

Valdez, V., Álvares, F., Layna, J. F., González, J. L., Herrera, J., Lucas, J. d., . . . Rosalino, L. M. (2022). Raccoon (*Procyon lotor*) in Iberia: Status update and suitable habitats for an invasive carnivore. *Journal for Nature Conservation*, 66, 126142. doi:<https://doi.org/10.1016/j.jnc.2022.126142>

Available at: <https://www.sciencedirect.com/science/article/abs/pii/S1617138122000152>

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## Raccoon (*Procyon lotor*) in Iberia: status update and suitable habitats for an invasive carnivore

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### Abstract

Raccoons are American carnivores, considered invasive across several countries worldwide, especially in Europe. In the Iberian Peninsula, previous studies on raccoons documented several breeding populations in Spain a decade ago and only two confirmed records from isolated individuals in Portugal. Given the need for updating its Iberian distribution and identifying suitable areas with higher invasion risk, we compiled presence records from established breeding populations and isolated individuals. By using a Maxent approach based on breeding records, we forecasted the suitable habitats in Iberia with higher invasion risk for raccoons and identified the related environmental drivers. Overall, we collected 1039 records of raccoon presence throughout the Iberian Peninsula, including 980 records from established breeding populations. Their origin is probably linked to escapes from captivity. Climatic conditions, linked to both drier and wetter environments, and proximity to water bodies were the main predictors of suitable areas for raccoon's expansion from the currently established breeding nuclei in Iberia. The forecasted high probability areas showed a wide, but fragmented distribution concentrated on four main areas: central, central-north, central-east, and north-west Iberia. NW Portugal seems to be the area with higher invasion risk in the country, although field surveys showed no evidence of raccoon presence yet. However, there are several records in Spain near the Portuguese border, comprising isolated individuals and breeding populations. Therefore, it is crucial to ensure regular monitoring of areas with high invasion risk, particularly those near facilities with captive raccoons that often act as a source of feral individuals, to assure early detection and effective control for the expansion of this invasive carnivore.

**Keywords:** Conservation, Invasive Species, Maxent, Portugal, Spain, Species Distribution Models

## Introduction

Invasive species are an increasing ecological problem worldwide, inducing changes in many of the world ecosystems, and being one of the five major causes of biodiversity decline globally (Reid et al., 2005). They can also be responsible for important socio-economic impacts, as often induce damages to agriculture, livestock and other human-related activities (Pyšek & Richardson, 2010). Therefore, prevention is the best approach to avoid the introduction, expansion and establishment of invasive species and reduce the magnitude of their ecological and economic impacts. Otherwise, their control should take place at an early stage of invasion, as these are only more easily controllable if actions target the founder populations, which are often small and more prone to local extinction (Keller et al., 2011; Mehta et al., 2007; Pyšek & Richardson, 2010).

Raccoons (*Procyon lotor*) are carnivores native from North and Central America, widely used in the pet trade and fur market (Biedrzycka et al., 2014). These activities can lead to accidental escapes or even intentional releases of some captive animals into the wild (Beltrán-Beck et al., 2012; Biedrzycka et al., 2014; García et al., 2012; Kauhala, 1996; Mori et al., 2015; Polaina et al., 2021). The scale and impact of these releases are so high that this carnivore is listed as one of the 100 most invasive species in Europe (Winter, 2009). Raccoons are widely distributed as invasive species, with breeding populations occurring in several countries, such as Japan, Russia, Iran, Azerbaijan, Uzbekistan and much of Europe (Beltrán-Beck et al., 2012; Bencatel et al., 2019; Farashi et al., 2013; Frantz et al., 2005; García et al., 2012; Ikeda et al., 2004; Louppe et al., 2019). This carnivore is already widely spread across Central and Eastern Europe, with a reported presence in at least 27 countries (Salgado, 2018). The consequences associated with the raccoon's widespread invasion encompasses several conservation concerns, such as competition with native species for resources (Ikeda et al. 2004), as well as increased predatory pressure upon small vertebrates due to its high diet adaptability, thus adding risks to native species already threatened (Bartoszewicz et al., 2008; Ikeda et al., 2004; Salgado, 2018). In particular, this carnivore poses a worrying threat to the tree and, especially, the ground-nesting birds, due to the high predation of eggs and nestlings (Bartoszewicz et al., 2008; Ellis et al., 2007; García et al., 2012; Ikeda et al., 2004; Kauhala, 1996). Furthermore, raccoons also constitute a threat to public health, being a transmission vector of *Balisascarys procyonis*, a nematode fatal to humans, and also of other dangerous zoonoses that can affect both humans and animals (Bartoszewicz et al., 2008; Beltrán-Beck et al., 2012; Ikeda et al., 2004). Lastly, this species induces economic impacts on human activities, causing damages to poultry and agriculture productions (Beltrán-Beck et al., 2012; Ikeda et al., 2004).

In the Iberian Peninsula, raccoons were first detected in 2001, in the province of Madrid, and since then have been spreading and increasingly recorded across Spain (García et al., 2012; Salgado, 2015). The largest population is distributed between the provinces of Madrid and Guadalajara (central Spain), whose founders seem to be originated from, at least, two distinct episodes of introduction (Alda et al., 2013; García et al., 2012). Records of free-living raccoons have been also detected in other regions located in northern and southern Spain, indicating a series of independent introduction events throughout the country (Fernández-Aguilar et al., 2012; García et al., 2012). However, the published information regarding raccoons in Iberia is sparse and outdated, with the last assessment on its population status dating from almost 10 years ago (e.g. García et al. 2012; Alda et al. 2013), which can result in inadequate management actions, especially targeting a species with high capacity of expanding its range (Louppe et al., 2019). Furthermore, the dense river network in the Iberian Peninsula may act as an important dispersion corridor (Alda et al., 2013; García et al., 2012), increasing the risk of natural colonisations into areas located nearby population edges, including Portugal, a country where the species was thought to be still absent in previous assessments (Salgado, 2018). However, the presence of free-living raccoons was recently reported in Portugal, comprising only two confirmed records of single individuals detected in 2008 and 2014, in the coastal region of NW Portugal (Bencatel et al., 2018, 2019). These events suggest that the current situation of this invasive carnivore in Portugal may be quite different from what is known for Spain. Therefore, a

successful invasion event can still be prevented or controlled, due to the early stage of the invasion process, if areas with high expansion risk are detected and monitored.

