield College	1 vertical line
	6304	Male	Bakersfield College	1 horz. line
	6306	Male	Bakersfield College	1 spot
	6069	Female	Bakersfield College	2 spots
	6240	Male	CSU Bakersfield	1 horz. line
	6241	Female	CSU Bakersfield	2 horz. lines
	6285	Male	CSU Bakersfield	1 vertical line
	6309	Female	CSU Bakersfield	2 vertical lines
	6286	Female	CSU Bakersfield	2 spots
	6288	Female	CSU Bakersfield	front legs
	6242	Female	CSU Bakersfield	rear legs
	6292	Female	Jewetta	1 horz. line
	6295	Male	Jewetta	1 vertical line
	6245	Female	Jewetta	2 spots
	6298	Female	Jewetta	1 spot
	Unm#1 - Jewetta	unknown	Jewetta	unmark
	6290	Male	7 Oaks Golf Course W	1 horz. line
	6243	Female	7 Oaks Golf Course W	1 vertical line
	6291	Male	7 Oaks Golf Course W	2 spots
	6244	Male	7 Oaks Golf Course W	2 horz. lines
	6065	Male	7 Oaks Golf Course W	front legs
	6246	Female	7 Oaks Golf Course W	unmark
	6287	Male	State Farm Field	1 horz. line
6310	Male	State Farm Field	unmark	
<i>Lokern</i>	6305	Male	North - D680	1 horz. line
	6293	Male	North - D680	1 vertical line
	6307	Male	North - D680	2 spots
	6299	Male	North - D680	unmark
	6289	Male	North - D704	1 horz. line
	Unm#1 - 704	unknown	North - D704	unmark
	Unm#2 - 704	unknown	North - D704	unmark
	Unm#1 - 405	unknown	South - D405	unmark
Unm#2 - 405	unknown	South - D405	unmark	

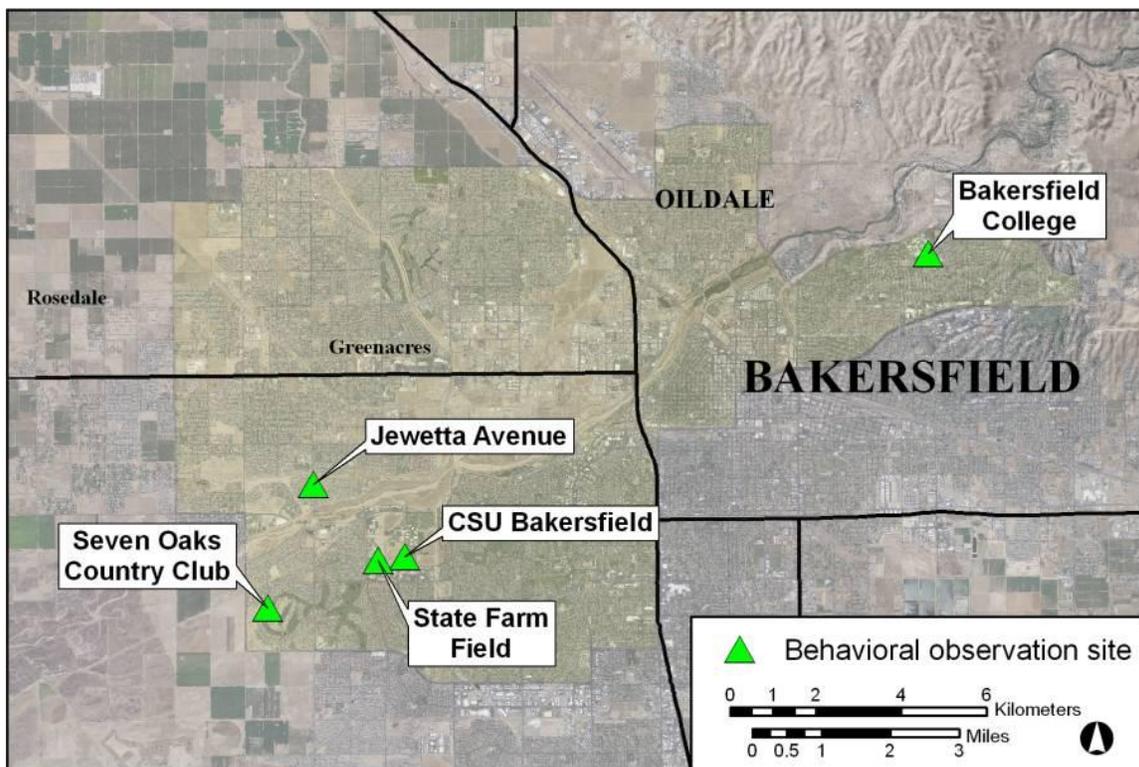


Figure 5 - Natal den site locations of San Joaquin kit fox pups observed in Bakersfield.

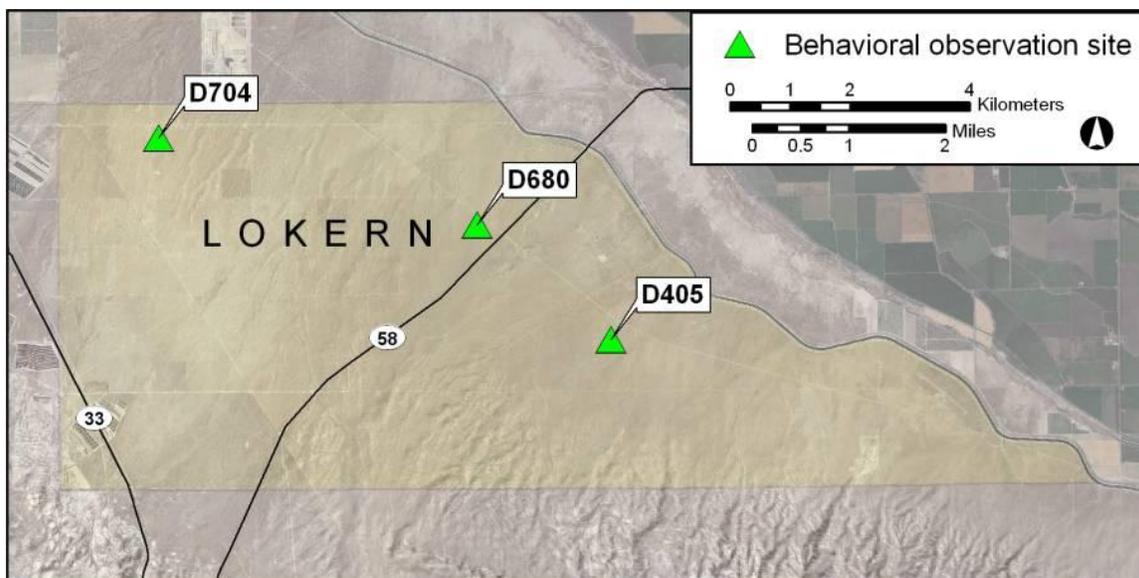


Figure 6 - Natal den site locations of San Joaquin kit fox pups observed in Lokern.

Experiment One behavioral observations ran from May 15, 2006 to July 3, 2006 in Bakersfield, and from June 7, 2006 to July 8, 2006 in the Lokern Natural Area. Trapping to fit radio-collars onto observed pups ran from July 19 to August 31, 2006 in the urban environment. During this period 20 pups were radio-collared (Table 4). Trapping to fit radio collars at the Lokern study site ran from October 5, 2006 to January 15, 2007. During this period only 1 of the previously trapped and observed pups was recaptured for

collaring (Table 4). Additional foxes were collared at this time at the Lokern study site for Experiment Two (see section entitled “Experiment Two” below for details).

**Table 4: Pups radio-collared for Experiment One for survival, movement and reproductive data.**

<b>Study Site</b>	<b>Fox ID</b>	<b>Sex</b>	<b>Location</b>
<i>Bakersfield</i>	6294	Male	Bakersfield College
	6304	Male	Bakersfield College
	6306	Male	Bakersfield College
	6069	Female	Bakersfield College
	6241	Female	CSU Bakersfield
	6285	Male	CSU Bakersfield
	6309	Female	CSU Bakersfield
	6286	Female	CSU Bakersfield
	6288	Female	CSU Bakersfield
	6242	Female	CSU Bakersfield
	6292	Female	Jewetta
	6295	Male	Jewetta
	6245	Female	Jewetta
	6298	Female	Jewetta
	6243	Female	7 Oaks Golf Course W
	6291	Male	7 Oaks Golf Course W
	6244	Male	7 Oaks Golf Course W
	6065	Male	7 Oaks Golf Course W
	6246	Female	7 Oaks Golf Course W
	6287	Male	State Farm Field
6310	Male	State Farm Field	
<i>Lokern</i>	6299	Male	North – D680

## Experiment Two

Foxes in Bakersfield and the Lokern Natural Area were trapped in 2006/2007 and 2008/2009 for behavioral assessment of boldness. The locations of trapped foxes at each study site are shown in Figure 7 and Figure 8. Table 5 and Table 6 show the ID numbers, age class, sex and location of foxes radio-collared and observed for Experiment Two. Behavioral observations of 27 known individuals and 5 unidentified foxes took place between December 19, 2006 and April 29, 2009 in Bakersfield. Observation of 27 known individuals and 2 unidentified foxes were conducted in Lokern between January 8, 2007 and April 13, 2009. A number of additional foxes were collared for observation in each study site but died prior to being observed. However, these foxes were included in the Trap/Handling behavioral assessment.

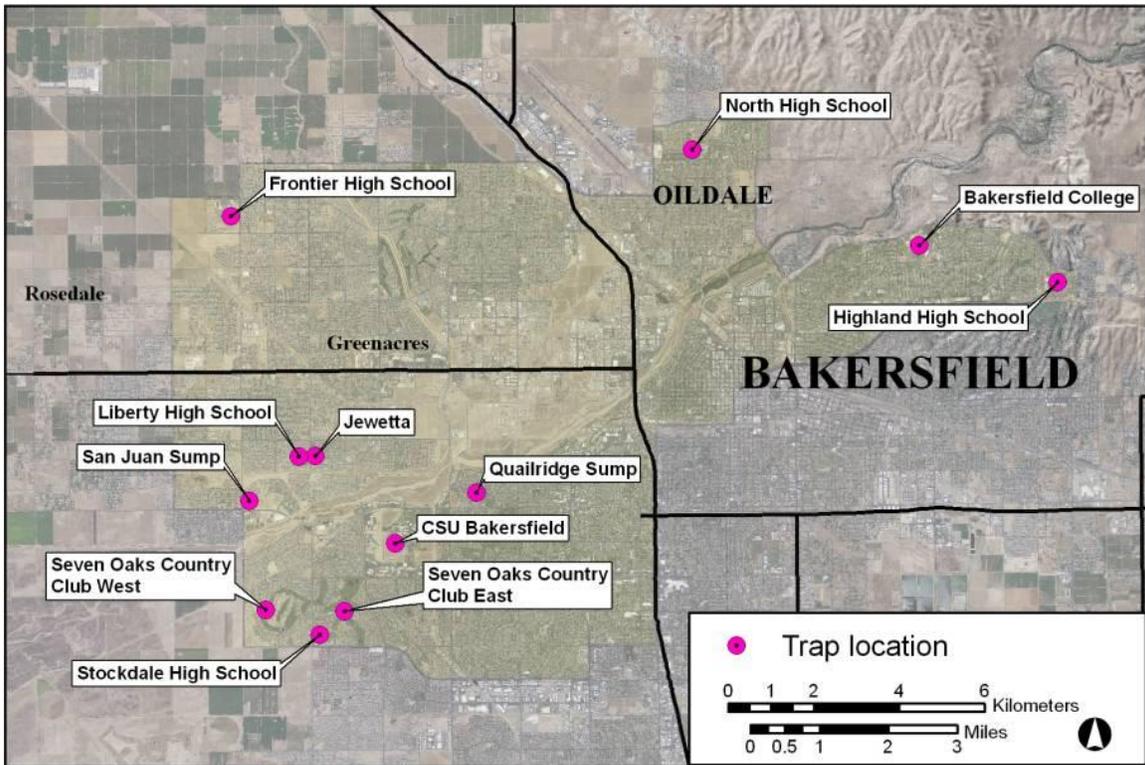


Figure 7 - Location of foxes trapped and radio-collared in Bakersfield for Experiment Two observations. See Table 4 for fox identification numbers at each site.

