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Notes from the Editor

I am excited to announce that this spring issue of the 105th volume is the debut of the *California Fish and Game* Journal's new subtitle: "Journal for the Conservation and Management of California's Species and Ecosystems." This new subtitle more effectively highlights the purpose of CFG, which is to provide information on the biology, ecology, conservation, and management of species native to California, including fish and plants, not just wildlife. Over the past decades, as prominent environmental and conservation issues in our state have changed, the Department's roles have changed in response. This was reflected by the Department's name change in 2012 from "Fish and Game" to "Fish and Wildlife" to more appropriately encompass all of the species we protect; the Journal's subtitle has been updated for a similar purpose.

We have a new Associate Editor on our team: Dr. Robert Sullivan. Dr. Sullivan completed his first three degrees at Humboldt State University: a B.S. in Biology, an M.S. in Biology, and an M.S. in Wildlife and Natural Resource Management. He then did his PhD in Biology at the University of New Mexico and post-doctoral research as the Curator of Mammals at the New Mexico Museum of Natural History. Dr. Sullivan has had a diverse research background in everything from salmonids to herpetofauna to rodents and marine mammals, among others. He began his career with CDFW in 2007 as an Environmental Scientist in the Timberland Planning Program, and in 2009 moved to the Wildlife Program at the North Coast Wildlife Area Complex, which he continues today. We are excited for Dr. Sullivan to join our team of Associate Editors!

Remember that we are working on three special issue this year: cannabis, fire, and human recreation and their impacts on fish and wildlife resources in the state. Please pass the word along to those you know who do research on these topics. If you would like to find out more about our Special Issues, please see our webpage: https://www.wildlife.ca.gov/Publications/Journal/Special-Issues.

Ange Darnell Baker, PhD
Editor-in-Chief
California Fish and Game

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Length-weight relationship and condition factor of butterfish *Peprilus medius* (Peters, 1869) in the southeast Gulf of California

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Butterfish *Peprilus medius* (Peters 1869), a species in the Stromateidae family (Perciformes, Stromateoidei), are distributed in the Pacific Ocean from Mexico to Peru and the Galapagos Islands (Fischer et al. 1995). This species lives in shoals and inhabits benthopelagic habitats, including soft bottoms (sand and mud) and reef environments, to a depth of approximately 200 m (Rojas and Zapata 2006; Ulloa et al. 2007; Salas and Alvarado 2008; Anónimo 2009; Chirichigno and Cornejo 2001 in Inga et al. 2008; Herrera et al. 2010; Domínguez-Domínguez et al. 2014).

Throughout its range, the fishery of *P. medius* benefits the socioeconomic systems of fishing communities. The resource is primarily harvested by artisanal fisheries, a commercial seine fishing fleet, and as bycatch in the shrimp trawl fishery (Inga et al. 2008; Martínez-Muñoz 2012; Reyes-Lucas and Reyes-Vega 2015). There is minimal fishery regulation and population monitoring in most locations where harvest occurs. In recent years, the commercial importance of *P. medius* has grown due to its quality, low price and availability throughout the year. *P. medius* is marketed for fresh human consumption in Peru (Inga et al. 2008), Mexico (Martínez-Muñoz 2012) and Colombia (Moreira-Arcentales 2012), and exported frozen from Peru (Inga et al. 2008). Also, *P. medius* is used as a feed component for fish aquaculture (e.g., Prado 2010, Martínez-Lagos 2003). In Mexico, *P. medius* is also used as bait, being highly important to other artisanal fisheries.

Currently, Peru is the only country that has established a research and monitoring plan to improve management of *P. medius*, so the biological knowledge of *P. medius* is scarce (Rey-Rey 2007). Due to the growing popularity of this species, information is essential to support timely decisions and effective fishery management (Torres-Lara 1991). Additionally, biological information may be useful because the species is a potential bioindicator (Sielfeld et al. 2010) of warm-anomalous environmental conditions, and it could provide information to support timely decisions for mitigation and prevention purposes for ecosystems management.

To contribute to the biological information of *P. medius*, we present unpublished estimates of the length-weight relationship (LWR) and condition factor of *P. medius* from the southeastern Gulf of California, Mexico. We also review the available information about LWR of *P. medius* along its distribution range. Variations of LWR parameters of *P. medius* and condition factor were explored for potential differences between seasons, growth stanzas and locations (Froese 2006). Results of the LWR analysis are a valuable tool to convert length observations into weight estimates, which are necessary for biomass estimates obtained from analytical models. Results are also useful in fitting the von Bertalanffy growth equation in weight and in calculating indicators of the fish's physiological condition (Jones et al. 1999, Frota et al. 2004, Froese 1998, Froese 2006). Such indicators are also crucial for determining the health of a population (Jones et al. 1999). The *b* value of LWR also can be used as indicator of body shape or condition at the time of sampling (Froese 2006).

For this study, monthly samplings on artisanal landings of *P. medius* from southeast of Gulf of California, Mexico, were conducted from December 2011 to October 2012 (2011-2012 season), and September 2014 to November 2015 (2014-2015 season). The artisanal fishery catches *P. medius* with gillnet (3-5 in. mesh size) along the coast (Figure 1). The total weight (g, W) and total length (cm, L) of individuals from a random sample of captured fish were measured. Sex and gonadal development were defined by morphochromatic inspection of gonads according to Maldonado-Amparo et al. (2017), in order to separate juvenile from adult organisms, with the aim of assessing potential differences in LWR parameters between sexes and different ontogenetic stages of *P. medius.*

The length-weight relationship (LWR) was calculated by using power function $W \sim a\ L^b$, where a and b parameters were estimated by linearization of power function as $\log W = \log a + (\log L)\ b$ (Ricker 1975; Froese 1998; Froese et al. 2011). Differences in intraspecific LWR can be attributed to variations in ecological factors and differences between ontogenetic stages (Froese, 2006). With this in mind, we estimated LWR separately for overall data from both sampling seasons (AS-all) and separate sampling seasons for the following groups: all sample (AS), adult males (AM, sexual mature males), adult females (AF, sexual mature females) and juveniles (J, females and males sexually immature). Additionally, a throughout review of the published literature of *P. medius* LWR parameters was conducted, in order to assess potential latitudinal variations of this species LWR.

The relative condition factor (K_n) was estimated using the equation proposed by Le Cren (1951): $K_n = W/W_e$, where W_e is the total weight calculate by LWR for AS-all, and it was used to explore and compare groups condition within the overall sample (Froese 2006). The confidence limits (CL) of parameters (a and b) and K_n mean (by group) were estimated as parameter± [1.96*(standard error of parameter)] (Sokal and Rohlf 2009). The confidence

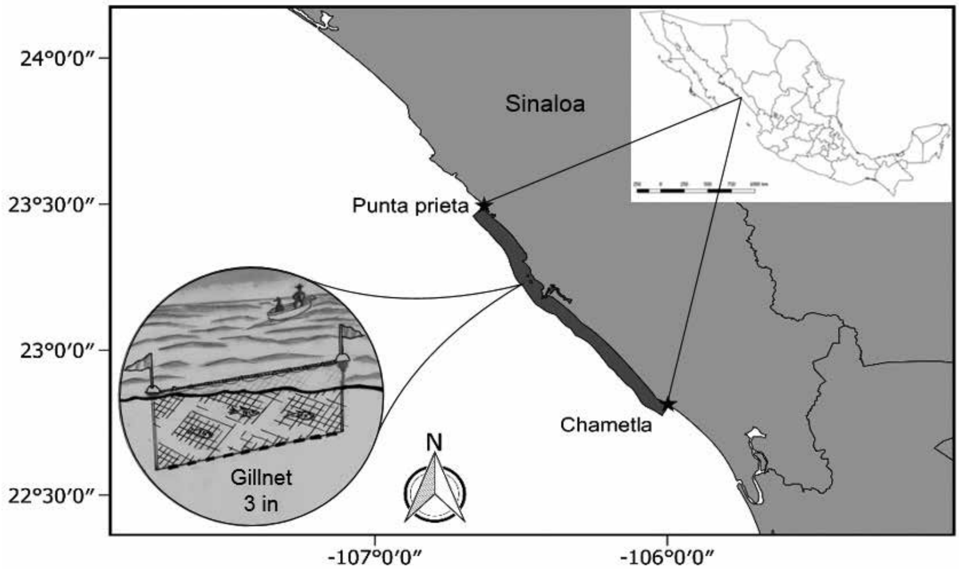

Figure 1.—Fishing area and fishing gear of *Peprilus medius* in the southeast Gulf of California.

limits (CL, 95%) were used to compare *b* with *b* = 3 to define: negative-allometric growth if b<3, isometric growth if b=3 and positive-allometric growth if b>3 (Froese 2006). K_n between groups were compared by Analysis of Variance (ANOVA) and a post-hoc Duncan test (Sokal and Rohlf 2009).

The overall sample group (AS-all) was integrated by 922 specimens with sizes that ranged from 15 to 30 cm of *L* and 50 to 312 g of *W* (Table 1). All adjusted models of LWR explained more than 65% of variance (Table 1) and the lower determination coefficients are due a high natural variation of *W* in each *L* (Figure 2).

Most of the upper (95%) values of the CLs for the *b* values were less than 3, indicating there was a negative allometric growth for all groups, except juveniles during the 2014-2015 season (Table 1, Figure 3). For all groups, the *b* values were higher during the 2014-2015 season (Figure 3). All *b* values presented an inverse trend with respect to parameter *a* values (Figure 3). All LWR presented *a* values less than 0.1.

Available LWR of *P. medius* indicate that this species presents the three growth types (negative-allometric, isometric and positive-allometric). Our work is the first report of isometric growth (only a set of juveniles; Table 1). This variation can be the result of different ecological and biological processes, such as growth stanzas (ontogenetic stages), and intraspecific differences in gonadal development status (Schneider et al. 2000, Frota et al. 2004, Froese 2006). Most analyzed groups of *P. medius* presented negative-allometric growth (*b*<3) in the southeastern Gulf of California (Table 1). This indicates that large specimens change their body shape to become more elongated (Froese 2006) in our study area, indistinctly in most of the different ontogenetic stages or seasons.

Other stromatids with negative-allometric growth reports are *P. paru* (Passos et al. 2012, Segura et al. 2012) and *P. snyderi* (Bautista-Romero et al. 2012). The positive-allometric growth was also reported for *P. simillimus* and *P. snyderi* (Rodríguez-Romero et al. 2009).

The relative condition factor (K_n) was significantly different between groups

Table 1.—Length-weight relationships of *Peprilus medius*.

Category	Period	n	Interval LT (cm)	Interval WT (g)	LWR parameters a (CL 95%)	LWR parameters b (CL 95%)	Gt	R^2	Place	Author
All sample	D 2011 to O 2012 y S 2014 to N 2015	922	15-30	50-312	0.0261 (0.0200-0.0322)	2.7607 (2.6854-2.8360)	-A	0.8381	SE Gulf of California	This work
All sample	D 2011 to O 2012	591	16-30	50-309	0.0082 (0.0287-0.0609)	2.5775 (2.4587-2.6963)	-A	0.7443	SE Gulf of California	This work
Adult males	D 2011 to O 2012	214	17.5-26.5	61-208	0.0566 (0.0172-0.0960)	2.4910 (2.2617-2.7203)	-A	0.6536	SE Gulf of California	This work
Adult females	D 2011 to O 2012	246	17-27.5	58-309	0.0660 (0.0290-0.1030)	2.4609 (2.2767-2.6451)	-A	0.7193	SE Gulf of California	This work
Juveniles	D 2011 to O 2012	41	16-22.5	50-141	0.0463 (0.0083-0.0843)	2.5309 (2.2514-2.8104)	-A	0.8813	SE Gulf of California	This work
All sample	S 2014 to N 2015	331	15-28.8	55-312	0.0265 (0.0175-0.0355)	2.7637 (2.6551-2.8723)	-A	0.9006	SE Gulf of California	This work
Adult males	S 2014 to N 2015	174	17.2-28.7	71-270	0.0211 (0.0115-0.0307)	2.8249 (2.6785-2.9713)	-A	0.9000	SE Gulf of California	This work
Adult females	S 2014 to N 2015	143	18-28.8	73-312	0.0322 (0.0134-0.0510)	2.7161 (2.5301-2.9021)	-A	0.8679	SE Gulf of California	This work
Juveniles	S 2014 to N 2015	13	15-24.18	55-180	0.0407 (-0.0122-0.0936)	2.6386 (2.1888-3.0884)	I	0.8959	SE Gulf of California	This work
All sample	D 2009	203	5-18	NR	0.0315	2.6771	NR	0.9700	Salvador	Anonymous, 2009
All sample	O 2004 to M 2006	18	12-18.7	45-188	0.0250	3.0730 (2.6530-3.4930)	+A	0.9370	BCS, México	Rodríguez-Romero et al., 2009
All sample	J_N 2005 to J_L 2006	624	3.5-29.6	1-267	0.0080	3.0890 (3.0645-3.1135)	+A	0.8820	SE Gulf of California	Nieto-Navarro et al. 2010
All sample	1995 to 1996	541	6.5-27.5	NR	0.1024	2.2962	NR	0.8228	Colombia	Zapata-Padilla, 2011

J_N, June; J_L, July; D, December; O, October; S, September; N, November; M, March; n, data number; LT, total length; WT, total weight; LWR, length-weight relationship; a, intercept; b, slope; GT, growth type; I, isometric growth; -A, negative allometric growth; +A, positive allometric growth; CL, confidence limit; R^2, coefficient of determination; SE, southwest; BCS, Baja California Sur state; NR, data not reported.

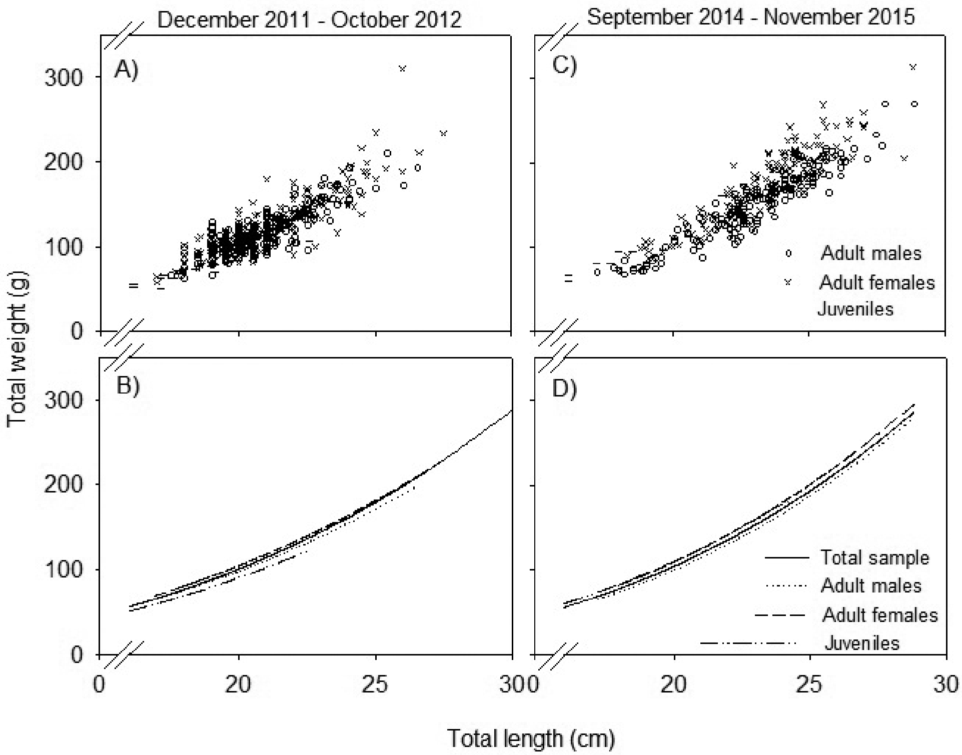

FIGURE 2.—Observed data and LWR regression fit for December 2011 – October 2012 (A and B, respectively) and for September 2014 – November 2015 (C and D, respectively).