Polaina et al. (2021) predicted a future range reduction of raccoons in Europe, whatever the forecasted climate and land-use change. However, half of Iberia was also highlighted as an “uncertain climatic hotspot” for invasive alien terrestrial vertebrates (Polaina et al., 2020), where the outcome of the synergistic effect of high climatic but low environmental adequacy on invasive vertebrate landscape colonization is unknown. Such uncertainty, linked to lack of data on invasive species adaptations and resilience to regional environmental contexts, urges the need to access, based on new and updated presence data, which regions of Iberia constitute a possible available spatial niche for invasive species, such as the raccoon. The great majority of raccoons’ presence records in the Iberian Peninsula are located along rivers and wetlands, which seem to offer an abundance of food and refuge for this invasive carnivore. Native vertebrate preys are often more abundant in these Iberian riverine environments (Rosalino et al. 2009), and these systems have also a generally high abundance of the introduced red swamp crayfish (*Procambarus clarkii*) that became an important nutritional source for many carnivores in Iberia (Gherardi 2006; García et al. 2012; Almeida et al. 2012; Barrientos et al. 2014; Melero et al. 2014).

Considering the current knowledge gaps related to the invasive raccoon presence in the Iberian Peninsula, we aim to: 1) update the current distribution pattern of this species in Iberia, based on an extensive compilation of all available records until 2020, including reference to free-living and captive animals; 2) identify the environmental drivers that determine the presence of raccoon’s breeding populations in Spain and, based on those, predict the regions with a higher expansion risk from the current established breeding nuclei in the Iberian Peninsula; and 3) highlight the most suitable areas for raccoons in Portugal that are more vulnerable to an invasion and should be prioritized for monitoring its presence, and conduct the first field survey in this country to enhance the detection of an early invasion. We hypothesize that: 1) the Iberian distribution of the raccoon is currently wider than in 2010, when the last census was conducted (García et al., 2012), with presence records already located near the Portuguese border; 2) Raccoon presence is mostly determined by climatic conditions and proximity to water bodies with high productivity, given its high water and food requirements (Lotze & Anderson, 1979; Stuewer, 1943); and 3) the northwestern region of Portugal is the most vulnerable area to a raccoon invasion since it presents a high density of permanent rivers (Stuewer, 1943), higher precipitation (Farashi & Naderi, 2017) and few confirmed records of single individuals (Bencatel et al., 2018, 2019).

## Materials and Methods

### Study area

The study area encompasses the continental part of the Iberian Peninsula (SW Europe), including Portugal and Spain (Fig. S1). Iberia’s southwest is mostly characterized by plains, while the northern half and the southeast regions have several mountain ranges. The Iberian Peninsula has three distinct climatic zones: 1) the Atlantic zone, in the northwest, characterized by mild temperatures and abundant rainfall in the wet season; 2) the Mediterranean area, in the South and East, with milder winters with less precipitation and a hot and dry season; 3) the Continental zone, in a more central region, with hotter and drier summers and more rigorous winters (Lorenzo-Lacruz et al., 2013; Sillero et al., 2012). Many major rivers, such as Minho, Lima, Douro, in the north, Tagus in the centre, and Guadiana in the south, have their source in Spain and reach the Atlantic Ocean in Portugal, often forming estuarine systems (Fig. S1). Derived from its privileged position, Iberia is a pathway between Europe and Africa and one of the main Pleistocene glacial refugia in Europe (Gómez & Lunt, 2007), harbouring a diversified fauna, flora, and habitats, including several endemic species (Blondel et al., 2010). The Iberian Peninsula is included in the Mediterranean basin biodiversity hotspot with a high number of endemic species, containing 0.9% of the world’s endemic vertebrates (Myers et al., 2000). Furthermore, Iberia shelters around 50% of the European terrestrial vertebrate species, with a rate of endemism of 31% (Williams et al., 2000).

## Raccoon data collection

We collected records of raccoon's presence from published documents identified through a literature review, but also from other different reliable sources (e.g. research experts, unpublished reports, zoological collections, GBIF, etc.), including data gathered in raccoon eradication programs conducted in Spain. Presence data was only considered when the exact location of detections was available, and we restricted the collected data to the period between 2010 and 2020, in Spain (starting when the last published survey ended the data collection; e.g. García et al. 2012), while we collected all the data available from Portugal. The compiled records of free-living raccoons were classified as associated with breeding populations (e.g. where evidence of pregnant females or cubs were reported) or to occasional detections of isolated individuals, i.e. where there was no evidence of an established breeding population present at the time. The collected data from free-living occurrences comprised records from captures, camera-trap photos or videos, footprints, scats, direct sightings, animals detected with a scent-detection dog, roadkills, radio-tracked animals, and photos (J. F. Layna *unpub. data*; CBC, S.L./Comunidad de Madrid *unpub. data*; S. Palazón/Generalitat de Catalunya *pers. comm.*; Fernández-Aguilar et al. 2012; Generalitat Valenciana 2012, 2013; Layna et al. 2013; Vazquez 2013; Morán et al. 2015; Prieto et al. 2015; Suances et al. 2018; Bencatel et al. 2019; Dana et al. 2020). Dubious records associated with unreliable sources (e.g. unconfirmed sightings without photographic proof) or linked to some degree of uncertainty in raccoon's identification were only considered for Portugal (as possible records, since few data was available), but were excluded from the modelling procedures.

We also compiled information on captive raccoons, both in private (e.g. pet owners) and public collections (e.g. zoos), by contacting Iberian wildlife centres that kept or still keep raccoons in captivity. Whenever possible we obtained information regarding the number of captive individuals, sex, origin, and reproductive status. Furthermore, we identified which of these facilities provide a high abundance of assessable animal food that may attract free-living or recently escaped raccoons to their vicinity. We collected data on raccoon sightings in Portugal from the public by using a citizen science approach. To encourage the general public to provide sighting data, Wilder, a Portuguese online magazine, wrote two articles related to this study, one to publicize and explain the project goals (<https://www.wilder.pt/historias/biologos-estao-a-estudar-risco-de-expansao-do-guaxinim-em-portugal/>) and another to help people know how to identify a raccoon in the wild (<https://www.wilder.pt/seja-um-naturalista/como-identificar-um-guaxinim/>), with a request to send us occurrence records, together with any material that could help us certify the data as a raccoon presence.

## Drivers of raccoon presence in Iberia

We selected 12 environmental variables with documented relevance on this species ecology and believed to influence raccoon's occurrence in the Iberian Peninsula (Table 1). These variables were chosen based on published literature reporting habitat selection and ecological requirements of introduced (including within the Iberian Peninsula) and native raccoon populations (Baldwin et al., 2006; Bartoszewicz et al., 2008; Duscher et al., 2018; Farashi et al., 2013, 2016; Farashi & Naderi, 2017; García et al., 2012; Louppe et al., 2019; Mori et al., 2015; Stuewer, 1943). To complement this process regarding important presence drivers, we gathered expert-based opinions from Spanish wildlife biologists with extensive work on this carnivore ecology and highly experienced in raccoon population monitoring in the Iberian introduced range.

