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Progress report on modeling geographic distribution of *Rhipicephalus sanguineus* s.l. in Tunisia

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I State of the art

Ticks are the second most important vectors of pathogens to both humans and animals after mosquitos (Dantas-Torres et al., 2012). Since decades, the geographic distribution of ticks is expanding in several parts of the world, this could be explained in part by changes in the climate (Taylor, 2012). Climate changes are more evident in the northern than in the southern hemisphere; they are characterized by an average increase of surface earth temperature of 0.74°C during the last century and a mean worldwide rise in sea level of 1.8 mm per year since 1961 (Semenza and Menne, 2009). In the Mediterranean Basin, the main features of climate change resulted in increased precipitation and drought in winter and summer, respectively (Beugnet and Chalvet-Monfray, 2013). *Rhipicephalus sanguineus* group tick are the most widespread ticks in the world; they are adapted to both temperate and tropical regions (Dantas-Torres et al., 2013).

Rhipicephalus sanguineus s.l. are vectors of several pathogens (Dantas-Torres et al., 2012) namely: *Ehrlichia canis*, *Anaplasma platys*, *Bartonella henselae*, *Mycoplasma canis*, *Mycoplasma ovis*, *Rickettsia rickettsii*, *Rickettsia conorii*, *Babesia* spp. and *Theileria* spp., *Cercopitifilaria* spp., *Dirofilaria repens* ...

Two groups of *Rh. sanguineus* ticks were described, namely tropical and temperate lineages. The tropical lineage is represented by *Rh. sanguineus* collected from tropical regions such as Thailand and Brazil, while the temperate lineage corresponds to *Rh. sanguineus* ticks collected in Spain, France and Italy (Dantas-Torres et al., 2013). This subdivision implies a difference, at the geographic level, in behaviour facing the abiotic factors (Jones et al., 2017).

The main host of *Rh. sanguineus* is dogs but they are collected from several domestic animal species, in close contact with dogs, such as cattle, sheep and goats. In Europe, it was shown that since the sixties, the suitable area for the reproduction of *Rh. sanguineus* expanded to north Europe by 669% (Beugnet et al., 2011).

In Tunisia, domestic animals are exposed to multiple pathogens, leading to high economic losses especially for poor farmers and a handicap to market access. *Rhipicephalus* spp. ticks are competent vectors of wide range of pathogens that have economic impact by affecting animal health like *Anaplasma* spp., and *Babesia* spp., and/or public health impact like Crimean Congo Hemorrhagic Fever virus...etc. Studies on ticks and tick-borne pathogens (TBP) in domestic animals in Tunisia are scattered and need to be considered using an ecological approach. Studying tick-host-environment interactions is required to implement an efficient control program against ticks and TBP.

Tunisia has a big variety of bioclimatic area, ranging from the humid to the Saharan, in the northern and the southern country, respectively. The climate is Mediterranean being wet in the north with annual rainfall reaching 1,500 mm and dry in the south with an average yearly precipitation not exceeding 100 mm. Environmental variables are known to be effective contributors to habitat suitability of almost all tick species. Among those variables, annual precipitation (Bio12), mean temperature of the driest quarter (Bio9), minimum temperature of the coldest month (Bio6), and mean Normalized Difference Vegetation Index (NDVI) are involved in tick geographic distribution and considered in modeling it

(Kessler et al., 2019). While the regional geographic distributions of tick species were investigated by several studies, more localized geographic distribution estimates have never been performed in Tunisia and the effect of environmental factors on TBP has never been studied before.

In the present work, we built a model to predict habitat suitability of ticks of *Rhipicephalus sanguineus* group in order to raise awareness about ticks and tick-borne pathogens in Tunisia. This study could also be extrapolated to neighbouring countries, mainly Algeria and Libya when ground data becomes available.

II Study hypothesis

Biotic (hosts densities, host availability, vegetation) and abiotic factors (temperature, precipitation and hygrometry) influence ticks geographic distribution in Tunisia in the northern, central and southern parts of the country.

