The Eco-Epidemiology of Tick-Borne Hemoparasitic Diseases Using Geographic Information Systems

Mohammad Mehdi Sedaghat,1 Khadijeh Shemshad,2 Karim Kamali,3 Zabihollah Charrahy,4 Akbar Biglarian,5 Javad Rafinejad*1

1. Department of Medical Entomology and Vector Control, School of Public Health, Tehran University of Medical Sciences, Tehran, Iran
2. Department of Medical Entomology, Faculty of Health, Mazandaran University of Medical Sciences, Sari, Iran
3. Department of Entomology, Science and Research Branch, Islamic Azad University, Tehran, Iran
4. Department of Education, Jehad-e-Daneshgahi of Tehran University, Tehran, Iran
5. Department of Biostatistics, University of Social Welfare and Rehabilitation (USWR), Tehran, Iran

Introduction

Ticks and tick-borne diseases are responsible for mortality and morbidity in livestock, wildlife, pets and humans in different parts of the world. However, detection is difficult with multiple infection cases and low infection rates in carrier animals [1-4]. The researches of the diseases distribution within populations have included a spatial factor. New methods, such as geographic information systems (GIS) and spatial statistics methods help epidemiologists to address the spatial prospects of disease and transmission rates more thoroughly and less subjectively [5-8]. The emergence of tick-borne infections in the different parts of Iran has been increased in recent years. In Iran the agent of several ixodid ticks are responsible for different important diseases [9, 10].

Hemoparasitic diseases could have a variety of direct and indirect impacts on ecosystems and economics. The favorite microclimates for the survival and multiplication of different ticks have been identified through field and laboratory surveys [11, 12]. The digital data acquired from satellite images are derived from the reflected or emitted energy from the ground, water, or vegetation in different wavelengths. Using modern geoinformation technologies integrated with landscape epidemiology can lead to the development and implementation of new disease-surveillance measures. The landscape epidemiology theory offers the opportunity to use the landscape as a key factor to the spatial and temporal distribution of disease risk identification.

Key environmental factors, such as attitude, temperature, rainfall and humidity affect on influence the presence, development, activity and longevity of pathogens, vectors and zoonotic reservoirs of infection, and their interactions with humans [5-8]. In order to be infected, hosts must enter a habitat in which infected ticks are present. Such areas can be accurately identified topographically [2, 3]. Our study used a GIS and statistics analysis to evaluate the spatial distribution of tick-borne hemoparasitic disease. There are several investigations on geographical information system enables to understand changes in space and time of tick and tick-borne diseases and its current and future disease control programs [4-15].
The main aim of this survey was to prepare hemoparasitic diseases risk maps in Qazvin province based on geographical information system of ixodid ticks distribution and their hosts and to isolate the hemoparasites from both ticks and cattle whole blood to make ecological comparisons between areas of infection and areas with no evidence of infection, in relation to climatological, topographical and geographical factors.

Materials and Methods

Qazvin province that is located in the north-west of the central plateau of Iran has five counties including Qazvin, Abyek, Boeen-Zahra, Alborz and Takistan (Fig. 1). This province is divided into two mountainous and plain areas with three main climatological zones including moderate, humid, and cold [13]. The climate of the province, especially in north parts is under the effect of altitude and proximity to the south wind of the Caspian sea. The northern parts climatic conditions are cold and snowy in winters and temperate in summers. In the southern parts, the climate is mild with relatively cold winters and warm summers [13]. In this province, the majority of the livestock are of a domestic race and low percentages are mixed breeds, including Holstein [14]. Meteorological data from April to August 2010, including the monthly means, daily minimum and maximum temperatures, relative humidity, wind speed, and rainfall in different agroclimatic and topographic zones were obtained from Iran Meteorological Organization. Temperature and relative humidity of the studied areas were recorded at the time of sampling using thermometers and hygrometers.

The total numbers of 10,316 cattle in sixty-six farms in 35 villages distributed over the whole different geographic and topographical regions of Qazvin province were inspected for ticks and hemoparasitic tick-borne diseases. For each farm, data were recorded. In April to September 2010, each sampled animal was carefully checked for ticks’ specimens using whole body collection and were kept alive till transporting them to the laboratory. The study area covers all counties of the Qazvin province. Satellite-derived global area coverage normalized difference vegetation index (NDVI) data were obtained for selected adjacent areas from Iranian Space Agency. A LANDSAT image of an area 100×100 km was obtained and georectified. Host distribution and their numbers were obtained, too. Most data have been digitized and analyzed using geographic information system software, ARC VIEW/INFO software GIS version 3.1 (Environmental Systems Research Institute, Inc., Redlands, CA) on personal computers. A handheld global positioning unit (GPS) (Garmin eTrex Legend®, Garmin International Inc., Olathe, KS, USA) was used to locate the selected sampling sites in the field. Geocoded study sites to individual points in a new map layer were done. Data were exported from ArcView.