## Species distribution modelling

We used a Maxent approach, based on the theory of maximum entropy applied to presence-only data (De Martino & De Martino, 2018). In the modelling procedure, we selected as presence data the records only corresponding to established breeding populations, because isolated animals recently escaped could provide wrong assumptions regarding habitat preferences. Evidence of reproduction proves that the habitat in which they were recorded is suitable enough for raccoons to breed (i.e. presence of mates and enough

resources to rear offspring). The 12 environmental predictors were incorporated into a Geographical Information System, as different layers, built using the software QGIS (v 3.6.2) (QGIS Development Team 2019) and R (v 3.6.0) (R Core Team 2019), and converted into the same geographic coordinate system (projection EPSG:3035-ETRS89/LAEA Europe) and scale (pixels of 1km<sup>2</sup>), after being rasterized.

To test correlations between environmental variables, we used the *usdm* package (Naimi et al., 2014) in R (v 3.6.0) (R Core Team 2019) to estimate each variable's Variance Inflation Factor (VIF). We chose the threshold value VIF<5 as a criterion to consider that a variable was not inducing multicollinearity (Zuur et al., 2010). Consequently, three variables (mean precipitation of the wettest quarter, annual mean temperature, and mean temperature of the warmest quarter) were removed from the modelling procedure, as they did not meet the VIF<5 criteria. We then assessed the spatial distribution of our data to evaluate geographical clustering. We corrected this sampling bias following the recommended systematic sampling approach described by Fourcade et al. (2014), as this method reduces the spatial aggregation and improves the performance of species distribution models (Fourcade et al., 2014).

**Table 1-** List of environmental predictors used in the modelling procedure for raccoons, their category, acronym (Code name), description, type of influence (+: positive; -: negative), reasoning supporting their selection, and data source.

Category	Environmental Variables	Code Name	Description	Influence	Reasoning	Data Source
Land Cover	Distance to agricultural fields (km)	DAF	Distance to: Rice fields, Vineyards, Fruit trees, and berry plantations, Olive groves, Annual crops associated with permanent crops, Complex cultivation patterns, Land mainly occupied by agriculture with significant areas of natural vegetation, and Agroforestry areas	-	Associated with the high availability of food resources and/or human waste (Ikeda <i>et al.</i> 2004; Bartoszewicz <i>et al.</i> 2008).	Corine Landcover ( <a href="https://land.copernicus.eu/pan-european/corine-land-cover/clc2018">https://land.copernicus.eu/pan-european/corine-land-cover/clc2018</a> )
	Distance to urban areas (km)	DUA	Distance to: Continuous urban fabric, Discontinuous urban fabric, Industrial or commercial units, Road and rail networks and associated land, Port areas, Airports Mineral extraction sites, Dumpsites Construction sites, Green urban areas, and Sport and leisure facilities	-		
	% of tree cover in a grid cell	TC	Percentage of Broad-leaved forest, Coniferous forest, and Mixed forest in each pixel (1 km <sup>2</sup> )	+	Associated to shelter and dispersal (Bartoszewicz <i>et al.</i> 2008; Fischer <i>et al.</i> 2017). Supported by expert-based opinion.	
	Distance to water bodies (km)	DWB	Distance to the nearest river, lake, lagoon, etc.	-	Associated with the high availability of food resources and shelter (Stuewer 1943; Bartoszewicz <i>et al.</i> 2008). Supported by expert-based opinion.	<a href="https://www.diva-gis.org/gdata">https://www.diva-gis.org/gdata</a> ; <a href="https://www.miteco.gob.es/es/cartografia-y-sig/ide/descargas/agua/ried-hidrografica.aspx">https://www.miteco.gob.es/es/cartografia-y-sig/ide/descargas/agua/ried-hidrografica.aspx</a>
	% of riparian vegetation in a grid cell	RV	Percentage of riparian vegetation in each pixel (1 km <sup>2</sup> )	+		<a href="https://land.copernicus.eu/local/riparian-zones/riparian-zones-delineation">https://land.copernicus.eu/local/riparian-zones/riparian-zones-delineation</a>
Topographic	Altitude (m)	ALT	Altitude a.s.l.	-	Associated with less suitable bioclimatic conditions for occurrence (Mori <i>et al.</i> 2015). Supported by expert-based opinion.	<a href="http://srtm.csi.cgiar.org/srtmdata/">http://srtm.csi.cgiar.org/srtmdata/</a>
Climatic	Mean temperature of the warmest quarter (°C)	TWQ	Average values for the years 1970-2000 (for the 1 km <sup>2</sup> pixel)	-	Associated with a wide range of favourable bioclimatic conditions for occurrence (Duscher <i>et al.</i> , 2018; Louppe <i>et al.</i> , 2019).	Fick and Hijmans 2017
	Mean temperature of the coldest quarter (°C)	TCQ		+		
	Annual mean temperature (°C)	AMT		+		
	Annual mean precipitation (mm)	AMP		+		
	Mean precipitation of the wettest quarter (mm)	PWQ		+		
	Mean precipitation of the driest quarter (mm)	PDQ		+		

Modelling analyses were performed using the Maxent software (v 3.4.1) (S. J. Phillips et al., 2019). We used a bootstrap approach with 75% of the records as training data, while the remaining 25% were used as test data and we selected 10,000 background points (Barbet-Massin et al., 2012; Phillips & Dudík, 2008). This procedure was replicated 10 times, and therefore each time we run the model, a different set representing 75%-25% proportion of the records was selected. By averaging the 10 runs we obtained the final model. We chose a regularization multiplier of 1 so that the model would not give an over localized prediction, and the output format of cloglog, as it gives a probability of presence between 0 and 1, being easier to interpret (S. Phillips, 2017). All other settings were kept as default.

We also tested three different bias files to account for two distinct sources of bias. Maxent does not consider uneven sampling effort (Silva et al., 2020). Therefore, to account for a possible highly limited sampling effort in the non-detection areas, we tested two different scenarios. Thus, following Kramer-Schadt et al. (2013) suggestions, we created two bias files. The cells with presence records were assigned the value of 1 (100% of sampling effort) and those with no presence records the values of 0.1 (10% of sampling effort) or 0.01 (1% of sampling effort). Furthermore, many invasive species do not fulfil the Maxent assumption that the species are in equilibrium (Gallien et al., 2012) and, non-detection may be linked to the stage of the invasion process and not to the unsuitability of the location. Thus, we tested the effect of this type of bias by including in the modelling procedure a third bias file. In this approach, we used the output from the global environmental approach model produced by Louppe et al. (2019) for Iberia, which was used as baseline information data from raccoon's native and invasive range. We then weighted our pseudo-absences by applying the formula proposed by Polaina et al. (2020) to the Louppe et al. (2019) raccoon favourability map for Iberia. We assessed models performance and identified the best model based on the averaged Area Under the Curve (AUC) of a Receiver Operating Characteristic (ROC) plot and AUCdiff (AUCtrain–AUCtest), a measure of overfitting (Warren & Seifert, 2011). We also produced a binomial map using maxSSS (which maximizes the sum of sensitivity and specificity) as it is a good threshold for presence-only datasets (Liu et al., 2013, 2016). We then assessed which variables contributed the most for the best model by examining their relative importance according to their permutation importance, which focuses on the drop of AUC when each variable is excluded, providing a precise ranking of the environmental variables chosen (Phillips, 2017; Phillips et al., 2006; Searcy & Bradley Shaffer, 2016). Also, we assessed how variables' variation influenced the probability of raccoon's breeding populations occurrence by creating response curves (Elith et al., 2011; Phillips, 2017).

