Clinical Case Report

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"Message from a turtle": otitis with *Salmonella arizonae* in children

Case report

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Abstract

Rationale: *Salmonella enterica* subsp *arizonae* is a common gut inhabitant of reptiles (snakes are the most common reservoir, but it also occurs in turtles). Alhough human cases owing to this organism are exceedingly rare, it may occasionally infect young infants and immunocompromised individuals with a history of intimate associations with reptiles. Our case is the 20th one among the infections with *S arizonae* in children, but the 2nd one of otitis and the first of mastoiditis. The other cases had different anatomical locations, such as gastroenteritis, osteomyelitis, meningitis, ankle infection, wound infection, and sinusitis.

Patient concerns and diagnosis: We report a rare case of otitis with *Salmonella* in a previously healthy adolescent, which was most likely acquired after bathing in a lake. The ear infection was complicated with mastoiditis. Audiometric testing showed a moderately conductive hearing loss (60 dB on pure-tone average).

Intervention: Standard therapy for *S arizonae* was initiated. The surgery revealed a "hidden" cholesteatoma. Surgical management comprised of canal wall up mastoidectomy with attico-antrotomy and posterior tympanotomy followed by tympanoplasty.

Outcomes: Daily postoperative dressing care of the incision, along with antibiotic lavage of the external auditory canal packing, ensured a favorable evolution. The functional gain was important; the 1-month postsurgical pure tone audiogram indicated nearly normal hearing (a mean of 25 dB for air conduction thresholds).

Lesson: *Salmonella enterica* serotype *arizonae* is a rare cause of human infection, being a common organism in reptiles, like snakes and turtles. Young children are at a particular risk for acquiring such infections. Our study might encourage further epidemiologic investigations into these infections to generate a more effective strategy among public health agencies.

Abbreviations: CT = Computed tomography, EAC = External auditory canal, PTA = pure-tone average, S. enterica = Salmonella enterica, TM = Tympanic membrane.

Keywords: children, cholesteatoma, mastoiditis, otitis, *S arizonae*

1. Introduction

Salmonella is a Gram-negative facultative anaerobic bacilli that belongs to the *Enterobacteriaceae* family, and it is usually responsible for food-borne diseases. Approximately 99% of human isolates are *Salmonella enterica* subsp *enterica*. Less-frequent subspecies are: *S enterica* subsp *houtenae* (IV), *S enterica* subsp *diarizonae* (IIIb), *S enterica* subsp *salamae* (II), *S enterica* subsp *arizonae* (IIIa), and *S enterica* subsp. *indica* (VI). The *Arizona* group, subgenus III of genus *Salmonella*, has received various names (*Salmonella arizonae, Arizona arizonae, Arizona hinshawii)* and was finally reclassified as *S enterica* subsp *arizonae* (monophasic strains, IIIa) and *S enterica* subsp *diarizonae* (diphasic strains, IIIb). Subspecies IIIa and IIIb are still identified as "Arizona" by some automated ID systems. The distinguishing biochemical features include the ability to ferment lactose, utilize malonate, liquefying gelatin, and the inability to grow in the presence of KCN.[1–4]

Here, we describe a patient who developed otitis with *S arizonae* after bathing in a lake. This should be the first report of an immunocompetent child who developed otitis complicated with mastoiditis.

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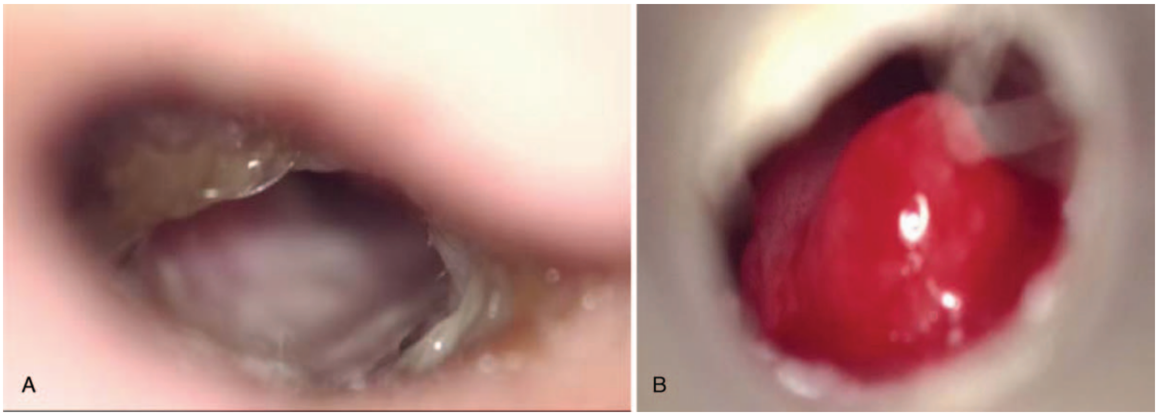

Figure 1. Local ear examination.

