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Research Paper

Phylogenetic Relationships of the Scorpion *Apistobuthus susanae* From Khuzestan Province Based on Mitochondrial DNA Cytochrome Oxidase I (*COXI*) Gene Sequences

Safie Bahri[1], Bahman Shams Esfandabad[1*], Abbas Ahmadi[1], Hedieh Jafari[2]

1. Department of Environment, Ar.C., Islamic Azad University, Arak, Iran.
2. Department of Venomous of Animal, Razi Vaccines and Serum Research Institute, Ahvaz, Agricultural Research Education and Extension Organization (AREEO), Ahvaz, Iran.

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ABSTRACT

Introduction: This study aims to analyze the morphometric and phylogeny of *Apistobuthus susanae* scorpions using mitochondrial DNA sequences. Information regarding the morphology and biology of scorpions is very limited, and the foundation of this information is the identification of habitats, morphology, and morphometrics of scorpions in different regions.

Materials & Methods: In this study, the morphometric analysis based on 32 morphological characteristics of *A. susanae* and DNA sequencing of *COXI* was performed. Phylogenetic tree was constructed using MEGA 10 software. Both male and female specimens of A. susanae were collected from four cities: Hamidiyeh, Masjed Soleyman, Ramhormoz, and Andimeshk in Khuzestan Province.

Results: According to the results, in all cases, the average sizes of female specimens were larger than those of male specimens. Except for the traits Cl, CHL, ML, TIW, MTIL, MTIILL, MTIVL, MTIIW, MTIVW, and MTIH, all other features studied were identical in both male and female specimens (P>0.05). The results of the analysis of morphometric values of *A. susanae* were compatible with the phylogenetic tree and supported the morphometric classification. Out of 614 nucleotides of the *COXI* gene amplified for 10 *Apistobuthus* samples and one sample of *Androctonus crassicauda* as an outgroup, 558 sites were conserved (90.97%), 41 sites were variable (6.76%), and 15 sites (2.25%) were parsimony-informative. The analysis of the average genetic distance within the species showed that two specimens from Hamidiyeh had the least divergence (0%), and two specimens from Ramhormoz and Andimeshk had the greatest inter-specimen genetic divergence (0.1%).

Conclusion: The greatest intraspecific divergence was observed in the specimens from Andimeshk and Masjed Soleyman, while the least genetic divergence was found in the Ramhormoz samples.

*** Corresponding Author:**
Bahman Shams Esfandabad, Assistant Professor.
Address: *Department of Environment, Ar.C., Islamic Azad University, Arak, Iran.*
Tel: *+98 (912) 3039490*
E-mail: *bahmanshams1357@iau.ac.ir*

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1. Introduction

Understanding the evolutionary origins of scorpions, one of the most important and unique groups of arthropods, has long been shrouded in mystery, with numerous disagreements existing on this topic. The Buthidae family of scorpions is the largest and most widespread scorpion family in the world [1]. However, limited information is available regarding phylogenetic relationships within this family. The population composition of Iranian scorpions is one of the most diverse in the region of West Asia [2]. On the other hand, morphotaxonomy is limited in its ability to accurately define species boundaries and distinguish between species. It is well-established that molecular data allow understanding of diversity, distribution, and intraspecific evolution in scorpions [3]. According to the latest morphometric studies, there are 18 scorpion genera in Iran, belonging to three families: Buthidae, Scorpionidae, and Hemiscorpidae. Of these, 14 genera have been reported in Khuzestan Province [4].

Apistobuthus susanae (Lourenço 1998), a scorpion species from the Buthidae family, is an invasive species commonly found in hot, dry, desert, and sandy regions, and is easily recognizable by its rounded second tail segment. This yellow-colored scorpion has a narrow tail and long legs with setae to facilitate movement over sand. The venom of this species may have effects upon stinging, but the likelihood of death is very low [5].

Molecular data have provided much information regarding the phylogeographic histories of many animal species. The mitochondrial DNA cytochrome oxidase I (*COXI*) gene sequences are one of the most widely used molecular markers for species identification. This enzyme is key in aerobic metabolism. The evolution of this gene is so rapid that it can identify phylogenetic changes not only among closely related species but also within a specific species [6]. Phylogenetic analysis using *COXI* gene sequences has been widely performed by various research groups across different animal taxa, including the *Puntius* genus in the Cyprinidae family [7] and 42 *Culicoides* species (Diptera, Ceratopogonidae) across three continents [8]. This gene is widely accepted as a DNA barcode for precise and easy species identification. In this study, mitochondrial DNA sequences were used for the phylogenetic analysis of *A. susanae* scorpions.

