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Hematological and Biochemical Parameters of Macropod Progressive Periodontal Disease in Wild Western Gray Kangaroos

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CASE REPORT
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ABSTRACT

Macropod progressive periodontal disease (MPPD), known as *Lumpy Jaw*, poses a persistent and potentially fatal threat in Western gray kangaroos when they are kept in captivity. Such a condition leads to the development of osteomyelitis and sepsis in Western gray kangaroos (*Macropus fuliginosus*). This case study presented the inaugural examination of hematological and biochemical aspects of MPPD with a progression toward sepsis in a captive environment. The primary objective of this research was to pinpoint hematological and biochemical indicators associated with severe MPPD in a Western gray kangaroo held in captivity. The study employed various methods, including clinical, radiographical, hematological, and biochemical analyses, as well as microbiological study methods. The case was a 2.5-year-old male wild Western gray kangaroo with fever (39.7 °C), dehydration, dyspnea, tachycardia, and involuntary jaw clenching due to stress and agitation. The kangaroo had a history of lethargy, anorexia, swelling of the soft tissues of the lower jaw on the left side, and tenderness during palpation. A radiograph of the head revealed mandible proliferative lesions. The hematological and biochemical examinations indicated an increase in the total count of leucocytes, level of neutrophils, number of erythrocytes, hematocrit level, and lymphopenia. Increased activity of alkaline phosphatase, amylase, and creatinine elevated azotemia. There was a decrease in the content of albumin, glucose, and total bilirubin. The bacteria, consisting of *Fusobacteriaceae* spp., *Porphyromonadaceae* spp., and *Bacteroidaceae* spp., were found and identified in all samples. However, this comprehensive diagnosis of MPPD based on clinical signs, radiography, and especially hematological and biochemical parameters of the septic process can be helpful in diagnosis and treatment.

Keywords: Macropod Progressive Periodontal Disease, *Macropus fuliginosus*, Hematological and Biochemical parameters

INTRODUCTION

The welfare of exotic pets is becoming increasingly important in the professional work of biologists and veterinarians (Sotohira et al., 2017; Rendle et al., 2020a). Exotic species are characterized by various lifestyles, behaviors, and reproduction, necessitating special conditions for their care, maintenance, and feeding (Sherwen et al., 2015; Kido et al., 2018).

Among these exotic species is a wild Western grey kangaroo (*Macropus fuliginosus*), which historically inhabited Australia and the island of Tasmania until the 19th century. Today, kangaroos are also kept in national parks and private zoos worldwide (Sotohira et al., 2017).

In contrast to periodontal disease in humans, macropod progressive periodontal disease (MPPD) frequently advances to necrotizing osteomyelitis affecting the mandible or maxilla. This progression involves the development of sequestra and the extensive growth of subperiosteal bone, ultimately causing deformities in the jawbone. While the pathogenesis of MPPD is deemed comparable to that in humans, the uncommon occurrence of osteomyelitis, suppurative inflammation, and necrosis in adjacent soft tissues observed in macropods sets it apart from similar conditions in humans (Yip et al., 2021). The common cause of pathologies is often attributed to keeping animals in captivity. This requires special knowledge and professional skills from veterinarians to make an accurate diagnosis and conduct treatment. Limited published data on the results of hematological and biochemical changes during severe MPPD in kangaroos are available. Some authors have reported the risks of developing this pathology (Kido et al., 2013; Rendle et al., 2020b). Therefore, this study aimed to present the clinical signs and changes in hematological and biochemical indicators of MPPD in Western Gray Kangaroos.

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CASE REPORT

Ethical approval

This investigation was approved according to the Law of Ukraine (No. 3447-IV of February 21, 2006), according to the requirements of an Ethical Permit (Consensus Guidelines on Animal Ethics and Welfare for Veterinary Journals – International Association of Veterinary Editors, Geneva, Switzerland, 2010).

Case presentation and clinical signs

A male wild Western grey kangaroo *(Macropus fuliginosus)* aged 2.5 years was referred to the Animal Central Clinic City of Kyiv, Ukraine. It weighed 52.3 kg, measured 0.95 m in length with a 0.93 m tail, and a height of 1.27 m. The kangaroo had a history of lethargy, anorexia, hypersalivation, and anxiety in history. Physical examination of the kangaroo revealed fever (39.7 °C), dehydration, shortness of breath and tachycardia, and involuntarily clenching the jaw due to stress and anxiety. The initial dental exam indicated excessive secretion of frothy saliva, soft tissue edema on the left side of the lower jaw, and pain on palpation. A radiograph of the head revealed a large radiodense mass in the left part of the lower jaw. Proliferative lesions of the lower jaw with intraosseous opacity (osteolysis) were seen, which created expanding lesions (Figure 1). Hematological studies indicated significant changes in the main parameters (Table 1).

