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The roles of phylogeny and climate in shaping variation in life-history traits of the newt genus *Triturus* (Caudata, Salamandridae)

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Assessing the origin of trait variation during evolutionary history is an important first step in understanding evolutionary diversification. Here, we tested the influence of shared ancestry and climate, and the interplay of both, on the variation of ten life history traits in *Triturus* newts. We showed that (1) climate alone has driven the evolution of variation in five life history traits, (2) phylogenetic signal partly explains the variation in two traits (vitellus diameter and snout–vent length of larvae at metamorphosis), and (3) the interplay of shared ancestry and climate explains the variation in one trait (snout–vent length of larvae at metamorphosis). This study highlights the coarse-grained influence of shared ancestry and climate on the structure of phenotypic trait variation in *Triturus* and provides a handle for more detailed, fine grained studies on the evolution of phenotypic trait variation.

Introduction

Determining the role of environmental selection pressures in the evolution of phenotypic trait variation in related species is a major challenge for evolutionary ecologists. Constraints, defined as "any limitation on phenotypic evolution that results from past evolutionary change in a particular lineage" (Schwenk 1995) and often referred to as phylogenetic or intrinsic constraints, are important factors in shaping the effects of environmental selection pressures. Phylogenetic signal (or phylogenetic effect) means that closely related species tend to be phenotypically similar and is an outcome of constrained phenotypic evolution (Revell *et al*. 2008).

Many studies (in morphology, life history, behaviour, physiology and ecology) have focussed on elucidating patterns of trait variation across species driven by environmental selection pressures, while taking phylogenetic signal into account (e.g. Freckleton *et al*. 2002, Blomberg *et al*. 2003, Desdevises *et al*. 2003). However, the majority of "phylogenetic correction" methods attributes most of the explained variation in a trait to phylogeny and considers only the residual as explained by ecology (Westoby *et al.* 1995). Westoby *et al.* (1995) pointed out that part of the variation of a trait may not be related only to phylogeny or to ecology, but to both phylogeny and ecology "at the same time" as a potential outcome of phylogenetic niche conservatism. Phylogenetic niche conservatism is a tendency of lineages to retain their niche-related traits through speciation events (Wiens *et al.* 2010). For example, sister species could independently develop adaptive phenotypes, but, as they tend to inhabit the same kind of environment as their ancestors, they develop the same plesiomorphic trait.

To summarize, the drivers behind trait variation across species can be partitioned into tree main components: (1) purely phylogenetic (intrinsic), where species traits evolve under constraints, (2) purely ecological (extrinsic), where species traits evolve in response to ecological drivers alone, and (3) phylogenetically structured environmental variation, where species traits related to niches are retained after speciation (Desdevises *et al.* 2003, Diniz & Bini 2008).

Although life history traits are closely related to fitness and therefore tightly connected to ecological factors, many papers confirmed correlation of different life history traits and species' phylogenetic history in different animal groups (e.g. Stearns 1983, 1984, Gotelli & Pyron 1991, Miles and Dunham 1992, Freckleton *et al.* 2002, Blomberg *et al.* 2003). Here, we focus on the role of climate and phylogeny in shaping life history traits in the genus *Triturus* (Amphibia, Caudata, Salamandridae), which is a well-recognized model system for evolutionary studies (e.g. Arntzen *et al.* 2007, Ivanović *et al.* 2012). For the genus *Triturus*, variation in life history traits has been studied in detail (*see* Appendix), and the phylogeny is well understood (Wielstra & Arntzen 2011). However, the life history traits of *Triturus* have never been analysed in a framework of the interplay of evolutionary history and climate.