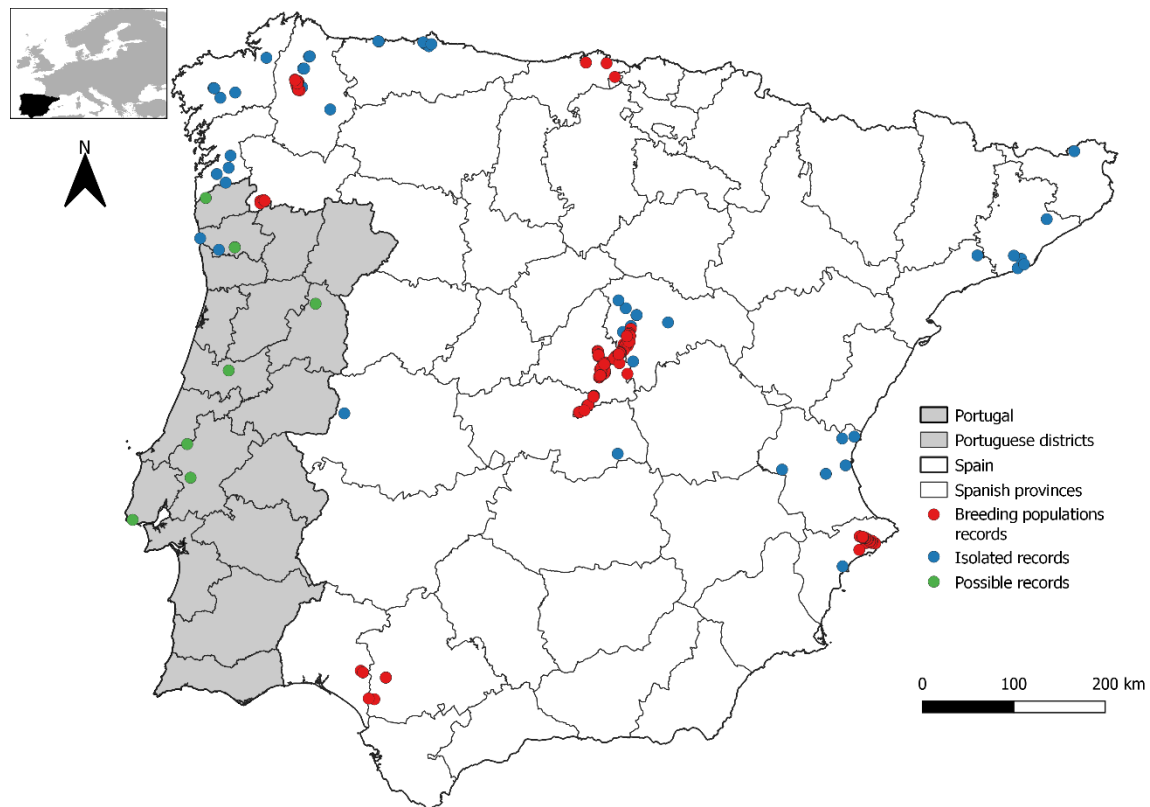
## Field survey to assess raccoon's occurrence and model accuracy in Portugal

Based on the areas identified by the model as having a higher probability of expansion risk as well as the proximity to confirmed and possible records near the Portuguese border, we selected two main areas in Portugal to conduct an intensive field survey to assess this species presence: NW Portugal, along the rivers Ave, Este, Cávado, Lima, Minho, Coura, Tamente and Salas; and in central-eastern Portugal, along the rivers Ponsul and Erges, both Tagus' tributaries (Fig. S2; see Fig. 4 for predicted suitable areas for raccoons). Since raccoons show high activity during summer and autumn to accumulate fat and endure the winter (Mech et al., 1968; Hoffman, 1979), we conducted sign surveys between 13<sup>th</sup> and 17<sup>th</sup> September 2020, optimizing the detection probability. We sampled 24 transects located along the river margins, comprising a total of 40.8 km, in search of presence signs, such as scats and footprints. A major part of these surveys, comprising a total of 31.4 km, was conducted with the help of a scent-detection dog, trained specifically to detect the presence signs of raccoons. Furthermore, we monitored the areas along river Erges (Tagus river watershed, Castelo Branco District) with 18 camera-traps (9 of them located in the riparian vegetation), to enhance the probability of confirming raccoon presence. A total of 13 camera-traps were placed on July 29<sup>th</sup> and the other 5 cameras on August 26<sup>th</sup>, being all active until 24<sup>th</sup> October (total effort of 1984 night-traps). All the fieldwork activities were properly authorized by the Institute for Nature Conservation and Forests – ICNF (Licenses 418/2020/PERTURB and 419/2020/PERTURB).

## Results

### Compiled information on raccoon's presence

We compiled a total of 1039 confirmed presence records of raccoons in the Iberian Peninsula (Tables S1 and S2), from which 980 records (94%) referred to locations where there was evidence of breeding populations (Fig. 1). The remaining 59 records (6%) corresponded to isolated individuals, with no evidence of belonging to breeding populations and which were scattered throughout several Iberian regions. Although records from breeding populations are located mostly in the provinces of Madrid and Guadalajara, in central Spain ( $n=773$ ; 79%; Table. S1), we also gather similar breeding data from several other provinces located in northern and southern Spain (e.g. Lugo, Ourense, Cantabria, Biscay, Toledo, Huelva, Seville and Alicante) (Fig. 1; Table. S1). Most of the raccoon records in Spain correspond to individuals captured in the scope of eradication programs ( $n= 861$ ; 83%; Table S1). In Portugal, we compiled a total of 10 presence records of isolated individuals, including two confirmed and two possible records already reported in previous studies, and six other possible records collected in the scope of this study, which were reported by people who sighted free-living animals with a morphological description compatible with raccoons, although without photographic proof to confirm the veracity of the occurrences (Fig. 1).



*Figure 1- Location of compiled presence records of raccoons in the Iberian Peninsula, including from breeding populations (red circles), detected isolated individuals (blue circles) and possible records of reported isolated sightings in Portugal without a photographic proof (green circles).*

We identified 16 facilities, including zoos and private animal collections, reporting to have raccoons in captivity, in the recent past (last decade) or currently (2020) (Fig. 2; Table S3). Several of these facilities could be associated with the presence of free-living raccoons, since they may act as sources of escaped individuals, inducing new introductory events, or function as important alternative food sources for free-living raccoons (e.g. facilities with animal feeders providing a constant and abundant source of



available food) (Fig. 2). The origin of raccoons in captivity was mostly related to delivered or confiscated individuals by public authorities, which were allocated to these facilities. The capture of free-living individuals in the scope of eradication programs is also an important source of captive raccoons in, at least, two of those facilities. Escapes of raccoons were confirmed to occur in, at least, two Spanish public zoos (in Lugo and Alicante, the latter currently closed), although in other facilities located in the Spanish provinces of Madrid, Toledo, Seville, Biscay, and Asturias similar incidents may have also occurred, yet not confirmed (Table S3). In Portugal, we identified three known locations where raccoons are currently kept in captivity (two public and one private) and an additional private facility where this species was present during the last decade (but not currently), although none of them had known escapes of individuals (Fig. 2).