2. Case report

A 16-year-old boy presented to our pediatric clinic ("Sf. Maria" Children's Hospital, Iasi, Romania) because of discharge from his right ear. No other associated symptoms (fever, pain, or headache) were present. There were no other pathological clinical signs present at the physical examination. The boy lives in a village, within a working class family, in North East Romania. His history revealed that this is the third episode of otorrhea in the last 6 months. One episode required antibiotic treatment (with Amoksiklav) and the other had a spontaneous remission. Routine blood tests did not reveal any abnormalities. The routine microscopic examination of urine and feces as well as the immunity tests was normal.

An otomicroscopic left ear examination revealed purulent secretion in the external auditory canal with skin inflammation. After the ear suction, a soft polypoid mass appeared through a posterior-superior perforation of the tympanic membrane. Inflammation and polypoid degeneration of the middle ear mucosa, as well as otic erosion, were visible, suggesting a chronic suppurative otitis (Fig. 1). The presence of purulent secretions is suggestive for a bacterial etiology, which needs to be confirmed through bacteriological examination.

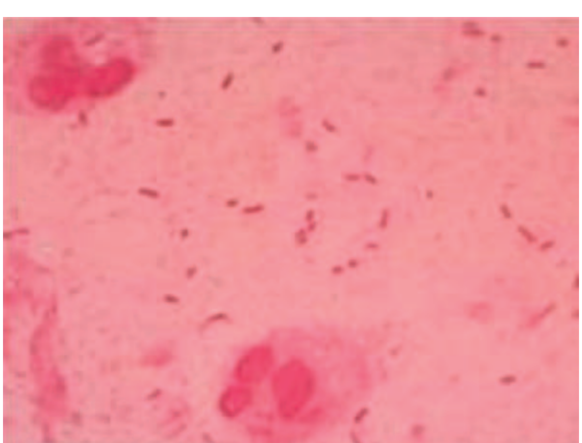

Figure 2. Gram-stain smear of ear secretion showing two polymorphonuclear neutrophils and Gram-negative bacilli *Salmonella arizona* (1000×).

Audiometric testing showed a moderately conductive hearing loss (60 dB on pure-tone average).

Microscopic examination of the direct Gram-stained smear of ear secretion showed inflammation and Gram-negative bacilli. Isolation was done by plating on blood with 5% sheep agar and MacConkey agar, respectively. After overnight incubation on MacConkey agar, the isolate showed lactose-negative colonies that became pink colonies after another 24-hour incubation. The Gram-stain smear of the MacConkey culture showed Gram-negative bacilli (Fig. 2). With the conventional minimal biochemical test, the isolate was identified as *Salmonella* and it showed agglutination with Salmonella polyvalent O antiserum. This was further confirmed by 3 biochemical identifications: the first one, performed with NBP32 (Microscan, Beckman-Coulther) found a Salmonella/Arizona group; the second one performed with MALDI-TOF mass spectrometry identified *Salmonella* spp; the third one, performed with Vitek2 (Bio-Merieux) confirmed *S enterica* subsp. *diarizonae*. The serotyping (performed at National References Laboratory, INC Cantacuzino Bucuresti, Romania) identified the serotype O:38:r:- and lisotyping found "lysed by phage O1," which are unique to the *S enterica* subsp *diarizonae* type (thus excluding other possibilities from the analysis). Therefore, the final taxonomic framing according to the WHO taxonomy[1] confirmed the etiology of otitis in this case, as being attributable to *S enterica* subsp *diarizonae*.

The isolate was subjected to antimicrobial susceptibility testing by disk diffusion method and minimal inhibition concentration, according to the recommendation of the Clinical and Laboratory Standards Institute,[35] and was found sensitive to ampicillin, ciprofloxacin, trimethoprim-sulfamethoxasol, and third-generation cephalosporin.

Following confirmed isolation of this organism from the ear discharge, we came back to the patient's history to trace any possible contact with reptiles. We found out that the boy was bathing in a village lake, where turtles were common inhabitants.

