**Biodiversity Species and Ecological Distribution of Scorpions in the City of Darmian, Southern Khorasan, Iran**

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**Abstract**

**Background and purpose:** Scorpionism is a public health problem in Darmian City. This study was conducted to determine the biodiversity and faunistic of scorpions in order to discover the health preventive features of scorpion sting among the residents of these regions.

**Methods:** A nightly and daily actively searching on distribution and biodiversity of scorpions was carried during April 2015 to March 2016 in Darmian, Iran.

**Results:** A total of 685 scorpions including five species belonging to Buthidae family were identified. The highest abundance was observed in warmer months, and *Mesobuthus eupeus* (54.5%) was the most abundant species. The researchers found a positive correlation between abundance and temperature and a negative correlation between abundance and humidity. The scorpions were observed with more nocturnal activity and were well-adapted with more abundance to the plain, rural, and outdoors environments. Regarding constancy index, all species except *A. crassicauda* were classified as constant species (C=100). There was no significant difference in the richness of scorpions in different time collections (day or night), geographical areas (plain or mountainous), regions collected (urban or rural), and places (indoors or outdoors). The Shannon and dominant indices were found to be not significantly different between mentioned sites except in time collection.

**Conclusions:** Species composition of scorpions revealed that scorpions were constantly active throughout the year which can play an important role in sting events. So, familiarity of healthcare experts and indigenous people with existing species and their seasonal activities can largely reduce the risk of scorpion stings.

**Keywords:** Biodiversity Species; Fauna; Scorpion; Rarefaction; Darmian

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1. Introduction

Scorpions as one of the most important taxa occupy various areas (1). These Taxa with nearly 2000 known species have been scattered in many terrestrial ecosystems (2, 3). Among these numerous species of scorpions only few species have been concerned in envenoming of humans. But scorpion stings and deaths from it with the approximate rate of 1,230,000 and 3250 cases per year, respectively (4-6) is recognized as considerable health problem in the tropical and subtropical regions including Iran (7, 8).

In the study of biodiversity of scorpions, temporal and spatial scales play an effective role (1, 2, 9). On the other hand, ecological conditions could have an effective relationship with scorpionism and health problems (6, 7). Therefore, assessment of the patterns of distribution and biodiversity of scorpions could be very important in health problem (7, 10).

Iran as one of the largest countries in the Middle East has a variety of climates that provide suitable conditions for a huge diversity of organisms, such as scorpions (11). Scorpions are abundantly distributed in many parts of Iran, and at least four families including 18 genera and 51 species are identified (5, 6, 11). Annually, 40,000-50,000 people are stung by these species in Iran (6). Among these, some genera, such as Androctonus and Hémiscorpius are known as medically important species in Iran (11). Therefore, information about scorpions’ fauna in Iran can be very useful in running control programs of scorpions and scorpionism (6). Southern Khorasan Province (32.8653° N 59.2164° E) is located in the east of Iran. Darmian city (32°50′13″ N, 59°54′11″ E) covering 5,797 km² of Southern Khorasan is located in the East of the province. Based on Southern Khorasan Environmental Protection Agency (2016), Darmian is located in lowland and mountainous area. Vegetation of the area mainly consists of steppe-desert and grassland. Weather conditions are usually warm and dry, or cold and dry, which could provide favorable conditions for the activity of scorpions. But unfortunately no studies concerning scorpion biodiversity have so far been carried out in Darmian city.

The present study is the first complete project on the biodiversity and community level attributes of scorpions in Darmian. The study was conducted as actively searching using UV light at night and daily collection from the shelters (permanent + temporary) as sampling methods. The main objective of this study was to investigate the community of scorpions from two different habitats (plain and mountainous regions) within urban and rural areas. Also, indoor and outdoor places were considered for further study. In addition to determining the biodiversity parameters (richness, Shannon, and Dominant indices), abundance, and seasonal variations of scorpions were calculated as the other purpose of our study during 12 months. Therefore, the current study was conducted in order to take preventive health measures of scorpion sting among the residents of these regions.

2. Materials and Methods

2.1. Study area

Darmian city is characterized by two different topographical scales (plain and mountainous). Based on climate patterns, vegetation and geographical locations, urban and rural areas, plain and mountainous areas, 10 sites (Table 1) were systematically selected by randomized
cluster sampling method. For each site, two transect (residential and non-residential premises) were designed. Totally, twenty transects (60m×60m) were selected for sampling efforts, and scorpions were separately captured within residential and non-residential premises during 12 months (April 2015 – March 2016).