III Objectives

The objective of this work is to model *Rhipicephalus sanguineus* s.l. geographic distribution in Tunisia using environmental factors (temperature and precipitation) and biotic factors (hosts density) and to produce recommendations to field veterinarians, medical doctors and decision makers that are specific to the Tunisian context. The resulting risk maps can also be used to draft a policy brief to the attention of officials in the animal health sector.

IV Methodology

4.2. Systematic review

4.2.1. Literature selection

All published papers ($n=40$) and DVM (doctor in veterinary medicine) dissertations ($n=15$) – available at the library of the National School of Veterinary Medicine of Sidi Thabet (Tunisia) - stored in “Database of ticks in livestock species in Tunisia” (<http://geoagro.icarda.org/ticks/>), were reviewed. Additional records were included from the database published as supplementary material by Estrada-Peña and de la Fuente (2016). Only studies reporting *Rh. sanguineus* s.l. group were selected regardless of the animal species from which ticks were collected.

4.2.2. Occurrence data preparation

Each occurrence record was defined by its geographic coordinates (longitude and latitude). From the published papers, coordinates corresponding to *Rh. sanguineus* s.l. occurrence were extracted and checked in *Google Earth* (www.google.com/earth). The data preparation was done according to the

recommendations of Hijman and Elith (2017). For tick occurrences cited without any GPS coordinates, the centroid coordinates of the smallest administrative subdivision of the mentioned locality was extracted. All the GPS coordinates were converted to decimal degree unit. As different published studies mentioned the same localities, duplicated coordinates were considered only once. We also included the records from the field during a 2-years study on ticks and tick-borne pathogens in sheep (Khamassi Khbou et al., 2021). A total number of 78 occurrences coordinates of *Rh. Sanguineus* in Tunisia were used.

4.3. Abiotic variables selection

The most relevant environmental variables affecting ticks' geographic distribution were identified from the literature; they are temperature and precipitation (Vajana et al., 2018). The 19 bioclimatic variables (Table 1) freely available were used in raster format from the WorldClim website (<https://www.worldclim.org/data/worldclim21.html>) version 2.1. with a spatial resolution of 30 arc-seconds and in the un-projected latitude/longitude coordinate reference system (WGS84 datum).

These bioclimatic variables are presented as monthly values of temperature and precipitation estimated from the averages between 1970 and 2000 global data (Fick and Hijmans, 2017).

To avoid overinterpretation due to bias sampling, both road distance and road roughness near the sampling records were also included according to the recommendations of Merow et al. (2013).

Table 1: Bioclimatic variables available from the WorldClim website and used in the present Maxent model

Bioclim variable	Code
BIO2	Mean diurnal range (mean of monthly (max temp - min temp))
BIO4	Temperature seasonality (standard deviation ×100)
BIO10	Mean temperature of warmest quarter
BIO15	Precipitation seasonality (coefficient of variation)
BIO17	Precipitation of driest quarter

4.4. Biotic variables selection

We considered the following domestic animal species: cattle (*Baus taurus*), sheep (*Ovis aries*) and equids (*Equus spp.*) as the main hosts of *Rh. sanguineus* s.l. (Estrada-Peña et al., 2013). As dogs depend largely on food provided by humans (Wandeler et al., 1993), we considered that their geographic distribution is superposed to that of humans, so we only took into account human population geographic distribution.

4.5. Species geographic distribution models

Predicting the risk of tick infestation may improve the effectiveness of tick control programs and reduces their costs for both animal health decision makers and farmers (Beugnet et al., 2009). Mathematical

models combining field, spatial and climatic data were showed to be good for predicting tick and tick-borne pathogens spatial and temporal geographic distribution (Estrada-Peña et al., 2016). Indeed, it was showed that the geographic distribution of *Rh. Sanguineus* is expanding to the northern part of the Mediterranean Basin (Beugnet & Chalvet-Monfray, 2013). In Europe, the most favourable climatic zone to the reproduction of *Rh. sanguineus* expanded by almost 670% since 1960s (Beugnet et al., 2011). In Tunisia, as *Rh. sanguineus* is a vector of several tick-borne pathogens of medical and veterinary importance (*Rickettsia conorii*, *Ehrlichia canis*, *Babesia vogeli*, *Anaplasma* spp....) (Khrouf et al., 2014; M'ghirbi & Bouattour, 2008; Rjeibi et al., 2016), it will be of paramount interest to predict its geographic distribution.