2. Materials and Methods

2.1. Sample collection

Scorpion specimens were collected from four cities: Hamidiyeh, Masjed Soleyman, Ramhormoz, and Andimeshk located in Khuzestan Province, which has a hot and dry climate, using geographic information system (GIS) tools (Table 1, Figure 1). Since scorpions are nocturnal, sampling was done at night using ultraviolet light flashlights and long-handled forceps. The specimens were collected from the tail region and placed in containers containing ethanol. To preserve the specimens' appearance and prevent structural changes, 96% ethanol was injected into the scorpion's abdomens, and the specimens were stored in sealed glass containers containing 70% ethanol and 5% glycerin. A total of 8 male and 8 female specimens were collected. Due to the scorpions' specific habitat in the psammophilic climate, there were limitations in sampling. This study was conducted following the academic ethics of the Department of Environmental Science and Desert Area Management, Faculty of Engineering and Agriculture, Islamic Azad University, Arak Branch.

2.2. Morphology

In the laboratory, each sample was placed separately on graph paper, and morphological characteristics were examined using an Olympus SZ-CTV stereomicroscope, with the help of a caliper accurate to one-tenth of a centimeter. In this study, morphological and morphometric indices were calculated based on the indices defined by Lamoral (1979) [9]. A summary of the morphometric ratios is provided in Table 2. Statistical analysis of the data was performed in SPSS 23 using The Independent T-Test, With A significance level of <0.05.

2.3. Molecular analysis

2.3.1. Tissue collection

Two grams of muscle tissue from the scorpion's tail region were collected using sterile tools and preserved in 90% ethanol. To extract DNA, the tissue was mixed with deionized water and transferred into 1.5 mL tubes. The tubes containing the samples were centrifuged at 2,000 rpm for 10 minutes at 4 °C. DNA was extracted from the pellet.

2.3.2. DNA extraction

The pellet was mixed with 600 µL of resuspension buffer and 60 µL of 10% SDS solution. It was then centrifuged at 1,000 rpm for 5 minutes. The supernatant was