A contrast-enhanced computed tomography scan of the head, following the clinical diagnosis of chronic suppurative otitis, revealed: ethmoidal cells partially filled by a hyperdense material, polypoid thickening of the right frontal, maxilar and sphenoidal sinus mucosa, mastoid cells filled with a solid density material, and trabecular destruction (Fig. 3).

The patient received local and intravenous antibiotics (ceftriaxone 1 g/day) for 10 days, as well as surgical treatment. Surgical management comprised of canal wall up mastoidectomy with

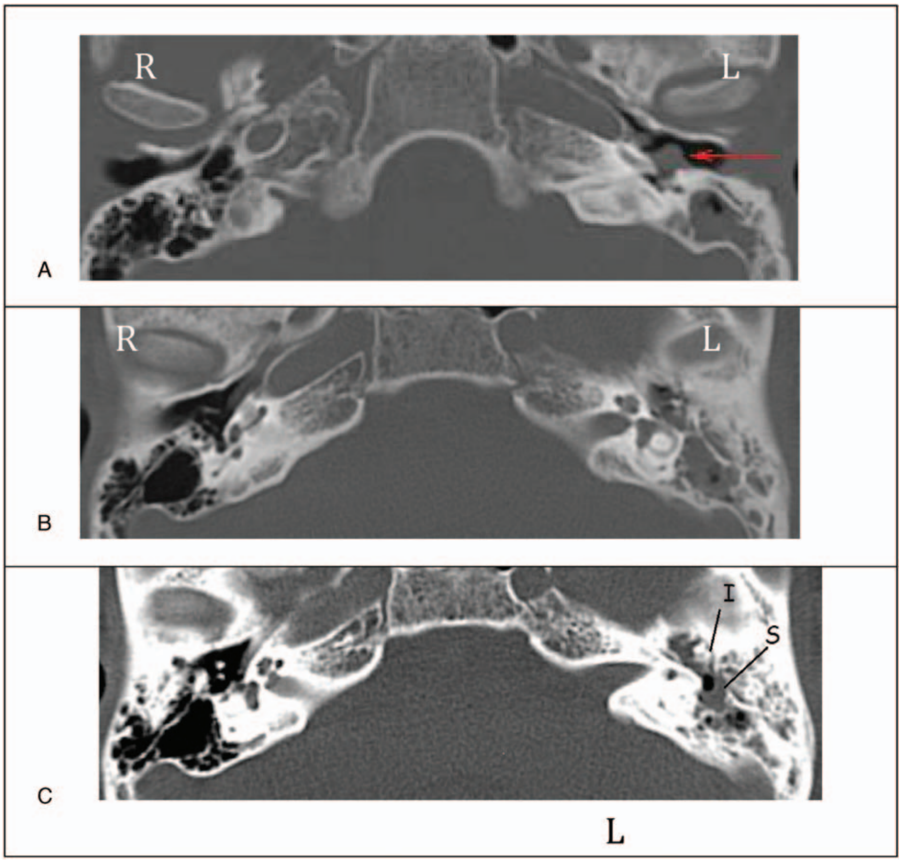

Figure 3. Craniocerebral computed tomography scan.

attico-antrotomy and posterior tympanotomy followed by tympanoplasty. The mastoid cavity and middle ear was occupied by a white pearly mass—cholesteatoma—surrounded by polypous degeneration of the mucosa and puss-like fluid. The short process of the incus, as well as the superior part of the malleus head, appeared eroded. After drilling, the incus with a diamond burr the ossicular chain was reconstructed by incus reposition. The scutum erosion was drilled with diamond burr; the defect was repaired using conchal cartilage, and an underlying tympanoplasty was performed. Daily postoperative dressing care of the incision line, along with antibiotic lavage of the external auditory canal (EAC) packing ensured a favorable evolution.

The bacterial culture from the ear liquid collected during the operation was negative. The clinical recovery was very good. The functional gain was important, with the 1-month postsurgical pure tone audiogram indicating nearly normal hearing (a mean of 25 dB for air conduction thresholds)

3. Discussion

The Arizona group organism was first reported in infected reptiles in 1939 by Caldwell and Ryerson.[5] It was initially considered to be pathogenic only in reptiles, especially in snakes (78.8% of them harboring it),[6] but later it was discovered to be responsible for severe outbreaks in turkeys and sheep.[7] Other animals, including poultry, rats, and dogs, have also been involved in human infection.[8–10]

