Author Query Form

Journal code: AMRE

Transmittal No.: AMRE1442

Dear Author,

During the preparation of your manuscript for typesetting, some questions have arisen. They are listed below. Please check your proof carefully and mark all corrections at the appropriate place in the proof (e.g., by using on-screen annotation in the PDF file) or compile them in a separate list. Note: if you opt to annotate the file with software other than Adobe Reader then please also highlight the appropriate place in the PDF file.

Any queries or remarks that have arisen during the processing of your manuscript are listed below and highlighted by flags in the proof.

Location in article	Query / Remark: Click on the Q link to find the query's location in text Please insert your reply or correction at the corresponding line in the proof
	No queries

Short Note

Genetic diversity and gene flow decline with elevation in the near eastern fire salamander (*Salamandra infraimmaculata*) at Mount Hermon, Golan Heights*Kathleen Preißler^{1,**}, Eliane Küpfer², Fabian Löffler¹, Arlo Hinckley³, Leon Blaustein⁴,
Sebastian Steinfartz¹

Abstract. The Near Eastern fire salamander (*Salamandra infraimmaculata*) reaches its southern distribution range in Israel. Although the population structure has been analysed in central Israel and at the southern distribution limit, we lack knowledge on populations in the northern area, such as along Mount Hermon. *S. infraimmaculata* occurs at Mt. Hermon along an altitudinal gradient and appears to be fragmented by urban and agricultural landscape. We studied the genetic structure of four populations based on microsatellite loci and the mitochondrial D-loop to determine the genetic diversity and connectivity between populations. We observed moderate gene flow at lower parts, i.e. from Tel Dan and Nimrod Castle to Baniyas indicating extant but limited connectivity. Genetic diversity and gene flow declined along the altitudinal gradient at Mt. Hermon, reaching rock-bottom levels in the highest located population of Nimrod Pool. The observed isolation-by-elevation gradient might induce a higher extinction risk for the highland populations of *S. infraimmaculata*.

Keywords: D-loop, genetic fitness, Israel, microsatellites, migration barrier.

The Near Eastern fire salamander (*Salamandra infraimmaculata*) is one of the southernmost occurring *Salamandra* species. Its patchy distribution encompasses parts of Turkey, Syria, Iran, Iraq, Lebanon, and Israel. Due to the loss of

suitable habitat and breeding sites it is classified as “Near Threatened” by IUCN criteria (Papenfuss et al., 2009; Bogaerts et al., 2013). The species is reaching the southern periphery of its distribution range in Northern Israel where it is restricted to three peripheral population clusters: Galilee, Mt. Carmel and Mt. Hermon (see fig. 1) and is considered endangered (Dolev, Perevolotsky, and Lachman, 2004). Recent genetic population studies revealed a restriction of connectivity between populations in Galilee and Mt. Carmel, possibly driven by land use change, such as settlements and agriculture, and a decrease in genetic diversity towards the southern distribution limit (Blank et al., 2013; Sinai et al., 2019). But the dispersal and migration behaviour of *S. infraimmaculata* is also strongly

1 - Institute for Biology, Molecular Evolution and Systematics of Animals, University Leipzig, Talstraße 33, 04103 Leipzig, Germany

2 - Zoological Institute, Evolutionary Biology, Technische Universität Braunschweig, Mendelssohnstraße 4, 38106 Braunschweig, Germany

3 - Conservation and Evolutionary Genetics Group, Estación Biológica de Doñana (EBD-CSIC), Avda. Americo Vespucio, 26, 41092 Sevilla, Spain

4 - Department of Evolutionary and Environmental Ecology, University of Haifa, Haifa, Israel

*This contribution is dedicated to Leon Blaustein who passed away on June 23rd, 2020 to remember him as an expert on Israeli fire salamanders.