2.2. Study design
The required sample size (n) was calculated by 25% of prevalence rate of scorpion stings in the province (p=0.25), 95% confidence interval at 5% (α=0.05), desired absolute precision (d=0.04), and Z-score (Z=1.96), through the following formula:

\[ n = \frac{Z^2 \cdot p(1-p) \cdot d^2}{}\]

where, n=required sample size; p=expected prevalence; d=desired absolute precision. Totally, 450 scorpions (n=450) were determined for minimum sampling required.

2.3. Scorpion sampling
Samplings were randomly conducted by walking along transects. Scorpions were collected by actively searching each site during the day and at night (with an ultraviolet flashlight) (1, 12). Sampling effort per transect was limited to 120 min during the day and 120 min at night. All sites were sampled monthly during April 2015 to March 2016. All samples were preserved in 75% ethanol and were transferred to the Entomology Laboratory of Mazandaran University of Medical Sciences, Sari, Iran. The scorpions were identified using appropriate keys (13, 14), and stored in the collection of Medical Entomology Museum of Mazandaran University of Medical Sciences, Sari, Iran.

2.4. Data Analysis
- Categories of Dominance
To evaluate the dominance structure of scorpions, Heydemann’s classification was used (15). This classification has five ranks of dominance: eudominant (more than 30%), dominant (10–30%), subdominant (5–10%), rare (1–5%), and sub-rare species (less than 1%). Constancy of species (2) was also calculated through the following formula:

\[ C = \frac{P \times 100}{N} \]

where, P = number of samples in which the species is present; N = total number of samples. Species were then classified in three levels of constancy as: constant (when present in > 50% of samples), accessory (present from 25% to 50%) or rare (< 25% of samples).

Species richness, Dominance, and Diversity were also calculated through Margalef Index \( D_M = \frac{s-1}{lnN} \), Simpson's dominance \( D = \lambda = \sum_{i=1}^{S} P_i^2 \) and Shannon indices \( H' = \sum_{i=1}^{S} P_i(lnP_i) \), respectively. To compute whether species were dispersed uniformly crosswise over microhabitats, Evenness Index was utilized (17-19); where N equaled the total number of individuals in the sample, S = the number of species in the sample, \( P_i = \frac{n_i}{N} \), \( n_i = \) number of individuals of taxon ith. Diversity t test was then performed by comparing the Shannon and Simpson diversities in two samples (20). To verify the sampling sufficiency to assess true richness and to determine the observed and estimated richness of scorpions, rarefaction curves were used using the following formula:

\[ E(Sn) = \sum_{i=1}^{S} \left[ 1 - \frac{(N-N_i)}{(n_i)} \right] \]

where N= total number of individuals in the sample, S= total number of species, and N_i= number of individuals of species number i.(21-24).
Statistical analyses were performed using PAST Software, Version 3.02 (25).

### 3. Results

During the study period, the researchers collected 685 specimens representing five species belonging to the Buthidae family. The most abundant species was *Mesobuthus eupeus* (54.5%). It was followed by *M. caucasicus* (20%), *Compsobuthus matthiesseni* (15.3%) and *Orthochirus scrobiculosus* (7%).

The least abundant species was *Androctonus crassicauda* (3.2%). Dominance structure of scorpions showed *M. eupeus*, and *A. crassicauda* were eudominant (54.5%), and rare (3.2%) species, respectively (Table 2). At the species level, *M. eupeus* was the most common species in all months, except January and March (Figure 1).

