# Modelling High-Risk Areas for African Horse Sickness Occurrence in Mainland China Along Southeast Asia

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# Abstract

African horse sickness (AHS) is a transboundary and non-contagious arboviral infectious disease of equids. Infected Culicoides biting midges can spread the African horse sickness virus, and Culicoides imicola (C.imicola) is one of the important transmission vectors. The disease has spread without any warning from the sub-Saharan Africa towards the Southeast Asian countries. Therefore, it is imperative to predict the distribution of the AHS infection risk along the Sino-Southeast Asian borders. The reported AHS outbreaks were extracted from the archive of the Food and Agriculture Organization from December 22, 2005 to September 1, 2020. The occurrence records of C.imicola were mainly obtained from published literature. Subsequently, the maximum entropy algorithm was used to model AHS and C.imicola separately and to research the relationship among bioclimate variables, land cover characterization, horse distribution density, and the prevalence of AHS infection. Finally, we combined the AHS risk prediction with the suitability map of C.imicola to model the risk areas for AHS occurrence in Mainland China. The models showed the mean area under the curve (AUC) as 0.935 and 0.910 for AHS and C.imicola, respectively. Using jackknife analysis, we determined the important factors affecting the AHS outbreak as horse distribution density, mean temperature of the wettest quarter, and precipitation of the coldest quarter. The mean temperature of coldest quarter contributed most to the occurrence of C.imicola, followed by precipitation of coldest quarter and global land cover characterization. The overlay of the AHS and C.imicola prediction map shows that the areas southwest of Hainan and southeast of Fujian are at high risk of AHS occurrence under current conditions. Furthermore, the border sectors of Yunnan and Guangxi also presented relatively high risk.

# Title: Modelling High-Risk Areas for African Horse Sickness Occurrence in Mainland China Along Southeast Asia

Running title: Risk areas for African Horse Sickness in China

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# Summary

African horse sickness (AHS) is a transboundary and non-contagious arboviral infectious disease of equids. Infected *Culicoides* biting midges can spread the African horse sickness virus, and *Culicoides imicola* (*C.imicola* 

) is one of the important transmission vectors. The disease has spread without any warning from the sub-Saharan Africa towards the Southeast Asian countries. Therefore, it is imperative to predict the distribution of the AHS infection risk along the Sino–Southeast Asian borders. The reported AHS outbreaks were extracted from the archive of the Food and Agriculture Organization from December 22, 2005 to September 1, 2020. The occurrence records of *C.imicola* were mainly obtained from published literature. Subsequently, the maximum entropy algorithm was used to model AHS and *C.imicola* separately and to research the relationship among bioclimate variables, land cover characterization, horse distribution density, and the prevalence of AHS infection. Finally, we combined the AHS risk prediction with the suitability map of *C.imicola* to model the risk areas for AHS occurrence in Mainland China. The models showed the mean area under the curve (AUC) as 0.935 and 0.910 for AHS and C.imicola, respectively. Using jackknife analysis, we determined the important factors affecting the AHS outbreak as horse distribution density, mean temperature of the wettest quarter, and precipitation of the coldest quarter. The mean temperature of coldest quarter contributed most to the occurrence of *C.imicola*, followed by precipitation of coldest quarter and global land cover characterization. The overlay of the AHS and *C.imicola* prediction map shows that the areas southwest of Hainan and southeast of Fujian are at high risk of AHS occurrence under current conditions. Furthermore, the border sectors of Yunnan and Guangxi also presented relatively high risk.

Keywords: African Horse Sickness; Culicoides imicola ; MaxEnt; Modelling

#### 1. Introduction

African horse sickness (AHS) is a transboundary and non-contagious infectious disease of equids, and its pathogenic virus is the AHS virus (AHSV) of the genus *Orbivirus* of the family Reoviridae (P. G. Howell, 1962). AHS is a typical arboviral disease, in the field infected *Culicoides* biting midges (Diptera: Ceratopogonidae) can spread the AHSV (P. S. Mellor, 1996). The clinical signs of AHS are usually classified into four forms (Robin, 2019). Because of the high mortality and the transboundary epidemic potential of AHSV, the World Organisation for Animal Health (OIE) classifies AHS as a listed notifiable disease (OIE, 2020).

