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Distribution of tadpoles of *Ollotis occidentalis* (Amphibia: Anura: Bufonidae) along the Río Salado, Puebla, México

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Abstract. We examined monthly variation in the characteristics of river sections along the Río Salado (Puebla, Mexico) and how these factors were associated with the distribution of the tadpoles of the toad *Ollotis occidentalis*. Tadpoles were observed in the river in March 2007, and from November 2007 through February 2008 and were only found in the main river channel. Sections of the Río Salado with tadpoles were deeper, wider, and longer than sections without tadpoles. Dissolved oxygen levels were higher and salinity was lower in river sections with tadpoles compared to the sections without tadpoles. There was no difference in temperature between sections with and without tadpoles. Tadpoles were found in river sections that contained more vegetation than river section without tadpoles. Our results suggest that the distribution of tadpoles of *O. occidentalis* is related to the permanence of water, the chemical nature of the water, and the presence of vegetation.

Keywords. Dissolved oxygen, *Ollotis occidentalis*, pool size, salinity, tadpoles.

INTRODUCTION

One of the major goals of ecology is to understand what determines species distributions. For pond-, pool-, or stream-breeding amphibians the relevant question is often “Why are tadpoles found in some locations but not others?” In some cases, the distribution of amphibians appears driven by the chemical or physical characteristics of the pool, pond, or stream (e.g., Eason and Fauth, 2001; Richards, 2002; Rocha et al., 2002; Halverson et al., 2003; Welch and MacMahon, 2005; Girish and Krishnamurthy, 2009). In other cases, it appears that the distribution can be explained by the biotic community (e.g., Blaustein and Margalit, 1995; Eason and Fauth, 2001). In addition, the interaction of the chemical or physical environment with the biotic environment can potentially determine the success of amphibian larvae, and thus could affect their distribution (e.g., Sadinski and Dunson, 1992; Warner et al., 1993; Skelly, 1996; Smith et al., 2006).

We investigated the distribution of the tadpoles of *Ollotis* [*Bufo*] *occidentalis* along the Río Salado (Puebla, Mexico) which, as its name suggests, can have relatively high salinity. Amphibians are typically not found in saline conditions, and are usually thought to be unable to withstand elevated salinities (Ultsch et al., 1999). However, several species of amphibians (*Exerodonta xera, Hyla arenicolor, Lithobates spectabilis*, and *O. occidentalis*) are known to occur in the Río Salado. We examined monthly variation in water chemistry (salinity, dissolved oxygen), water temperature, physical characteristics (size, distance from main channel), and vegetative cover of river sections and we evaluated how these factors differed between sections of the Río Salado with and without tadpoles of *O. occidentalis*. *Ollotis occidentalis* occurs from south-central to northern Mexico. In southern Puebla, *O. occidentalis* occurs in pine forest and fry tropical deciduous forests (Oliver-López et al., 2000). In the Valle de Zapotitlán Salinas, *O. occidentalis* breeds during the rainy season in slow moving water or in pools on the edges of rivers (Oliver-López et al., 2000; Smith and Lemos-Espinal, 2010). Very little is known about the ecology and natural history of this toad beyond reports on its reproduction (Duellman, 1961; Oliver-López et al., 2000; Oliver-López and Ramírez-Bautista, 2002; Smith and Lemos-Espinal, 2010).

MATERIALS AND METHODS

Study Area

The Río Salado (see Fig. 1) runs through El Valle de Zapotitlán Salinas in southeastern Puebla, Mexico (see Woolrich Piña et al. 2005 for additional details). Pools along the Río Salado are seasonal, forming during the dry season as the water level in the Río Salado falls. El Valle de Zapotitlán is part of the Valle de Tehuacan-Cuicatlán that lies in central Mexico, which is considered to be an ecologically important region due to its high levels of biodiversity and endemism (Dávila-Aranda et al., 1993). Surveys of the herpetofauna have reported between 22 and 32 species of amphibians and reptiles (Martín del Campo and Sánchez-Herrera, 1979; Canseco-Márquez and Gutiérrez-Mayén, 1996). More recently, there have been studies examining the effects of habitat deterioration on amphibians and reptiles (Mata-Silva, 2000) and the reproduction of the anurans in the Valle de Zapotitlán Salinas (Oliver-López et al., 2000; Oliver-López and Ramírez-Bautista, 2002). Woolrich Piña et al. (2005) provide a summary of the amphibians and reptiles of El Valle de Zapotitlán Salinas.