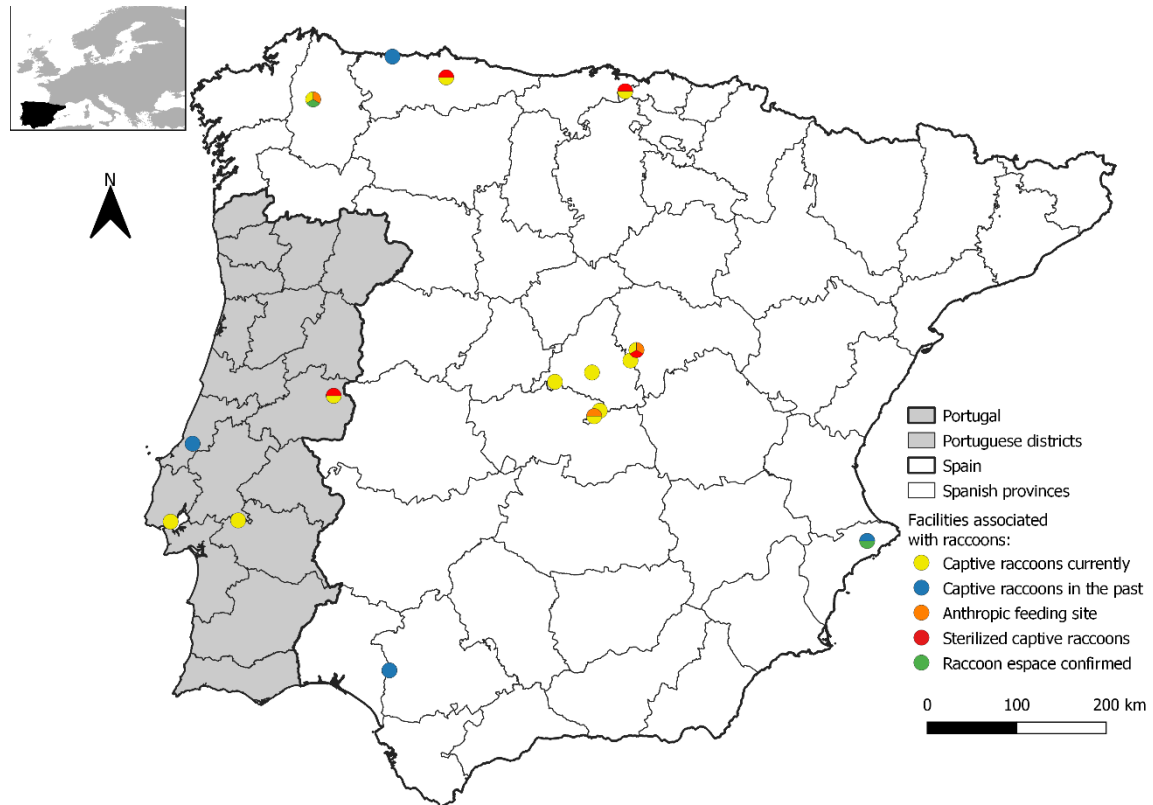


Figure 2- Location of known facilities with captive raccoons currently (2020) or in the past (last decade) in the Iberian Peninsula, including facilities reported to have sterilized individuals, known escapes of captive individuals, and/or provide constant and abundant food resources near areas with free-living raccoon populations (anthropogenic feeding sites).

## Environmental predictors of raccoon's breeding presence

The best model was the one with the bias file in which cells without occurrences were assigned the value 0.01 (i.e. 1% of the sampling effort of occurrence cells). The produced model reached a training AUC=0.9453, AUCdiff=0.0083, and the maxSSS threshold used was 0.6825 (Table S4). According to this model, the presence of raccoons breeding populations in Iberia is mostly influenced by climatic variables and proximity to watercourses. Distance to water bodies (DWB) was the variable with greater importance in the model, reaching a Permutation Importance of 25%, followed by annual mean precipitation (AMP) with 20.4%. Mean precipitation of the driest quarter (PDQ) and altitude (ALT) ranked third and fourth with 14.8% and 14.6%, respectively (Table S5). Both lower and higher precipitation regimes, as well as areas close to water bodies, had an important influence in predicting raccoons' breeding populations, while altitude had a negative effect (Fig. 3).