($F_{(3,921)}$=16.8536, P<0.001). The general trend was that *P. medius* had better condition in 2014-2015 season with respect to 2011-2012 season in the southeastern of Gulf of California (Figure 3). The major difference in condition factor was between juveniles, followed by the differences between adult females. The condition factor of adult males was similar between seasons (Figure 3). Changes in fish condition can be the result of good versus poor feeding success, disease or reproduction (Helfman et al. 2009). Juveniles do not invest energy in reproduction; therefore, they could enhance notably their condition in favorable ecological context. Meanwhile adults use part of their energy in gonadic development, thus possibly their condition improvement is minor (as in females) or not significant (as in males).

Reproductive activity was also more intense during 2014-2015 season. Most specimens' gonads were mature or in partial spawning phase (71.71%), and only 4.18% were immature, 10.93% in development, 4.5% post-spawning and 8.68% in resting phase. Conversely, in 2011-2012 season, the gonads were predominantly immature (45.53%) or in development process (47.86%), and a minor percentage presented as mature or partial spawning appearance (6.61%).

According to the Oceanic Niño Index (NOAA 2018), warm oceanic environmental conditions occurred during the 2014-2015 season, which is consistent with the higher K_n and b values and more intense reproductive activity observed in that season. Conversely, during 2011-2012 season the Pacific Ocean was slightly cooler-than-normal. This suggests

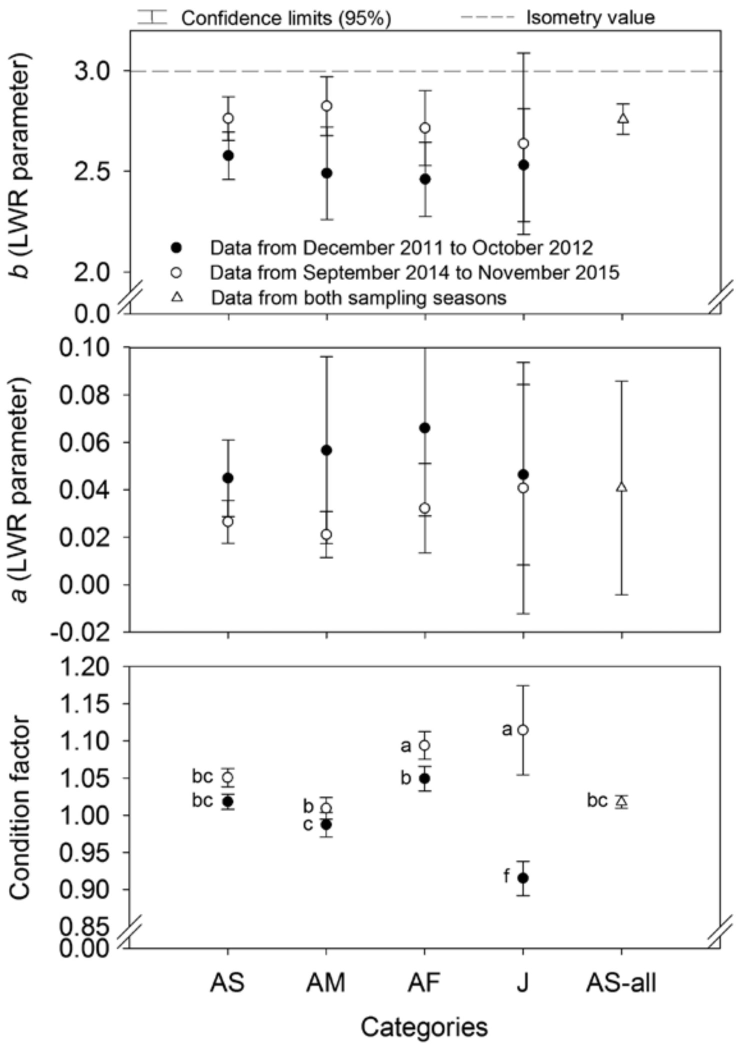

FIGURE 3.—Estimation and confidence interval of Length-weight relationships parameters and relative condition factor of *Peprilus medius*.

that warm-anomalous environmental phenomenons are favorable for *P. medius*, and in consequence, the population could enhance its condition and its reproductive activity. Additionally, it has been reported that *P. medius* can also change its distribution patterns during warm periods, and can be found outside its typical distribution range (Sielfeld et al. 2010).

The *b* values presented positive latitudinal arrangement being higher at subtropical areas with respect to tropical zones (Figure 4). This suggests that Bergmann's rule could occur in *P. medius* if higher *b* values are considered as indicators of better condition, based on the similar trends between b values and K_n (Figure 3). The Bergmann's rule postulates that body mass increases towards higher latitudes (Meiri 2011). To strengthen this idea it is necessary to assess *b* coefficient in more places over the entire latitudinal distribution range of *P. medius*.

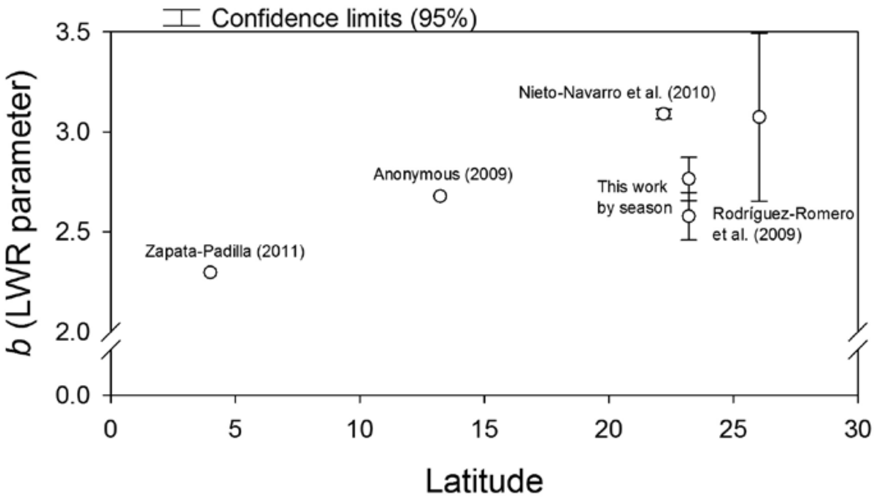

FIGURE 4.—Latitudinal variation of previously published data and results of this study for values of the *b* parameter in the LWR of *Peprilus medius.*

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The Rio Grande leopard frog (*Lithobates berlandieri*) and other introduced and native riparian herpetofauna of the Coachella Valley, Riverside County, California

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The Coachella Valley of southeastern California has gradually developed a riparian herpetofauna since the conversion of desert land to agricultural and urban landscapes and the formation of the Salton Sea in the early 1900s. Most of these species originally spread from the Colorado River to the Imperial Valley through natural channel shifts long ago, and more recently through the All-American Canal. Some of these riparian species then moved northward into the Coachella Valley of Riverside County, situated at the north end of the Salton Sea, namely, Woodhouse's Toad (*Anaxyrus woodhousii*), Great Plains Toad (*Anaxyrus cognatus*), Rio Grande Leopard Frog (*Lithobates berlandieri*), Spiny Softshell (*Apalone spinifera*), and Checkered Gartersnake (*Thamnophis marcianus*). The Red-eared Slider (*Trachemys scripta elegans*) was introduced from captivity, and the American Bullfrog (*Lithobates catesbeianus*) may have arrived from multiple sources. The most recent immigrant is the Rio Grande Leopard Frog. This species has colonized the agricultural southern part of the Coachella Valley and has begun to move into the urbanized northern portion. This frog has the potential to negatively affect native anurans if it spreads further. The current distribution, timing of introduction, and suspected origins of all riparian herpetofauna in the Coachella Valley are discussed.

Key words: amphibians, Coachella Valley, herpetofauna, introduced species, reptiles, Rio Grande Leopard Frog

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This article documents the spread of the Rio Grande Leopard Frog (*Lithobates berlandieri*) and other riparian species into the Coachella Valley of southeastern California. Riparian reptiles and amphibians of various origins have over time colonized the artificial wetlands now scattered across the previously arid Coachella Valley. The established assemblage now consists of the following species: American Bullfrog (*Lithobates catesebeianus*), Rio Grande Leopard Frog, Great Plains Toad (*Anaxyrus cognatus*), Woodhouse's Toad (*Anaxyrus woodhousii*), Spiny Softshell (*Apalone spinifera*), Red-eared Slider (*Trachemys scripta elegans*), and Checkered Gartersnake (*Thamnophis marcianus*).

The Coachella Valley is situated at the northwestern edge of the Colorado Desert and is separated from the coastal plain of California by the San Bernardino Mountains to the north, the Santa Rosa and San Jacinto Mountains to the west, and San Gorgonio Pass between those ranges (Figures 1 and 2). Its southern border is the north end of the Salton Sea. Mean annual precipitation varies from 81 mm in Mecca to 88 mm in Indio and 123 mm in Palm Springs (USCD 2018). Natural wetlands consist of a small number of valley-edge palm oases and springs surrounded by desert and supported by artesian water welling up through earthquake faults or perennial streams flowing a short distance onto the valley from nearby mountains. Thousand Palms Oasis, Dos Palmas Oasis, and the Whitewater River are examples. In the early 1900s, there were still a handful of artesian springs on the valley floor. Three such springs near Indio, Thermal, and Mecca were searched for by California Department of Fish and Wildlife biologists, who concluded that they no longer exist and were probably capped and appropriated for irrigation (Black 1980). The Coachella Valley floor is now mostly irrigated and developed, with both agricultural and urban land uses. Agriculture began in the late 1800s in the upper Coachella Valley once settlers discovered the abundant artesian water, aided by the arrival of the railroad in 1876, facilitating crop transport (Downs 2015). The cities of Palm Springs, Palm Desert, and others gradually replaced the farmland. Country clubs and golf resorts abound, most of which have ponds that are potential habitat for riparian reptiles and amphibians.

The lower Coachella Valley is mainly agricultural and is similar to a swath of canal-irrigated farmland from the Imperial Valley down into Mexico, and from there east to the Colorado River (Figure 1). The Coachella Valley is isolated from the Imperial Valley by the hypersaline Salton Sea and along its sides by about 35 km of barren desert, though a small number of springs have potential use as stepping stones for riparian species.

The Coachella Canal, constructed in the 1940s, bridges this gap and brings water to the Coachella Valley as an extension of the All-American Canal from the Colorado River. A network of feeder lines, holding ponds, and open ditches take water to and from the farms, eventually ending up in the low-lying Salton Sea. Most of the ponds, canals, and ditches develop algal mats, cattails, and other adventitious plants, but are periodically cleaned out to expedite water movement. Additional riparian habitat is found in fish farms, duck-hunting ponds, and poorly drained areas in agricultural fields and groves.

The Whitewater River is the main waterway through the Coachella Valley. The upper reach of the Whitewater River where it exits the mountains is characterized by swift, cold water, with little vegetation along its banks. It flows through the desert of the far upper Coachella Valley, where it terminates at a series of infiltration basins that supplement the aquifer for desert cities. Rainfall is normally so low that there is no surface flow from these basins for approximately 9 km where the dry channel reaches the edge of Palm Springs. From there, the Whitewater River is a mostly a dry flood control channel with small perennial seeps from urban and golf course runoff. These urban seeps can function as refugia for herpetofaunal survival and/or breeding. Surface flows return in Indio as discharge from a wastewater treatment plant. The Salton Sea itself is too saline to support any herpetofauna, but small marshes at the mouth of the Whitewater River and other drain outfalls into the Sea constitute important habitat for introduced species, as well as for the endangered native desert pupfish (*Cyprinodon macularius*) (Keeney 2012).

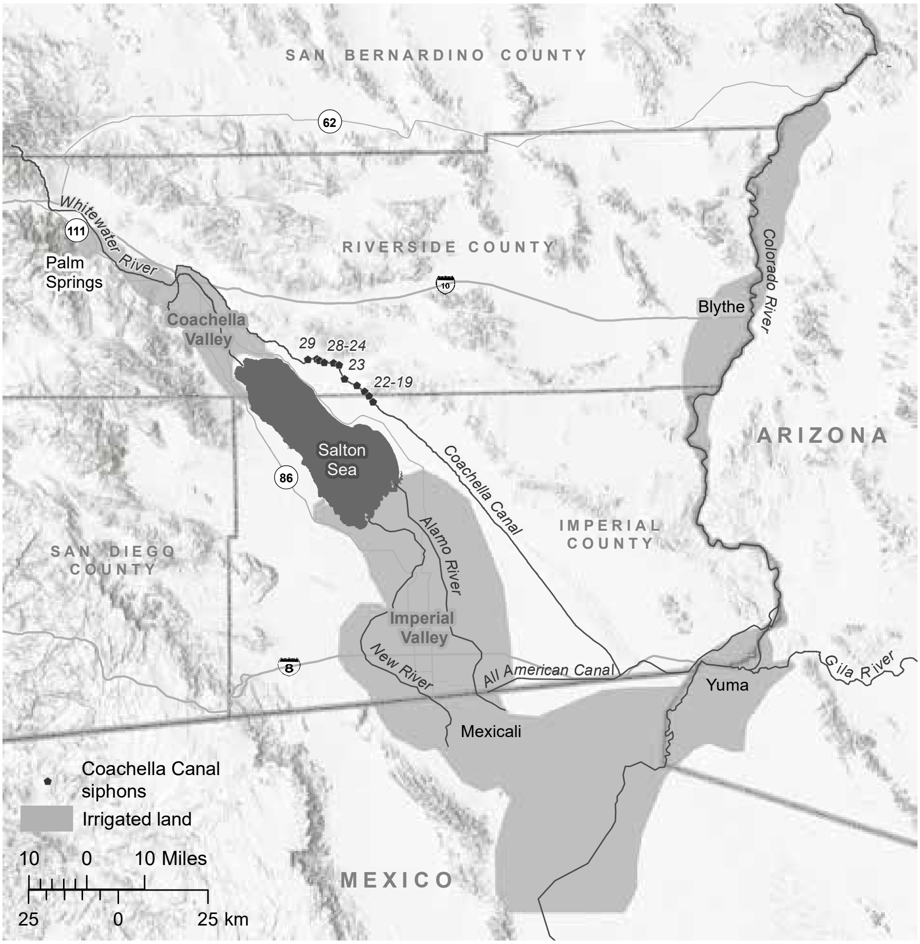

Figure 1.—Area overview.

While most of the introduced herpetofauna do not pose an ecological threat, the most recent immigrant to the Coachella Valley, the Rio Grande Leopard Frog (RGLF) is of concern. This native of Texas and northern Mexico was accidentally introduced to the lower Colorado River near Yuma, Arizona, first detected in 1981 (Platz et al. 1990). For further history on the early spread of RGLF and possible interactions with other species of leopard frogs, see Discussion section and Hillis (1981), Kocher and Sage (1986), Jennings and Fuller (2004), Sredl (2005), Rorabaugh and Servoss (2006), and Thomson et al. (2016).