The genus *Triturus* is a monophyletic clade divided into two coherent groups (Fig. 1), marbled newts (*T. marmoratus*, *T. pygmaeus*) and crested newts (*T. carnifex*, *T. cristatus*, *T. dobrogicus*, *T. macedonicus*, *T. karelinii*). It should be noted that *T. karelinii* comprises more than one species but as the taxonomy is currently not yet settled (*see* Wielstra *et al.* 2013a, 2013b) we treat it as a single unit; this should not affect our conclusions as there are as yet no known differences among the potential species involved (Arntzen 2003). The *Triturus* species show species-specific ecological demands (e.g. duration of the annual aquatic period being one of the most pronounced difference, Arntzen 2003, Arntzen *et al.* 2007, Ivanović *et al.* 2012) which is reflected in differences in many life history traits among *Triturus* species (*see* Fig. 1).We include the Pyrenean brook newt (*Calotriton asper*) as an outgroup. Despite its sister relationship to *Triturus*, *C. asper* has a completely different ecology, being adapted to a benthic life in mountain streams (Griffiths 1996), in contrast to *Triturus* newts, which congregate in stagnant water bodies to court, mate and lay eggs.

The aim of this study is to trace life history traits over a phylogeny and explore the drivers behind life history trait variation, which is a crucial step in providing important insights into whether the evolution of life history traits had an important role in the radiation of *Triturus* species. In order to do so, we explore the structure of life history trait variation in the genus *Triturus* by testing for (1) variation explained by shared ancestry, (2) variation explained by climate, and (3) variation explained by both, shared ancestry and climate.

Material and methods

Phylogeny, climate and life history data

The phylogeny of the *Triturus* species used in this study (Fig. 1) was obtained from Wielstra and Arntzen (2011). Phylogeny was expressed in the form of principal coordinates via a Principal Coordinate Analysis (PCoA: Gower 1966) computed from the patristic distance matrix. Then, following Desdevises *et al.* (2003), principal coordinates (PCs) were subjected to a forward selection procedure to select PCs that significantly contributed to the explanation of the dependent variable (life history trait). PCs selection procedure was performed for each life

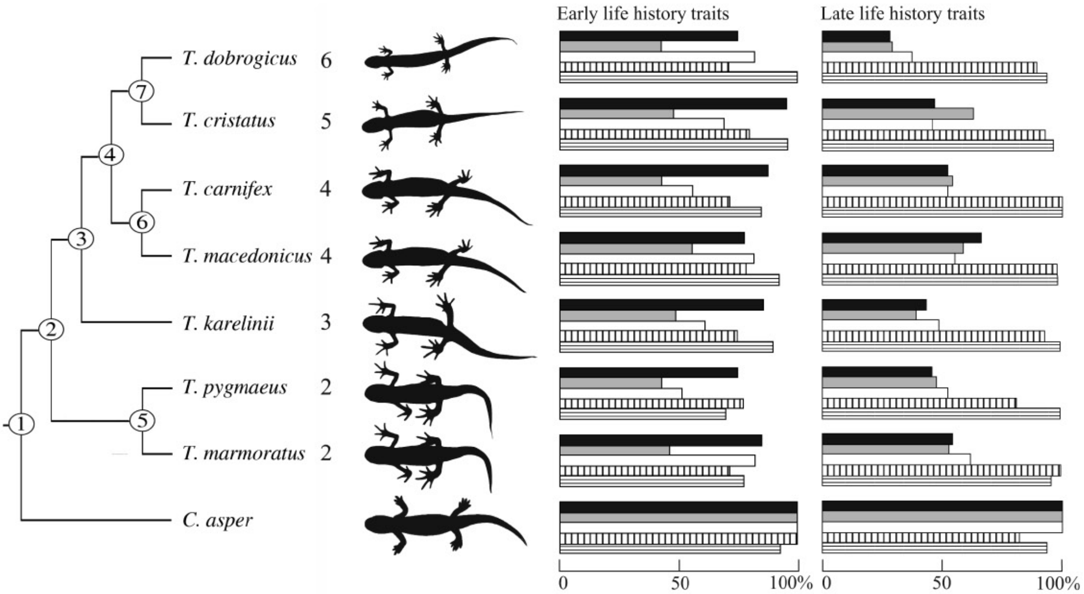

Fig. 1. The phylogeny of the genus *Triturus* according to Wielstra and Arntzen 2011 (left); numbers next to the species names represent the duration of the annual aquatic period (in months) and cartoons represent body shape (based on Vukov *et al.* 2011). Relative variation of life history traits is presented with horizontal bars (right). Early life history traits: black = length of egg (mm), grey = vitellus diameter (mm), white = duration of embryonic period (days), vertical lines = length of larvae immediately after hatching (mm), horizontal lines = snout–vent length (SVL) at metamorphosis. Late life history traits (average for both sexes): black = average age, grey = longevity, white = age at maturation, vertical lines = average SVL (from tip of the snout to the posterior end of the cloaca), horizontal lines = sexual size dimorphism (SSD) index.