Methods

We conducted surveys along a 2 km segment of the Río Salado (Fig. 1) monthly from February 2007 to June 2008 to determine the distribution of tadpoles of *O. occidentalis* and to characterize conditions along the Río Salado. The tadpole of this species could be actually differentiated from those of other co-occuring species in the field. We surveyed the first 10 – 19 sections of river or isolated pools we encountered on each of our surveys. We generally began the surveys in the same location and proceeding in the same direction along the river and so many of the river sections were similar among visits; however, on some visits the starting location and direction moved varied

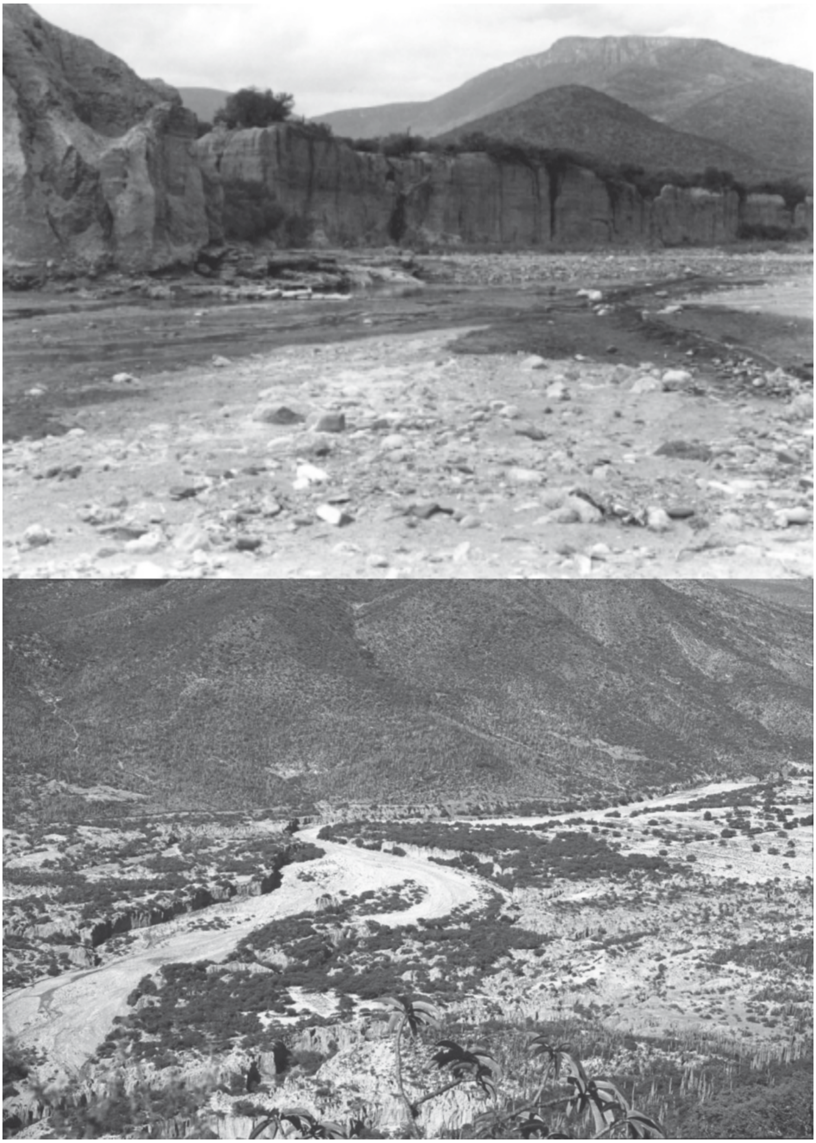

Fig. 1. Photographs of the study site along the Río Salado, Puebla, Mexico. Note that the close-up photograph was taken during one of the wetter months of the year.

(e.g., when different researchers conducted a monthly survey). In addition, because isolated pools dried up during some parts of the year, the selected isolated pools may have varied. Our impression is that while selection of river sections and isolated pools may have caused some of the variation observed among months, we do not believe this to be a major factor in explaining monthly variation since there appeared to be much more homogeneity among sites within monthly samples than among monthly samples.

