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Peritonitis Secondary to Multiple, Full-thickness Jejunal Wall Perforations in a California Sea Lion

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Abstract

A juvenile, female California Sea Lion (*Zalophus californianus)* that had stranded at Fisherman's Cove, Laguna Beach, California in late March of 2012, presented in critical condition from severe malnutrition (starvation). Despite aggressive treatment, the seal died the following morning. Gross postmortem examination revealed the carcass to be markedly depleted of subcutaneous and visceral adipose stores, characteristic of severe, chronic malnutrition (starvation). In addition, there was a small, cervical abscess; marked omental and mesenteric hyperemia; multiple full-thickness perforations of the jejunal wall, with marked secondary abdominal effusion and moderate mesenteric lymph node enlargement. Cytological and microbiological evaluations of the abdominal exudate were consistent with marked, chronic-active, multiple-species, bacterial peritonitis. Histological findings included multiple, coalescing foci of pneumonia; diffuse hepatocellular vacuolation; and extensive, necrotizing, subacute jejunitis with multiple full-thickness perforations of the jejunal wall and secondary effusive peritonitis. While the definitive cause of the jejunal lesions is uncertain, it can well be hypothesized to be a result of conspecific trauma, i.e. unweaned sea lion pup loses mother, tries to attach itself to another female sea lion, and is attacked by abdominal biting.

Case Report

On March 27, 2012, beach-goers reported a female, juvenile California Sea Lion (*Zalophus californianus)* pup stranded in a cave on the concrete-block jetty at Fisherman's Cove, Laguna Beach, California. The sea lion was prevented from returning to the water by the presence of people and dogs around the jetty. A Pacific Marine Mammal Center (PMMC) rescue team was dispatched to the scene, where an initial evaluation suggested severe malnutrition. The seal was impounded and transported to PMMC for further evaluation and care.

Following admission, a physical examination was conducted. The seal ("Ella") was ambulatory, but moved reluctantly and only when encouraged to do so. She responded aggressively to tactile stimuli as well as human movements in the vicinity of her head, and she readily attempted to bite staff members when they crossed her line of sight. Her vital signs were all with normal limits, i.e. heart rate 120 bpm; respiratory rate 12 bpm, blood glucose 98 mg/dl and rectal temperature 100.2F. Severe dehydration was evident, and palpation suggested subcutaneous adipose tissue depletion and moderate muscle wasting. The abdomen was slightly distended. A limited neurologic examination revealed mental dullness.

A provisional diagnosis of maternal disassociation and subsequent Stage 3 malnutrition (starvation) was made. Aggressive rehydration and nutritional support therapy were initiated, but the pup was found dead early the following morning and a postmortem examination was conduct.

Postmortem Examination

Gross Pathology

A small, superficial, healing abrasion was noted on the left fore-flipper. A ~2.0 cm, 'melanized', circular cicatrix was noted on the right distal cervical area. Subcutaneous and visceral adipose stores were almost completely depleted.

In the thorax, there was bilateral, ventral, gravitational, pneumonic congestion, with a few scattered foci of apparent consolidative pneumonia. The lungs also contained multifocally dessiminated, raised, 1-3 mm lesions, consistent with *Parafilaroides sp* (Nematoda) infestation, common in pinnipeds.

The abdominal cavity (*Figure 1*) contained 435 ml of a fetid, thick, opaque, 'mustard' colored peritoneal effusion. Samples of the effusion were collected for bacteriology, cytology, and fluid analysis. The greater omentum was markedly edematous and hyperemic, with moderate to marked, multifocally disseminated hemorrhages. Mild-moderate liver enlargement and mustard discoloration was consistent with starvation-related lipidosis (Bunch 2000). Two 20 cm lengths of the mid-jejunum were discolored dark-grey, principally along their antimesenteric borders, suggesting severe, vascular congestion-hemorrhage. Scattered within these areas were 5-10 mm, ragged-edged, dark red, full-thickness perforations (*Figures 2 & 3*). The remainder of the bowel appeared normal, with the exception of mild-moderate, postmortem gas distension. The bowel contained no ingesta. The kidneys, urinary bladder, spleen, and reproductive tract were normal in appearance. The mesenteric lymph nodes were moderately enlarged. Lymph node impression smears were taken for cytologic examination.