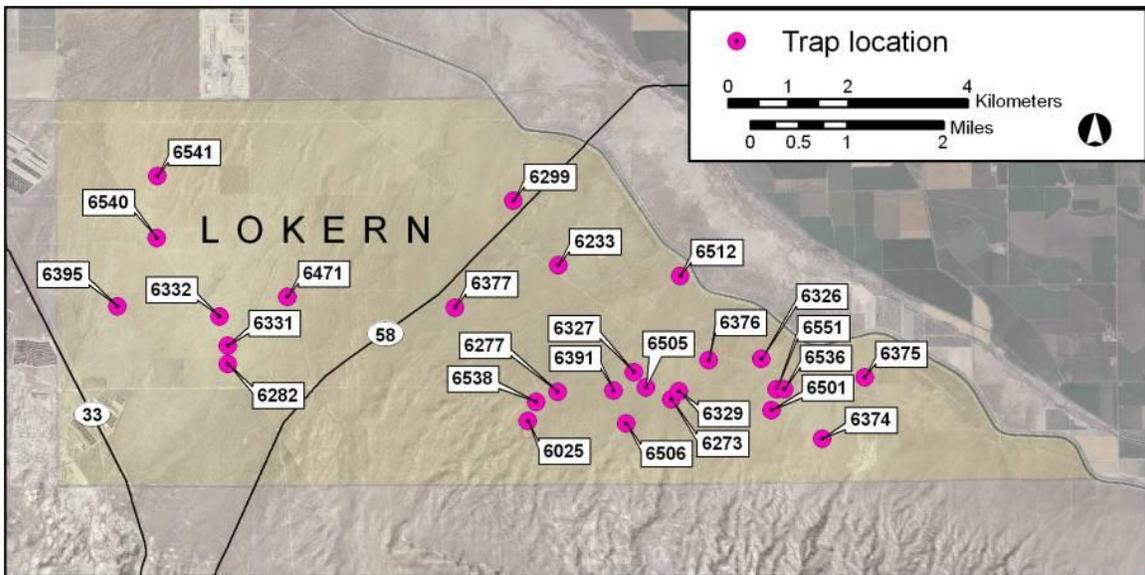


Figure 8 - Location of foxes trapped and radio-collared in the Lokern Natural Area for Experiment Two observations.

**Table 5: Urban San Joaquin kit foxes observed for Experiment Two.**

<b>Study Site</b>	<b>Fox ID</b>	<b>Age</b>	<b>Sex</b>	<b>Location</b>
<i>Bakersfield</i>	5678	adult	female	Stockdale High School
	6065	yearling	male	7 Oaks West
	6243	yearling	female	7 Oaks West
	6246	yearling	female	7 Oaks West
	6067	yearling	female	7 Oaks East
	6069	yearling	female	Bakersfield College
	6392	adult	male	Bakersfield College
	6241	yearling	female	CSU Bakersfield
	6309	yearling	female	CSU Bakersfield
	6244	yearling	male	CSU Bakersfield
	6398	adult	female	CSU Bakersfield
	6266	adult	female	CSU Bakersfield
	6286	yearling	female	CSU Bakersfield
	6288	yearling	female	CSU Bakersfield
	6292	yearling	female	Jewetta
	6298	yearling	female	Jewetta
	6365	adult	female	Highland High School
	6351	adult	male	Highland High School
	6362	adult	male	Liberty High School
	6527	adult	female	Liberty High School
	6532	adult	male	Frontier High School
	6369	yearling	male	Frontier High School
	6366	adult	female	North High School
	6530	yearling	female	North High School
	6531	adult	male	North High School
	6534	adult	male	Quailridge Sump
	6535	adult	male	San Juan Sump

**Table 6: Natural lands San Joaquin kit foxes observed for Experiment Two.**

Study Site	Fox ID	Age	Sex	Location
<i>Lokern</i>	6025	adult	female	South of Hwy 58
	6233	adult	female	South of Hwy 58
	6273	adult	female	South of Hwy 58
	6277	adult	female	South of Hwy 58
	6326	adult	female	South of Hwy 58
	6327	adult	female	South of Hwy 58
	6329	adult	female	South of Hwy 58
	6374	yearling	male	South of Hwy 58
	6375	adult	male	South of Hwy 58
	6376	yearling	female	South of Hwy 58
	6377	adult	female	South of Hwy 58
	6391	yearling	male	South of Hwy 58
	6501	adult	female	South of Hwy 58
	6505	adult	female	South of Hwy 58
	6506	adult	female	South of Hwy 58
	6512	adult	female	South of Hwy 58
	6536	adult	female	South of Hwy 58
	6538	yearling	male	South of Hwy 58
	6551	yearling	male	South of Hwy 58
	6282	adult	female	North of Hwy 58
	6299	pup	male	North of Hwy 58
	6331	adult	female	North of Hwy 58
	6332	adult	female	North of Hwy 58
	6395	adult	female	North of Hwy 58
	6471	adult	female	North of Hwy 58
	6540	yearling	female	North of Hwy 58
	6541	adult	female	North of Hwy 58

## ***OBSERVATIONAL DATA***

### **Boldness Scores - Experiment One**

A total of 32 hours of observational data were collected for Experiment One from five dens in Bakersfield and three in Lokern. Boldness scores were calculated for each dye-marked individual. Mean scores were calculated at dens with un-marked individuals. Behavioral activities representing overly bold, bold, shy, and overly shy were identified within the ethogram and categorized into behavior type. The number of occurrences of behavioral activities within each behavior type was counted. Boldness scores were calculated using modifications from a previously developed method whereby the number of occurrences of overtly bold activities were multiplied by 3; bold activities by 2; shy activities by 1; and overtly shy activities by -1. These four values were summed to give a score for each individual per stimulus. A total boldness score for each individual was calculated by summing the scores for the three stimuli. High scores represented high

levels of boldness and low scores represent lower levels of boldness. Boldness scores are shown in Table 7.

Analysis of boldness scores for all foxes across the four stimuli showed that individual foxes were consistent in their behavior ( $W = 0.43$ ,  $X^2_{32} = 55.04$ ,  $P < 0.01$ ; Kendall's Coefficient of Concordance, corrected for tied ranks; Siegel 1956). Therefore, a fox that scored highly for boldness in one observation scored highly in all four observations.

**Table 7: Individual fox boldness scores obtained from Experiment One novel object tests in Bakersfield and Lokern.**

Location	Den	Fox ID	Observation Type				Total Score
			Non Stimulus 1	Non-Stimulus 2	PBS	PTS	
<i>Bakersfield</i>	Bakersfield	6294	-6	88	-41	-60	-19
		College	6304	-9	-37	-54	-60
		6306	-60	-60	-60	-60	-240
		6069	-60	19	4	-41	-78
	CSUB	6240	30	103	5	-60	78
		6241	-58	105	-57	-60	-70
		6285	15	100	18	-60	73
		6309	2	-60	27	-60	-91
		6286	57	114	14	-60	125
		6288	-57	-60	-1	-60	-178
		6242	-5	-60	-17	-60	-142
		Jewetta	6292	36	41	15	-60
	6295		104	90	139	-60	273
	6245		27	-1	-60	-60	-94
	6298		100	91	120	-60	251
	unm2		32	46	-60	-60	-42
	State Farm	6287	111	15	-24	-60	42
		6310	4	28	-21	-60	-49
	Seven Oaks	6290	7	-7	-58	-60	-118
		West	6243	21	14	-56	-34
	6291		-15	-7	-42	-60	-124
	6244		-10	21	-60	-60	-109
	6065		4	23	-60	-60	-93
6246	-27		-36	-60	-60	-183	
<i>Lokern</i>	Den 680	6305	-21	95	-60	-60	-46
		6293	-26	90	-36	-60	-32
		6307	-35	87	-25	-60	-33
		6299	-32	64	-38	-60	-66
	Den 704	6289	-11	-54	-60	-60	-185
		unm1	-18	-35	-60	-60	-173
		unm2	-55	-33	-60	-60	-208
	Den 405	unm1	-47	-60	-60	-60	-227
		unm2	-60	-60	-60	-60	-240

## Boldness scores - Experiment Two

A total of sixty-one hours of focal observation data were collected for Experiment Two from Bakersfield and Lokern. Observations were transcribed by calculating the amount of time spent by each individual on the behaviors listed in Table 2. As foxes were not visible for equal time periods within observation periods, data were transformed into percent data per category of the time visible.

Boldness scores were calculated by categorizing behavioral activities within the ethogram as bold or shy (Table 8). The behavioral categories ‘Not Emerged from Den’ and ‘Left Den Site’ were excluded from the analysis as it was not possible to ascertain whether these categories were motivated by variables external from the novel object test, for example, motivation to hunt. Bold behaviors were given a weighting of 2 and shy behaviors were given a weighting of -1. Scores were then summed across the categories to give a total score for each fox observed. Again, higher scores represented foxes that performed a greater proportion of bold-type behavior, and lower scores represented foxes that performed more shy-type behavior (Table 9).

**Table 8: Shy and bold behavior classification for Experiment Two Novel Object Tests.**

Bold Behaviors	Shy Behaviors
Investigating Novel Object	Observe novel object
Investigating General	Resting Alert
Resting Relaxed	Retreat
Approach	Back In Den
Grooming	

Table 9: Experiment Two boldness scores – Bakersfield and Lokern populations.