### Table 1. Location of the sampling sites, with emphasis on climate and geographical location

<table>
<thead>
<tr>
<th>Region</th>
<th>Area</th>
<th>Climate</th>
<th>Mean annual precipitations (mm)</th>
<th>The average annual temperature (°C)</th>
<th>Average annual humidity</th>
<th>Vegetation</th>
<th>Geographical location</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tabas</td>
<td>Plain</td>
<td>Warm and dry</td>
<td>0</td>
<td>16.8</td>
<td>33.2</td>
<td>Steppe desert</td>
<td>54 11 32 47 1474</td>
</tr>
<tr>
<td>Gazik</td>
<td>Plain</td>
<td>Warm and dry</td>
<td>11</td>
<td>16.2</td>
<td>34.3</td>
<td>Steppe desert</td>
<td>60 14 33 58 13 60</td>
</tr>
<tr>
<td>Asadieh</td>
<td>Plain</td>
<td>Warm and dry</td>
<td>143.8</td>
<td>16.7</td>
<td>34</td>
<td>Steppe desert</td>
<td>60 01 32 55 15 00</td>
</tr>
<tr>
<td>Darmian</td>
<td>Mountain</td>
<td>Cold</td>
<td>0</td>
<td>12.6</td>
<td>33</td>
<td>Meadow</td>
<td>59 58 32 55 20 00</td>
</tr>
<tr>
<td>Noghab</td>
<td>Plain</td>
<td>Warm and dry</td>
<td>13</td>
<td>15.9</td>
<td>35.3</td>
<td>Steppe desert</td>
<td>59 56 33 59 14 76</td>
</tr>
<tr>
<td>Boorang</td>
<td>Plain</td>
<td>Warm and dry</td>
<td>0</td>
<td>16.2</td>
<td>34.4</td>
<td>Steppe desert</td>
<td>59 44 33 01 14 74</td>
</tr>
<tr>
<td>Gasak</td>
<td>Plain</td>
<td>Warm and dry</td>
<td>0</td>
<td>16.6</td>
<td>33.7</td>
<td>Steppe desert</td>
<td>59 40 33 02 14 75</td>
</tr>
<tr>
<td>Ghahestan</td>
<td>Mountain</td>
<td>Cold</td>
<td>9.5</td>
<td>14.7</td>
<td>37.5</td>
<td>Steppe desert</td>
<td>59 42 33 10 20 10</td>
</tr>
<tr>
<td>Khan</td>
<td>Mountain</td>
<td>Cold</td>
<td>0</td>
<td>14.6</td>
<td>37.6</td>
<td>Steppe desert</td>
<td>59 48 33 11 20 15</td>
</tr>
<tr>
<td>Takhteh</td>
<td>Mountain</td>
<td>Cold</td>
<td>0</td>
<td>15.3</td>
<td>36.3</td>
<td>Steppe desert</td>
<td>59 45 33 15 14 75</td>
</tr>
</tbody>
</table>

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Comparison of biodiversity components of scorpions

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Figure 1. Seasonal dynamics of the different species of scorpions during the study period (April 2015-March 2016) at different sites of Darmian, Iran

The results of the study showed that night (51.5%), plain (66.1%), rural (72.8%), and outdoor (59.1%) collections caught more scorpions than day (48.5%), mountainous (33.9%), urban (27.2%) and indoors (40.9%) collections, respectively (Table 2).

Table 2. Diversity parameters ($H'$, $D$, $D_{Mg}$, $E$) and abundance (N) of scorpions in different types of scenarios in the Darmian city

<table>
<thead>
<tr>
<th>Species</th>
<th>Time Collection</th>
<th>Geographical area</th>
<th>Region collected</th>
<th>Places</th>
<th>Total</th>
<th>Dominance of structure</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Night (N, %)</td>
<td>Day</td>
<td>Plain</td>
<td>Mountainous</td>
<td>Urban</td>
<td>Rural</td>
</tr>
<tr>
<td>M. euepeus</td>
<td>181 (26.4%)</td>
<td>192 (28%)</td>
<td>257 (16.9%)</td>
<td>105 (15.3%)</td>
<td>268 (22.5%)</td>
<td>154 (32%)</td>
</tr>
<tr>
<td>M. caucasicus</td>
<td>55 (8%)</td>
<td>50 (14%)</td>
<td>47 (6.6%)</td>
<td>32 (4.5%)</td>
<td>73 (9.3%)</td>
<td>43 (6.3%)</td>
</tr>
<tr>
<td>C. matthiesseni</td>
<td>27 (7.3%)</td>
<td>21 (8%)</td>
<td>26 (3.8%)</td>
<td>8 (4.7%)</td>
<td>40 (6.3%)</td>
<td>15 (9.1%)</td>
</tr>
<tr>
<td>O. scrobiculosus</td>
<td>8 (14%)</td>
<td>14 (2%)</td>
<td>10 (3.2%)</td>
<td>10 (1.2%)</td>
<td>12 (2.2%)</td>
<td>4 (8%)</td>
</tr>
<tr>
<td>A. crassicauda</td>
<td>8 (12%)</td>
<td>20 (2.9%)</td>
<td>2 (3.2%)</td>
<td>4 (1.5%)</td>
<td>12 (1.8%)</td>
<td>4 (0.6%)</td>
</tr>
<tr>
<td>Total</td>
<td>353 (51.5%)</td>
<td>332 (48.5%)</td>
<td>453 (33.9%)</td>
<td>232 (17.2%)</td>
<td>186 (27.2%)</td>
<td>499 (27.3%)</td>
</tr>
</tbody>
</table>