In April and September, 2010, blood samples were collected on each of the sixty six selected farms from a total of 10,316 goats, cows and sheeps, which were grazing in the areas. Samples were collected from randomly selected animals (N=165). Details of each sampled animal were recorded in individual files. The blood was stored at -20°C. DNA samples were extracted from wild hard ticks and blood samples collected from Qazvin provinces. 18S rRNA-based PCR targeted *Theileria/Babesia* species were done to detect the probability of tick-borne hemoparasitic pathogen. Then a direct polymerase chain reaction was performed with the specific primers. The confirmed cases of tick-borne hemoparasitic diseases were analyzed and confirmed by direct sequencing of polymerase chain reaction productions.

The results of the present study were analyzed using Chi-square test. A p-Value less than 0.05 were considered significant.

Results

The distributions of the ixodid ticks in the study area were *Boophilus annulatus*, *Haemaphysalis concinna*, *Haemaphysalis sulcata*, *Hyalomma anatolicum*, *Hyalomma asiaticum*, *Hyalomma detriment*, *Hyalomma dromedarii*, *Hyalomma marginatum*, *Hyalomma schulzei*, *Rhipicephalus bursa* and *Rhipicephalus sanguineus*. Satellite imaginary investigation showed that there were river, pool and basin in all villages with tick infestation. Tick infestation and *Theileriosis* were seen more in mountainous areas in comparison to hilly areas (Fig. 2). It was attempted to select the villages in both mountainous and hilly areas. Result of the study in different topographical regions based on relative humidity showed that when the RH (%) reached to lowest point in august the highest frequencies of hard ticks were seen (Fig. 3). Villages with high tick infestation were not located on the same isotherm or isoveporate regions. Seventy one, and 315 specimens were collected from villages with 945-1485 and 1911-1485 meters above sea level, respectively (df=1, *p*<0.005). Soil taxonomy of the study areas were consisted of ten different soil covers. 139, 121, 73, 41, and 12 specimens were collected from the areas with rock outcrops/entisols, inceptisols, rock outcrops/entisolsinseptisols, rock outcrops/ inceptisols, and entisolsinseptisols, respectively (df=4, *p*<0.005). Satellites imaginary results showed that Qazvin province has seven different plant covers. One hundred seventy nine specimens were collected from Dryland agriculture with Caspian land cover, 105 specimens from irrigated agricultural lands and 102 specimens from pastures (df=2, *p*<0.001). None specimens, 7 and 17 specimens collected from areas with annual rainfall of 350 mm, 250 mm, 300 mm, respectively. Statistical results showed that there were correlation between ticks frequency and iso-rain areas (df=2, *p*<0.001).

A total number of sixteen cases of *Theileriosis* were identified through the PCR assay in 2010. DNA was extracted from wild hard ticks (Table 1) by kit and proteinase K and phenol/ chloroform/ isooamyl alcohol. Quality of extracted DNA was evaluated by agarose gel electrophoresis and quantified using UV spectrophotometry (A260) and measuring the ratio of
A260/A280. Detection of hemoparasitic pathogens in hard tick vectors by PCR. The specific primer pair P1: 5’ cacagggaggtagtgacaag 3’ and P2: 5’ aagaatttcacctatgacag 3’ was used in the PCR that were designed from hyper variable region V of 18S rRNA gene of Babesia spp. and Theileria spp. [15]. The amplicons were recognized by size on agarose gel electrophoresis. The 426-430 bp fragment size of PCR product was generated in 165 blood samples and 287 wild ticks' specimens tested. 4 (1.4%) hard ticks were positive by PCR assay. Theileria spp. 18S rRNA gene sequenced and registered in GenBank Accession No. JN412658-JN412673.