**Corresponding author;

e-mail: kathleen.preissler@uni-leipzig.de

Figure 1. Population structure analysis of four *S. inframaculata* sites located in northern Israel. Map not to scale. (A) Distribution of sampling sites Tel Dan (TD), Banias (Ba), Nimrod Castle (NC) and Nimrod Pool (NP) across area and elevation. (B) Median joining network of mitochondrial haplotypes in comparison with individuals from Galilee and Mt. Carmel. Hatch marks indicate the number of mutations. (C) Genetic structure assuming $K = 2-4$ as inferred by STRUCTURE. Bar plot depicts individual proportions of assignment to each cluster. (D) Principal component analysis with the first two components displayed. Each dot represents an individual. Populations are color-coded according to STRUCTURE $K = 3$ but note that Banias is here closer to Tel Dan. An inertia ellipse is spanning the individuals of each population.

restricted by low elevation and steep slope (Ker-shenbaum et al., 2014; Sinai et al., 2019). Popu-lations along the southern foothills of Mt. Her-mon, such as the Tel Dan population are separ-ated from the Galilee by the Hula valley and appear to be fragmented by urban and agricul-tural landscapes. Moreover, populations follow an altitudinal gradient along the range of Mt. Hermon.

We aim to evaluate the genetic population structure and connectivity of *S. inframacu-lata* populations at Mt. Hermon based on nu-clear (microsatellite loci) and mitochondrial (D-loop) haplotype variation, with a special focus on the effect of elevation. Our data will provide information on the degree of genetic isolation between populations and its causes, thus being informative for local conservation of this threat-ened species.

In 2013/2014, We took tail clips from fire salamander larvae as described elsewhere (Segev et al., 2015) from four sites: Tel Dan Nature Reserve (“TD”), Banias (“Ba”), Nimrod Castle (“NC”) and Nimrod Pool (“NP”). See table 1 for detailed sample information. We stored the tissue separately in 1.5 ml Eppendorf tubes filled with 96% EtOH at -20°C until sample processing and extracted genomic DNA follow-ing a salt extraction protocol.

The mitochondrial D-loop was amplified and sequenced using published primer combinations (L-Pro-ML 5'-GGCACCCAAGGCCAAAATTCT-3'; H-12S1-ML 5'-CAAGGCCAGGACCA AACCTTTA-3') (Steinfartz, Veith, and Tautz, 2000). We checked, edited, and aligned the D-loop sequences using CodonCode Aligner v8.0.2 (CodonCode Corporation, www.codoncode.com). For the detailed PCR pro-tocol, see supplementary material. In order

Table 1. Estimates of genetic diversity parameters for each *S. inframaculata* population: TD-Tel Dan, B-Banias, NC-Nimrod Castle, NP-Nimrod Pool located at different elevations. N: number of individuals analysed per marker system, P_A : private alleles, A_r : allelic richness, H_o : observed heterozygosity, H_e : expected heterozygosity, F_{is} : inbreeding coefficient and 95% confidence interval.

Site		TD	Ba	NC	NP
Coordinates	Latitude	33°14'53N	33°14'38N	33°15'08N	33°14'56N
	Longitude	35°39'10E	35°41'15E	35°42'45E	35°45'10E
Elevation [m.a.s.l.]		192	328	743	1022
N	D-loop	22	0	15	35
	Msats	25	7	21	46
A		46	42	43	38
P_A		5	3	2	7
A_r		3.14	3.42	2.92	2.55
H_o		0.54	0.62	0.51	0.39
H_e		0.56	0.62	0.53	0.42
F_{is}		0.039	−0.001	0.026	0.07
F_{is} 95% CI	low	−0.069	−0.201	−0.064	0.0056
	high	0.1368	0.1309	0.1132	0.1309

to present the full Israeli haplotype network, we incorporated *S. inframaculata* sequences from Galilee and Mt. Carmel sampled in a separate project (see sample information in supplementary table S1). The alignment was checked and exported in MEGA7 (Kumar, Stecher, and Tamura, 2016). The median-joining haplotype network was created in PopArt v1.7 (Leigh and Bryant, 2015).

