

Status of Striped Hyaena (Hyaena hyaena) in Hatay and Şanlıurfa-Turkey

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ARTICLE INFO	ABSTRACT		
Research Articles	In the present work, an 18 month survey, involving face to face interviews with the local people and deployment camera-traps, was carried out on about 600 km2 areas in Turkey. Field surveys and camera-trapping resulted in strong evidences that striped hyenas travel and live		
Received 03 May 2017 Accepted 04 July 2017	around Hatay (Syria border) and Şanlıurfa Provinces in Turkey. However, a comprehensive study was not carried out about the status of striped hyenas in this region. Our results showed that striped hyenas were frequently observed around the wasteland and the chicken farm in		
Keywords: Striped hyena Hyaena hyaena	Hatay Province. Alternatively, they live in small groups in Şanlıurfa Province. Several caves suitable for striped hyenas were identified by footprints, feces and other animal remnants		
Ecology Cytochrome oxidase b	observed in and before those caves. We performed molecular characterization of striped hyenas in Turkey for the first time using Cytb mitochondrial DNA isolated from hair, ear and carcass tissues. Sequences of Cytb DNA sequences from10 different striped hyena samples		
DNA barcoding	from Turkey were found to be identical to each other. Comparison of the sequences with the previously reported Cytb sequences, including prehistoric ones, showed that Cytb gene was highly conserved among the Hyaena hyaena species. During the field surveys we also observed		
Corresponding Author: E-mail: eatay@mku.edu.tr	that habitat destruction and fragmentation are detected on high level in the studied regions due to the intensive agricultural areas, settlements and quarries. Illegal hunting, frequent cave usage by shepherds, extensive porcupines hunting, water pollution from olive oil production facilities and highway crashes have been negatively affecting striped hyena population in		
	Hatay and Şanlıurfa regions. It appears that a comprehensive study and protection plan should be exerted to preserve the habitat of striped hyenas.		

Türk Tarım - Gıda Bilim ve Teknoloji Dergisi, 5(10): 1165-1172, 2017

Çizgili Sırtlanların (Hyaena hyaena) Hatay ve Şanlıurfa'daki Durumu

ÖZET		
Bu çalışmada, Türkiye'de yaklaşık 600 km2'lik alanda 18 ay süresince uygulanan yerli halkla yüz yüze görüşmeler, anket ve foto kapanların yerleştirilmesini içeren bir araştırma gerçekleştirilmiştir. Sahada yapılan anketler ve foto kapan çalışmaları, çizgili sırtlanların Hatay		
ile Suriye sınırı civarında ve Şanlıurfa'da güçlü şekilde yaşadıkları konusunda belirgin kanıtlarla sonuçlandı. Ancak, bu bölgede çizgili sırtlanların durumu hakkında kapsamlı bir çalışma yapılmamıştır. Elde edilen sonuçlarımız çizgili sırtlanın sıklıkla çorak arazide ve		
Hatay ilinde tavuk çiftliği etrafında görüldüğünü göstermiştir. Alternatif olarak, Şanlıurfa İli'nde küçük gruplar halinde yaşıyorlar. Çizgili sırtlanlara uygun birkaç mağara, bu mağaralarda ve sonrasında gözlemlenen ayak izleri, dışkılar ve diğer hayvan kalıntıları ile tespit edilmiştir. Çizgili sırtlanlara uygun birkaç mağara ve sonrasında yapılan gözlemlerde ayak izleri, dışkılar ve diğer hayvan kalıntıları tespit edilmiştir. Saç, kulak ve karkas dokularından izole edilen Cytb mitokondrial DNA'yı kullanarak, çizgili sırtlanların moleküler karakterizasyonunu Türkiye'de ilk defa gerçekleştirdik. Türkiye'den 10 farklı çizgili sırtlan örneğinden Cytb DNA sekanslarının dizilerinin birbirlerinin özdeşi oldukları bulunmuştur.		
Tarih öncesi olanlar da dahil olmak üzere daha önce rapor edilen Cytb dizilerin karşılaştırılması sonucu Ctyb geninin Hyaena hyaena türleri arasında oldukça muhafaza edildiğini göstermiştir. Arazi çalışması sırasında, yoğun tarım alanları, yerleşimler ve taş ocakları nedeniyle incelenen bölgelerde habitat tahribatının ve parçalanmanın yüksek düzeyde olduğu gözlemlenmiştir. Hatay ve Şanlıurfa illerinde, yasadışı avlanma, çobanlar tarafından sıklıkla mağaranın kullanımı, yaygın kirpi avlanması, zeytinyağı üretim tesislerinden gelen su kirliliği ve karayolu kazaları çizgili sırtlan nüfusunu olumsuz etkilemektedir. Çizgili sırtlanların yaşam alanını korumak için kapsamlı bir çalışma ve koruma planının uygulanması		