Materials and Methods

Survey methods.—From 2005-2018, we conducted opportunistic searches by car and on foot throughout the Coachella Valley for riparian herpetofauna, in both daylight and nighttime, particularly during rainy periods. For RGLF, we conducted systematic searches of Dos Palmas Preserve (site R5) in 2005, 2008, and 2018. The survey area on all three years consisted of all the ponds and ditches from the headquarters northward to the Coachella Canal, and the small set of ponds south of the headquarters, totaling about 160,000 m^2. We spent three to four hours starting at dusk slowly walking the berms between all the ponds, spotlighting the banks, algal mats, and edges of cattail stands, counting all individuals seen and heard. To document the potential role of the Coachella Canal in the spread of RGLF into Riverside County, we visually examined the Canal from Siphon 19 in Imperial County across the county line up to Siphon 26, including the wildlife drinkers adjacent to each siphon (Figure 1). To monitor the potential spread of RGLF into the upper Coachella Valley via the Whitewater River, we established reference locations in the river northwest of their documented range. These locations are urban seeps in La Quinta (sites A8-11), Palm Desert (sites A12-13), and Indian Wells (site A19). After a five-year hiatus, due in part to low rainfall that would inhibit overland dispersal, we resumed targeted searches in 2017 to determine if RGLF had moved upstream. Turtles were visually located with binoculars during daylight hours, both opportunistically and by focused searches of the Coachella Canal, golf course ponds, and other wetlands.

Museum and database searches.—We used VertNet.org to search online museum databases for all target species in Riverside and Imperial counties. Our museum search located pertinent specimens from the California Academy of Sciences (CAS), the Los Angeles County Museum (LACM), the Museum of Vertebrate Zoology at Berkeley, California (MVZ), the National Museum of Natural History (NMNH), the San Bernardino County Museum (SBCM), and the San Diego Natural History Museum (SDNHM). We also queried the California Natural Diversity Database (CNDDB) and iNaturalist.org. We deposited voucher specimens at LACM (see Appendix A).

Taxonomy.—Since taxonomic opinions are beyond the scope of this paper, we follow the taxonomy of Scientific and Standard English Names of Amphibians and Reptiles of North America, Eighth Edition (Crother 2017).

Results

Rio Grande Leopard Frog.—The account below chronicles the spread of this species northwestward from the Coachella Canal near the Imperial County border. Refer to Figures 1 and 2 for locations.

In 2005, leopard frogs were found at shallow pools emanating from leaks in the Coachella Canal, 14 km below the Riverside County border in Imperial County, (C. McGaugh, Amec Foster Wheeler Inc., personal communication). Ranid tadpoles were in one such pool on 16 January 2006, and a subadult frog was photographed and confirmed as RGLF by J. Rorabaugh, U.S. Fish and Wildlife Service.

RGLF was first detected in Riverside County in 2005 at ponds at Dos Palmas Preserve adjacent to the Coachella Canal (N. Moorhatch, Amec Foster Wheeler, Inc., personal communication). Table 1 illustrates the initial small numbers of RGLF at the preserve in 2005 followed thereafter by population increases. The RGLF in 2005 were concentrated in one pond, and in subsequent years spread throughout the preserve.

In January 2007, B. Claypool found two RGLF infected with chytrid fungus near Mecca (Lovich et al. 2008), providing the first published record of this frog in Riverside County.

In April 2008, we found three of the Coachella Canal wildlife drinkers had been colonized by RGLF, at about 1, 3, and 7 km north of the Imperial County line at Siphons

Table 1.—Numbers of Rio Grande leopard frog (RGLF) and American Bullfrog (AMBU) at Dos Palmas Preserve, site R5. Frogs of all ages were counted, both seen and heard.

Date	RGLF	AMBU	Unidentified
10 May 2005	6	44	8
24 July 2008	61	66	11
11 July 2018	91	32	8

21, 22, and 23 (Figure 1). We captured an individual for species verification at Siphon 23 (site R4) and photographed a RGLF egg mass in the drinker at Siphon 21. We also observed RGLF in 2008 on the banks of the canal from Siphon 19 in Imperial County up to Siphon 26 in Riverside County. An irrigation pond next to the canal in Mecca had at least a dozen RGLF, including juveniles (site R12), while ponds farther away had no frogs.

From 2009-2011, another set of RGLF occurrences were documented about 8 km southwest of Mecca in irrigation ponds and a fish farm near Oasis.

In 2012, searches were resumed following late summer thunderstorms, and a third area was found to have been colonized by RGLF, this one in Indio, about 22 km north of Mecca, adjacent to the Coachella Canal. In August 2012, we found multiple adult and juvenile RGLF at golf course ponds and overflow seeps 100-300 m from the Coachella Canal (sites R13-R16) as well as in the Coachella Canal itself.

In 2012, we surveyed seeps at and around the Coachella Canal undercrossing at the Whitewater River in Indio and documented RGLF in the river, including several individuals dispersing into the river from a pond in a plant nursery (Sites R17-19). Surveys both upstream and downstream from that site were negative for RGLF (sites A4, SO14, A15, and A17).

After several years of low rainfall, we resumed searches in 2017 after substantial rains and found RGLF still had apparently not moved up the Whitewater River into the resort communities but had moved into the lower Whitewater River (site R28) and drain outfalls at the edge of the Salton Sea (R26 and 27).

In summary, from 2005-2018, RGLF moved into the Coachella Valley, likely via the Coachella Canal, dispersed widely and reproduced successfully. We found RGLF broadly distributed in the lower agricultural part of the Coachella Valley, as well as locally in Indio in areas adjacent to the Coachella Canal. They do not appear to have colonized all available habitat within their current range.

American Bullfrog.—Currently, bullfrogs occur throughout both the upper and lower Coachella Valley, and can be quite abundant locally, such as in the Whitewater River, fish farms, and Dos Palmas Oasis (Figure 2). Bullfrogs were found in the unlined part of the Coachella Canal when it was drained in 2007 (J. Crayon, California Dept. Fish and Wildlife, personal communication). Bullfrogs have spread farther northwest into the upper Coachella Valley than RGLF, where most of our bullfrog sightings are from small seeps and wet portions of the Whitewater River as it winds through golf resorts and urban areas.

Great Plains Toad.—At present, this species is most common in the agricultural southern Coachella Valley (Figure 3). Breeding congregations have been found primarily in date palm groves but also at the edges of field crops, both of which are flood irrigated. The permanent irrigation ponds do not appear to be attractive to Great Plains Toad, though one basin that had been dry for some time and started to grow grassy vegetation attracted a chorus after it was refilled (site G2). Currently, the upper Coachella Valley supports Great

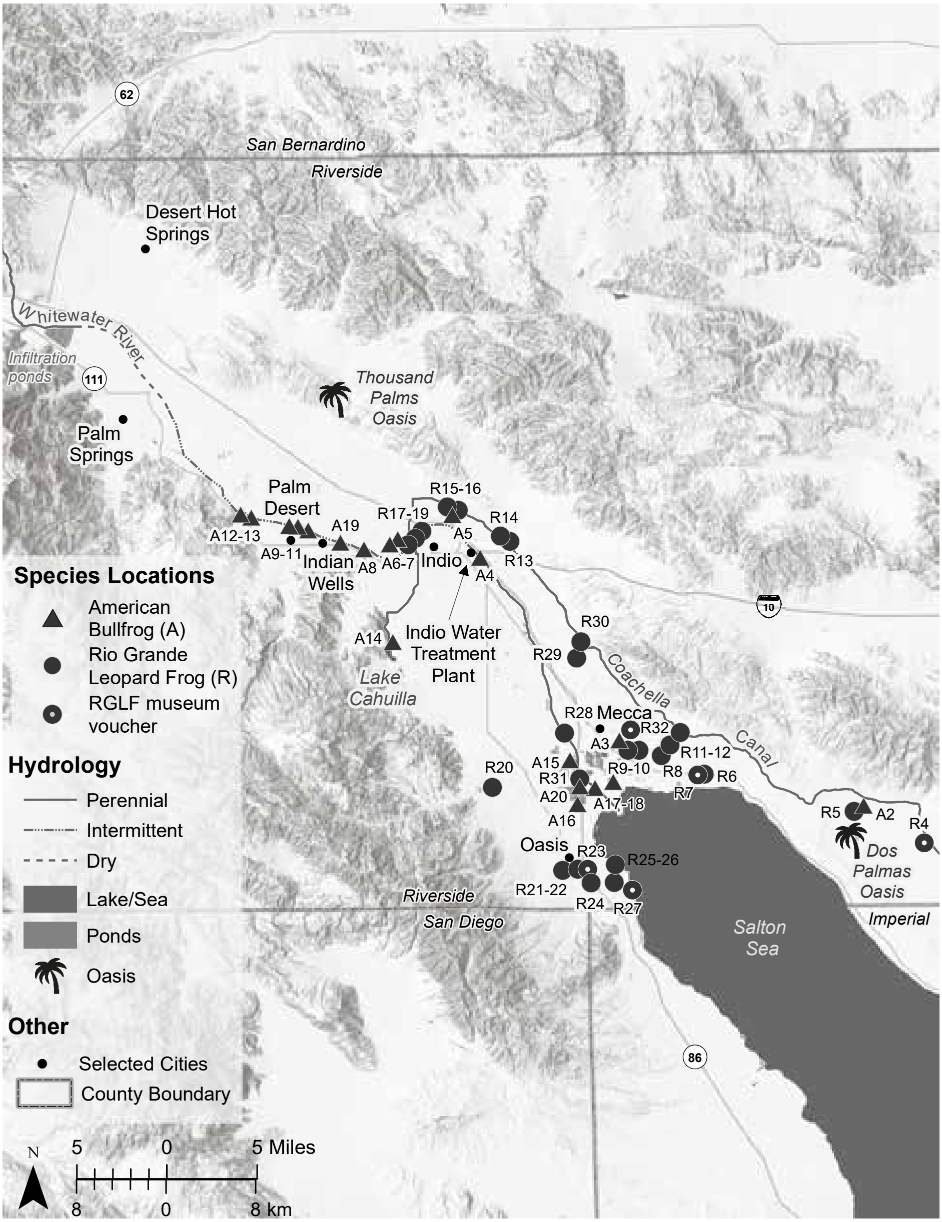

Figure 2.—Location map for Rio Grande leopard frog and American Bullfrog. Coordinates and site details are listed in Appendix B. All records shown are detections (solid symbols) and vouchered specimens or photos (hollow symbols) from this study.

Plains Toad only locally, such as in suitable grassy sections of the Whitewater River channel (e.g., site G10), but there are many museum specimens from the 1930s to the 1970s from the upper valley, including north of Interstate 10. Examples are Indio: 1952, MVZ 563; Palm Springs: 1964, LACM 74549; Whitewater Canyon: 1965, LACM 171036; near Thousand Palms Oasis: 1975, SBCM 2401. These records may reflect the decline of farming and spread of urbanization in the upper valley. Figure 3 shows only our recent locations for this species.

Woodhouse's Toad.—At present, this species is common throughout the lower agricultural part of the valley (Figure 3) where it is seen more frequently than Great Plains Toad, with individuals on the roads at night through citrus groves and vineyards from spring through fall. Woodhouse's Toad is reported to be increasing in the Palm Springs area in the urban northern Coachella Valley (Sullivan 2005) and is found sparingly north of I-10 (See Figure 3, iNaturalist records). Woodhouse's Toad was previously found occasionally at Thousand Palms Oasis, though not in recent years (G. Short, Center for Lands Management, personal communication) and 1987 (MVZ 233366). Adults call and form small congregations in irrigation ponds and can be found in small numbers in canals and ditches. Woodhouse's Toad bred at Palm Island Drive, where tadpoles and transformed toadlets

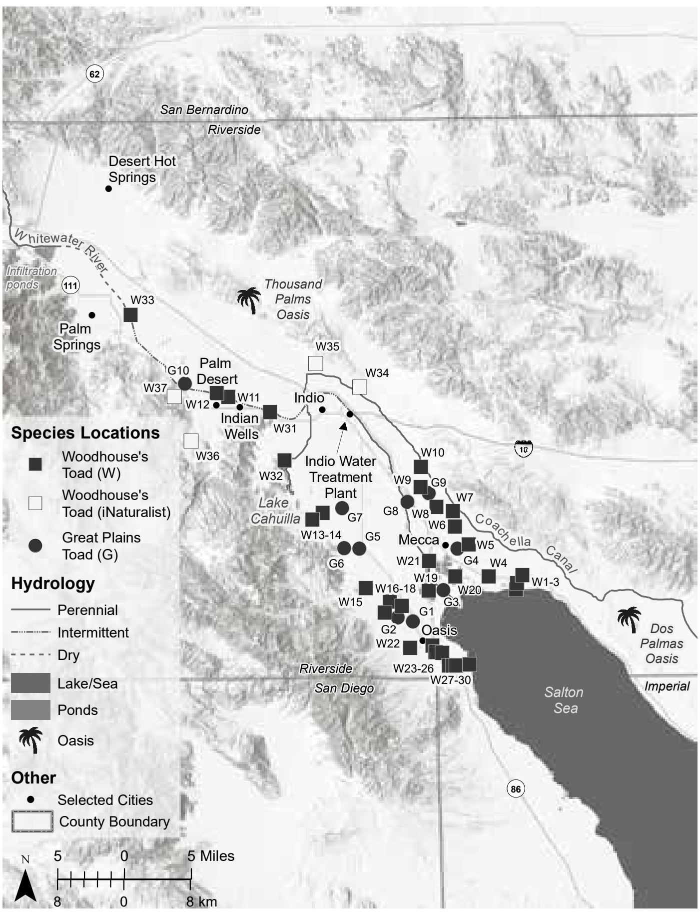

FIGURE 3.—Location map for Great Plains toad and Woodhouse's toad. Coordinates and site details are listed in Appendix B. Museum specimens mentioned in the text are not mapped. All records shown are detections from this study (solid symbols) or iNaturalist records (hollow symbols).

were found at the edge of the Salton Sea in the drain's fresh water where it passed through the barnacle and salt-encrusted shoreline (site W1), and subadults and adults were found in similar conditions at Ave. 84 (site W31). A 2016 record on iNaturalist from Whitewater Canyon (not mapped) indicates Woodhouse's Toad may have moved up the Whitewater River channel from the Coachella Valley.

Spiny Softshell.—Spiny Softshells are currently found along the length of the Coachella Canal, and presumably moved up into the Coachella Valley soon after the canal become operational in 1948. Our sightings of this species from 2006-2009 in the Coachella Canal encompass its entire length in Riverside County, from near the county line to within 2 km of its terminus at Lake Cahuilla (Figure 4). Softshells can be seen basking on the concrete banks of the canal in the morning or resting just below the surface. Twenty-three softshells were seen in a drain leading to the northern part of the Salton Sea (site SO11). They are found in the Whitewater River at the Indio Wastewater Treatment Plant outfall (site SO14), and downstream to the edge of the Salton Sea (site SO12). We have not observed eggs or hatchlings in the Coachella Valley.