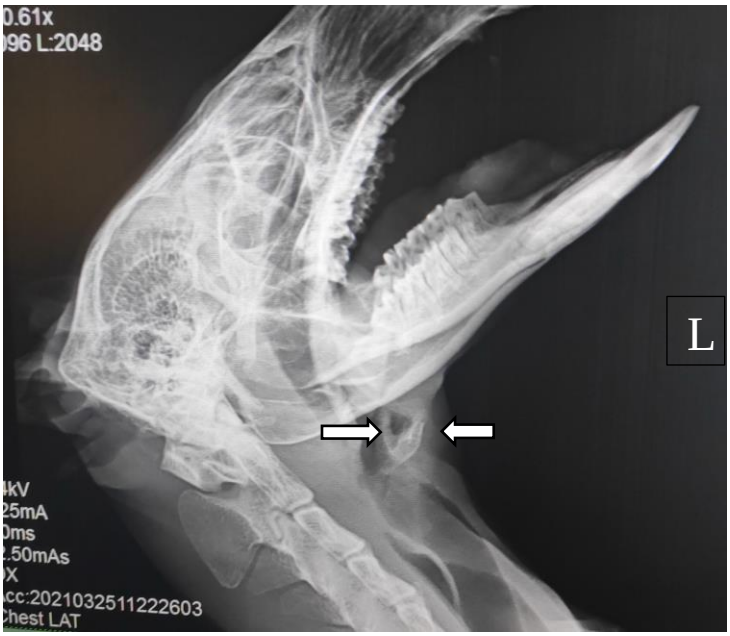

Figure 1. The lateral view of the skull of a wild Western grey kangaroo aged 2.5 years with signs of macropod progressive periodontal disease. The mandible proliferative lesions with intraosseous opacity (osteolysis) and periosteal new bone formation create expansile lesions (white arrows; lateral plane). Bar = 10 mm.

In particular, the acute inflammatory process of the periodontium was accompanied by a marked increase in the count of erythrocytes (6.05×10^{12}/L) and level of hematocrit (0.94 L/L). The laboratory outcomes revealed leukocytosis (13.17×10^{9}/L) with neutrophilia (88.4 %) and lymphopenia (6.1 %). Abnormalities of serum biochemistry included an increase in the concentration of total protein and globulin and a decrease in the albumin level (Table 2). Biochemical changes in the patient's body were characterized by an increase in alkaline phosphatase activity (at 4.45 mmol/L), alanine aminotransferase (at 2.52 mmol/L), and amylase (at 21.16 mmol/L). The activity of creatinine (194.48 μmol/L) and blood urea nitrogen level (at 16.07 mmol/L) were increased. The content of phosphorus and calcium exceeded the upper reference values. The laboratory results determined a decrease in the content of albumin, level of glucose, and total bilirubin (Table 2).

Microbial associations of bacteria were identified in the samples collected from the gums and tooth using a sterile curette or a swab from the subgingival margins. Bacterial species were identified in the microbiological laboratory. The microbiological laboratory utilized non-selective (Wilkens Chalgren Agar, WCA), selective, and enriched media. This included glucose-enriched thioglycollate medium (BBL™ Thioglycollate Medium, Enriched with Vitamin K1 and Hemin, and also Calcium Carbonate), anaerobic kanamycin-vancomycin blood agar (B.D. ™ Schaedler Kanamycin-Vancomycin Agar with 5% Sheep Blood (Schaedler-KV Agar) for selective isolation of gram-negative anaerobes, and Bacteroides Bile Esculin (BBE) agar. The colony morphology was studied using a stereoscopic microscope (Zeiss Discovery.V12 APO Stereo Motorized Microscope Stereoscope - A.V.), which was also used for the tentative

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differentiation of bacteria. Microbial strains (*Fusobacteriaceae* spp., *Porphyromonadaceae* spp., and *Bacteroidaceae* spp.) play a significant role in the pathogenesis of MPPD (NCBI, 2012).

The diagnosis of MPPD was established through a comprehensive assessment, incorporating the case history, clinical signs, hematology, and biochemical parameters. To provide intensive therapy, the animal was transferred to the surgical department of the biotechnological university (Kyiv, Ukraine). As reported by the surgical department, a successful operation was conducted to extract the diseased teeth. The operation was performed under general anesthesia successfully. Following the surgery, antibiotic therapy was administered using clindamycin (Cleocin, Med-Vet International, USA) in a single dose of 20 mg/kg intravenously (*v. coccygeal ventralis*) every 12 hours for 14 days.

