

## Drought as a Risk Factor to the Occurrence of Tuberculosis-like Lesions Detected during Red Deer Post-Mortem Inspection: A Pilot Study

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### ABSTRACT

Tuberculosis (TB) is one of the infectious diseases which causes more concern worldwide. TB is a multi-host disease, that include domestic and wild animals. From wild species, red deer (*Cervus elaphus*) is considered as a key host in several countries, as in Portugal. In the Mediterranean ecosystem, one of the risk factors to TB infection in red deer population is the climate, including drought periods. The aim of this study was evaluating the influence of drought periods in one Portuguese Mediterranean ecosystem (Idanha-a-Nova county) in the occurrence of Tuberculosis-like lesions observed during post-mortem inspection of hunted wild red deer in this region. In short, our study confirms, statistically, a positive influence of drought periods in the increase of Tuberculosis-like lesions found during post-mortem inspection in wild red deer (p-value<0.05; IC=95%), having the exposed animals almost twice the probability [Odds ratio= 1,96; (1,22; 3,15)] of having these lesions in the following hunting season; pointing out this factor as an alert for game managers to timely implement preventive measures for TB mitigation and highlight the importance of the post mortem inspection as a smart tool to be used to screen sanitary profile of hunted wild animals.

### Keywords

*Cervus elaphus*, drought, large game, red deer, Tuberculosis-like lesions

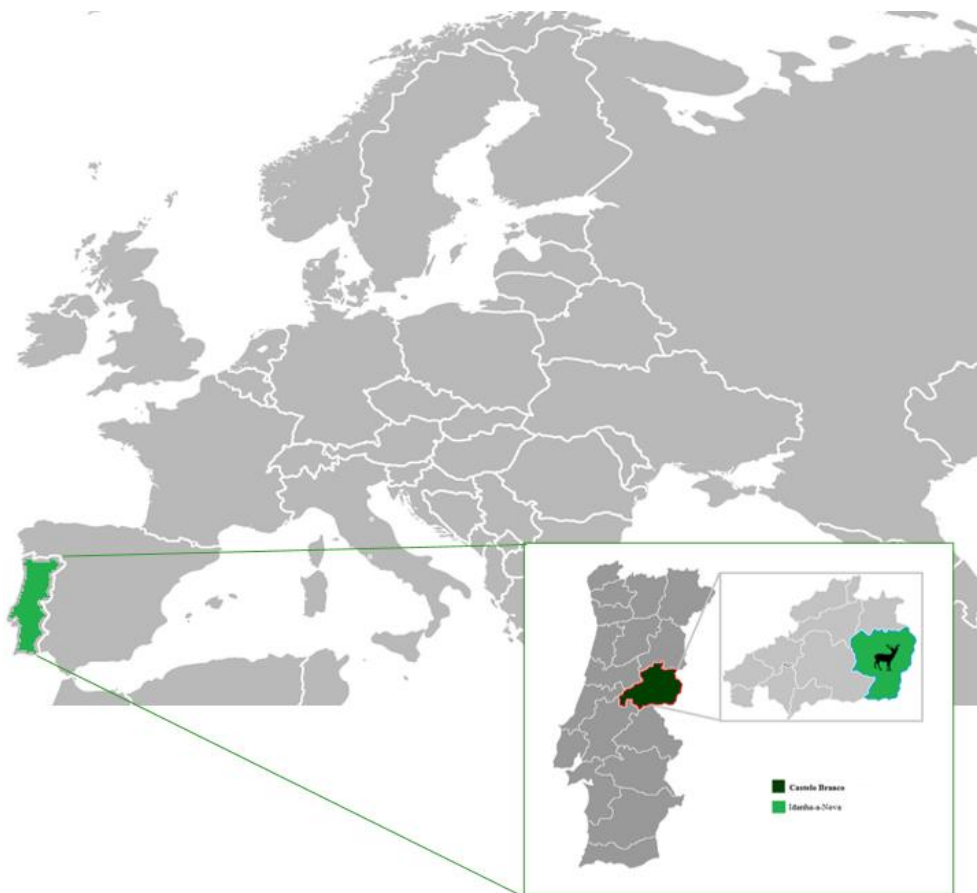
### Introduction

Wild animals are susceptible to infection by most of the same pathogenic agents that affect domestic animals (Palmer, 2013). From those, Animal Tuberculosis (TB), a multi-host disease caused by mycobacteria of Mycobacterium tuberculosis complex (MCT), is one of the infectious diseases which causes more concern worldwide (Humblet et al. 2009; Cunha et al. 2011). Amongst the known wild TB hosts distributed worldwide are animals of Cervidae family (MSU, 2010; Fitzgerald & Kaneene, 2013). In Europe, specifically in the Iberian Peninsula, the most abundant cervid, that also presents the highest occurrence of TB infection, is red deer (*Cervus elaphus*) (Palmer, 2013). Areas with Mediterranean climate where extensive livestock cohabit with many TB wildlife hosts are described as the regions with a higher prevalence of TB both in wild and domestic animals (Vicente et al. 2013). In Portugal, studies in red deer hunted in the southeast of central region, revealed a TB prevalence of 10.3%, ranging from 4.55% to 25.93% infected red deer (Vieira-Pinto et al. 2011). Literature refers some potential risk factors to the TB infection in red deer from Mediterranean ecosystem: low genetic variability, high population density, the habitat, spatial aggregation, and permanence of fenced areas (Vicente et al. 2007, 2013; Queirós et al. 2016). The

existence of a hunting industry in some parts of Iberian Peninsula enhance all these risk factors for red deer population (Vicente et al. 2013).