history trait independently. Significant PCs were used as independent variables in a variation partitioning model. Patristic distances, PCoA and significance of PCs were calculated using the functions *cophenetic*, *pcoa*, and *lmorigin* from the *ape* library (Paradis *et al*. 2004) of the R statistical package (R Development Core Team 2006).

As climatic data we used biocliomatic variables available from the WorldClim database ver. 1.4 (http://www.worldclim.org, Hijmans *et al*. 2005). Data values were extracted from these data layers for 4480 *Triturus* newt occurrences [311 *T. karelinii*, 139 *T. macedonicus*, 120 *T. carnifex*, 1695 *T. cristatus*, 136 T. *dobrogicus*, 1529 *T. marmoratus*, and 559 *T. pygmaeus*, locality data from Wielstra *et al*. (2012) and 491 Pyrenean brook salamander (*C. asper*) occurrences (GBIF)]. GBIF refers to the data obtained via the GBIF Data Portal (http://www.gbif.org/) originally supplied by the following: (1) Ministerio de MedioAmbiente, y Medio Rural y Marino. Dirección General de Medio Natural y Política Forestal. Inventario Nacional de Biodiversidad 2007, Anfibios; (2) Museum of Vertebrate Zoology Berkeley; (3) Service du Patrimoine naturel, Muséum national d'Histoire naturelle, Paris; (4) Centre d'estudis de la neui de la muntanyad'Andorra (CENMA), Institut d'Estudis Andorrans; (5) Smithsonian National Museum of Natural History; (6) University of Navarra, Museum of Zoology. Considering recommendations to use only biologically meaningful data based on life history knowledge of the model system (Austin 2007, Guisan & Thuiller 2005), we selected several uncorrelated (Pearson's $r < 0.7$) climatic variables for further analyses (*sensu* Wielstra & Arntzen 2012): bio11 = mean temperature of the coldest quarter, bio16 = precipitation of the wettest quarter, bio17 = precipitation of the driest quarter. The median values of climate variables for each species were used as independent variables in a variation partitioning method.

Life history data of the *Triturus* newt species and *C. asper* were obtained from the published literature (*see* Appendix). If the data were

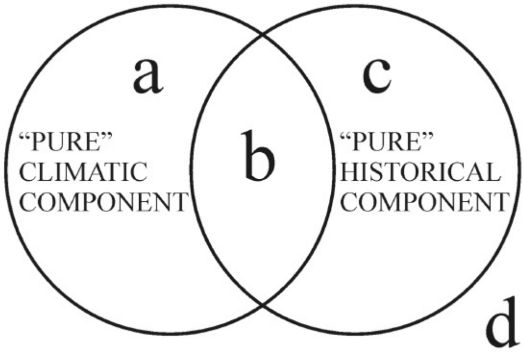

Fig. 2. Partitioning of a dependent variable *y* by two sets of indipendent variables (circles). The left circle (fraction [a + b]) is explained by climate, the rigth circle (fraction [b + c] is explained by phylogeny. The partitioning method allows the estimation of the overlapping fraction of variation (fraction [b]), "pure" components (fraction [a] and [c]) and the remaining unexplained variation [d].

obtained for more than one population per species, parameters were averaged across populations. Ten life history traits were included in our dataset: five early life history traits: length of egg (mm), vitellus diameter (mm), duration of embryonic period (days), length of larvae immediately after hatching (mm), snout–vent length (SVL) at metamorphosis (mm); and five late life history traits : average age (years), longevity (years), age at maturation (years), average SVL (from tip of the snout to the posterior end of the cloaca) (mm), and a sexual size dimorphism (SSD) index. The SSD index (hereafter referred to as SSD) was estimated as the mean size of the females divided by the mean size of the males. Late life history traits (except SSD) were analysed separately for males and females. Life history traits were used as dependent variables in a variation partitioning method.