We used a Maxent (maximum entropy) model to predict the geographic suitability of *Rh. sanguineus* s.l. distribution in Tunisia. The main reasons for using the Maxent model are (i) adaptation to the few records we have (n=78), (ii) adaptation to presence only data, (iii) adaptation to R software (Muscarella et al., 2014). Maxent model is one of the several species geographic distribution models (SDM) that measures the relationship between species presence in a given area and its environmental characteristics (Phillips et al., 2004). The Maxent model is largely used for modelling vectors' geographic distribution with good performances compared to other models such as generalized linear models (GLM) (Carvalho et al., 2017). The advantage of using the Maxent models is their flexibility and protection against overfitting (Muscarella et al., 2014). R package ENMeval (Muscarella et al., 2014) was used to model *Rhipicephalus sanguineus* s.l. occurrence (ψx) from presence only data, by maximizing the probability of occurrence according to the logit-linear model, as described by Vajana et al. (2018). Multiple models are generated, with one or a combination of features (linear, quadratic, threshold, hing, etc) multiplied by a coefficient that allows to globally optimize the balance between model fit and complexity (regularization multiplier) (Morales et al., 2017). The best model is the one with the smallest Akaike Information Criterion value (AIC) and the highest Area Under the Curve (AUC), that express the capacity of the model to identify the best environmental factors for tick suitability (Morales et al., 2017).

4.4. Raster and vector data

A vectorized map of Tunisia map was downloaded from DIVAGIS database (URL: www.diva-gis.org) (WGS 84 datum). All raster files were transposed into Africa Albers Equal Area Conic projection to guarantee a constant pixel size and meet the main assumption of the statistical technique used to model *Rhipicephalus sanguineus* s.l. occurrence.

Data on domestic animals geographic distribution in Tunisia was retrieved from Gilbert et al. (2018). Data on human density was extracted from the Center for International Earth Science Information Network - CIESIN, University of Columbia (2018) website (<http://www.ciesin.org/>) .

V Results and discussion

5.1. Modelling *Rhipicephalus sanguineus* geographic distribution according to the biotic factors

Taken together, the density of cattle and sheep, explain 80% of *Rhipicephalus sanguineus* geographic distribution in Tunisia, with a lesser extent the density of equids and humans (Figure 1.a.). According to these biotic factors model, the north of Tunisia is the area with the most *Rh. sanguineus* suitability, followed by the north-eastern region (Figure 1.b.).