Fox ID	Age	Sex	Shy Behavior						Bold Behavior						Total Score	
			Observe novel object	Resting Alert	Retreat	Back In Den	Σ shy percent	Shy score (x -1)	Investigating Novel Object	Investigating General	Resting Relaxed	Approach	Grooming	Σ bold percent		Bold score (x 2)
<b>Bakersfield</b>																
5678	A	F	4.1	2.7	1.1	0.0	7.9	-7.9	0.3	1.6	0.2	1.3	5.5	8.9	17.8	10.0
6065	Y	M	1.3	4.3	0.3	0.0	5.9	-5.9	0.7	4.8	4.1	1.1	6.2	16.9	33.7	27.8
6067	Y	F	7.5	19.2	2.5	0.0	29.2	-29.2	13.3	0.0	0.0	2.5	0.0	15.8	31.7	2.5
6069	Y	F	0.0	2.0	0.0	98.0	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6241	Y	F	5.6	7.4	11.1	0.0	24.1	-24.1	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-24.1
6243	Y	F	0.1	0.6	0.0	99.3	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6244	Y	M	5.1	42.1	0.0	2.0	49.2	-49.2	0.0	2.8	8.3	0.0	26.7	37.7	75.4	26.2
6246	Y	F	0.3	0.0	0.0	99.7	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6266	A	F	0.0	2.6	0.0	94.3	96.9	-96.9	1.8	1.2	0.0	0.2	0.0	3.1	6.3	-90.6
6286	Y	F	0.7	0.5	0.2	0.0	1.5	-1.5	1.8	2.5	30.9	0.5	3.3	38.9	77.8	76.3
6288	Y	F	2.3	6.3	0.0	0.0	8.5	-8.5	39.2	29.0	0.0	2.8	0.0	71.0	142.0	133.5
6292	Y	F	0.0	0.0	0.0	100.0	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6298	Y	F	0.6	0.0	0.0	15.4	16.0	-16.0	5.0	2.2	0.6	0.9	72.0	80.8	161.6	145.6
6309	Y	F	0.3	1.0	0.5	0.0	1.8	-1.8	1.7	2.9	0.0	2.0	0.0	6.6	13.1	11.3
6351	A	M	14.8	42.6	7.4	0.0	64.8	-64.8	0.0	0.0	0.0	16.7	0.0	16.7	33.3	-31.5
6362	A	M	2.3	14.8	0.7	26.3	44.1	-44.1	2.3	8.9	0.0	0.2	1.3	12.7	25.5	-18.6
6365	A	F	2.4	8.7	3.3	32.9	47.3	-47.3	0.0	2.0	0.0	2.4	0.0	4.4	8.9	-38.4
6366	A	F	2.8	21.9	0.0	0.0	24.8	-24.8	0.0	1.9	41.7	0.3	12.2	56.1	112.3	87.5
6369	Y	M	0.7	3.9	0.0	0.0	4.7	-4.7	2.4	0.0	0.8	0.5	14.7	18.4	36.8	32.1
6392	A	M	1.8	2.0	0.7	95.0	99.5	-99.5	0.0	0.2	0.0	0.2	0.0	0.5	1.0	-98.5
6398	A	F	8.7	0.4	0.0	73.3	82.5	-82.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-82.5
6527	A	F	5.3	23.2	0.0	0.0	28.5	-28.5	0.0	0.8	0.0	0.2	0.0	1.0	2.1	-26.4
6530	Y	F	3.1	7.0	0.4	87.9	98.4	-98.4	0.0	0.3	0.0	1.0	0.1	1.5	2.9	-95.5
6531	A	M	6.0	9.0	0.4	68.1	83.4	-83.4	1.0	9.6	0.0	4.4	0.0	15.0	29.9	-53.5
6532	A	M	62.1	37.9	0.0	0.0	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6534	A	M	1.8	39.7	0.0	0.0	41.6	-41.6	0.0	1.1	36.1	0.2	21.0	58.4	116.7	75.2
6535	A	M	1.7	0.0	0.0	93.9	95.5	-95.5	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-95.5
<b>Lokern</b>																
6025	A	F	2.2	16.7	0.3	72.1	91.3	-91.3	0.0	0.5	0.0	0.4	0.0	0.9	1.8	-89.5
6233	A	F	0.8	0.0	0.0	99.2	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6273	A	F	28.9	65.6	0.0	0.0	94.4	-94.4	0.0	5.6	0.0	0.0	0.0	5.6	11.1	-83.3
6277	A	F	0.1	0.1	0.0	99.8	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6282	A	F	2.5	0.0	0.0	97.5	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6299	P	M	1.7	4.2	0.0	94.2	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6326	A	F	23.0	11.9	0.4	63.8	99.1	-99.1	0.0	0.9	0.0	0.0	0.0	0.9	1.8	-97.3
6327	A	F	3.9	3.9	0.4	88.3	96.4	-96.4	0.0	0.0	0.0	0.3	0.0	0.3	0.6	-95.8
6329	A	F	0.5	0.0	0.0	99.5	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6331	A	F	7.2	88.7	0.6	0.0	96.5	-96.5	0.0	0.4	0.0	0.0	0.3	0.7	1.4	-95.1
6332	A	F	1.2	0.0	0.9	0.0	2.1	-2.1	0.0	0.0	0.0	0.0	0.0	0.0	0.1	-2.1
6374	Y	M	2.9	7.7	0.0	0.0	10.6	-10.6	2.1	2.4	0.0	2.1	11.9	18.5	37.1	26.5
6375	A	M	5.9	0.8	0.0	93.4	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6376	Y	F	1.3	1.3	0.0	97.5	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6377	A	F	0.1	0.0	0.0	99.9	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6391	Y	M	15.2	17.1	0.1	0.0	32.4	-32.4	1.9	2.5	0.0	3.1	3.6	11.1	22.2	-10.2
6395	A	F	4.3	21.8	0.0	73.9	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6471	A	F	0.2	0.0	0.3	99.5	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6501	A	F	29.9	24.6	0.5	0.0	55.0	-55.0	0.0	0.1	9.6	1.3	0.4	11.5	23.0	-32.0
6505	A	F	0.5	0.4	0.0	99.1	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6506	A	F	41.6	36.5	5.2	6.7	90.1	-90.1	0.0	0.0	0.0	9.9	0.0	9.9	19.8	-70.3
6512	A	F	3.0	1.1	0.6	0.0	4.6	-4.6	0.8	2.0	0.0	1.4	0.0	4.2	8.5	3.9
6536	A	F	15.7	26.5	6.4	0.0	48.6	-48.6	2.5	3.4	0.0	5.4	0.0	11.3	22.6	-26.0
6538	Y	M	8.1	17.6	0.0	74.3	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6540	Y	F	22.9	12.1	1.4	0.0	36.4	-36.4	12.9	1.4	0.0	10.7	0.0	25.0	50.0	13.6
6541	A	F	2.2	34.1	0.0	63.7	100.0	-100.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0	-100.0
6551	Y	M	21.8	5.0	3.3	0.0	30.1	-30.1	16.2	0.0	0.0	6.4	0.0	22.5	45.1	15.0

## Boldness scores – Trap and Handling Behavior

Trap and handling behavior data were collected for 87 foxes in Bakersfield and 67 foxes in Lokern. Boldness scores were calculated from the behavior of each fox during the trapping and handling process. Behavior was recorded on a simple ‘yes or no’ basis regarding whether a particular behavior was performed. The data were then transcribed as binary data. Types of behavior recorded were classified as shy or bold, and given a weighting according to category (Table 10). Shy behavior was given a weighting of -1, bold behavior was given a weighting of 1. Boldness scores were calculated by summing the occurrences of shy and bold behavior to give a boldness value (Table 11). As there was a likelihood that bold and shy behavior may cancel one another out for some foxes, data were transformed by adding 0.5 to each boldness score and the resultant value was divided by the number of shy/bold behaviors performed by each individual. Several foxes had data collected from multiple capture instances. The data obtained from the first recorded capture was used to calculate the boldness score for each individual fox.

**Table 10: Trap/Handling Behavior Data – Bakersfield.**

<b>Bold Behaviors (assigned value of 1)</b>	<b>Shy Behaviors (assigned value of -1)</b>
Biting at cage or bag	Warning bark (combined for trap and bag)
Struggling	Growl/snarl (combined for trap and bag)
Attempt to escape	Scream (combined for trap and bag)
Entered bag calmly	Running backwards & forwards
	Moving away from handler

## Comparison of boldness scores from Experiments 1 and 2 and trap/handling data

Boldness scores were calculated using three different methods: Experiment One, Experiment Two, and the Trap/Handling Behavior. These three methods of data collection differed considerably in terms of the effort and duration required to obtain the data. In order to determine whether the data obtained were comparable across the three methods, data were analyzed for consistency across all three measures. As the numbers of foxes were not consistent for each experiment, a number of different tests were conducted.

Twelve individuals had boldness scores collected using all three methods (Table 11). Consistency for levels of boldness in this instance were analyzed using Kendall’s Coefficient of Concordance as this allows for comparison across greater than two measures. Data were ranked from highest to lowest and corrected for tied ranks. Boldness scores were found to be consistent across the three tests, indicating that the 12 foxes were ranked equally in terms of boldness regardless of which method of data collection was used ( $W = 5.02$ ,  $X^2_{11} = 165.66$ ,  $P < 0.001$ ; Kendall’s Coefficient of Concordance, corrected for tied ranks).

There were no further foxes that had boldness scores for Experiments One and Two only. A total of 21 foxes had boldness scores obtained from Experiment One and the Trap/Handling data, and 52 foxes had boldness scores obtained from Experiment Two

and the Trap/Handling data (Table 11). Consistency for boldness measures between these two sets of data was analyzed using non-parametric Spearman Rank Correlations. No correlation was found between Experiment One and the Trap/Handling data or between Experiment Two and the Trap/Handling data.

**Table 11: Boldness scores obtained from Experiments One and Two, and Trap/Handling Data.**

<i>Bakersfield</i>				<i>Lokern</i>			
Fox ID	Experiment 1	Experiment 2	Trap/Handling (1st capture only)	Fox ID	Experiment 1	Experiment 2	Trap/Handling (1st capture only)
5678		9.96	0.25	6025		-89.50	0.30
6065	-93.00	27.83	0.25	6233		-100.00	0.25
6067		2.50	0.25	6273		-83.33	-0.07
6069	-78.00	-100.00	0.30	6277		-100.00	-0.07
6240	78.00			6282		-100.00	-0.38
6241	-70.00	-24.07	0.25	6289	-185.00		
6242	-142.00		0.08	6293	-32.00		
6243	-55.00	-100.00	0.25	6299	-66.00	-100.00	0.25
6244	-109.00	26.19	1.25	6305	-46.00		
6245	-94.00		0.50	6307	-33.00		
6246	-183.00	-100.00	-0.38	6326		-97.26	-0.17
6266		-90.55	0.13	6327		-95.76	-0.07
6285	73.00		-0.50	6329		-100.00	-0.17
6286	125.00	76.32	0.25	6331		-95.10	-0.38
6287	42.00		-0.50	6332		-2.05	-0.88
6288	-178.00	133.52	-0.83	6374		26.48	0.13
6290	-118.00			6375		-100.00	0.25
6291	-124.00		1.50	6376		-100.00	-0.25
6292	32.00	-100.00	0.50	6377		-100.00	0.25
6294	-19.00		-0.25	6391		-10.22	-0.07
6295	273.00		0.13	6395		-100.00	-0.38
6298	251.00	145.60	0.50	6471		-100.00	-0.25
6304	-160.00			6501		-32.04	-0.10
6306	-240.00		-0.10	6505		-100.00	-0.25
6309	-91.00	11.29	0.25	6506		-70.29	-0.50
6310	-49.00		0.25	6512		3.86	-0.88
6351		-31.48	-0.10	6536		-26.04	0.30
6362		-18.57		6538		-100.00	-0.50
6365		-38.45	0.25	6540		13.57	-0.38
6366		87.55	0.25	6541		-100.00	-0.38
6369		32.12	-0.17	6551		15.03	-0.17
6392		-98.53	-0.38	6035			-0.1
6398		-82.47	0.50	6061			0.083
6527		-26.39		6062			-0.167
6530		-95.50	-0.17	6066			0.125
6531		-53.49	-0.25	6068			0.7
6532		-100.00	-0.38	6071			-0.25
6534		75.17	0.63	6145			0.25
6535		-95.53	-0.25	6158			0.417
J5	-42.00			6239			0.214
1111			-0.375	6268			-0.1
2222			0.25	6270			0.125
6063			0.214	6283			0.083
6064			0.25	6328			0.3
6235			0.3	6330			0.125

