Kafkas Univ Vet Fak Derg
19 (6): 1061-1063, 2013
DOI: 10.9775/kvfd.2013.9275

Journal Home-Page: http://vetdergi.kafkas.edu.tr
Online Submission: http://vetdergikafkas.org

CASE REPORT

Edwardsiella tarda Associated Subcutaneous Abscesses in A Captive Grass Snake (*Natrix natrix*, Squamata: Colubridae)

Lóránd Béla KÖBÖLKUTI [1,†] Gábor Árpád CZIRJÁK [1,2,†] Miklós TENK [3]
Attila SZAKÁCS [1] Attila KELEMEN [4] Marina SPINU [1]

† *These authors contributed equally*

[1] Department of Infectious Diseases, Faculty of Veterinary Medicine, University of Agricultural Sciences and Veterinary Medicine, Mănăştur str. 3-5, 400372 Cluj-Napoca, ROMANIA

[2] Department of Wildlife Diseases, Leibniz Institute for Zoo and Wildlife Research, Alfred-Kowalke-Str. 17, 10315 Berlin, GERMANY

[3] Central Agricultural Office, Veterinary Diagnostic Directorate, Tábornok u. 2, 1149 Budapest, HUNGARY

[4] Vivarium, Faculty of Biology and Geology, University Babeş-Bolyai, Clinicilor str. 5–7, 400006 Cluj-Napoca, ROMANIA

Makale Kodu (Article Code): KVFD-2013-9275

Summary

Edwardsiella tarda is an opportunistic pathogen associated with aquatic environments. It has been identified as part of the normal flora of the digestive tract of healthy crocodiles, alligators, turtles, lizards and snakes. However, data on its association with different clinical diseases in reptiles, especially in snakes, is scarce. A female grass snake *(Natrix natrix)* was presented with low body condition and with multiple subcutaneous nodules. After surgical cleaning of a nodule, *E. tarda* was cultivated in pure culture from the abscesses. To the author's best knowledge, this is the first reported clinical case linked to *E. tarda* in snakes and one of the first case reports in grass snake.

Keywords: *Subcutaneous abscesses, Captivity, Edwardsiella tarda, Natrix natrix*

Bir Küpeli Yılanda (*Natrix natrix*, Squamata: Colubridae) *Edwardsiella tarda* İle Birlikte Seyreden Subkutan Apse Olgusu

Özet

Edwardsiella tarda sulak çevrelerle ilgili oportunist bir patojendir. Sağlıklı timsahlar, kaplumbağalar, kertenkeleler ve yılanların sindirim sisteminin normal florasının bir parçasıdır. Ancak, sürüngenlerde, özellikle yılanlarda, farklı klinik hastalıklarla olan ilişkilerine yönelik veriler sınırlıdır. Bir dişi çayır yılanı *(Natrix natrix)* düşük vücut kondüsyon ve çok sayıda subkutan nodüllerle birlikte sunuldu. Nodülün birinin cerrahi temizliğini takiben, apselerden *E. tarda*'nın saf kültürü izole edildi. Yazarın bildiğine göre, bu ilk defa rapor edilen yılanlarda *E. tarda* ile ilgili klinik olgudur ve çayır yılanında ilk raporlardan biridir.

Anahtar sözcükler: *Subkutan apseler, Gözetim, Edwardsiella tarda, Natrix natrix*

INTRODUCTION

Bacterial species of the genus *Edwardsiella* are associated with aquatic habitats, being frequently isolated from fresh-water fishes, amphibians, reptiles and their environment [1]. Two members of the genus, *E. tarda* and *E. ictaluri*, are important fish pathogens [2], while *E. hoshinae* was isolated from wild birds, lizards and turtles without reported pathogenicity [3,4].

E. tarda causes serious systemic disease, especially in eels *(Anguillidae)* and catfishes *(Ictaluridae)*, however recent studies on natural- and experimental infections suggested that probably all fish species are susceptible under certain conditions (e.g. stress, pollution) [2]. Besides fishes, the bacterium causes enteric disease in different aquatic bird species and invades opportunistically the sick and injured marine mammals [5,6]. It also has zoonotic potential, being associated with both gastrointestinal and extraintestinal infections in humans [7]. In reptiles, the

İletişim (Correspondence)
+49 30 5168 214
czirjak@izw-berlin.de

bacteria was isolated both from oral cavity and cloaca of healthy individuals, however studies on its association with different clinical symptoms, especially in snakes, are scarce.

This report presents for the first time the clinical expression of *E. tarda* infection in snakes.

CASE HISTORY

A captive female grass snake *(Natrix natrix,* Squamata: Colubridae*)* was referred to the Department of Infectious Disease of the Faculty of Veterinary Medicine Cluj-Napoca. The animal belonged to the Vivarium of the University Babeş-Bolyai and was captured by fishermen in Mărtineşti, Cluj County, Romania. The animal was kept in a terrarium replicating the natural environment of the species and it was fed weekly with small crucian carps *(Carassius carassius)* and tadpoles.