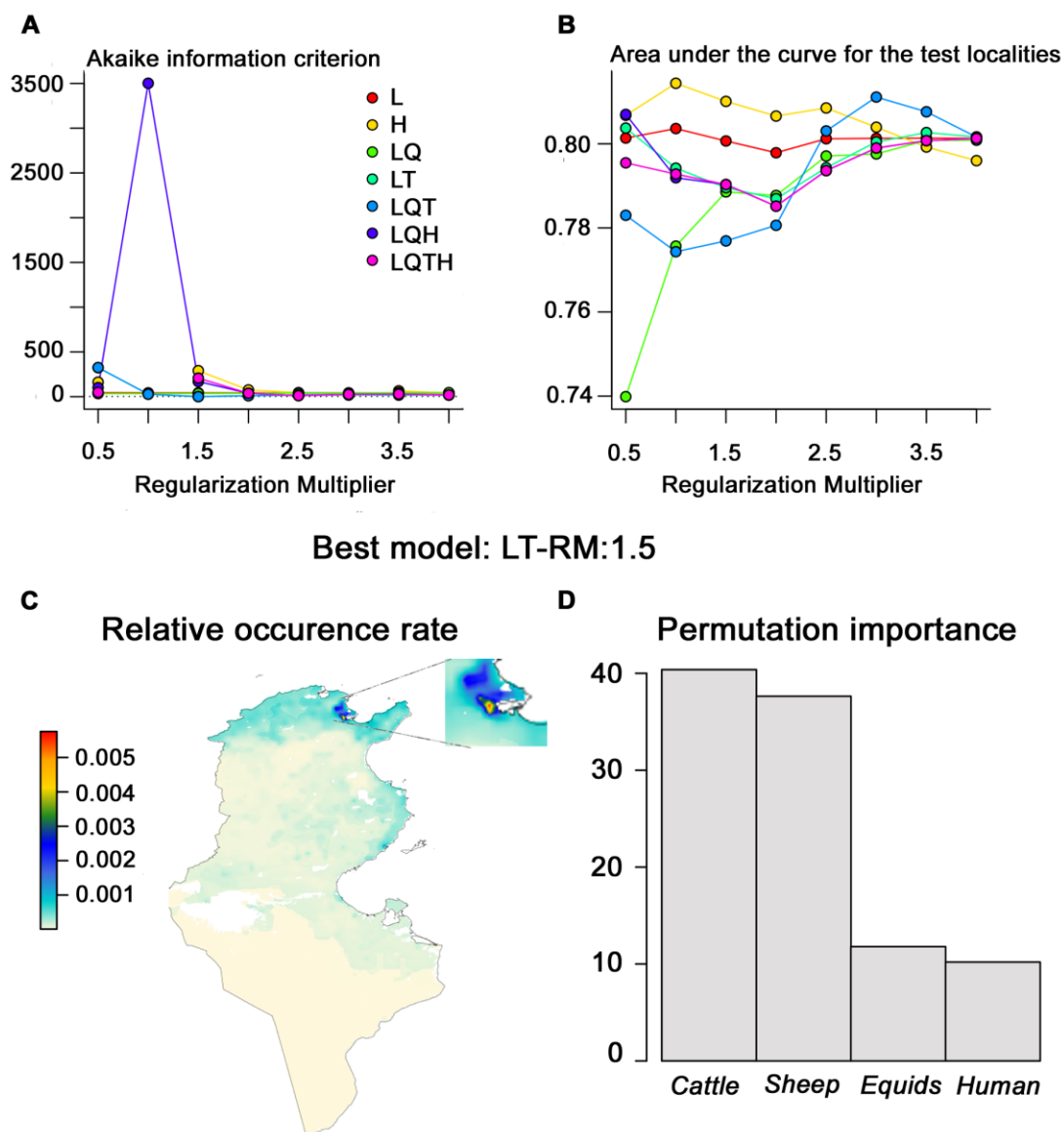


Figure 1: Both evaluation metrics: the AIC in **A** and the AUC (Area under the curve) in **B** show that the best model is a combination between linear and threshold (LT) features and a regularization multiplier (RM) = 1.5. **C:** The map shows the suitability area for *Rhipicephalus sanguineus* s.l. (higher values (warmer colors) indicate higher suitability) with maximum relative occurrence rate in the region

of Tunis and surroundings. **D**: cattle and sheep geographic distribution explain together almost 80% of the *Rhipicephalus sanguineus* suitability.

5.1. Modelling *Rhipicephalus sanguineus* geographic distribution according to the abiotic factors

This model explored the involvement of the main bioclimatic variables in the tick biology (Table 1). The Principal Component Analysis (PCA) showed that the first three axis (PCA1, PCA2 and PCA3) explained almost 97% of the bioclimatic variance (Table 2). The best model was defined by linear and quadratic features and a regularization multiplier=2.5. As shown in Figure 2, the PCA1 explained more than 70% of the model. The PCA1 comprises temperature variables represented by: Bio2: Mean diurnal range (mean of monthly (max temp - min temp)), Bio4: Temperature seasonality, and Bio10: Mean temperature of the warmest quarter. PCA2 and PCA 3 comprise mainly precipitation variables, represented by Bio 15 and Bio17, which appear also as significant determinant in tick geographic distribution. According to these abiotic factors, the most suitable geographic area for *Rhipicephalus sanguineus* s.l. is the northern and eastern coastal regions of Tunisia.