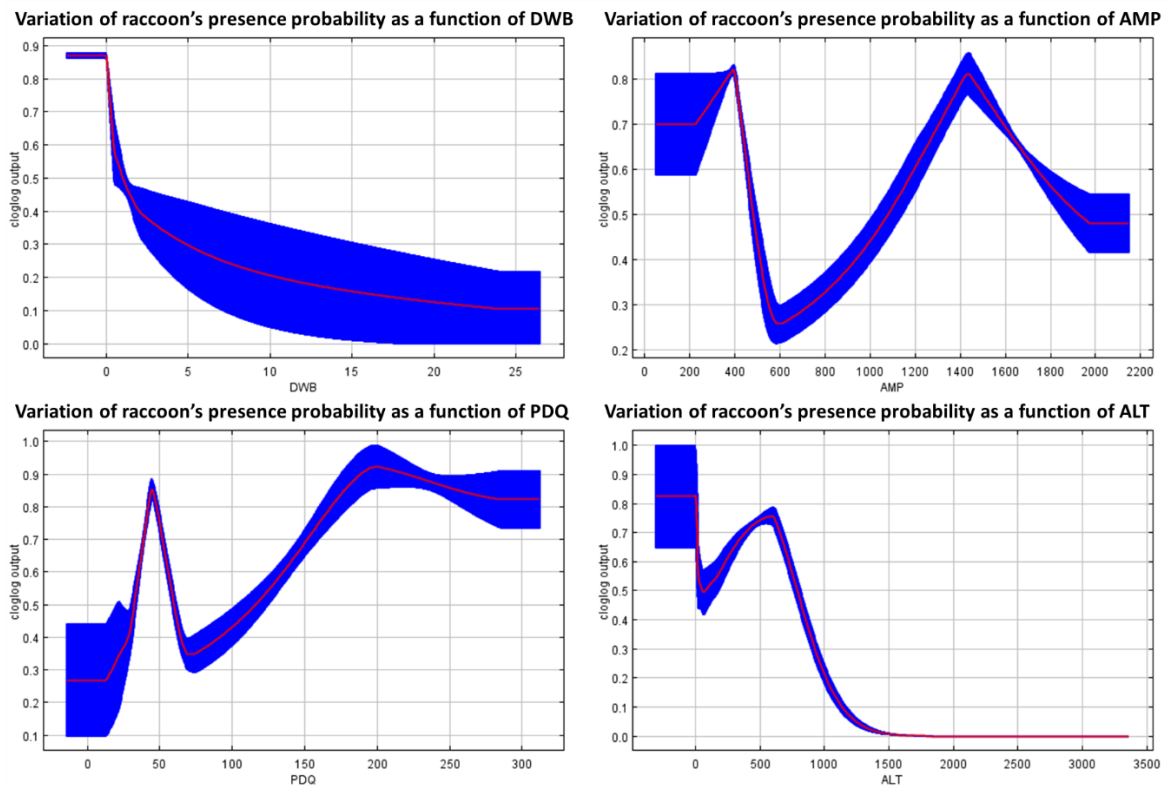


Figure 3- Response curves of the most important variables to raccoons' breeding populations (DWB: distance to water bodies; AMP: annual mean precipitation; PDQ: mean precipitation of the driest quarter; ALT: altitude). The red line indicates the mean response of the 10 replicates, while the blue area represents the standard deviation.

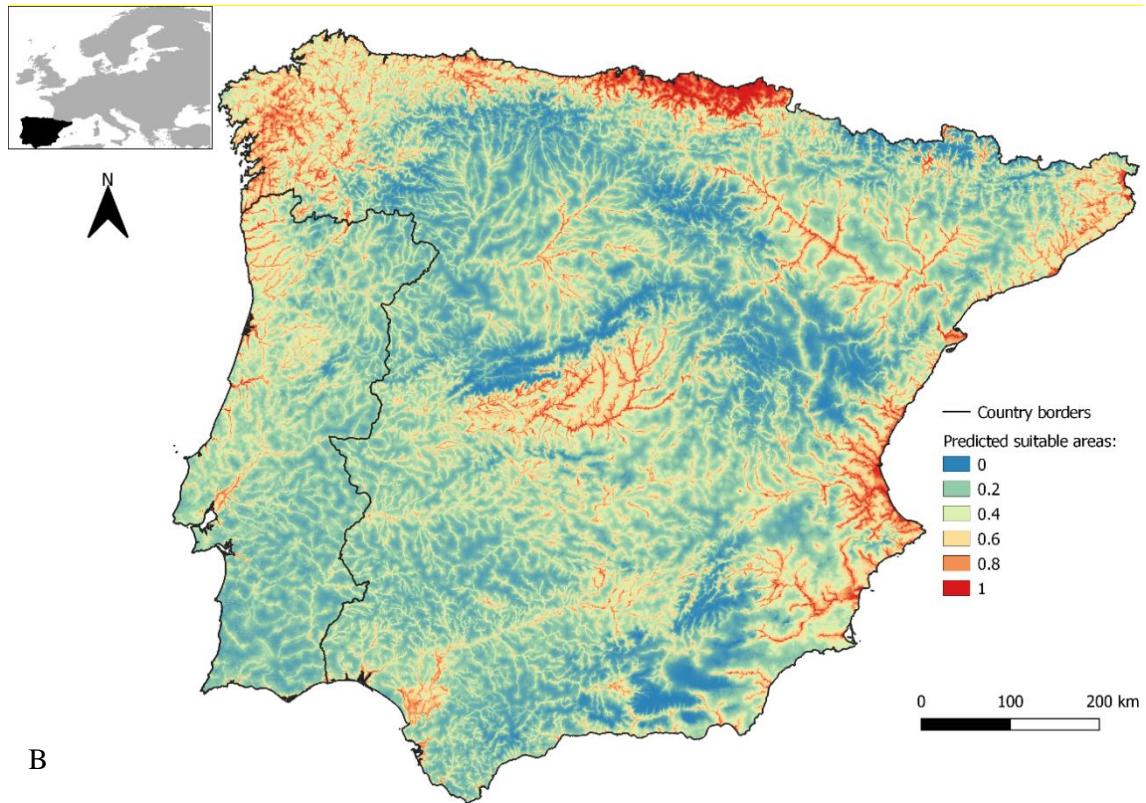
## Predicted areas for raccoon invasion risk in Iberia

Based on the produced model for known breeding populations, the watersheds in the north, northwestern, central, and southeastern regions of the Iberian Peninsula seem to have a higher predicted probability for raccoon's invasion risk (Fig. 4). In Portugal, the main river basins in the northwest have a higher probability of a successful invasion from this species. A comparison between the estimated probability range of raccoons in both Iberian countries highlights that Spain presents more and wider areas with better-predicted conditions for the invasion risk of this carnivore, given the presence of known breeding populations. Furthermore, 92% of all records regarding breeding populations are within the threshold of the suitable habitats ( $>0.6825$ ) predicted by our model.

## Field survey to assess raccoon's occurrence and model accuracy in Portugal

During surveys of the four watersheds in the northwest region (Minho, Lima, Cávado and Ave; Fig. S2) and one in central-east Portugal (Tagus; Fig. S2), no clear evidence of raccoon presence was confirmed. However, the scent-detection dog gave a weak response sign in the Minho river (at the northern border between Portugal and Spain), which might be related to the presence of a raccoon. The dog is trained to perform three distinct signals facing a possible presence of raccoons. In the first and the most intense one, the dog shakes its body for several seconds indicating with accuracy a raccoon presence. The second one happens for less time and indicates a possible presence of a raccoon in the area. As for the third and last, the signal lasts for a smaller period and usually does not reflect the presence of a raccoon, but something else in the area that triggered this slight behaviour by the dog. This latter poorly reliable signal was the one performed by the dog in an area near Minho river, suggesting a very unlikely raccoon's presence, despite the proximity of a reported sighting of this carnivore across the river, in Spain.