<i>Bakersfield</i>				<i>Lokern</i>			
Fox ID	Experiment 1	Experiment 2	Trap/Handling (1st capture only)	Fox ID	Experiment 1	Experiment 2	Trap/Handling (1st capture only)
6249			0.25	6333			-0.167
6250			-0.375	6334			0.125
6256			0.25	6335			0.3
6275			0.75	6336			-0.167
6297			0.5	6378			0.417
6308			-0.1	6387			-0.375
6312			0.25	6388			0.083
6313			0.3	6393			0.25
6314			-0.375	6394			0.214
6315			0.214	6396			0.3
6316			-0.375	6400			0.3
6317			-0.167	6502			0.083
6318			-0.167	6503			0.25
6319			-0.75	6508			-0.167
6320			-0.071	6510			-0.167
6337			1.25	6513			-0.1
6338			-0.167	6515			0.625
6339			-0.167	6516			-0.75
6340			0.25	6517			0.083
6363			0.25	6537			0.214
6364			0.083	6539			0.125
6367			-0.1	6542			-0.167
6370			0.25	6552			0.125
6371			0.7	6553			0.125
6372			0.25	6554			0.3
6373			0.3	6555			-0.167
6379			-0.167	6556			0.214
6380			-0.167				
6381			0.25				
6382			0.3				
6383			-0.75				
6384			0.5				
6385			0.125				
6386			-0.167				
6389			0.063				
6504			1.25				
6507			0.083				
6509			0.25				
6511			0.125				
6528			0.625				
6529			0.3				
6533			0.125				
6543			0.25				
6545			0.125				
6547			-0.167				
6548			-0.167				
6549			1.5				
6565			0.25				
6566			0.25				
6567			-0.375				

## Comparison of boldness scores between habitats

### Experiment One

Boldness scores between the urban Bakersfield site and the natural lands Lokern site were compared using two sample *t*-tests assuming unequal variances. Foxes at the urban study site exhibited significantly higher boldness scores for the baseline and novel object data combined, and a trend for higher levels of boldness for the baseline data (Table 12). Urban foxes were significantly bolder in the presence of novel stimuli (both novel stimulus tests combined) and further analyses determined that urban foxes showed greater levels of bold behavior in the presence of the PBS food stimuli than natural lands foxes but were equally shy as natural lands foxes in the presence of the PTS predator stimulus (Table 12). In addition, the range of boldness scores was higher in the urban habitat than in the natural lands habitat. An index of relative size was calculated by dividing the urban by the natural lands range to determine the magnitude of difference in range between the two sites (Table 13).

**Table 12: Comparison of Experiment One boldness data collected from San Joaquin kit foxes in urban and natural lands habitats (*t*-test assuming unequal variances).**

Urban vs. Natural Lands	Mean Score		<i>t</i>	df	<i>P</i>
	Urban	Natural			
All Observations	-40.5 (SD128.9)	-134.4 (SD 88.4)	2.38	21	<0.05
Baseline	33.9 (SD 91.2)	-23.4 (SD 78.2)	2.11	17	0.09
Novel Stimuli	-74.3 (SD 54.2)	-111 (SD 13.9)	2.05	29	<0.01
PBS (food)	-16.2 (SD 54.4)	-51 (SD 13.9)	2.05	29	<0.01
PTS (predator)	-58.1 (SD 6.4)	-60 (SD 0.0)	1.43	23	NS

**Table 13: Range of Experiment One boldness data collected from San Joaquin kit foxes in urban and natural lands habitats.**

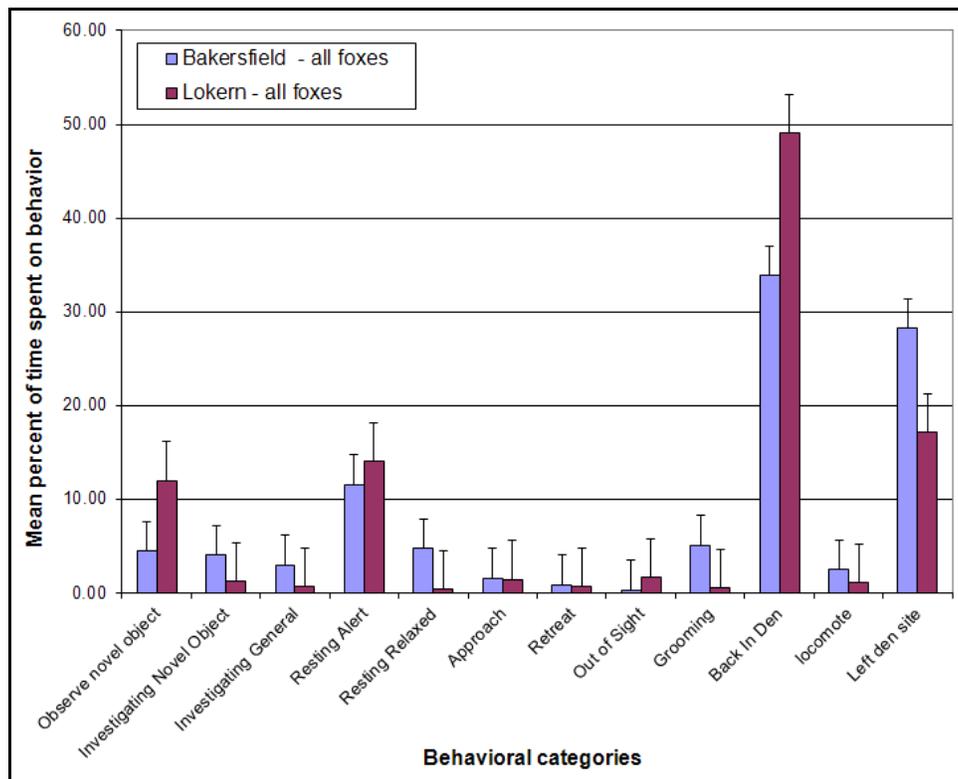
Population	Range				
	All Observations	Baseline	Novel Stimuli	PBS	PTS
Urban	513	314	199	199	26
Natural Lands	208	194	35	35	0
Index or relative size	2.5	1.6	5.7	5.7	26

### Experiment Two

Percent occurrence data were compared for foxes from the urban and natural lands environment (Figure 9-Figure 13). While results were not significantly different for percent occurrence of the various behavioral categories, strong trends were observed for urban foxes spending more time investigating (general), resting relaxed, and grooming in the presence of the novel object (Table 14).

**Table 14: Comparison of overall percent occurrence time spent on behavioral activities by San Joaquin kit foxes in urban vs natural lands habitats.**

Behavior Categories	Means		<i>t</i>	df	<i>P</i>
	Urban	Natural Lands			
Investigating (general)	2.66	0.71	1.69	29	0.10
Investigating novel object	2.58	1.35	0.73	38	NS
Resting relaxed	4.55	0.36	1.85	27	0.08
Approach	1.39	1.52	-0.15	52	NS
Grooming	6.03	0.60	1.87	27	0.07
Resting alert/vigilant	11.10	14.73	-0.73	45	NS
Observe novel object	5.24	9.16	-1.24	52	NS
Retreat	1.06	0.75	0.53	44	NS
Back in den	36.52	52.68	-1.33	52	NS



**Figure 9. Percent occurrence of behavior for all foxes in response to Experiment Two novel object tests in urban and natural lands habitats.**

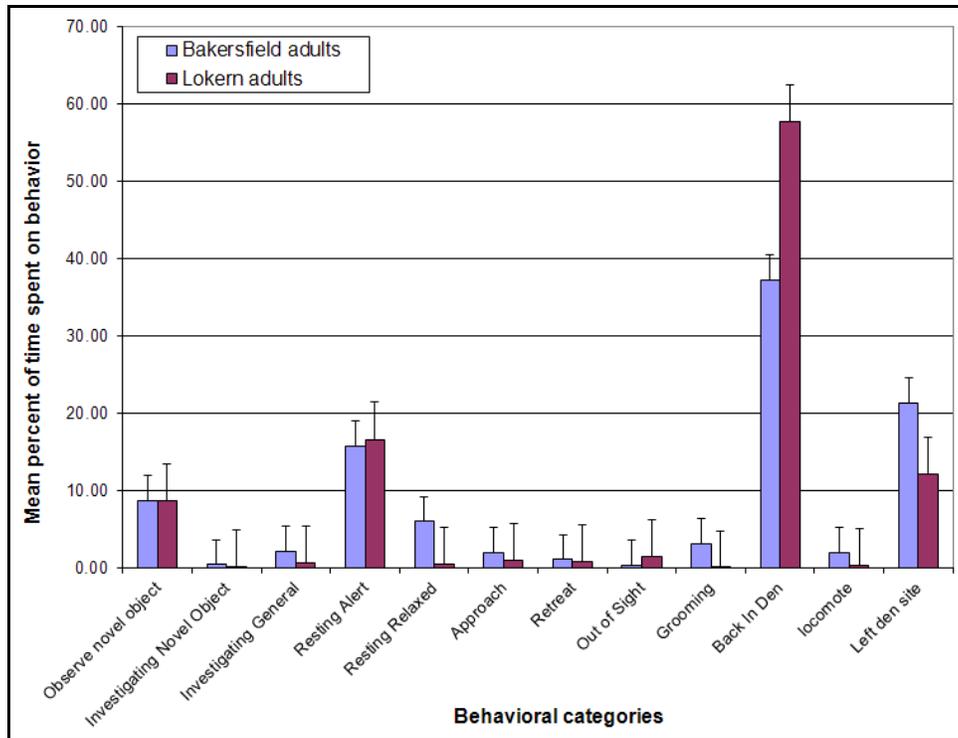


Figure 10. Percent occurrence of behavior for yearling foxes in response to Experiment Two novel object tests in urban and natural lands habitats.