AHSV has nine serotypes, all of which are endemic in sub-Saharan Africa, and they periodically invade Europe and Asia (Carpenter, Mellor, Fall, Garros, & Venter, 2017). AHSV serotype 9 (AHSV-9) has caused epizootics in the Asian region (Pakistan and India) and more than 300,000 equines died during 1959–1961 (P. G. Howell, 1960). Then, AHSV-4 caused outbreaks in the Spain, Portugal, and Morocco between 1987 and 1990 (Rodriguez, Hooghuis, & Castano, 1992). In February 2020, the first incidence of AHS was reported in Thailand; this was the first outbreak caused by AHSV-1 outside of sub-Saharan Africa. As of September 1, a total of 17 AHS outbreaks have been recorded with 610 equines infected and with 568 deaths. The total morbidity and mortality of the 17 outbreaks were 22.30% (610/2735) and 20.77% (568/2735), respectively, and the case fatality rate was as high as 93.11%. Recently, Malaysia reported the first AHS outbreak in August 2020. Therefore, the outbreak of AHS in Thailand may pose a major threat to Southeast Asia and even other Asian countries.

The epidemic area and seasonality of AHS occurrence are related to vector epidemiology (Robin, 2019). Culicoides biting midges are important transmission vectors of arboviral diseases worldwide; they transmit the agents that cause diseases in humans and domestic and wild animals. Culicoides imicola (C.imicola) is the only confirmed field transmission vector of AHSV (P. S. Mellor & Boorman, 1995). C.imicola is present across most of the inhabited world, including Africa, southern Europe, and southern Asia (Guichard et al., 2014; Meiswinkel, 1989). The vector is present round the year, and these regions are also potential risk areas of AHS occurrence. Rawlings et al. (Rawlings, Pro, Pena, Ortega, & Capela, 1997) proposed that global climate change may cause C. imicola to expand northward. This will lead to a wider geographical distribution of the AHSV, thereby increasing the risk of exposure to diseases.

The maximum entropy (MaxEnt) model is a machine learning method, which is used to analyze with the presence-only point data (Phillips, Anderson, & Schapire, 2006). It has become the ecological niche modelling tool for species and epidemic disease distribution studies (Alkhamis, Hijmans, Al-Enezi, Martínez-López, & Perea, 2016; Gao & Cao, 2019; Liu et al., 2019). As the AHS vector, maps of *C. imicola* distribution have been

developed for Senegal, Spain, and Portugal (Ciss et al., 2019; Peters et al., 2014; Ramilo, Nunes, Madeira, Boinas, & da Fonseca, 2017). However, there is limited research on the risk distribution for AHS and the suitable habitat for C.imicola in China. The aim of this study is to identify high risk areas for AHS occurrence in Mainland China and to assess the relationship between the AHS infection and environmental factors. The model could help stakeholders to make decisions for C.imicolamanagement and for the epidemiological control of AHS.

# 2. Materials and methods

# 2.1. Research area

China is a vast country located in East Asia; it covers an area of 9.6 million  $\text{km}^2$  and consists of 34 provincelevel administrative units. In this study, Mainland China was included in the research scope. Our area of interest is the Yunnan, Guangxi, Guangdong, Fujian, and Hainan provinces in Southern China along Southeast Asia, located approximately between N 18°–30° and E 97°–120° (Figure 1).

# 2.2. Data collection and processing

We obtained the *C. imicola* presence points (n = 1045) from literature (Leta, Fetene, Mulatu, Amenu, Jaleta, Beyene, Negussie, Kriticos, et al., 2019; Ye, Liu, Li, Qiu, & Liu, 2019) and the Global Biodiversity Information Facility database (*https://www.gbif.org/*). The reported AHS outbreaks were extracted from the archive of Food and Agriculture Organization (FAO) of the United Nations from December 22, 2005 to September 1, 2020 (n = 203).