A



B

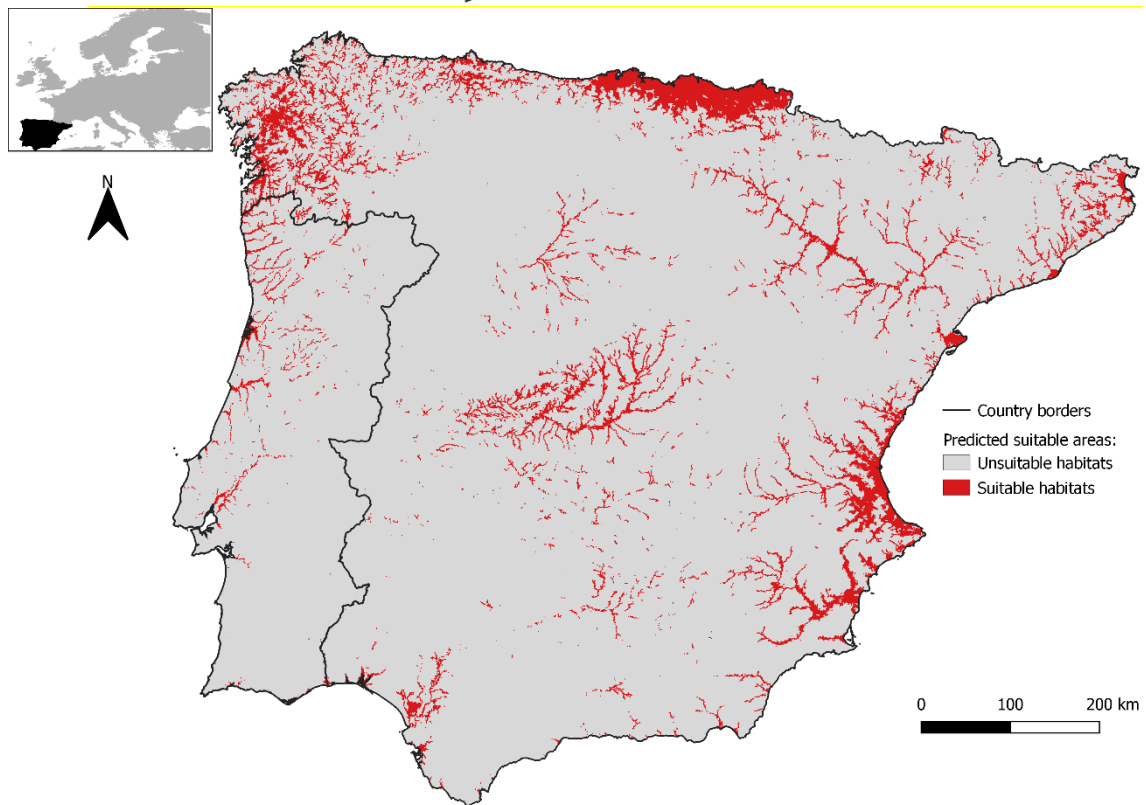


Figure 4- A) Continuous prediction map of suitable areas for raccoons' invasion in the Iberian Peninsula, produced using Maxent. Warmer colours mean a higher probability of invasion risk from known breeding populations. B) Binomial prediction map created using maxSSS as a threshold. Grey indicates the predicted areas unsuitable and red the predicted areas suitable for raccoons.

## Discussion

Our findings allowed us to update the population status of raccoons in the Iberian Peninsula and identify the areas with a higher risk of being occupied by raccoon's breeding populations, in both Iberian countries. This is particularly important for Portugal, where no breeding records are yet known. This information has strong management and conservation implications, by contributing to enhancing the knowledge on the current population status and ecological requirements of this invasive carnivore in the Iberian Peninsula. Overall, we highlighted the presence of breeding raccoon populations in central, northwestern, and southeastern Spain and showed that raccoons' presence seems to be promoted by the proximity to water bodies.

Raccoons' suitable areas are mainly located in Iberia's North, Central and Southeastern regions. According to Louppe et al. (2019), the alien European populations of raccoons did not yet colonize all the environmental niches available, and therefore an expansion of this species range in some European landscapes is still expected. Our results for the Iberian Peninsula seem to corroborate this prediction. Most of the suitable areas identified in our study as high invasion risk regions match those identified by Polaina et al. (2020) as "uncertain climatic hotspots" for invasive alien terrestrial vertebrates in Iberia. These authors showed that central and northern Iberia areas may present climatic conditions to be an invasion hotspot, but lacked the data to confirm it. Our data and modelling results validate their prediction. Although Farashi et al. (2016) predicted that raccoons in Iberia would only encounter favourable occurrence conditions in the north of Spain and central Portugal, our study provides a much more spatially detailed prediction for raccoon's expansion in Iberia, because we did not restrict our modelling to bioclimatic variables, but tested also landcover and topographic drivers. Additionally, we used breeding populations' data to assure that the predicted areas encompass specific characteristics for individuals to establish their range and breed successfully, since isolated records in introduced ranges may be linked to biotic and abiotic conditions (or niches) that do not reflect the ecological requirements for the establishment of this invasive carnivore. However, the results for southeastern Iberia should be carefully interpreted since rivers (one of the most important predictors of raccoon presence; see below) in that region are generally dry in the summer, which could restrain raccoons' presence, and thus reduce the local suitability for this carnivore. We could not include this seasonal variation in river flow into our model since there was no environmental variable available that could provide information on Iberian rivers seasonality.

We updated the information regarding the current raccoon population status in Iberia and assessed facilities with raccoons in captivity. Considering the previous raccoon population assessment conducted in Spain (García et al. 2012), two new breeding populations were now recorded in Galicia (NW Spain), with one of them located near the border with NW Portugal. Several isolated records of free-living raccoons were also reported particularly in Spain, with numerous new locations (e.g. A Coruña, Asturias and Girona), including near the Portuguese border as well, although the real number of records must be higher, as the collected data resulted from opportunistic surveys and not from systematic monitoring covering all Iberian regions. The actual number of captive raccoons in Iberia should also be higher since pet raccoons are difficult to detect and many public zoos did not answer our request for information, which all together prevented the use of this data in our model. Nevertheless, based on this evidence, it seems that the source and persistence of free-living populations of raccoons in Spain can also be highly related to zoological facilities rather than escaped pet individuals as previously suggested (Alda et al., 2013; García et al., 2012). Most presence records are still found in the largest and oldest population, in central Spain, along the Tagus basin (Guadalajara, Madrid, and Toledo provinces; see Fig. 1 and Fig. S2), suggesting a slow geographical expansion of this population, which managed to increase due to the available resources, despite several eradication programs during the last few years (J. F. Layna *unpub. data*; CBC, S.L./Comunidad de Madrid *unpub. data*; Generalitat Valenciana 2012, 2013; Vazquez 2013; Morán et al. 2015; Prieto et al. 2015; Suances et al. 2018). Here, six public facilities keep raccoons in captivity, and some (in Madrid and Toledo) might have been the source of this increasing population, as suggested by genetic data (Alda et al. 2013). Some of these zoological facilities (e.g. Guadalajara) provide an abundant and permanent source of food to their captive animals, attracting many free-living raccoons that can have access to it, thus supporting high densities of this carnivore in the neighbouring areas.