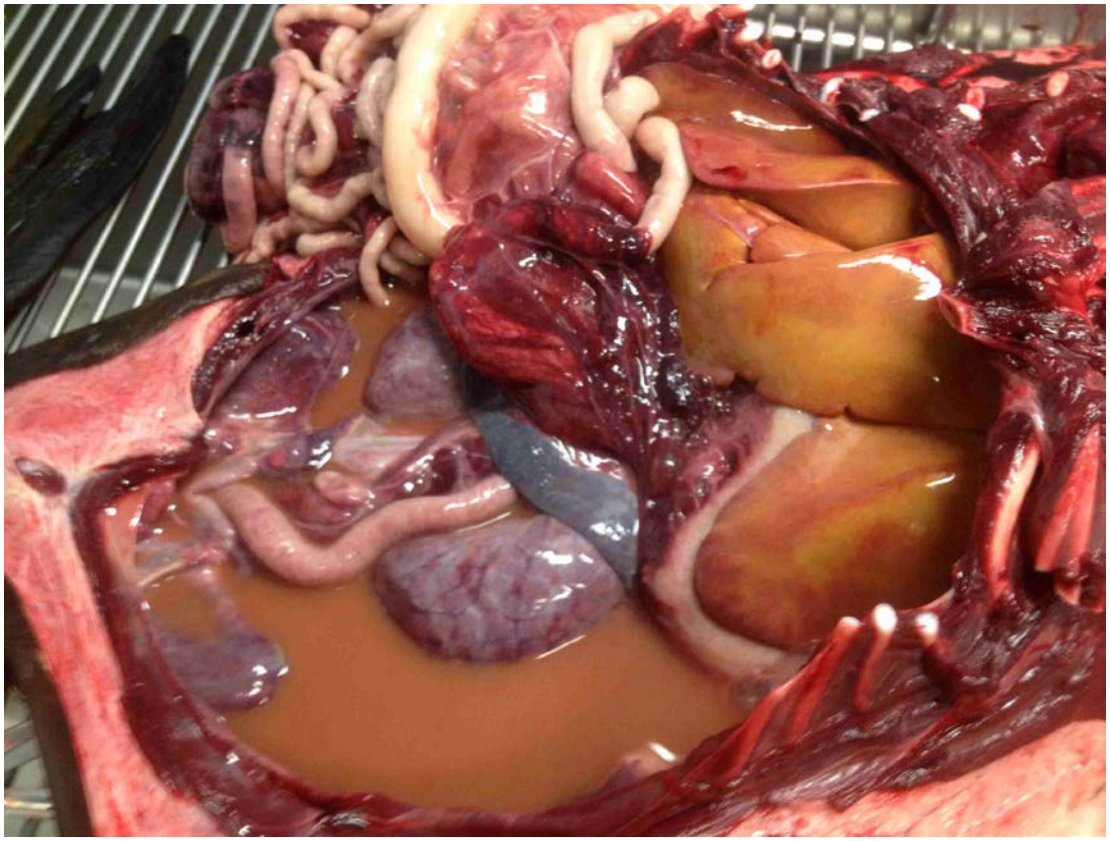
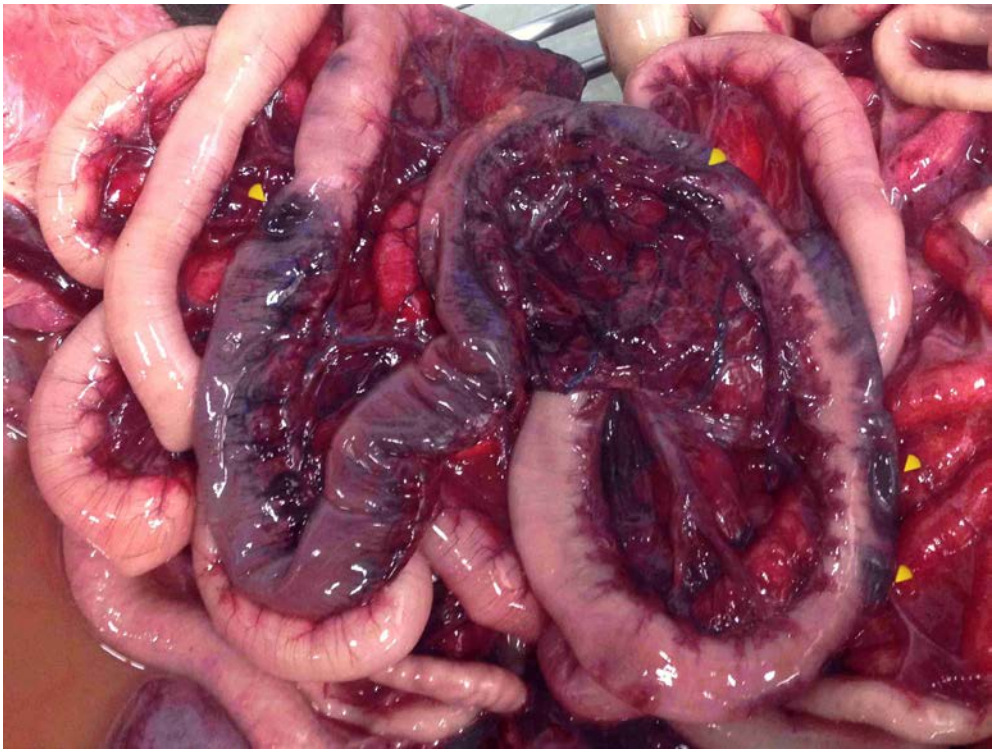