The life history traits and climate variables were ln-transformed to linearize their relationship. Departure from normality did not have to be assessed because all tests of significance were performed through permutational procedures (9999 permutations) that do not assume normality. A permutation test is also highly recommended because it preserves a low Type I error rate (false positive results) and increases power when handling small sample size data ($n < 10$) (Bishara & Hittner 2012).

Variation partitioning method

In order to estimate what proportion of the life history traits are correlated with climate alone, phylogeny alone, and jointly with climate and phylogeny, we calculated several fractions (Fig. 2): (1) [a + b], (2) [b + c] and (3) [a + b + c] (Desdevises *et al.* 2003). The first fraction was defined as the regression of each life history trait to each climatic variable. The second fraction was defined as the regression of each life history trait to the PCs (represents phylogeny). The third fraction was defined as the regression of each life history trait to the significant PCs and climatic variables. Factors [a] ("pure" climatic component) and [c] ("pure" historical component) were found by two partial regression equations. Fraction [a] was found by regressing climate to PCs and computing residuals. The partial regression was obtained by regressing life history traits on these residuals. Fraction [c] was found by calculating residuals from the regression of each individual PC on each climate variable. Next, life history trait was regressed on those residuals via multiple linear regressions. As an unbiased estimator of the contribution of the fractions of variation in the variation partitioning model, the adjusted coefficient of determination (r^2) was used (Peres-Neto *et al.* 2006). Variation partitioning and the tests of significance of the fractions were computed using the function *lmorigin* in the *ape* library (Paradis *et al.* 2004) of R (R Development Core Team 2006). All tests were performed through a permutation procedure using 9999 permutations.

Variation partitioning was performed for each life history trait separately by using each of the three climatic variables as an exploratory variable separately (10 life history traits × 3 climate variables = 30 variance partitioning procedures). PCs, a second set of exploratory variables, were used according to the results of the PC selection procedure described above.

Results

Mean values of the dependent variables (life history traits) and median values of the independent exploratory variables (climatic variables) are

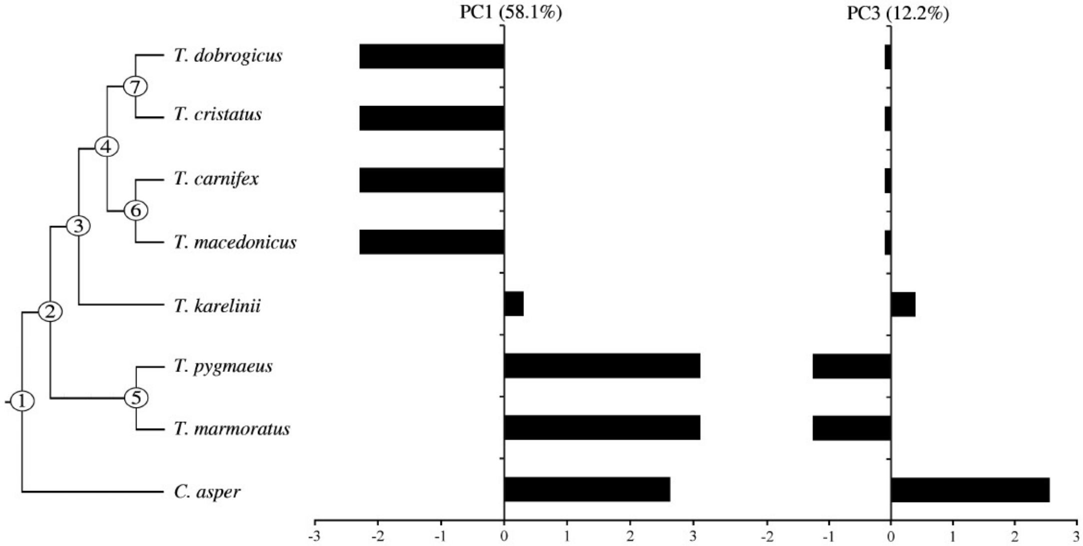

Fig. 3. Phylogeny (left) was expressed in the form of principal coordinates (right: PC1 and PC3 scores) that significantly contribute to the explanation of the life history traits variation.