Biodiversity indices

<table>
<thead>
<tr>
<th></th>
<th>$H'$</th>
<th>$D$</th>
<th>$D_{Mg}$</th>
<th>$E$</th>
</tr>
</thead>
<tbody>
<tr>
<td>$t$ test</td>
<td>3.9</td>
<td>521.39</td>
<td>&lt; 0.0001</td>
<td>0.64</td>
</tr>
<tr>
<td>$df$</td>
<td>2.6</td>
<td>454.63</td>
<td>0.01</td>
<td>0.69</td>
</tr>
<tr>
<td>$p$</td>
<td>-0.28</td>
<td>313.31</td>
<td>0.78</td>
<td>0.73</td>
</tr>
<tr>
<td>$D_{Mg}$</td>
<td>0.27</td>
<td>0.39</td>
<td>0.32</td>
<td>0.74</td>
</tr>
<tr>
<td>$E$</td>
<td>0.09</td>
<td>0.38</td>
<td>0.36</td>
<td>0.77</td>
</tr>
<tr>
<td>$D_{Mg}$</td>
<td>0.64</td>
<td>0.08</td>
<td>0.59</td>
<td>0.71</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th></th>
<th>$t$ test</th>
<th>$df$</th>
<th>$p$</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>-4.4</td>
<td>428.63</td>
<td>&lt; 0.0001</td>
</tr>
</tbody>
</table>
A. crassicauda was the only species found to be rare, with a constancy index of $C = 5$, while other species were classified as constant species ($C = 100$). Scorpions were more collected during the warmer months (Figure 2). Monthly Scorpion abundances were significantly and negatively correlated with monthly mean humidity (Pearson correlation coefficient, $p < 0.01; r_s = -0.85$). Positive significant correlation was also observed between scorpion abundances and temperatures ($p < 0.01; r_s = 0.88$). M. eupeus showed a peak of activity during May and September, while a small number of individuals were recorded throughout winter (Figure 3).

Species richness did not differ in different parts of the study, while the Shannon Index was significantly ($P < 0.001$) higher at night ($H' = 1.41$) than in day ($H' = 1.2$) (Table 2). Evenness Index of scorpions was also higher at night ($E = 0.82$) than in day ($E = 0.66$), while Dominance Index showed a lower value at night ($D = 0.27$) than in day.
There was no significant difference between biodiversity indices in plain and mountainous areas, urban and rural regions, as well as indoors and outdoors places ($P>0.01$) (Figure 4).

Right-hand of rarefaction curves can be used to compare richness at different spatial and temporal scales. Monthly rarefaction curves in different seasons indicated that with the same sample size or specimens, the highest richness belonged to April and January. Hence, with the lowest sampling effort, the maximum species was obtained. In summer, reaching the greatest number of species is more likely to happen in July and August compared to September, while equal richness was observed in all months of the autumn (Figure 5).
4. Discussion

Iran has the highest rate of scorpion stings as compared to other countries in the Middle East, and in the meantime, Buthidae family have been incriminated as the main venomous scorpions in Iran (26). As the only observed family in Darmian, Buthidae has many deadly species with high compatibility to dry environment (27-29).

M. eupeus as the most dominant species in our study has widespread distribution in Middle East and Central Asia, including many parts of Iran. This species is one of the most important genera in terms of envenomation and has close association with human life (8, 30, 31). In the current study, M. eupeus along with other members of this family were observed with the...
highest activity and density during the warmer months (May 2015 – September 2015). As a result, by reducing moisture during the warmer month, we encountered more abundance of scorpions (11). On the other hand, the effects of high temperature on increasing plant growth and subsequently herbivorous insects create favorable conditions for the scorpion activity, and consequently improve scorpion communities (2, 32, 33). These topics amplify the consideration of medical importance of envenoming to native people during spring and summer in Darmian.