We used 14 polymorphic microsatellite loci (Sal E8, Sal E7, Sal E6, Sal 3, Sal E14, Sal E11, G6, SST-A6-I, SST-A6-II, SST-E11, SST-B11, SST-C3, SST-C2, SST-F10) as described in Steinfartz et al. (2004) and Hendrix et al. (2010) to infer nuclear-based population structure. For the detailed PCR protocol, see supplementary material. We genotyped the microsatellite loci with GeneMarker v3.0.0 (SoftGenetics®). We discarded individuals with more than 15% missing data and loci with more than 7% of missing alleles from further analysis. Alleles were checked for scoring errors, allele dropouts, and null alleles with MICRO-CHECKER v2.2.3. Allelic richness (A_r), the number of alleles per locus (N_a), the inbreeding coefficient (F_{is}), and observed (H_o) and expected (H_e) heterozygosities were calculated for

each sampling site using the R statistics (R Core Team, 2019) package “*diveRsity*” (Keenan et al., 2013). We tested for isolation-by-distance (IBD) and isolation-by-elevation (IBE) as a correlation between genetic distance (F_{st} , pairwise differences) and geographic distance (euclidean) or elevational distances (pairwise) using the Mantel test implemented in Arlequin v3.5.2.2. Population structure was estimated in STRUCTURE v2.3.4 (Pritchard, Stephens, and Donnelly, 2000) by running an admixture model with LOCPRIOR assuming correlated allele frequencies for 10 iterations of $K = 1-5$ with a burn-in of 100,000 and an MCMC of 500,000. STRUCTURE HARVESTER (Earl and vonHoldt, 2012) identified the best fitting K to explain the population structure based on Delta K and Mean $LnP(K)$ (Pritchard, Stephens, and Donnelly, 2000; Evanno, Regnaut, and Goudet, 2005). We used CLUMPP v1.1.2 (Jakobsson and Rosenberg, 2007) and distruct v1.1 (Rosenberg, 2003) to visualize the population’s probability of assignment to each cluster. STRUCTURE population clustering was corroborated by a principal component analysis (PCA, R package “*adeigenet*” (Jombart, 2008)). A hierarchical analysis of molecular variance between/within

strata (AMOVA, 1000 permutations) performed in Arlequin simulated the impact of STRUCTURE predicted clustering on the genetic variance.

The analysis of the mitochondrial D-loop revealed two haplotypes in the sampled Mt. Hermon populations and the reference sites from Mt. Carmel and Galilee. Haplotype I is only shared by individuals from Galilee ($n = 6$) and Carmel ($n = 31$) whereas haplotype lineage II is found in all sampled populations (Galilee $n = 37$, Carmel $n = 10$, TD $n = 22$, NC $n = 15$, NP $n = 35$, fig. 1A). Two mutational steps separate the two haplotypes (T-C and C-T). Sequencing failed for all samples from Banias.

PCR failed for microsatellite loci Sal E7, Sal E14 and Sal G6 resulting in their omission from the dataset. The analysis with MICROCHECKER indicated that loci C3, B11, F10 and E11 show signs of null alleles due to homozygote excess. However, this is not consistent across populations. Indicated errors due to stuttering were re-checked and validated. Deviations from Hardy-Weinberg Equilibrium were not consistent across loci or populations (supplementary table S2).