Our results show that, in the Iberian Peninsula, raccoons seem to have a clear preference for areas close to water bodies. Water is known to be an important predictor of raccoons' presence due to the abundance of food resources (e.g. *P. clarkii*, aquatic birds, etc.) and refuge conditions, such as hollow trees (Bartoszewicz et al., 2008; Beasley et al., 2007; García et al., 2012; Gehrt & Fritzell, 1998; Lotze & Anderson, 1979; Stuewer, 1943). Several studies in both native and invasive ranges have found that proximity to watercourses represents a significant and influential variable in predicting raccoon's distribution (Baldwin et al., 2006; Farashi et al., 2013; Farashi & Naderi, 2017; Heske & Ahlers, 2016). Additionally, previous studies have already highlighted that the Iberian rivers play a fundamental role in the dispersion of this invasive species (Alda et al., 2013; García et al., 2012), and therefore might lead to the expansion of its range to the suitable areas identified by our model. Precipitation seems to have also an important role in providing suitable habitats for this species and this pattern is aligned with the findings of previous studies (Farashi et al., 2016; Farashi & Naderi, 2017; Khosravifard et al., 2020; Louppe et al., 2019). Since raccoons prefer environments where water is highly available, it seems logical that precipitation correlates to their distribution. However, in Iberia, breeding populations of raccoons seem to be established in both drier and wetter environments. The preference for areas near water bodies, but with lower precipitation values, might be linked to a higher predation success, when aquatic prey species, such as the highly abundant and widespread red swamp crayfish, a primary prey of raccoons in Iberian ecosystems (García et al., 2012), are restricted to small stretches of rivers, or too shallow areas, due to a low volume of water in riverine systems. A different situation may be linked to wetter environments, in which water is abundant all year. Wetter environments usually have higher plant productivity (Murray-Tortarolo et al. 2017) and can support a more complex land cover, which provides a more stable availability of food and refuge throughout the year, and thus benefiting raccoons. Additionally, we corroborate raccoons avoidance for high altitudes, probably due to less favourable climatic conditions, resulting in food and shelter shortenings (Duscher et al., 2018; Mori et al., 2015).

These findings highlight an imminent threat to Portugal since its northwest region was identified as the most suitable and vulnerable area to a raccoon invasion, although we found no evidence of established breeding populations during field surveys. The Minho river, which constitutes the northwestern border of Portugal with Spain, presents, according to our model, a highly suitable habitat, and thus might constitute a major invasion route. There are several isolated records in Spain near this river (200 m to 16 km), and a possible, but unconfirmed, record on the Portuguese margin. The Lima river might also be an important invasion route since it crosses both countries and harbours a small breeding population in a major tributary in Spain, close to the Portuguese border (5 km), which seems to still be present despite the eradication efforts (Alberto Gil and Xosé Pardavila, *pers. comm.*; See Fig. S2 for rivers' locations). In central Portugal, the Tagus river and its tributaries could also be important in the dispersion of this invasive carnivore towards Portugal since the largest breeding population established in Spain (Madrid and Guadalajara) persists within the upper section of Tagus watershed and an isolated record of a single captured individual was detected in a tributary of this river (western Cáceres) close to the Portuguese border (8 km), although its suitability is lower. Previous studies suggested that large and wide rivers may act as geographical barriers for raccoons dispersion (Cullingham et al., 2009; Mari L. Fischer et al., 2015). However, in central Spain, radio-tracked raccoons have easily crossed the wide Tagus river (J. F. Layna, *unpub. data*), suggesting that Iberian rivers might not constitute natural barriers for a possible raccoon dispersion.

Being generalist carnivores (Kauhala, 1996), raccoons may constitute a wider threat to Iberian native species in the future, namely in Portugal, in case they manage to establish a breeding population. Thus, besides the detection and control of free-living individuals, it would be of extreme importance to properly assess raccoons invasion impact on Iberian ecosystems, since there is scarce knowledge on this carnivore negative effects as an invasive species in Europe (Salgado, 2018). Additionally, if this predator expands its invasive range and occupies the areas selected by our models, particularly in the northern and central regions of the Iberian Peninsula, the impacts on native ecosystems might be significant. The increasing presence of raccoons may cause competition for food and foraging areas with several Mediterranean carnivores of conservation concern, such as the Eurasian otter (*Lutra lutra*) and the European polecat (*Mustela putorius*) (Salgado 2018). Also, predation by raccoons in Iberian landscapes might increase the extinction risk of several threatened species associated with riparian environments, such

as the European pond turtle (*Emys orbicularis*), the Mediterranean pond turtle (*Mauremys leprosa*), aquatic birds (Alvarez, 2008; Blanco & González, 1992; Cabral et al., 2005; García et al., 2012), and, when available, molluscs bivalves (Layna et al., 2013; Simmons et al., 2014), particularly the freshwater pearl mussel (*Margaritifera margaritifera*), considered an endangered species in the Iberian Peninsula (Moorkens et al., 2018).

Monitoring NW Portugal should be prioritized since it borders Spanish areas currently occupied by raccoons and has several rivers with suitable conditions for their occurrence. Thereby, as already implemented in Spain, we recommend the urgent approval by the Portuguese authorities of an action plan targeting raccoons, especially because it is still in a pre-invasion phase. The first step should consider a prevention plan to regularly monitor the areas with high invasion risk identified in our study, especially those close to the border with Spain, where presence records are known. Simultaneously, special attention to the Spanish raccoon population must be maintained to assure an early-warning system that identifies the presence of new individuals close to the Portuguese border. Finally, this plan should also define the regular monitoring of the areas near facilities that keep raccoons in captivity, since they can be a source of escaped animals and free-living breeding populations, as reported in Spain, as well as reinforce the current legislation that imposes the sterilization of captive raccoons. Another fundamental aspect to be included in a Portuguese action plan should be the articulation with all the management actions already in place in several Spanish communities facing the raccoon problem to guarantee an efficient transboundary Iberian management of this “invasive” problem.

## Acknowledgements

We thank the regional governments of Castilla-La Mancha and Madrid, Confederación Hidrográfica del Tajo, Fundación Biodiversidad (Ministerio de Medio Ambiente de España), Zoo de Marcelle, Xunta de Galicia, Santiago Palazón (Generalitat de Catalunya) and Junta de Extremadura for providing records of free-living raccoons in Spain. Regarding information on captive raccoons, we thank to Karpin Fauna, Parque de la Vida, La Fundación Internacional para la Protección de los Animales Raúl Mérida, Zoológico El Bosque, Quinta Layla, Jardim Zoológico de Lisboa, Parque dos Monges, Monte Selvagem and Parque Biológico de Gaia. For the modelling procedures we thank the valuable help from André Silva and João Carvalho. For the fieldwork in Portugal, we thank the help of Laureano Prieto and his dog Gretta, as well as Gonçalo Matias. Finally, we thank to the association “NATIVA” for promoting a crowdfunding initiative to partially fund our fieldwork, to “Wilder” for divulging our study to the public as well as allowing a citizen-science approach to collect raccoon sightings, and to The Navigator Company for all logistic support related with the camera-trapping campaigns. Thanks are due to FCT/MCTES for the financial support to cE3c (UIDB/00329/2020), through national funds, and the co-funding by the FEDER, within the PT2020 Partnership Agreement and Compete 2020. This work was also financially supported by the project POCI-01-0145-FEDER-028204 funded by FEDER, through COMPETE2020 - Programa Operacional Competitividade e Internacionalização (POCI), and by national funds (OE), through FCT/MCTES.

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