shown in Tables 1 and 2. Results for the life history traits where all fractions are non-significant were not presented in the result section and are available upon request.

The phylogenetic (historical) component

The phylogeny of newts is expressed as principal coordinates, and PC1 and PC3 are selected as coordinates that contribute to the explanation of the life history traits. PC1 and PC3 represent 58.1% and 12.2%, respectively, of the total variance of the PCoA-transformed patristic distance matrix. PC3 contributes to the explanation of most of the life history traits while PC1 contributes to SVL at metamorphosis and age at maturation (Table 3). PC1 presents differences between *C. asper* + marbled newts and crested newts, while PC3 presents differences between *C. asper* and *Triturus* newts (Fig. 3).

After strict permutation procedures, only the variation of vitellus diameter and snout–vent length at metamorphosis contained a significant

Table 1. Mean values of early life history traits and median values of climatic variables.

Species	Length of egg (mm)	Vitellus diameter (mm)	Duration of EP (days)	Length of hatched larvae (mm)	SVL at metamorphosis (mm)	bio11 (°C)	bio16 (mm)	bio17 (mm)
Triturus dobrogicus	3.75	1.74	19.97	9.18	37.10	0.24	219	114.5
T. cristatus	4.78	1.95	16.84	10.34	35.59	–2.35	223	112
T. carnifex	4.38	1.75	13.65	9.25	31.50	3.06	300.5	151
T. macedonicus	3.89	2.28	19.89	10.14	34.28	1.68	292	129
T. karelinii	4.29	1.99	14.89	9.65	33.25	1.62	237	110
T. pygmaeus	3.75	1.75	12.50	10.00	25.88	8.57	224	32
T. marmoratus	4.25	1.88	20.00	9.25	28.74	5.07	256	110
Calotriton asper	5.00	4.08	24.33	12.93	34.38	0.53	311.5	229.5

EP = embryonic period, SVL= snout–vent length, bio11 = mean temperature of the coldest quarter, bio16 = precipitation of the wettest quarter, bio17 = precipitation of the driest quarter.

phylogenetic signal (fraction [b + c], Table 3 and Fig. 2). However, the "pure" phylogenetic component (fraction [c]) was not significant (Table 3), which means that the phylogenetic component for these life history trait variations overlaps to a great extent (fraction [b]) with the climatic component. Only for snout–vent length at metamorphosis variation, when the precipitation of the wettest quarter was used in a full model ([a + b + c]), a "pure" historical component was extracted ($r^2 = 0.582$, $p = 0.007$; Table 3). Although tests show that fractions [a], [b] and [c] were not significant for all traits for which a signal was confirmed, we noticed a potentially important role of fraction [b] in the phylogenetic signal ([b + c]). The phylogenetic signal in the variation of vitellus diameter is a consequence of the large vitellus in *C. asper* as compared with that in *Triturus* newts, while the signal in snout–vent length at metamorphosis is a consequence of smaller size at metamorphosis of marbled newts as compared with other newts.

The climatic component

A significant portion of the snout–vent length at metamorphosis variation was explained by the mean temperature of the coldest quarter (fraction [a + b], $r^2 = 0.788$; *see* Table 3). Precipitation of the wettest quarter explained a significant portion of the variation of average age in males ($r^2 = 0.606$) and females ($r^2 = 0.603$) and age at maturation for males ($r^2 = 0.483$) (fraction [a + b]; *see* Table 3). The portion of duration of embryonic period variation was explained by the precipitation of the driest quarter (fraction [a + b], $r^2 = 0.370$; *see* Table 3). However, the "pure" climatic component (fraction [a]) was significant only for age at maturation for both males and females (Table 3).