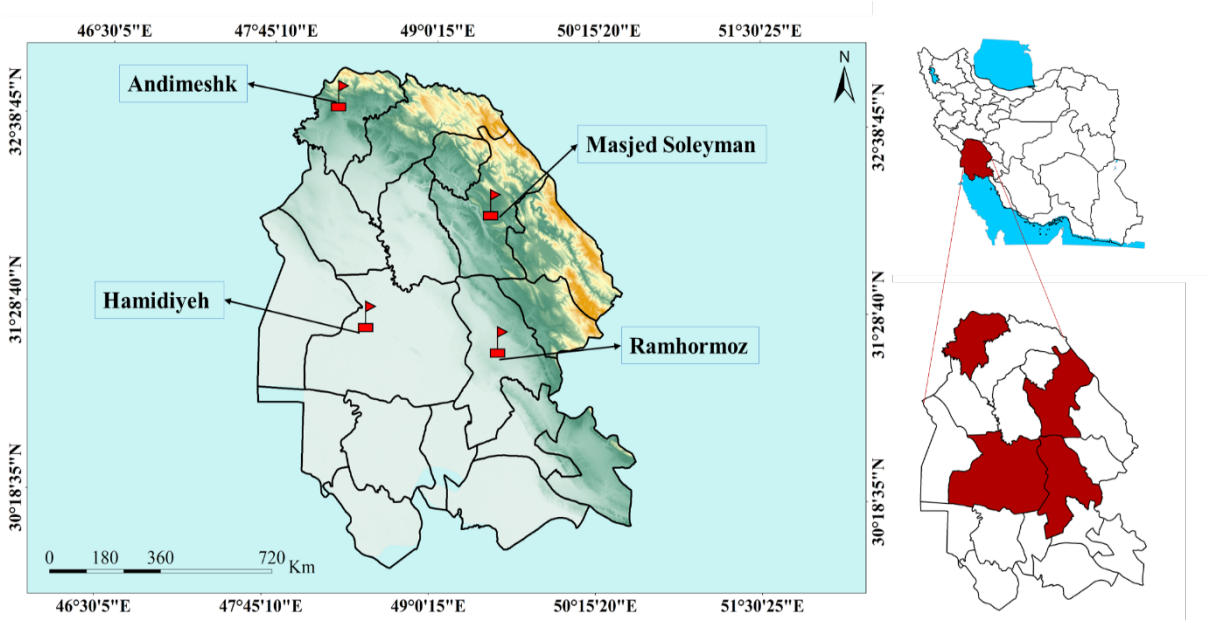

Figure 1. Geographic distribution of sampling points in Khuzestan Province

transferred to a new microtube, 500 µL of chloroform was added, and the tube was centrifuged again at 10,000 rpm for 5 minutes. The supernatant was transferred to a new microtube, and 10 µL of 5 M NaCl and 1,200 µL of cold ethanol were added. After centrifuging at 10,000 rpm for 5 minutes, the supernatant was discarded, and the pellet was dried. Then, 50 µL of sterile deionized water was added to the pellet.

2.3.3. Polymerase chain reaction (PCR)

PCR was performed to amplify the cytochrome *oxidase I* gene fragment using primers designed by Gantenbein et al. (1999) [10]. The reaction was carried out in a final volume of 25 µL containing 1 × PCR buffer, 1 mM dNTPs, 1.5 mM magnesium chloride, forward and reverse primers at 1 µM each, and 1 ng of Taq polymerase. The thermal program included an initial denaturation at 94 ˚C for 1 minute, followed by 30 seconds at 94 ˚C, 30 seconds at 50–60 ˚C, and 50 seconds at 72 ˚C for 30 cycles. The final extension was at 72 ˚C for 5 minutes. The PCR product was visualized by electrophoresis on a 1% agarose gel, and sequencing was performed with the specific primers.

2.3.4. Genetic diversity analysis

The obtained sequences were first reviewed and manually corrected using SeqScape software. Then, the sequences were aligned using the ClustalW algorithm in BioEdit software. The number of haplotypes, polymorphic sites, haplotype diversity, and nucleotide diversity were determined using DNAsp software and BLAST.

2.4. BLAST analysis

The BLAST results from NCBI indicated that the obtained gene sequence from *A. susanae* showed 100% similarity with *Apistobuthus pterygocercus* and 89-90% similarity with other sequences. The sample presented in the BLAST program, consisting of 627 base pairs, showed zero gaps and complete similarity to the *A. pterygocercus* scorpion sequence (pairwise alignment of the input gene with the different gene sequences, their number, and percentage of overlap, which is 100%). Gene alignment in the BLAST program for the most similar sequences to the target sample, showing 100 gene lines, is displayed (solid red lines represent complete alignment of the input gene with the available aligned genes).

2.5. Statistical analysis

The Mean±SE were calculated, and univariate and multivariate statistical analyses were performed in SPSS Inc., Illinois, USA, version 23, at a significance level of less than 0.05.

3. Results

3.1. Morphology result

A. susanae is typically yellowish; the prosoma (carapace), mesosoma, metasoma segments, and venter are

Table 1. Geographic coordinates of sampling sites

City	Longitude	Latitude
Hamidiyeh	31°26′56′′	48°30′17′′
Masjed Soleyman	31°35′45′′	48°57′18′′
Ramhormoz	31°13′54′′	49°15′26′′
Andimeshk	31°21′44′′	48°37′36′′

generally yellowish. The aculeus is blackish, and the pedipalps are reddish. There are also some dark longitudinal spots on the mesosoma. The color of the male specimens was darker than that of the female. The Mean±SD of each measured feature by gender are presented in Table 2. In all cases, the mean measurements of female specimens were higher than those of male specimens. Except for the traits CL, CHL, ML, TIW, MTIL, MTIILL, MTIVL, MTIIW, MTIVW, and MTIH, all other features examined were identical between male and female specimens ($P>0.05$).

Non-matching letters indicate a significant difference between male and female specimens ($P<0.05$).

The difference in the dorsal and ventral views of *A. susanae* scorpion between the male specimen (A) and female specimen (B) is shown in Figure 2.

3.2. Molecular results

To assess the PCR, the product obtained from the reaction was examined on an agarose gel. Samples that indicated successful amplification of the target gene and optimal reaction conditions were selected for sequencing (Figure 3).