The first case of human infection by *S arizonae* (presenting with gastroenteritis) was recognized in 1944.[11] The most common form of infection is gastroenteritis[12] with 73% of illnesses occurring in the first 3 months of exposure. It requires an incubation of 2 to 48 hours and is characterized by fever, headache, abdominal pain, vomiting, and diarrhea. Stools are expulsive, copious, and frequently mixed with blood and mucus.[13] There is a substantial risk of dehydration owing to the severity of diarrheal illness. Molecular techniques, such as a Salmonella-specific PCR, may prove a valuable tool for confirming the etiology, allowing timely implementation of appropriate therapy, to prevent morbidity and mortality owing to this rare human pathogen.[9]

Arizona group infection is uncommon among adult patients, having an in-hospital mortality rate of 5.6%[14] and it usually occurs in individuals with underlying disease or immunodeficiency. A carrier state is not uncommon, and a recurrence of S *enterica* subsp *arizonae* sepsis in immunocompromised patients after 1 year has been reported.[13] These infections are often invasive and lead to complications such as meningitis, septicemia, and osteomyelitis.[15] Lee et al reported in 2016[14] the largest case series infected by *S arizonae* (in Taiwan): 18 adults of a total of 485 patients with *Salmonella* species infection, during a 1-year period.

The reported infection in infants occurs as a result of intimate contacts with reptile pets. Little information is available on their distribution in extraintestinal infections, particularly in children. Blood and urine were the most common extraintestinal sites.

Abbot et al retrospectively analyzed salmonellae data (>75,000 isolates) collected by a large state laboratory during 25 years. The Arizona group (subspecies IIIa, IIIb), had isolate totals of 463 (35%) and 443 (33%), respectively. Overall,

Table 1

***Salmonella enterica* subsp *arizonae* human infections with varying clinical pictures.**

Case no	Age	Sex	Underlying disease	Exposure to reptile	Infective syndrome	Author, year
1.	7 y	Girl	Sickle cell anemia	Unknown	Otitis media	Aleksic et al, 1978[17]
2.	2 y	Boy	Langerhans cell histiocytosis	Unknown	Osteomyelitis	Fischer RH, 1953[18]
3.	2.5 y	Girl	Sickle cell anemia	Unknown	Osteomyelitis	Hruby et al, 1973[19]
4.	8 w	Girl	None	Unknown	Meningitis	Ichord et al, 1980[20]
5.	11 y	Boy	None	Pet snake	Osteomyelitis	Croop et al 1984[13]
6.	15 y	Girl	SLE	Rattlesnake capsule	Septicemia, sinusitis	Kraus et al, 1991[21]
7.	12 y	Girl	Lymphocytic leukemia	Rattlesnake pills	sepsis	Kraus et al, 1991[21]
8.	6 y	Boy	None	Rattlesnake meat	gastroenteritis	Kelly et al, 1995[22]
10.	8 w	Boy	Netherton syndrome	Pet snake	gastroenteritis	Sanyal et al, 1997[31]
11.	7 mo	Girl	None	Pet Iguana	Osteomyelitis	Nowinski et al, 2000[24]
12.	10 y	Boy	None	Snake bite	Wound infection	Bello et al, 2001[25]
13.	3 mo	Girl	Microcephaly	Pet reptiles	Gastroenteritis	Mahajan et al, 2003[7]
14.	1 y	Boy	Sickle cell anemia	Unknown	Osteomyelitis	Ogden and Light, 1979[26]
15.	2 y	Boy	Sickle cell anemia	Unknown	Osteomyelitis	Ogden and Light, 1979[26]
16.	14 y	Boy	None	Unknown	Ankle arthritis, Gastroenteritis	Foster and Kerr, 2005[27]
17.	10 mo	Boy	None	Unknown	Hip osteoarthritis	Schneider et al, 2009[28]
18.	13 days	Girl	None	Denied	Meningitis	Lakew et al, 2013[29]
19.	32 days	Girl	None	Unknown	Meningitis	Tuan et al, 2015[30]
20.	16 y	Boy	None	Turtles	Otitis, mastoiditis	This study

patients were more likely to have had subspecies II–IV isolated from feces (n = 947) than from an extraintestinal site (n = 395; $P < .001$). Only patients from whom subspecies IIIa were isolated were equally likely to have an extraintestinal infection as opposed to diarrhea only. Sources of extraintestinal infections included cerebrospinal fluid, blood, urine, cervix, bile, wounds and abscesses, and the respiratory tract.[16]

