

Early maturation and high growth rates in an Eastern Treefrog, *Hyla orientalis* Bedriaga, 1890, population after the species' expansion in Kaliningrad Oblast (East Baltic Sea region)

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Global anthropogenic climate change has become the most dominant factor affecting the distribution and abundance of amphibians in recent decades (Luedtke et al., 2023). A number of studies have found that amphibians are dying out even in areas with seemingly intact habitats, including nature reserves and national parks (Lada et al., 2011; Kuzmin, 2013; Kukushkin, 2023). This also applies to populations of the Eastern Treefrog, *Hyla orientalis* Bedriaga, 1890. *H. orientalis* occurs in Eastern Europe, Anatolia, Caucasus, where it occurs isolated from its main range – the forest belt of the southern Caspian region (Litvinchuk et al., 2006; Stöck et al., 2012; Dufresnes et al., 2016a, b). In populations at the eastern edge of the species' distribution in Russia, Ukraine, and Belarus, declines and local extinctions have been recorded (Litvinchuk and Üzüüm, 2022). Similarly, in Russia, *H. orientalis* is almost completely extinct in Belgorod Oblast and has become very rare in Orel and Kursk Oblasts (Kuzmin, 2013; Degtyarev, 2021). It is thought that the main reason for the extinction of *H. orientalis* populations in the eastern part of its range is due to aridification (Lada et al., 2011; Kuzmin, 2013; Kukushkin, 2023).

However, conversely, a range expansion of *H. orientalis* has been recorded in the East Baltic Sea region. For example, *H. orientalis* was very rare in the Kaliningrad region of Russia, and was only found around its southernmost border (Wulff, 1765; Pagast, 1941; Grishanov and Belyakov, 2000; Grishanova and Grishanov, 2022). More recently, at least 50 new localities have been recorded, located up to 30–40 km to

the north (Litvinchuk et al., 2024; our data) (Fig. 1). An increase in average annual temperature and precipitation along the southern Baltic Sea coast is likely to have contributed to this (Stont and Demidov, 2015). This paper presents a first assessment of the age structure and demographic patterns of *H. orientalis* in one of the new populations that is part of the species' range expansion in the southern Baltic Sea region.

Material and methods

Adult animals were captured during the breeding season (8–10 June 2023 and 8–10 May 2024) near the Bolshakovskoye settlement (54.4383°N, 20.6544°E, elevation 46 m) in the Bagrationovsky District, Kaliningrad Oblast (Russian Federation) (Fig. 1). In this area, *H. orientalis* uses flooded depressions in maize and wheat fields to breed (Fig. 2). From 27 adult males, the snout-vent length (SVL) was measured, and toe clips were taken at the third phalanx of the fourth finger of the right hind limb to determine the age of animals through skeletochronology (Smirina, 1989). The animals were released at the site of capture after sampling.

Statistical processing and visualisation of the data were performed using OriginPro 2024 software (OriginLab, USA). The mean and standard deviation ($M \pm SD$), as well as the range (min–max) of the examined characteristics were calculated. The asymptotic size (SVL_{max}) and growth coefficient (k) were calculated using non-linear estimation, applying the von Bertalanffy growth model (Bertalanffy, 1938). The correlation between body length and age in *H. orientalis* individuals was further assessed using linear regression analysis. The annual survival rate (S) and life expectancy (ESP) based on the age distribution were calculated using the formulas of Robson and Chapman (1961) and Seber (1973).

To compare the demographic characteristics of *H. orientalis* from the new population, data obtained from native populations available in the literature (searched through Scopus, Web of Science, and ResearchGate)

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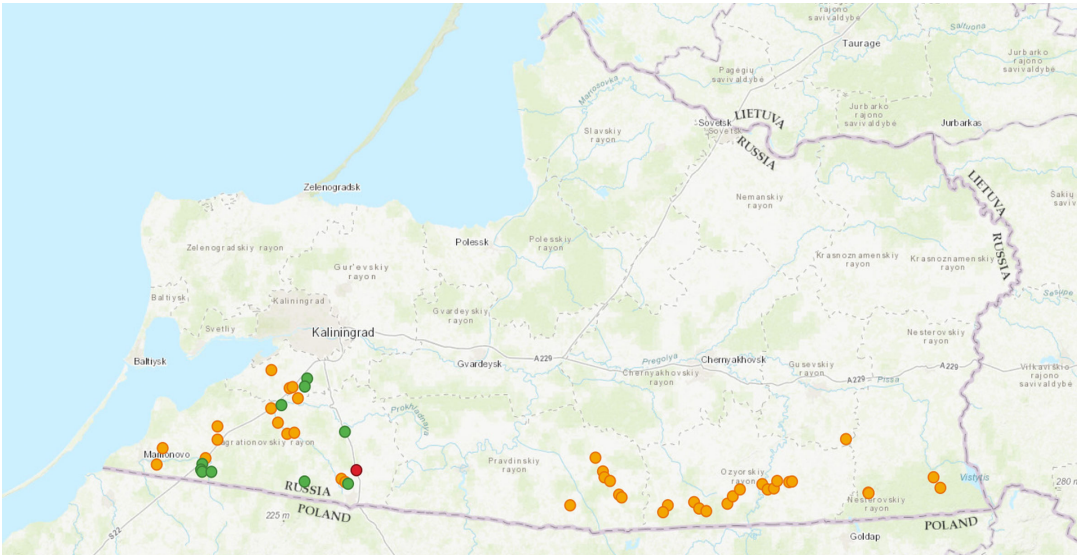