Population differentiation was greatest between NP and all other sites. Ba-TD (pairwise $F_{st} = 0.069$, $P < 0.01$), Ba-NC ($F_{st} = 0.076$, $P < 0.01$), Ba-NP ($F_{st} = 0.244$, $P < 0.01$), TD-NC ($F_{st} = 0.103$, $P < 0.01$), TD-NP ($F_{st} = 0.229$, $P < 0.01$, NC-NP ($F_{st} = 0.191$, $P < 0.01$). This genetic divergence is mainly driven by geographic distance as well as by elevation as indicated by significant patterns of IBD (Pearson's $r = 0.67$, $P = 0.037$) and IBE (Pearson's $r = 0.58$, $P = 0.049$, supplementary fig. S1). Moreover, elevation had a strong effect on genetic diversity (A_R : Pearson's $r = -0.88$, $P = 0.11$; H_o : $r = -0.84$, $P = 0.15$; H_e : $r = -0.86$, $P = 0.13$). This negative correlation, however, was not significant due to the low size of sampled locations ($N = 4$). Located at the highest elevation, individuals at NP show the lowest genetic diversity and experience significant inbreeding (table 1, the 95% CI of the difference between means

did not include zero). The presence of more private alleles ($N = 7$) at NP than in the other populations (table 1) supported the isolation of this population. Although we used a rare-fraction method, the low sample size analyzed from Ba could still have biased the results as indicated by the higher diversity estimates compared to TD which is occurring at a lower elevation.

Population structure analysis suggested a clustering of sampled individuals in two or three distinct genetic groups ($K = 2$: mean $\ln P(K) = -2091.83$ SD 0.59, $\Delta K = 485.64$; $K = 3$: mean $\ln P(K) = -2027.87$ SD 2.71, $\Delta K = 12.75$, $K = 4$: mean $\ln P(K) = -1998.57$ SD 7.6, $\Delta K = 7.02$, $K = 5$: mean $\ln P(K) = -2022.60$ SD 28.88; fig. 1A, supplementary fig. S2). ΔK peaked at $K = 2$, suggesting absent gene flow between sites TD, B, NC to NP (fig. 1B). However, the standard deviation of the mean $\ln P(K)$ was smallest at $K = 3$ before it rapidly increased at $K = 4$ to 5. This assumption marks Ba a hybrid zone between TD and NC; NP remains isolated (fig. 1B). The strong genetic divergence of NP is corroborated by the PCA ($PC1 = 22.9\%$, $PC2 = 7.57\%$; fig. 1C). By clustering the sampling sites into $K = 2$ and 3, the AMOVA derived molecular variance among sites drops from 17.86% ($K = 1$) to 8.17% and 6.62% respectively (supplementary table S3); confirming an extant population structure. The simulation of $K = 4$ indicates a higher gene flow from NC to Ba than from TD to Ba (fig. 1B). Further splitting ($K = 5$) confirms this pattern (supplementary fig. S3).

Our results indicate a substantial population structure of *S. infraimmaculata* at the southern foothills of Mt. Hermon. Moreover, elevation seems to have a strong impact on population structure as here the genetic isolation from other populations results in an increase of inbreeding and lower genetic diversity indices.

The sharing of the common D-loop haplotype I by Mt. Hermon populations and populations from the Galilee and Mt. Carmel indicate historical gene flow and/or initial colonization of *S. infraimmaculata* at its southern distribution range. The presence of a private haplotype in the Galilee and Mt. Carmel, however, shows that this connection has been interrupted and that populations of these areas follow their own evolutionary trajectories already for a significant amount of time. Populations at the low, medium and high elevation range of the Mt. Hermon region display different patterns of genetic structure. Low (TD, Ba) and medium (NC) elevation sites show signs of genetic coherence with moderate degrees of F_{st} differentiation and gene flow. While these populations form a single genetic unit compared to the high elevation site (NP) for $K = 2$, a further splitting of K supports the existence of separate genetic groups with substantial intermixing between sites (fig. 1). Thus, it seems that migration and dispersal of salamanders is possible through human-mediated landscape structures at low and medium elevations. Our local findings contradict the view that dispersal of *S. infraimmaculata* is limited through low elevations as found in the range of Galilee (Kershenbaum et al., 2014; Sinai et al., 2019).