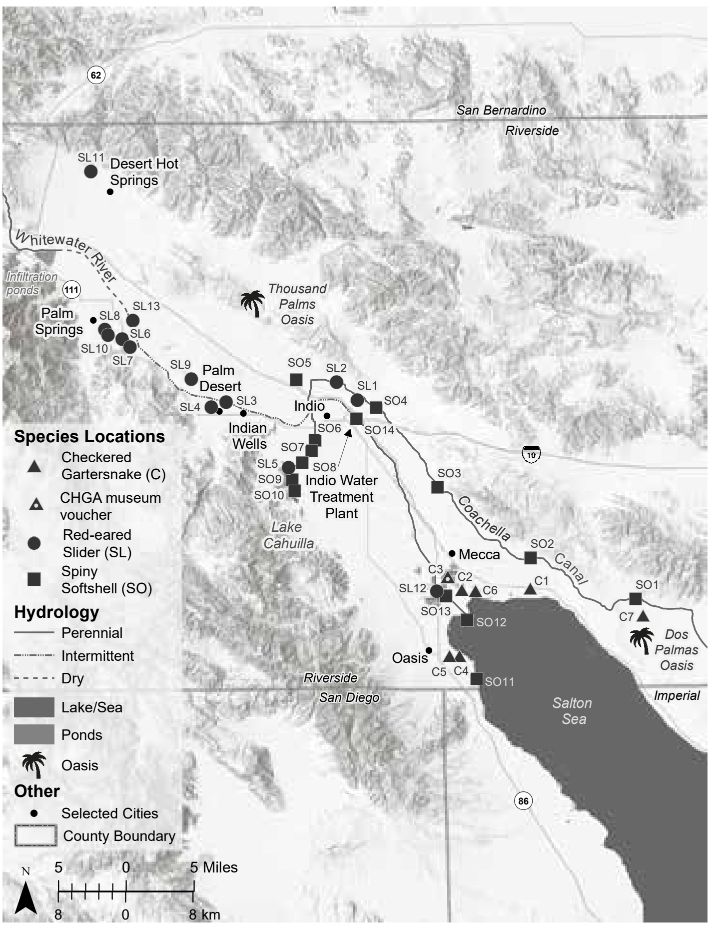

Figure 4.—Location map for Spiny Softshell, Red-eared Slider, and Checkered Gartersnake. Coordinates and site details are listed in Appendix B. All records shown are detections (solid symbols) and vouchered specimens (hollow symbols).

Red-eared Slider.—Red-eared Slider is more known from the coastal slope than from desert communities, but we found that this species is widely distributed in the upper Coachella Valley (Figure 4). It can be quite abundant in golf course and other ponds where algae and at least a modicum of emergent vegetation can grow. For example, the ponds at the Palm Desert Civic Center (SL4) were drained in 2015 and about 300 sliders of all ages were removed (B. Chuck, City of Palm Desert, personal communication). We found one slider in the Whitewater River in an urban seep pool in an otherwise dry section of the river (site SL3). Only one of our records is from the Coachella Canal (site SL2), but this points to the canal as another potential conduit for dispersal. We obtained proof of attempted breeding on 18 July 2018, with a clutch of 17 slider eggs near Mecca, laid in damp soil found between two ponds (site SL12).

Checkered Gartersnake.—We have undated recent sightings of this species at Dos Palmas Preserve adjacent to the Canal (J. Cornett, Palm Springs Desert Museum, personal communication) and from the Grant Street drain east of Mecca (J. Crayon, California Dept. of Fish and Wildlife, personal communication). We contribute three additional records from the vicinity of Mecca, in or near irrigation drains, and two records adjacent to the northwest part of the Salton Sea (Figure 4). It appears to be established but uncommon in suitable habitat in the southernmost agricultural part of the Coachella Valley.

Species Not Established.—Western Toad (*Anaxyrus boreas*).—This montane and cismontane species was found in residential Palm Springs in 2018 (two iNaturalist records), and in Indio in 2005 (this study, LACM 160934). A Western Toad was reported to have been seen near Thousand Palms Oasis in the 1980s (C. Barrows, University of California, Riverside, personal communication). These may be strays from the foothills, or from heretofore undetected breeding locations.

California Treefrog (*Pseudacris cadaverina*).—Tadpoles and a recently transformed frog at the Palm Springs water infiltration ponds on 18 July 2018 probably originated as eggs or larvae washed down the Whitewater River. Treefrogs probably of this species previously occupied Thousand Palms Oasis but were last seen in the 1970s following impacts of intense flooding and introduced species (C. Barrows, University of California, Riverside, personal communication).

Baja California Treefrog (*Pseudacris hypochondriaca*).—This montane and cismontane species has become established at other desert locations in California (Stebbins 2003). Two iNaturalist records are from residential Palm Springs and the City of Thousand Palms, both in 2017. Well-irrigated landscaping and ponds for breeding may allow this species to become established in the Coachella Valley.

Couch's Spadefoot (*Scaphiopus couchii*).—The California Natural Diversity Database (CNDDB) has two recent sight records of this species in appropriate habitat in Riverside County at the southeastern edge of the Coachella Valley from 1993 and 2007, and a 2007 record in nearby Imperial County. The two Riverside County locations are adjacent to the Coachella Canal, suggesting this species is following the same conduit northward as RGLF and other herpetofauna. Potential breeding habitat is found along the Coachella Canal where rainwater backs up against the berms. The 1993 CNDDB record reports breeding on 2 March, but this would be very unusual for what is an obligate summer breeder throughout its range. Nonetheless, establishment should be looked for in flooded desert habitat at the edges of the lower Coachella Valley.

African Clawed Frog (*Xenopus laevis*).—The California Department of Fish and Wildlife has received unverified reports of this species in the Coachella Valley, and it has been documented once from Imperial County (USFWS 2017). Clawed frogs were introduced into the United States in large numbers in the 1930s-1940s for medical use, laboratory study, and the pet trade, and since 1968 have become broadly established in coastal and inland southern California as well as Tucson, Arizona (Dodd 2013, USFWS 2017). Due to the ability of this species to tolerate elevated temperatures and brackish and poor-quality water (Munsey 1972), it would not be surprising if this species is eventually found in the Coachella Valley.

Discussion

The Coachella Valley herpetofauna is mix of species of different origins, most of which were either native to or introduced to the lower Colorado River Valley (LCRV). At least some of the species native to the LCRV also ranged westward to the Imperial Valley (see Figure 1). Great Plains Toad, Woodhouse's Toad, Sonoran Desert Toad (*Incilius alvarius*), Lowland Leopard Frog (*Lithobates yavapaiensis*), and Checkered Gartersnake may have arrived when the Colorado River flooded through a partially constructed canal in 1905-1906, entering the Imperial Valley and creating what is now the Salton Sea (McCollum 2000). However, several natural flooding events from the Colorado River into the Salton Sink took place during the 1800s and earlier. The Colorado overflowed into the Salton Sink in 1840, 1849, 1852, 1859, 1867, and in 1891 when a 48 km-long temporary lake formed (Ibid.). As described by T. F. Cronise (1868), in 1840, the Salton Sink was "…partially submerged by the waters of the Colorado. The New River, through which a portion of those waters now finds its way to the sea, had no existence until that year. A number of large lagoons remained for several years after that inundation."

It is not known exactly when these species made it to Imperial County, but most of them either did not spread north to the Coachella Valley or didn't persist there. However, one species, the Great Plains Toad, native to the LRCV (Camp 1915, Grinnell and Camp 1917) has been in the Coachella Valley for at least a century. The first museum record of Great Plains Toad in the region was a series collected by Charles Camp in Mecca in 1908, only three years after the formation of the Salton Sea (Storer 1925). It is possible that this species was able to move the approximately 30 km from northern Imperial County, and then become common enough to be noticed by the collector, all in only two to three years. One of the specimens collected by Camp was an adult measuring 82 mm in length. To reach adult size would take about four years (Ibid.) meaning that juvenile toads or large tadpoles would have to have been swept all the way to Mecca in 1905 when the Colorado River flowed into the Salton Sink.

A more plausible explanation for Great Plains Toad entering the Coachella Valley is entry during one or more of the natural flooding events in the 1800s or earlier, which would make it a species native to the Coachella Valley. Great Plains Toads could have survived dry periods in habitat afforded by temporary pools and permanent springs. Along the western edge of the Coachella and Imperial Valleys the 1853-54 Blake expedition (Blake 1858), encountered native Americans using spring water to grow crops and found "…a dense growth of weeds over a wide area near the mountains...The ground upon which they grew was moist and miry being supplied with water by numerous springs."

Indeed, Lowland Leopard Frog, previously known from the isolated San Felipe Springs on the western edge of the Salton Sink may have been a relic from earlier contact with the Colorado River system, perhaps from Lake Cahuilla, one of the major predecessors of the Salton Sea (Ruibal 1959).

The completion of the All-American Canal in 1940 and the Coachella Canal in 1948 to bring water from the Colorado River (Imperial Irrigation District 2018) facilitated the spread of other species up into the Coachella Valley. Between 1948 and 2007 much of the Coachella Canal was unlined (San Diego County Water Authority 2016), adding potential breeding habitat to the possibility of passive transport.

Woodhouse's Toad was found in the early 1900s in the Imperial Valley, (e.g. 1929, El Centro, CAS-SU(Amp) 3033-34) and was collected at Harpers Well in 1939 in the vicinity of San Felipe Creek, upstream from cultivated land in the western Imperial Valley (MVZ 31539). The only record for the Coachella Valley in Glaser (1970) is from Mecca. It is undated but can be assumed to be from the 1960s as Glaser believed Woodhouse's Toad was just beginning to enter Riverside County from Imperial County. The earliest museum records for this species in the Coachella Valley are from 1962, North Shore, LACM 88516; 1963, Mecca, LACM 88518 and Indio, LACM 88517. There are many subsequent records. Numerous collections of Great Plains Toads in the Coachella Valley from the 1930s to the 1960s are not paralleled with records of Woodhouse's Toads, suggesting a lack of collecting effort does not explain the lack of pre-1960s Woodhouse's Toads in museum collections. It appears this species did not get established or persist in the Coachella Valley with the 1905-06 or earlier floodwaters that created temporary wetlands in the Salton Sink. It is now widely distributed in the Coachella Valley.

The history of American Bullfrog's initial entry into southeastern California has been well-documented (Dill 1944) and summarized (Jennings and Hayes 1994). Bullfrogs were introduced deliberately to the LCRV in the 1920s and expanded rapidly from the 1930s to the 1950s. We suspect this species has taken the same route to the Coachella Valley as other riparian species, namely, the canals from the Colorado River and agricultural land in Mexico and Imperial County. However, bullfrogs quickly became so common in coastal southern California and elsewhere in the state that we cannot rule out direct introductions to the Coachella Valley in the latter half of the 20th century. By the late 1960s, it had colonized the Imperial Valley, but there was no evidence of the species in the Coachella Valley at that time (Stebbins 1966). Similarly, Glaser (1970) does not list this species in the Coachella Valley, and museum searches for specimens from that era were negative. Stebbins (1985) indicated bullfrogs had finally made it to the Coachella Valley, where they are now widespread.

Spiny Softshells were introduced to the Colorado and Gila rivers in Arizona around 1900 (Miller 1946, Bury and Luckenbach 1976). From the Colorado River, they presumably moved through irrigation canals and/or the New and Alamo Rivers from Mexico to Imperial County. Museum records show that this species had colonized the Imperial Valley by the 1940s and 1950s. Examples include 1942-45, Calipatria, CAS-SU 11755; 1952, shore of Salton Sea near Mullet Island, CAS-SU 15143; and 1955, 5 km NE of Holtville, MVZ 78792. Softshells only recently spread from the Imperial Valley up to the Coachella Valley in Riverside County. Glaser (1970) reports no records from Riverside County, but that "…a dead softshell was noted by Dr. Ernest Karlstrom in 1956 on the southeastern shore of the Salton Sea, about 12 miles south of the Riverside County Line." Stebbins (2003) does not list spiny softshell as occurring north of the Imperial Valley, but California Department of Fish and Game (2005) lists this species in both Imperial and Riverside counties around the Salton Sea. We found this turtle now occupies the Coachella Canal, the lower Whitewater River, and drains feeding into the Salton Sea.

Red-eared Sliders are native to the southeastern United States. This turtle has been introduced around the world through the pet trade, including the Pacific slope of California where it often occurs in urban areas (Ernst and Lovich 2009). Most introductions are considered to be individuals released into ponds and reservoirs by pet owners (Spinks et al. 2003). The question of whether these turtles are breeding in the wild is complicated by continued releases of sliders by pet owners. Breeding in California is perhaps more often assumed than proven (e.g. California Department of Fish and Wildlife undated) though breeding has been confirmed in northern California (Bettelheim et al. 2006), and we documented attempted breeding in the Coachella Valley.

Evidence suggests the Checkered Gartersnake is a very recent addition to the Coachella Valley. They are native to the lower Colorado River (Yarrow 1882 cited in Grinnell and Camp 1917): specimen from 1855, Fort Yuma, USNM852. The first published record for the Coachella Valley is from Mecca in 1997 (Hollingsworth and Prosser 1997) about 2 km from the Coachella Canal. This species probably entered the Coachella Valley through the Coachella Canal, but the paucity of records makes it impossible to pinpoint when this species arrived.

Rio Grande Leopard Frog arrived in the Coachella Valley at the turn of the 21st century, with compelling evidence that they came via the Coachella Canal. From its first discovery in 1981 near Yuma, Arizona on the lower Colorado River (Platz et al. 1990), RGLF was subsequently found in Imperial County in farmland in the LCRV (Clarkson and Rorabaugh 1989) and further west in the Imperial Valley (Jennings and Hayes 1994). Rorabaugh et al. (2002) predicted its spread northward through the Coachella Canal into Riverside County. This species has also moved up the Salt and Gila Rivers of Arizona from the LCRV (Ibid.). We have shown in Results that RGLF is now well-established in the Coachella Valley where it has room and habitat to expand further.

Wildlife biologists have tried to control numbers of bullfrog at Dos Palmas because of their potential predation on desert pupfish. Preserve managers must now contemplate the potential risks of both species of introduced frogs. Adult RGLF in Arizona were found to feed predominantly on invertebrates and often on young leopard frogs but not fish (Platz et al. 1990), and in Texas they fed almost exclusively on terrestrial invertebrates (Parker and Goldstein 2004). Thus, they might not pose a serious threat to Desert Pupfish, which so far persist at Dos Palmas despite the presence of both species of frogs (J. Miner, Bureau of Land Management, personal communication). On the other hand, bullfrogs occasionally feed heavily on Desert Pupfish (Marsh and Sada 1993) despite few examples of predation on fish reported in prey studies of bullfrogs (e.g., Korschgen and Moyle 1955, Stewart and Sandison 1972, Clarkson and deVos Jr. 1986, Casper and Hendricks 2005). Similarly, RGLF, as generalist feeders, might prey on Desert Pupfish under certain conditions. The presence of RGLF at Dos Palmas and in drains at the edge of the Salton Sea therefore potentially puts Desert Pupfish at risk.