To determine the influence of the environmental variables on the AHS distribution and C.imicola, we considered 19 bioclimate variables, 12 land cover variables, and global horse distribution density as risk factors in the models (Table 1). Bioclimate variables were obtained from the WorldClim database (http://worldclim.org/version1). Global land cover characterizations were obtained from the archives of the United States Geological Survey Earth Resources Observation and Science center (https://www.usgs.gov/). The global horse distribution density was downloaded from the FAO database (http://www.fao.org/livestock-systems/).

All the occurrence data were rarefied at  $10 \text{ km}^2$  (Radosavljevic & Anderson, 2014) to minimize the spatial autocorrelation using the SDM Toolbox (Brown, Bennett, & French, 2017). Thus, 728 spatially rarefied occurrence records of *C. imicola* and 110 locations of AHS were used in the MaxEnt modelling of this study (Figure 1).

To avoid the multi-collinearity of environmental variables, Pearson correlation analyses were performed using the SPSS 22.0 software. Each pair of variables had a correlation value  $|\mathbf{r}|$  [?] 0.80, and one of the variables was considered to remove from the final model (Ma et al., 2019; Wu, Sharp, Zhao, Shirato, & Jiang, 2007). Finally, two sets of variables were included in the AHS model and the *C.imicola* model (see Table 1). In ArcGIS 10.2, all the environmental variables were resampled to the ASCII raster grids at a resolution of 2.5 arcmin.

# 2.3. MaxEnt modelling

MaxEnt (version 3.4.1) was used for modelling (http:// biodiversityinformatics.amnh.org/open\_-source/maxent/). In the modelling, 25% of the occurrence points were randomly set as test points, and the remaining 75% were training points (Kramerschadt et al., 2013). Each model ran ten repetitions, and the average logical output was used for the final prediction (Conley et al., 2014). To account for the sampling bias (Kramer-Schadt et al., 2013), we created a bias file and 10,000 background points were taken into the MaxEnt models as "pseudo-absence" data.

The AUC of the receiver operating characteristic assesses the predictive performance of the model; a high value (0-1) corresponds to a better predictive model (Phillips et al., 2006). To evaluate the importance of the environmental variables in modelling, the jackknife test and percent contribution of variables were used as indicators of MaxEnt. Finally, we followed the methods used by the previous researchers (Fekede, van

Gils, Huang, & Wang, 2019; Liu et al., 2020), which combined the AHS risk prediction with the *C.imicola* suitability map to model the risk areas for AHS occurrence in Mainland China. The result maps were visualized using ArcGIS 10.2.

# 3. Results

The AUC value is 0.935 in the AHS model and 0.910 for C.imicola (see Figure S1). The contributions of the important environmental variables to the AHS model and C.imicola model are shown in Table 2, and Figure 2 presents the results of the jackknife test. In the AHS model, the horse distribution density was identified as the most important variable for model construction, followed by the mean temperature of wettest quarter and precipitation of coldest quarter. In the model for C.imicola, the mean temperature of coldest quarter contributed the most, and the precipitation of coldest quarter and global land cover characterization were strongly associated with the C.imicola prediction.

Figure S2 presents the predicted probability risk for AHS and the suitable habitat for *C.imicola* in Mainland China. Figure 3 shows the overlay of the AHS and *C.imicola* prediction maps. Clearly, the southwest of Hainan and southeast of Fujian are high risk areas of AHS occurrence under the current conditions. Furthermore, the border sectors of Yunnan and Guangxi present relatively high risks.