Figure 1. Distribution records of *Hyla orientalis* in the Kaliningrad Oblast. Orange circles are localities represented in Litvinchuk et al. 2024. Green circles present our new findings, while the red circle (Bolshakovskoye settlement, Bagrationovskiy District) highlights our study site.

were used (Table 1). Across the eight total populations for which comparative data could be obtained, Spearman’s rank correlation coefficient was used to determine the correlation between demographic indicators (presented in the Table 1) and bioclimatic variables extracted from the WorldClim 2.1 database (Fick and Hijmans, 2017) with NextGIS QGIS (v 24.11.0) with a 30 arc

seconds spatial resolution. To eliminate predictor collinearity before assessing the correlation between demographic indicators and bioclimatic variables, we calculated pairwise Pearson’s correlation coefficients for all bioclimatic variables. For correlated pairs ($|r| > 0.70$), we excluded the variable that more correlated with others. The resulting dataset contained

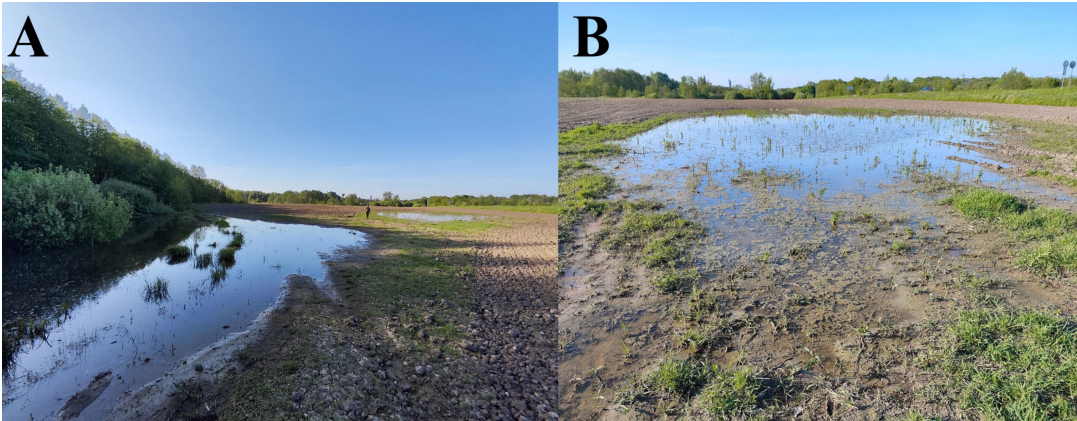


Figure 2. The breeding locality of *Hyla orientalis* presenting our study site (Bolshakovskoye settlement, Bagrationovskiy District, Kaliningrad Oblast), consisting of (A) a shaded pond located at the edge of a field and forest, and (B) an open pond in the middle of the field with increased sun exposure, in which most of the *H. orientalis* individuals were found. Photos by Roman Ivolga, May 2024.

Table 1. Age and size of matured males of the *Hyla orientalis* from different localities. n – number of individuals, SD – standard deviation, S – annual survival rate, ESP – life expectancy, SVL – snout-vent length, SVL_{max} – asymptotic size, SE – standard error, k – growth coefficient