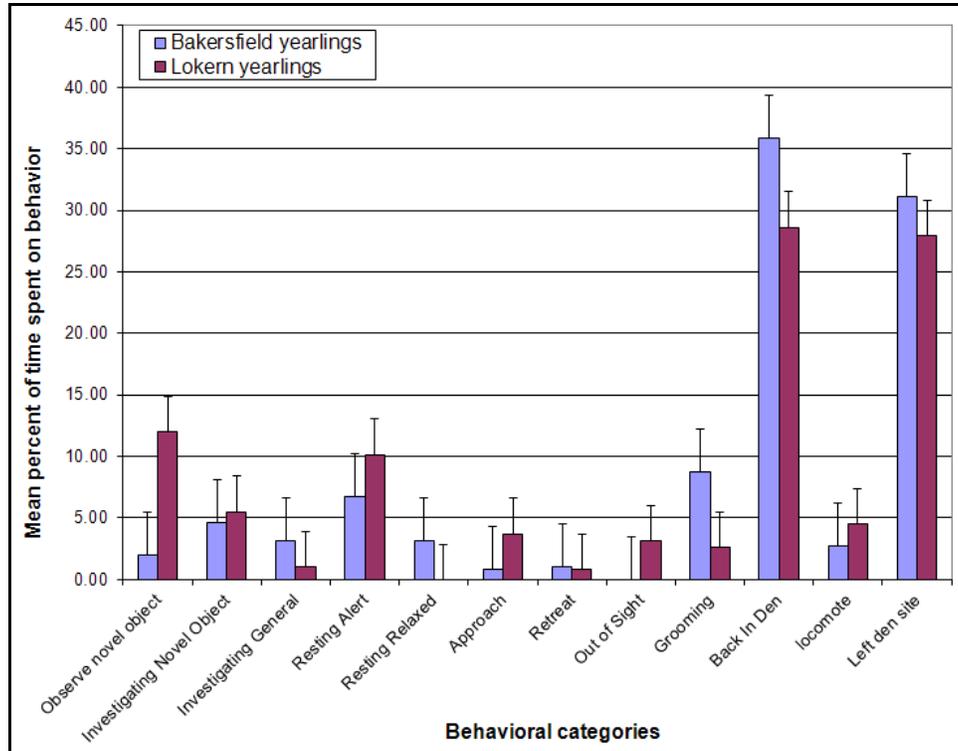


Figure 11. Percent occurrence of behavior for yearling foxes in response to Experiment Two novel object tests in urban and natural lands habitats.

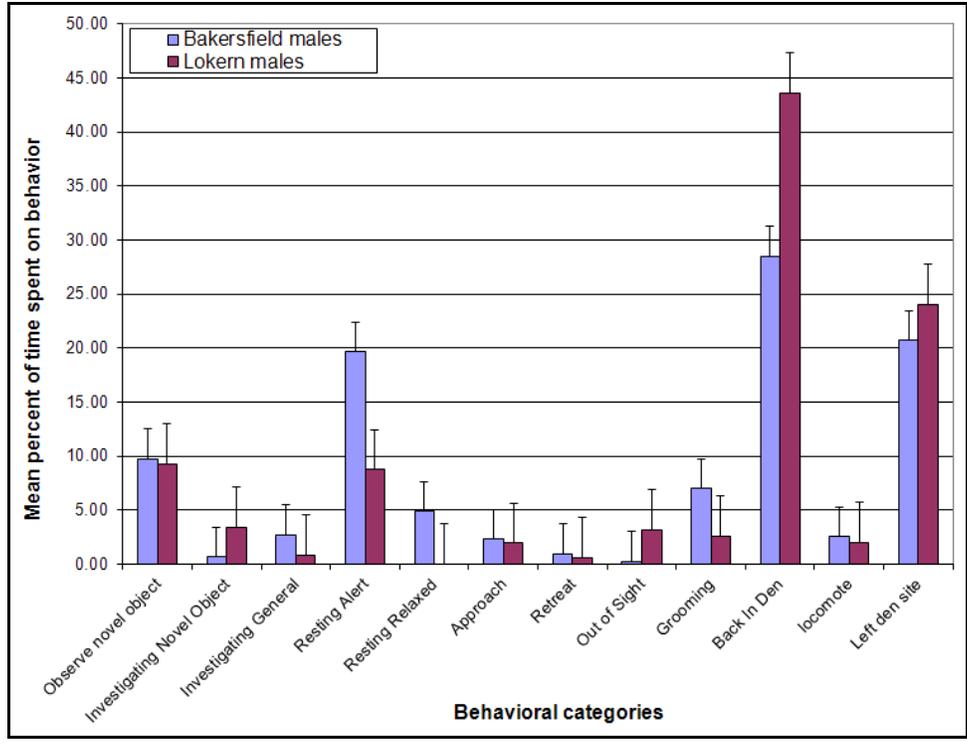


Figure 12. Percent occurrence of behavior for male foxes in response to Experiment Two novel object tests in urban and natural lands habitats.

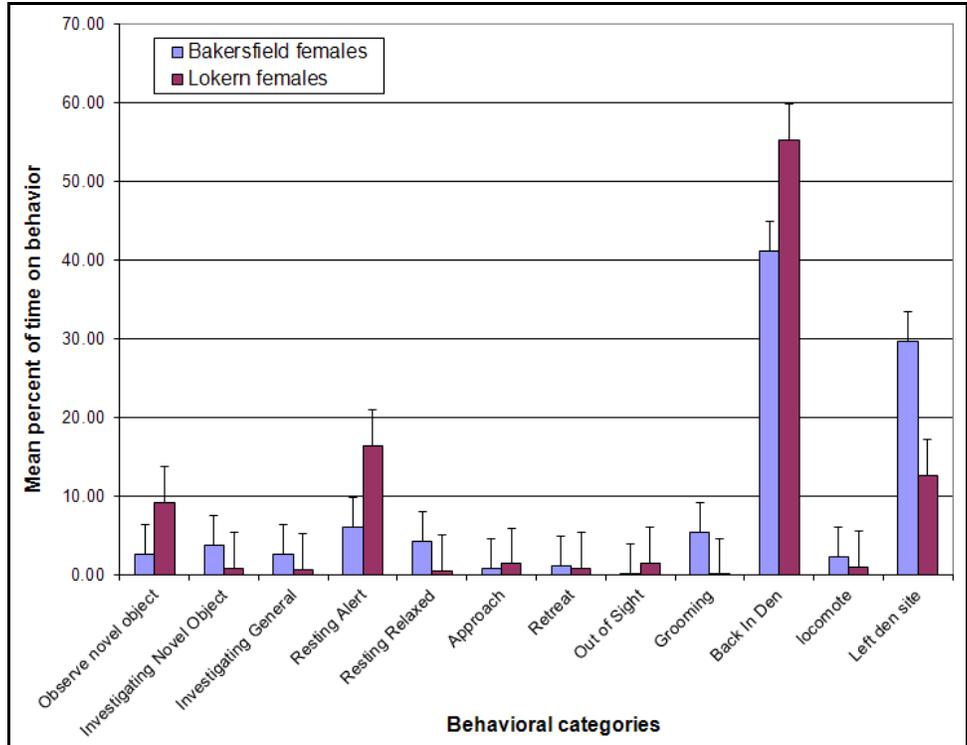


Figure 13. Percent occurrence of behavior for female foxes in response to Experiment Two novel object tests in urban and natural lands habitats.

Analyses of Experiment Two boldness scores of foxes in the two habitats demonstrated significantly higher levels of boldness for individuals in the urban environment (Table 15). Further analyses demonstrated adult foxes were significantly bolder in the urban environment than in the natural lands environment but while mean boldness scores were higher for urban yearlings, there was no significant difference in levels of boldness between the two habitats. Females were significantly bolder in the urban habitat, however, while the mean boldness score was higher for urban males than for natural lands male foxes this difference was not significant.

**Table 15: Comparison of Experiment Two boldness scores in urban vs natural lands San Joaquin kit foxes.**

Urban vs. Natural Lands	Mean Score		<i>t</i>	df	<i>P</i>
	Urban	Natural			
All foxes	-19.51	-68.25	2.85	43	<0.01
Adults	-35.60	-79.38	2.30	17	<0.05
Yearlings	-4.60	-25.86	0.64	14	NS
Males	-23.63	-44.79	0.66	11	NS
Females	-17.10	-74.95	2.60	22	<0.05

Range of boldness scores was compared between habitat types for all individuals, and by age and sex. The range of boldness scores was higher in the urban habitat for all criteria (Table 16). An index of relative size was calculated by dividing the urban by the natural lands range to determine the magnitude of difference in range between the two sites.

**Table 16: Range of Experiment Two boldness scores collected from San Joaquin kit foxes in urban and natural lands habitats.**

Population	Range				
	All Foxes	Adults	Yearlings	Males	Females
Urban	245.60	187.55	245.60	175.17	245.60
Natural Lands	126.48	103.86	126.48	126.48	113.57
Index of relative size	1.9	1.8	1.9	1.4	2.2

### Trap/Handling Behavior

Boldness scores were calculated for all foxes captured within the two habitats for the duration of the study. Analysis of the boldness scores illustrated that foxes in the urban habitat had significantly higher boldness levels than foxes within the natural lands habitat (Table 17). Furthermore, comparative analysis demonstrated that adult urban foxes were significantly bolder than adults in the natural lands, urban females overall were bolder than natural lands females, and there was no significant difference in boldness between urban and natural lands males. No difference was found between levels of boldness for yearlings or pups. As the sample size was larger for this data set, data were able to be broken down further for analysis. Adult females in the urban environment were significantly bolder, but no significant differences were found between adult males in the differing habitats, or between yearlings or pups of either sex.

**Table 17: Comparison of Trap/Handling boldness scores in urban vs natural lands San Joaquin kit foxes.**

Urban vs. Natural Lands	Mean Score		<i>t</i>	df	<i>P</i>
	Urban	Natural			
All foxes	0.19	0.06	2.52	149	0.01
Adults	0.20	0.05	1.67	63	0.03
Yearlings	-0.02	0.07	-0.57	7	NS
Pups	0.24	0.11	1.30	32	NS
Males	0.18	0.14	0.51	82	NS
Females	0.21	-0.01	2.99	57	0.004
Adult Males	0.16	0.16	0.02	32	NS
Yearling Males	-0.09	0.15	-1.22	5	NS
Pup Males	0.25	0.14	0.57	5	NS
Adult Females	0.26	-0.04	1.70	31	<0.0001
Yearling Females	0.17	0.01	0.73	1	NS
Pup Females	0.21	0.09	0.35	22	NS

Range of boldness scores were compared between habitat types for all individuals, and by age and sex. The range of boldness scores was higher in the urban habitat for all criteria (Table 18). An index of relative size was calculated by dividing the urban by the natural lands range to determine the magnitude of difference in range between the two sites.

**Table 18: Range of Trap/Handling boldness scores collected from San Joaquin kit foxes in urban and natural lands habitats.**

Population	Range					
	All Foxes	Adults	Yearlings	Pups	Males	Females
Urban	2.25	1.63	1.25	2.25	2.25	2.25
Natural Lands	1.45	1.45	0.47	0.79	0.95	1.05
Index of relative size	1.6	1.1	2.7	2.8	2.4	2.1

### Comparison of boldness scores within habitats

Data from each method of obtaining boldness scores were analyzed to determine whether individuals varied significantly in levels of boldness *within* each habitat type. Data were analyzed to determine whether variation existed between individuals, and according to sex or where applicable, age.

#### Urban Environment

Descriptive statistical analysis of boldness scores obtained from pups within the urban habitat demonstrated individual variation between boldness scores obtained across all three measures (Table 19).

