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Evaluation ecological niche between Platyceps rhodorachis and P. karelini (Serpentes: **Colubridae) in Iran**

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Abstract

Platyceps rhodorachis (JAN, 1863) and P. karelini (BRANDT, 1838) are both members of the P. rhodorachis complex species, which is widely distributed in Iran and includes many local populations in the country. These two species are molecularly and to some extent morphologically valid. However, hybrids between P. karelini and P. rhodorachis have been described, but so far their ecological differentiation have not been evaluated. In this study, the ecological niche models was predicted for these two members of the P. rhodorachis complex using bioclimatic layers and geographical coordinates. Possible habitat models show the distribution density of these two species in the southern (including some islands in the Persian Gulf) regions, and some areas in northeastern Iran. The results of niche similarity tests (identity and niche overlap tests) based on the criteria of environmental species, in order to assess the degree of species differentiation, indicate the degree of differentiation between these two sister species and raises the possibility of a hybrid belt in southern Iran.

Key words: *Platyceps*, Common Cliff Racer, Ecological niche modeling, Niche differentiation, Snake.

INTRODUCTION

The issue of species delimitation has become a contentious issue over the last half century (De Queiroz, 2007). Biologists believe that the delimitation of species depends on the identification of biological diversity at the species level (Carstens et al., 2013). Numerous operational techniques based on molecular or phenotypic data have been designed to aid the understanding of biological species, however in some cases post- or pre-zygotic barriers are broken (Mayr 1978; Tobias et al., 2010). Because selection acts on phenotypes and any mutualistic units, the concept of ecological species is proposed as a solution to describe species boundaries (Van Valen, 1976). Interest in describing, understanding, and predicting geographic and environmental distributions of species is very old (Grinnell, 1917; Wallace, 2016). According to Dobson (1995), "Biodiversity is the diversity and variability of living organisms and ecological complexes and the sum of the different types of organisms that inherit an area". Understanding how climate change affects the distribution of different animal species has always been one of the main challenges for conservation biologists (Root

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& Schneider, 2006). A set of techniques commonly called species distribution model (SDM) (Guisan & Zimmermann, 2000; Guisan & Thuiller, 2005), and the related Ecological Niche Modeling has experienced significant growth over the past decade and serves as one of the fundamental strategies for determining potential species habitats (Peterson, 2006). The fundamental importance of identifying species distribution areas as a biogeographical and ecological concept and the myriad applications that can be used through these methods explain the growing interest in this field (Peterson *et al.*, 2011). Bioclimatic envelope models is one of the neutral terms that has been developed to use these models (Araújo & Peterson, 2012).

The concept of ecological species, as mentioned before, has a lot to do with the climate and ecosystem of the species distribution region, so the study of species biodiversity in Iran is very important due to the existence of different formations in geological, climatic and soil arrangement (Agapow *et al.*, 2004; Jowkar *et al.*, 2016). Habitat loss is the greatest threat to reptile and amphibian populations, and climate change due to human activities is exacerbating it (Fisher *et al.*, 1968; Devos & Wiles, 1973; Ashrafzadeh *et al.*, 2019). Detection of the protective status of reptiles is faced with a lack of information for various reasons. Modern computer processing and modeling methods with the integration of mechanical models and distribution data of the target species make it possible to gain a better understanding of species limiting factors(Root & Schneider, 2006; Yousefkhani, 2019; Ghelichy Salakh *et al.*, 2020). The MaxEnt principle is one of the most popular methods for modeling species distribution, which is based on ecosystem data and species presence area. The high power of presence prediction, low cost and ease of use have attracted many researchers to use the method (Hijmans *et al.*, 2005; Warren & Seifert, 2011).

Colubrid Snakes make up about two-thirds of the world's snakes (Dessauer, 1967). Racer snakes of the genus *Platyceps* distributed from SW Croatia to Central Asia (Kyrgyzstan), Himalayas (probably westernmost Nepal) and parts of North and Northeast Africa (Schaetti *et al.*, 2014; Sinaiko *et al.*, 2018). So far, 29 species of this genus have been described, of which six species are distributed in Iran (Schaetti *et al.*, 2014; Rajabizadeh, 2018). Among Iranian representatives, *P. karelini, P.ventromaculatus* and *P. rhodorachis* are defined as complex species (Schätti & Mccarthy, 2004). *Platyceps ventromaculatus* (Gray, 1834) is somewhat isolated from *P. rhodorachis* complex in terms of distribution range, but the separation of the distribution range of the other two species has not been considered so far (Yildiz, 2011). All research on *P. rhodorachis* complex has so far focused on taxonomic and phenotypic studies and no study has been done on the extent of distribution and habitat suitability (Perry, 1985; Khan, 1997; Schätti & McCarthy, 2004; Schaetti *et al.*, 2014; Sinaiko *et al.*, 2018).

