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Identifying Environmental Refuges ("Coldspots") from Infection by *Batrachochytrium dendrobatidis* of Amphibians in Eastern Europe

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Abstract: Amphibians are the most threatened group of vertebrates. While habitat loss poses the greatest threat to amphibians, a spreading fungal disease caused by *Batrachochytrium dendrobatidis* (Bd) is seriously affecting an increasing number of species. Although Bd is widely prevalent, there are identifiable heterogeneities in the pathogen's distribution that are linked to environmental parameters. Our objective was to identify conditions that affect the geographic distribution of this pathogen using species distribution models (SDMs), with a special focus on Eastern Europe. SDMs can help identify hotspots for future outbreaks of Bd, but perhaps more importantly identify locations that may be environmental refuges ("coldspots") from infection. In general, climate is considered a major factor driving amphibian disease dynamics, but in particular temperature has received increased attention. Here, 42 environmental raster layers containing data on climate, soil and human impact were used. Mean annual temperature range (or 'continentality') was found to have the strongest constrain on the geographic distribution of this pathogen. Using the partial dependence visualization module in the R package 'embarcadero', a number of corresponding coldspots were identified.

Keywords: amphibians; coldspots; *Batrachochytrium dendrobatidis*; fungal diseases; infection; pathogen; distribution; GIS modelling.

1. Introduction

Amphibians are the most threatened group of vertebrates with a third of currently known species endangered with extinction and although habitat loss clearly poses the greatest threat to amphibians, a newly recognized fungal disease is seriously affecting an increasing number of species [1]. This disease caused by the chytrid fungus *Batrachochytrium dendrobatidis* (Bd) has been linked to the declines of amphibian species globally and represents the greatest documented loss of biodiversity attributable to a pathogen [2]. Although Bd is widely prevalent, there are identifiable heterogeneities in the pathogen's distribution that are linked to environmental parameters [3]. In this respect, species distribution models (SDMs) have proven to be useful tools for predicting Bd distribution and elucidating the importance of a wide range of environmental covariates considered to affect Bd occurrence. The first developed Bd SDMs were global in scope [4, 5]. Using SDMs, our objective was to identify conditions that constrain the geographic distribution of this pathogen in Eastern Europe in an aim to identify hotspots for future outbreaks of Bd, but perhaps more

importantly identify locations that may be environmental refuges ("coldspots") from infection [6]. Undoubtedly, both aspects are essential for proactive conservation planning [7].

2. Materials and Methods

Localities for *Bd* were gathered from GBIF (<https://www.gbif.org>) and the literature [8, 9; etc.]. Because many uncertainties are associated with SDM projections, particularly when it comes to building a SDM for a species expanding its home range in a new area, we used for the analysis only records of European localities. In total there were 648 such records. These were filtered out by enforcing a distance of 50 km between records; we used this filtering process because ecological niche models are sensitive to sample bias [10]. In the end, the total number of records was reduced to 116. To build models, environmental values at localities of known occurrence are determined and then used to identify geographic regions that have similar combinations of environmental values. Several types of environmental variables at a geodetic resolution of 5 arc minutes have been used as proxy for the fundamental niche [11]: 1) the Bioclim dataset (<https://www.worldclim.org/bioclim>), 2) several eco-attributes, like: human fragmentation, -accessibility and -appropriation (<https://databasin.org>), 3) the ENVIREM dataset (<https://envirem.github.io/>), and 4) the Global Soil Dataset (<http://globalchange.bnu.edu.cn/research/soilw/>) (see also: [12]). SDMs were generated by employing Bayesian additive regression trees (BART), a powerful machine learning approach. Running SDMs with BARTs has recently been greatly facilitated by the development of an R package, 'embarcadero' [13], including an automated variable selection procedure being highly effective at identifying informative subsets of predictors. Also the package includes methods for generating and plotting partial dependence curves and visualization called spatial partial dependence plots, which reclassifies predictor rasters based on their partial dependence plots, and show the relative suitability of different regions for an individual covariate. Habitat suitability values range from 0 to 1. Model performance was assessed using measures of accuracy: the area under the receiver-operator curve (AUC, [14]) and the true skills statistic (TSS, [15]).