#### 4. Discussion

In our models, the temperature and precipitation contributed the most among all the variables. Temperature and rainfall are determining factors in the activity, abundance, and survival of *Culicoides*(P. S. Mellor, Boorman, & Baylis, 2000). Previous studies have demonstrated that temperature is positively correlated with the *Culicoides* activity, and adult vector activity was suppressed at low temperatures (Carpenter et al., 2011; M. Murray, 1987; Searle et al., 2014). At the same time, high temperatures favored larval development leading to faster population growth (Kitaoka, 1982; Vaughan & Turner, 1987). Furthermore, temperature can also influence the AHSV infection rates and the vector *Culicoides* virogenesis rates (Wellby, Matthew, Peter, & Research, 1996). At high temperatures, the vector virogenesis rates were high (Verhoef, Venter, & Weldon, 2014; Wittmann, Mello, & Baylis, 2002); the incidence of vector-bone diseases were low in cold temperatures (Venter & Meiswinkel, 1994). However, laboratory studies have proved that individual *Culicoides* survived for a relatively short lifespan at very high temperatures (Wellby et al., 1996; Wittmann et al., 2002).

On the other hand, rainfall can also influence the activity of C.imicola, which will decrease at low moisture levels (Walker, 1977). In Australia, after rainfall, there was an increase in the feed time of many *Culicoides* midges. The feeding frequency influences the host-biting rates; therefore, the population transformed to one capable of explosive transmission (M. D. Murray, 1986). At a suitable temperature, the abundance of vectors is often more closely related to the rainfall. Diarra found that the largest amount of C.imicolaabundance appeared in the year of the greatest rainfall in Senegal (Diarra et al., 2014). In addition, rainfall influences the spread of AHS by governing the availability of larval habitat and regulating the survival and dispersal of adult *Culicoides* (Purse, Carpenter, Venter, Bellis, & Mullens, 2015). The water content can determine the suitable semiaquatic habitat for the larva (Meiswinkel, 1997), and more rain may produce more or suitable habitats. However, if the habitats are flooded, *C. imicola* will drown (Nevill, 1967).

Horse density and land cover were also key factors in the jackknife test, and there was high probability of *C.imicola* presence in the "Urban and built-up" and "Closed shrublands" areas. The horse density was positively correlated with the AHS occurrence (Riddin, Venter, Labuschagne, & Villet, 2019); other studies (Bakhoum et al., 2016; Fall et al., 2015; Garros et al., 2011) also showed the very complex relationship between the AHS vectors and livestock. Female *Culicoides* depend mostly on blood meals as a food source (de Beer, Boikanyo, Venter, & Mans, 2020; Slama, Haouas, Mezhoud, Babba, & Chaker, 2015). In dense urban and built-up areas, where the urban population and livestock sector is continually expanding, the transmission probability of AHS may be greatly increased.

Closed shrublands are dominated by shrubs or short statured trees, which results in a moist and shaded

microenvironment that is very suitable for livestock breeding; this environment also leads to the development of arbovirus vectors (Diarra et al., 2018; Diarra et al., 2015). In addition, shrublands have highly permeable and organic soils, which are beneficial for the growth and development of larvae. Notably, the male *Culicoides* mainly feed on the soft stems of plants; therefore, they need vegetation cover nearby to survive.

Our area of interest is in the tropical and subtropical monsoon climate, and this study shows that they are almost at relatively high risk of AHS infection. The tropical monsoon climate is characterized by high temperatures throughout the year with two seasons: a dry season and a rainy season. The mean temperature of the coldest month is not less than 16 degC, and there is no frost all year round. The subtropical monsoon climate has high temperatures and rain in summer, and the mean temperature of the coldest month ranges from 0 degC–15 degC. Hainan and Fujian are not extremely cold and neither are they too dry; therefore, the weather is suitable for the survival and development of *Culicoides*. Leta has suggested high suitability for *C.imicola* in the tropical and subtropical area (Leta, Fetene, Mulatu, Amenu, Jaleta, Beyene, Negussie, & Revie, 2019)