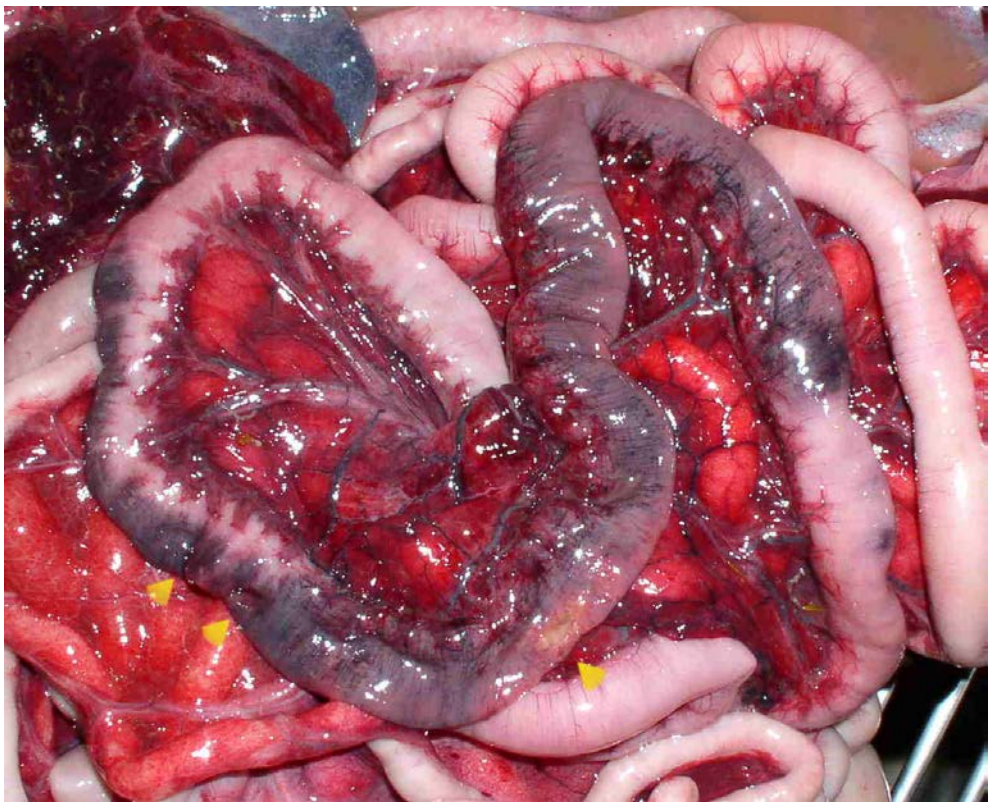