Table 1. Hematological parameters of an adult male wild Western grey kangaroo (*Macropus fuliginosus*) with macropod progressive periodontal disease

Parameter	Unit	Normal mean (ranges)	MPPD
Erythrocytes	$\times10^{12}$/L	2.99 (1.52–4.83)	6.05
Leukocytes	$\times10^{9}$/L	7.1 (2.16–14.36)	13.17
Neutrophil	%	55.83 (10.17–68.2)	88.4
	$\times10^{9}$/L	2.47 (0.45–5.7)	11.64
Lymphocyte	%	27.0 (9.39–61.32)	6.10
	$\times10^{9}$/L	3.85 (1.34–8.75)	0.81
Eosinophils	%	5.16 (0.37–6.04)	4.50
	$\times10^{9}$/L	0.56 (0.04–1.41)	0.61
Monocytes	%	7.33 (0–8.1)	0.92
	$\times10^{9}$/L	0.13 (0-0.52)	0.12
Basophils	%	4.66 (0–5.1)	0.10
	$\times10^{9}$/L	0.01 (0-0.09)	0.01
Haemoglobin	µmol/L	1290.21 (967.11-1640.14)	1600.0
Haematocrit	L/L	0.27(0.14-0.41)	0.94
MCV	fl	91.12(62.15-107.91)	82.0
MCH	fmol	2.79 (1.39-4.59)	1.77
MCHC	mmol/L	30.89 (21.31-50.31)	20.10
Thrombocyte	$\times10^{9}$/L	155.94 (66.95-286.1)	258.0

MCV: Mean corpuscular volume, MCH: mean corpuscular hemoglobin, MCHC: mean corpuscular hemoglobin concentration, MPPD: Macropod progressive periodontal disease

Table 2. The blood biochemical parameters of an adult male wild Western grey kangaroo (*Macropus fuliginosus*) with macropod progressive periodontal disease

Parameters	Normal mean (ranges)	MPPD
Albumin (mmol/L)	0.71 (0.70-0.72)	0.57
Alkaline phosphatase (mmol/L)	1.62 (1.61-1.63)	4.45
Alanine aminotransferase (mmol/L)	1.27 (1.2-1.35)	2.52
Amylase (mmol/L)	3.01 (2.66-3.56)	21.16
Total bilirubin (µmol/L	7.5 (7-8)	0.61
Blood urea nitrogen (mmol/L	8.60 (8.4-8.8)	16.07
Ca^{2+} (mmol/L	2.19 (2.17-2.22)	2.34
P (mmol/L)	1.96 (1.77-2.15)	2.57
Creatinine (µmol/L)	125.0 (121.0-129.0)	194.48
Glucose (mmol/L)	8.35 (7.0-9.7)	4.10
Na^{+} (mmol/L)	161.00 (157.0-165.0)	139.0
K^{+} (mmol/L)	4.0 (3.1-4.9)	4.80
Total protein (g/L)	58.0 (56.0-60.0)	80.10
Globulin (g/L)	10.50 (8.0-13.0)	42.10

MPPD: Macropod progressive periodontal disease

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DISCUSSION

Macropod progressive periodontal disease (MPPD), commonly referred to as lumpy jaw in Western gray kangaroos, tends to be a persistent and potentially lethal condition in captive kangaroos (Vogelnest and Portas, 2019; Purcarea and Sovaila, 2020; Rendle et al., 2020a). The MPPD is a severe complication involving the inflammation and infection of bone tissue. In the context of the jaw, this condition can lead to the formation of sequestra and the proliferation of subperiosteal bone, ultimately resulting in bone deformities, which may manifest as the characteristic 'lumpy jaw.' Gingivitis and periodontitis, if left untreated or in specific cases, can lead to complications such as periodontitis-osteomyelitis, where the infection and inflammation extend from the gums and periodontal tissues into the underlying bone, potentially contributing to osteomyelitis (Antiabong et al., 2013; Rendle et al., 2020b).

The distinct purulent and necrotizing lesions are believed to result from infection from anaerobic bacteria, such as *Fusobacterium necrophorum* (Antiabong et al., 2013; Kido et al., 2013; Rendle et al., 2018; Ward et al., 2018). The disease initiates as periodontitis with the invasion of the mucosa by saprophytic bacteria, such as *Corynebacterium pyogenes*, and *Dichelobacter nodosus*, which then extends to adjacent bones, resulting in osteomyelitis (Yip et al., 2021). Protective factors and the progenitor microbiome are important in local immune protection of the mucous membrane (Zhelavskyi, 2021). These lesions typically manifest in the jaws, feet, and less commonly, in stomach. Secondary infections can develop in the intestinal wall, lung, liver, and brain. While wallabies are not uniquely susceptible to *Fusobacterium necrophorum*, predisposing factors such as trauma, fecal contamination of the environment, and stress play a crucial etiological role.