**Table 19: Descriptive statistics of boldness scores obtained from San Joaquin kit foxes in the urban habitat for all three boldness measures.**

	Mean	Standard Deviation	Standard Error	Variance
<u>Experiment 1</u>				
All Observations	-40.5	128.9	26.3	16612.6
Baseline	33.9	91.2	18.6	8315.5
Novel Stimuli	-74.3	54.2	11.1	2939.1
PBS	-16.2	54.4	11.1	2954.3
PTS	-58.1	6.4	1.3	41.4
<u>Experiment 2</u>				
All foxes	-19.5	76.1	14.6	5788.9
Adults	-35.6	62.7	17.4	3928.8
Yearlings	-4.6	86.3	23.1	7452.5
Males	-23.6	62.9	19.9	3954.3
Females	-17.1	84.7	20.5	7165.9
<u>Trap/Handling</u>				
All foxes	0.19	0.40	0.04	0.16
Adults	0.20	0.31	0.05	0.09
Yearlings	-0.02	0.41	0.16	0.17
Pups	0.24	0.47	0.07	0.22
Males	0.18	0.43	0.06	0.18
Females	0.21	0.36	0.06	0.13

Analysis of variance of boldness scores of urban foxes obtained from Experiment One demonstrated that there was significant difference between boldness scores of pups in different dens ( $F_4 = 3.097$ ,  $P < 0.05$ ). Post hoc Fishers PLSD tests determined that pups from the ‘Jewetta’ den were significantly bolder than pups from the ‘Bakersfield College’ natal den ( $P < 0.005$ ) and the ‘Seven Oaks West’ natal den ( $P < 0.005$ ). In addition, there was a trend towards the Jewetta pups being bolder than pups from the ‘CSUB’ natal den ( $P = 0.07$ ). Furthermore, analysis of variation of boldness scores between male and female pups within the urban habitat determined sex had no effect on the boldness levels of pups.

The variance of boldness scores (5788.393) among urban foxes obtained from Experiment Two was significantly different from the hypothesized value of 1 ( $\chi^2 = 150498.2$ ,  $df = 26$ ,  $P < 0.0001$ ). In addition, boldness scores within each of the habitat types were compared to see if there were differences between foxes of different sex or age. Analyses using  $t$ -tests assuming unequal variances determined that there were no significant differences between adult and yearling foxes or between male and females foxes in the urban habitat.

The variance of boldness scores (0.16) among urban foxes obtained from Trap/Handling data was significantly different from the hypothesized value of 1 ( $\chi^2 = 14.3$ ,  $df = 88$ ,  $P < 0.0001$ ). In addition, boldness scores within each of the habitat types were compared to see if there were differences between foxes of different sex or age. Analyses using  $t$ -tests assuming unequal variances determined that there were no significant differences between boldness scores obtained from Trapping/Handling data for adult and yearling foxes, or between male and females foxes in the urban habitat.

## Natural lands Environment

Analysis of variance of boldness scores of natural lands foxes obtained from Experiment One (Table 20) demonstrated that there was significant difference between boldness scores of pups in different dens ( $F_2 = 4.55$ ,  $P < 0.05$ ). Post hoc Fishers PLSD test determined that pups from the ‘D680’ den were significantly bolder than pups from both the ‘D704’ natal den ( $P < 0.05$ ) and the ‘D405’ natal den ( $P < 0.05$ ). All foxes in the natural lands habitat that were trapped and sexed were male, and therefore no comparison of boldness levels of foxes of different sexes was possible for boldness data collected for Experiment One.

**Table 20: Descriptive statistics of boldness scores obtained from San Joaquin kit foxes in the natural lands habitat for all three boldness measures.**

	Mean	Standard Deviation	Standard Error	Variance
<i>Experiment 1</i>				
All Observations	-134.4	88.4	29.5	7806.8
Baseline	-23.4	78.2	26.1	6122.5
Novel Stimuli	-111.0	13.9	4.6	194.5
PBS	-51.0	13.9	4.6	194.5
PTS	-60.0	0.0	0.0	0.0
<i>Experiment 2</i>				
All foxes	-68.2	45.9	8.8	2105.9
Adults	-79.4	35.0	7.8	1225.4
Yearlings	-25.9	58.7	23.9	3440.9
Males	-44.8	61.6	8.7	1575.5
Females	-75.0	39.7	8.7	1575.5
<i>Trap/Handling</i>				
All foxes	0.060	0.251	0.030	0.063
Adults	0.049	0.278	0.042	0.078
Yearlings	0.074	0.165	0.046	0.027
Pups	0.107	0.235	0.071	0.055
Males	0.141	0.236	0.044	0.056
Females	-0.001	0.248	0.040	0.061

The variance of boldness scores (2105.9) among natural lands foxes obtained from Experiment Two was significantly different from the hypothesized value of 1 ( $\chi^2 = 54768.8$ ,  $df = 26$ ,  $P < 0.0001$ ). In addition, boldness scores within each of the habitat types were compared to see if there were differences between foxes of different sex or age. Analyses using  $t$ -tests assuming unequal variances determined that there were no significant differences in boldness scores obtained from Experiment Two between male and females foxes in the natural lands habitat, but there was a trend for higher boldness scores in yearlings than in adults ( $t_6 = -2.12$ ,  $P = 0.07$ ).

The variance of boldness scores (0.63) among urban foxes obtained from Trap/Handling data was significantly different from the hypothesized value of 1 ( $\chi^2 = 4.22$ ,  $df = 67$ ,  $P < 0.0001$ ). In addition, boldness scores within each of the habitat types were compared to see if there were differences between foxes of different sex or age. Analyses using  $t$ -tests assuming unequal variances determined that there were no significant differences

between boldness scores obtained from Trapping/Handling data for adults, yearling or pups, but that the Trapping/Handling data did show higher levels of boldness in males than in female foxes across all ages ( $t_{62} = 2.5$ ,  $P < 0.01$ ) and for adult male and female foxes ( $t_{40} = 2.5$ ,  $P < 0.05$ ).

## Boldness scores and survival

### Experiment One

Radio-telemetry data provided data on survival, and movements of individual foxes re-trapped and radio-collared following Experiment One observations. As only one Lokern fox was re-trapped and fitted with a radio-collar the remaining data analysis for Experiment One is focused on urban foxes. Survival data were categorized according to two variables: surviving until the first breeding season (classed as Dec 1); and survival through the first pup-rearing season (June 1) (Table 21). Survival data were available for two Lokern pups; the fox that was re-trapped and radio-collared, and a fox whose carcass was collected opportunistically.

Experiment One boldness scores were not significantly correlated with survival to either Dec 1 or June 1 for all foxes from the two study sites. In addition, there was no significant correlation for survival and boldness for foxes in the urban environment to either Dec 1 or June 1, but foxes that died did have higher bolder scores than foxes that survived to June 1 for both total boldness score and non-stimulus boldness scores (Figure 14).

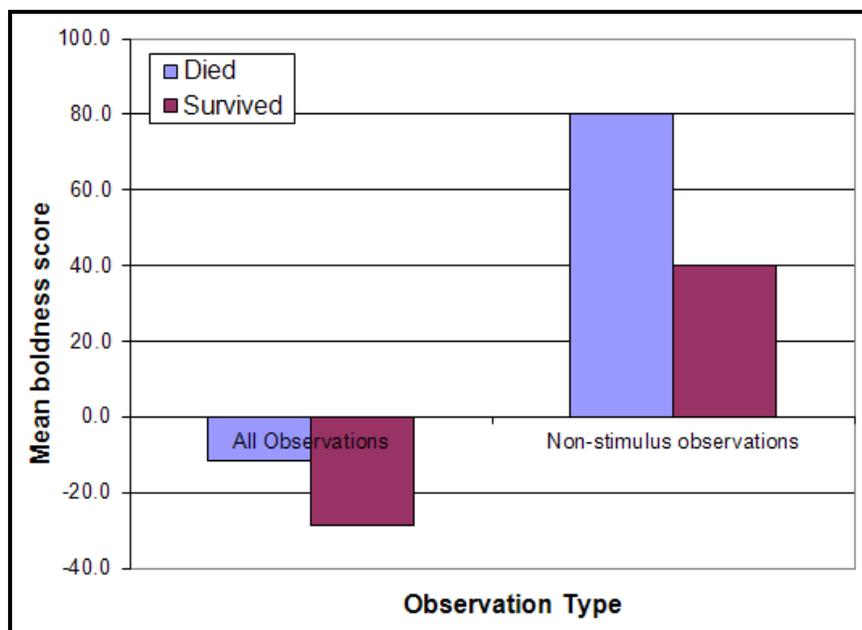


Figure 14. Experiment One boldness scores and survival of urban foxes through June 1, 2007.

Table 21: Survival data for Experiment One foxes.

Study Site	Fox ID	Sex	Mortality Date	Last date on air/heard	Last known date alive	Survived to 1st Dec	Survived to 1st June 07	
<b>Bakersfield</b>	6294	Male	2006-12-22		2006-12-22	survived	dead	
	6304	Male		not collared	2006-06-27	unknown	unknown	
	6306	Male		11/9/2006	2006-11-09	unknown	unknown	
	6069	Female		6/26/2009	2009-06-26	survived	survived	
	6240	male	2006-07-05	not collared	2006-07-05	dead	dead	
	6241	Female		6/26/2009	2009-06-26	survived	survived	
	6285	Male	2007-10-23		2007-10-23	survived	survived	
	6309	Female		6/26/2009	2009-06-26	survived	survived	
	6286	Female		6/26/2009	2009-06-26	survived	survived	
	6288	Female		4/19/2007	2007-04-19	survived	unknown	
	6242	Female		4/5/2007	2007-04-05	survived	unknown	
	6292	Female	2008-04/24		2008-04-24	survived	survived	
	6295	Male			2006-10-20	unknown	unknown	
	6245	Female	2007-01-08		2007-01-08	survived	dead	
	6298	Female	2007-08-09		2007-08-09	survived	survived	
	unmarked	unknown			not collared	2006-06-30	unknown	unknown
	7ow6290	Male			not collared	2006-06-20	unknown	unknown
	6243	Female	2007-07-13		2007-07-13	survived	survived	
	6291	Male			11/9/2006	2006-11-09	unknown	unknown
	6244	Male			6/8/2009	2009-06-08	survived	survived
	6065	Male			10/10/2007	2007-10-10	survived	survived
	6246	Female			9/29/2008	2008-09-29	survived	survived
	6287	Male			9/3/2008	2008-09-03	survived	survived
6310	Male			1/23/2007	2007-01-23	survived	unknown	
<b>Lokern</b>	6305	Male		not collared		unknown	unknown	
	6293	Male		not collared		unknown	unknown	
	6307	Male		not collared		unknown	unknown	
	6299	Male	2006-10-25		2006-10-25	dead	dead	
	6289	Male	2006-07-27		2006-07-27	dead	dead	
	unmarked	unknown			not collared	unknown	unknown	
	unmarked	unknown			not collared	unknown	unknown	
	unmarked	unknown			not collared	unknown	unknown	
	unmarked	unknown			not collared	unknown	unknown	

## Experiment Two

Foxes trapped and radio-collared for Experiment Two observations were monitored on a weekly basis for survival and movement data. Survival data were classed as dead or alive at the time they went off air. Following radio-collaring, a small number of foxes were not located during the study due to either leaving the study sites or collar malfunctions. These foxes were not included in the analysis and are listed in the table as ‘unknown’ (Table 22).