Figure 1. Abdomen. Enlarged, mustard-colored liver; marked mesenteric hyperemia and vascular congestion, and marked peritoneal effusion.



Figures 2 and 3. Abdomen. Moderate, mesenteric hyperemia and congestion, with an extensive section of jejunal hemorrhage and multifocal, full-thickness mural perforations (yellow arrow heads)



Abdominal Effusion Characterization and Cytology

The abdominal effusion had a total protein of 300 mg/dL and ~500 leukoctes/ul (AimStrip®, 10-SG). Wright's Giemsa stained smears revealed large numbers of polymorphonuclear (~50-60%) and mononuclear (~40-50%) cells undergoing nuclear degenerative vacuolative change. Both neutrophils and monocytes were not uncommonly found to have phagocytized cocci and coccobacilli. These finding are indicative of an exudate.

Impression smear of the right and left mesenteric lymph nodes revealed tightly-packed, broad sheets of small lymphocytes, with moderate numbers of scattered, medium-to-large lymphoid cells, a significant number with dark blue cytoplasm indicative of immune stimulation.

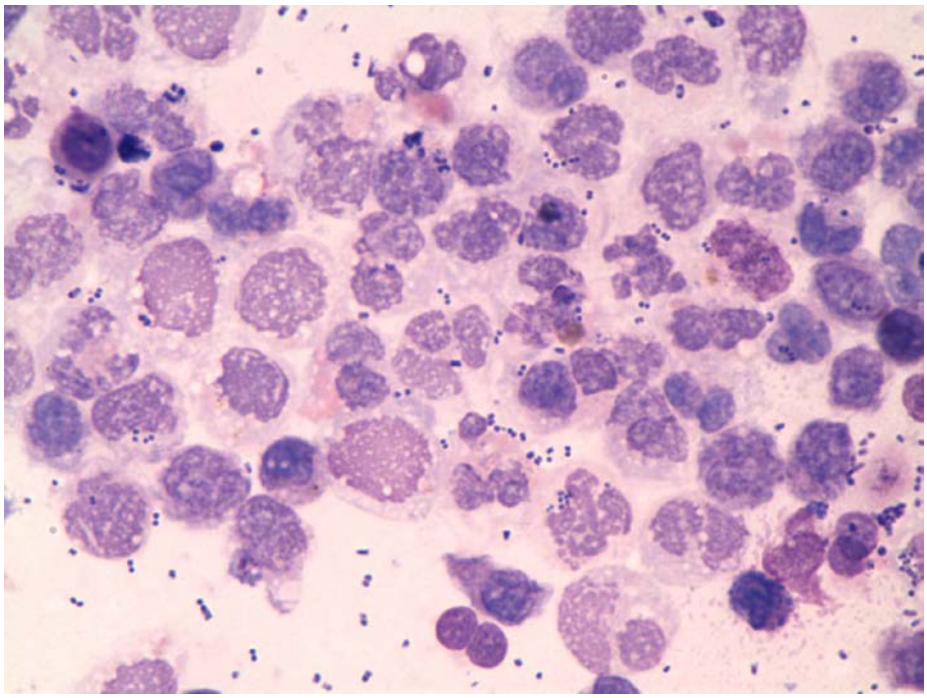


Figure 3. Cytology of peritoneal effusion. Leukocytes are abundant and undergoing varying degrees of degenerative change . The leukocyte differential count was 54% neutrophils and 46% mononuclear cells (lymphoid cells, monocytes, and macrophages). Many mononuclear cells had engulfed varying amounts of cellular debris and bacteria. (Wright's Giemsa Stain)

Histology

A cutaneous-subcutaneous section of cervical skin containing an apparent, subcutaneous abscessation exhibited moderate, diffuse hyperemia and hemorrhage with mixed inflammatory cells infiltration, surrounding an subcutaneous abscessation with a well-formed wall. The abscess cavity was filled with numerous, dissecting, septate tags and moderate amounts of a mixed inflammation and hemorrhage.