Table 2: Predictors used to model *Rhipicephalus sanguineus* s.l. geographic distribution

Principal component analysis	Bioclimatic variables
PCA 1	Bio10, Bio2, Bio4
PCA2	Bio15, Bio17, Bio4
PCA3	Bio15, Bio17, Bio2

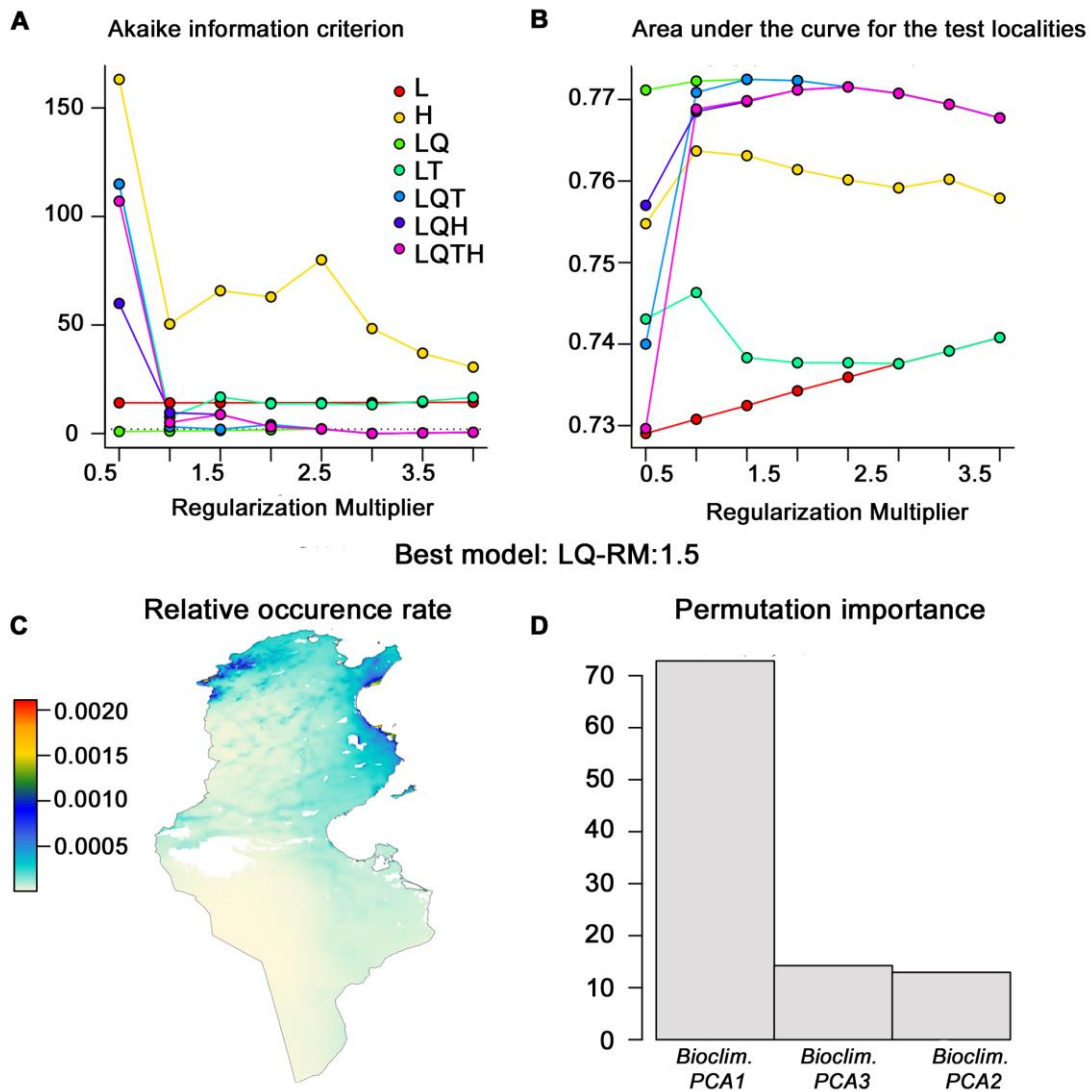


Figure 2: D: Principal components analysis 1, 2 and 3 explain >97% of the bioclimatic (Bioclim.) variance, with PCA1 (Bio10, Bio2 and Bio4) having the maximum contribution. The final model is a combination of both linear and quadratic features (LQ) with a regularization multiplier of 1.5. **C:** The map shows the maximum suitability area for *Rhipicephalus sanguineus* s.l. according to the abiotic factors (proportional to the intensity of the blue colour on the map).