Boldness scores and survival data were analyzed for within population relationships. Unpaired *t*-test analyses indicated no relationship between boldness scores and survival data for either urban or natural lands foxes.

**Table 22: Survival data for Experiment Two.**

Study Site	Fox ID	Boldness Score	Survival Status	
Bakersfield	5678	10.0	Alive	
	6065	27.8	Unknown	
	6067	2.5	Unknown	
	6069	-100.0	Alive	
	6241	-24.1	Alive	
	6243	-100.0	Dead	
	6244	26.2	Alive	
	6246	-100.0	Alive	
	6266	-90.6	Unknown	
	6286	76.3	Alive	
	6288	133.5	Unknown	
	6292	-100.0	Dead	
	6298	145.6	Dead	
	6309	11.3	Alive	
	6351	-31.5	Alive	
	6362	-18.6	Alive	
	6365	-38.4	Alive	
	6366	87.5	Alive	
	6369	32.1	Dead	
	6392	-98.5	Unknown	
	6398	-82.5	Dead	
	6527	-26.4	Dead	
	6530	-95.5	Alive	
	6531	-53.5	Alive	
	6532	-100.0	Unknown	
	6534	75.2	Alive	
	6535	-95.5	Alive	
	Lokern	6025	-89.5	Alive
		6233	-100.0	Alive
		6273	-83.3	Alive
		6277	-100.0	Alive
		6282	-100.0	Alive
		6299	-100.0	Dead
6326		-97.3	Unknown	
6327		-95.8	Alive	
6329		-100.0	Unknown	
6331		-95.1	Unknown	
6332		-2.1	Unknown	
6374		26.5	Alive	
6375		-100.0	Alive	
6376		-100.0	Dead	
6377		-100.0	Dead	
6391		-10.2	Alive	
6395		-100.0	Alive	
6471		-100.0	Alive	
6501		-32.0	Alive	
6505		-100.0	Dead	
6506		-70.3	Alive	
6512		3.9	Alive	
6536		-26.0	Alive	
6538		-100.0	Alive	
6540		13.6	Unknown	
6541	-100.0	Alive		
6551	15.0	Alive		

**Trap/Handling**

There were a number of foxes that were trapped and collared for either Experiment One or Experiment Two in both the urban and natural lands but died before the behavioral observation could take place (Table 23). Analyses were conducted to determine whether there was a relationship between trap/handling boldness scores and survival. Only foxes that were radio-collared and therefore actively tracked were included in these analyses. Inclusion of foxes that had been assessed for trap/handling boldness and picked up as mortalities opportunistically were not included, as it was felt they may bias the results.

**Table 23: Trap/handling boldness scores and survival data for urban and natural lands foxes.**

Study Site Bakersfield	Boldness Score	Survival	Study Site Lokern	Boldness Score	Survival
5678	0.25	Alive	6025	.300	Alive
6069	0.30	Alive	6035	-.100	Dead
6241	0.25	Alive	6062	-.167	Dead
6243	0.25	Dead	6068	.700	Dead
6244	1.25	Alive	6233	.250	Alive
6245	0.50	Dead	6239	.214	Dead
6246	-0.38	Alive	6270	.125	Dead
6285	-0.50	Dead	6273	-.070	Alive
6286	0.25	Alive	6277	-.070	Alive
6292	0.50	Dead	6282	-.380	Alive
6294	-0.25	Dead	6299	.250	Dead
6298	0.50	Dead	6327	-.070	Alive
6309	0.25	Alive	6328	.300	Dead
6351	-0.10	Alive	6333	-.167	Dead
6365	0.25	Alive	6335	.300	Dead
6366	0.25	Alive	6374	.130	Alive
6369	-0.17	Dead	6375	.250	Alive
6398	0.50	Dead	6376	-.250	Dead
6530	-0.17	Alive	6377	.250	Dead
6531	-0.25	Alive	6391	-.070	Alive
6549	1.50	Dead	6395	-.380	Alive
			6400	.300	Dead
			6471	-.250	Alive
			6501	-.100	Alive
			6505	-.250	Dead
			6506	-.500	Alive
			6512	-.880	Alive
			6516	-.750	Dead
			6536	.300	Alive
			6538	-.500	Alive
			6539	.125	Dead
			6541	-.380	Alive
			6551	-.170	Alive
			6552	.125	Dead
			6553	.125	Dead
			6554	.300	Dead
			6555	-.167	Dead

An unpaired  $t$ -test revealed no significant difference in trap/handling boldness levels for foxes that died and those that survived in the urban environment. However, a strong trend was observed for higher levels of boldness in foxes that died in the natural lands environment ( $t_{33} = -2.006$ ,  $P = 0.0527$ ). This relationship is further illustrated in Figure 15.

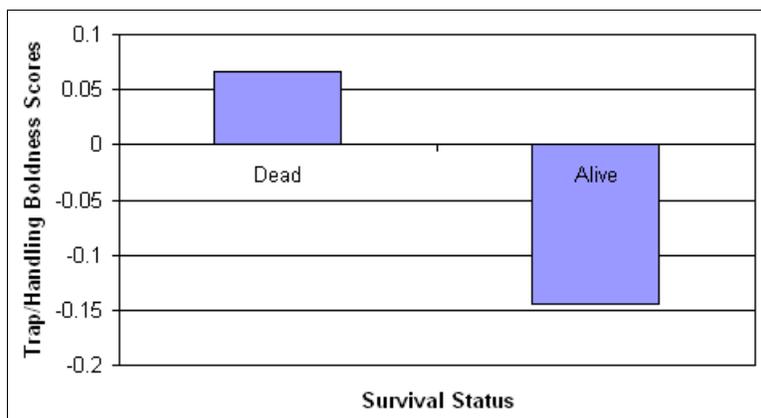


Figure 15: Trap / Handling boldness scores and survival status of natural lands foxes

## Boldness scores and reproduction

### Experiment One

Radio-collared foxes in the urban environment were tracked through at least one breeding season and assessed to determine whether they had paired and reproduced. No observed foxes reproduced in the first year, and therefore several individuals were re-collared and assessed for reproduction in 2008 and, where available, 2009 (Table 24).

Boldness scores and reproductive data for urban foxes were analyzed using non-parametric Mann Whitney  $U$ -tests. No significant correlation was found between boldness score and reproduction, or boldness score and number of pups for either 2008 or 2009.

**Table 24: Reproductive data for Experiment One urban foxes observed over three breeding seasons.**

Year	Fox ID	Sex	Paired	Reproduced	Age at reproduction	No. of pups
2007	6069	f	yes (6392)	no		
	6241	f		no		
	6285	m		no		
	6309	f		no		
	6286	f	yes (6244)	no		
	6288	f		no		
	6242	f		no		
	6292	f		no		
	6298	f		no		
	6243	f		no		
	6244	m	yes (6286)	no		
	6065	m		no		
	6246	f	poss	no		
	6287	m		no		
2008	6069	f	yes (6392)	no		0
	6241	f		no		0
	6309	f		no		0
	6286	f	yes (6244)	yes	y	8
	6292	f		no		0
	6244	m	yes (6286)	yes	y	8
	6246	f	yes	yes	y	3
	6287	m		no		0
2009	6069	f	yes (6392)	yes	a	4
	6241	f		no		0
	6309	f		no		0
	6286	f	yes (6244)	yes	a	3
	6244	m	yes (6286)	yes	a	3

## Experiment Two

Radio-collared foxes in the urban and natural lands habitats were tracked through one breeding season and assessed to determine whether they had paired and reproduced. Reproductive status is shown for both urban and natural lands foxes (Table 25).

Analysis of reproductive data determined there was no significant relationship between boldness score and reproductive success or between boldness score and the number of pups produced in the urban environment. However, analyses of reproductive success and boldness in the natural lands environment indicated a strong trend for higher boldness leading to increased reproductive success (unpaired  $t$ -test;  $t_{17} = 2.058$ ,  $P = 0.053$ ), and foxes with higher levels of boldness had significantly higher numbers of pups in their litters than foxes with lower boldness scores ( $F_{18} = 4.729$ ,  $P < 0.05$ ).

**Table 25: Reproductive data for urban and natural lands Experiment Two kit foxes.**

Bakersfield Foxes	Boldness Score	Reproduced during Exp 2	No. pups produced	Lokern Foxes	Boldness Score	Reproduced during Exp 2	No. pups produced
5678	10.0	Yes	?	6025	-89.5	Yes	3
6065	27.8	Unknown	-	6233	-100.0	No	0
6067	2.5	Unknown	-	6273	-83.3	Yes	4
6069	-100.0	Yes	4	6277	-100.0	No	0
6241	-24.1	No	0	6282	-100.0	Yes	1
6243	-100.0	No	0	6299	-100.0	No	0
6244	26.2	Yes	8	6326	-97.3	No	0
6246	-100.0	Yes	3	6327	-95.8	No	0
6266	-90.6	Unknown	-	6329	-100.0	Unknown	-
6286	76.3	Yes	8	6331	-95.1	Unknown	-
6288	133.5	No	0	6332	-2.1	Unknown	-
6292	-100.0	No	0	6374	26.5	Yes	-
6298	145.6	Unknown	-	6375	-100.0	Yes	2
6309	11.3	No	0	6376	-100.0	Unknown	-
6351	-31.5	Unknown	-	6377	-100.0	Unknown	-
6362	-18.6	Yes	?	6391	-10.2	Yes	3
6365	-38.4	Unknown	-	6395	-100.0	No	0
6366	87.5	Yes	3	6471	-100.0	No	0
6369	32.1	Unknown	-	6501	-32.0	Yes	3
6392	-98.5	Yes	4	6505	-100.0	No	0
6398	-82.5	Unknown	-	6506	-70.3	No	0
6527	-26.4	Yes	?	6512	3.9	No	0
6530	-95.5	Yes	4	6536	-26.0	Yes	2
6531	-53.5	Yes	4	6538	-100.0	Yes	3
6532	-100.0	Unknown	-	6540	13.6	Unknown	-
6534	75.2	Yes	3	6541	-100.0	No	0
6535	-95.5	Yes	?	6551	15.0	Yes	4

### Boldness scores and distance moved

The maximum distance between capture point and location point was calculated for each individual fox using ArcView GIS (version 3.2). Correlation analysis tested to see if there were significant relationships between the distance moved and the boldness scores of foxes within populations. Unpaired *t*-tests tested for significance between survival and distance moved.

### Experiment One

Distance analysis indicated that there was a trend towards a positive correlation between the maximum distance from capture point and boldness score (Kendall Rank Correlation,  $Z = 1.687$ ,  $P < 0.1$ ) for foxes in the urban habitat. No distance analysis was conducted for the natural lands as there were only data for one fox available.

## Experiment Two

Distance analysis of Experiment Two foxes did not indicate a significant relationship between boldness score and distance moved for foxes in the urban or natural lands environment.

### Trap/Handling

Distance analysis of boldness scores obtained from trap/handling data did not indicate a significant relationship between boldness score and distance moved for foxes in the urban or natural lands environment. However, a trend was observed in the urban environment towards foxes that died being those that had moved a greater distance from the capture point than those that had survived ( $t_{17} = -1.777$ ,  $P < 0.1$ ; Figure 16).