The Mediterranean climate, also named as dry subtropical climate, is characterized by alternating periods of drought and rainfall. This characteristic can influence the transmission of TB between red deer and another cohabitant specie. According to Vicente et al. (2013), drought can promote a direct or indirect contact between intra and inter-species due to the considerable decrease of food and water supplies during this time.

Due to this fact, the main objective of this study was to analyse the influence of drought in the occurrence of TB infection in hunted wild red deer, during 8 hunting seasons (2009/2010 to 2016/2017), in one of the most important large game hunting area and a TB high-risk area (Idanha-a-Nova county: lat 39° 55'N; long 7° 14'W; 1416,0 km<sup>2</sup>) localized in the southeast of central Portugal. Idanha-a-Nova is a county with a temperate Mediterranean climate, dry summer, and rainy winter with marked drought periods (Vieira-Pinto et al. 2011). Regarding landscape traits, this area has a marked territory of mountains mixed with large plateaus (Figure 1).



**Figure 1.** Map of area of study – Idanha-a-Nova county.

## Methods

During 8 hunting seasons (September to February from 2009/2010 to 2016/2017), Tuberculosis--like lesion (TBL) data was collected during post-mortem inspection of free-ranged red deer hunted in Idanha-a-Nova county. This is a county with high prevalence of TB laboratorial confirmed in game species (Cunha et al. 2011; Vieira-Pinto et al. 2011). For this study, it was considered a Tuberculosis-like lesion (TBL) all caseous or caseocalcareous tubercles with different sizes presents in lymph nodes and organs (Gortazar et al. 2003; Zanella et al. 2008). Animal was considered positive when at least one TBL was found. The post-mortem inspection was always performed by the same veterinarian, following the same procedure already described (Vieira-Pinto et al. 2014) and briefly presented: incision and examination of the retropharyngeal lymph nodes; visual examination and palpation of the lungs; incision and examination of the left bronchial and mediastinal lymph nodes; incision of mesenteric lymph nodes; and incision of precrural and pre-scapular lymph nodes. During this period, a total of 1289 hunted wild red deer were evaluated. None sample from red deer hunted in fenced areas and subject to artificial supplementation was included in this study.

Data about drought index in Portugal was obtained by means of public records published by Instituto Português do Mar e da Atmosfera, I.P. (IPMA, I.P.). This climate index defined, by IPMA (2019), ranges from rainfall scores (extreme, severe, moderate, or weak rainfall) to drought scores (weak, moderate, severe, or extreme drought). This drought index is a complete and multivariate index that include more than one specific climatic factor, based on the concept of water balance taking into account data on the amount of precipitation, air temperature and available water capacity in the soil.

Data was organized in a two-by-two table in order to analyse if the occurrence of a previous drought period before the beginning of each hunting season was a risk factor for the occurrence of cases of TBL in wild red deer. Fisher's exact Test was used to evaluate the significance of the association between variables and Odds ratio to measure the strength of association between them. The probability value ( $p$ )  $<0.05$  was considered as statistically significant. Both were carried using the program EpiTool version 0.5-6.