5.3. Global model

The geographic distribution of *B. taurus* (permutation importance > 50%) and *O. aries* (permutation importance = 30%) are the most important variables in the global model (Figure 3). Both climatic variables (PCA3: Bio15, Bio17, Bio2) and human density are of minor involvement in the final model. The final model is a combination of both linear and threshold features (LT) with a regularization multiplier=2.

The suitability map obtained using MaxEnt model, shows that the occurrence of *Rh. sanguineus* s.l. in Tunisia, follows a North-South gradient with the highest suitability in the coastal part of Tunisia, then few suitability foci in the South (marked by the light green colour on the map). The urban region of Tunis (marked in yellow and red colours) and its surroundings (marked in dark blue) is the highest

suitability area for the tick. Another very small suitability area exists near Sfax, in the coastal, central area of the country (Figure 4).

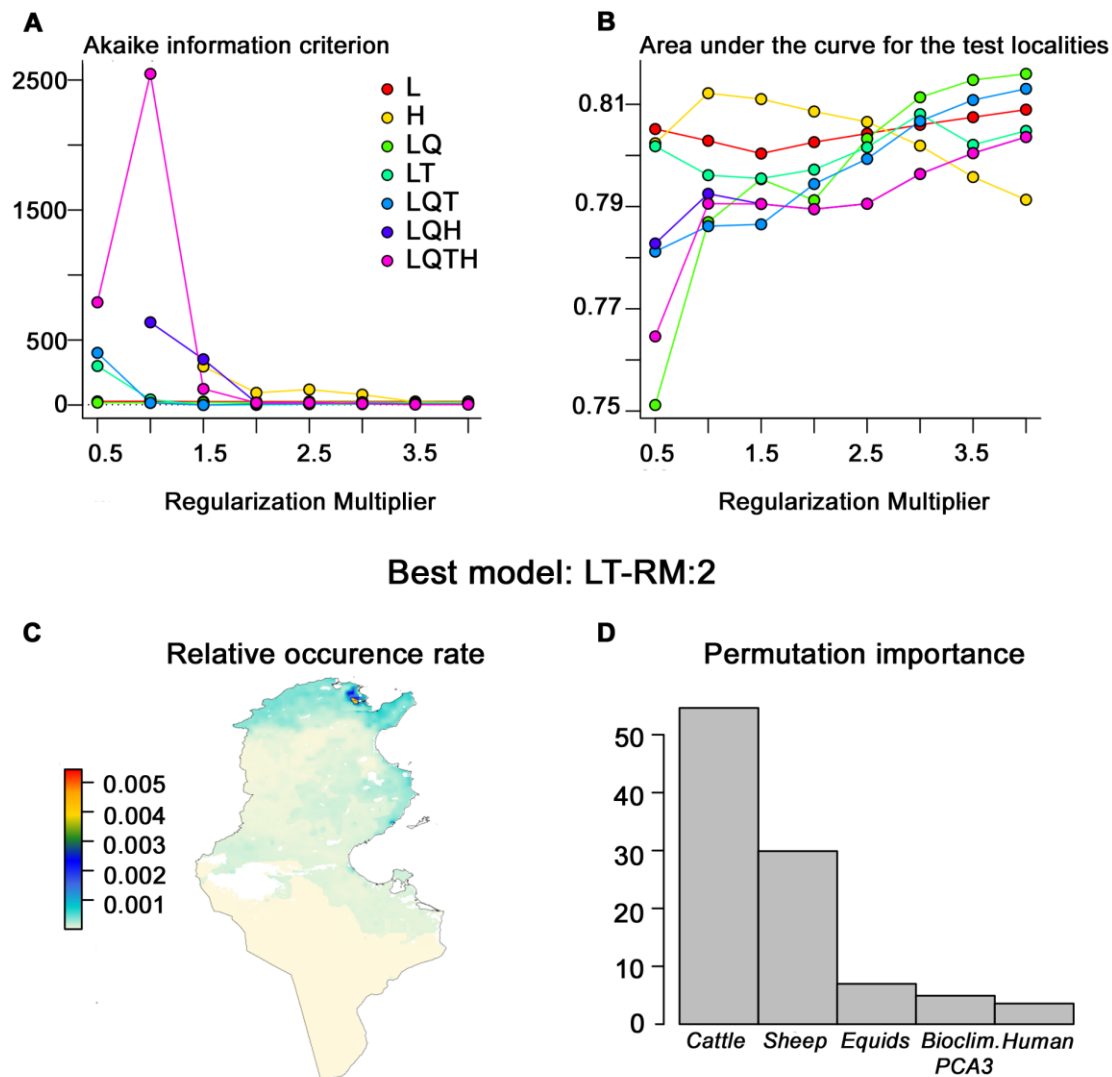


Figure 3: Both evaluation metrics: the AIC in **A** and the AUC in **B** show that the best model is a combination between linear and threshold (LM) features and a regularization multiplier (RM) = 2. **C:** The map shows the suitability area for *Rhipicephalus sanguineus* s.l. (higher values (warmer colours) indicate higher suitability) with maximum relative occurrence rate in the region of Tunis and surroundings and less suitability in coastal area of the north and the east (light green color). **D:** The domestic animals density explains together most of the *Rhipicephalus sanguineus* suitability, and at lesser extent the Bioclimatic variables of the Principal Component Analysis n°3 (Bioclim. PCA3) and human density.

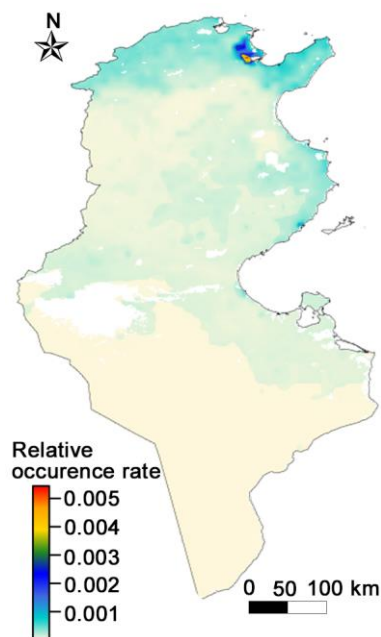


Figure 4: The geographic suitability for *Rhipicephalus sanguineus* group according to biotic and abiotic factors

5.3. Discussion and concluding remarks