DOI: https://doi.org/10.24925/turjaf.v5i10.1165-1172.1295

Introduction

Being a large set of carnivores, the hyena family is represented by only 4 species on earth (Hyaena hyaena, Parahyaena brunnea, Crocuta crocuta, Proteles cristata) (Mills and Hofer, 1998; Koepfli et al., 2006; Yıldırım, 2010; Sheng et al., 2014). Regarding their general characteristics, striped hyenas have similar natural distributions; they prefer steps, semi-deserts, rocky fields and valleys with sparse trees (Mills and Hoffer, 1998). They avoid deserts, high altitude areas, dense bushes and forests. Striped hyenas prefer living inside caves (Mills and Hoffer, 1998; Yıldırım, 2010). Just like the rest of the world, these animals strive for survival with humans in their co-habitat. These animals also participate in the ecosystem by eating dead and decaying animal carcasses (Mills and Hofer, 1998; Bunaian et al., 2001; Singh, 2008; Stein et al., 2013). Dietary preferences of these species include spined (Bon et al., 2012) and non-spined animals, various fruits, vegetables and human-sourced organic wastes (Wagner, 2006). Being a dietary opportunist, hyenas are omnivore carcass eaters. When starved, they can feed on melons, watermelons, grapes and some other vegetables (Mills and Hoffer, 1998; Yıldırım, 2010).

Striped hyena lives on rocky mountains and valleys located in semi-deserts, steps, bushes and crooked pine forests of Turkey (Yıldırım, 2010). Striped hyaena is a carnivore with dog-like posture (Abi-Said, 2004). The back is inclined to the tail with vertical black stripes on sides of the body. Overall body color is pale-gray or beige. There are 5 - 9 distinguished vertical stripes on the body, vertical and horizontal light-black stripes on front and behind legs. Body mass is around 26-41 kg and 26-34 kg in females (Mills and Hofer, 1998; Yıldırım, 2010). The striped hyena gives birth to between one and four (average three) off springs after approximately 90 days of pregnancy (Wagner, 2006). The offspring begins feeding on meat after 30 days. They are reported to be fed on mother's milk until they are four to five months old. In Turkey, striped hyenas mate in January and February. They give birth in April and May and females are mature for mating when they are almost two - three years old (Wagner, 2006, Mills and Hofer, 1998; Yıldırım, 2010).

These species are distributed over North and Middle Africa, Anatolia, Arabian Peninsula, Middle East, Caucassia, Middle Asia and India (Rieger, 1981; Wagner, 2006; Bunaian et al., 2001; Mills and Hofer, 1998; Yıldırım, 2010). Kumerloeve, (1982) reported the first information on the presence of striped hyenas in Central, East, West and South-East Anatolia. Kasparek et al., (2004) indicated that the striped hyenas have a nonuniform distribution in Turkey and have been recorded in Çanakkale, İzmir, Antalya, Hatay and South-East Anatolia of Turkey in last 25 years. According to their distribution over a wide range of areas, regional adaptations and morphological changes, striped hyaenas are represented by the following species; Hyaena hyaena barbara (Northwest Africa), Hyaena hyaena dubbah (Northeast Africa), Hyaena hyaena sultana (Arabian Peninsula), Hyaena hyaena syriaca (Syria, Anatolia and Caucassia), Hyaena hyaena hyaena (India) (Rieger, 1981; Mills and Hofer, 1998; Qarqaz et al., 2004; Singh, 2008; Yıldırım, 2010).

The International Union for the Conservation of Nature (IUCN) has classified the striped hyenas as near threatened (Arumugam et al., 2008). Despite the important role of the striped hyenas in ecosystems, little effort has been devoted in studying them. Habitat changes across their range seem a possible cause of declining populations (Ripple et al., 2014). Evaluation of the population status and their habitat features at the local and regional level is essential for planning a conservation plan for viable populations. Monitoring these animals in their habitats appears to be a better strategy for such conservation approaches. Unfortunately, few studies have looked into the ecology of striped hyenas (Singh, 2008; Yıldırım, 2010).

In recent years, camera trapping has been broadly employed to study range of vertebrates (Cutler and Swann, 1999) and to collect quantitative information (O'Connell et al., 2011). Camera trapping is an efficient way to understand the biology of species and habitat relationships (Carter et al., 2012; Schuette et al., 2013). Striped hyenas can be identified individually especially from their fur patterns, and camera traps can be used to estimate their population density (Gupta et al., 2009; Harihar et al., 2010).

DNA barcoding, which uses short and standardized DNA sequences (again typically from a mitochondrial gene), can also be used to identify the known species and to discover new species (Herbert et al., 2004; Savolainen et al., 2005). The mitochondrial gene cytochrome c oxidase I (COI) has been frequently used for animal species after it was reported that the region could serve as a genetic barcode for animals (Hebert et al., 2003; Taylor and Harris, 2012). Other target DNA region was cytochrome b (cytb), which has been used for the phylogenetic analyses of mammals (Castresana, 2001, Quérouil et al., 2001; Tougard et al., 2001; Lecompte et al., 2002; Montgelard et al., 2002). Sequencing the mitochondrial genes such as cytb and comparing it with those of other species in the GenBank database have been frequently employed to genetically identify a particular species (Inoue and Akomo-Okoue, 2015). Arif et al., (2011) successfully differentiated striped hyenas from other carnivores using 130 bp part of COI region. Foran et al., (2015) used cytb and other mitochondrial primes for species identification and differentiated spotted hyena from other species.