Table 2. Studied morphometric ratios

Morphometric Ratio	Description
CL/CAW	The length of the carapace to the width of the anterior part of the carapace
CL/CPW	The length of the carapace to the width of the posterior part of the carapace
CAW/PW	The width of the anterior section to the posterior section of the carapace
X/Y	The distance from the middle eye to the anterior and posterior edges of the carapace
Mt(I)l/W	The ratio of the length of segment I to the width of the segment
Mt(I)l/H	The ratio of the length of segment I to the height of the segmen
Mt(II)L/W	The ratio of the length of segment II to the width of the segment
Mt(II)L/H	The ratio of the length of segment II to the height of the segment
Mt(III)L/W	The ratio of the length of segment III to the width of the segment
Mt(IV)L/W	The ratio of the length of segment IV to the width of the segment
Mt(IV)L/H	The ratio of the length of segment IV to the height of the segment
Mt(V)L/W	The ratio of the length of segment V to the width of the segment
Mt(V)L/H	The ratio of the length of segment V to the height of the segment
CHL/ML	The length of the pincer to the length of the hand
MFL/ML	The length of the movable finger to the length of the hand
TIL/W	The length to the width of the telson

Table 2. Comparison of the mean morphometric traits of male and female *A. susanae* scorpions collected from Khuzestan Province

Morphometric Variable	Female	Male	Sig.
Cl	10.31±1.64[b]	7.18±0.27[a]	P<0.05
cpw	8.9±0.75[a]	7.54±0.7[a]	P>0.05
Caw	5.47±0.52 [a]	4.87±0.16[a]	P>0.05
X	5.2±1.06[a]	3.78±0.12 [a]	P>0.05
Y	4.76± 0.72[a]	3.32±0.19[a]	P>0.05
CHL	17.76±1.20[b]	15.32±0.57[a]	P<0.05
ML	5.38± 0.58 [b]	4.97±0.08[a]	P<0.05
MFL	13.79±0.71[a]	12.68±0.44[a]	P>0.05
THL	8.41±0.41[a]	7.80±0.34[a]	P>0.05
TLH	2.95±0.24[a]	2.45±0.16[a]	P>0.05
TIW	2.53± 0.22 [b]	2.41±0.2[a]	P<0.05
Mt(I)L	6.08±0.43[b]	5.83±0.16[a]	P<0.05
Mt(II)L	7.26±0.51[a]	6.79±0.16[a]	P>0.05
Mt(IIIL)L	6.73±0.4[b]	6.52±0.21[a]	P<0.05
Mt(IV)L	7.33±0.61[b]	7.19±0.24[b]	P<0.05
Mt(V)L	22.32±13.01[a]	8.74±0.20[a]	P>0.05
Mt(I)w	5.42±0.59[a]	4.63±0.28[a]	P<0.05
Mt(II)w	7.19±0.65[b]	6.75±0.36[a]	P<0.05
Mt(IIIL)w	4.66±0.4[a]	4.2±0.26[a]	P>0.05
Mt(IV)w	3.33±0.31[b]	3.13±0.16[a]	P<0.05
Mt(V)w	3.34±0.28[a]	3.06±0.16[a]	P>0.05
Mt(I)H	3.58±0.37[b]	2.77±0.18[a]	P<0.05
Mt(II)H	4.31±0.33[a]	3.41±0.21[a]	P>0.05
Mt(IIIL)H	3.75±0.28[a]	3.02±0.15[a]	P>0.05
Mt(IV)H	2.86±0.2[a]	2.59±0.14[a]	P>0.05
Mt(V)H	2.94±0.27[a]	2.39±0.12[a]	P>0.05

Figure 2. Dorsal and ventral views of the male and female *A. susanae* scorpions collected from Khuzestan Province

3.2.1. Phylogenetic analysis of the apistobothus genus based on *COXI* gene sequences

3.2.1.1. Sequence characteristics and divergence rates

Among the 614 nucleotides of the amplified *COXI* gene from 10 *A. susanae* samples and one sample of *Androctonus crassicauda* as an outgroup, 558 positions were conserved (97.9%), 41 positions were variable (6.76%), and 15 positions (2.52%) had informative content. The average intra-species genetic distance showed that species 1 and 5, isolated from the Hamidiyeh region, had the lowest divergence (0%), suggesting they are genetically closer. The greatest inter-species genetic divergence (0.1%) was observed between species from the Ramhormoz [4] and Andimeshk [8] regions, indicating the largest genetic separation between these two species. Moreover, the average intra-species genetic distance across different regions showed that the greatest intra-species divergence was found in species from Andimeshk, while the samples from Masjed Soleyman and Ramhormoz had the least genetic divergence within the species (Table 2).