We have no evidence that RGLF has negatively affected the other anurans in the Coachella Valley, most of which are non-natives. While there is no immediate risk to native anurans from the colonization of the Coachella Valley by RGLF, there are still potential future impacts. The Lowland Leopard Frog is the only native ranid previously known from extreme southeastern California, but it is now considered extirpated from the state (Black 1980, Sredl 2005). Lowland Leopard Frog probably declined or disappeared from the LCRV and the Imperial Valley before RGLF became established (Vitt and Ohmart 1978, Jennings and Hayes 1994, Jennings and Fuller 2004, Thomson et al. 2016). Nonetheless, RGLF is considered a competitive risk in Arizona, where it is encountering native ranids in the Gila and Salt River drainages, to Lowland Leopard Frog in particular (Rorabaugh et al. 2002). In some areas of Texas, RGLF is sympatric with other leopard frog species without hybridization (Hillis 1981), but in other areas they hybridize with Southern Leopard Frogs, (*Lithobates sphenocephalus*) (Kocher and Sage 1986) and Plains Leopard Frogs (*Lithobates blairi*) (Platz 1972). Local displacement of Plains Leopard Frog by RGLF was documented in Texas (Platz 1981).

Whether or not RGLF will displace or otherwise negatively affect native foothill anurans is a question that has not been tested. The recent reappearance of the federally Threatened California Red-legged Frog (*Rana draytonii*) at the Whitewater Preserve, (Backlin et al. 2017) only about 23 km upstream from Palm Springs may be a test of possible negative effects of the spread of RGLF. The dispersal ability of RGLF is impressive: colonization of a water tank 1.6 km from the nearest water source was documented in Arizona (Rorabaugh 2005), and RGLF moved an estimated 16 km/year along intermittent reaches of the Gila River (Platz et al. 1990). As pointed out by J. Rorabaugh (personal communication) colonization of Whitewater Canyon could facilitate eventual spread throughout the foothills of the San Bernardino Mountains and eventually the coastal slope of California where it could encounter a variety of native anurans. The coastal slope could also be reached westward through San Gorgonio Pass.

If RGLF continues to spread in the densely populated northern Coachella Valley, the chance of intentional or unintentional transport by people increases. RGLF is continuing to spread in the American Southwest and should be monitored closely.

Within the Coachella Valley, the riparian herpetofauna may eventually change in species composition though competition, habitat changes, and additional introductions. Colonization of the valley floor by native foothill species should be looked for in the future, as well as spread of valley species into the foothills.

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Appendix A. Museum Voucher Specimens

Anaxyrus boreas—LACM 160934. Roadkill. Varner Ave., 1.3 mi W of Monroe St., N side of I-10. Indio, Riverside Co., CA, USA. 33.743799ºN, -116.256617ºW. Coll. D. Goodward, 21 February 2005.

Lithobates berlandieri—LACM 178031. Hwy. 111 between Palm Island Drive and Cleveland St., Mecca, Riverside Co., CA, USA. 33.534206ºN, -115.97975ºW. Coll. D. Goodward, 06 September 2009. (site R7).

LACM 178032. 0.2 mi. east of Highway 86S on Avenue 81, Oasis, Riverside Co., CA, USA. 33.462534ºN, -116.087603ºW. Coll. D. Goodward, 07 March 2010. (site R23).

LACM 178033. Coachella Canal, from wildlife drinker, siphon 23, North Shore, Riverside Co., CA, USA. 33.470796ºN, -115.750433ºW. Coll. M. Wilcox, 05 April 2008. (site R4).

LACM PC 2409. Photo. Irrigation pond, 66th Ave., 2.2 km east of Mecca, Riverside Co., CA, USA. 33.568472ºN, -116.053348ºW. D. Goodward, 24 October 2007. (site R32).

LACM PC 2410. Photo. East end of irrigation drain, 84th Ave., 5.6 km southeast of Oasis, Riverside Co., CA, USA. 33.441859ºN, -116.043466ºW. D. Goodward, 27 September 2017. (site 27).

Thamnophis marcianus—LACM 178035. Roadkill, Lincoln Street, 1.7 mi. south of Mecca, Riverside Co., CA, USA. 33.547121ºN, -116.079122ºW. Coll. D. Goodward, 05 February 2009. (site C3).

Appendix B. Specific Site Locations and Data

Sites with more than one species are listed under each species found there, i.e., some locations are listed more than once. Coordinates format is decimal degrees. Date format is month/day/year.

SITE	LATITUDE	LONGITUDE	DATE	DESCRIPTION
Rio Grande Leopard Frog				
R1	33.373441	-115.610177	10/07/2005	Tadpoles, one subadult frog in seepage pond, leakage from Coachella Canal, Imperial County, about 10 km southeast of county line.
R2	33.439248	-115.690848	04/05/2008	Five frogs, siphon 21 of Coachella Canal.
R3	33.453935	-115.713531	04/05/2008	Five frogs and egg mass, siphon 22 of Coachella Canal.
R4	33.470796	-115.750433	04/05/2008	One adult, siphon 23 of Coachella Canal. Voucher specimen.
R5	33.503712	-115.830013	several	Dos Palmas Preserve, see Table 1 for dates. RGLF and bullfrogs.
R6	33.534741	-115.97385	09/06/2009	One adult, Cleveland Street ditch, north of Highway 111.
R7	33.534206	-115.97975	09/06/2009	One adult, Highway 111 south of Palm Island Drive, rainy night. Voucher specimen.
R8	33.550376	-116.017809	03/07/2010	One juvenile, Wheeler Street, 500 m south of 68th Avenue, near irrigation pond. Rainy night.
R9	33.554403	-116.036027	03/07/2010	One adult, 68th Avenue at Colfax Street. Rainy night.
R10	33.554718	-116.046815	03/07/2010	One adult, 68th Avenue, 260 m west of Grant Street. Rainy night.
R11	33.561966	-116.009453	03/07/2010	One subadult in irrigation pond, 800 m north of 68th Avenue on Garfield Street.
R12	33.569087	-116.000624	04/12/2008	Several in irrigation pond near Coachella Canal, east end of 66th Avenue.
R13	33.725323	-116.163814	08/23/2012	About 20, mostly juveniles, a few adults. The Vineyards resort. In ponds and on lawns.
R14	33.726748	-116.164187	08/01/2009	Four adults, in grates on Coachella Canal, Dillon Road crossing. None in grates further west, up to Jackson Street.
R15	33.74539	-116.212003	08/29/2012	Several adults and subadults, Indio public golf course, in pond near entrance.
R16	33.747948	-116.212473	08/29/2012	Several adult RGLF and subadult bullfrogs, north edge of Indio public golf course, seep ponds with overflow from golf course plus recent rainfall.
R17	33.726388	-116.253049	08/29/2012	One RGLF calling, one bullfrog seen, leakage seep from Coachella Canal, 350 m southeast of Fred Waring Drive.
R18	33.723652	-116.251809	09/13/2011	Several frogs in La Hacienda plant nursery, Miles Street, near cattail-filled pond.

SITE	LATITUDE	LONGITUDE	DATE	DESCRIPTION
R19	33.722166	-116.254334	09/13/2011	Dozens of subadults and juveniles, exiting plant nursery on Miles Street, three on bank down to Whitewater channel, no frogs in channel puddles below bridge. Rainy night.
R20	33.525526	-116.177338	05/20/2009	Four subadults, west end of 72nd Avenue, in irrigation pond.
R21	33.462559	-116.100051	09/05/2009	One juvenile and one adult on 81st Avenue, 260 m west of Harrison Street, by fish farm. Rainy night.
R22	33.462803	-116.094784	07/10/2011	One subadult, 81st Avenue and Buchanan Street, on bank of irrigation pond.
R23	33.462503	-116.087363	03/07/2010	One adult, 350 m east of Highway 86S on 81st Avenue. Near date grove, rainy night. oucher specimen.
R24	33.455545	-116.088069	07/30/2009	One adult RGLF in irrigation pond, 82nd Avenue, just east of Highway 86S. Eight Woodhouse's Toads, some calling.
R25	33.448161	-116.061219	03/07/2010	Two adults, 83rd Avenue and Johnson Street. Rainy night.
R26	33.462629	-116.059949	09/29/2017	Four (2 adults, 2 subadults), east end of 81st Avenue, edge of drain, from end of pavement down to dense cattails.
R27	33.441859	-116.043466	09/29/2017	One large juvenile, end of 84th Avenue at lower end of cattails, just before the drain opens to the Salton Sea shore. Photo voucher.
R28	33.568757	-116.10771	09/29/2017	Three juveniles, Whitewater River, bank openings downstream from 66th Avenue bridge.
R29	33.629336	-116.095153	10/05/2017	Four juveniles, Buchanan Street, 190 m north of 58th Avenue, in irrigation pond.
R30	33.642229	-116.091074	10/05/2017	One juvenile, east end of Airport Boulevard, on bank of Coachella Canal.
R31	33.531736	-116.093205	07/18/2018	About 40 RGLF, all ages, hunt club, Mecca, 71st Avenue and Buchanan Street. A few bullfrogs also present.
R32	33.568472	-116.053348	Several, duration of study	Several, in irrigation pond 2.2 km east of Mecca, 66th Avenue at Euclid Street, several dates. RGLF photo voucher on 24 September 2007. Same site as G4, A3, W5.
American Bullfrog				
A1	33.423123	-115.682425	04/26/2003	One, Hot Mineral Spa Road in Imperial County, in small pond.
A2	33.503712	-115.830013	05/10/2005	Dos Palmas Preserve, see Table 1 for dates, bullfrogs and RGLF.
A3	33.568472	-116.053348	Several, duration of study	Several, in irrigation pond, 2.2 km east of Mecca, 66th Avenue at Euclid Street, several dates. Same as G4, R32, W5.
A4	33.711119	-116.197108	09/15/2012	A few bullfrogs calling, wildlife ponds adjacent to Indio Water Treatment Plant, no RGLF. Islands of bulrushes.

SITE	LATITUDE	LONGITUDE	DATE	DESCRIPTION
A5	33.747948	-116.212473	08/29/2012	Several, different ages, north edge of Indio public golf course in shallow overflow ponds.
A6	33.726388	-116.253049	08/29/2012	One bullfrog, one RGLF, leakage seep from Coachella Canal, 350 m southeast of Fred Waring Drive. Same location as R17.
A7	33.724954	-116.254269	08/29/2012	Two adults, Whitewater River, 480 m south of Fred Waring Drive, seep with Arroweed and cattails.
A8	33.717504	-116.299602	09/14/2011	Four, Whitewater River seep, 420 m west of Washington Street. After summer rain, extensive marshy habitat.
A9	33.734501	-116.355949	09/15/2011	Three adults, Whitewater River channel, Cook Street, seep just below road crossing.
A10	33.735408	-116.360823	09/29/2017	Five adults, Whitewater River channel, seep upstream from Cook Street.
A11	33.736231	-116.365157	09/15/2011	One subadult, Whitewater River channel, Cook Street, uppermost seep.
A12	33.744513	-116.40779	09/14/2011	Five heard calling, Whitewater River channel, just below Bob Hope Drive.
A13	33.7463	-116.411553	09/14/2011	About 25 small bullfrog tadpoles, Whitewater River channel, seep above Bob Hope Drive.
A14	33.643563	-116.272286	07/20/2011	Several heard calling, 600 m north of Lake Cahuilla in cattail-lined pond.
A15	33.541551	-116.098351	05/12/2007	Sixty-five bullfrogs (no RGLF), fish farm along Highway 86S at 72nd Avenue.
A16	33.511668	-116.095572	03/07/2010	Thirteen juveniles on road at night, near hunt club pond on Buchanan Street, 150 m north of 74th Avenue.
A17	33.524658	-116.078981	05/02/2009	Multiple observations, Lincoln Street crossing, Whitewater River. Listed date is one of many.
A18	33.529445	-116.06129	08/09/2009	Bullfrogs encountered multiple times, Johnson Street ditch, west from Highway 111. Listed date is one of many.
A19	33.723761	-116.322089	11/05/2017	Twenty-three juveniles, Whitewater River channel, below Renaissance Esmeralda Resort, 450 m west of Miles Avenue. Cattails and open water.
A20	33.531736	-116.093205	07/18/2018	Four adult bullfrogs with many RGLF, hunt club, Mecca. 71st Avenue and Buchanan Street.
Great Plains Toad				
G1	33.491745	-116.11798	07/30/2009	Ten adults, 77th Avenue, 500 m west of Pierce Street, date grove under flood irrigation.
G2	33.499228	-116.129511	07/30/2009	Six adults (also 20 Woodhouse's Toads), Fillmore Street and 86th Avenue. Great Plains Toads in clumps of flooded grass in recently filled irrigation pond.
G3	33.525295	-116.082995	03/10/2008	Chorus heard west of Whitewater River at Lincoln Street, habitat unknown.

SITE	LATITUDE	LONGITUDE	DATE	DESCRIPTION
G4	33.568472	-116.053348	Several, duration of study	Several calling adults, irrigation pond 2.2 km east of Mecca, 66th Avenue at Euclid Street, Same as R32, A3, W5.
G5	33.568815	-116.185153	07/30/2009	Chorus of about twelve, 66th Avenue, 100 m west of Harrison Street, in date grove.
G6	33.569471	-116.204952	07/30/2009	Chorus of about 6, 66th Avenue 800 m east of Jackson Street, in flooded date grove.
G7	33.613025	-116.206984	07/30/2009	One adult, 60th Avenue and Calhoun Street, next to date groves.
G8	33.618961	-116.122224	05/05/1995	Chorus, Oasis Date Gardens, Highway 111, vicinity of 59th Avenue, Thermal, in flooded date grove and in nearby puddles.
G9	33.628319	-116.09446	08/01/2009	Adults and tadpoles, 58th Avenue and Buchanan Street, vineyard with flooded furrows. Citrus and field crops nearby.
G10	33.7463	-116.411553	09/14/2011	One adult, Whitewater River channel, 350 m upstream from Bob Hope Drive. Recent rains, grassy habitat.
Woodhouse's Toad				
W1	33.423123	-115.682425	04/26/2003	Hundreds of tadpoles and new toadlets, irrigation drain at end of Palm Island Drive, on wet sand, Salton Sea shore.
W2	33.503712	-115.830013	03/07/2010	One, South end of Palm Island Drive, rainy night.
W3	33.538519	-115.973864	03/30/2002	One adult, Cleveland Street ditch north of Highway 111. Three on 09/07/2002.
W4	33.537387	-116.017778	03/07/2010	One adult, Wheeler Street, 300 m south of 70th Avenue. Rainy night.
W5	33.568472	-116.053348	08/01/2009	Several adults, irrigation pond 2.2 km east of Mecca, 66th Avenue at Euclid Street. Same as G4, A3, R32. Several visits.
W6	33.591631	-116.061128	08/29/2012	One adult, Johnson Street 800 m south of 62rd Avenue. Rainy night.
W7	33.612887	-116.060622	03/07/2010	One adult, Johnson Street and 60th Avenue. Rainy night.
W8	33.61327	-116.084706	03/07/2010	One adult, 60th Avenue 640 m west of Lincoln Street. Rainy night.
W9	33.629336	-116.095153	08/01/2009	Twenty adults, Buchanan Street at 58th Avenue, irrigation pond, same as R29.
W10	33.656628	-116.104464	08/01/2009	One on road, east end of 54th Avenue, vineyards.
W11	33.734501	-116.355949	10/05/2017	One subadult Woodhouse's Toad, four adult bullfrogs, Cook Street seep, Whitewater River channel.
W12	33.736231	-116.365157	10/05/2017	One subadult, upper Cook Street seep.
W13	33.605441	-116.365157	02/21/2005	One, 160 m east of Monroe Street on 61st Avenue. Recent rains.