Surveys took place in the morning hours (e.g., 0800 to 1200 h). Each river section (identified as a single continuous length of river of similar water flow, either a pool with still water or a section of river with greater flow between two pools) or isolated pool (a pool without direct connection to the main channel of the Río Salado) was visually searched for the presence of larval amphibians (the river sections were small enough, and the water clear enough and shallow enough to allow for the easy detection of any tadpoles). All river sections were contiguous with, or in the case of isolated pools, parallel to, the main river channel of the river (i.e., we did not skip sections of the river during our surveys).

We measured the distance between each isolated pool and the main channel of the Río Salado. The main channel consisted of the primary path of the Río Salado and which contained water throughout its length and throughout the year. For surveyed section in the main channel, this distance was recorded as 0 m. We also measured the physical dimensions (length and width) of each river section. For each sampled river section or isolated pool we measured maximum depth, salinity, temperature, dissolved oxygen, and pH. We measured water chemistry and temperature approximately 20 – 30 cm from the river's edge and at a depth of 10 – 20 cm. Salinity, dissolved oxygen, and temperature were measured using a YSI Model 85 Handheld DO/conductivity meter. We used DF Universal Indicator Paper pH 1-14 to measure pH. In addition, we visually estimated the percent cover of aquatic vegetation in each pool.

To examine the differences in the characteristics of river sections where we found tadpoles and sections where we did not find tadpoles, we used a nested MANOVA with month and tadpole presence (nested within month) as the independent variables and the physical and chemical parameters as the dependent variables. We limited this analysis to the November 2007 to February 2008 surveys (the months when tadpoles were present; see below). A significant MANOVA was followed by univariate nested ANOVAs to examine each dependent variable in greater detail and to determine which of the measured variables differed over time and between sections with and without tadpoles. Due to the dynamic nature of the Río Salado (drying of pools and changes in water flow), we were not able to follow specific river sections from month to month, especially pools isolated from the main river channel (see above). We, therefore, could not use repeated measures ANOVAs.

RESULTS

Monthly variation

Depth of the river sections varied among months, with the lowest values found in August 2007 and an increase from this low through June 2008, with a temporary decrease in March 2008 (Fig. 2A). Width of the river sections also differed among months (Fig. 2B), as did the length of river sections (Fig. 2C). Width and length were both higher early in the study period (e.g., March to August 2007) and lower from September 2007 through June 2008.

The distance of a river section or isolated pool to the main river channel varied among months, with peaks in July and August 2007 and December 2007 (Fig. 2D). Dissolved oxygen was fairly constant but showed distinctly low values in June 2007, and February to June 2008 (Fig. 2E). Salinity showed a significant trend to higher values throughout the study period (Fig. 2F). Water temperature varied significantly among months (Fig. 2G). The amount of vegetative cover in sections was low for most of the study period with distinct periods of high vegetative cover in March 2007 and from November 2007 to February 2008 (Fig. 2H).