Even if infection occurs more often than in adults, there are not many case reports of *S arizonae* infection in children either: to the best of our knowledge only 20 pediatric patients, below 18 years of age, from 1952 until now[7,13,16–31] (Table 1) have been reported. These patients have generally been older children, some of them having an underlying severe disease such as leukemia, histiocytosis, SLE, Netherton syndrome, or sickle cell disease. Other epidemiologic infectious disease studies have included children with *S arizonae* among other bacterial infections: Kolo et al[32] have found 9 pediatric cases with bacteremia with *S arizonae* (5 of them being infected with HIV), of 102 bateriemic patients during a 6-month period in Nigeria. Meyer Sauteur[33] identified 9 cases of *S arizonae* among 182 children with reptile-associated salmonellosis, most of them (177 children) being previously healthy children.

Abbots' data[16] showed an equivalent number of cases (Salmonella subspecies II-IV) in children <1 year of age and in persons 11 to 60 years of age (from isolates obtained from fecal samples), but the prevalence of extraintestinal infections occurred in the older patient groups. These patients, as owners or handlers of exotic pets, may be exposed to a greater inoculum, whereas children <1 year primarily acquire infections secondarily from fomites or surfaces, such as sinks used previously to bathe reptiles or by transmission from handler to child.[34]

There were no reported human *S enterica* subsp *arizonae and diarizonae* infection in Romania. The cases that have been reported by Köbölkuti et al[36] were only in Viper snakes (both captive and free living).

4. Conclusions

S enterica serotype *Arizonae* is a rare cause of human infection, being a common organism in reptiles, like snakes and turtles. It can cause not only gastroenteritis but also osteomyelitis, otitis, mastoiditis, meningitis, osteoarthritis, or septicemia. Young children are at a particular risk of acquiring such infections. Therefore, proper history should be obtained in such cases. These results might encourage further epidemiologic investigations into these infections. In order to use this information to generate a more effective strategy that public health agencies and the exotic pet industry can implement to reduce the extent of disease caused by these organisms.

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References

[1] Grimont PAD, Weill FX. Antigenic Formulae of the Salmonella Serovars. 9th ed.2007;WHO Collaborating Centre for Reference and Research on Salmonella,
[2] Tindall BJ, Grimont PA, Garrity GM, et al. Nomenclature and taxonomy of the genus Salmonella. Int J Syst Evol Microbiol 2005;55:521–4.
[3] Le Minor L, Genus III. Krieg NR, Holt JG. Salmonella Lignieres 1900. Bergey's Manual of Systematic Bacteriology, vol 1 Williams & Wilkins Co, Baltimore:1984;427.
[4] Patricia Fields. National Salmonella Reference Laboratory CDC. 10th Annual PulseNet Update Meeting April 5, 2006.
[5] Caldwell ME, Ryerson DL. Salmonellosis in certain reptiles. J Infect Dis 1939;65:242–5.
[6] Di Bella S, Capone A, Bordi E, et al. Salmonella enterica ssp. arizonae infection in a 43-year-old Italian man with hypoglobulinemia: a case report and review of the literature. J Med Case Rep 2011;5:323.
[7] Mahajan RK, Khan SA, Chandel DS, et al. Fatal Case of Salmonella enterica subsp. arizonae Gastroenteritis in an Infant with Microcephaly. J Clin Microbiol 2003;41:5830–2.
[8] Kolker S, Itsekzon T, Yinnon AM, et al. Osteomyelitis due to Salmonella enterica subsp. arizonae: the price of exotic pets. Clin Microbiol Infect 2012;18:167–70.
[9] Lee Y-C, Hung M-C, Hung S-C, et al. Salmonella enterica subspecies arizonae infection of adult patients in Southern Taiwan: a case series in a non-endemic area and literature review. BMC Infect Dis 2016;16:746.