In this study, we examined the current distribution of two species (*P*.rhodorachis and *P*. karelini) using ecological niche criterion, to examine the degree of ecological differentiation between the two sister species. Also, modeling was used to predict the potential distribution of both species in Iran and the degree of niche space overlap between them. Finally, it is predicted that despite the overlap in the distribution range, the focus area of these two species are different from each other and we will discuss the effect of non-living environmental factors (temperature, precipitation, altitude and slope) on the extent of this separation. Also, we will determine the potential hybridization region by using niche modeling.

MATERIAL AND METHODS

More of the occurrence records are related to the field studies of the research team of Imam Hossein University and Hakim Sabzevari University between 2003 and 2008, which include 126 points. Otherwise, the rest are obtained from the literatures (Schätti *et al.*, 2012; Schaetti *et al.*, 2014). In total, 158 records of presence belonging to both species (50 records for *P. karelini* and 108 records for *P. rhodorachis*) (Appendix 1) were used in this study. The current 19 bioclimatic variables were loaded from WorldClim (www.worldclim.org) with an accuracy of 30 seconds and used to model both

species (Fick & Hijmans, 2017). The slope layer was created using ArcGIS 10.6.1 from the original altitude layer in 30 arc-second resolution and all layers were cut using ArcGIS 10.6.1 for Iran. Correlation matrix for bioclimatic variables was obtained using ENMtools v1.3 software and Pearson correlation coefficient was obtained using ENMTools V 1.3 software and variables with Pearson correlation coefficient higher than 0.75 were removed due to high correlation. (Benesty *et al.*, 2009; Warren *et al.*, 2010). Finally, three bioclimatic variables and two geographical variables were selected for analysis as follows: BIO6 (Min temperature of coldest month of the year (°C), BIO11 (Mean temperature of coldest quarter of the year (°C), BIO18 (Precipitation of warmest quarter of the year (mm), altitude and slope.

The species potential distribution model was implemented using the maximum entropy method in Maxent v3.4.4 (Elith *et al.*, 2011). The ENMeval package in R v4.0.4 was used to obtain the optimal Maxent settings (Muscarella *et al.*, 2014). Maximum 500 iterations, convergence threshold was set to the lowest (0.01) due to the wide range of species distribution, regularization multiplier 0.5 and 15 replicates with cross-validation method (Heidari, 2019). The area under receiver operating characteristic curve (AUC) were considered as accuracy criteria between 0 and 1, the amount of lower than 0.5 represents the random model, and a value closer to 1.0 shows high accuracy of the predicted model (Townsend Peterson *et al.*, 2007).

To measure habitat differentiation between species, niche overlap and identity test were measured using ASCII files on ENMTools 1.3 (Nakazato *et al.*, 2010). In these tests, two required criteria were measured, namely Schoener's D (Giannini & Goloboff, 2010), and Hellinger's-based I (Schoener & Gorman, 1968). Schoener's D calculates the suitable range based on the probability of occupied grid cells, whereas Hellinger's-based I refers to the probability of ecological niche models (Warren *et al.*, 2010). Identity tests were conducted for each species based on 100 pseudoreplicates. Finally, Schoener's D and Hellinger's-based I indices of true calculated niche were compared with the null distribution of 100 pseudoreplicates (Warren *et al.*, 2008; Gholipur, 2018). These indices ranged between 0 (no overlap) and 1 (complete overlap).

RESULTS

Ecological niche modeling results

Based on the literature and records, the distribution range of both species; *P. karelini* and *P. rhodorachis* overlaps, and there are reports of hybridization among their different populations (Туниев, 2000; Schätti *et al.*, 2012). To evaluate the performance of the Maxent model, the area under the curve (AUC) was used. The area under the curve (AUC) is a quantitative indicator to show the efficiency and accuracy of the model prediction (Elith *et al.*, 2006). The AUC values for *P. karelini* and *P. rhodorachis* are 0.812±0.166 (mean ± standard deviation) and 0.845±0.255 respectively, which is acceptable considering the occurrence records. The potential distribution model for *P. karelini* shows the high distribution suitability of this species throughout Iran, except the Zagros and northwestern heights of the country; The main distribution for *P. karelini* is in the southeastern in Sistan and Baluchestan Province and to some extent eastern and northeastern of Iran (Figure 1: A). For *P. rhodorachis*, the Niche distribution model shows a high density in the southern coast strip of Iran, Khuzestan and western Golestan province (Figure 1: B); both species are mainly distributed in coastal strip in the south. BIO6 (Min Temperature of Coldest Month) in *P. karelini* and altitude in *P. rhodorachis* are the most important factors determining the forecasting model (Table 1).

Niche overlap between *P. karelini* and *P. rhodorachis* indicated that their niche similarity was upper than 0.5 (Hellinger's-based I = 0.89 and Schoener's D = 0.65) however, due to the hybridization between these two sister species, this amount is a significant difference. Identity test showed the expected ecological niche interference between two sister species. The results are visible in the diagram in Figure 2. The results of the identity test (i = 0.89, D = 0.65) show the relative separation of the distribution for these two species, this separation can also be seen in the range of the obtained prediction model (Figure 1).

