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A Case of *Mycoplasma haemofelis* Infection in a Korean Domestic Shorthair Cat

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Abstract : A six-months-old male Korean domestic shorthair cat was presented with fever, tachypnea, anorexia, and weight loss and admitted to Lee Seung Jin Animal Medical Center. During the routine physical examination, clinical signs such as mild dehydration and jaundice in the sclera were present. The complete blood count (CBC) and serum chemistry result showed anemia, thrombocytopenia, neutrophilia, and hyperbilirubinemia. Radiography revealed hepatomegaly and splenomegaly. Blood smear and microscopic examination showed severe hemolysis and anisocytosis. We sent the blood sample to the Neodin Veterinary Laboratory for PCR analysis to conduct a test to find out *Ehlichia*, feline hemoplasmas (haemobartonella), feline leukemia virus (FeLV), feline immunodeficiency virus (FIV) and anaplasma infection. According to PCR examination, the blood of this cat was positive for feline hemoplasmas (*Mycoplasma haemofelis*), but negative for other pathogens. The patient was prescribed doxycycline for 4 weeks and prednisolone for 1 week. The free of feline hemoplasmas infection was confirmed by PCR recheck after six months.

Key words **:** Hemotrophic mycoplasma, *Mycoplasma haemofelis*, *Haemobartonella*, Feline hemoplasmas, regenerative anemia.

Introduction

Hemotrophic mycoplasmas are gram negative, non acid fast, intra erythrocytic and arthropod transmitted bacteria (1). Previously, these organisms were classified as rickettsia in the genera *Hemobartonella* (1). Hemotrophic mycoplasma infection has a worldwide distribution among feline population. Hemotrophic mycoplasma has three species such as *Mycoplasma* (*M.*) *haemofelis, M. haemoinutume, and M. turiensis.* In Korea, there is only one report for the infectious rate of feline hemoplasmas in randomly picked sample from feral cat population using molecular detection method (6). No reports has been made on clinical case yet. In this report, it is solely focused on explanation for *M. haemofelis* infection case in the Korean domestic shorthair cat patient.

Case

A 6-months-old, male domestic shorthair cat named "Toto" was presented to the Lee Seung Jin Animal Medical Center with chief complaint of respiratory distress and anorexia. The cat was rescued in Kyungju city about a month ago and was very friendly toward people and did not exhibit any signs of health problem at that time. But 10 days after vaccination with FELOCELL®, the patient had intermittent fever and respiratory distress. The patient was treated and hospitalization at the other animal clinic but the respiratory distress got worsen after fluid therapy.

At the physical examination, it was revealed that the cat had signs of jaundice in the sclera, pale gum mucosa, and presence of vesicular lung sound. Radiographic examination revealed cardiomegaly, fissure line in the pleural cavity, hepatomegaly and splenomegaly (Fig 1). We waited for any changes in clinical sign without prescribing any medication to rule out overhydration which was caused by fluid therapy. The next day, cardiomegaly and fissure line disappeared but splenomegaly and hepatomegaly were still present in the radiograph (Fig 2). Respiratory distress was improved drasti-

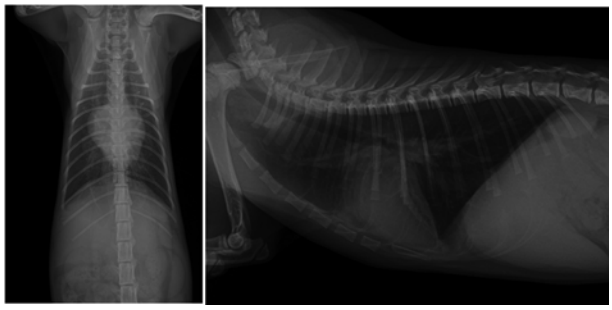

Fig 1. Radiographic examination on the first day of admission. Fissure line and cardiomegaly was detected through chest radiograph.

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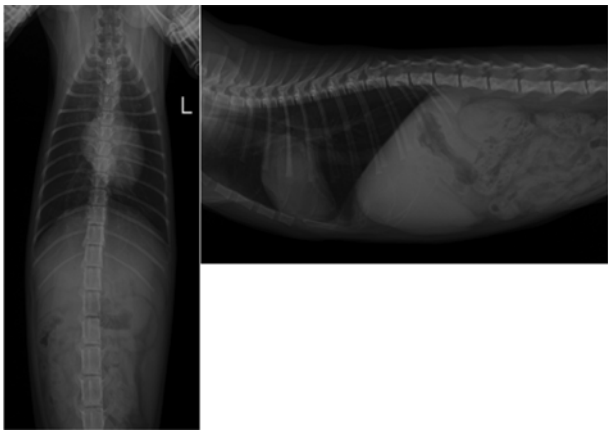

Fig 2. Radiographic examination the next day. Fissure line diminished but apparent splenomegaly and hepatomegaly present.