Discussion

The variation in five out of ten tested life history traits among *Triturus* species can be explained by climate or phylogeny (vitellus diameter, duration of embryonic period, snout–vent length at metamorphosis, age at maturation and average age). Only two of these five life history traits (snout–vent length at metamorphosis and vitellus diameter) showed a phylogenetic signal.

Differentiation within life history traits is likely to have been driven by natural selection considering their tight link to the environment. Temperature has a profound impact on life history traits of newts, where higher temperatures cause smaller egg size and vitellus diameter, a shorter embryonic period, a smaller size of hatched larvae, a shorter life span (i.e. the mean population age is lower) and earlier maturation (e.g. Griffiths & de Wijer 1994). Precipitation, together with temperature, also has a strong influence on life histories of newts. Under more unstable precipitation seasonality and at a higher temperature of the coldest quarter, larvae are smaller at metamorphosis. In general, individu-

Table 2. Mean values of late life history traits.

	Average age (years)		Longevity (years)		Age at maturation (years)		SVL (mm)		SSD
Species	males	females	males	females	males	females	males	females	
Triturus dobrogicus	3.58	3.69	6.50	7.00	2.00	3.00	64.77	66.38	1.02
T. cristatus	6.12	6.00	15.75	13.25	3.00	3.13	66.46	69.55	1.05
T. carnifex	6.65	6.85	11.50	13.50	3.50	3.50	70.03	76.48	1.09
T. macedonicus	8.80	8.30	14.00	13.00	3.80	3.60	69.29	74.24	1.07
T. karelinii	5.50	5.63	9.00	9.00	3.25	3.25	65.33	70.34	1.08
T. pygmaeus	5.86	5.94	12.00	10.00	3.50	3.50	56.95	61.59	1.08
T. marmoratus	6.64	7.38	12.00	12.33	3.75	4.50	71.50	74.12	1.04
Calotriton asper	11.98	13.78	22.00	24.00	6.15	7.20	59.70	60.81	1.02

SVL = snout–vent length, SSD = sexual size dimorphism, m = males, f = females.

als that are exposed to unpredictable environments, like habitats with less predictable rainfall and higher evaporation, tend to metamorphose earlier, to decrease the risk of the pond drying before metamorphosis is reached (which would mean death) (Merilä *et al.* 2000, Wellborn *et al.* 1996). Also, a general principle is that at higher altitudes (with higher precipitation), longevity (i.e. mean population age) is higher and sexual maturation is delayed. This is an adaptation to lower metabolic rates due to lower temperatures. Under these conditions newts show a lower investment to reproduction and, conversely, a higher investment to self-maintenance, and additionally have a lower mortality risk due to longer hibernation (e.g. Marunouchi *et al.* 2000, Miaud *et al.* 2000, Liao & Lu 2012, Oromi *et al.* 2012).

In order to follow the strict statistical rule of excluding redundant variables from the analyses, we tested the effect of only a single temperature variable on the variation of life history traits among *Triturus* species: mean temperature of the coldest quarter. This climatic variable has a strong, positive effect on snout–vent length at metamorphosis, suggesting that newt species that live in regions with colder winters have larvae that metamorphose at larger sizes. The observed pattern could be a reflection of a higher survival rate during winter for larger individuals (Martof 1956). *Triturus dobrogicus* and *T. cristatus*, which inhabit areas with the coldest winters, have the largest metamorphs, while marbled newts, which inhabit areas with mild and warm winters, have the smallest metamorphs. We have to keep in mind that smaller size at metamorphosis could just be a reflection of small egg size and fast development at warmer temperatures where breeding sites are likely to dry up quickly (e.g. Bradford 1990). Although marbled newts have the smallest size at metamorphosis in *Triturus*, our data (mean values per species) do not show that they have smaller eggs or that they inhabit areas with much warmer climate than other newts. An exception is *T. pygmaeus*, which inhabits areas with a few degrees (°C) higher mean temperatures during the warmest quarter of the year (data not shown).