In this study, the current status of carnivore striped hyenas were disccussed, as they are decreasing in numbers rapidly in Hatay and Şanhurfa Provinces, with their population sizes, distribution, habitats, diets and dietary sources, relationships and interactions with humans, the threats and dangers on them as well as the precautions and countermeasures to be taken for their protection. Also, mitochondrial cytochrome oxidase b (cytb) gene barcoding of Turkish striped hyenas was revealed and its relationship with other striped hyenas analyzed.

Material and Methods

Field Survey

The study was carried out between January 05, 2012 - July 05, 2013. About 600 km² of natural, agricultural and residential areas in Altınözü Wildlife Development Field, Antakya, Kırıkhan, Yayladağı and Birecik (Şanlıurfa) counties were surveyed to spot striped hyenas in their natural habitat. In the study field surveys were conducted, including the presence of the species, their shelters, caves, feces, food prints, and tissue samples. All the places on which striped hyenas were seen that recorded and located on the map (Fig. 1). Therefore, this study aimed at identfying and analzing the distribution, habitats, and shelters of striped hyenas.

Direct and indirect observation methods were used in the field studies. Indirect observation method included the interviews about striped hyenas with the locals, hunters, shepherds, workers in central waste yard and chicken farm. In Hatay Province, face to face interviews were made with 25 shepherds, 10 hunters and 45 local country people. Seven questions were directed to them as follows:

- Do you recognize striped hyena?
- Have you ever seen striped hyenas, if yes, how many times and where?
- How often did you see striped hyenas?
- Did you see striped hyenas on day or at night?
- Do you know anything about caves or shelters where striped hyenas live in? If yes, where are they?
- Have you encountered any dead striped hyena?
- Have you ever heard about striped hyena attacks to human or animals?

In Birecik county, field surveys were not be able to be carried out since striped hyenas have sedentary life in a particular location which is undisclosed due to the safety reasons. Therefore, according to interview results field surveys were initiated and cameras (Bushnell Trophy Cam) were deployed to sites for 5 days and were transferred to new sites for 5 day intervals for 18 months. This procedure was previously applied for 8 day periods for detecting wild animals (Kays et al., 2010). The study area was divided into four counties where striped hyenas were likely to be seen on corresponding latitudes and longitudes (Fig. 1). Therefore, a total of 44 camera deployment was performed in Altınözü, Antakya, Yayladağı and Kırıkhan Counties with 23, 11, 8, and 2 times, respectively (Fig. 1). Direct observation method was also included in the search of striped hyena, striped hyena carcass, their hair, foot prints, feces, food scraps and their shelters (Sing, 2008). Wastelands, chicken farms, caves, stream beds and paths that could be used by striped hyenas around Altınözü, Antakya, Kırıkhan, Yayladağı and Birecik Counties were searched for their appearances. Therefore, camera-traps were placed around those regions to investigate the distribution of striped hyenas. In order to attract striped hyena, parts of chicken, fish, sheep and goat carcasses have been placed to areas. Camera-traps were placed in a distance of 3-5 meters to those baits. It was picked up after 5 days and their records were investigated. A total of 25 spots in 3 out of 4 counties were investigated by indirect methods. For indirect observation, fine sand was spread in and before caves, on paths and around rivers to search for footprints. Footprints and feces were also sought on dirt roads, paths, sandy stream beds and around wasteland and chicken farms. Parallel line and cross (X) methods were used to determine the footprints (Yıldırım, 2010). When detected, striped hyena foot prints were pictured. Footprint records were preferably taken right after rainy weathers.

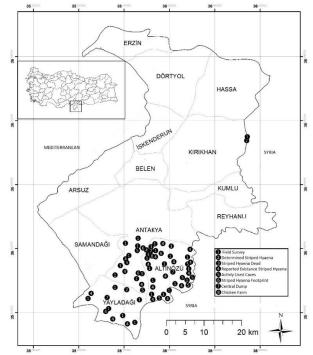


Figure 1 The study areas and distribution of Striped Hyaena (*Hyaena hyaena*) in Hatay, Turkey

DNA Isolation

Carcass (2 samples), hair (7 samples) and ear (1 sample) tissues of striped hyaenas were used for molecular analyses. BiospeedyTM DNA Isolation Kit (Bioeksen Ltd. Co., Turkey) was used for the DNA extraction. Briefly, 200 mg of the tissue sample was homogenized at 6000 rpm for 2 minutes in a buffer (2% CTAB -hexadecyltrimethylammonium bromide-, 100 mM TrisHCl pH 8, 20 mM EDTA, 1.4 M NaCl) containing 0.1 mm diameter glass beads. The samples were then incubated at 98°C for 10 minutes. The samples were centrifuged and the supernatant was combined with a binding buffer (final concentration of 3.4 M Guanidinium thiocyanate, 8 mM Tris-HCl pH 8.0, 25% isopropanol). The extracted DNA was captured using silica columns and then washed twice with a buffer containing 20 mM NaCl, 2 mM Tris-HCl, pH 7.5; 80% v/v Ethanol. The DNA was eluted in 100 mM Tris-HCl pH 8.0 and stored at -20°C.