The nucleotide sequence numbers, number of nucleotides, haplotype counts, average nucleotide differences, nucleotide diversity, haplotype diversity, and the number of polymorphic sites for the 10 *A. susanae* samples studied are summarized in Table 3.

3.2.2. Maximum likelihood intra-species phylogenetic tree analysis

In this study, using MEGA 10 software and the Maximum Composite Likelihood model, a phylogenetic tree of *A. susanae* samples from Khuzestan Province was constructed (Figure 4).

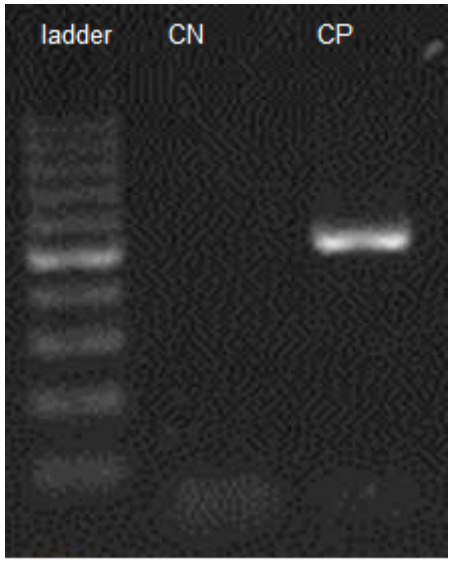

Figure 3. Electrophoresis of the PCR product for amplification of the cytochrome oxidase I gene

Table 2. Average genetic distance between species and intra-species in the mitochondrial *COXI* gene of *A. susanae*

Apisto1									
Apisto_2	0.002								
Apisto_3	0.002	0.002							
Apisto_4	0.003	0.003	0.002						
Apisto_5	0.000	0.002	0.002	0.003					
Apisto_6	0.005	0.005	0.003	0.005	0.005				
Apisto_7	0.002	0.002	0.000	0.002	0.002	0.003			
Apisto_8	0.008	0.008	0.008	0.010	0.008	0.008	0.008		
Apisto_9	0.002	0.002	0.000	0.002	0.002	0.003	0.000	0.008	
Apisto_10	0.002	0.002	0.000	0.002	0.002	0.003	0.000	0.008	0.000

3.2.3. Phylogenetic tree of the Buthidae family based on nucleotide alignment of the *COXI* gene

Based on the analysis of *COXI* gene sequences of *A. susanae* scorpions collected from Khuzestan Province, which were registered in the GeneBank with accession number PV931979, and sequence alignment with sequences from the GeneBank, the constructed phylogenetic tree shows that *Apistobothus* sp. from Khuzestan, together with *A. pterygocercus*, formed a sub-clade (0% genetic distance). These species were grouped with *Androctonus australis*, *Androctonus amoreuxi*, and *Androctonus bicolor* in a clade. Additionally, the genetic distance of each species compared to the others is shown in Figure 5.

4. Discussion

Despite extensive efforts in identifying scorpions in Iran, information regarding their biology and morphological differences is still very limited and perhaps incomplete. In particular, data on sexual dimorphism, the expression of sexual behaviors, and the recognition of intraspecific differences are of special importance (Lourenco, 1999) [11]. Behavioral and ecomorphological adaptation has led scorpions to adjust to specific climatic conditions. In scorpions, except for a few species that exhibit sexual dimorphism, there is no significant difference between males and females due to the absence of distinct sexual organs in other species [12].

Based on the results obtained, the body length in females was significantly larger than in males, which is related to the increase in the size of the abdomen and the enlargement of the reproductive system due to the

Table 3. The number of nucleotide sequences, number of nucleotides, number of haplotypes, average nucleotide differences, nucleotide diversity, haplotype diversity, and the number of polymorphic sites within species in the 10 *A. susanae*

A.susanae	
Number of nucleotide sequences	5
Number of nucleotides	614
Number of haplotypes	4
Average nucleotide difference	2.700
Nucleotide diversity	0.00897
Number of separating sites (polymorphic sites)	7
Haplotype diversity	0.900