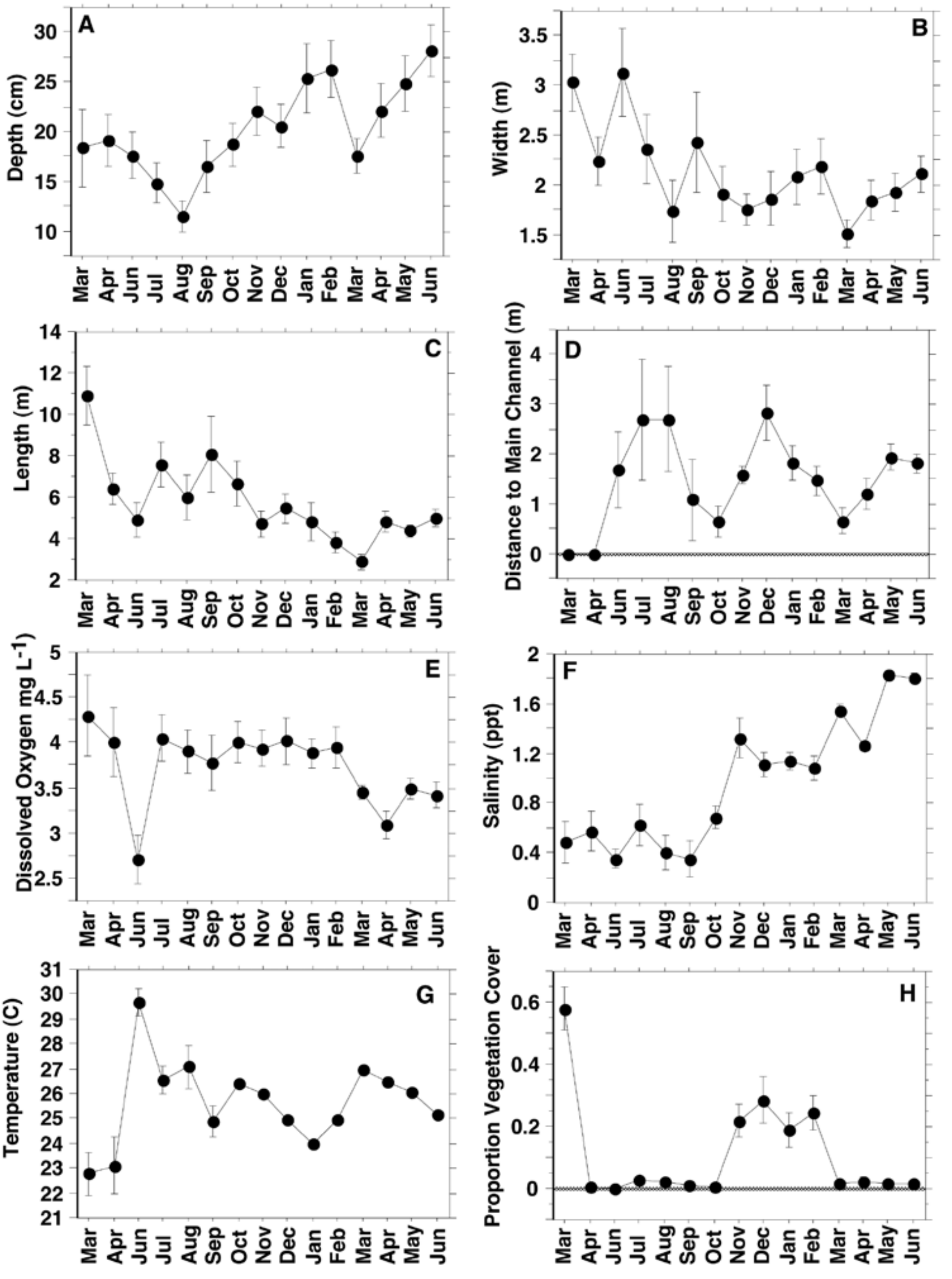

Fig. 2. Variation in characteristics of surveyed river sections throughout the study period (February 2007 to June 2008) along the Río Salado. Means are given ± 1 SE.

Comparison of sections with and without tadpoles

Tadpoles were observed in the river in March 2007 and from November 2007 through February 2008 (Fig. 3). To further investigate the factors that might influence the distribution of tadpoles in the Río Salado, we limited our analyses to the November 2007 to February 2008 surveys (i.e., a single contiguous breeding season).

The nested MANOVA found significant differences in the river sections where we found tadpoles and sections where we did not find tadpoles (Wilks' $\lambda = 0.005$, $F_{32,186} = 18.7$, $P < 0.0001$). There was also a significant month effect (Wilks' $\lambda = 0.009$, $F_{24, 146} = 24.84$, $P < 0.0001$).

Water depth varied among months (Fig. 4A; $F_{3,57} = 3.96$, $P = 0.012$), and tadpoles were found in deeper river sections (Fig. 4A; $F_{4,57} = 24.3$, $P < 0.0001$).

Width of the river sections did not vary significantly from month to month (Fig. 4B; $F_{3,57} = 1.41$, $P = 0.25$). Sections of the river where tadpoles were found were wider than those lacking tadpoles (Fig. 4B; $F_{4,57} = 5.98$, $P = 0.0004$).

River section length did not change from month to month (Fig. 4C; $F_{3,57} = 1.17$, $P = 0.33$). Sections with tadpoles were longer than sections without tadpoles (Fig. 4C; $F_{4,57} = 5.88$, $P = 0.0005$).