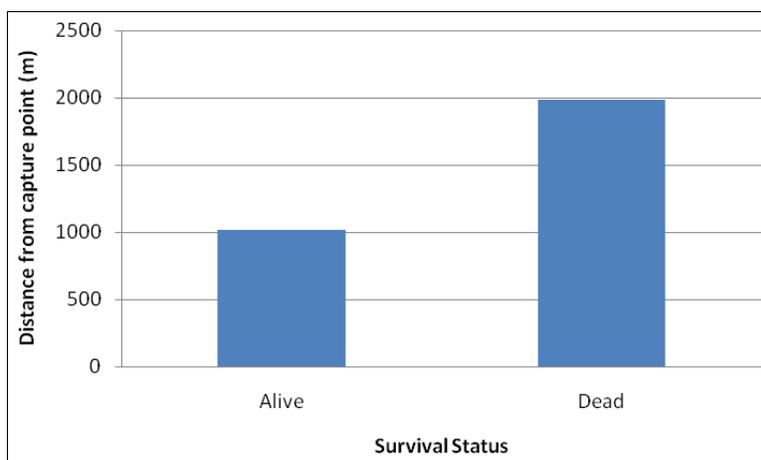


Figure 16: Distance moved from capture point and survival status for urban foxes.

### Data Summary

A summary of all data analyses is presented in Table 26 for ease of reference. Results for all three measures of boldness, survival status, reproduction and distance moved from capture point are included in the table.

**Table 26: Summary of data analyses presented throughout results section.**

Site	Analysis	Experiment 1	Experiment 2	Trap/Handling
Urban vs	All observations	*	-	-
Natural Lands	Baseline	‡	-	-
	Novel Stimuli	**	-	-
	PBS	**	-	-
	PTS	NS	-	-
	All foxes	-	**	**
	Adults	-	*	*
	Yearlings	-	NS	NS
	Pups	-	-	NS
	Males	-	NS	NS
	Females	-	*	**
	Adult males	-	-	NS
	Yearling males	-	-	NS
	Pup Males	-	-	NS
	Adult females	-	-	****
	Yearling females	-	-	NS
	Pup females	-	-	NS
Urban	Variance	*	****	****
	Male vs Female	NS	NS	NS
	Adult vs Yearling	-	NS	NS
	Adult vs Pup	-	NS	NS
	Yearling vs Pup	-	NS	NS
Natural Lands	Variance	**	****	****
	Male vs Female	-	NS	**
	Adult vs Yearling	-	‡	NS
	Adult vs Pup	-	-	NS
	Yearling vs Pup	-	-	NS
	Adult male vs adult female	-	-	*
Urban	Survival & boldness	NS	NS	NS
Natural Lands	Survival & boldness	NS	NS	‡
Urban	Boldness & reproduction	NS	NS	NS
	Boldness & No. of pups	NS	NS	NS
Natural Lands	Boldness & reproduction	-	‡	NS
	Boldness & No. of pups	-	*	NS
Urban	Boldness & distance	NS	NS	NS
Natural Lands	Boldness & distance	-	NS	NS
Urban	Survival & distance	NS	NS	‡
Natural Lands	Survival and distance	-	NS	NS

‡ =  $P < 0.01$  (trend), \* =  $P < 0.05$ , \*\* =  $P < 0.01$ , \*\*\* =  $P < 0.001$ , \*\*\*\* =  $P < 0.0001$ , - = Not applicable

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## DISCUSSION

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### *COMPARISONS OF BOLDNESS MEASURES*

While the three boldness measures did not exhibit significant correlations to one another, each of the three measures produced the same results in terms of differences in boldness between the populations, and in some aspects within the populations. Each of the boldness measures clearly detected the differences in boldness between the two populations with high levels of significance.

Despite being the most in-depth method of collecting boldness data, Experiment One provided the least information in terms of comparative analyses. This was partly due to an inability to compare boldness between populations due to the issues associated with the Lokern observations and re-trapping success, but also due to the nature of the observation which excluded the opportunity for comparison across age classes. In addition, Experiment One was an extended process that was labor-intensive. This methodology would not be suited for selection of reintroduction candidates during a reintroduction program, and its use is not recommended.

Experiment Two appeared to be better suited for use in behavioral profiling of wild foxes for several reasons, the main one being that observation subjects are trapped and collared prior to the behavioral observation taking place. This is particularly beneficial in the natural lands where foxes have larger home ranges and more den site availability, thus making it harder to target individuals when trapping. Radio-collaring prior to behavioral profiling both substantially increases the likelihood of obtaining fitness data following profiling and would allow for locating and recapturing an individual that had been selected for relocation. In addition, this test is far less labor-intensive, and therefore cheaper and faster to conduct; both considerations to take into account should an actual reintroduction effort take place in the future.

The Trap/Handling method of obtaining data was the least labor-intensive and provided the greatest sample size for this study. In addition, comparison of results obtained from Experiment Two and from the Trap/Handling boldness scores are similar for many categories of analyses. However, the data obtained were limited in some aspects, as exemplified by certain significant trends being detected in Experiment Two but not being detected using the Trap/Handling data. However, this was the first time this type of data had been collected, and further refinement of the data collection and boldness score calculation may result in a more informative measure. If refined, this method of collecting boldness data could be useful for analyzing the behavioral composition of a reintroduced population when determining what types of additional founders should be introduced.

### **Boldness and Kit Fox Reintroductions**

The aim of this study was to determine whether behavior variation in terms of shyness and boldness existed between populations of San Joaquin kit foxes in two very divergent habitats and to assess the potential for these populations to serve as source populations

for reintroduction of kit foxes to vacant habitats. Of particular interest was the potential suitability of urban kit foxes for introduction into non-urban environments.

The results of this investigation have clearly demonstrated that variation in boldness is present, both between the urban and natural land populations, and within the two populations. The urban population demonstrated significantly higher levels of boldness than the natural lands population. This difference was particularly evident among adults. However, there was no significant difference in boldness levels between populations for both yearlings and pups indicating that levels of boldness show variance within litters in both habitat types, and that selection for optimal behavioral attributes for conditions within a given habitat type are most evident in later life-stages. Boldness is considered an adaptive trait (Wilson 1998), thus similar patterns of variation between yearling and pup kit fox in differing populations demonstrates the potential for adaptation in response to environmental change.

Furthermore, boldness is subject to natural selection. Analysis of the variation of boldness levels within each of the two populations revealed a wider range of boldness scores present in the urban habitat than the natural lands habitat. Foxes in natural lands habitats may be subject to greater selection pressures, such as high predation levels and fluctuating food abundance, and this might produce the narrower range of variation in boldness observed in this population compared to that in the urban environment.

Because boldness is subject to natural selection, foxes with optimal levels of boldness relative to selection pressures present at a release site are likely to be more successful following reintroduction. Molecular genetic studies have demonstrated that high levels of genetic diversity facilitate adaptation of founder populations to selective pressures (Frankham, Ballou & Briscoe 2002), thereby increasing likelihood of establishment of a self-sustaining population. This suggests that incorporating high levels of personality variation, which has a genetic basis, in a founding population may be as important as other factors (e.g., genetics, sex, age, health) when identifying candidates for reintroduction (Watters & Meehan 2007).

When considering source populations for proposed relocation efforts, one approach is to select a population from an area with habitat conditions most similar to those on the reintroduction site. Individuals from this population are more likely to possess optimal behavioral attributes, including boldness levels, for conditions on the relocation site. However, given that conditions are unlikely to be identical between the source and release sites, an alternate approach is to strive for a founding population with a more diverse array of boldness levels. This will increase the adaptive capacity of the population and hopefully improve the probability of successful population establishment. Thus, a better approach might be to include animals from multiple populations, including urban foxes. Such a mixed population would include individuals more closely adapted to the conditions present on the reintroduction site as well as individuals with behavioral attributes that will facilitate adaptation to a new environment.

Finally, for a variety of reasons, it may not be possible to use fox populations from natural lands as source populations. In particular, the removal of a sufficient number of animals required to attempt a reintroduction might jeopardize the viability of these populations, most of which are already small in size and many of which are still declining. In fact, a rigorous population viability analysis should be conducted before

even considering using a population as a source of individuals for relocation in order to assess the potential demographic impacts of removing individuals. If using populations in natural lands appears inadvisable, then foxes from urban populations may be the only alternative. Based on this investigation, urban foxes appear to possess behavioral attributes that potentially make them suitable as relocation candidates. In particular, they appear to have a higher tendency to investigate new resources compared to non-urban foxes. This would facilitate the discovery and use of foods and dens present on a reintroduction site. Also, urban foxes appear to exhibit a wariness of potential dangers similar to that observed in non-urban foxes. Thus, the ability of relocated urban foxes to avoid dangers in natural lands, such as predators, might be similar to that of non-urban foxes that have previous experience with such dangers. Additionally, urban foxes exhibit a much broader range of behavioral attributes, particularly boldness levels, and therefore a founding population consisting of urban foxes likely would have a relatively high adaptive capacity, which would increase the probability of successful population establishment.

## **RECOMMENDATIONS**

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1. When considering a potential source population for a kit fox reintroduction effort, the behavioral attributes of the population should be considered equally with other attributes or considerations.
2. The behavioral attributes of the any potential source population should be considered with respect to conditions on the reintroduction site.
3. A population viability analysis of the potential source population should be conducted to determine whether the removal of foxes for relocation will adversely impact the viability of the population, or what number of foxes could be safely removed.
4. Behavioral assessments, particularly a measurement of boldness level, should be conducted on all candidate foxes prior to selection for inclusion in the founding population. Such assessments may require capturing, transmittering, and monitoring foxes to facilitate replicate assessments and, if selected, to facilitate recapture for relocation.
5. Foxes selected for the founding population should comprise a wide range of behavioral attributes to maximize the adaptive capacity of the population and increase the probability of a successful reintroduction.
6. Use of urban foxes may be preferable for a relocation and reintroduction because they exhibit apparently suitable behavioral attributes and because doing so would avoid any adverse impacts to populations in natural lands.
7. An experimental relocation involving urban foxes is recommended to assess relocation strategies and assess the suitability of urban foxes for reintroduction efforts.

## LITERATURE CITED

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## APPENDIX A

**Eartag Number:**..... **Date:**..... **Handler**.....

**Sex:** Male/Female

**Site:** Natural/Urban

**Age:** Adult/Pup/Yearling

### Vocalisations in trap

1. Warning bark (low pitched ‘whump’ sort of noise)

Yes                  No

2. Growl/snarl

Yes                  No

3. Scream (high pitched bark or yelp)

Yes                  No

4. Defecated in trap

Yes                  No

### Movement in Trap

1. Running backwards and forwards

Yes                  No

2. Crouching down and staying still

Yes                  No

3. Moving away from handler

Yes                  No

4. Biting at cage

Yes                  No

5. Entered into bag calmly

Yes                  No

### Vocalisations in bag

1. Warning bark

Yes                  No

2. Growl/snarl

Yes                  No

3. Scream

Yes                  No

### Behavior in Bag and during handling

1. Struggling

Yes                  No

2. Remaining still

Yes                  No

3. Defecated

Yes                  No

4. Biting at bag

Yes                  No

5. Attempt to escape (e.g. eyes uncovered)

Yes                  No