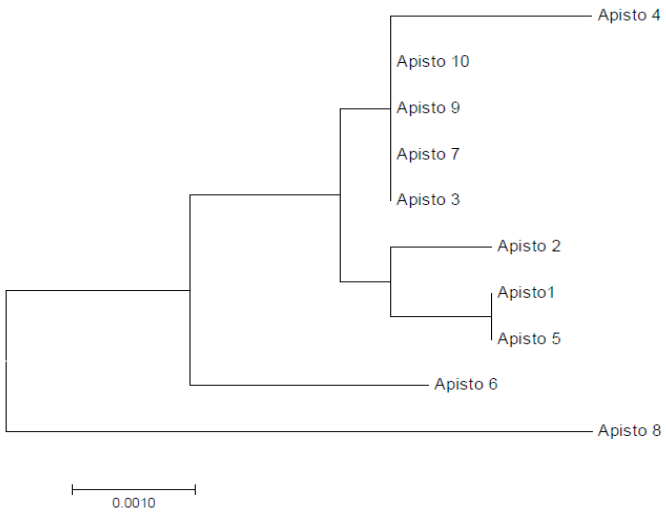

Figure 4. 50% majority-rule consensus maximum likelihood tree based on the *COXI* gene

formation of embryos and the increase in the number of offspring. Levy and Amitai (1980) reported that male bodies are narrower than those of females, which is consistent with the present study. On the other hand, the larger carapace length in females is due to the positioning of the offspring on the mother's back [13, 14]. According to the results, carapace length, chela length, and segment length played the most significant role in distinguishing males and females. In Lane and Crosskey's study (1996), as in the present study, the segments of the tail were used to determine sex, with males being differentiated from females [15]. In Dehghani and Tirgari's study (2012), the size of the fifth abdominal segment and the number and length of the pedipalps were used to differentiate males and females, with the fifth abdominal segment size being larger in females compared to males on the dorsal surface [16].

Levy and Amitai (1980) reported that male bodies are narrower than those of females [13]. Additionally, in studies such as that of Nemat Elahi et al. (2018), other indices, like pectinal length, the number of pectinal teeth, and the distance between the two bases of the pectines, were used to determine sex in the species *Hottentotta*

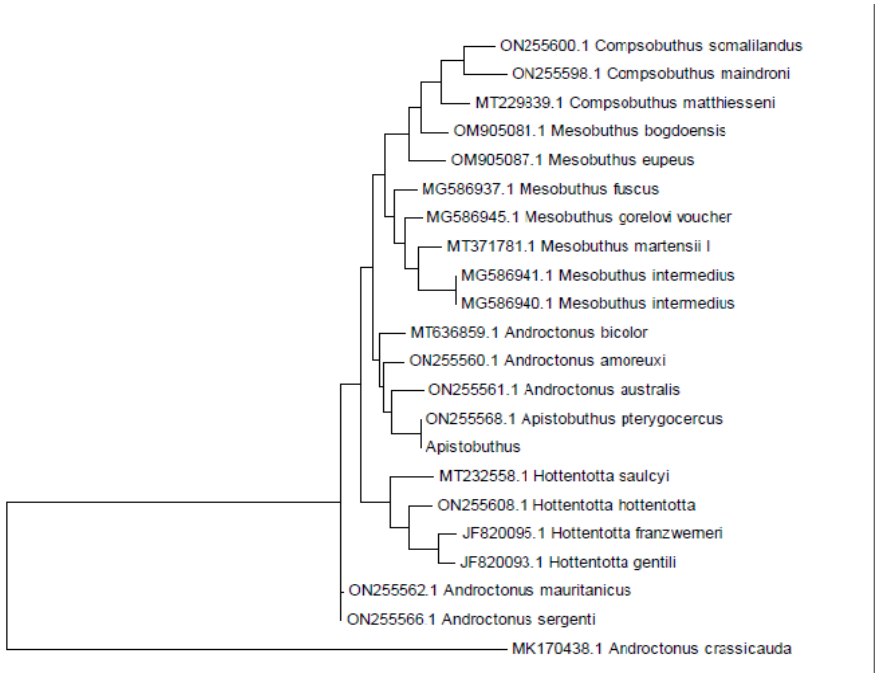

Figure 5. Genetic distance of each species compared to the other species studied

saulcyi, and they stated that the best time for sex determination is during mating [17]. Booncham et al. (2007) reported that in *Heterometrus laoticus*, a wider carapace and a larger seventh mesosomal segment were key factors for distinguishing males from females [14]. These differences can be used to better differentiate sex in future studies of *A. susanae*.