The high elevation population NP (>1000 m) displays for all types of analyses a strong genetic differentiation with regard to low and medium elevation sites. We would like to emphasize that the geographic distance between NC and NP is in the same range as for NC and TD. Elevation gradients are common physical barriers to migration in other animals (Polato et al., 2017; Valbuena-Ureña et al., 2018) and plants (Reis et al., 2015) due to associated high energetic costs and higher predation risk (Blank et al., 2013). We observed that the increasing genetic isolation manifested in declining allelic richness and heterozygosity along the gradient entailing increased inbreeding. Addis et al. (2015) found the same pattern in boreal

toads (*Bufo boreas*). The high number of private alleles present in NP are most likely the result of strong genetic drift which is typical for small and highly isolated populations. In how far the evolution of these new neutral alleles are a proxy for environmental adaptation at higher elevations remains unclear.

Climate change and the increasing landscape fragmentation in Israel are likely to further drive the process of isolation of *S. infraimmaculata* populations if open migration corridors with suitable steppingstone habitats are not present. Low genetic diversity and presumably small population size render the studied Mt. Hermon populations prone to extinction in case of extreme events, such as drought or infectious diseases (Bar-David et al., 2007; Markert et al., 2010; Martel et al., 2014). This high extinction pattern is common among peripheral populations due to suboptimal habitat e.g. in animals (bustard (*Otis tarda*) in Africa (Palacín et al., 2016), great crested newt (*Triturus cristatus*) in Great Britain (Miró et al., 2017)) and plants (forest trees (Fady et al., 2016)) alike. However, peripheral populations are important in evolutionary terms as high selective pressures lead to significant evolutionary changes (Fady et al., 2016). To conclusively determine the degree of isolation and potential threat, detailed estimates about population sizes and migration rates based on genome-wide loci analysis are crucial. Our study lines up in a number of publications demonstrating the isolation, low genetic diversity, and potential small population sizes of the peripheral *S. infraimmaculata* populations in Israel amplifying the need for conservation measures.

Acknowledgements. We feel deep grief at the loss of our colleague Leon Blaustein, a great scientist and wonderful person. We would like to thank Rabea Dabous, Eitan Nisim and the Rangers from the Tel Dan Nature Reserve for providing access to the reserve. Special thanks to Sergé Boegarts for helpful discussions. We thank Meike Kondermann and Christian Wartenberg for conducting laboratory work and Daniel J. Goedbloed for helping in fieldwork. We are grateful for assistance and permits issued by the Israeli Nature and Parks Authority (INPA, 2013/40174, 2013/40284

and 2014/40672). The German-Israel Project-cooperation (DIP, DFG reference number BL 1271/1-1, STE 1130/8-1) funded the study.

Author contribution. EK, SS, LB developed the concept. EK and AH conducted the fieldwork. KP and FL performed the statistical analyses. KP wrote the manuscript with input by SS.

Supplementary material. Supplementary material is available online at:

<https://doi.org/10.6084/m9.figshare.13221008>

References

- Addis, B.R., Lowe, W.H., Hossack, B.R., Allendorf, F.W. (2015): Population genetic structure and disease in montane boreal toads: more heterozygous individuals are more likely to be infected with amphibian chytrid. *Conserv. Genet.* **16**: 833-844.
- Bar-David, S., Segev, O., Peleg, N., Hill, N., Templeton, A.R., Schultz, C.B., Blaustein, L. (2007): Long-distance movements by fire salamanders (*Salamandra atra*) and implications for habitat fragmentation. *Isr. J. Ecol. Evol.* **53**: 143-159.
- Blank, L., Sinai, I., Bar-David, S., Peleg, N., Segev, O., Sadeh, A., Kopelman, N.M., Templeton, A.R., Merilä, J., Blaustein, L. (2013): Genetic population structure of the endangered fire salamander (*Salamandra atra*) at the southernmost extreme of its distribution. *Anim. Conserv.* **16**: 412-421.
- Bogaerts, S., Sparreboom, M., Pasmans, F., Almasri, A., Beukema, W., Shehab, A., Amr, Z. (2013): Distribution, ecology and conservation of *Ommatotriton vittatus* and *Salamandra atra* in Syria. *Salamandra* **49**: 87-96.
- Dolev, A., Perevolotsky, A., Lachman, E. (2004): Vertebrates in Israel: the red book.
- Earl, D.A., von Holdt, B.M. (2012): STRUCTURE HARVESTER: a website and program for visualizing STRUCTURE output and implementing the Evanno method. *Conserv. Genet. Resour.* **4**: 359-361.
- Evanno, G., Regnaut, S., Goudet, J. (2005): Detecting the number of clusters of individuals using the software STRUCTURE: a simulation study. *Mol. Ecol.* **14**: 2611-2620.
- Fady, B., Aravanopoulos, F.A., Alizoti, P., Mátyás, C., Wühlich, G. von, Westergren, M., Belletti, P., Cvjetkovic, B., Ducci, F., Huber, G., Kelleher, C.T., Khaldi, A., Kharrat, M.B.D., Kraigher, H., Kramer, K., Mühlethaler, U., Peric, S., Perry, A., Rousi, M., Sbay, H., Stojnic, S., Tijardovic, M., Tsvetkov, I., Varela, M.C., Vendramin, G.G., Zlatanov, T. (2016): Evolution-based approach needed for the conservation and silviculture of peripheral forest tree populations. *For. Ecol. Manage.* **375**: 66-75.
- Hendrix, R., Hauswaldt, S.J., Veith, M., Steinfartz, S. (2010): Strong correlation between cross-amplification success and genetic distance across all members of 'True Salamanders' (Amphibia: Salamandridae) revealed by *Salamandra atra*-specific microsatellite loci. *Mol. Ecol. Resour.* **10**: 1038-1047.
- Jakobsson, M., Rosenberg, N.A. (2007): CLUMPP: a cluster matching and permutation program for dealing with label switching and multimodality in analysis of population structure. *Bioinformatics* **23**: 1801-1806.
- Jombart, T. (2008): adegenet: a R package for the multivariate analysis of genetic markers. *Bioinformatics* **24**: 1403-1405.
- Keenan, K., McGinnity, P., Cross, T.F., Crozier, W.W., Prodöhl, P.A. (2013): diveRsity: an R package for the estimation and exploration of population genetics parameters and their associated errors. *Methods Ecol. Evol.* **4**: 782-788.
- Kershenbaum, A., Blank, L., Sinai, I., Merilä, J., Blaustein, L., Templeton, A.R. (2014): Landscape influences on dispersal behaviour: a theoretical model and empirical test using the fire salamander, *Salamandra atra*. *Oecologia* **175**: 509-520.
- Kumar, S., Stecher, G., Tamura, K. (2016): MEGA7: molecular evolutionary genetics analysis version 7.0 for bigger datasets. *Mol. Biol. Evol.* **33**: 1870-1874.
- Leigh, J.W., Bryant, D. (2015): popart: full-feature software for haplotype network construction. *Methods Ecol. Evol.* **6**: 1110-1116.
- Markert, J.A., Champlin, D.M., Gutjahr-Gobell, R., Grear, J.S., Kuhn, A., McGreevy, T.J., Roth, A., Bagley, M.J., Nacci, D.E. (2010): Population genetic diversity and fitness in multiple environments. *BMC Evol. Biol.* **10**: 205.
- Martel, A., Blooi, M., Adriaensen, C., Van Rooij, P., Beukema, W., Fisher, M.C., Farrer, R.A., Schmidt, B.R., Tobler, U., Goka, K., Lips, K.R., Muletz, C., Zamudio, K.R., Bosch, J., Lötters, S., Wombwell, E., Garner, T.W.J., Cunningham, A.A., Spitzen-van der Sluijs, A., Salvidio, S., Ducatelle, R., Nishikawa, K., Nguyen, T.T., Kolby, J.E., Van Bocxlaer, I., Bossuyt, F., Pasmans, F. (2014): Recent introduction of a chytrid fungus endangers western Palearctic salamanders. *Science* **346**: 630-631.
- Miró, A., O'Brien, D., Hall, J., Jehle, R. (2017): Habitat requirements and conservation needs of peripheral populations: the case of the great crested newt (*Triturus cristatus*) in the Scottish highlands. *Hydrobiologia* **792**: 169-181.
- Palacín, C., Martín, B., Onrubia, A., Alonso, J. (2016): Assessing the extinction risk of the great bustard *Otis tarda* in Africa. *Endanger. Species Res.* **30**: 73-82.
- Papenfuss, T., Disi, A., Rastegar-Pouyani, N., Degani, G., Ugurtas, I., Sparreboom, M., Kuzmin, S., Anderson, S., Sadek, R., Hraoui-Bloquet, S., Gasith, A., Elron, E., Gafny, S., Kiliç, T., Gem, E., Kaya, U. (2009): *Salamandra atra*. IUCN Red List Threat. Species 2009.