Primer design: PCR primers were designed based on comparison of the available *Hyaena hyaena* mithocondrial cytochrome b (cytb) gene sequences retrieved from Genbank. The sequences were aligned using Clustal Omega (www.ebi.ac.uk) and the primers were designed manually.

5'-AACAACCGCCTTTTCATCAG-3' forward and 5'-

GGGTTGGCTGGTGTGTGTAGTT-3' reverse primers were designed to amplify 606 bp region of the ctyb. The specifity of the primers were tested using Primer-Blast (www.ncbi.nlm.nih.gov).

Real time PCR (Q-PCR): BiospeedyTM EvaGreen Master Mix (Bioeksen Ltd. Co., Turkey) and Biorad CFX Connect (Biorad Inc., U.S.A.) were utilized for all reactions. Reaction mixes contained 25 ng template DNA, 6mg/ml BSA, 5 mg/ml PEG 400, 0.25% Tween 20, 20 mM Tris-HCl pH 8.4, 50 mM KCl, 1.5 mM MgCl₂, 0.2 mM dNTP mix, 0.1U Proof Reading Hot-Start DNA Polymerase and 200 nM of each primer. The following thermocycling program was applied: 98° C, 3 min; 35 cycles of 10 s at 95° C, 5 s at 55° C and 20 s at 72° C. A melt curve analysis was performed from 65° C to 95° C to determine if only one amplified product was generated during Q-PCR. Q-PCR runs were analyzed using Biorad CFX Connect Software.

DNA sequencing and phylogenetic analysis: The Q-PCR products larger than 100 bp were purified using BiospeedyTM PCR Product Purification Kit (Bioeksen Ltd. Co., Turkey). The purified DNA was sequenced using the ABI prism Big Dye Terminator Cycle Sequencing Ready Reaction Kit on an ABI Prism 377 DNA sequencer (Applied Biosystems, USA). The analyzed sequence was in Chromas (www.technelysium.com) and manually checked for reading errors. Homology search of the sequence in DNA databases performed with BLAST was (www.ncbi.nlm.nih.gov). The sequence was submitted to the GenBank and; an accession number (KU881801) was assigned by the GenBank. The sequence and its closest matches in GenBank were aligned using Clustal Omega (www.ebi.ac.uk) followed by manual adjustments. Only unambiguously aligned base positions were used in the analysis. Neighbor-joining method was performed with the MEGA software (www.megasoftware.net) to construct an unrooted phylogenetic tree. The percentage of replicate trees in which the associated taxa clustered together in the bootstrap test (100 replicates) is shown next to the branches. The tree is drawn to scale, with branch lengths in the same units as those of the evolutionary distances used to infer the phylogenetic tree. The evolutionary distances were computed using the Poisson correction method and are in the units of the number of amino acid substitutions per site. All positions containing gaps and missing data were eliminated.

Results

Field Survey and Camera-Trapping Study

The striped hyenas are actually seen rare in Hatay Province while a flock of striped hyenas has sedentary life around Birecik County of Şanlıurfa, Turkey. However, we successfully spotted two striped hyenas in Hatay province by direct and indirect observation methods. Thereby, a total of 44 camera placements resulted in two striped hyena appearances around Kırıkhan and Yayladağı Counties while two dead bodies were found in Altınözü location near the central dump and a chicken farm where existence of striped hyenas was reported in two interviews and footprints were observed around. There were also six caves used actively in this county. The existence of striped hyena was also reported in Yayladağı County near the Syrian border, however, no traces were observed. On the other hand, one camera placement resulted in cameracapture of striped hvena in Kırıkhan County right by Syria border. This was interesting since there were not any reports of their existence in this area before. Ecology of striped hyenas in their natural habitat was also evaluated, and results were discussed in detail with respect to each site.

DNA Barcoding of Striped Hyaenas from Turkey

In order to investigate the effect of geographical distribution of the genetic diversity of hyena species, Cytb DNA sequence of dead hyena was compared to that of other hyena sequences. Results showed that Cytb DNA sequences of the studied 10 different striped hyena samples were identical to each other. The obtained Cytb DNA sequence was deposited to the database, which can be accessed from the GenBank (KU881801). Sequence of KU881801 was compared with the previously reported Cytb sequences (Table 1 and Fig. 2), and a phylogenetic tree was constructed. The evolutionary history was inferred using the Neighbour-Joining method. The optimal tree with the sum of branch length = 0.011 is shown. Sequences retrieved from prehistoric samples of Germany (AY048787), Greece (DQ157578), Russia (DQ157576), Angola (DQ157582) and Syria (DQ157577) clearly separated from existing of Turkey are (KU881801), Germany (AF153055, AY048788), France (JF894376) and U.S.A. (AY926678).