The *COXI* gene is a mitochondrial gene that is maternally inherited, and because it does not undergo recombination, it makes it easier to trace the maternal lineage of animal species [18]. Mitochondrial genes exhibit higher mutation rates compared to nuclear genes, and their evolutionary changes occur at a faster rate. Due to the high species diversity of scorpions in Khuzestan Province, scorpion stings and fatalities caused by them are of significant concern in the region. The present study investigates the phylogeny based on DNA sequences of the *COXI* gene of *A. susanae* scorpions, an invasive species collected from four regions of Khuzestan Province in southwestern Iran. The findings of this study indicate divergence between the species from Masjed Soleyman and Andimeshk. The genetic diversity observed between the populations of the Ramhormoz and Andimeshk regions can be attributed to the differing climatic conditions (temperature, relative humidity) between the two regions.

Additionally, this scorpion species was found to be closely related to *Androctonus* sp. species, as they all cluster in the same clade, suggesting a close genetic relationship between these two genera. Studies by Billington and Hebert (1991) and Quijano-Ravell et al. (2019) in Mexico, which investigated new species within the *Centruroides* sp. (Buthidae family) using mitochondrial *16S rRNA* gene sequences, used a genetic distance of around 10% between species as a criterion to confirm a new species. In the current study, however, the maximum genetic divergence observed was 0.1%, which suggests that all identified scorpions, including *A. susanae* identified in the GenBank, belong to the same species [19, 20].

In the study by Pirmoradi et al. (2023), which investigated the genetic diversity of scorpions in *Hottentotta* sp. in Khuzestan Province using *COXI* and *12sRNA*, it was reported that based on *12sRNA*, all *H. saulcyi* species (HS4, HS6, and HS7), except for species HS5, grouped in Cluster B, while two species of *Hottentotta Zagrosensis* (HZ6 and HZ1) were placed in cluster A. Additionally, Jolodar et al. (2023) in their study on the genetic diversity of the genus Scorpio based on *16sRNA* sequencing, constructed a phylogenetic tree with two clusters (A and B), which were closely related to *Scorpius maurus*. Both studies indicate that gene flow is influenced by geographic barriers, which is consistent with intraspecific divergence observed in the present study. In fact, the separation of species occurs due to distance and the presence of natural barriers. Therefore, geographic factors affect gene flow and genetic diversity, leading to species divergence [3, 21].

5. Conclusion

The results of this study show that genetically, *A. susanae* scorpions from the Masjed Soleyman and Andimeshk regions exhibit divergence.

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Compliance with ethical guidelines

This paper adheres to the academic ethics of the Department of Environmental Science and Desert Area Management Faculty of Engineering and Agriculture, Arak Branch, Islamic Azad University, Arak, Iran.

Data availability

The data that support the findings of this study are available from the corresponding author upon reasonable request.

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Authors' contributions

Conceptualization and study design: Bahman Shams Esfandabad and Hedieh Jafari; Data acquisition, analysis and data interpretation: All authors; Writing: Bahman Shams Esfandabad, Safie Bahri, and Abbas Ahmadi; Statistical analysis: Bahman Shams Esfandabad, Abbas Ahmadi, and Hedieh Jafari; Administrative, technical, and material support: Bahman Shams Esfandabad and Safie Bahri.

Conflict of interest

The authors declared no conflict of interest.

Reference

[1] Fet V. Fauna and zoogeography of scorpions (Arachnida: Scorpions) in Turkmenistan. In: Fet V, Atamuradov KI, editors. Biogeography and Ecology of Turkmenistan. Dordrecht: Springer Netherlands; 1994. [DOI:10.1007/978-94-011-1116-4_31]

[2] Dehghani R, Haghi FM, Mogaddam MY, Sedaghat M, Hajati H. Review study of scorpion classification in Iran. J Entomol Zool Stud. 2016; 4(5):440-4. [Link]

[3] Pirmoradi S, Jolodar A, Jafari H. Genetic diversity of Hottentotta sp. scorpions (Scorpionidae: Buthidae) in Khuzestan, Iran. Arch Razi Inst. 2023; 78(1):15-24. [DOI:10.22092/ari.2022.359194.2385] [PMID]