[10] Hoag JB, Sessler CN. A comprehensive review of disseminated Salmonella arizona infection with an illustrative case presentation. South Med J 2005;98:1123–9.
[11] Seligmann E, Saphra L, Wassermann M. Occurrence of some unusual Salmonella types in man including a new type, Salmonella georgia. Am J Hyg 1944;40:227–31.
[12] Schutzman SA, Petrycki S, Fleisher GR. Bacteremia with otitis media. Pediatrics 1991;87:48–53.
[13] Croop JM, Shapiro B, Alpert G, et al. Arizona Hinshawi osteomyelitis associated with a pet snake, Pediatr. Infect Dis 1984;3:188.
[14] Cohen JI, Bartlett JA, Corey GR. Extra-intestinal manifestations of salmonella infections. Medicine 1987;66:349–88.
[15] Makin G, Abu Harb M, Finn A, et al. Letter. Lancet 1996;348:200.
[16] Abbot SL, Ni FCY, Janda M. Increase in extraintestinal infections caused by Salmonella enterica subspecies II-IV. Emerg Infect Dis 2012;18:637–40.
[17] Aleksic S, Rohde R, Aleksic V, et al. A new fimbrial antigen as a cause for a complete O-inagglutinability of various Arizona strains. Zentbl Bakteriol 1978;241:427–37.
[18] Fischer RH. Multiple lesions of bone in letterer sieve disease: report of a case with culture of paracolon Arizona bacilli from bone lesion and blood followed by response to therapy. J Bone Joint Surg 1953;35:445–65.
[19] Hruby MA, Honig GR, Lolekha S, et al. Arizona hinshawii osteomyelitis in sickle cell anaemia. Am J Dis Child 1973;125:867–8.
[20] Ichord R, Brook I, Contrani G. Arizona hinshawii bacteraemia and meningitis in a child: a case report. J Clin Pathol 1980;33:848–50.
[21] Kraus A, Guerra BG, Alarcon SD. Salmonella arizona arthritis and septicemia associated with rattlesnake ingestion by patients with connective tissue diseases. A dangerous complication of folk medicine. J Rheumatol 1991;18:1328–31.
[22] Kelly J, Hopkin R, Rimsza ME. Rattlesnake meat ingestion and Salmonella arizona infection in children: case report and review of the literature. Pediatr Infect Dis J 1995;14:320–2.
[23] Mermin J, Hoar D, Angulo FJ. Iguanas and Salmonella marina infection in children: a reflection of increasing incidence of reptile-associated salmonellosis in the United States. Pediatrics 1997;99:399–402.
[24] Nowinski RJ, Albert MC. Salmonella osteomyelitis secondary to iguana exposure. Clin Orthrop Relat Res 2000;372:250–3.
[25] Bello CSS, Singh S, Al Waley A, et al. Salmonella arizonae infection from snake bite. Ann Saudi Med 2001;21:352–4.
[26] Ogden JA, Light TR. Pediatric osteomyelitis: II. Arizona hinshawii osteomyelitis. Clin Orthop Relat Res 1979;139:110–3.
[27] Foster N, Kerr K. The snake in the grass—Salmonella arizonae gastroenteritis in a reptile handler. Acta Pediatr 2005;94:1165–6.
[28] Schneider L, Ehlinger M, Stanchina C, et al. Salmonella enterica subsp. arizonae bone and joints sepsis. A case report and literature review. Orthop Traumatol Surg Res 2009;95:237–42.
[29] Lakew W, Girma A, Triche E. Salmonella enterica serotype arizonae meningitis in a neonate. Case Rep Pediatr 2013;2013:813495, 3 pages.
[30] Tuan ĐQ, Hung PH, Mai PX, et al. Salmonella meningitis: a report from National Hue Central Hospital, Vietnam. Jpn J Infect Dis 2015;68: 30–2.
[31] Sanyal D, Douglas T, Roberts R. Salmonella infection acquired from reptilian pets. Arch Dis Child 1997;77:345–6.
[32] Kolo OO, Galadima M, Daniyan SY, Saidu##TB, et al. Bacteremia in children infected with HIV/AIDS in Minna, Niger State, Nigeria. Br Microbiol Res J 2015;9:1–7.
[33] Meyer Sauteur PM1, Relly C, Hug M, et al. Risk factors for invasive reptile-associated salmonellosis in children. Vector Borne Zoonotic Dis 2013;13:419–21.
[34] Bertrand S, Rimhanen-Finne R, Weill FX, et al. Salmonella infections associated with reptiles: the current situation in Europe. Euro Surveill 2008;13: pii: 18902.
[35] Clinical and laboratory standards institute, Performance Standards for Antimicrobial Susceptibility Testing: twentieth Informational Supplement, M100S, 2017.
[36] Köbölkuti L, Spinu M, Cadar D, Czirjak G, Kiss T., Classical And Molecular Monitoring Of The Prevalence Of Salmonella Spp. Carriage In Free- Living And Captive Native Vipera Snakes In Romania, Lucrări Stiinłifice Medicină Veterinară Vol. Xlii (1), 2009, Timişoara, 294.