Distance to the main channel varied from month to month (Fig. 4D; $F_{3,57} = 6.39$, $P = 0.0008$). Sections of the river that contained tadpoles were closer to the main river channel than sections that did not contain tadpoles (Fig. 4D; $F_{4,57} = 17.0$, $P < 0.0001$). In fact, no tadpoles were observed in sections that were not along the main river channel.

Dissolved oxygen levels remained relatively constant among months (Fig. 4E; $F_{3,57} = 0.4$, $P = 0.75$). Dissolved oxygen levels were higher in river sections where we found tadpoles compared to the sections without tadpoles (Fig. 4E; $F_{4,57} = 42.8$, $P < 0.0001$).

Salinity varied among months (Fig. 4F; $F_{3,57} = 7.85$, $P = 0.0002$). Tadpoles were found in river sections that had significantly lower salinity than river sections where no tadpoles were found (Fig. 4F; $F_{4,57} = 360.2$, $P < 0.0001$). The difference in salinity between sections with and without tadpoles was highest in November 2007 and then stabilized from December 2007 to February 2008 (Fig. 4F).

Temperature showed a clear variation among months with a low in January 2008 (Fig. 4G; $F_{3,57} = 942.3$, $P < 0.0001$). The temperature of sections in which we found tadpoles

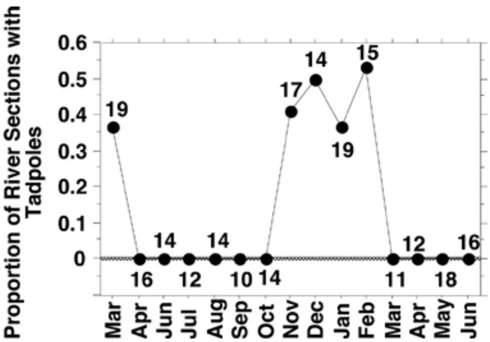

Fig. 3. Proportion of surveyed river sections with tadpoles during monthly surveys of the Río Salado from February 2007 to June 2008. Number of surveyed river sections is provided for each survey.