[4] Navidpour S, Studies B. An annotated checklist of scorpions in south and southwestern parts of Iran. Int J Fauna Biol Stud. 2015; 2(3):9-15. [Link]

[5] Mirshamsi O, Sari A, Elahi E, Hosseinie S. Phylogenetic relationships of Mesobuthus eupeus (C. L. Koch, 1839) inferred from COI sequences (Scorpiones: Buthidae). J Nat Hist. 2010; 44(47-48):2851-72. [DOI:10.1080/00222933.2010.512400]

[6] Jolodar A. Molecular characterization and phylogeny analysis based on sequences of cytochrome oxidase gene from Hemiscorpius lepturus of Iran. Iran J Vet Med. 2019; 13(1):59-67. [DOI:10.22059/ijvm.2018.245802.1004883]

[7] Garg RK, Dubey N, Batav N, Pandey P, Singh RK. Mitochondrial COI gene sequence analyses of Puntius ticto and compared with seven species of genus Puntius of family Cyprinidae: A finding for phylogenetic positioning and DNA barcoding as model study for cryptic species. J Proteomics Bioinformatics. 2017; 10(9):214-21. [DOI:10.4172/jpb.1000445]

[8] Augot D, Mathieu B, Hadj-Henni L, Barriel V, Zapata Mena S, Smolis S, et al. Molecular phylogeny of 42 species of Culicoides (Diptera, Ceratopogonidae) from three continents. Parasite. 2017; 24:23. [DOI:10.1051/parasite/2017020] [PMID]

[9] Lamoral BH. The scorpions of Namibia. Annals of the Natal Museum. 1979; 23: 497-784.

[10] Gantenbein B, Fet V, Largiader CR, Scholl A. First DNA phylogeny of Euscorpius Thorell, 1876 (Scorpiones, Euscorpiidae) and its bearing on taxonomy and biogeography of this genus. Biogeographica. 1999; 75(2):49-65. [Link]

[11] Lourenço WR, Goodman SM. Taxonomic and ecological observations on the scorpions collected in the forest of Ankazomivady-Ambositra and on the "RS d'Ivohibe", Madagascar. Rev Biol Trop. 1994; 47(3):475–82. [Link]

[12] Keegan HL. Scorpions of medical importance. Jackson: Mississippi University Press; 1980. [Link]

[13] Levy G. Amitai P. Scorpions. Jerusalem: The Israel Academy of Sciences and Humanities; 1980.

[14] Booncham U, Sitthicharoenchai D, Pradatsundarasar AO, Prasarnpun S, Thirakhupt K. Sexual dimorphism in the Asian Giant Forest Scorpion, Heterometrus laoticus Couzijn, 1981. Int J Sci. 2007; 4(1):42-52. [Link]

[15] Lane RP, Crosskey R. Medical insects and arachnids. Netherlands: Springer Netherlands; 2012. [Link]

[16] Dehghani R, Tirgari S. [Differential parameters of male and female Odontobuthus doriae (Persian)]. Feyz Med Sci J. 2012; 28:11-16. [Link]

[17] Nemat Elahi S, Shayista-Far A, Poserkello A. [Investigating quantitative traits to detect the sexual dimorphism of the scorpion Hottentotta sulcyi (Persian)]. Sci Res J Exp Anim Biol. 2018; 8(3):71-8. [Link]

[18] Oshaghi MA. mtDNA inheritance in the mosquitoes of Anopheles stephensi. Mitochondrion. 2005; 5(4):266-71. [DOI:10.1016/j.mito.2005.05.001] [PMID]

[19] Billington N, Hebert DN. Mitochondrial DNA diversity in fishes and its implications for introductions. Can J Fish Aquat Sci. 1991; 48(Supplement 1):80-94. [DOI:10.1139/f91-306]

[20] Quijano-Ravell AF, de Armas LF, Francke OF, Ponce-Saavedra J. A new species of the genus Centruroides Marx (Scorpiones, Buthidae) from western Michoacán State, México using molecular and morphological evidence. Zookeys. 2019; 859:31-48. [DOI:10.3897/zookeys.859.33069] [PMID]

[21] Jolodar A, Pourhosseini B, Jafari H. Genetic diversity within Scorpio genus (Scorpion: Scorpionidae) from Iran: Preliminary evidence based on 16s rRNA sequence. Arch Razi Inst. 2023; 78(5):1462-72. [DOI:10.32592/ARI.2023.78.5.1462] [PMID]