At the clinical examination, the snake presented low body condition and several subcutaneous nodules of different sizes, dispersed almost all over the body surface *(Fig. 1)*.

After local anaesthesia with 2% lidocaine, one of the abscesses was opened and microbiological samples were collected in order to establish the microbial agents involved and to identify the optimal antibiotic treatment based on disk diffusion susceptibility test. The swabs were streaked for isolation onto Columbia agar supplemented with 5% sheep blood for fastidious Gram-negative and Gram-positive bacteria and onto Drigalski agar to select Gram-negative lactose-positive bacteria. After 24 h of incubation at 37°C, a pure culture of lactose-negative, Gram negative bacteria was obtained. The isolate was identified as *E. tarda* using Analytical Profile Index (API) 20E biochemical test strips in accordance with the manufacturer's directions (BioMérieux, France). The sensitivity of the isolate to various antimicrobials commonly used in exotic practices was evaluated as mentioned before.

Fig 1. Female grass snake *(Natrix natrix)* with multiple subcutaneous abscesses *(white arrows)*

Şekil 1. Dişi çayır yılanında *(Natrix natix)* çok sayıda subkutan apse odakları *(beyaz oklar)*

Based on susceptibility results a treatment was initiated with Enrofloxacin (10 mg/kg b.w. im; Bayer Corporation) for nine days, 5 ml Duphalyte (Pfizer) and 5 ml Glucose 5%, both administered by intra-coelomic route every second day for 2 weeks.

Despite the treatment, the condition and the health status of the animal deteriorated and ten days later, due to ethical considerations, we were forced to euthanize the snake using ketamine 20 mg/kg b.w. followed by freezing [8].

DISCUSSION

Previous studies suggested that *E. tarda* is part of the normal flora of the digestive tract of reptiles [9], the bacteria being isolated from American crocodiles *(Crocodylus acutus)* [10], American alligators *(Alligator mississippiensis)* [11], Chinese soft-shelled turtles *(Trionyx sinensis)* [12], Geoffroy's toadhead turtles *(Phrynops geoffroanus)* [4], tokay geckos *(Gekko gecko)* [13] and Brook's House geckos *(Hemidactylus brookii)* [14], garter snakes *(Thamnophis* spp.) [15], South American rattlesnakes *(Crotalus durissus terrificus)* [16] or tiger snakes *(Notechis scutatus)* [17]. Besides the healthy carrier status of different reptile species, septicaemia associated with *E. tarda* was reported in captive crocodiles and in Chinese soft-shelled turtles [18,19]. To our best knowledge, the present study is the first to associate clinical symptoms with *E. tarda* infection in snakes, causing multiple subcutaneous abscesses in a captive grass snake *(Natrix natrix)*.

Similarly to earlier studies reporting clinical edwardsiellosis in reptiles [18,19], the described animal was also a captive one. It has been shown that captivity increases the levels of corticosterone, which leads to decreased immunocompetence [20]. Such captivity related immunosuppression could have been underlying the infection with this opportunistic pathogen.

Although *E. tarda* was found as part of the bacterial flora in several snake species [15-17], previous attempts to isolate this microorganism from free-living grass snakes failed [21]. Since the captive grass snake was fed with tadpoles and fishes, both taxa being known as reservoirs for *E. tarda* [2,22], there is a strong argument for a food -borne infection in this case.

The grass snake is one of the most frequent snake species in Europe, and it is a common native reptile brought to veterinarians either as a wildlife casualty [8] or as pet [23]. Despite these facts, there are only two studies on bacterial diseases of this species [23,24]. While one of them reported the susceptibility of the grass snake to experimental infection with different *Mycobacterium* species [24], the second

presented a clinical case similar to the one described in the present study, associating *Salmonella typhymurium* with clinical symptoms [23]. Both reports stressed the importance of dermatologic problems in captive snakes.

The present study not only described a new clinical case in a common European native snake species, but also reports the first edwardsiellosis case in snakes.

REFERENCES

1. Sakazaki R: Genus XI. *Edwardsiella* Ewing and McWhoter 1965. **In,** Brenner DJ, Krieg NR, Staley JT (Eds): Bergey's Manual of Systematic Bacteriology. Vol. Two, Part B - The Gammaproteobacteria. 2nd ed., 657-661, Springer, 2005.

2. Plimb JA: *Edwardsiella* Septicaemias. **In,** Woo PTK, Bruno DW (Eds): Fish Diseases and Disorders. Vol. 3: Viral, Bacterial and Fungal Infections. 1st ed., 479-521, CAB International, 1999.

3. Grimont PAD, Grimont F, Richard C, Sakazaki R: *Edwardsiella hoshinae*, a new species of Enterobacteriaceae. *Curr Microbiol*, 4 (6): 347-351, 1980.