The geographic distribution of *Rhipicephalus sanguineus* s.l. showed a North-South gradient, with the highest suitability in northern and coastal regions. Despite the low effect of climate in the geographic distribution of *Rhipicephalus sanguineus* s.l., the geographic suitability is concordant with the highest temperatures moderated by the effect of the Mediterranean Sea. Furthermore, the geographic distribution seems to be driven mainly by the density of domestic mammals rather than human/dog density or climatic variables.

Our findings showed that cattle, sheep and horses contribute largely to *Rhipicephalus sanguineus* s.l. geographic distribution and this is in agreement with the observations of Estrada-Peña et al. (2013). In their analysis, they found that cattle, sheep and horses' contribution as host for *Rhipicephalus sanguineus* group was estimated to almost 36.5%, very close to that of dogs estimated at 37.1%. Indeed, livestock play an important role in carrying *Rhipicephalus sanguineus* tick even where dogs are present.

The main limitation of current work is the size of the data. Further data records are required to better characterize the ecological niche of *Rhipicephalus sanguineus* group in Tunisia and provide adapted preventive measures against this tick and the pathogens it transmits. One of the main lessons of this work is the calibration of the method (in particular the appropriateness of the MaxEnt model to study ticks geographic distribution) generating the risk maps which can be now extended to other important tick species that may pose important public health risks. Ticks of the *Hyalomma* genus is a typical example.

VI Workplan for 2021

The workplan for 2021 will be dedicated to the finalization of data analyses and the publication of a journal paper on *Rhipicephalus sanguineus* s.l. geographic distribution in Tunisia.

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