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INSTITUTE FOR INTERNATIONAL COOPERATION IN ANIMAL BIOLOGICS

IOWA STATE UNIVERSITY College of Veterinary Medicine



OIE Collaborating Centre for

- Diagnosis of Animal Disease and Vaccine Evaluation in the Americas
 Day-One Veterinary Competencies
- and Continuing Education



Etiology

Salmonella spp. are members of the family Enterobacteriaceae. They are Gram negative, facultatively anaerobic rods. *Salmonella* species are classified into serovars (serotypes) based on the lipopolysaccharide (O), flagellar protein (H), and sometimes the capsular (Vi) antigens. There are more than 2500 known serovars. Within a serovar, there may be strains that differ in virulence.

A number of *Salmonella* serotypes have been found associated with reptiles and/or amphibians including, the *S. enterica* subsp. *enterica* serovars Chameleon, Java, Marina, Poona, Stanley and Typhimurium, among others. *S. bongori, S. enterica* subsp. *salamae, S. enterica* subsp. *arizonae, S. enterica* subsp. *diarizonae, S. enterica* subsp. *houtenae, S. enterica* subsp. *indica,* are usually found in poikilotherms (including reptiles, amphibians and fish) and in the environment. Some of these organisms are occasionally associated with human disease.

Geographic Distribution

Reptile-associated salmonellosis occurs worldwide.

Transmission

Salmonella spp. are mainly transmitted by the fecal-oral route. They are carried asymptomatically in the intestines of infected reptiles and are continuously or intermittently shed in the feces. *Salmonella* is most commonly transferred between reptiles by contact with contaminated feces of other reptile or contaminated food, water or soil. Transmission may also occur in utero, perinatally, or by ingestion of contaminated prey. Transovarian passage has also been reported.

People are often infected by ingesting organisms in animal feces. Direct transmission occurs through handling of a reptile and indirect transmission occurs by contact with an object contaminated by a reptile, its feces or contaminated food or water. Clothing in contact with reptiles has served as a source of transmission, as have claw scratches and bites. Contact with feeder rodents, intended as reptile and amphibian food, has also been linked to cases of salmonellosis in humans. Less commonly, cases have been reported after the ingestion of sundried, uncooked rattlesnake meat and blood transfusions.

Salmonella spp. can survive for long periods in the environment, particularly where it is wet and warm, and can be isolated for prolonged periods from surfaces contaminated by reptile feces. *Salmonella* has been reported to survive 89 days in tap water, 115 days in pond water, within dried reptile feces from cages 6 months after removal of the reptile and from aquarium water 6 weeks after removal of a turtle.

Disinfection

Salmonella spp. are susceptible to many disinfectants including 1% sodium hypochlorite, 70% ethanol, 2% glutaraldehyde, iodine-based disinfectants, phenolics and formaldehyde. They can also be killed by moist heat (121°C for a minimum of 15 min) or dry heat (160-170°C for at least 1 hour).

Infections in Humans

Incubation Period

The incubation period for *Salmonella* gastroenteritis in humans is from 6 to 73 hours. Enteric fever usually appears after 10 to 14 days. Symptoms can last from 24 hours to 12 days.

Clinical Signs

In humans, salmonellosis varies from a self-limiting gastroenteritis to invasive infections that can lead to septicemia. Whether the organism remains in the intestine or disseminates depends on host factors as well as the virulence of the strain. Asymptomatic infections can also be seen.

Salmonellosis acquired from reptiles is often severe, and may be fatal due to septicemia or meningitis. Most cases of reptile-associated salmonellosis are seen in children under 10 and people who are immunocompromised. Infants, especially those that are bottle fed, are highly susceptible.

Gastroenteritis is characterized by nausea, vomiting, cramping abdominal pain and diarrhea, which may be bloody. Headache, fever, chills and myalgia may also be seen. Severe dehydration can occur in infants and the elderly. In many cases, the symptoms resolve spontaneously in 1 to 7 days. Deaths are rare except in very young, very old, debilitated or immunocompromised persons.