Organism	GenBank Accession No	Similarity to KU881801	Reference
Hyaena hyaena isolate Cerza	JF894376	566/566(100%)	Bon et al., (2012)
Hyaena hyaena	AY928678	566/566(100%)	Koepfli et al., (2006).
Hyaena hyaena isolate TP-Chris	AY048788	566/566(100%)	Albert, (2001)
Hyaena hyaena isolate X-321	AY048787	563/566(99%)	Albert, (2000)
Hyaena hyaena from Germany	AF153055	209/209(100%)	Albert, (2000)
Hyaena hyaena from Greece	DQ157578	180/181(99%)	Rohland et al., (2005)
Hyaena hyaena from Russia	DQ157576	180/181(99%)	Rohland et al., (2005)
Hyaena hyaena from Angola	DQ157582	179/181(99%)	Rohland et al., (2005)
<i>Hyaena hyaena</i> from Syria	DQ157577	179/181(99%)	Rohland et al., (2005)

Table 1 Comparison of KU881801 sequence with the previously reported cytb sequences

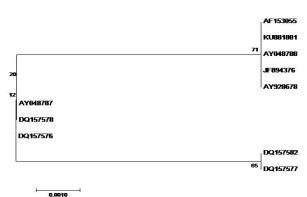


Figure 2 Genetic relationship of KU881801 with the previously reported cytb sequences

Discussion

The Evaluation of Sites Narlıca-Kuruyer-Enek of Antakya County

Some caves determined in the western and southern slopes of Adadağı Hill at the south of Narlıca District. According to the interviews, these caves and their surroundings were investigated for the existence of striped hyenas and at the two places footprints of striped hyena were found. In the south-western, the north-western and the north-eastern sides of Adadağı Hill, there are three quarries which could easily distract striped hyenas. The western side of the hill has a quarry and is more suitable to shelter striped hyenas. In the southern parts of the same territory, Mercimek Hill and Mağara Hill are other suitable habitats for striped hyenas. These locations were detected to have safe paths to reach the wasteland in Altınözü County. Küncülü Defile next to Altınözü county border is a narrow passage that striped hyenas generally use to visit the wasteland and chicken farm. On December 18th, 2008, a female striped hyena was killed by a car crash on this defile on Antakya-Altınözü highway (Fig. 3). In some cases around those hills, too many feeding remnants such as swine, sheep and goat head bones were encountered along with footprints of striped hyenas. According to the locals, striped hyenas were mostly seen in summer months, and the three quarries which use dynamites to shatter rocks affect striped hyenas not to settle and populate securely.

The Evaluation of Altınözü County

Altınözü County has big rocky hills located between Altınözü County Centre, Çetenli and Karsu Villages. The area is surrounded by olive trees and fruit groves. Striped hyenas could take shelter in this natural field. Around this area, there are also several caves which have entrances as big as 0.50-2.0 meters long, rendering hyenas with hiding and resting place. Some of these caves seemed to be used actively by foxes, coyotes, beech martens and badgers. Camera-traps also pictured boars, coyotes, foxes, badgers, beech martens, some field mice, lesser moles, levant vipers, water snakes, mountain black snakes around this area. Pazar Creek creates a valley on the southern and the south-eastern part of the rocky area. In fact, some caves in this valley were thought to be natural fields for hyenas to hide and take shelter. Also Pazar Creek would render a water supply and bushes which would give hyenas a chance to hide easily. Previously, from this rocky area a hunter (from Çetenli Village, Altınözü) caught three young striped hyenas and donated them to a Zoo. However, we encountered no trace of striped hyenas in this rocky area although some shepherds and locals said they could distinguish and encounter striped hyenas at night time from time to time.

The Evaluation of Altınözü Hills

Altınözü Hills are situated on the southeast of Altınözü county centre to the Syria border and are naturally protected areas. This large area shows so many similarities to habitats of striped hyenas (Yıldırım, 2010). There are valleys on this area which interconnect to others creating important passages leading to the wasteland and the chicken farm. Forest fires and explosions in Syria (caused by political disturbance) make striped hyenas to migrate to these hill sides. Nowadays, it is reported that there are more striped hyenas around these areas according to interviews with locals and shepherds encountered during field surveys.

Degraded pine forest fields, shrubberies and hallow valleys on southern lines of Kerim Hill, Marangoz Hill and Çamlıkaya Hill constitute a proper living space for striped hyenas. There are several caves in those valleys rendering shelters to striped hyenas. During field surveys, many feeding remnants, probably consumed by striped hyenas, and footprints of hyenas were found in front of and within caves (Fig. 4). This region is very important for Wildlife Development Field and sheltering wild animals such as striped hyenas due to limited human activities, highway rarity and the closeness to Syrian border.