		Percent contribution		Permutation importance	
Variables	Description of variables	P. karelini	P. rhodorachis	P. karelini	P. rhodorachis
BIO6	Min Temperature of Coldest Month of the year (°C)	51.8	20.00	37.4	7.30
BIO11	Mean Temperature of Coldest Quarter of the year (°C)	19.7	32.4	45.7	32.1
BI018	Precipitation of Warmest Quarter of the year (mm)	0.40	0.50	0.00	1.70
Altitude	Height above sea level (m)	26.5	45.6	16.9	58.8
Slope	Land slope	1.60	1.50	0.00	0.00

TABLE 1. Permutation importance and Percent contribution of variables used in MaxEnt model for two species of *P. karelini* and *P. rhodorachis*.



FIGURE 1. Predicted potential distributions of *P. karelini* (A) and *P. rhodorachis* (B) using MaxEnt, Habitat suitability is shown on the map using a gradient and the white circles indicate specimens collecting sites.



FIGURE 2. Results of the identity test. Red arrows refer to the actual niche overlap as calculated by ENMTools (D and I). The bars (with two different patterns) are calculated by replicates with identity test mode.

DISCUSSION

Review of recent papers suggests that ecological theory is rarely explicitly considered (Austin 2007). Species distribution models (SDMs), which are commonly used to obtain hypotheses about the distribution or realization of species potential, has grown significantly since 1995, when this field came into focus. Current linkages between SDM practice and ecological theory are often weak, hindering progress. What is clear is that the use of SDM is essential for applications such as bioprotection planning and uncertainty in SDMs arises from data deficiencies (eg, samples of species occurrences that are small, biased or lacking absences) (Barry & Elith, 2006). Although the conservation status of Colubrid snakes has not been studied due to their widespread distribution and the least concern for this group has been predicted, but due to the overlap of the distribution range in the most suitable habitats of this group with human activities, this group may considered as "endangered" in the not too distant future (Petros Lymberakis *et al.*, 2009). Recently, new studies have been performed on the P. rhodorachis complex and this group was revised and new species recommended (Perry, 2012; Schaetti et al., 2014). Platyceps rhodorachis (Jan, 1865) has the highest range of distribution in this genus and overlaps with most congeners and there are significant cases of crossbreeding with them, especially with *P. karelini* (Khan, 1997; Туниев, 2000; Sinaiko *et al.*, 2018). Based on the literature and records, the distribution range of both species; *P. karelini* and *P.* rhodorachis overlaps, and there are reports of hybridization among their different populations (Туниев, 2000; Schätti et al., 2012), our results confirm this and the range of distribution obtained is very similar to reports and literatures. Platyceps karelini (BRANDT, 1838) has three subspecies, all three of which are distributed in Iran. There is no report in the literature on the distribution of this species in southern Iran and no comprehensive study of the distribution of this species has been done so far in Iran. The available studies are limited to the initial faunistic studies and checklists of snakes in this area, which due to the observed variations in this genus, there is a possibility of misidentification of the species (Rastegar-Pouyani et al., 2011). Comparison of ecological niches among different populations can show the niche differentiation and also detect distinct groups along the whole distribution range within the species complex (Cayuela *et al.*, 2009; Guisan *et al.*, 2013). The predicted distribution pattern in both species has a high overlap with their reported distribution range, however areas of the predicted model are reported for the first time. In P. karelini, Ardabil and Hormozgan provinces, and especially Sistan and Baluchestan, have been identified as compatible areas for the distribution of this species (Figure 1: A). In P. rhodorachis, Ardabil province and southwest of Khuzestan province have been identified as areas with high compatibility for distribution that Ardabil province had not been reported before (Figure 1: B). Altitude is the most important factor determining the likely distribution of *P. rhodorachis*, while in *P.* karelini the minimum temperature of the coldest month has the greatest effect on the prediction model (Table 1).

In the coastal areas of Sistan and Baluchestan province, the temperature is always above zero and due to the high evaporation volume is always sultry and higher than the temperature of the province. The same situation is observed in the coastal areas of Hormozgan province, but due to less monsoon winds, its intensity has decreased. These conditions justify the desirability of *P. karelini* distribution in these areas (Figure 1) (Chaichitehrani & Allahdadi, 2018). *Platyceps rhodorachis* distribution prediction model indicates the high importance of altitude factor, according to the prediction model, this species has the highest distribution merit at low altitudes and close to sea level. The precipitation seasonality in Hormozgan province affects the type of vegetation in the region (Abolhasan & Maryam, 2013; Nohegar *et al.*, 2015), and this issue directly affects the distribution of this species in this area. Probably the distribution of this species in this area is seasonal and reports of hybrids between *P. rhodorachis* and *P. karelini* in this area confirms this argument (Schätti *et al.*, 2012).

The general rule is that the new application of ecological niche modeling greatly facilitates species identification, and thus helps to identify additional species diversity and newer species

speciation events, it also provides a way to identify the possible hybridization region between the two sister species and to explain overlapping distribution patterns. We expect this approach to create new opportunities to identify potential distribution areas and protected areas.

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