Enteric fevers are a severe form of systemic salmonellosis. Although most cases are caused by *S. typhi*, a human pathogen, other species can also cause this syndrome. Gastrointestinal disease may be the first sign, but it usually resolves before the systemic signs appear. The symptoms of enteric fever are non-specific and may include fever, anorexia, headache, lethargy, myalgia and constipation. This disease can be fatal, due to meningitis or septicemia, if not treated quickly.

Focal infections such as septic arthritis, abscesses, endocarditis or pneumonia are occasionally seen. Many tissues and organs can be affected.

Reiter's syndrome may be a sequela in some cases of gastroenteritis. This syndrome is characterized by mild to severe arthritis, nonbacterial urethritis or cervicitis, *conjunctivitis* and small, painless, superficial mucocutaneous ulcers. Reiter's syndrome occurs in approximately 2% of cases of salmonellosis. It is also seen after other enteric infections. Reiter's syndrome usually resolves in 3 to 4 months, but approximately half of all patients experience transient relapses for several years. Chronic arthritis can occur in some cases.

Communicability

Salmonellosis can be transmitted to other people or to animals in the feces. Humans shed bacteria throughout the course of the infection. Shedding can last for several days to several weeks, and people may become temporary carriers for several months or longer. Approximately 0.3-0.6% of patients with non-typhoidal *Salmonella* infections shed the bacteria in the feces for more than a year. Antibiotic treatment can prolong shedding. Over 90% of reptiles may carry *Salmonella*, and up to five different serotypes have been isolated from a single reptile.

Diagnostic Tests

Salmonellosis can be confirmed by isolating the organisms from feces or, in cases of disseminated disease, from the blood. *Salmonella* will grow on a wide variety of selective and non-selective media including blood, MacConkey, eosin-methylene blue, bismuth sulfite, Salmonella-Shigella, and brilliant green agars. Enrichment broths can increase the probability of isolating the organism. Intensive methods to detect *Salmonella* (pre-enrichment) are

primarily designed for food analysis but are sometimes used clinically, to resuscitate stressed organisms and increase the probability that small numbers of organisms will be detected.

Salmonella spp. are identified with biochemical tests, and the serovar can be identified using serology for the somatic (O), flagellar (H) and capsular (Vi) antigens. Phage typing or plasmid profiling is also used for some serovars. Further characterization, if needed, can be carried out at a reference laboratory. PCR and other genetic techniques may also be available.

Treatment

Salmonellosis in humans can be treated with a number of antibiotics including ampicillin, amoxicillin, gentamicin, trimethoprim/sulfamethoxazole and fluoroquinolones. Many isolates are resistant to one or more antibiotics, and the choice of drugs should, if possible, be based on susceptibility testing. Antibiotics are used mainly for septicemia, enteric fever or focal extraintestinal infections. Focal infections may require surgery and prolonged courses of antibiotics.

In the elderly, infants and immunosuppressed persons, who are prone to septicemia and complications, antibiotics may be given for gastroenteritis. However most healthy people recover spontaneously in 2 to 7 days and may not require antibiotic treatment. Antibiotics do not usually shorten this form of the disease. They also prolong the period of bacterial shedding and increase the development of antibiotic-resistant strains. Symptomatic treatment of dehydration, nausea and vomiting may be required.

Prevention

To prevent reptile-associated *Salmonella* infections, the Centers for Disease Control and Prevention (CDC) have issued recommendations for the handling of reptiles. Some states have legislation requiring pet store owners to communicate the increased risks of salmonellosis to customers purchasing reptiles.

Standard recommendations include:

- Frequent and thorough hand washing with soap and water after handling reptiles or reptile cages.
- Following contact with reptiles, clothing should be changed, especially before close contact with infants.
- Eating, drinking or smoking while handling reptiles or their environments should be avoided.
- People who are at increased risk for infection or serious complications of salmonellosis (e.g., children < 5 years of age and immunocompromised persons) should avoid contact with reptiles.
- Reptiles should not be kept in childcare centers.
- Reptiles should be kept out of households with children younger than 1 year of age and persons with weak immune systems.