There were also footprints of a striped hyena on a newly cultivated field at a distance of 300-400 meters in the southeast of the Central Wasteland situated in Altınözü County. The Central Waste yard is located on Uç Tepe local which is at the north of Karalidağı Hill at the northwest side of Altınözü County. The waste yard is the catchment basin for the subject species. There is also a full-capacity working chicken farm in a large acreage. Daily disposals of this farm (Fig. 5) are thrown to the wasteland which is near to the farm. These chicken remnants constitute an important part of wild animals' alimentation. Information about chicken disposals to the wasteland was confirmed by workers of the chicken farm. Such a wasteland should be a proper area for feeding of striped hyenas which are well known to eat dead animal carcasses.

The Evaluation of Some Fields around Antakya and Yayladağı County

Şenköy, Hanyolu, Çayır, Bozlu, Şakşak and Yukarıokçular Villages in Yayladağı County are considered to be suitable sites for roaming of striped hyenas. According to interviews with locals and shepherds during field surveys, striped hyenas were seen around this region from time to time. Some shepherds reported that some hyenas had visited Yayladağı County wasteland for scavenging food. Many caves, in which there were many dead animal remnants brought by other animals, were also encountered around this region during field surveys.



Figure 3 A female Striped Hyena killed in a traffic accident



Figure 4 A cave in where the bones were found



Figure 5 Chicken carcasses and dump and used for feeding Striped Hyaena



Figure 6 A striped hyena in Şakşak Village (Hatay) in 2013

Several footprints of striped hyenas have been detected in or around those caves. These caves are thought to be used actively by striped hyenas. Officers of Nature Protection and National Parks Antakya Branch monitored a striped hyena in Şakşak Village at 2013 with camera-traps (Fig. 6).

The Evaluation of Şanlıurfa-Birecik County Fields

In Şanlıurfa and Birecik, striped hyenas generally prefer caves around semi desert fields and sparse vegetation fields with rocky and steep surroundings. According to field surveys, there are 8 actively used caves (Figs. 7, 8).

Although the definite number was not obtained, 15 individuals are thought to be living in this region. Striped hyenas living in this region tend to use caves on north during summer and on south during winter months. Apart from males, female individuals do not leave their shelters during reproductive seasons. Striped hyenas living around this region prefer to search for food on fields, wastelands and places close to human settlements. They feed especially on animal carcasses, porcupines, various reptiles, bugs, watermelons, various vegetables and figs. Most important factors threatening striped hyenas in this region are traffic accidents, hunting, entry of hunters and shepherds to caves and the invasion of habitats of animals by humans. At least two striped hyenas were killed by traffic accidents around this region in 2014 (Nature Protection and National Parks of Şanlıurfa Region, 2014).

Molecular Identifaction of Striped Hyeana

Barcoding of wild animals is a useful tool to track animals even using animal parts such as hairs, faces, urines. In the present study, universal primers successfully amplified Ctyb region of mitochondrial DNA (mtDNA) and similarity among other striped hyenas from different geographical region was analyzed. In a study, wolf range expansion in France and Switzerland has been traced back using mtDNA isolated from faces and hair samples and successfully distinguished maternal lineages of wolf (Valière et al., 2003). Compared Cytb sequences of striped hyenas were found to be highly conserved within the species from very distant geographical locations. Prehistoric striped hyenas from Syria and Angola seem to have the same maternal ancestry. On the other hand, sequences of existing hyena samples had identical to those prehistoric European hyena samples. The existing hyenas interestingly had conserved cybt sequences regardless of the geographical distance even though striped hyenas have more nomadic nature. The Cytb sequence from Turkey is less similar to the sequences from geographically close locations such as Syria and Greece. Genetic distance between cave and spotted hyenas was found to be lower than that with striped hyenas (Rohland et al., 2005). Contrary to striped hyenas wolves show more genetic diversity as the distance gets larger (Mech, 1987).

Threats on Striped Hyenas and Their Habitats

Habitat destruction and fragmentation is on high level in the studied regions due to the intensive agricultural areas, settlements and quarries. Shattering rocks with heavy equipment and dynamite usage of quarries disturb striped hyenas which search for food, take shelter and reproduce. In such cases striped hyenas were forced to leave their caves.

Too many gun cartridges were found on fields, and their pictures were taken. During field surveys, several hunters were encountered, whose ages are well below 18 having no legal permission to hunt. There are also small animals such as porcupines which become a source of food for striped hyenas. Unfortunately, porcupines are frequently hunted and eaten by locals. Therefore, illegal hunting appears to be a big threat for striped hyenas.

Ovine husbandry is one of means of livelihood of locals. Many shepherds were encountered during the field studies, and they stated they frequently use caves, possibly sheltering striped hyenas, during cold and rainy weather. Additionally, they make fire in caves and fields where striped hyenas roam.

Altmözü County is the biggest olive growing and processing region in Hatay. Especially during olive oil production and processing season, so much olive and olive oil waste are disposed by factories to small rivers around this area. Pollution of water sources directly and indirectly threatens wild animals including striped hyenas.