Regarding precipitation, we tested the effect of precipitation of the wettest and driest quarter of the year on the variation of life history

Table 3. Results of variation partitioning of life history traits between phylogenetic and climatic component. *p* values indicating significance are set in boldface. SVL = snout–vent length.

	[a + b + c]		[a + b]		[b + c]		[a]		[b]		[c]	
	r^2	*p*	r^2	*p*	r^2	*p*	r^2	*p*	r^2	*p**	r^2	*p*
Mean temperature of the coldest quarter (bio11)												
Vitellus diameter (PC3)	0.739	**0.0488**	0.080	0.5128	0.735	**0.0296**	0.004	0.5600	0.076	n/a	0.659	0.1230
SVL at metamorphosis (PC1 & PC3)	0.825	**0.0165**	0.788	**0.0010**	0.717	**0.0291**	0.108	0.4490	0.680	n/a	0.037	0.8070
Precipitation of the wettest quarter (bio16)												
Average age (females) (PC3)	0.575	**0.0443**	0.603	**0.0021**	0.242	0.2792	0.333	0.1210	0.270	n/a	–0.028	0.6630
Average age (males) (PC3)	0.560	**0.0360**	0.606	**0.0017**	0.198	0.1495	0.362	0.1020	0.244	n/a	–0.046	0.7240
Age at maturation (females) (PC1 & PC3)	0.852	**0.0206**	0.339	0.0758	0.705	0.0528	0.147	**0.0060**	0.192	n/a	0.513	0.1420
Age at maturation (males) (PC1)	0.774	**0.0010**	0.483	**0.0051**	0.192	0.1541	0.582	**0.0065**	–0.099	n/a	0.291	0.2000
Vitellus diameter (PC3)	0.728	**0.0263**	0.276	0.1040	0.735	**0.0296**	–0.007	0.7620	0.283	n/a	0.452	0.0665
SVL at metamorphosis (PC1&PC3)	0.753	**0.0423**	0.161	0.8715	0.717	**0.0291**	0.036	0.5080	0.125	n/a	0.592	**0.0070**
Precipitation of the driest quarter (bio17)												
Vitellus diameter (PC3)	0.689	0.1300	0.240	0.1232	0.735	**0.0296**	–0.046	0.8280	0.286	n/a	0.449	0.1210
Duration of embryonic period (PC3)	0.253	0.1890	0.370	**0.0489**	0.183	0.1601	0.070	0.3010	0.300	n/a	–0.117	0.8430
SVL at metamorphosis (PC1&PC3)	0.673	0.0791	0.405	0.0683	0.717	**0.0291**	–0.044	0.4840	0.449	n/a	0.268	0.3700

**p* values not available because r^2[b] was calculated as r^2[b + c] – r^2[c].

traits among *Triturus* species. Precipitation of the wettest quarter had a strong positive effect on the average age and age at maturation, suggesting that newt species that live in regions with high precipitation (usually during winter) are older and mature later. Regarding *Triturus* species, *T. dobrogicus* has the "youngest" populations, with individuals that mature early, while *T. macedonicus* has the "oldest" populations, with individuals that mature late. *Calotriton asper* has much older individuals in populations that mature later in comparison with *Triturus*. Seasonal temperature regimes have a major impact on growth and sexual maturation, as amphibians in colder climates (high altitudes and latitudes) grow more slowly and reach maturity later than amphibians in warmer climates (e.g. Liao & Lu 2012). Our analysis did not show a direct connection between mean temperature of the coldest quarter and age and maturation for *Triturus* species, but higher precipitation is usually present at higher altitudes and therefore could be treated, cautiously, as a proxy for seasonal temperature regimes. Precipitation of the driest quarter had a strong positive effect on duration of the embryonic period, suggesting that newt species that live in drier regions have a shorter embryonic period (such as T. *pygmaeus*). *Calotriton asper* has a much longer embryonic period in comparison with *Triturus*. Again, this prolonged embryonic period is best explained in the literature by low temperatures during embryonic development at high altitudes (Griffiths & de Wijer 1994). Precipitation of the driest quarter can be used as a proxy, again cautiously, for such seasonal temperature regimes.