Table 1. The changes of hematologic parameters in a cat with *Mycoplasma haemofelis* for 24 days

Parameters	Normal range	Unit	Day 1	Day 5	Day 12	Day 24
WBC	5-11	$*10^3$	14.5	18.4	7.3	6.8
RBC	5-10	$*10^6$	2.95	4.45	6.6	7.42
HGB	8-17	g/dL	5.3	7.5	10.4	11.0
HCT	27-47	%	15.1	24.0	34.4	37.2
PLT	180-430	10^3	133	338	386	463
MCV	40-55	um^3	21	54	52	50
MCH	13-17	pg	17.8	16.8	15.8	14.8
MCHC	31-36	g/dL	34.9	31.1	31	29.5
RDW	17-22	%	15.3	18.3	17.1	16.8
MPV	6.7-11.1	um^3	12.1	12.8	11.1	12.9
LYM	0-100	%	14.0	25.0	47.0	53.0
MON	0-100	%	1.8	2.2	2.4	2.5
GRAN	0-100	%	84.2	72.8	50.6	44.5
LY	1-4	10^3	2.0	4.6	3.4	3.5
MO	0-0.5	10^3	0.2	0.4	0.1	0.1
GRA	3-12	10^3	12.3	13.4	3.8	3.2

cally but anorexia was still in progress.

The blood sample was revealed severe anemia (HCT 15.1%) and hyperbilirubinemia (T bilirubin 2.7 mg/dl). Leukocytosis (WBC 14,500) and thrombocytopenia (PLT 134,000) were also presented (Table 1-2). The blood showed severly hemolysis so we couldn't find any other features without anisocytosis through blood smear examination at the first day. Three days later, we made blood smear again and sent the slide to Neodin Vet Lab for evaluation.

The differential diagnosis of feline hemolytic anemia includes autoimmune hemolytic anemia, hemoplasmas, babesiosis, cytauxzoonosis, heinz body hemolytic anemia, microangiopathic hemolytic anemia and pyruvate kinase deficiency. We sent the blood sample to Neodin Vet Lab in order to identify pathogens through polymerase chain reaction (PCR) methods. The same sample was also tested with IDEXX Test SNAP triple combo kit for feline heart worm, feline leukemia virus (FeLV), and feline immunodeficiency virus (FIV).

The cytology examination showed severe form of anisocytosis of RBC and numbers of Howell Jolly Body (Fig 4). Unfortunately the slide was evaluated after two days medication with doxycycline, so we couldn't find any mycoplasma pathogens on RBC surface. Certain numbers of eccentric RBC were also detected. The result of FeLV and FIV came out negative on SNAP kit and PCR analysis.

Table 2. Serum chemistry changes in a cat with *Mycoplasma haemofelis* for 24 days

Parameters	Normal range	Unit	Day 1	Day 5	Day 12	Day 24
T.protein	5.7-7.8	g/dL	6.4	8.4	7.4	
globulin	2.8-5.1	g/dL	3.8	5.1	4.2	
Albumin	2.3-3.5	g/dL	2.6	3.3	3.2	
ammonia	23-78	ug/dL	37	23		
BUN	17.6-32.8	mg/dL	18.0	27.5		
Creatinine	0.8-1.8	mg/dL	1.2	1.0		
ALP	77-358	U/I	152	194		
GPT	23-109	U/I	34	87		
GOT	18-51	U/I	36	29		
Glucose	71-148	mg/dL	130	107		
Chol	89-176	mg/dL	123	213	219	
T.Bilirubin	0.1-0.4	mg/dL	2.7	0.5	0.2	0.1
GGT	1-10	U/I	15	9		
Ca	8.8-11.9	mg/dL	10.1	12.2		
IP	2.6-6.0	mg/dL	5.0	8.0	7.2	

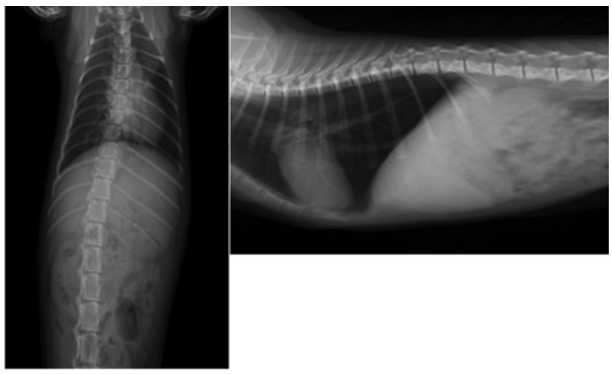

Fig 3. Radiographic examination taken five days after treatment. Spleen and Liver regained its normal anatomical size.