Sections of the left and right lungs exhibited varying-sized foci of alveolar consolidation by moderate to marked neutrophils, monocytes and macrophages with common phagocytosis of cellular and inflammatory debris by macrophages. Additionally, a few, scattered foci of gravid adult *Parafilaroides sp*. were noted, but without any associated inflammation.

With the exception of a multifocal, mild, mixed, serosal leukocytosis, pathology was not noted in kidney sections. Several sections of liver were examined and all exhibited severe, diffuse, hepatocellular vacuolation. In the greater omentum there was marked, diffuse, vascular congestion; mild to moderate, multifocally dessiminated, interstitial hemorrhage and mild to moderate, mixed (neutrophils and mononuclear cells) inflammatory cell infiltration. The grossly obvious, discolored dark-grey areas along the mesenteric border of the jejunum exhibited diffuse, moderate-severe, congestion and hemorrhage with moderate mural necrosis and mucosal sloughing (Figure 4). In the ileum there were multifocal areas of diffuse, mild-moderate infiltration of mixed inflammatory cells. The distal portions of the ileum and throughout the colon contained scattered embedded *Corynosoma sp*. (Figure 5). The mesenteric lymph nodes exhibited diffuse, moderate, reactive lymphoid hyperplasia.

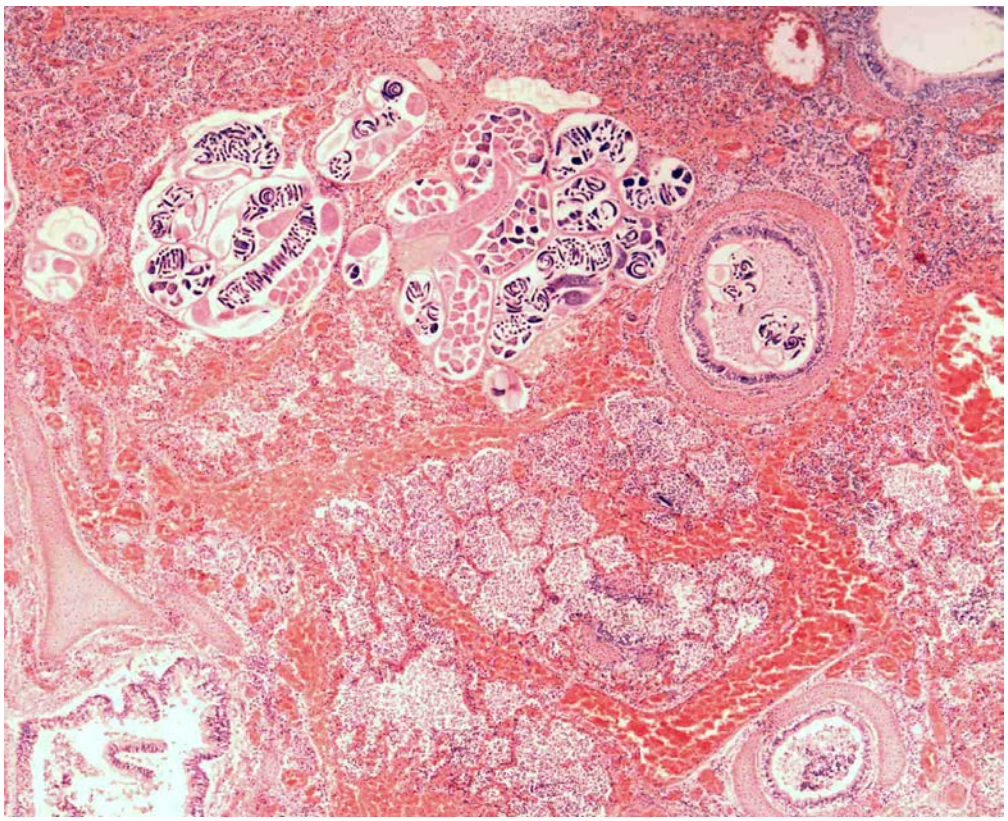


Figure 4. Lung. Note the intense dependent, vascular, postmortem congestion. Along the top of the photomicrograph are alveoli packed with multiple gravid, female *Parafilaroides sp*. nematodes filled with short basophilic larvae. The bronchi at ~2:00pm also contain two gravid, female *Parafialroides sp.*

