NATURAL HISTORY NOTE

Hind Limb Malformation in the Foothill Yellow-legged Frog, Sonoma County, California

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Physical malformations in amphibians have been recently reported (Sessions and Ruth 1990, Douran et al. 1998, Alvarez et al. 2021, Alvarez et al. 2023a, 2023b), and more specifically anurans have shown physical anomalies that originate from parasites (Tidd 1962, Johnson and Sutherland 2003, Kupferberg et al. 2009). Lotic habitats appear to support anurans that show malformations (Johnson and Chase 2004). However, Kupferberg et al. (2009), found that the copepod *Lernaea cyprinacea* can parasitize larval forms of foothill yellow-legged frogs (*Rana boylii*) in a lentic environment, the South Fork of the Eel River, Mendocino County, California, which led to malformations of the eyes, snout, and limbs.

The trematode parasite *Ribeiroia* spp. is also known to infect anuran larvae in California (Johnson et al. 2013, 2019) and appears to cause malformations during transition from larval to frog life stages (Johnson et al. 2002, 2013, 2019). Typically, these malformations occur in lentic habitats that are eutrophic, such as cattle stock ponds and marshes, that support the planorbid snail (family Planorbidae), which serves as the primary intermediate host for the parasite (Johnson and Chase 2004, Budria and Candolin 2014, JAA and DGC pers. obs.). Anurans are the secondary intermediate host and malformations occur most commonly at the developing limbs where the parasite enters the body (Johnson et al. 2002, 2019).

Here we describe a malformation of a juvenile foothill yellow-legged frog. Although extensive investigations have not occurred, the increasing reports of malformations in the species suggest that this phenomenon may be prevalent. This is of particular concern because the foothill yellow-legged frog is in decline, which has resulted in protections under the California and federal Endangered Species Acts (Patterson 2019, USFWS 2023).

Typically, foothill yellow-legged frogs utilize lotic habitats with cool, clear-water conditions (Storer 1925, Zweifel 1955), and only rarely occur in eutrophic, lentic habitats (Wilcox and Alvarez 2019). Therefore, we would anticipate the number of locations and frequency of occurrence of malformations of amphibians in lotic and oligotrophic conditions to be relatively low. Nevertheless, Alvarez et al. (2021) reported an idiopathic malformation of the hind limb of a post-metamorphic foothill yellow-legged frog in a tributary to Sonoma Creek in Sonoma County, California. Although we expected this to be an anomalous observation, we report a limb malformation in a post-metamorphic foothill yellow-legged frog from the mainstem Sonoma Creek, Sonoma County, California.

On 27 and 28 October 2023, we visually searched a 100 m section of Sonoma Creek, at an elevation of 400 m, from a small picnic area to an open creek crossing, near the headwaters. This site was located

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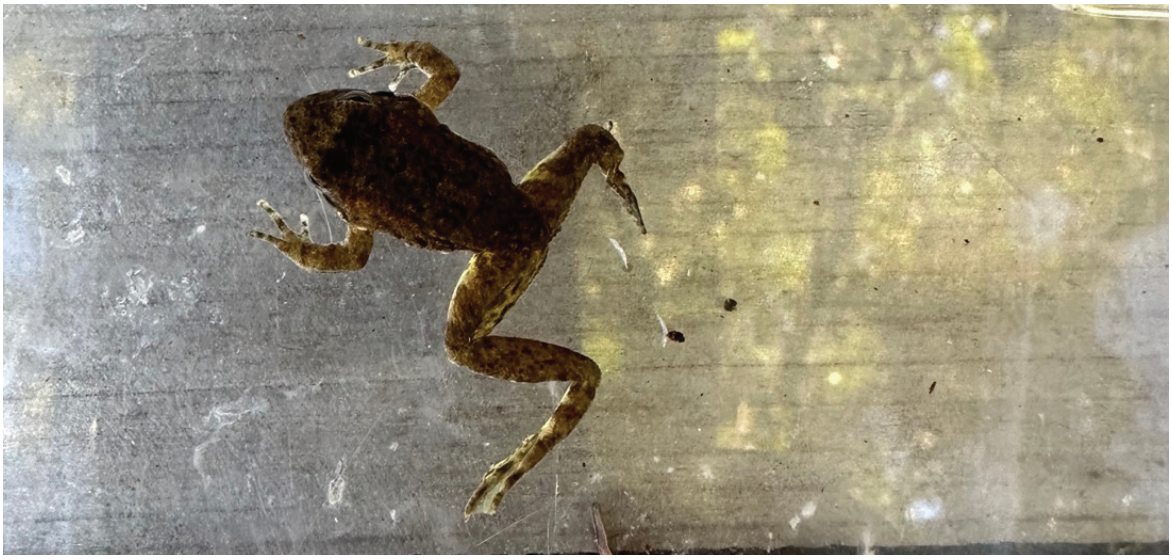

Fig. 1. A post-metamorphic foothill yellow-legged frog (*Rana boylii*), resting in water in a clear bin, showing a right rear malformed leg. Photo by authors.