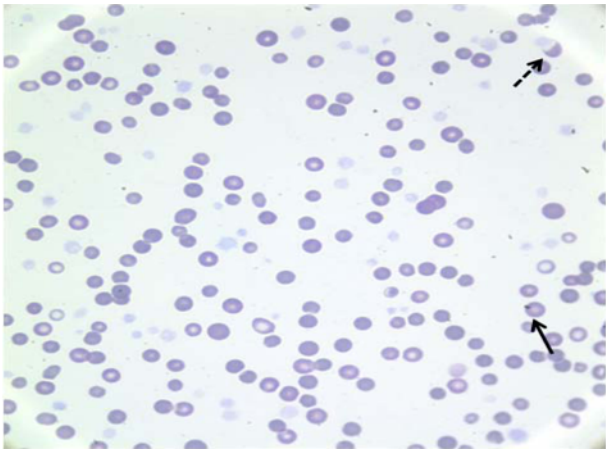

Fig 4. Microscopic examination of peripheral blood smear sample. Anisocytosis of RBC is quite severe and Howell jolly body was detected (solid arrow). Certain numbers of RBC can be thought as eccentrocyte(dotted arrow).

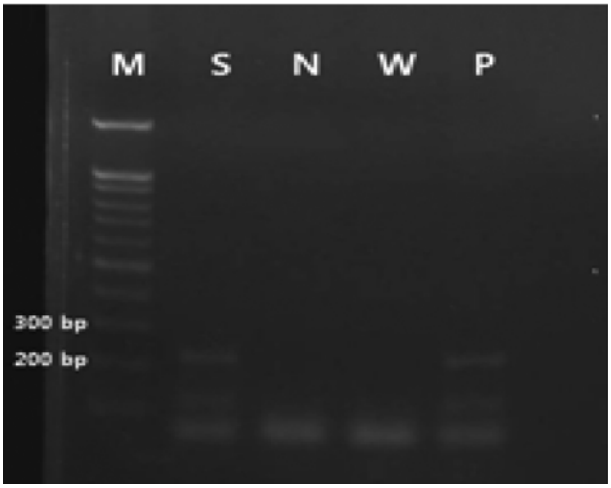

Fig 5. PCR product of blood sample from a cat (provided by Neodin Vet Lab). Blood sample revealed positive result for *Mycoplasma haemofelis*. Lane 1; molecular weight marker (100 bp), S; blood sample, N; negative control, W; distilled water, P; *M. haemofelis* positive control.

The PCR demonstrated 208 bp *M. haemofelis* specific band (Fig 5). However, PCR analysis showed all negative results for Erhlichia, FeLV, FIV, anaplasma.

Initially, the patient was treated for one week with doxycycline 5 mg/kg BID for treatment of the organism, and prednisolone 0.5 mg/kg BID to control of immune mediated hemolysis. Patient regained normal appetite and became active in three days after initial treatment. The HCT increased and icterus diminished a week later. After 5 days, the liver and spleen become normal size in the radiograph (Fig 3). The patient developed diarrhea after taking doxycycline for three weeks. But shortly after cessation of doxycycline treatment, patient's stool was in normal form after one month. The patient have maintained normal body condition and was confirmed free of infection after PCR re-check.

Discussion

Hemoplasmas is the organism causing feline infectious hemolytic anemia. In the past, feline hemoplasmas was classified as *Rickettsia* but based on phylogenetic analysis, it is re-classified as *Mycoplasma*. Those are mainly referred to as the feline haemoplasmas (1). These are non acid fast, gram negative bacteria that is attached to the surface membrane of the erythrocyte. These are important agent that causes mild to severe hemolytic anemia.

