TREATMENT AND CONTROL OF AN OUTBREAK OF SALMONELLOSIS IN HATCHLING NILE CROCODILES *(CROCODYLUS NILOTICUS)*

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ABSTRACT

The therapeutic and managemental steps taken to bring a severe outbreak of salmonellosis in Nile crocodiles *(Crocodylus niloticus)* under control are described. All the crocodiles were initially given intramuscular injections with kanamycin on alternating days for 8 d, coupled with adjustment of the ambient temperature to 29°C. The holding pens were cleaned and disinfected with 2% formalin at the onset of treatment. Daily scrubbing and disinfection was continued throughout the treatment period. Severely affected crocodiles were separated and force-fed a liquid diet. All crocodiles were vaccinated with an inactivated calf paratyphoid vaccine 10 d after the onset of treatment and again one month later. The initial treatment was followed by a 30-week period of in-feed medication with oxytetracycline. Response to the initial treatment was dramatic, although some mortalities still occurred in the force-fed group for one month. The following year's hatchlings were fed heat-treated meat from first feeding onwards to avoid the possibility of introducing *Salmonella* spp. via the feed.

Key words: Crocodiles, *Crocodylus niloticus, Salmonella* spp., control, disinfection, temperature

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INTRODUCTION

Commercial raising of Nile crocodiles *(Crocodylus niloticus)* for the production of leather is an accepted farming practice in South Africa. Problems and management associated with this type of farming are unique and quite different from those encountered with captive crocodilians kept for display purposes.

Being poikilothermic, crocodiles have developed elaborate ways of behavioural thermoregulation centered around the optimal use of energy obtained from food[4]. Free-ranging crocodiles regulate body temperature by temperature selection within thermal gradients in the environment. As with many other poikilotherms, the rate of metabolism and hence growth increases within limits with increasing temperature, provided adequate food is present. Non-feeding crocodilians will seek out cooler temperatures within their environment in order to conserve energy until they feed again. Should a crocodile not be able to avoid high temperatures when not feeding, the situation would be expected to elicit a stress response. Maximum growth is obtained by keeping the crocodiles at constantly high temperatures with daily feeding. The complex behavioural response of thermal selection within an environment of varying temperatures is thus withheld from these crocodiles.

Although it has been demonstrated that alligators show a behavioural fever response towards an infection[4], the immune-defence response appears to increase with temperatures up to a maximum above which it declines. In alligators the leucocyte response to infection reaches a maximum at a body temperature of 30°C. At 35°C body temperature the immune-defence response was rendered ineffective (Glassman & Bennet 1978, quoted by Lang[4]). It therefore appears that the behavioural fever response to infection is limited within the available temperature gradient and once a disease has taken a chronic course with loss of appetite, the animal's response is further complicated by its need to conserve its energy reserves and thus possibly seeking out cooler temperatures.

There have been relatively few documented reports of salmonellosis in farmed crocodiles. The disease appears to be common, with serious economic implications. Foggin[2] reported 70/108 bacterial isolations made from post mortem cases of farmed crocodiles in Zimbabwe to have been *Salmonella* spp.. Friedland[3] also isolated *Salmonella* spp. from septicaemic crocodiles held in intensive rearing units in Lebowa. Contaminated feed, poor hygiene in feed preparation and feeding, stagnant water, and overcrowding resulting in high bacterial concentrations are all regarded as predisposing factors for salmonellosis[1].

Ongoing mortality occurred in a batch of almost 2 000 crocodiles, which hatched on a farm in the Eastern Transvaal in December 1988, soon after they had been put into a closed environment house and had started feeding. Feed consisted mainly of minced meat obtained from feedlot cattle which had died from various causes. According to the farmer, the carcasses had been handled under poor hygienic conditions before arriving at the crocodile farm. Bone meal, carcass meal and a vitamin and mineral premix were added to the minced meat. Bacterial cultures taken at autopsy from a number of these crocodiles, revealed the involvement of several *Salmonella* spp., including *Salmonella typhimurium*. It was suspected that the original source of infection may have been the feedlot carcasses. The farmer treated ill crocodiles with sulphamonomethoxine (Daimeton B, Centaur) and later with gentamycin (Genta 50, Phenix) but with little success. Subsequently, enrofloxacin (Baytril, Bayer) and then sulpha-chloropyrazine (ESB3, Ciba Geigy) were used on the sick crocodiles, both with poor results. All these antibiotic treatments were given at