- Reptiles should not be allowed to roam freely throughout the home or living area.
- Reptiles and their equipment should be kept out of kitchens and other food-preparation areas.
- Kitchen sinks should not be used to bathe reptiles or to wash their dishes, cages or aquariums. If bathtubs are used for these purposes, they should be thoroughly cleaned and disinfected afterwards. It is best to use a dedicated plastic tub to bathe or swim reptiles.
- Reptiles should not be kissed
- Do not share food or drink with reptiles.

No human vaccines to prevent zoonotic or foodborne salmonellosis exist. A vaccine is available to prevent typhoid fever, an infection transmitted from person to person.

Morbidity and Mortality

Salmonellosis is common in humans, and the incidence of disease seems to be increasing in the U.S. Approximately seven percent (93,000 cases) of salmonellosis reported annually to the CDC are thought to be caused by reptiles. Many cases are unreported, so the actual incidence is thought to be higher. Salmonellosis can affect all ages, but the incidence and severity of disease is higher in young children, the elderly, and people who are immunocompromised or have debilitating diseases. Children under 10 and immunocompromised persons seem to have an increased risk of contracting severe disease from reptiles.

Approximately 500-600 fatal cases of salmonellosis are reported each year in the U.S. The overall mortality rate for most forms of salmonellosis is less than 1%; however, some serovars or syndromes are more likely to be fatal. During outbreaks, approximately 10% of all cases and 18% of cases in the elderly result in invasive disease. *Salmonella* gastroenteritis is rarely fatal in healthy people.

During the 1970s, small pet turtles were a major source of Salmonella infections in the United States, leading to an estimated 280,000 reported cases of salmonellosis each year. In 1975, the Food and Drug Administration (FDA) banned all interstate shipments of pet turtles with a carapace length less than 4 inches. This prevented almost 100,000 cases of salmonellosis among children 1 to 9 years of age each year. However, more recently, the popularity of reptiles as pets has led to an increase in the number of reptile-associated cases. During 1991-2001, the estimated number of households with reptiles doubled from approximately 850,000 to 1.7 million; an estimated 3% of households in the United States have a reptile. Additionally, regulations to prevent the transmission of zoonotic diseases in childcare centers vary from state to state: only 50% of states have regulations requiring children and staff to wash their hands after handling animals, and less than 25% of states have regulations expressly banning reptiles from childcare centers.

Infections in Animals

Species Affected

Salmonella spp. have been found in all species of reptiles that have been investigated. Infection has been documented in turtles, tortoises, snakes and lizards (including chameleons and iguanas).

Incubation Period

The incubation period in animals is highly variable. In many cases, infections become symptomatic only when the animal is stressed.

Clinical Signs

Clinical disease in reptiles seems to be uncommon. Salmonella spp. are often carried asymptomatically. Clinical disease may appear when animals are stressed by factors such as transportation, crowding, food deprivation, exposure to cold, a concurrent viral or parasitic disease, sudden change of feed, or overfeeding following a fast. In some cases, oral antibiotics may also precipitate disease.

Syndromes that have been reported include septicemia (characterized by anorexia, listlessness and death), osteomyelitis, osteoarthritis, pneumonia, gastroenteritis and cutaneous/subcutaneous abscesses. Progressive, fatal bone infections have been seen in snakes. In one group of free-living turtles, the symptoms included emaciation, lesions of the plastron, a discolored carapace and intestinal, respiratory and hepatic lesions. *Salmonella* spp. have also been implicated in sporadic deaths among tortoises in zoos.

Post-Mortem Lesions

The necropsy lesions, are not pathognomonic.

Fibrinonecrotizing gastroenteritis may be present, and may involve the formation of diptheritic membranes in the lumen of the intestine. Fibrinonecrotizing plaques may be present in the oral cavity, and pneumonia and/or liver lesions may be present.