The main transmission is by arthropods and fleas (1). Hemoplasmas can be transmitted from mother cat to their kittens and also horizontal transmission is possible by trauma induced during inter-cat aggregation or by blood transfusion (1). The clinical signs are various in genus. *M. haemofelis* causes severe anemia, whereas *M. haemoinutum* does not usually induce clinical signs (4). It has also been suggested that co-infection with *M. haemominutum* and feline retrovirus may result in severe anemia (4).

The organism attaches to the erythrocyte and directly damages the surface. It can cause immune mediated injury that leads to a shortened erythrocyte lifespan and hemolysis (1).

The clinical signs is anemia with fever in the early stage of disease. As other hemolytic anemia progresses, symptoms such as fatigue, depression, anorexia, weight loss, pale mucosal membrane, icterus, and dehydration are present. Some cats show signs of respiratory distress and tachypnea (1).

CBC revealed regenerative anemia, sometimes with thrombocytopenia. During acute phase of anemia, nonregenerative reaction can be seen because of acute hemolytic reaction. The organisms were unculturable in vitro despite of numerous attempts in other studies (1). Blood smear may reveal the organisms on the surface of erythrocyte but is very insensitive for the definite diagnosis. *M. haemofelis* is only seen 50% of the time during the acute phase of disease and *M. Haemominatum* is rarely seen. The organisms may appear as blue stained, small particle, cocci or rods on the erythrocyte membrane (2). The radiograph and ultrasonograph revealed hepatomegaly and splenomegaly. Recently, PCR has become the "gold standard" for diagnosing this infection. FeLV/FIV test are performed because of the possibility of retroviral co-infection (3).

Antibiotics such as doxycycline (5 mg/kg PO q12hr) is primary treatment for Hemoplasma infection. Doxycycline is usually prescribed for 3-4 weeks but oral medication can cause esophageal stricture and gastrointestinal problem such as diarrhea (5). The other antibiotics, marbofloxacin has been proven to be effective but may not eliminate infection completely (5). Oral corticosteroid is recommended in the case of severe anemia (5). Occasionally, blood transfusion is required for patient to recover from severe anemia (1). Progression of the disease should be checked by CBC. After treatment, complete elimination of disease is confirmed by PCR.

The prognosis is variable. If the cat have concurrent dis-

ease, prognosis is poor (5). Elimination of fleas and ticks can help to prevent transmission (5).

In Korea, there were no clinical report of feline *hemoplasma* infection up to these days. But in 2007, 4.2% of *M. haemofelis*, 10.3% of *Candidatus* mycoplasmas *haemominatum*, and 5.3% of the two organisms co-infected were reported among 331 feral cats in Korea (6). The infected cats can be chronic carriers for long period of time without any significant clinical signs. But if the cat got stress from either by vaccination or any other environmental conditions, the clinical symptoms can reveal within certain period of time, may be in many years later (1).

This case is suggested that *M. Haemofelis* infection in young Korean domestic cat has good prognosis if being treated properly according to the treatment protocol.

Acknowledgments

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한국 토종 단모종 고양이에서 *Mycoplasma Haemofelis* 감염 증례

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요 약 : 6개월령의 한국토종단모고양이가 발열, 빈호흡, 식욕부진, 체중감소로 내원하였다. 고양이는 10일 전 Fellocel® 백신을 접종받은 이후 증상을 보였다. 신체검사 결과 공막의 황달과 호흡곤란을 호소했다. 혈액혈구검사와 혈액화학검사에서 빈혈, 혈소판감소증, 백혈구증가증, 고빌리루빈혈증이 관찰되었다. 혈액도말 검사와 기타 검사를 통해 심한 용혈을 확인할 수 있었고 고양이 용혈성 빈혈을 감별진단 하기위해 *Erhlichia*, Hemobartonella, FeLV, FIV, Anaplasma PCR 검사를 의뢰하였다. PCR 검사 결과 *Mycoplasma haemofelis* 특이 208 bp의 양성밴드가 확인되었고, 다른 질병은 음성으로 진단되었다. 고양이는 4주 동안 Doxycycline을 사용하였고 급성기 1주 동안에는 prednisolone을 같이 사용하였다. 임상증상은 약물투여 4일째부터 호전되기 시작하였고 이후 PCR 재검을 통해 완치를 확인하였다. 이것은 *M. haemofelis* 감염 고양이의 진단을 확인하고 완치한 국내 첫 증례이다.

주요어 : 고양이 마이코플라즈마, *Mycoplasma Haemofelis*, 헤모바토넬라, 재생성 빈혈