SITE	LATITUDE	LONGITUDE	DATE	DESCRIPTION
W14	33.598199	-116.245097	02/21/2005	One, 62nd Avenue, 100 m east of Madison Street. Recent rains.
W15	33.525526	-116.177338	05/20/2009	Woodhouse's Toads calling from irrigation pond west end of 72nd Avenue, Same as R20.
W16	33.510651	-116.145933	07/30/2009	Two on 74th Avenue, 130 m east of Polk Street.
W17	33.503476	-116.147456	03/07/2010	One adult, Polk Street, 750 m south of 74th Avenue, rainy night.
W18	33.503476	-116.129511	07/30/2009	Twenty Woodhouse's Toads and six Great Plains Toads at irrigation pond, Fillmore Street 500 m north of 76th Avenue. Same as G2.
W19	33.523156	-116.095564	05/02/2009	One adult on road, 3 calling, Buchanan Street and 72nd Avenue, hunt club ponds.
W20	33.53761	-116.061241	09/10/2004	Two, Johnson Street ditch, south of Highway 111, near cattails.
W21	33.554616	-116.094793	02/21/2005	Two, on Highway 86S at 68th Avenue. Recent rains.
W22	33.462517	-116.116573	03/07/2010	One immature, 81st Avenue 370 m west of Pierce Street.
W23	33.462598	-116.09151	07/30/2009	One adult on 81st Avenue at Highway 86S. Also 02/21/2005, one in front of Oasis Palms RV Park, 100 m to the west. No recent rain.
W24	33.45565	-116.088069	07/30/2009	Eight Woodhouse's Toads, some calling, in irrigation pond at west end of 82nd Avenue. One adult RGLF also.
W25	33.45565	-116.086927	07/30/2009	One adult calling in pond, 100 m east of site W24.
W26	33.454584	-116.07868	07/30/2009	One, Lincoln Street near 82nd Avenue.
W27	33.441151	-116.070173	07/30/2009	Two calling, irrigation pond, 84th Avenue 440 m east of Highway 86S.
W28	33.440901	-116.065095	07/30/2009	Three calling, irrigation pond, 84th Avenue 380 m west of Johnson Street.
W29	33.440978	-116.061148	09/29/2017	Two adults, 84th Avenue at Johnson Street.
W30	33.441859	-116.043466	09/29/2017	One large juvenile, east end of 84th Avenue, at end of drain where it opens onto the Salton Sea shore. An adult on barnacle sand nearby. Three adults calling here April 2016.
W31	33.717296	-116.299624	09/14/2011	Two subadult Woodhouse's Toads, four bullfrogs, Whitewater River channel above Washington Street crossing. After summer rain.
W32	33.665024	-116.281483	10/20/2017	One dead on golf pathway, Silver Rock Resort.
W33	33.82396	-116.480536	10/27/2017	One adult in yard, Cimarron Resort, Palm Springs.
W34	33.744674	-116.186492	08/24/2011	iNaturalist photographic record, Indio.
W35	33.778539	-116.247966	04/01/2016	iNaturalist photographic record, Indio Hills.
W36	33.683940	-116.404821	05/19/2014	iNaturalist photographic record, Palm Desert.
W37	33.746816	-116.414503	07/09/2018	iNaturalist photographic record, Rancho Mirage.

SITE	LATITUDE	LONGITUDE	DATE	DESCRIPTION
Spiny Softshell				
SO1	33.521505	-115.839148	12/09/2006	Three adults, siphons 28-29, draining unlined Coachella Canal.
SO2	33.566264	-115.97277	03/01/2009	Three on banks of Coachella Canal, just east of Cleveland Street crossing.
SO3	33.642721	-116.09121	07/07/2005	One on bank of Coachella Canal, Airport Boulevard crossing.
SO4	33.728637	-116.168843	03/04/2009	Three on bank of Coachella Canal, at Dillon Road crossing.
SO5	33.758582	-116.270777	03/04/2009	One on bank of Coachella Canal, east of Madison Street crossing.
SO6	33.694013	-116.247274	08/04/2009	One on bank of Coachella Canal, between 48th-49th Avenues.
SO7	33.682913	-116.251711	08/04/2009	One on bank of Coachella Canal, south of 50th Avenue crossing.
SO8	33.670263	-116.263788	08/02/2009	Two on bank of Coachella Canal, 52nd Avenue crossing.
SO9	33.651664	-116.277034	08/02/2009	One on bank of Coachella Canal, by golf course, southwest of 54th Avenue crossing.
SO10	33.639815	-116.274006	07/20/2011	One on bank of Coachella Canal, 160 m north of Lake Cahuilla.
SO11	33.437695	-116.043473	08/17/2009	Twenty-three adults and large juveniles, no small juveniles, end of 85th Avenue, in irrigation drain.
SO12	33.500318	-116.05452	09/05/2009	One adult, in Whitewater River at mouth.
SO13	33.526241	-116.081357	04/28/2007	Four adults, on sandy bank of Whitewater River above Lincoln Street crossing. Also 3 on 03/23/2008.
SO14	33.716759	-116.194167	09/14/2012	3 adults, Indio Water Treatment Plant outfall, Whitewater River.
Red-eared Slider				
SL1	33.736775	-116.192789	07/08/2011	One, Lago Vista Street and 43rd Avenue, golf course pond.
SL2	33.755844	-116.219285	09/08/2011	One adult sunning on bank of Coachella Canal, west of Jackson Street crossing.
SL3	33.735408	-116.360823	09/29/2017	One large juvenile, Upper Cook Street seep, Whitewater River channel.
SL4	33.730446	-116.380175	10/21/2017	Sixty-five adults and juveniles, Palm Desert Civic Center ponds.
SL5	33.665024	-116.281483	2016-17	Several, Silver Rock Resort, La Quinta. Reported by workers.
SL6	33.80337	-116.493672	10/24/2017	Thirty-one, mostly adults, Taquitz Creek Golf Resort, pond near Water Park.
SL7	33.794719	-116.483958	10/24/2017	Four, Taquitz Creek Golf Resort, small pond near Golf Club Drive and Cree Street.
SL8	33.813517	-116.515785	10/29/2017	Sixteen, all sizes, Compadre Road 265 m south of Ramon Road, Palm Springs. Golf course pond with duckweed.

SITE	LATITUDE	LONGITUDE	DATE	DESCRIPTION
SL9	33.760324	-116.405363	11/05/2017	Five, Eisenhower Medical Center ponds.
SL10	33.807802	-116.511959	12/07/2017	Two adults, pond at Bel Air Greens (abandoned), west of El Cielo Road, 300 m north of East Sonora Road.
SL11	33.982508	-116.532768	03/01/2018	Twenty-three adults and subadults, Mission Springs Golf Resort, Desert Hot Springs, golf course pond.
SL12	33.531736	-116.093205	07/18/2018	One small juvenile, 6 adults, one clutch of eggs, Hunt Club, Mecca, Buchanan Street and 71st Avenue.
SL13	33.823568	-116.480064	08/01/2018	Nine of various sizes, in golf course pond, Cimarron Golf Resort, west end of McCallum Way, Cathedral City.
SL14	33.736828	-116.360803	07/03/2018	Nine, First Tee Golf Course, Cook Street at north edge of Whitewater River channel, Palm Desert.
Checkered Gartersnake				
C1	33.533972	-115.973818	10/19/2002	One immature, Cleveland St. Ditch, north of Highway 111, cattails, other vegetation, shallow water.
C2	33.533126	-116.061128	05/25/2007	DOR, Johnson Street, 780 m south of 70th Avenue, John Green, photo.
C3	33.547121	-116.079122	05/02/2009	DOR, Lincoln Street, 860 m south of 68th Avenue. Voucher specimen.
C4	33.462648	-116.064923	05/17/2008	DOR, 81st Avenue, 330 m west of Johnson Street.
C5	33.462651	-116.077742	03/12/2008	DOR, 81st Avenue, 1.5 km west of Johnson Street. Chet McGaugh. photo.
C6	33.531932	-116.043748	Undated, after 2000	Grant Street drain, J. Crayon, California Department of Fish and Wildlife, personal communication.
C7	33.503712	-115.830013	Undated, 1990s	Dos Palmas Preserve, J. Cornett, Palm Springs Desert Museum, personal communication.

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Serum chemistry of wild, free-ranging mountain lions (*Puma concolor*) in the eastern Sierra Nevada, California, USA

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We report descriptive statistics and reference intervals for serum chemistry from 43 unique mountain lions captured in the eastern Sierra Nevada, California, for ecological or genetic investigations during 1993–2004. We tested for differences between males and females, and winter (when mule deer [*Odocoileus hemionus*] were most abundant in diets) and summer (when smaller prey were more common in diets). Differences in direct bilirubin and total protein occurred between the sexes, whereas seasonal differences occurred in CO_2, A/G ratio, Ca and, again, direct bilirubin. Subjects that were bayed with hounds and captured after short chases exhibited lower levels of creatine kinase than those captured using other methods or that had been pursued long distances. Retrospective comparisons with previously published information also revealed differences in mean levels of Na, cholesterol, and creatine kinase among mountain lions captured after baying with hounds in those independent investigations.

Key words: comparative biochemistry, cougar, geographic variation, mountain lion, puma, *Puma concolor*, reference interval, serum chemistry

There is intense interest, both political and biological, in mountain lion (*Puma concolor*) as a keystone species, highly adaptable predator, and potential threat to human safety (Torres et al. 1996, Bleich and Pierce 2005, Torres 2005, USFWS 2000, 2007). Despite widespread and increasingly detailed research on population genetics, ecology, and the role of mountain lions in ecosystem function, there is a paucity of information on blood parameters (Pierce and Bleich 2003). Such data are limited to few reports in the professional literature that are based on captive individuals (Currier and Russell 1982), wild animals (Currier and Russell 1982, Dunbar et al. 1997, Foster and Cunningham 2009), or combinations of wild and captive animals (Currier and Russell 1982).

Mountain lions are among the most widely distributed mammals in the Western Hemisphere, with an historical range from northern British Columbia to southernmost Argentina and Chile, and occupied suitable habitat from coast to coast in North America (Young and Goldman 1946, Williams 2018). Thus, variation in serum chemistry across the range of the species would not be unexpected (Dunbar et al. 1997), and the value of obtaining data from multiple study areas previously has been emphasized (Dunbar et al. 1997, Pierce and Bleich 2003).

We captured mountain lions from a population occupying a localized and remote area of California, which since has been identified as genetically distinct from other populations of mountain lions in that state (Ernest et al. 2003, Gustafson et al. 2019). Seasonal variation in availability of primary prey (Villepique et al. 2011), the potential for sex-specific differences, and our large sample of wild, free-ranging animals provided an opportunity to examine blood serum chemistry of those cryptic felids occupying a rural and largely pristine region. Moreover, increasing attention to pathogen spillover from domestic cats (*Felis catus*) to mountain lions at the urban-wildland interface (Paul-Murphy et al. 1994, Foley 1997, Bevins et al. 2012, Carver et al. 2016, Kellner et al. 2018) made it especially valuable to document reference intervals in this sparsely populated part of California.

Study Area

Our primary study area, the Round Valley Mule Deer Winter Range (~450 km^2; 37°25' N, 118°36' W), was located along the eastern slope of the Sierra Nevada, Mono and Inyo counties, California, a rural area with a mean population density of <2 persons/km^2 (Duncan 1993). The Sierra Nevada is a massive mountain range reaching elevations in excess of 4,400 m, and extends 640 km in a north-south direction while separating the Great Basin from the San Joaquin and Sacramento valleys to the west (Storer and Usinger 1968). Mule deer (*Odocoileus hemionus*) comprise the primary prey base for mountain lions in the eastern Sierra Nevada (Bleich and Taylor 1998, Pierce et al. 1999, 2000a, 2000b, 2004; Villepique et al. 2011). An endemic subspecies of bighorn sheep (*Ovis canadensis sierrae*), and tule elk (*Cervus elaphus nannodes*)—which are not native to the region—also are occasional prey of mountain lions (McCullough 1969, Johnson et al. 2013, Villepique et al. 2015), as are domestic livestock (Villepique et al. 2011). Mountain lions showed a functional response to the concentration of mule deer on winter range, as evidenced by a marked increase in the frequency of deer remains in lion feces during winter compared to summer, and a corresponding increase of smaller mammals in lion feces during summer (Villepique et al. 2011).

Mountain lions occupy the eastern Sierra Nevada year-round, although some individuals migrate with mule deer to summer ranges (Pierce et al. 1999). Density of mule deer during winter (November–April) in Round Valley was much greater than during summer (May–October) because the majority of wintering mule deer disperse northward to higher elevations or through mountain passes to summer ranges west of the Sierra Crest, returning to winter ranges each year during autumn (Kucera 1992, Monteith et al. 2011). The mule deer population in Round Valley during winter declined substantially, from approximately 6,000 (~13/km^2) in 1985, reaching its nadir of about 1,000 (~2/km^2) in 1991; in 1992, the population began to increase slowly, and trended upward through the remainder of our investigation (Pierce et al. 2012). The mean number of mountain lions occupying the winter range declined from 6.1 in winter 1992-1993 to 0.6 in winter 1998-1999, lagging the decline in the deer population by about 7 years (Pierce et al. 2012, Pierce and Bleich 2014).

Methods

We captured mountain lions for ecological (Pierce et al. 1999, 2000a, 2000b, 2004, 2012; Villepique et al. 2011, 2015), behavioral (Bleich et al. 1996, Pierce et al. 1998), and genetic (Ernest et al. 2003, Gustafson et al. 2019) investigations from 1991 to 2004, and obtained serological information on lions captured during 1993–2004. Our capture protocol followed then-current guidelines of the California Department of Fish and Game (CDFG; Jessup et al. 1986) and animal care and use protocols of the American Society of Mammalogists (*ad hoc* Committee on Acceptable Field Methods in Mammalogy 1987, Kirkland 1998), and was approved by the Institutional Animal Care and Use Committee at the University of Alaska Fairbanks (Pierce 1999).

We immobilized animals with Telazol® (tiletamine HCl and zolazepam HCl; Fort Dodge Animal Health, Fort Dodge, IA) after they were brought to bay by hounds (Young and Goldman 1946) or captured with foot snares (Logan et al. 1999); one individual was caught accidentally in a leg-hold trap set legally for other species, as described by Andreason et al. (2018). Following immobilization, we restrained each animal with hobbles and covered the eyes with a blindfold, obtained morphometrics and weight, and conducted a thorough physical examination. We fitted each mountain lion with a VHF or GPS telemetry collar (Bleich et al. 2000), and used venipuncture to collect 50 cc of blood from the medial saphenous vein, and transferred samples immediately to appropriate vacutainer tubes for serum chemistry and hematology, as well as for anticipated genetic investigations. Blood samples were transported directly from the field (≤4 hr) to Northern Inyo Hospital, Bishop, California, where samples were processed and analyzed upon arrival (Vitros Chemistry System®, Ortho Clinical Diagnostics, Raritan, NJ); funds were not available for processing through a commercial veterinary laboratory. At least one investigator remained with each study animal until it became mobile and departed the capture site.

Ten individuals were represented >1 time among our samples. Thus, we used Mann-Whitney tests to compare variables between males and females, and between winter (November–April) and summer (May–October). If no statistically significant difference ($P > 0.05$) existed between the sexes or between seasons, variables were pooled prior to further analysis. Where differences did occur, we present values for males and females separately and in combination, as well as separately for winter and summer and in combination.