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far below recognised therapeutic levels and were applied for an inadequate period. In April 1989 an untyped *Salmonella* sp. was again isolated from the sick crocodiles. This isolate had been found to be resistant to the most commonly used antibiotics. An antibiogram however, indicated good sensitivity towards kanamycin, which was subsequently used but was found to reduce mortalities only temporarily. At this stage, the farmer started chlorinating incoming water at 5 ppm chlorine.

Up to 15 crocodiles were dying per day and the appetite of the whole group was severely suppressed despite a constant ambient temperature of 35°C. At this stage the author was consulted. Post mortem examination on 10 crocodiles indicated that the majority of crocodiles probably died of salmonellosis. The purpose of this paper is to document the treatment and management regimens by which the outbreak was brought under control.

MATERIALS AND METHODS

Treatment

Treatment with kanamycin (Kanamyn, Phenix) was continued at 20 mg kg^{-1} body mass by intramuscular injection repeated every 48 h for 8 d. All 1 500 crocodiles still alive, were treated, whether they appeared ill or not.

The weakest crocodiles were sorted into a sick bay where they were force-fed from a syringe on alternating days. A liquid diet consisting of egg, liver and a vitamin and mineral premix was given. The remaining crocodiles were fed as usual during the treatment period. Once crocodiles in the force-fed group appeared to regain strength, they were released back into the pens.

An inactivated calf paratyphoid vaccine (V.R.I. Onderstepoort) was administered by intramuscular injection to all the crocodiles at a dose of 0,2 ml per crocodile 10 d after the onset of treatment. Vaccination was repeated one month later.

Two days after the last kanamycin injection, the crocodiles were put onto a 3-week course of medicated feed containing oxytetracycline at a dose of 75 mg kg^{-1} body mass. This was mixed with the carcass meal, bone meal and vitamin and mineral premix before being added to the minced meat just prior to feeding. At this stage the crocodiles were being fed daily.

Management

Close examination revealed a sticky layer of fat covering the cement surfaces of the holding pens. This feed residue remained, despite daily scrubbing, as no fat solvent was used. After moving the crocodiles, the pens were scrubbed down with 0,1% Teepol (Teepol Orange Concentrated Detergent, Cera Oil S.A.). All the extractor fans and doors were opened and the pens were then scrubbed down with a 2% formalin solution followed by thorough rinsing before the crocodiles were returned. Thereafter the pens were scrubbed daily with Teepol, followed by the addition of 100 ppm iodophore (Adcodyne, Adcock Ingram) to the pen water for a period of one hour. Scrubbing utensils were stored in 200 ppm iodophore. An iodophore footbath was also installed at the entrance to the crocodile house.

The normal ambient temperature of the house was thermostatically controlled and kept constant at 35°C. At the onset of treatment, the thermostat setting was reduced to 29°C. The lower temperature was maintained throughout the initial 8 d treatment period. Culture of one of the *Salmonella* isolates at various temperatures had indicated a distinct drop in growth vigour between temperatures of 29°C and 25°C.

As the most likely source of infection appears to have been the feed, the following year's hatchlings (1989) were fed on heat-treated meat from first feeding onwards. Minced meat was heated to 80°C for a period of 10 min and then allowed to cool slowly. Once cool, the carcass and bone meal and the vitamin and mineral premix were added. The feed was then once again put through a clean mincer which produced a fairly dry pellet.

RESULTS

Response to the initial treatment with kanamycin and the lowered temperature was good, with mortalities dropping to fewer than one per day by the end of the initial 8 d. Mortalities mainly occurred in the group being force-fed. During the 3 weeks that the crocodiles were on in-feed oxytetracycline treatment, mortalities continued to decrease and feed consumption increased dramatically. At no stage was there any indication of an adverse response to either kanamycin or oxytetracycline. The vaccination was also well tolerated. Approximately 50% of force-fed crocodiles recovered and were returned to the pens with the healthier crocodiles. The remainder did not respond and took up to one month to die. Three weeks after the start of the initial treatment, several dead crocodiles were presented for post mortem examination. Severe peritonitis, necrotic enteritis and focal liver necrosis were the outstanding lesions. These crocodiles were severely emaciated and gave the impression of having been chronically ill. A month after the initial treatment, mortalities had become rare and the crocodiles were all eating well and appeared healthy.