4. Ferronato BO, Marques TS, Souza FL, Verdade LM, Matushima ER: Oral bacterial microbiota and traumatic injuries of free-ranging *Phrynops geoffroanus* (Testudines, Chelidae) in southeastern Brazil. *Phyllomedusa*, 8 (1): 19-25, 2009.

5. White FH, Simpson CF, Williams Jr. E: Isolation of *Edwardsiella tarda* from aquatic animal species and surface waters in Florida. *J Wildlife Dis*, 9 (3): 204-208, 1973.

6. Coles BM, Stroud RK, Sheggeby S: Isolation of *Edwardsiella tarda* from three Oregon sea mammals. *J Wildlife Dis*, 14 (3): 339-341, 1978.

7. Janda JM, Abbot SL: Infections associated with the genus *Edwardsiella*: the role of *Edwardsiella tarda* in human disease. *Clin Infect Dis*, 17, 742-748, 1993.

8. Maclean B: Dealing with casualty wild amphibians and reptiles. *Vet Nursing J*, 27 (6): 220-225, 2012.

9. Roggendorf M, Müller HE: Enterobacteria of reptiles. *Zentralbl Bakteriol Orig A,* 236 (1): 22-35, 1976.

10. Charruau P, Pérez-Flores J, Pérez-Juárez JG, Cedeño-Vázquez JR, Rosas-Carmona R: Oral and cloacal microflora of wild crocodiles *Crocodylus acutus* and *C. moreletii* in the Mexican Caribbean. *Dis Aquat Org*, 98, 27-39, 2012.

11. Johnston MA, Porter DE, Scott GI, Rhodes WE, Webstre LF: Isolation of faecal coliform bacteria from the American alligator *(Alligator mississippiensis)*. *J Appl Microbiol*, 108, 965-973, 2010.

12. Sugita H, Deguchi Y: Microflora in the gastrointestinal tract of soft-shelled turtle *Trionyx sinensis*. *Bull Jap Soc Sci Fisheries*, 49 (2): 197-201, 1983.

13. Tan RJ, Lim EW, Ishak B: Intestinal bacterial flora of the household lizard, *Gecko gecko. Res Vet Sci,* 24 (2): 262-263, 1978.

14. Gugnani HC, Oguike JU, Sakazaki R: Salmonellae and other enteropathogenic bacteria in the intestines of wall geckos in Nigeria. *Antonie van Leeuwenhoek*, 52 (2): 117-120, 1986.

15. Goldstein EJC, Agyare EO, Vagvolgyi AE, Halpern M: Aerobic bacterial oral flora of Garter snake: Development of normal flora and pathogenic potential for snakes and humans. *J Clin Microbiol*, 13 (5): 954-956, 1981.

16. Ferreira Jr RS, Siqueira AK, Campagner MV, Salerno T, Soares TCS, Lucheis SB, Paes AC, Barraviera B: Comparison of wildlife and captivity rattlesnakes *(Crotalus durissus terrificus)* microbiota. *Pesq Vet Bras*, 29 (12): 999-1003, 2009.

17. Iveson JB: Strontium chloride B and E.E. enrichment broth media for the isolation of *Edwardsiella, Salmonella* and *Arizona* species from tiger snakes. *J Hyg (Lond)*, 69, 323-330, 1971.

18. Buenviaje GN, Ladds PW, Melville L, Manolis SC: Disease husbandry associations in farmed crocodiles in Queensland and the Northern Territory. *Aust Vet J*, 71, 165-173, 1994.

19. Pan XY, Hao GJ, Yao JY, Xu Y, Shen JY, Yen WL: Identification and pathogenic facts studying for *Edwardsiella tarda* from Edwardsiellosis of *Trionyx sinensis*. *Freshwater Fisheries,* 40 (6): 42-47, 2010.

20. Johnstone CP, Reina RD, Lill A: Interpreting indices of physiological stress in free-living vertebrates. *J Comp Physiol B,* 182, 861-879, 2012.

21. Cooper JE, Needham JR, Lawrence K: Studies on the cloacal flora of three species of free-living British reptile. *J Zool, Lond (A)*, 207, 521-525, 1985.

22. Sharma VK, Kaura YK, Singh IP: Frogs as carriers of *Salmonella* and *Edwardsiella*. *Antonie van Leeuwenhoek*, 40, 171-175, 1974.

23. Rostami A, Memarian I, Zahraei Salehi T, Dehghan MM, Masoudifard M, Mohammadi E, Ashrafi Tama I: The report of *Salmonella typhimurim* in *Natrix natrix*: detection and identification by culture and multiple PCR methods. **In,** Wibbelt G, Kretzschmar P, Hofer H, Seet S (Eds): *Proceedings of the International Conference on Diseases of Zoo and Wild Animals 2009.* pp.312-316, Leibniz Institute for Zoo and Wildlife Research, 2009.

24. Griffith AS: The susceptibility of the water (or grass) snake *(Trepidonotus natrix)* to the avian tubercle bacillus and to reptilian strains of acid-fast bacilli. *J Hyg (Lond)*, 41 (3): 284-288, 1941.