Figure 7 A Striped Hyaena in Şanlıurfa Birecik (2013)



Figure 8 A Striped Hyena in Şanlıurfa Birecik (2012)

Highways and cross roads the animal passages and many times traffic accidents involving striped hyenas as well as other wild animals are unavoidable. Each year two- five striped hyenas are recorded to be killed by traffic accidents around the region we studied.

Suggestions for Protection of Striped Hyenas

Population of striped hyenas on earth, thought to be between 5-14 thousands (Mills and Hofer, 1998). This species was included in the 2008 Red List of International Union for Conservation of Nature (IUCN) as near threatened species (Yıldırım, 2010). Since striped hyenas constantly enter and exit through Syria and Hatay border, there are not a definite individual number of animals living in Hatay. However, combining the field data involving footprints and camera-trap results with interviews it is considered that about 20 individuals live in Hatay. Therefore, to protect this rare species in this region, some suggestions listed below should be considered urgently:

- Protection of striped hyenas should not be limited to Altınözü Wildlife Development Field; instead, protection plans should be imposed much to large areas and focus on protecting the species, not to the area particularly. Therefore, locals should be frequently informed about protection of striped hyenas and their ecology by short seminars.
- Feeding and reproducing habitats and shelters of striped hyenas are frequently intruded by various trespassers. These small living areas should be protected without disturbing the other biological niches. Hunters, villagers and shepherds should be warned not to enter the caves. Land hunting and hunting with various traps should be monitored on Wildlife Protection Field region and strict regulations should be exerted for illegal hunting.
- Locals should be warned on not to fire stubbles, yellowed grass and scrubs in or around caves where striped hyenas may exist.
- According to interviews around Hatay and Şanlıurfa, there are some untrue stories in a way that striped hyenas attack people and ovine, and they open new graves to eat corpses. The threats referred above will negatively affect striped hyenas living in Hatay and Şanlıurfa and their habitats, so this would lead to decrease their population and make them become extinct.
- At some seasons, striped hyenas may have food shortage. Therefore, to maintain their survival animal wastes obtained from slaughterhouse should be left around places where striped hyenas live.
- Warning signs, involving information about protecting striped hyenas, should be placed along the highways close to the passages. In addition, overpasses and underpasses should be built on important passage points of striped hyenas.
- Quarries should be audited frequently not to use dynamites especially during reproductive season of striped hyenas.
- Since this species is rare and spread on large areas, an urgent and comprehensive long term inventory and monitoring plans should be implemented for protection of striped hyenas.

This study is one of the most comprehensive studies on striped hyenas around Şanlıurfa and Hatay Regions, the northernmost boundary of their distribution area. In the present study, nomadic nature of striped hyenas in Hatay Region was investigated and possible living areas were identified through camera-trap method as well as field surveys involving locals. In addition, for the first time, striped hyenas of Turkey were genetically characterized. The results of this study are expected to help further monitoring the striped hyenas and protect them with their habitats.

Acknowledgements

Thanks to Nature Protection and National Parks Antakya Branch and Dear İsmet Ceyhun Yıldırım for sharing pictures on this article.

References

- Abi-Said MR. 2004. Breeding potential of the striped hyaena, Hyaena hyaena syriaca
- Matschie, 1910 (Carnivora) in captivity. Zoo in the Middle East 33:79-86.
- Albert R. 2000. Genetic identification of the geographic origin of spotted hyaenas (*Crocuta crocuta*). Zoo. Gart 70: 1-10.
- Albert R. 2001. Gene structure and gene flow in selected populations of spotted hyaena (*Crocuta crocuta*). Thesis Freie Universita et Berlin.
- Arif IA, Khan HA, Sadoon Al, Shobrak M. 2011. Limited efficiency of universal mini-barcode primers for DNA amplification from desert reptiles, birds and mammals. Gen Mol Res 10: 3559-3564.
- Bon C, Berthonaud V, Maksud F, Labadie K, Poulain J, Artiguenave F, Wincker P, Aury J, Elalouf J. 2012. Coprolites as a source of information on the genome and diet of the cave hyena. Proceedings Royal Soc B 1-6.
- Bunaian F, Hatough A, Ababaneh D, Mashaqbeh S, Yousefand M, Amr Z. 2001. The Carnivores of the Northeastern Badia, Jordan. Türk J Zool 25:19-25.
- Castresana J. 2001. Cytochrome b phylogeny and the taxonomy of great apes and mammals. Molecular Bio and Evolution 18: 465-471.
- Foran DR, Fischer AB, Stoloff ME. 2015. A Comparison of Mitochondrial DNA Amplification Strategies for Species Identification. J Forensic Inves 3: 1-7.
- Hebert PDN, Cywinska A, Ball SL, DeWaard JR. 2003. Biological identifications through DNA barcodes. Proceedings of the Royal Society of London Series B-Bio Sci 270: 313-321.
- Herbert PN, Stoeckle MY, Zemlak TS, Francis CM. 2004. Identifi cation of birds through DNA barcodes. Pub Lib of Sci Bio 2: 1657–1663.
- Inoue E, Akomo-Okoue EF. 2015. Application of DNA barcoding techniques to mammal inventories in the African rain forest: droppings may inform us of the owners. Tropics 23: 137-150.
- Kasparek M, Kasparek A, Gözcelioğlu B, Çolak E, Yiğit N. 2004. On the status and distribution of the Striped Hyaena, *Hyaena hyaena*, in Turkey. Zoo in the Middle East 33: 93-108.
- Kays R, Tilak S, Kranstauber B, Jansen PA, Carbone C, Rowcliffe MJ, Fountaine T, Eggert J, He Z. 2010. Monitoring wild animal communities with arrays of motion sensitive camera traps. arXiv preprint arXiv: 1009-5718.

