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Diagnosis of the principal infectious diseases in reptiles: pathology, traditional and molecular microbiology, and serology

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Reptile medicine is a branch of veterinary medicine, which underwent a major expansion starting in the 70’s and reaching, during these past few years, a first maturity that is witnessed by the increasing number of subspecialties that have recently blossomed from it. Among those, infectious disease and pathology represents one of the areas that showed a remarkable development also thanks to the new diagnostic and investigation tools that became available along the way. The numbers of microorganisms that is possible to detect, isolate and identify in reptiles is basically impossible to calculate, and even more to characterize. The major effort that has been done in these past few years was directed in attempting to link a microorganism observed within the context of a disease, to the actual disease in a causative manner. This is a long, tedious and expensive process, which, given the limited resources available, has been possible to adopt only for few microorganisms, mostly viruses and bacteria. Although, these microorganisms represents only a very small part of the realm of potential pathogens that crawl daily on the skin and in the viscera of reptiles, the remarkable depth of the investigations that were conducted to characterize some of them (and their interaction with the host), and the dedication of the people that has spent countless hours in this pursue, has allowed to shed light on critical aspects of the pathophysiologic of these animals such as the function of their immune-system and the variation of other physiological parameters within the context of the host-pathogen interaction. The ‘byproduct’ of these investigations and of these research projects has been the development of multiple diagnostic tests which allowed to refine our knowledge about the features of the microorganism along with that of the host response kinetic, dynamic and specificity. The new data and information that have been acquired represent often a baseline that can be partially transferred to other reptile systems of host-pathogen interaction.

Although it is not possible to review all the investigations that have been conducted in this field, here, we will give a brief overview of the most significant findings that have been collected within the context of the main research projects. Among others, Herpesvirus and Mycoplasma sp infection of tortoises, inclusion body diseases of Boid snakes, Ophidian paramyxovirus and lizard adenovirus are diseases that have been extensively investigated and represent true examples of how far we have come in this area of veterinary medicine.

Tortoise herpesvirus and Mycoplasma sp are the two proven pathogens better known and characterized in chelonians. Tortoise herpesvirus has been shown to be a worldwide pathogen for tortoises, causing a disease characterized by a sudden onset and unpredictable recurrence, most likely secondary to the ability of this virus to undergo latency (Origgi et al., 2004/2006). Mycoplasma sp is a causative agent of a chronic and debilitating upper respiratory disease of tortoises (URTD) which has been well characterized in the Gopherus agassizii and which is considered one of the factors that might have contributed to the severe decline of the free-ranging populations of Gopherus agassizii in the Southwest United States during the last 30 years (Jacobson et al., 1991; Brown et al., 1994). More detailed descriptions and features of these two diseases can be found in other abstracts of these proceedings [Herpesvirosi delle testuggini: 1998-2008. Tutto quello che abbiamo imparato in dieci anni di ricerca; Scienza medica veterinaria e conservazione. L’esempio delle Testuggini del deserto (Gopherus agassizii)].

Inclusions body disease (IBD) of Boid snakes is a chronic disease characterized by multiple clinical signs including neurological and respiratory signs, stomatitis, lymphoproliferative disorders and round-cell tumors. In the late 1970’s and extending into the mid-1980’s, Burmese pythons (Python molorus bivittatus) were the most common boid snake seen with IBD, showing clinical signs such as head tilts, disequilibrium, and opisthotonos. Later on, more specifically from the early 1990’s, the cases seen in Boa constrictors outnumbered those seen in Burmese and other pythons. In Boa constrictors, in addition to neurological signs, affected snakes were reported to regurgitate food items within several days of feeding. Mortality can occur within several weeks or months after the appearance of the first clinical signs. In Burmese pythons the disease has more chronic features, with primary neurological signs such as flaccid paralysis of their entire bodies. Regurgitation is not seen in Burmese pythons (Jacobson, 2007). The histological hallmark of the disease is the presence of intracytoplasmic eosinophilic inclusions. Although there are evidences suggesting the role of a retrovirus as etiologic agent, the causative relationship between the presumptive pathogen and the disease has not been confirmed yet (Schumacher et al., 1994). The role of other viral agents has been investigated but no causative relationship was assessed for any of these other candidate microorganisms either. The absence of
a well-defined and characterized causative agent has severely limited the diagnostic potential for this disease. At the moment, the only diagnostic tool that can conclusively confirm the presence of the disease is the observation of the intracytoplasmic inclusions by histopathology. An ELISA test has been developed to detect Boas (Boa constrictor occidentalis) immunoglobulin (Lock et al., 2003), but it will not be able to be used for the diagnosis of IBD until the causative agent will be conclusively identified and the specific antigen will be used in the test. Molecular tests are potentially available and very easily applicable, once the putative causative agent will be identified. The best therapeutic tool for this disease at the moment is prevention. A quarantine of at least 6 months is absolutely necessary and the exchange of snakes with uncertain or debatable origin should be avoided.

Ophidian paramyxovirus is another well-characterized disease of snakes. The first paramyxoviral out-break occurred in Switzerland in the late 70’s in a colony of Lancehead vipers (Bothrops moojeni). Following this first report, the disease has been observed in several Boidae, Elapidae, Colubridae and Crotalidae (Jacobson, 2007). The causative agent is a Paramyxovirus, a single-stranded, negative-sense RNA virus that has been isolated and characterized. Frequent gross lesions observed in infected snakes are diffuse hemorrhage with collection of caseous necrotic debris within the lumen of the airways of the lung and in the air sacs. The lung parenchyma is frequently thickened and edematous. In terminally ill snakes, blood or caseous-purulent material might be expelled from the glottis. Classic historical features of the disease are the proliferative response of the alveolar cells and the presence of intracytoplasmic eosinophilic inclusions. Neurological signs can be commonly observed in sick snakes, while neurological disease has been observed only occasionally (Jacobson, 2007). A serological test (hemagglutination inhibition), and a molecular test (Reverse transcription polymerase chain reaction-RT-PCR), for the detection of the exposure or the presence of the virus respectively, have been developed. These tests are commonly used for the preventive screening of collections of captive snakes and populations of wild snakes.

Adenoviruses have surfaced as significant pathogens in lizard and among others in Bearded dragons (Pogona vitticeps). Lethargy, weight loss, diarrhea, and sudden death are clinical signs commonly observed in adenovirus-infected Bearded dragons (Julian et al., 1982, Jacobson et al., 1996). The classical lesions are seen in the liver and consisting of foci of coagulative necrosis with intranuclear inclusions. Similar inclusions have also been observed within the enterocytes of the small intestine (Jacobson, 2007). Recently, the viral etiologic agent of the necrotizing hepatitis of the Bearded dragons has been characterized and it has been determined to be a member of the genus Atadenovirus along with other adenovirus detected in different species of lizards (Wellehan et al., 2004).

In the last 30 years we have made giant steps, starting a long journey with few if any, diagnostic tools, plenty of anecdotal information and very limited scientific data. Today, we have great investigation and diagnostic tools that are updated, renewed and replaced in real time, exponentially accelerating the time of discoveries and of the gathering of new information. It is now time to shift gear and start to answer the bigger pathogenetic questions of several of the diseases that have been investigated so far and of the many to come. The bio-informatic and bio-technological support is expected to be even more efficient in the future, making the next part of the journey as exciting as it has never been.

References