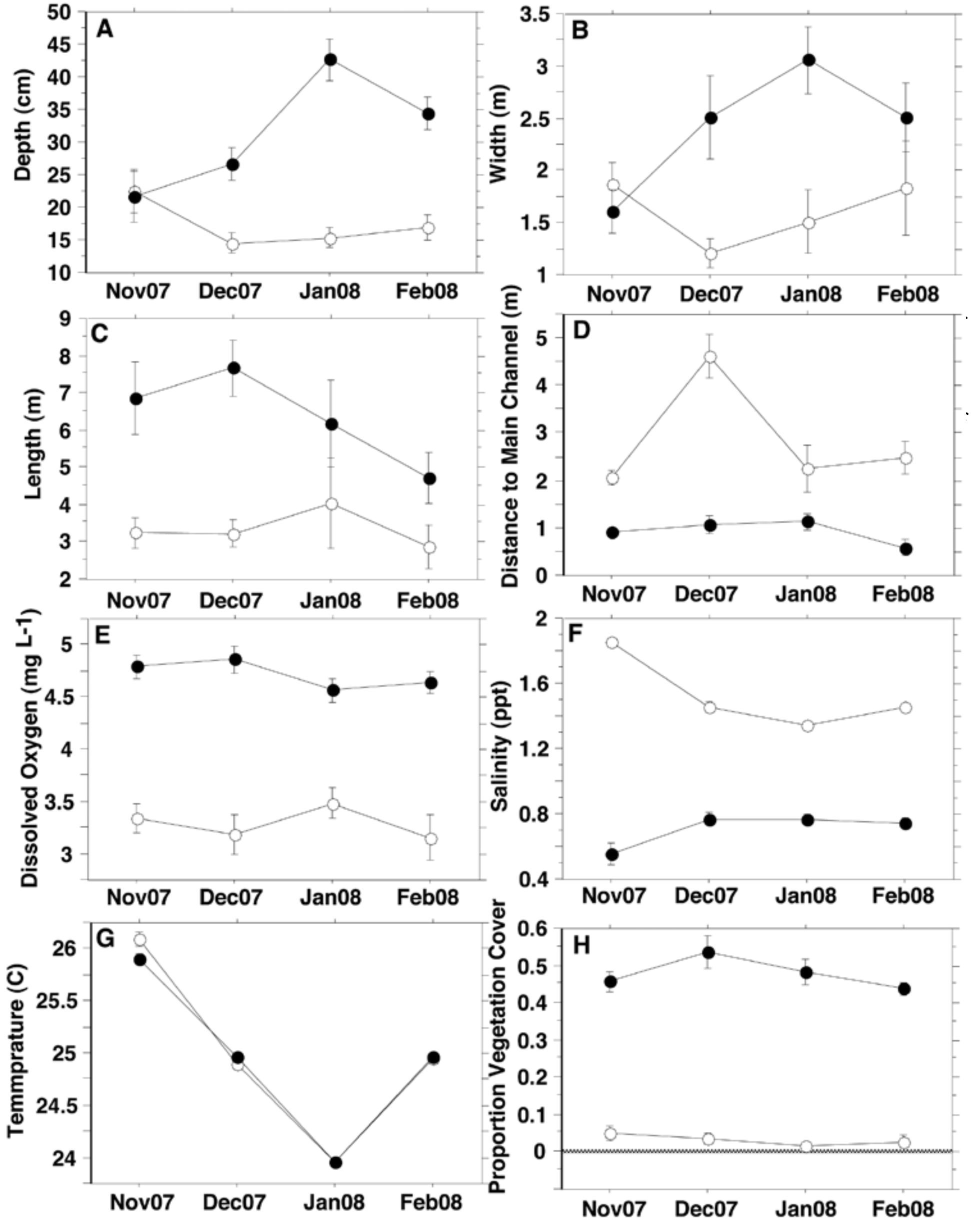

Fig. 4. Mean characteristics of river sections of the Río Salado with (closed circles) and without (open circles) tadpoles. Means are given ± 1 SE.

are also known to be less prevalent in areas with higher salinity (e.g., Davenport and Huat, 1997; Smith et al., 2007) or to avoid oviposition in water with higher salinity (Haramura, 2008). However, salinity did not affect the distribution of *Buergeria japonica* tadpoles

(Haramura, 2007). In contrast, the abundance of *Rhinella marina* increased with salinity in introduced populations on Puerto Rico (Ríos-López, 2008). It appears that at least some species of bufonids show local adaptation to salinity, and are able to tolerate higher levels of salinity as tadpoles (e.g., Gómez-Mestre and Tejedo, 2003, but see Beebee, 1985).

We also found dissolved oxygen levels were higher in areas of the river with tadpoles than in areas of the river without tadpoles. This is consistent with previous results suggesting that *Anaxyrus terrestris* tadpoles use microhabitats with higher levels of dissolved oxygen (Noland and Ultsch, 1981). Dissolved oxygen also appears to be important in determining species richness of tadpoles in ponds along the Middle Paraná River in Argentina (Peltzer and Lajmanovich, 2004).

Tadpoles also used sections of the river with higher levels of vegetative cover. For the most part, the vegetation consisted of algae, and thus this relationship of tadpole presence and vegetative cover could reflect greater food availability for the tadpoles. Alternatively, the presence of more vegetative cover could simply reflect better quality water (i.e., lower salinity, higher dissolved oxygen content, greater permanence).

Given the apparent importance of water permanence and water chemistry in determining the distribution of tadpoles of *O. occidentalis* in the Río Salado, it is worth examining the potential conservation implications of the salt factories ("*salineras*") that are located along the Río Salado, including just upstream of our study area. These *salineras* divert water from the Río Salado to harvest the salt by evaporation. Thus these *salineras* have the potential to greatly alter both the amount of water and the chemistry of the water of the Río Salado. Such alterations may have impacts on the amphibians of the Río Salado, either directly through changes in water levels or water chemistry, or indirectly by sublethal effects of changes in water chemistry (e.g., Squires et al., 2008). It is not clear whether or not there is any current impact of the *salineras* on the Río Salado; however, any future increases in the number of extent of *salineras* should be carefully considered in light of the potential impacts on the native fauna, including the amphibians, living in the Río Salado.

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