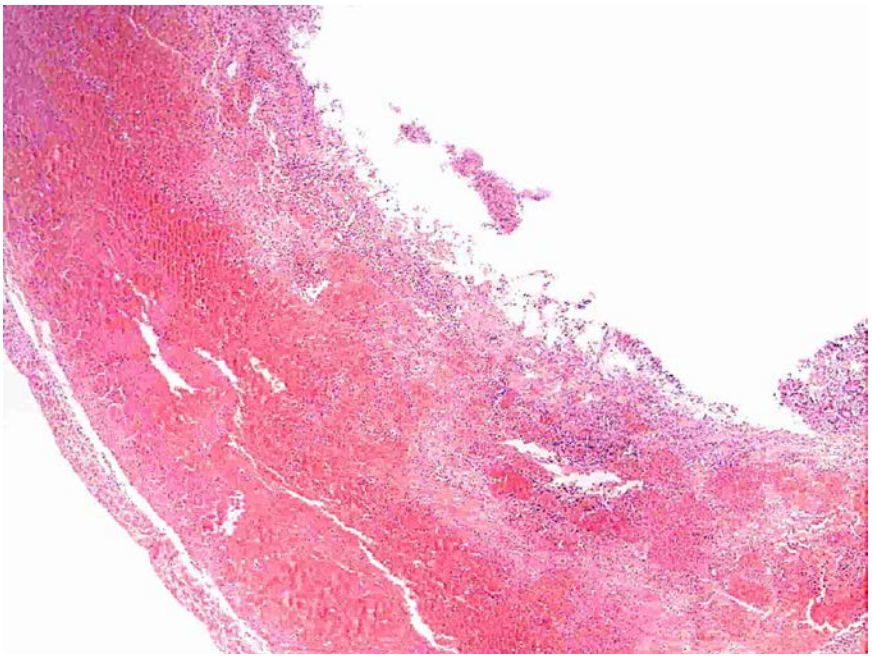


Figure 4. Jejunum. Full-thickness histology section of the jejunum, immediately lateral to the full-thickness mural rent. The left side is serosal which is mild to moderately enlarged by inflammatory cells, while the right side is the necrotic and markedly inflamed mucosa.

Bacteriology

Enterococcus sp., Shewanella algae (a gram-negative bacillus), and *Escherichia coli, biotypes 1* and *2* were isolated from the peritoneal effusion (IDEXX Laboratories). *Escherichia coli, Biotype 1* was resistant to Amoxicillin, Clavamox®, Piperacillin, Ciprofloxacin, Enrofloxacin, Tetracycline, Chloramphenicol, and Trimethoprim-Sulfa; the isolate was sensitive to Amikacin, Gentamycin, and Cephalexin, with intermediate sensitivity to Tobramycin. *Escherichia coli, Biotype 2* was resistant to Amoxicillin, Clavamox®, Piperacillin, and Cephalexin; the isolate was sensitive to Amikacin, Gentamycin, Tobramycin, Ciprofloxacin, Enrofloxacin, Tetracycline, Chloramphenicol, and Trimethoprim-Sulfa. The *Enterococcus* isolate was resistant to Amoxcillin, Clavamox®, Gentamycin, Ciprofloxacin, Enrofloxacin, and Tetracycline; it was sensitive to Chloramphenicol. *Shewanella algae* was resistant to Amoxcillin, Clavamox®, and Cephalexin; the isolate was sensitive to Piperacillin, Amikacin, Gentamycin, Tobramycin, Ciprofloxacin, Enrofloxacin, Tetracycline, Chloramphenicol, and Trimethoprim-Sulfa.