Locality	Year	Latitude	Longitude	Elevation (m)	n	Mean age	SD	Age at maturity	Longevity	S	ESP	Mean SVL	SD	SVL _{max}	SE	k	SE	Reference
Borjomi Canyon, Georgia	1992–1993	41.8333	43.3833	900–1200	33	4.41	1.49	3	12	0.628	3.19							Gokhelašvili, Tarkhišvili, 1994
Kantari, Rize, Turkey	2010	40.9667	40.8667	800	32	4.88	0.83	2	7	0.659	3.43	43.56	2.56					Altunışık, Özdemir, 2013
Konaklı, Antalya, Turkey	2012	36.5925	31.8806	18	32	2.7	0.6	1	4	0.617	3.11	35.3	1.8					Özdemir et al., 2012
İslampaşa, Rize, Turkey	2012	41.0369	40.5508	26	27	3.5	0.8	1	5	0.601	3.01	37.2	3.2					Özdemir et al., 2012
Gelibolu area, Turkey	2012	40.6278	26.9264	32	66	2.5	0.9	2	5	0.606	3.04	37.3	3.1					Özdemir et al., 2012
Chornobyl area, Ukraine	2016–2018	51.4	30.05	100	191	3–4		2	9					–				Burraco et al., 2024
Primorskiy, Dagestan, Russia	2022	41.8428	48.5772	–10	13	3.4	1.33	1	6	0.721	4.08	40.13	3.648	41.54	1.107	0.89	0.292	Kidov et al., 2023
Bolshakovskoye, Kaliningrad Oblast, Russia	2023–2024	54.4383	20.6544	46	27	2.07	0.874	1	4	0.527	2.61	43.69	2.875	45.26	0.624	1.975	0.243	present study

four bioclimatic variables: annual mean temperature, mean diurnal range (=mean of monthly (max temp-min temp)), mean temperature of wettest quarter, and annual precipitation.

Results

In the obtained samples, the lines of arrested growth (LAGs) were easy to read, allowing us to determine the age of individuals with a high degree of confidence. Since adult males (seen to be vocalising and showing a pigmented vocal sac) with a single LAG (bone edge) were present in the studied sample, it can be argued that they begin to participate in breeding as early as one year of age. The mean age of the animals studied was 2.07 ± 0.874 years, ranging from 1 to 4 years. The modal age of *H. orientalis* was 2 years (37% of the sample) (Fig. 3A), and the life expectancy (ESP) of individuals after sexual maturity was 2.6 years ($S = 0.527$). Out of all of the demographic characteristics of the *H. orientalis* males from the localities presented in the Table 1, none were correlated with bioclimatic variables.

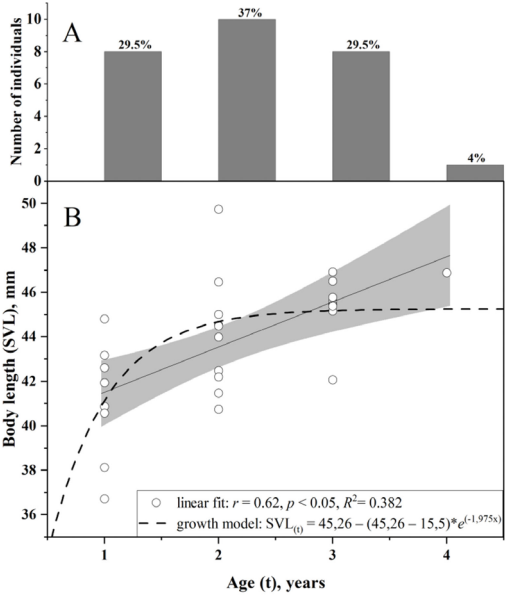


Figure 3. Age structure and growth of *Hyla orientalis* in our study population (Bolshakovskoye settlement, Bagrationovsky District, Kaliningrad Oblast). (A) Histograms highlight the amount and proportions individuals found in each age class. (B) Relation between body length (SVL) and age, using both linear regression ('linear fit') and approximating its growth trajectory using the von Bertalanffy's model ('growth model').

The SVL of *H. orientalis* varied between 36.71–49.74 mm (mean 43.69 ± 2.875 mm) and was correlated with ($r = 0.62$; $p < 0.05$), but not predictive of ($R^2 = 0.382$), animal age. The mean SVL of the *H. orientalis* males from the localities presented in the Table 1 correlated with annual mean temperature ($r = -0.92$, $p < 0.05$). The von Bertalanffy growth model was estimated to reliably predict the growth of *H. orientalis* ($R^2 = 0.872$), with an asymptotic size (SVL_{\max}) of 45.26 ± 0.624 mm ($p < 0.001$) and a growth coefficient (k) of 1.975 ± 0.243 ($p < 0.001$) (Fig. 3B).