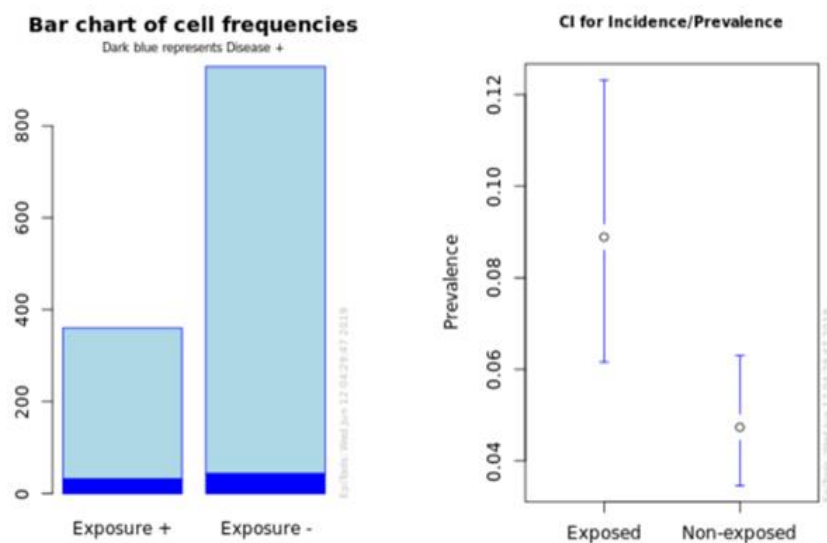
## Results

Of the 8 hunting seasons analysed, 3 (2009/2010; 2012/2013; 2015/2016) followed a scored drought period. During these 3 seasons a total of 360 animals were analysed during post-mortem inspection in the spot. From those, 8,89% (32/360) were TBL positive. In opposite, in the remains 5 hunting seasons, 4,74% (44/929) red deer were identified as positive (Table 1).

**Table 1.** Distribution of red deer TBL positive by hunting season and by exposure to previous drought period.

Previous drought period	Hunting season	Red deer evaluated	Red deer TBL positive	% TBL positive	
<b>Exposed</b>	2009-2010	166	17	32	8,89%
	2012-2013	107	8		
	2015-2016	87	7		
<b>Non-exposed</b>	2010-2011	366	18	44	4,74%
	2011-2012	269	11		
	2013-2014	145	10		
	2014-2015	79	2		
	2016-2017	70	3		
<b>Total</b>		1289	76	5,90%	

Briefly, statistical analyses showed that the pre-existence of a drought period before the hunting season revealed a significant association ( $p < 0.05$ ;  $IC=95\%$ ) with the occurrence of TBL observed in red deer detected during post-mortem inspection (Figure 2). Additionally, after one drought period the probability of TBL prevalence in red deer in the next hunting season was 1,96 [Odds ratio= 1,96 (1,22; 3,15)] times higher compared to a hunting period not previously exposed to a drought period. A previous drought period had a relative risk of 1,88 and a population attributable risk of 0,01.



**Figure 2.** Illustrative charts of frequencies and incidence/prevalence.

## Discussions

Our results, pointing out to the importance of drought on TB infection in wild population of red deer from Idanha-a-Nova county. In this Mediterranean ecosystem, summer season seems to be the main limitative period for the growth, feed, and reproduction of red deer populations. According to several authors, dry weather may affect body condition of red deer, that becomes more susceptible to TB infection or to develop a systemic spread of pre-existent TB (Michel et al. 2006; Vicente et al. 2013). In addition, extensive drought periods promote an aggregation of species, especially in specific points of natural resources, increasing an intra and/or inter-species contact and transmission of infectious agents (Di Marco et al. 2012). In natural conditions is extremely difficult to find a wild population with only one specie and the natural coexistence of several species can be a factor of promotion of the TB transmission, especially in a multi-host system such as Portugal (Queirós et al. 2016).

In conclusion, in Mediterranean ecosystem, drought periods have a marked influence in the dynamic of TBL occurrence in red deer population. Our study contributes to define that drought periods have a positive effect in the increase of the TBL occurrence detected during post-mortem inspection of wild red deer from Idanha-a-Nova county. Adding important information to previous published studies about georeferenced spatial-temporal distribution of wildlife population and TB prevalence in Idanha-a-Nova county (Vieira-Pinto et al. 2011; Vicente et al. 2013; Madeira et al. 2017; Aranha et al. 2021).

These preliminary results have a great use for hunting managers that should predict an increase of TBL occurrence after a drought period and prevent it with timely application of control measures for infectious agents' dissemination. Additionally, the authors would like to underline post mortem inspection as an affordable and cheap tool to be used to screen sanitary profile of hunted wild animals allowing to evaluate the success of control and mitigation measures implemented in field, especially in cases like Idanha-a-Nova county, that is a TB high-risk area. Also, according to Vicente et al. (2013), data from sanitary inspection post mortem collected during hunting seasons may allow to stablish an TBL pattern to the species that may indicate possible source/route of infection (respiratory or oral) and help in locating potential risk points of contamination (Di Marco et al. 2012; Nugent et al. 2015).

## Conclusion

In short, in the Mediterranean habitat, the drought periods must be considered as a risk factor to TBL occurrence detected during the red deer postmortem inspection. This climate data, together with results from postmortem inspection, should always be considered by hunting managers as part of a surveillance system for TB in wild red deer populations.

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