Communicability

Salmonella spp. are shed in the feces of both symptomatic and asymptomatic animals. Reptiles shed the organism continuously or intermittently, and should always be considered a potential source of *Salmonella*.

Diagnostic Tests

Salmonellosis can be confirmed by isolating the organisms from feces or, in cases of disseminated disease, from the blood. At necropsy, heart blood, bile, liver, spleen and mesenteric lymph nodes are collected. Intensive methods (pre-enrichment) to detect *Salmonella* are designed for food analysis but are sometimes used clinically. They can resuscitate stressed organisms and increase the probability that small numbers of organisms will be detected. Pre-enrichment, enrichment and selection of several colonies

may be particularly useful for reptiles, which can carry several species of *Salmonella* simultaneously.

Salmonella spp. are identified with biochemical tests, and the serovar can be identified by serology for the somatic (O), flagellar (H) and capsular (Vi) antigens. Phage typing or plasmid profiling is also used for some serovars. Further characterization, if needed, can be carried out at a reference laboratory.

Diagnosis of clinical cases and identification of carriers are complicated by the following factors: Because *Salmonella* spp. can be found in healthy carriers, isolation of these bacteria from the feces is not a definitive diagnosis of salmonellosis. Reptiles may shed *Salmonella* spp. intermittently. Currently, it is impossible to determine whether an individual reptile is *Salmonella*-free.

Most serologic tests detect a limited number of serovars or serogroups. Serology is of limited use in individual animals, as antibodies do not appear until two weeks after infection, and antibodies may also be present in uninfected animals. Polymerase chain reaction (PCR) and other genetic techniques may also be available.

Treatment

The Association of Reptile and Amphibian Veterinarians (ARAV) discourages veterinarians from treating reptiles with antibiotics to eliminate *Salmonella*. Attempts to treat reptiles with antibiotics to eliminate *Salmonella* carriage or to raise *Salmonella*-free reptiles have been unsuccessful. Antibiotics can favor the persistence of *Salmonella* spp. in the intestines after recovery, affect the intestinal flora, and increase the emergence of antibiotic-resistant strains.

In that both healthy and clinically affected animals may carry *Salmonella*, the isolation of this organism from lesions is difficult to interpret. Many isolates are resistant to one or more antibiotics, and the choice of drugs should, if possible, be based on susceptibility testing. Supportive care in affected animals can be initiated, including fluid replacement, temperature and humidity regulation, and nutritional support.

Prevention

All reptiles should be considered to be potential sources of *Salmonella*. In many cases, elimination of *Salmonella* infections is impractical, and control is limited to preventing clinical disease and/or the transmission of bacteria to humans. Clinical salmonellosis can be decreased by good hygiene and minimizing stressful events.

Morbidity and Mortality

In animals, asymptomatic *Salmonella* infections are common. Estimates of the carrier rate among reptiles vary from 36% to more than 80-90%, and several serovars can be found in a single animal. Some authorities consider most or all reptiles to be *Salmonella* carriers. Deaths or disease are occasionally reported in reptiles, but seem to be rare.

Internet Resources

Association of Reptile and Avian Veterinarians (ARAV) <u>http://www.arav.org</u>

Centers for Disease Control and Prevention (CDC) <u>http://www.cdc.gov/salmonella/</u>

CDC Special Advice for People at Extra Risk for Zoonoses http://www.cdc.gov/healthypets/extra_risk.htm

Food and Drug Administration: Pet Turtles Cute But Contaminated With Salmonella <u>http://www.fda.gov/ForConsumers/ConsumerUpdates/ucm</u> 048151.htm

Pathogen Safety Data Sheets and Risk Assessment– Public Health Agency of Canada http://www.phac-aspc.gc.ca/lab-bio/res/psds-ftss/indexeng.php#s

Medical Microbiology http://www.ncbi.nlm.nih.gov/books/NBK7627/

The Merck Manual <u>http://www.merck.com/pubs/mmanual/</u>

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