Eastern Yunnan is adjacent to Guangxi, in the south of the two provinces, there are many wide valleys and basins, which are mainly covered by low vegetation (small bushes and trees). According to the People's Government of Yunnan Province (*http://www.yn.gov.cn/*), by 2018, the forest area of Yunnan was approximately 23.11 million hectares, and the forest coverage rate was 60.3%. According to the data from the National Bureau of Statistics of the People's Republic of China (http://www.stats.gov.cn/), by 2018, Yunnan is the first largest province in terms of mule production in China; the yields of donkeys and horses are also in the top 10 (in 31 provinces). Guangxi is the fifth largest province in terms of horse production. Yunnan and Guangxi are suitable habitats for *C.imicola* because both these areas are characterized by dense vegetation and high density of livestock. Furthermore, border trade on land is frequent in the Yunnan and Guangxi areas11The 17th China–ASEAN expo will be held in Nanning, Guangxi, China from November 27 to 30, 2020.. Therefore, there may be two pathways for the spread of AHSV: the cross-border movement of infectious hosts and movement of infected *Culicoides* via livestock trade (Faverjon et al., 2015).

China has a thriving modern horse industry with a lot of attention being paid to horse breeding. The Chinese Equestrian Association joined the International Equestrian Federation in 1982. By 2019, there were approximately 2160 equestrian clubs in China. East China where Fujian is located has the largest number of these clubs (Horsemanship, 2019). Hainan is located at the junction of the Pacific Ocean and the Indian Ocean; it connects Northeast Asia to Southeast Asia. After the establishment of the Hainan Equestrian Association in 2015, the construction of a horse culture characteristic town has been completed. Currently, Hainan clearly encourages the development of horse racing as a sport and supports the creation of the international horse culture, sports, and tourism resorts (Yuanyuan & Yu, 2020). The planning and development of the Hainan Pilot Free Trade Zone will further promote economic and trade cooperation with overseas markets (Meng et al., 2018). Therefore, in addition to the infrastructure, such as racecourses, Hainan and Fujian need to further build an epidemic-free area, especially an AHS-free area.

Fujian and Hainan are coastal provinces of southern China; they have well developed maritime trade, and their sea ports are very important. The horses needed for modern equestrian and horse racing are mostly bred in developed countries such as Europe, North America, and Australia (Power, 2014). A major risk pathway for AHS is the movement of infectious equids, and for example, the AHS outbreak in Europe (1987–1990) was caused by the import of infected zebras from Namibia to Spain (P. S. Mellor & Boorman, 1995; Rodriguez et al., 1992). Previous studies have also shown that the AHSV outbreak in Iran in 1959 was associated with riverine trade (P. G. Howell, 1960). An additional uncontrollable route of AHS introduction into southern China is through infected *Culicoides* midges carried by the wind from AHS-affected regions. *Culicoides* midges can be transported from long distances (Reynolds, Chapman, & Harrington, 2006), and they may be blown as far as 700 km when the wind speed is suitable (Pedgley & Tucker, 1977), and there are no orographic obstacles (Bishop, Spohr, & Barchia, 2004). The wind carries the infected *Culicoides* from the epidemic area to other areas. After landing, these insects bite the local equids who, in turn, spread AHSV.

Although AHS has long been endemic in the sub-Saharan Africa, the transmission of AHSV is heterogeneous

spatially and temporally and occurs across a wide area related to the interactions among the vector, host, and environment. AHS is an epidemic with the highest fatality rate in horses; the relevant departments should pay enough attention to prevent AHS from invading China from Southeast Asia.

This research has certain limitations. The mechanism of transmission and infection of AHS is complex; bioclimatic variables, land cover characterization and horse distribution density are only a few of the influential factors. There are other factors, such as horse trade, that should also be considered. Our follow-up study will consider additional factors and provide detailed information.

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# Conflict of interest statement

The authors have no conflicts of interest to declare.

# **Ethics Statement**

The authors confirm that the ethical policies of the journal, as noted on the journal's author guidelines page, have been adhered to. No ethical approval was required as no animal experiment was involved in this study.

# **Data Availability Statement**

The data that support the findings of this study are available in section 2.2 Data collection and processing.

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# Figure legends:

Figure 1 Research area and occurrence of African horse sickness and Culicoides imicola.

Figure 2 Jackknife test of prediction models (A) African horse sickness; (B) Culicoides imicola.

Figure 3 Map of risk probability areas for African horse sickness occurrence in Mainland China.

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