Discussion

Young California sea lions (*Zalophus californianus*) frequently present to the Pacific Marine Mammal Center with varying degrees of malnutrition. In this case, a provisional diagnosis of maternal disassociation and chronic malnutrition was made. Despite rigorous nutritional therapy, survival of late-stage malnutrition is problematic, because the associated alterations of critical metabolism are increasingly irreversible.

In addition, this case also presented unusual pathology. Two large expanses of multifocal, mural necrosis and perforation were noted in the jejunum. The greater omentum was markedly edematous, hyperemic with many hemorrhages, consistent with severe regional circulatory disturbance. Despite an in-depth examination for initiating bite wounds or other penetrating abdominal trauma, none was found. Foci of mural jejunal necrosis and perforation also may be secondary to blunt force that causes trans-abdominal pressure and may rupture soft organs (Kolata 1993), but thorough external and internal examination was unrewarding. Other possible causes for the observed jejunal lesions include circulatory collapse and multifocal tissue anoxia, secondary to the hypometabolic state induced by severe malnutrition and dehydration; severe alterations in systemic and focal acid-base balance; deterioration of gut barriers, with secondary sepsis caused by bacteremia or bacterial toxemia; and overt organ failures.

A notable finding in this case is isolation of *Shewanella algae* in association with bacterial peritonitis. The marine environment is replete with *Shewanella sp.* (Ivanova et al 2003; Delong et al 1997; Kato et al 2001). In a recent study, *Shewanella* organisms were isolated from 694 marine invertebrates, seawater, and sediments in the North-West Pacific Ocean. (Ivanova et al 2003) Gram-negative gammaproteobacteria such as *Shewanella* are aerobic and facultative anaerobes that are found in aquatic habitats under high pressure. (MacDonell et al 1985; Bowman et al 1997; Leonardo et al 1999; Venkateswaran et al 1999; Kato et al 2001) They reduce iron (Gram 1994; Bowman et al 1997), and uranium and plutonium (Hau et al 2007), and thus are of interest to the United States Department of Energy, as a possible method to reduce the amount of radioactive waste in groundwater.

To our knowledge, this is the first illness-related isolation of *Shewanella algae* from a marine mammal, although the organisms have been found in surveys of microflora. (Johnson et al 2006) *Shewanella algae* and *Shewanella putrefaciens* have been reported with increasing frequency as suspected causal agents of infections in humans, typically involving ears, skin, and soft tissues. (Tsai et al 2008) In circulating human blood, infections can be very serious, requiring prolonged treatment and, at times, drastic measures such as limb amputation. (Krsnik et al 2002; Sharma et al 2010; Goyal et al 2011) Persons who are immunocompromised, particularly those with chronic liver disease and chronic leg ulcers, are at increased risk for bloodstream infections (sepsis). (Pagani et al 2003; Goyal et al 2011)

In a case study conducted by a hospital in Denmark, two persons with chronic lower leg ulcers developed *Shewanella algae* bacteremia (bloodstream infection) after going into the ocean during a very warm summer. Both survived, but one experienced extensive necrosis and loss of muscle tissue. (Dominguez et al 1996) In another study, conducted by a different Danish hospital, investigators found that, during an unusually warm summer, 67 people (70 percent being children between ages 3 and 15 years) developed ear infections associated with *Shewanella* after swimming or bathing in the ocean. (Holt et al 1997) Evidently, healthy children also may be at increased risk for infection.

In cases such as this young California Sea Lion, multiple bacterial isolates create difficulty with respect to assigning roles in disease processes. The organisms might act independently, dependently, or synergistically. Additional case reports and research studies will be required to understand the roles of the multiple bacterial isolates from this sea lion, and from similar cases.

Acknowledgments

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