3. Results

Both measures of accuracy showed the SDM performed very well (AUC = 0.92 and TSS = 0.73). The automated variable selection procedure identified an informative subsets of predictors, of which important were continentality ($^{\circ}\text{C}^*10$), minimum temperature of the coldest month ($^{\circ}\text{C}^*10$), Thornthwaite's aridity index [16, 17], pH (measured in a calcium chloride solution), and human appropriation [18, 19]. Temperature is considered one of the most important environmental factors driving chytridiomycosis [20], with its lower thermal limit being below 4°C [21]. Amongst the used covariates, continentality is perhaps the most distinguishable dimension of the climatic niche of *Bd* in the study area, featuring the seasonal amplitude in ambient temperature. From the picture (Figure 1) it is clear that large differences between low temperatures in the cold season and high temperatures in the hot season are limiting factors for the pathogen, with habitat suitability (the 'response') dropping from above 0.6 in the west of the continent to below 0.3 in the east (Figure 2). An indication that low temperatures limit the spread of *Bd* is the ascending character of the partial dependence curve plotted for the minimum temperature of the coldest month, showing a steep rise of habitat suitability (to over 0.5) on approaching the mark of 0°C . Earlier bioclimatic variables associated with precipitation were found to make high contributions to SDMs considering *Bd* [6]. In our case Thornthwaite's aridity index, commonly used for measuring aridity of an area and based on both precipitation and temperature, showed better performance than most other bioclimatic variables, maybe because of its compound character. The partial dependence curve built for this index clearly highlights wet, humid and marginally moist sub-humid (values between 1 and 31, or slightly over 32) climates as suitable for the pathogen. Of soil features pH turned out to be the most contributing to the SDM. Outbreaks of chytridiomycosis may be affected by pH, but the pH optimum (pH 6–7) for *B. dendrobatidis* is not outside common pHs of freshwater systems [21]. Our model indicated an

optimum of around 6.5 where the suitability was the highest. Finally, human appropriation, the only human-related covariate selected for its contribution to the SDM. Human appropriation provides a useful measure of human intervention into the biosphere through the appropriation of net primary production. Our model explicitly points towards areas of greater human intervention as areas more likely to be suitable for Bd.

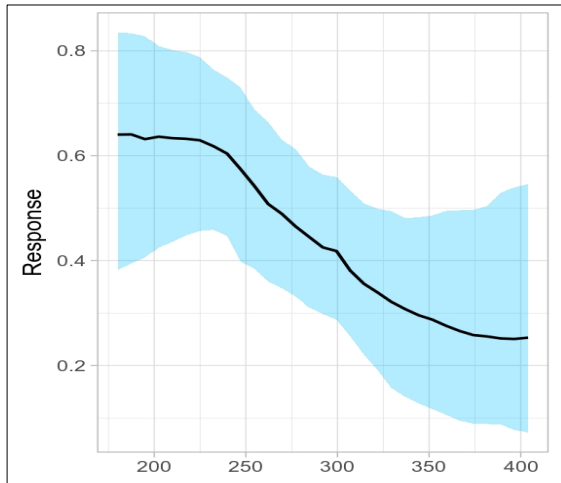


Figure 1. Partial dependence plot for 'continentality'; blue area = 95% confidence interval

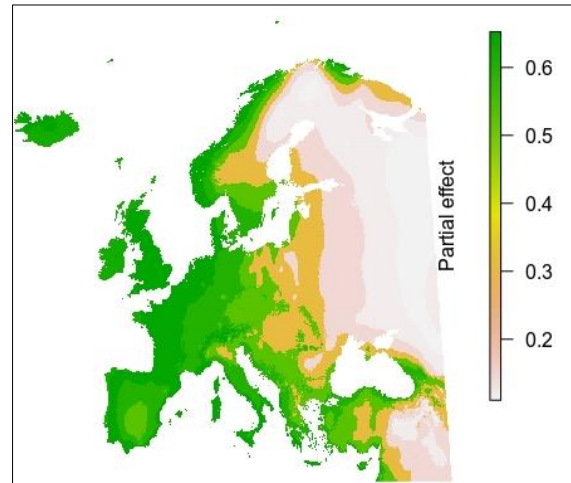


Figure 2. Spatial partial dependence plot for 'continentality', showing the relative suitability of different regions in Europe for *Batrachochytrium dendrobatidis*