We used Reference Value Advisor (Greffre et al. 2011), an Excel Spreadsheet add-in, to calculate descriptive statistics, analyze each variable for distribution and outliers, and to calculate reference intervals for this population. Reference Value Advisor used Tukey's Test to flag outliers and confirmed them with the Dixon-Reed Test to evaluate the distance from the outlier to the nearest value, divided by the whole range of values; in the absence of an obvious explanation for outliers, we retained them among data to be analyzed (Greffre 2009). Gaussian distribution was assessed in Reference Value Advisor with the Anderson-Darling Test and, as a result, reference intervals and the 90% CI around the upper and lower reference limits were calculated using the nonparametric method (Greffre et al. 2011). Where sample sizes were inadequate to calculate a reference interval, we present only the mean, *SD*, median, and range of values (Friedrichs et al. 2012).

We also summarized results of previous investigations, and conducted retrospective comparisons between analytes reported here and those reported by earlier researchers. We tested for differences between mean values of sodium, potassium, chloride, creatine kinase, total bilirubin, phosphorous, and cholesterol, for which mean values had been reported by Currier and Russell (1982), Paul-Murphy et al. (1994), or Dunbar et al. (1997). We back-calculated standard deviation (Higgins and Green 2011) from the mean and 95% confidence interval provided by Currier and Russell (1982) and then used Welch's Approximate *t*, which is robust to considerable departures from theoretical assumptions when two-tailed tests are employed and samples are large (Zar 1984), for these comparisons.

Results

We report results for 61 blood samples obtained from 43 (20 male, 23 female) unique mountain lions ≥6 months-of-age; descriptive statistics and reference intervals are based on samples ranging in size from 20 to 59 (Table 1). One male and one female were captured with foot snares, one female was accidentally caught in a leg-hold trap, and one female was sampled immediately after being pursued at length and dispatched because of human safety concerns. Blood samples were obtained immediately after the mountain lion was immobilized; in the latter case, however, the sample was obtained immediately following the animal's death. External physical examination and body weight (Roelke 1987, Dunbar et al. 1997), body conformation (our subjective index to body condition), and coat condition (Charlton et al. 1998) indicated that mountain lions included in these analyses were healthy and in good body condition. Further, none exhibited evidence of chronic disease, serious injury, or heavy infestation by external parasites.

Differences (Table 1) occurred between males and females in direct bilirubin (U_A = 577, z = -2.44, P = 0.015) and total protein (U_A = 556.5, z = -2.12, P= 0.034), whereas seasonal differences occurred in CO_2 (U_A = 190.5, z = 3.19, P = 0.001), A/G ratio (U_A = 201.0, z = 3.01, P = 0.003), calcium (U_A = 266.5, z = 1.97, P = 0.049), and again in direct bilirubin (U_A = 542, z = -2.64, P = 0.008). We identified four outliers for creatine kinase, but present results with and without those data. Creatine kinase for animals bayed with hounds ($\bar{x}$ = 554.3, range 148–1,545) was far lower than that involving other methods of capture or a long pursuit ($\bar{x}$ = 13,215.8, range 3,605–25,967); in the absence of those outliers, no difference existed in creatine kinase by sex or season (Table 1). Retrospective analyses indicated differences in mean values for creatine kinase, Na, and cholesterol among wild, free-ranging mountain lions captured in this investigation when compared to results from other independent reports (Appendix I, Appendix II).

Table 1.— Serum chemistry values for mountain lions captured in the eastern Sierra Nevada, Inyo and Mono counties, California, 1993–2004. Reference intervals and the 90% CI around the upper and lower reference limits were calculated using the method of Greffre et al. (2011); where sample sizes were inadequate to calculate a reference interval, we present only the mean, SD, median, and range of values (Friedrichs et al. 2012).

Analyte	*n*	Mean	SD	Median	Range	Reference Interval	90% CI Lower Limit	90% CI Upper Limit
Sodium (mEq/L)	59	158.3	40.2	153.0	141–461	144–309	141–148	157–461
Potassium (mEq/L)	59	4.55	1.85	4.20	3.6–17.6	3.60–12.95	3.60–3.65	5.20–17.60
Chloride (mEq/L)	59	123.3	27.3	121.0	106–327	108.5–227.0	106–112	126–327
CO_2 (mEq/L) (Annual)[a]	59	16.4	6.4	16.0	9–59	9.5–42.0	9.0–11.5	21.0–59.0
CO_2 (Summer)	20	14.2	2.0	14.0	11–18	—	—	—
CO_2 (Winter)	39	17.6	7.5	17.0	9–59	—	—	—
Glucose (mg/dL)	59	144.9	69.1	130.0	62–431	65–391.5	62.0–74.0	279.0–431.0
Blood Urea Nitrogen (mg/dL)	59	42.3	17.4	45.0	16–74	16.0–74.0	16.0–18.0	70.0–74.0
Creatine Kinase (U/L)[b]	57	1458.7	4208.2	514.5	148–25967	148.0–23072.8	148.0–186.7	3062.2–25967.0
Creatine Kinase[c]	53	554.3	336.9	493.5	148–1545	148.0–1467.3	148.0–180.0	1126.0–1545.0
SGPT (U/L)	59	77.9	54.3	65.0	19–364	30.0–316.0	19.0–45.0	169.0–364.0
GGT (U/L)	57	9.5	4.1	9.0	5–28	5.5–26.7	5.0–6.0	17.0–28.0
Alkaline Phosphatase (U/L)	59	37.3	30.0	28.0	10–187	11.0–144.5	10.0–12.5	81.0–187.0
Total Bilirubin (mg/dL)	58	0.47	0.88	0.30	0.1–6.8	0.10–4.33	0.10–0.10	0.85–6.80
Direct Bilirubin (mg/dL) (♀+♂)[d]	58	0.37	0.88	0.20	0–6.8	0–4.0	0.0–0.0	0.71–6.80
Direct Bilirubin (♀)	30	0.19	0.15	0.20	0–0.6	—	—	—
Direct Bilirubin (♂)	28	0.55	1.24	0.30	0–6.8	—	—	—
Direct Bilirubin (mg/dL) (Annual)[e]	58	0.37	0.88	0.20	0–6.8	0–4.0	0.0–0.0	0.71–6.80
Direct Bilirubin (Summer)	20	0.36	0.23	0.30	0–0.9	—	—	—
Direct Bilirubin (Winter)	38	0.37	1.08	0.20	0–6.8	—	—	—
Indirect Bilirubin (mg/dL)	58	0.07	0.12	0.0	0–0.6	0–0.51	0.0–0.0	0.30–0.60
Total Protein (g/dL) (♀+♂)[f]	59	7.48	1.76	7.30	5.5–20.2	5.75–14.15	5.50–6.40	8.00–20.20
Total Protein (♀)	31	7.13	0.61	7.20	5.5–8.0	—	—	—
Total Protein (♂)	28	7.88	2.44	7.45	6.7–20.2	—	—	—
Albumin (g/dL)	59	4.04	1.40	3.90	3.0–14.3	3.05–9.60	3.0–3.4	4.3–14.30

Table 1.—Continued.

Analyte	n	Mean	SD	Median	Range	Reference Interval	90% CI Lower Limit	90% CI Upper Limit
A/G Ratio (Annual)[g]	59	1.20	0.31	1.10	0.80–2.40	0.80–2.40	0.80–0.90	1.70–2.40
A/G Ratio (Summer)	20	3.73	0.27	3.80	3.0–4.0	—	—	—
A/G Ratio (Winter)	39	1.28	0.34	1.20	0.9–2.4	—	—	—
Calcium (mg/dL) (Annual)[h]	59	10.66	1.28	10.50	9.0–18.9	9.05–15.45	9.0–9.40	11.70–18.90
Calcium (Summer)	20	10.32	0.58	10.20	9.0–11.5	—	—	—
Calcium (Winter)	39	10.84	1.49	10.70	9.1–18.9	—	—	—
Phosphorus (mg/dL)	59	6.01	1.99	5.50	3.20–10.80	3.20–10.55	3.20–3.50	9.75–10.80
Triglycerides (mg/dL)	56	37.9	24.3	31.0	9.0–115.0	9.40–110.3	9.0–10.0	89.1–115.0
Cholesterol (mg/dL)	58	150.0	29.6	150.0	59.0–207.0	68.0–205.6	59.0–109.9	196.4–207.0
Creatinine (mg/dL)	56	1.89	0.67	1.75	0.80–3.60	0.84–3.47	0.80–1.00	3.07-3.60

[a] Seasonal difference ($U_A = 190.5$, $z = 3.19$, $P = 0.001$)
[b] Includes 4 outliers (3 ♀, 1 ♂) with exceedingly high CPK (see text for details)
[c] Outliers removed from sample
[d] Females and males differed ($U_A = 577$, $z = -2.44$, $P = 0.015$)
[e] Seasonal difference ($U_A = 542$, $z = -2.64$, $P = 0.008$)
[f] Females and males differed ($U_A = 556.5$, $z = -2.12$, $P = 0.034$)
[g] Seasonal difference ($U_A = 201.0$, $z = 3.01$, $P = 0.003$)
[h] Seasonal difference ($U_A = 266.5$, $z = 1.97$, $P = 0.049$)

Discussion

Significant differences in direct bilirubin and total protein between males and females reported here may not be biologically meaningful, but are presented for consideration by future investigators. Similarly, significant differences between seasons in direct bilirubin, calcium, CO_2, and A/G ratio may not be biologically meaningful. Nonetheless, these results could have foundations in differences between the sexes in diet or life history strategies (Pierce et al. 2000b, White et al. 2011), and remain open to further inquiry.

Availability of previously published information provided an opportunity to conduct a retrospective comparison of mean values for sodium, potassium, chloride, total bilirubin, phosphorus, cholesterol, and creatine kinase between this investigation and those reported by earlier researchers (Appendix I, Appendix II). Mean level of creatine kinase reported here was nearly identical in value to that for mountain lions bayed by hounds in Florida (Dunbar et al. 1997), but values reported by Currier and Russell (1982) were significantly less than those reported in this investigation or by Dunbar et al. (1997). Further, the mean value for creatine kinase reported by Currier and Russell (1982) did not fall within the reference interval for mountain lions occupying the eastern Sierra Nevada, and may reflect effects of differences in pursuit times (Harlow et al. 1992), ambient conditions (Kozakiewicz et al. 2018), handling protocols (Kock et al. 1987, Zahid et al. 2018), laboratory methods (Duncan et al. 1994), or genetic variation among populations (Yamin et al. 2007, 2008, 2010). Mean values for sodium and cholesterol also differed among the four investigations (Appendix II), and reasons for those differences remain open to further consideration. Mean values for sodium and cholesterol reported by Currier and Russell (1982) or Dunbar et al. (1997) did, however, fall within the reference interval calculated for mountain lions captured in the eastern Sierra Nevada.

Anthropogenic mortality is frequent among mountain lions (Wolfe et al. 2015, Andreasen et al. 2018), and an increase in domestic pets, including house cats, can be expected as human populations expand. This eventuality will lead to greater opportunities for mountain lions to contact humans and their domestic felids (Anderson et al. 2009, Bevins et al. 2012; but, see Carver et al. 2016). Although mountain lions preyed infrequently on domestic cats in the eastern Sierra Nevada (Villepique et al. 2011), we provide baseline reference intervals from a part of California where those iconic carnivores are much less apt to contract pathogens than at the urban-wildland interface, where there is increasing interest in spillover from domestic to wild felids (Foley 1997, Riley et al. 2004, Foley et al. 2013, Carver et al. 2016, Kellner et al. 2016, Kozakiewicz et al. 2018).

As noted by Barnes et al. (2008), baseline data are of importance when investigating the health status of free-ranging wildlife, and health monitoring is necessary to understand and manage threats (Deem et al. 2001). Our results provide reference intervals from a genetically defined population of mountain lions that likely has experienced fewer encounters with domestic cats than at the constantly growing urban-wildland interface, and values reported here can serve as a baseline against which to measure future changes among mountain lions occupying a rural and isolated region of America's most populous state. Indeed, other investigators have reported that prevalence rates of pathogens among mountain lions occupying the eastern Sierra Nevada were among the lowest reported from California (Girard et al. 2012, Foley et al. 2013).

Reference intervals normally are based on values obtained from individual animals.

In this investigation, 5 mountain lions were sampled twice, 3 were sampled 3 times, 1 was sampled 4 times, and 1 was sampled 5 times (median time between repeat captures = 18 months [range 4–38 months]). Thus, population-specific reference values reported here are based on a combination of intra-individual and inter-individual variation (Greffe et al. 2009).

Variation in weather, prey availability and its effect on diet composition, reproductive status, age, and differences in capture methods likely are meaningful representations of variability in the conditions that can affect serum chemistry of individuals (Ellervik and Vaught 2015). Although some mountain lions were sampled more than once and those data were used to calculate the reference values presented, resampling occurred under a variety of ecological settings that likely reflected individual responses to environmental or physiological variability. We acknowledge the potential for "nondemonic intrusion" (i.e., a chance event) that may have affected some individuals (Hurlburt 1984), but consider our use of multiple samples from the same individual to be representative of conditions likely to be encountered by individuals comprising our study population at some point in their lives. Multiple samples from each experimental unit (i.e., the individual mountain lion) can be desirable in that it increases the precision with which properties of each individual are estimated (Hurlburt 1984), and multiple samples from individual mountain lions were included in population-specific reference intervals reported by Currier and Russell (1982) and Dunbar et al. (1997).

The importance of local factors in explaining disease exposure (Carver et al. 2016) and calculations performed ancillary to this investigation support the desirability of obtaining serum chemistry values from mountain lions on local scales that reflect differing ecological settings (Pierce and Bleich 2003), landscape features (Kozakiewicz et al. 2018), or one or more of the numerous stressors currently facing wild felids (Kellner et al. 2018). Rather than assuming that reference values or descriptive statistics from a single location are representative for what historically was the most widely distributed terrestrial mammal in the western hemisphere (Logan and Sweanor 2001), differences among populations are to be expected and do exist (Appendix I, Appendix II). Our results further emphasize the value of detailed sampling at multiple locations and add substantially to the paucity of information on the serum chemistry of wild, free-ranging mountain lions that currently is available.

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We dedicate this paper to the memory of Philip E. (Pep) Partridge in recognition of his prominent role in helping the scientific community understand the ecological relationships among mountain lions, mule deer, and humans in the eastern Sierra Nevada. We thank numerous others and, especially, J. Ostergard, for help capturing mountain lions during our long-term investigations. D. Reed ensured expedient processing of samples at Northern Inyo Hospital (NIH), and N. Barbieri developed and maintained our serum chemistry database. L. Konde and J. Rudd (CDFG) and L. Weber and M. Armstrong (NIH) were instrumental in providing additional helpful information. We thank the CDFG Deer Herd Management Plan Implementation Program, National Rifle Association, Mule Deer Foundation, Safari Club International, Fish and Game Advisory Committee of Inyo and Mono Counties, California Deer Association, and the Institute of Arctic Biology at the University of Alaska Fairbanks for funding, and Northern Inyo Hospital and the University of California White Mountain Research Station for additional support. Manuscript preparation was supported by the Eastern Sierra Center for Applied Population Ecology. This is Professional Paper 111 from the Eastern Sierra Center for Applied Population Ecology.