Although life history traits are under strong influence of the environment, their variation patterns could be shaped by shared ancestry, which we detect as a phylogenetic signal in trait variation. We detected a phylogenetic signal in the variation of vitellus diameter and snout–vent length at metamorphosis. Phylogenetic signal in the variation of vitellus diameter is detected at the node between *C. asper* and *Triturus*, where *C. asper* has a larger vitellus. Phylogenetic signal of variation in snout–vent length at metamorphosis is detected at the node that separates marbled newts from *C. asper* on one side and crested newt on the other side, where marbled newts have a smaller snout–vent length at metamorphosis.

Since life history traits show a connection with climate, the variation patterns in life history traits found in *Triturus* could be a consequence of niche evolution. Given the ecological variation in *Triturus* observed today, climatic niche differentiation probably has an important role during the evolution of *Triturus* (Wielstra *et al.* 2012). We tried to quantify the shared influence of climate and phylogeny on life history trait variation patterns in *Triturus*, which represent the potential effects of niche evolution on life history traits. Only for snout–vent length at metamorphosis did we detect a significant influence of both fractions; climate (mean temperature of the coldest quarter) [a + b], and phylogeny [b + c]. However, we failed to extract a "pure" climatic [a] and a "pure" historical [c] component and their overlap [b] (*see* Fig. 2), which means there could be a large overlap between climate and phylogeny. This result implies that phylogenetic signal in the pattern of variation of snout–vent length at metamorphosis could potentially be a consequence of niche change along the marbled newt branch, where marbled newts have become adapted to warmer climate. Regarding phylogenetic signal in variation of the vitellus diameter, we detected a significant influence of phylogeny [b + c] but failed to extract a "pure" historical component. This means that the variation pattern of vitellus diameter is under the influence of phylogeny but we still have to elucidate if there is a shared influence of climate and phylogeny.

Our analyses of the influence of climate and a shared history on life history trait variation among *Triturus* confirmed the presence of a phylogenetic signal for two life history traits. However, we cannot rule out the presence of signals in the other eight life history traits. Besides climatic influences on life histories, other factors like differences in biotic interactions, and the abundance of natural enemies, competitors and food resources can influence variation in life history traits. We have to keep in mind that phenotypic plasticity also influences the variation in life histories. A study on environment–life history traits variation in the light of phylogeny for a wider set of species (e.g. the whole Salamandridae family) is a logical next step to better understand the evolution of life history traits.

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Appendix. List of data sources used in this study to obtain life history traits. Numbers correspond to those in the references list.

Species	Length of egg	Vitellus diameter	Duration of EP	Length of hatched larvae	SVL at metamorphosis	Average age	Longevity	Age at maturation	SVL
T. dobrogicus	12, 22	12, 22	12, 22, 33	12, 22	12, 10, 22	10, 18	10, 18	10, 18	48
T. cristatus	12, 22	12, 22, 32	12, 22, 27, 32, 33	12, 22, 27, 32	12, 3, 17, 22	28, 35	17, 21, 28, 35	17, 21, 28, 35	48
T. carnifex	19	19	13, 20	20	26	34, 42	34, 42	34, 42	48
T. macedonicus	12, 22, 31	12, 22, 31	12, 22	12, 22, 31	12, 22	11	11	11	48
T. karelinii	12, 22	12, 22	12, 19, 22, 33	12, 19, 22	4, 12, 22, 40, 46, 47	40, 46, 47	40, 46, 47	40, 46, 47	48
T. pygmaeus	24	24	24	24	15, 24	5	5	5	orig. data
T. marmoratus	37	37, 45	16, 41	16, 41	2, 25, 29, 30	5, 30	5, 21, 30	5, 21, 30	48
C. asper	1	14, 23, 43	8, 9, 23	7, 8, 23, 43	8, 43	36	36, 38, 39	36, 44	6