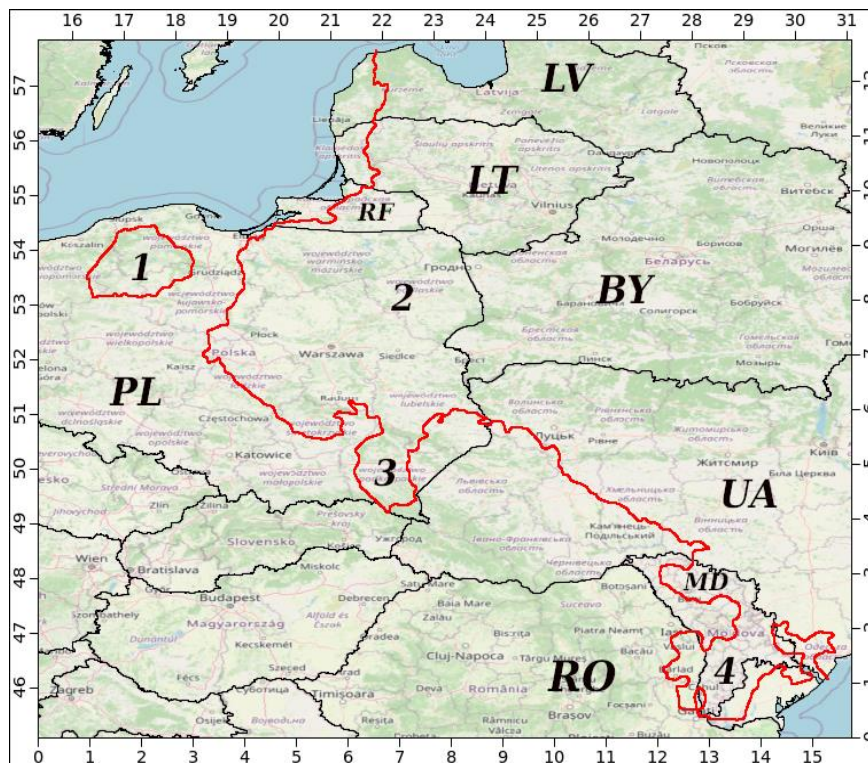


Figure 3. Map depicting contour lines (in red colour) delimiting areas in Eastern Europe where modelled habitat suitability (HS) for *Batrachochytrium dendrobatidis* is above or below the threshold of 0.2; coldspots ($HS < 0.2$) along the major line are numbered: 1 - Northern Poland (enclave); 2 - NE Poland; 3 - Subcarpathia; 4 - Moldovan; country abbr.: BY - Belarus, LV - Latvia, LT - Lithuania, MD - Moldova, PL - Poland, RO - Romania, RF - Russian Federation, UA - Ukraine.

3. Discussion

Using the SDM and an arbitrarily selected threshold of 0.2, we identified locations that may be environmental refuges ("coldspots") for amphibians from infection by Bd in Eastern Europe (Figure 3). These in particular are areas close to the dividing threshold in Poland and Moldova (shared also with Romania and Ukraine) where adequate management and conservation plans for protecting amphibians have to be designed in the first place. Results of the modelling allow assuming that large portions of Latvia, Lithuania and Ukraine, Kaliningrad Province of the Russian Federation will not be favorable for the pathogen; in the case of Belarus this seems to hold for the entire country.

4. Conclusion.

SDMs, on one hand, predict the geographic extent of a species and on the other allow identifying the contribution of habitat covariates in explaining that distribution. Applied to Bd, we find them useful to identify conditions that constrain the geographic distribution of this pathogen and identify locations that may be environmental refuges from infection.

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Conflicts of Interest: The authors declare no conflict of interest.

Abbreviations

The following abbreviations are used in this manuscript:

SDM: species distribution model

BART: Bayesian additive regression trees

Bd: *Batrachochytrium dendrobatidis*

TSS: true skills statistic

AUC: area under the receiver-operator curve

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