There is no reported death due to *M. eupeus* in Iran (11, 34), but some reports of several deaths from sting of this species has been observed in Algeria and Tunisia (8). Sting by *M. eupeus* makes direct extreme agony at the sting location and causes a variety of symptoms as thirst, dry mouth, headache, nausea, irritability, and restlessness by affecting sympathetic and parasympathetic nervous systems (11, 30, 34). So, prevention is necessary and medical care must be observed in the face of this species, especially in warmer month.

*M. caucasicus* as dominant species in our study commonly dispersed in the central and east of Iran, and it is reported that its sting is not so dangerous (11), while *C. matthiesseni* as the other dominant species in our study has a relatively large distribution in Iran (11), and sting by this scorpion leads to hematuria. Hence, people strongly prefer to avoid any contact with *C. matthiesseni* (11, 35). *O. scrobiculosus*, that is scattered in the south, southwest, and central of Iran (11), was observed as subdominant species in our study. Although its sting causes a relative pain without the requirement for medicinal maintenance, but one death caused by its sting has so far been reported in Iran (14). Therefore, a subject that should be considered is that, there is always the probability of stung for the people of this region by scorpions especially in warmer seasons.

A. *crassicauda*, also known as black scorpion, is the only species that has been classified as rare species. Although this species has been observed with low frequency in our study areas, this species is widely distributed in Iran, and is known as one of the most dangerous species in Iran with high rate of envenoming and with often deadly consequences (11, 36, 37). So, people in these areas should strongly consider the risks of exposure to this dangerous species.

The most abundant species are observed in outdoors, plains, rural areas, and slightly more at night. This suggests that these members of Buthidae family are more common in outdoors, plains, rural areas, and with a little more degree of nocturnal activity. Although it has been reported that some members of this family are common in indoors at night (38), some other studies noted that most scorpion stings occurred in outdoors at night (39). One of the main reasons which can be cited is that outdoors provide wide variety of favorable habitats for scorpions, such as rocks, boards, and vegetation. Also, control strategy is necessary to modify the entering areas of scorpion, and chemical control of scorpions in indoors (40) could be another reason for the less abundance of scorpions. It is known that scorpions are common in urban and rural areas of Iran (41), but in our study, rural areas showed the most abundant species than urban areas. It seemed that more vegetation in rural areas could be related to an increase in the abundance of insects, because it can provide more prays and lead to food availability for scorpions (2, 16).
In the current study, since the rarefaction curves for different months usually showed flattened asymptotic lines, so the estimations for abundances per month were not very critical, and as a result, increasing estimated densities of individuals per month usually did not increase the estimated number of species (richness) (42). Thus, enough sampling effort was made in all months to achieve the maximum species of scorpions. It is only possible that in February, a little more sampling effort was necessary to reach an asymptotic estimate of the species richness (9, 42).

Community diversity index (Shannon Index) at night showed the highest diversity level than day. Some species, then, can be seen, such as M. caucasicus, C. matthiesseni and O. scrobiculosus which can escape from daytime temperatures by moving below ground or into cracks. Species diversity was similar in other scenarios, (geographical areas, region collected, and places). It can be justified that long life span of scorpions along with factors such as low vagility, habitat specificity, extreme climate adaptability, food size specificity, cannibalism, adaptive radiation and predation by nocturnal predators may act as limiting features for species diversity (2, 43, 44). Thus, we probably cannot expect to observe a wide range of changes or differences in scorpion diversity.

5. Conclusion

Species composition of scorpions revealed that Darmian County with five species provides varying degrees of toxic species which are constantly active throughout the year. Although the present study showed that only a small number of scorpions like A. crassicauda are potentially life-threatening for human it should not be neglected that must of them have an important role in sting events (11). Therefore, the presence of the eudominant species, such as M. eupeus along with dominant species like M. caucasicus and C. matthiesseni may increase the risk of scorpion biting to the residents of the area under this study, mainly in spring and summer. Being aware of this subject can lead healthcare experts and indigenous people to become familiar with and know how to confront bites caused by native scorpions and consequently reduce the risk of scorpion stings.

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Conflicts of interest

The authors declare that they have no potential conflict of interest.

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