Table 1. Ticks collected from Qazvin province, Iran that were used for tick-borne hemoparasitic using PCR assay

<table>
<thead>
<tr>
<th>Genus</th>
<th>Counties</th>
<th>Total specimens examined</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Abeyek (N)</td>
<td>Boeen Zahra (N)</td>
</tr>
<tr>
<td>Boophilus</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Haemaphysalis</td>
<td>0</td>
<td>16</td>
</tr>
<tr>
<td>Hyalomma</td>
<td>7</td>
<td>59</td>
</tr>
<tr>
<td>Rhipicephalus</td>
<td>16</td>
<td>11</td>
</tr>
<tr>
<td>Total</td>
<td>287</td>
<td></td>
</tr>
</tbody>
</table>

Figure 1. Map of Qazvin province, different geographic distribution of studied areas (purple circles) in different climatic zones

Figure 2. The average wind speed, dew point, mean weather pressure of the studied areas in different agroclimatic and topographic zones of Qazvin province, during the survey from April to August 2010
Discussion

This study employed some environmental factors and a regional climate to investigate the present distribution of ixodid ticks in Qazvin province. Results showed that climate change affected the preferred habitat of the ticks. Increase in maximum temperatures could increase the frequency and distribution of ticks in the studied areas. Increase in temperature and decrease in percentage of relative humidity could accelerate development and shortening generation times, and as a result led to higher tick populations in the study areas. The hotter the climate, the more prevalence of tick borne hemoparasitic diseases was seen.

Using a GIS and spatial statistics, we investigated the spatial distribution of confirmed cases of tick-borne hemoparasitic diseases in both vector and livestock and identified areas of increased risk within an area highly endemic for tick-borne diseases. Such diseases have become recognized as serious health threats in different part of Iran. The spatial statistics analyses clearly yielded a nonrandom distribution of Theileria spp. within the Qazvin counties. Spatial filtering (smoothing) identified areas of increased risk centered in areas that river was existed. The increase in tick abundances were seen when the relative humidity and air pressure decrease, the increase in tick abundances were seen. Theileria infection mostly was seen in mountainous areas in comparison to hilly areas [16-23].

The decrease in risk for Theileriosis was seen in hilly areas and decreasing latitude. Gathering and including vector population data (including population density, distribution, and infection prevalence rates) and environmental variables in the risk analysis of tick-borne diseases. The relationship between TBDs and hard tick vectors, and landscape characteristics has been studied from remotely sensed and field-gathered data [24-26]. Increased TBDs risk has been well correlated with increased tick abundance and prevalence of infected ticks [24, 26, 27].

The spatial distribution of TBD rates is related with wide range of tick populations and pathogen prevalence. Local weather variations and the periodicity of weather patterns also play a role in tick-borne disease risk. The gathers of these factors leads to high variability of risk even in a tick-borne diseases hyperendemic area. The results of this survey, GIS and spatial statistics, provide an opportunity to clarify and quantify the health burden from tick-borne disease in an endemic area and a foundation to pursue further investigation into the environmental factors leading to increased disease burden. Using specific and geographically suitable risk-reduction measures, the use of analysis like spatial measures should become integral components in the epidemiologic clarifying and risk evaluation of tick-borne diseases [24, 26, 27].

Domesticated animals around act as hosts for the ticks. Climate changes will thus not only affect the preferred habitat of the ticks, but also the distribution of their hosts and these could prove to be major constraints to the predicted future distributions of these ticks and epidemiology of TBDs. Changes that occur in ticks’ distributions will depend strongly on changes in the distribution of their specific hosts. The above analysis and the fact that different tick species prefer different ecological and climatic requirements imply that the effect of climate change on the distribution of one tick species will not necessarily reflect the manner in which other
species would react, emphasizing the importance of assessing its effect on as many species as possible. It is evidenced that climate changes will lead to changes in the overall pattern of tick species richness [1, 26, 27]. These may include the effect of climate change on the hosts as discussed above, acaricide usage, land degradation and human population increase. Not all tick species reacted to the predicted climate changes in a similar way. However, specific information concerning how physiological tolerances and behavioral patterns influence species range.

The expected range expansions ticks could be in response to the predicted general decrease in maximum temperatures. Since temperature determines rates of invertebrate development, reproduction and mortality, a decrease in temperature could accelerate rates of population increase, especially in areas where these are currently limited by high temperatures [24, 26, 27] through a reduction in the desiccation and death of eggs. Its range expansion may, however, be in response to the predicted increase in minimum temperatures accelerating development and shortening generation times, possibly leading to higher tick populations in some areas [1, 26, 27]. Furthermore, the predicted decrease in maximum temperatures and increase in rainfall may result in shorter generation times and therefore more tick generations per year and higher tick populations.

Pastures are predicted to decrease in protein levels because of high temperatures and carbon dioxide concentrations, causing both high tick infestations and greater losses for each tick attachment in stressful situations [1, 24, 26, 27].

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Authors’ Contributions

All authors had equal role in design, work, statistical analysis and manuscript writing.

Conflict of Interest

The authors declare no conflict of interest.

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