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Appendix I: Mean and Standard Deviation of Serum Chemistry Values For Wild, Free-Ranging Mountain Lions Inhabiting Disparate North American Ecosystems in Florida, Colorado, and California, USA

	Bleich et al. (this paper)			Currier and Russell (1982)			Paul-Murphy et al. (1994)			Dunbar et al. (1997)		
Analyte	*n*	Mean	SD	*n*	Mean	SD	*n*	Mean	SD	*n*	Mean	SD
Sodium (mEq/L)[a]	59	158.3	40.2	17	144.6	2.42	—	—	—	94	152.6	3.4
Potassium (mEq/L)[a]	59	4.55	1.85	17	4.6	0.32	—	—	—	94	4.6	0.48
Chloride (mEq/L)[a]	59	123.3	27.3	17	114.2	4.84	—	—	—	94	115.5	4.3
CO_2 (mEq/L)[b]	59	16.4	6.4	—	—	—	19	12.53	1.75	94	14.33	4.00
Glucose (mg/dL)[b]	59	144.9	69.1	—	—	—	19	110.6	37.3	94	154.4	51.0
Blood Urea Nitrogen (mg/dL)[b]	59	42.3	17.4	—	—	—	19	32.9	6.4	94	37.7	14.1
Creatine Kinase (U/L)[a]	52[c]	554.3[c]	336.9[c]	17	108.0	55.75	—	—	—	88	515.1	415.1
SGPT (U/L)[b]	59	77.9	54.3	—	—	—	19	60.2	33.4	94	60.2	35.0
GGT (U/L)[a b]	57	9.5	4.1	—	—	—	—	—	—	80	1.6	1.4
SGOT (U/L)[a d]	—	—	—	17	68.0	22.09	—	—	—	94	73.4	77.8
Alkaline Phosphatase (U/L)	59	37.3	30.0	—	—	—	19	22.6	11.3	94	35.4	38.6
Total Bilirubin (mg/dL)	58	0.47	0.88	17	0.3	0.11	19	0.30	0.25	94	0.26	0.61
Direct Bilirubin (mg/dL)[a b e]	58	0.37	0.88	—	—	—	—	—	—	—	—	—
Indirect Bilirubin (mg/dL)[a b e]	58	0.07	0.12	—	—	—	—	—	—	—	—	—
Total Protein (g/dL)[b]	59	7.48	1.76	—	—	—	19	6.58	0.67	93	7.37	0.67
Albumin (g/dL)[b]	59	4.04	1.40	—	—	—	19	3.13	0.32	94	3.70	0.36
A/G Ratio[a b e]	59	1.20	0.31	—	—	—	—	—	—	—	—	—
Calcium (mg/dL)[b]	59	10.66	1.28	—	—	—	19	9.53	0.66	94	9.92	0.66
Phosphorus (mg/dL)	59	6.01	1.99	17	5.6	1.58	19	5.66	1.15	94	5.77	1.51
Triglycerides (mg/dL)[a b]	56	37.9	24.3	—	—	—	—	—	—	85	54.9	103.4
Cholesterol (mg/dL)	58	150.0	29.6	17	167	2.41	19	155.1	29.9	94	147.9	26.9
Creatinine (mg/dL)[b]	56	1.89	0.67	—	—	—	19	2.05	0.45	94	1.84	0.54

[a] Not reported by Paul-Murphy et al. (1994)
[b] Not reported by Currier and Russell (1982)
[c] Four outliers excluded (see text for explanation)
[d] Not reported by Bleich et al. (this paper)
[e] Not reported by Dunbar et al. (1997)

Appendix II: Pairwise Comparisons Of Serum Chemistry Values

Results of pairwise comparisons of mean serum chemistry values from populations of free-ranging mountain lions captured in widely disparate ecosystems (at sea level in Florida [Dunbar et al. 1997], exclusively at high elevation in the Rocky Mountains in Colorado [Currier and Russell 1987], and at intermediate elevations in the eastern Sierra Nevada in California [this paper]). A fourth group (Paul-Murphy et al. 1994) included mountain lions captured among a multitude of ecological systems in California. Two-tailed P-values for differences in means were calculated from data in Appendix I using Welch's Approximate t, which is robust to considerable departures from theoretical assumptions when two-tailed tests are employed and samples are large (Zar 1984); significant differences are indicated in bold font.

		Source		
Analyte	Source	Bleich et al. (this paper)	Currier and Russell (1982)	Paul-Murphy et al. (1994)
Sodium[a] (mEq/L)	Currier and Russell (1982)	**0.012**		
	Paul Murphy et al. (1994)			
	Dunbar et al. (1997)	0.281	**<0.001**	
Potassium[a] (mEq/L)	Currier and Russell (1982)	0.843		
	Paul Murphy et al. (1994)			
	Dunbar et al. (1997)	0.839	>0.999	
Chloride[a] (mEq/L)	Currier and Russell (1982)	0.095		
	Paul Murphy et al. (1994)			
	Dunbar et al. (1997)	0.143	0.312	
Creatine Kinase[a] (U/L)	Currier and Russell (1982)	**<0.001**		
	Paul Murphy et al. (1994)			
	Dunbar et al. (1997)	0.544	**<0.001**	
SGOT[a b] (U/L)	Currier and Russell (1982)			
	Paul Murphy et al. (1994)			
	Dunbar et al. (1997)		0.578	
Total Bilirubin[a] (mg/dL)	Currier and Russell (1982)	0.157		
	Paul Murphy et al. (1994)	0.192		
	Dunbar et al. (1997)	0.114	0.560	0.640
Phosphorus (mg/dL)	Currier and Russell (1982)	0.382		0.898
	Paul Murphy et al. (1994)	0.348	0.898	
	Dunbar et al. (1997)	0.429	0.685	0.722
Cholesterol (mg/dL)	Currier and Russell (1982)	**<0.001**		0.101
	Paul Murphy et al. (1994)	0.523	0.101	
	Dunbar et al. (1997)	0.661	**<0.001**	0.340

[a] Not reported by Paul-Murphy et al. (1994)
[b] Not reported by Bleich et al. (this paper)

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Book Review

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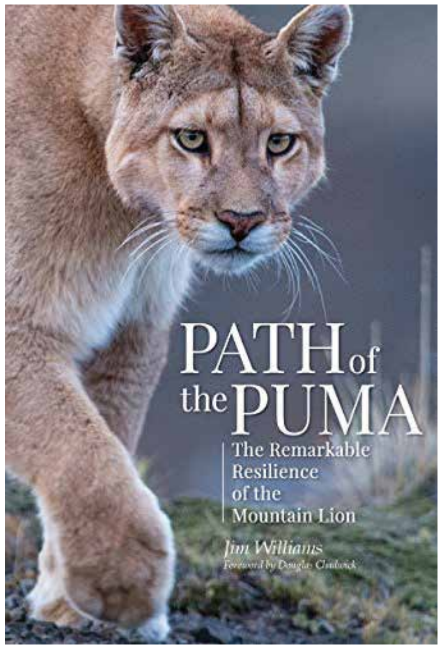

Jim Williams, an employee of Montana Fish, Wildlife and Parks for nearly 30 years, has written a very informative and entertaining summary of his career and his convictions as they relate to wildlife conservation in North and South America. As indicated by the title, the species of primary interest is the mountain lion (*Puma concolor*), but the conservation ethic espoused by Williams clearly extends to all species, the importance of habitat to their continued persistence, and the value of intact and functional ecosystems. Jim's concerns extend from the near-arctic reaches of North America to the southern part of South America, which encompasses the geographic range of what he refers to throughout the book as 'America's lion'.

The first half of the book is based largely on personal experiences and lessons learned during his career in wildlife conservation and management. Rather than being a detailed treatise on the ecology of mountain lions, Williams takes the reader through a history of his involvement with conservation, culminating with an emphasis on the need for management practices that will ensure the persistence of the mountain lion across its range. Williams has 'walked the walk' for nearly 3 decades and, thus, is well-qualified to 'talk the talk' of management and conservation, and the importance of both to the persistence of wild things and the habitat upon which they depend. He does this in a series of 21 chapters, each of which could be described as a stand-alone essay, and each of which is based on experiences and knowledge gained during a life-long career in conservation.

The first several chapters touch on personal aspects of Williams' career, beginning with an aerial observation of a mountain lion pursuing a bighorn sheep (*Ovis canadensis*) and the subsequent capture of that lion as part of an extensive investigation of predators and prey (Enk 1999, Enk et al. 2001). Chapter 2 describes his early life and interest in nature, a move from Iowa to California, school, first job as a professional, and his desire to enter a graduate program at Montana State University (MSU), where he successfully completed his graduate degree while investigating mountain lion ecology (Williams 1992). In Chapter 3, Jim provides detailed snippets of several events experienced while attending MSU, including insight into the politics of wildlife management in a rural, small-town setting, a subject that I gained a special appreciation for during my own career in wildlife conservation.

Conservation of individuals, habitat, and population connectivity are subjects addressed in chapters 4 and 5, where Williams emphasizes the value of each in easily understood language. Chapter 6 is dedicated to a description of the wildlands of northwestern Montana, an area referred to as the Crown of the Continent and, clearly, a region that has had a profound impact on Jim's thinking, his career, and his lifestyle. The near extirpation of the mountain caribou, an apparent ecotype of woodland caribou (*Rangifer tarandus caribou*), efforts to translocate mountain caribou for conservation purposes, the potential role of predation in efforts to ensure the persistence of that taxon in the absence of habitat loss, and the expansion of mountain lions from their then current range ("… but what struck me most was how far these lions had traveled east") are discussed in the next chapter.

I can relate well to the experiences that Williams describes in Chapter 8 (Locals Only) and Chapter 9 (Suburban Lions). Indeed, we each have experienced the sometimes-uncomfortable situations that can develop during town-hall meetings held in small, rural communities where residents, already suspicious of government, frequently resent any regulatory change that might impact hunting opportunity. Jim was addressing changes needed to ensure the persistence of mountain lions, and I was addressing changes deemed necessary to correct unacceptably low ratios of male to female mule deer. In both cases, however, public trust and understanding prevailed and, as noted by Williams, "… hunting isn't always about the science. Sometimes, it's about social license—about having the people's support for wild nature…". Williams goes on to emphasize that hunters and anglers may provide more practical support for the conservation of habitat and wildlife than all the dedicated conservation groups combined, an extremely important, albeit often-ignored, contention. And, Jim and I both have dealt with livestock depredation and potential human safety concerns associated with mountain lions. In Montana, a zone management strategy was adopted to minimize the potential for mountain lions to 'misbehave'; in California, a voter initiative banned the take of any mountain lion unless it already had killed livestock, a pet, or a human, or was perceived to be an imminent threat to public safety, and is a law that is unlikely ever to be changed (Bleich and Pierce 2005).

Beginning with chapter 10, the remainder of the book addresses conservation issues in the southernmost part of the mountain lion's geographic distribution. In my opinion, this is the most interesting part of the work, albeit largely because of my lack of familiarity with that part of South America known as Patagonia. Williams describes the beginnings of his involvement with conservation in that area and emphasizes the personal roles that well-intentioned individuals have played in attempting to piece back together a system that, previously unbeknownst to me, has been heavily fragmented by anthropogenic development and agricultural activities. He describes in some detail the ecological relationships between predators and prey, the vast stretches of grasslands and steppes, and the geography of this fascinating region. Prior to reading *Path of the Puma* I knew little of this vast area, the ecological problems that prevail, or the ongoing efforts to conserve what remains of a functional ecosystem and to restore what can be restored.

Patagonia has been ecologically damaged, not only by anthropogenic development, but also by the presence of exotic species that have altered the primary prey base of mountain lions and other carnivores in that part of the world. Indeed, following its introduction the European hare (*Lepus europaeus*) has spread largely across the southern half of South America (Bonino et al. 2010), and in so doing has become an important source of protein for the native canids, as well as other native felids that are themselves the subject of a separate chapter. Further, the introduction and spread of red deer (*Cervus elaphus*) has contributed to further alteration of the landscape, and domestic sheep and cattle, while altering the landscape, have largely replaced the native guanaco (*Lama guanacoe*) as the primary native grazer and formerly the most important prey of the mountain lion. Moreover, the desire to eliminate predation on domestic ungulates has played a major role in the ecology and status of mountain lions.

The presence of vast ranches owned by sometimes willing sellers or individuals primarily interested in conservation, when combined with the financial means, efforts, and persistence of others dedicated to the restoration of intact ecosystems, is gratifying and are examples of some of the successes that Jim Williams has been instrumental in facilitating. Indeed, Jim has been an important player in ongoing efforts to 'rewild' portions of Patagonia and ensure a future for America's lion in that part of the world, and through *Path of the Puma* he is sharing those ventures with others.

This book contains much about natural history, ecology, and conservation. Jim includes a chapter on wild felids that share portions of the landscape with mountain lions, and touches on the plight of the huemul (*Hippocamelus bisulcus*), an endangered South American cervid. Perhaps more meaningful, however, he includes information on the ways that some of the local populace that formerly made a living as *leoneros* (those that hunted lions to protect domestic livestock) have become involved in conservation and are now contributing to efforts to better understand and conserve the remaining ecological integrity of Patagonia. And, the many scientists, wealthy benefactors, and visionary conservationists dedicated to this goal are recognized for their efforts toward that end.

Williams makes a several generalizations, some of which I question—at least in the context presented—or appear to be contradictory and include statements regarding prey images of mountain lions (pp. 44, 60, 101; but see p. 122), and population regulation (p. 135, but see pp. 65, 98, 150; see also Pierce et al. 2000). There are other generalizations with which I strongly concur, however, including the advantages of maintaining mature males in harvested populations (p. 136), the need to minimize the presence of mountain lions in suburban neighborhoods (p. 148) and, especially, Jim's insistence that habitat protection must be the primary conservation focus (pp. 122, 132). Moreover, the importance of an understanding of natural history to conservation is emphasized through his admonition (p. 40) that, "… unless you're there, in the field, you miss the relationships that make nature work—the weather and the wind and the topography and the light that can explain *why* a cat was in a particular place at a particular time" is paramount advice to researchers but, unfortunately, is a topic that has been largely de-emphasized in wildlife curricula of late (Bleich and Oehler 2000, Bleich 2018).

The book is very well written and contained surprisingly few errors. The few minor glitches that were noted (e.g., use of lowercase initial letters for genera [p. 23], missing letters in words [pp. 58, 127], occasional misspellings of scientific names [pp. 60, 75], inconsistent spelling of surnames [pp. 137, 139], or inclusion of extraneous words [p. 284]) do not detract from the value of the text. I do believe an index would have been helpful, as would have been the inclusion of appropriate literature citations and a more detailed map of Patagonia.

As noted by Maurice Hornocker, "[*Path of the Puma*] … is a prime example of Jim

Williams' dedicated effort to inform and enlighten a broad audience on the ecological and cultural importance of this charismatic apex carnivore". From my perspective, Jim has been successful in doing so and those that are unfamiliar with a vast portion of the range of the mountain lion, as was I, will benefit especially from the information contained in the text.

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Information for Contributors

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Front.—Great Plains Toad (*Anaxyrus cognatus*). Photo by Gary Nafis, CaliforniaHerps.com CC BY-NC-ND 3.0.

Back.—Mountain lion (*Puma concolor*). CDFW file photo.

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