During differentiation, gingivitis, periodontitis, and periodontal abscesses initiated by plaque bacteria should be considered. According to the published studies, gingivitis is a reversible inflammation of the gingival margins, while periodontitis involves the periodontal ligament, connective tissue attachment, and loss of alveolar bone (Antiabong et al., 2013; Hoyer et al., 2020). Diagnosis primarily relied on clinical appearance, radiography, and hematology of suspected cases (Hao et al., 2022). Determination of α-amylase is of great clinical and diagnostic importance in diagnosing and monitoring acute and exacerbations of chronic pancreatitis. However, elevated serum α-amylase levels may be present in pancreatitis, inflammatory salivary glands, and various oral tissues (Dave et al., 2021). Alanine aminotransferase enzyme is found in many animal body cells. However, its highest concentration is in liver cells and kidneys, to a lesser extent, in the heart, pancreas, and skeletal muscles. An increase in transferase activity is a sign of the development of a systemic pathology (Dugar et al., 2020; Zhelavskyi et al., 2020; Zhelavskyi et al., 2023).

All wallabies are thoroughly examined for signs of MPPD during any handling procedure. Special attention is focused on the gingival mucosa where pinhead-sized sinuses might indicate extensive submucosal and perialveolar infection. The diagnostic potential of clinical hematology indicates raised fibrinogen levels and abnormalities of neutrophil morphology as more consistent findings in MPPD cases than in neutrophilia (Antiabong et al., 2013; Rendle et al., 2020b). Septic conditions lead to the malfunctioning of all systems and organs, resulting in severe pathology and frequently serving as the primary cause of the animal's demise. Hematological and biochemical parameters indicate the progression of pathology in afflicted animals (Rendle et al., 2020b; Jevon et al., 2021; Hao et al., 2022).

Treatment of kangaroos with MPPD is also carried out with antibiotics. Literature reports highlight the successful use of clindamycin (Watson et al., 2017; Birot et al., 2022). Currently, intravenous oxytetracycline at 10 mg/kg every day, plus oral metronidazole at 60-70 mg/kg is used daily for MPPD. Antibiotic therapy can continue until at least 2-3 weeks after clinical signs have resolved (Watson et al., 2017). Additionally, there is a mention of adjunctive therapy involving parenteral vitamin A. In cases where repeated handling of untamed animals for antibiotic administration causes stress, the use of long-acting oxytetracycline or amoxicillin every 3-4 days is recommended. Whenever possible, oral metronidazole is administered prophylactically to wallabies that have suffered trauma, and MPPD is a complication (Birot al., 2022).

Even minute sequestra of alveolar bone within granulating lesions significantly impede healing and must all be removed (Birot et al., 2022). To prevent continual re-contamination of an open or 'dry' socket by food material, a packing of zinc oxide/oil of cloves/oil of wintergreen/oil of cinnamon can be useful ('Chlorbutanol', Produits dentaires, S.A. Vevey). The chlorobutanol pack has strong analgesic and antiseptic properties and can be left *in situ* for up to 2 weeks without becoming putrid. In cases of extensive bony involvement, human destruction should be considered.

Macropod progressive periodontal disease can be identified during routine oral cavity examinations. Removal of calculus through ultrasonic or hand-scaling is recommended. In severe cases, parenteral antibiotic therapy is often necessary. Dental abscesses are frequent in macropods, often necessitating surgical debridement of the abscess and sometimes dental extraction (Rendle et al., 2020b; Birot al., 2022).

CONCLUSION

The insights from this case report can contribute to achieving a successful diagnosis outcome for macropod progressive

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periodontal disease in Western grey kangaroos by considering the clinical signs, radiography, and particularly hematological and biochemical parameters. Animal owners and veterinarians must consider all risk factors that can cause this disease.

DECLARATIONS

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Availability of data and materials

The data of the current study are available by request from the authors.

Authors' contribution

Mykola Zhelavskyi originated the presented concept, validated medical history, contributed to data collection, and conducted the experiment. Serhii Kernychnyi authored the manuscript and handled the submission process. Tamara Betlinska participated in designing and coordinating the study as well as providing assistance in drafting the manuscript. All authors reviewed and approved the final edition of the manuscript.

Ethical consideration

The authors considered all necessary ethical issues (e.g., plagiarism, consent to publish, misconduct, data fabrication and/or falsification, double publication and/or submission, and redundancy).

Competing interests

The authors declare that they have no competing interests.

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