The body temperature of the crocodiles was generally found to be approximately 2,5°C lower than the ambient temperature. The reduced ambient temperature therefore resulted in crocodile body temperatures of about 26,5°C.

The hygiene measures were well tolerated by the crocodiles, neither scrubbing nor disinfection having any noticeable adverse effects.

The following year's hatchlings readily accepted heat-treated meat and refused to take raw meat when given the choice 2 weeks later. Growth appeared to be good and by 3 months post hatching, the crocodiles were exceptionally healthy.

DISCUSSION

The pathology caused by salmonellosis in crocodiles is severe and may result in acute mortality. However in many cases the course of the disease may be protracted, affected animals dying from chronic manifestations, such as intestinal occlusion, up to 3 weeks after being identified as ill. This was particularly evident amongst the force-fed crocodiles. The disease may therefore be associated with a protracted period during which the diseased crocodiles also refuse to eat. Nevertheless, in this case the crocodiles had still been kept at the usual constant 35°C of their controlled environment. Environmental temperature was reduced to a level that was more conducive to the effective funtioning of the crocodiles' immune system. It also reduced the stress of a forced high metabolic rate in a non-feeding crocodile.

It appeared that the probable sources of infection were feedlot carcasses infected with *Salmonella* spp.. One would expect a mammalian pathogen to have a temperature preference close to that of its host. The lowering of the ambient temperature in the controlled environment house would have also adversely influenced bacterial growth.

The fairly long course of antibiotic therapy with kanamycin did not appear to impair the renal function of the crocodiles. A prolonged course of oxytetracycline-medicated feed was given to exclude complications by other opportunistic bacteria during the recovery period in the already weakened crocodiles.

High humidity and high temperatures make controlled environment rearing units ideal incubators for bacteria. Although the pens appeared spotless, the sticky fat layer on most of the surfaces of the pens may have provided a rich source of bacterial contamination as well as protection against disinfectants. Although iodophore disinfection after removal of the fat layer is probably not necessary during the normal operation of a closed environment unit, it provides a useful way of effectively disinfecting pens during a disease outbreak.

Once diseased crocodiles show advanced clinical symptoms of salmonellosis (sluggishness and eventually total inabili-

ty to move), the disease has progressed so far that the above measures alone are usually insufficient to ensure their recovery. The high individual value of these animals makes it worthwhile to force-feed the animals in order to supply fluids and to regain a positive energy balance. A survival rate of approximately 50% amongst these crocodiles, where most would have died despite correct antibiotic therapy, indicates a reasonable degree of success.

Whether vaccination had any effect in preventing a recurrence of the outbreak or whether improved hygiene and general health of the population were responsible for the further well-being of the crocodiles is not known. There is no reason to believe that crocodiles would not respond immunologically to inoculation with an antigen.

Crocodiles appear to be particularly susceptible to salmonellosis during the early post-hatching period and it is important not to expose them to *Salmonella* spp. during this period. Meat fed to the crocodiles was heat-treated to exclude the possibility of disseminating *Salmonella* infection. The hatchling crocodiles readily accepted this meat when it was fed immediately after hatching.

CONCLUSION

Antibiotic therapy alone is seldom successful in resolving an infectious bacterial problem in intensively housed populations of poikilothermic animals such as crocodiles and fish. Knowledge of the hosts' preferred environmental temperature while healthy or when diseased, the temperature requirements for maximal immune response as well as the temperature preference of the pathogen are needed to establish a sound treatment regimen. Furthermore, in densely stocked, controlled environment houses, adequate attention must be given to the overall hygiene of the house, particularly during disease outbreaks. Whilst intensive treatment, including force-feeding of individual crocodiles, is time-consuming, it is justifiable due to the high value of the individual animal. Vaccination with a bacterin vaccine is well tolerated and may provide a sound prophylactic measure when dealing with salmonellosis.

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