- Koepfli KL, Jenks SM, Eizirik E, Zahirpour T, Valkenburgh BV, Wayne RK. 2006. Molecular systematics of the Hyaenidae: Relationships of a relictual lineage resolved by a molecular supermatrix. Mol Phylo and Evol 38: 603–620.
- Kumerloeve H. 1982. Historical development of research and findings on Anatolian mammalians. İstanbul University Forest Faculty J 32: 265-273.
- Lecompte E, Granjon L, Peterhans JK, Denys C. 2002. Cytochrome b-based phylogeny of the Praomys group (Rodentia, Murinae): a new African radiation. Comptes Rendus Bio 325: 827-840.
- Mech LD. 1987. Age, season, distance, direction, and socialaspects of wolf dispersal from a Minnesota pack. In Mammali and Ispersal Patterns Chepko-Sade BD, Halpin ZT (Eds). Chicago: University of Chicago Press, pp. 55–74.
- Mills MGL, Hofer H. 1998. Hyaenas. Status Survey and Conservation Action Plan. IUCN/SSC Hyaena Specialist Group. IUCN, Gland, Switzerland and Cambridge, UK.
- Montgelard C, Bentz S, Tirard C, Verneau O, Catzeflis FM. 2002. Molecular Systematics of Sciurognathi (Rodentia): The mitochondrial Cytochrome b and 12S rRNA genes support the Anomaluroidea (Pedetidae and Anomaluridae). Mol Phylo and Evo 22: 220-233
- Nature Protection and National Parks of Şanlıurfa Region. 2014. Action plan for Striped hyenas (*Hyaena hyaena*) in Şanlıurfa Province. Orman ve Su İşleri Bakanlığı, pp. 1-36.
- Qarqaz AM, Abu Baker A, Amr ZS. 2004. Status and ecology of the Striped Hyaena, in Jordan. Zool in the Middle East 33:87-92.
- Rieger I. 1981. Hyaena hyaena. Mammalian Species 150: 1-5.
- Rohland N, Pollack JL, Nagel D, Beauval C, Airvaux J, Pääbo S, Hofreiter M. 2005. The population history of extant and extinct hyenas. Mol. Biol. Evol. 22: 2435–2443.
- Rohland N, Pollack JL, Nagel D, Beauval C, Airvaux J, Pääbo S, Hofreiter J. 2005. The population history of extantand extinct hyenas. Mol Bio and Evo 22: 2435-2443.
- Savolainen VR, Cowan S, Vogler AP, Roderick GK, Lane R. 2005. Towards writing the encyclopedia of life: An introduction to DNA barcoding. Philosophical Transactions of the Royal Society of London Series B, 360: 1805–1811.
- Sheng G, Soubrier J, Liu J, Werdelin L, Llamas B, Thomson VA, Tuke JH, Wu LJ, Hou XD, Chen QJ, Lai XL, Cooper A. 2014. Pleistocene Chinese cave hyenas and the recent Eurasian history of the spotted hyena, *Crocuta crocuta*. Mole Eco 23: 522–533.
- Singh P. 2008. Population density and feding ecology of the striped Hyaena (*Hyaena hyaena*) in relation to land use patterns in an arid region of Rajasthan. The Manipal University, pp. 1-48.
- Stein AB, Fuller TK, Marker LL. 2013. Brown Hyaena Feeding Ecology on Namibian Farmlands. South African J of Wildlife Res 43: 27-32.
- Taylor HR, Harris WE. 2012. An emergent science on the brink of irrelevance: a review of the past 8 years of DNA barcoding. Mole Eco Res 12: 377-388.
- Valière N, Fumagalli L, Gielly L, Miquel C, Lequette B, Poulle ML, Taberlet P. 2003. Long-distance wolf recolonization of France and Switzerland inferred from non-invasive genetic sampling over a period of 10 years. Animal Con 6: 83-92.
- Wagner AP. 2006. Behavioral ecology of the striped Hyaena (*Hyaena hyaena*). Montana State University, pp. 1-182.
- Yıldırım İC. 2010. Hatay yöresinde çizgili sırtlan (Hyaena hyaena L.) ekolojisi üzerine araştırmalar. Kahramanmaraş Sütçü İmam Üniversitesi, Fen Bilimleri Enst. Orman Müh. ABD, Kahramanmaraş, Türkiye.