Polato, N.R., Gray, M.M., Gill, B.A., Becker, C.G., Casner, K.L., Flecker, A.S., Kondratieff, B.C., Encalada, A.C., Poff, N.L., Funk, W.C., Zamudio, K.R. (2017): Genetic diversity and gene flow decline with elevation in montane mayflies. *Heredity* (Edinb). **119**: 107-116.

Pritchard, J.K., Stephens, M., Donnelly, P. (2000): Inference of population structure using multilocus genotype data. *Genetics* **155**: 945-959.

R Core Team (2019): R: a language and environment for statistical computing.

Reis, T.S., Ciampi-Guillardi, M., Bajay, M.M., Souza, A.P. de, Santos, F.A.M. dos (2015): Elevation as a barrier: genetic structure for an Atlantic rain forest tree (*Bathysa australis*) in the Serra do Mar mountain range, SE Brazil. *Ecol. Evol.* **5**: 1919-1931.

Rosenberg, N.A. (2003): distruct: a program for the graphical display of population structure. *Mol. Ecol. Notes* **4**: 137-138.

Segev, O., Polevikove, A., Blank, L., Goedbloed, D., Küpfer, E., Gershberg, A., Koplovich, A., Blaustein, L. (2015): Effects of tail clipping on larval performance and tail regeneration rates in the near eastern fire salamander, *Salamandra infraimmaculata*. *PLoS One* **10**: e0128077.

Sinai, I., Segev, O., Weil, G., Oron, T., Merilä, J., Templeton, A.R., Blaustein, L., Greenbaum, G., Blank, L. (2019): The role of landscape and history on the genetic structure of peripheral populations of the near eastern fire salamander, *Salamandra infraimmaculata*, in northern Israel. *Conserv. Genet.* **20**: 875-889.

Steinfartz, S., Küsters, D., Tautz, D. (2004): Isolation and characterization of polymorphic tetranucleotide microsatellite loci in the fire salamander *Salamandra salamandra* (Amphibia: Caudata). *Mol. Ecol. Notes* **4**: 626-628.

Steinfartz, S., Veith, M., Tautz, D. (2000): Mitochondrial sequence analysis of *Salamandra* taxa suggests old splits of major lineages and postglacial recolonizations of central Europe from distinct source populations of *Salamandra salamandra*. *Mol. Ecol.* **9**: 397-410.

Valbuena-Ureña, E., Oromi, N., Soler-Membrives, A., Caranza, S., Amat, F., Camarasa, S., Denoël, M., Guillaume, O., Sanuy, D., Loyau, A., Schmeller, D.S., Steinfartz, S. (2018): Jailed in the mountains: genetic diversity and structure of an endemic newt species across the Pyrenees. *PLoS One* **13**: e0200214.

Submitted: September 7, 2020. Final revision received: November 8, 2020. Accepted: November 10, 2020.
Associate Editor: Inigo Martínez-Solano.