Discussion

The demographic characteristics of *H. orientalis* are highly variable throughout its range (Gokhelaşvili and Tarkhnishvili, 1994; Özdemir et al., 2012; Altunişik and Özdemir, 2013; Kidov et al., 2023; Burraco et al., 2024). For example, the mean body length (SVL) in different populations was seen to range from 35.3 mm in Konaklı (Turkey; Özdemir et al., 2012) to 43.56 mm in Kantarlı (Turkey; Altunişik and Özdemir, 2013). The age of maturity of this species ranges from 1 year in lowland localities in the south of its range (Konaklı, İslampaşa (Turkey; Özdemir et al., 2012) and Primorskiy (Dagestan, Russia; Kidov et al., 2023)) to 3 years in a mountain population in Borjomi Canyon (Georgia; Gokhelaşvili and Tarkhnishvili, 1994). Unsurprisingly, the mean age of *H. orientalis* is lowest in populations with early maturity (2.5–2.7 years in Konaklı and İslampaşa (Turkey; Özdemir et al., 2012)) and highest in populations with relatively late maturity (4.41–4.88 years in Borjomi Canyon (Georgia; Gokhelaşvili and Tarkhnishvili, 1994) and Kantarlı (Turkey; Altunişik and Özdemir, 2013)). The longevity of *H. orientalis* shows the same trends: from 4 years in Konaklı (Turkey; Özdemir et al., 2012) to 12 years in Borjomi Canyon (Georgia; Gokhelaşvili and Tarkhnishvili, 1994). The life expectancy of male *H. orientalis* ranges from 3.01–3.04 years in İslampaşa and Gelibolu (Turkey; Özdemir et al., 2012) to 4.08 years in Primorskiy (Dagestan, Russia; Kidov et al., 2023) (Table 1).

The *H. orientalis* individuals we studied from a newly observed population in the southern Baltic Sea region were characterised by a relatively large body size (mean SVL = 43.69 mm), a high growth rate ($k = 1.975$), early sexual maturity (starting from 1 year of age), as well as a low mean age (mean age = 2.07 years) and lifespan (observed longevity = 4 years, ESP after maturity = 2.61 years). Similar trends have previously been observed for some other amphibian species when comparing

demographic parameters in individuals from native and introduced populations. For example, Asian Common Toads, *Duttaphrynus melanostictus* (Schneider, 1799), introduced to Madagascar were larger and younger than conspecifics from native populations (Guarino et al., 2023). Similarly, Fire-bellied Toads, *Bombina bombina* (Linnaeus, 1761), introduced into France were characterised by early maturity, were younger on average, but did not differ in body size from individuals from native habitats (Cogălniceanu et al., 2024).

The seemingly shortened lifespan fits into a general life-history theory that predicts that populations with early sexual maturation of individuals, high fecundity (Pianka, 1970), and an accelerated life cycle can promote range expansion (Blackburn et al., 2015; Capellini et al., 2015). This allows small groups to grow rapidly, increasing the number of individuals by reducing the period during which individuals are vulnerable to stochastic environmental changes (Pimm, 1991) and compensating for the likely reduced survival of adults in new habitats (Cayuela et al., 2022).

Increased locomotion and body size, coupled with high growth rates, may also contribute to accelerated expansion of the species, as these traits should allow individuals to travel longer distances in a shorter timespan. For example, it has been shown that the invasion of the Cane Toad, *Rhinella marina* (Linnaeus, 1758), into Australia was accompanied by a sharp increase in locomotion and (in some populations) the ability to maintain terrestrial activity under cold and/or dry conditions. (Kosmala et al., 2017). Hudson et al. (2016) found a positive relationship between hind limb length and obstacle crossing ability in toads of this species. Phillips et al. (2006) showed that *R. marina* with longer hind limbs moved faster and were the first to occupy new areas, and that individuals living in the periphery had longer limbs than toads from long-established populations.

The results we obtained in our study of the Eastern Treefrogs are consistent with other predictions of life-history traits that contribute to the successful establishment of amphibian populations following introduction and further expansion (Allen et al., 2017). However, increases in body size and decreasing in longevity are not always a result of species expansion, but may also depend on climatic variables in new habitats: e.g., mean annual precipitation (Oromi et al., 2016). Indeed, our analysis showed that the mean SVL of the *H. orientalis* males from the localities presented in the Table 1 decreased with increasing annual mean

temperature.

Thus, this study suggests that *H. orientalis* in a new population in the southern Baltic Sea region has a life history characterised by early sexual maturity, a high growth rate, and a short lifespan. It is plausible that these observed demographic features are the result of the recent range expansion of *H. orientalis* in the southern Baltic area, which have been noted to favour the spread of the species, while the large body size may be linked to local climatic conditions. However, since the study is based on a single population, these demographic patterns may also be due to the small sample size rather than true variability following the species' expansion. Further comparisons with longer-occupied sites would be needed to assess whether the hypothesised relation of range expansion and demographic features is consistent.

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