Fig. 2. Magnified view of the malformed limb showing extreme reduction in the upper and lower leg, as well as the foot and toes of a post-metamorphic foothill yellow-legged frog. Photo by authors.

approximately 1-km upstream of the tributary confluence where a foothill yellow-legged frog malformation was reported four years earlier by Alvarez et al. (2021). This portion of the creek is perennial, with numerous small pools and short riffles and runs, and a substrate of gravel, cobble, and occasional boulders. The riparian area was dominated by a dense canopy of white alder (*Alnus rhombifolia*), California bay (*Umbellularia californica*), Oregon ash (*Fraxinus latifolia*), and coast live oak (*Quercus agrifolia*), with an understory of poison oak (*Toxicodenron pubescens*), California blackberry (*Rubus ursinus*), sedges (*Carex* spp,), and cow parsnip (*Heracleum lanatum*). The surrounding uplands were undeveloped and used for passive recreation. The portion of Sonoma Creek that we focused on was 1–2 m wide, approximately 15 cm deep, and had occasional pools as deep as 1 m. Water conditions were flowing, clear, and cool (approximately 11–13º C), with filamentous algae (Charophyta) along the shore. Syntopic with foothill yellow-legged frogs were California giant salamanders (*Dicamptodon ensatus*), western toad (*Anaxyrus boreus*), and Pacific Chorus Frog (*Pseudachris regilla*), all of which appeared typical, and showed no sign of malformations.

We hand-captured five foothill yellow-legged frogs (one adult and four post-metamorphic juveniles). All of the frogs were placed in a clear plastic bin for inspection. The adult and three of the post-metamorphic frogs appeared to be morphologically and behaviorally normal. The fourth post-metamorphic foothill yellow-legged frog was observed along the creek bank and when we attempted to capture the frog, we noticed it moved slowly and jumped in an atypical fashion. Once placed in the examination bin, this frog hopped in small circles. Upon examination the right hind limb appeared to be malformed. We noted that the upper and lower leg, foot, and toes (i.e., femur, tibia, fibula, astragalus, calcaneum, and phalanges) were all reduced by approximately 60–70% (visually estimated), and surrounding musculature was markedly reduced or missing (Fig. 1 and 2). We closely examined the frog for any atypical growths, parasites, and/or injuries and found none. This malformation appeared morphometrically similar to the malformation observed by Alvarez et al. (2021) from the same creek system, including the same limb. We released the frog after the examination.

Our observation of a malformed foothill yellow-legged frog is the third report of malformations in this species (Kupferberg et al. 2009, Alvarez et al. 2021). However, this report stands out in that this is the second account in the same creek system, showing very similar malformations. In addition, the area in the report, along with Alvarez et al. (2021), is surrounded by undeveloped lands that would not provide nutrient inputs that typically cause eutrophication and the prevalence of parasitic *Ribeiroia*. This allows the possibility that a parasite that inhabits oligotrophic streams may be the cause of the observed malformation, such as copepod *Lernaea cyprinacea* reported by Kupferberg et al. (2009). To our knowledge, *Ribeiroia* spp. or *Lernaea cyprinacea* have not been reported from upper Sonoma Creek watershed. We did not determine

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the cause of the malformation we observed, and we acknowledge that other agents may be contributing to these malformations.

We contend that two observations of a similar malformation should elicit some effort to assess the rate of malformation in this system. A creek-wide survey during the post-metamorphic period could help to determine if these malformations are part of a pattern or simply coincidental. Due to the recent declines of some populations of the foothill yellow-legged frog in California (Patterson 2019), and the recent listing of the species as threatened or endangered by both State and federal resource agencies (Title 14, Section 670.1 [2020], 86-FR-73914 [2023]), we recommend further investigations into the possible causes and impacts of malformations in this species.

Acknowledgments—We thank C. Shaffer for providing a California State Parks collecting permit for the workshop that was conducted at this site. Was also thank T. Wagner, who assisted in the field. Field surveys were conducted under the auspices of California Department of Fish and Wildlife.

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