Case 1

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SIGNALMENT:
Five year old, female, giant African millipede (*Archispirostreptus gigas*)

HISTORY:
This animal presented for paralysis of the caudal half of her body. The animal was euthanized and submitted whole for histologic examination. She lived in a terrarium with nine other individuals that were unaffected.

GROSS FINDINGS:
No external lesions were noted. Internal examination was not performed grossly to preserve tissues for in situ histologic examination.

HISTOPATHOLOGIC/CYTOLOGIC FINDINGS:
Multifocal granulomas expanded the coelom, frequently occupying up to 50% of the coelomic cavity, predominantly in the dorsal aspect. Granulomas were most prominent in the midbody, with smaller multifocal granulomas in more caudal segments; granulomas were not identified in more anterior segments. Inflammation predominantly infiltrated the digestive tract, nerves, and tracheoles. Granulomas had a central core of abundant necrotic cell debris, melanin pigment, and small numbers of bacteria. Granulomas were surrounded by fibroplasia with hemocytes.

MORPHOLOGIC/ETIOLOGIC DIAGNOSIS:
Body as a whole: severe, chronic, multifocal to coalescing, granulomatous coelomitis with intralesional bacteria

DISCUSSION:

Invertebrates comprise 98% of all animal species, yet there is a paucity of literature regarding health and disease of most invertebrate species. Invertebrate pathology offers a unique challenge of interpreting and diagnosing lesions in animals for whom references on even normal histology, let alone pathology, are not widely available. There are several excellent texts that provide general foundations on invertebrate anatomy, histology, immunity, and medicine, including *Invertebrate Zoology, Microscopic Anatomy of Invertebrates, Invertebrate Immunity,* and *Invertebrate Medicine*, respectively.[1,4,7,8] Color HE histology references are not widely available, and the primary literature, including the *Journal of Invertebrate Pathology,* is often the most useful in obtaining such reference material.

The importance of obtaining representative histologic controls when evaluating the pathology of invertebrates cannot be overstressed. Many invertebrate species have symbiotic organisms, immune responses, and other structures for which there is no vertebrate homologue, such as zooxanthellae in cnidarians, and excretory concretions in gastropodian nephridia. As histologic controls or references are

often not available for rare or endangered invertebrate species, control histology for any species within the same taxonomic class, or in some cases phylum, often serves as a good proxy. Another hurdle in evaluating invertebrate pathology is processing exoskeletons for histology. Histology Consultation Services (Everson, WA) uses Rapid Cal (BBC Biochemcial, Mount Vernon, WA) to soften the exoskeleton prior to sectioning for histology with good results (Leroy Brown, pers. comm.). Other tips on processing these animals for histology can be found in *Invertebrate Medicine*.[1]

Giant African millipedes are arthropods in the class Diplopoda that live in African rainforests. These animals have nervous, digestive, respiratory, circulatory, reproductive, and musculoskeletal systems. The nervous system comprises a brain, from which extends a ventral nerve cord with lateral branches.[3] The digestive system consists of the fore-, mid-, and hindgut, as well as Malpighian tubules and fat body. Malpighian tubules are part of the digestive tract as they are attached to the intestines, but have excretory function.[2,5] The respiratory tract is rudimentary and consists of gas exchange through pores in the body wall that connect with a series of tubes called the tracheal system.[5] These animals have an open circulatory system with a muscular heart pumping hemolymph that bathes tissues in the coelom.[5] Millipedes are sexually dioecious and oviparous.[7]

Bacterial infections are common in millipedes and are often secondary to aberrant environmental conditions or cutaneous trauma. Associated bacteria are usually from the environment or millipede flora.[1] In this case, bacterial culture was not performed. In arthropods, bacterial culture can be performed by sampling hemolymph from a leg. Sampling from the coelom is less ideal as it is frequently contaminated by overgrowth of normal enteric flora, which occurs rapidly postmortem, and sometimes even antemortem in moribund arthropods.[6] Causative bacteria are frequently low in number, and plating directly onto culture media, rather than placing in transport media, may help yield positive culture results in paucicellular infections.[9]

Immune defense in arthropods is both cell mediated and humoral with many bactericidal components in hemolymph. Bacteria are killed by hemocyte phagocytosis, deposition of melanin and its intermediate products leading to free radical damage, and antibacterial substances in the plasma, such as lysozyme.[10,11] In this case, bacteria were demonstrably surrounded by hemocytes and melanin, primary components of the immune response that aid in destroying offending bacteria.

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