AgroLife Scientific Journal - Volume 6, Number 1, 2017
ISSN 2285-5718; ISSN CD-ROM 2285-5726; ISSN ONLINE 2286-0126; ISSN-L 2285-5718

CLINICAL AND PARACLINICAL PECULIARITIES OF EFFUSIVE FELINE INFECTIOUS PERITONITIS UNDER NON-SPECIFIC IGY TREATMENT: CASE STUDY

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Abstract

Feline infectious peritonitisis a disease determined by a coronavirus, affectingdomestic and wild cats and once triggered is fatal in almost 100%. The occurrence of the related pathology are yet to be fully determined; currently, a certain and confident in-clinicaldiagnosis is unavailable, as is the case for an efficient vaccine or a treatment that will restore to health.
These make the feline infectious peritonitis a subject currently undergoing intensive research studies.
The paper presents the case of a 5 months cat patient manifesting the rapid evolution effusive form of the disease. The aim of study was to evaluate if the variation of the 24 biological parameters studied was significant under treatment with immunoglobulins of avian origin. The patient tolerated well the supportive therapy based on egg yolk Immunoglobulin Y, without evidences of side effects.

Key words*: feline infectious peritonitis, immunoglobulin Y, immunomodulatory treatment.*

INTRODUCTION

Feline infectious peritonitis (FIP) is a severe multi-systemic immune-enhanced disease of cats, progressive and always deadly, associated with feline coronavirus (FCoV) (Brown et al., 2009; Cobzariu et al., 2015); also, it is a major factor in kitten mortality (Hartmann, 2005). In current Romanian practice, the difficulty of FIP confirmation caused under-diagnosis of disease in several cases (Cobzariu et al., 2015). Despite the weakness of the detection, Feline Enteric Coronavirus (FECV) is prevalent mainly in high-density environments, with limited clinical consequence (Kim et al., 2016).
FCoVs have two pathotypes: the feline enteric coronavirus (FECV) and the feline infectious peritonitis virus (FIPV). The two pathotypes recognize different primary replication site, this feature being related with the clinical outcome. It is accepted the theory that FIPV is originating from FECV infected cats by genetic alterations - gaining tropism for macrophages - and inducing the FIP disease (Bálintet et al., 2012; Tanaka et al., 2012; Kim et al., 2016).
FCoV diagnosis based on antibody titration, antigen detection or PCR-based tests can't differentiate the strains. Actually, histopathology and immuno-histochemistry are the reliable confirmatory procedure but laborious for in-clinical diagnosis (Cobzariu et al., 2015).
Until present is neither effective treatment, vaccine, nor diagnostic protocol able to discriminate the FECV from FIPV (Brown et al., 2009). However, a small ratio of cats develops FIP during the FECV infection. Reliable studies support the important role of immune system in the pathogenesis of FIP.

OPTIONS OF TREATMENT FOR FIP
Many strategies have been developed to cure FIP. Interferons inhibit the FIPV *in vitro* but not *in vivo*. Various immunosuppressants have been studied, but although these drugs increase life average, the FIPV infection remains fatal. Therefore, an effective vaccine and a therapeutic medicine to FIPV are still looking for (Tanaka et al., 2012).
The immunoglobulin Y (IgY) is present in birds and mainly in the egg-yolk (Bentes et al., 2015). The IgY is playing the functional role of

mammalian IgG (Kovacs-Nolan and Mine, 2004), but it present important physical and structural differences toward the IgG molecule and does not cross-react with mammalian immunoglobulins (Polanowski et al., 2012).
The chicken egg yolk contain from 5 to 25 mg/ml of IgY. In egg, IgY is stable for months, and once purified it may be stored for years. The chicken eggs present an ideal alternative source of immune globulins for mammals. Ethically and economically, producing antibodies in hens should be considered (Kovacs-Nolan and Mine, 2004). Moreover, chickens are developing high specific response against mammalian antigens, which never naturally are exposed to (Sudjarwo et al., 2014).
This research evaluated the effect of the use beside the classical protocol of supportive therapy based on egg yolk IgY.

MATERIALS AND METHODS

The study presents the case of a feline patient manifesting an altered health status, with a 30 days history of sickness; the clinical features involved repeated soft moist stool and a constantly abdominal swelling, despite the deworming treatment received.

Table 1. General info of the study case

Info	Patient
Species	*Feliscatus*
Breed	Domestic (Mixed) Breed
Age	5 months
Sex	Male
Hormonal status	Stray
Origin	Adopted from shelter
Vaccination status	Unvaccinated
Parasites infestation status	One single deworming
Where it lives	Exclusively in house
Type of food	Commercial food
Other cats in house	One adult cat, female, 10 years old

The evaluation was carried on the clinical status, hematologic and biochemistry parameters, including the Serum Proteins, mostly in respect to the life quality improvement.
Based on the clinical signs, it was presume the diagnosis of FIP effusive form:
- Low grade fever - 39.4°C;
- Loss of weight;
- Abdominal swelling and pain;
- Lethargy;
- Decreasing appetite.

The ultrasound revealed a non-adherent fluid in the abdominal cavity, in a moderate quantity; consequent the abdominal centesis, 350 mL of greenish-yellow fluid were extracted.
Additional investigations were performed in order to reinforce the presumptive diagnosis: the qualitative immunochromatographic test for IgG antibodies gave low-positive result and the immunofluorescence assay was also positive for antibodies, at 1:400.
For this particular case, the general status of the animal did not allowed performing the biopsy.
In order to screen the patient's evolution, a series of laboratory and clinical investigations was performed in days one and eleven: biochemistry, hematology andelectrophoresis of serum proteins.
The physical examination was carried out as prescribed by medical general procedures, pointing out other relevant clinical manifestations. Blood specimens were sampled, prior and after the treatment, in order to assess the hematological and biochemical parameters and the serum protein electrophoresis.
The blood samples for the hematological assays were collected on EDTA tubes and the samples for biochemical examinations and those for the electrophoresis of proteins were preserved in tubes containing Blood Coagulation Accelerator and Serum Separating Gel.
The investigated parameters were:
- Cells blood count: White Blood Cells, Granulocytes, Lymphocytes/Monocytes, Hemoglobin, Hematocrit, Mean Corpuscular Hemoglobin Concentration and Platelet Count.
- biochemical assays: Glucose (GLU), Urea (Blood Urea Nitrogen - BUN), Creatinine (CRE), Calcium, Alanine Aminotransferase (ALT), Alkaline Phosphatase (ALP), Gama Glutamyl Transferase (GGT), Total Bilirubin (Bil T), Amylase (AMYL) and Pancreatic Lipase (LIPA).

Periodical were performed the abdominal fluid centesis; the fluid was greenish-yellow, shoed an advanced degree of opalescence and had multiple fibrin clots. Four days prior the treatments start, were extracted 350 mL of fluid, as well as 500 mL in day one and 540 ml in day eight.

The avian immunoglobulin (IgY) treatment as supportive therapy has been established for ten days period. The IgY used is a polyclonal IgY, obtained from chickens inoculated with the bacteria listed below: *Acinetobacter baumannii*, *Streptococcus pneumoniae*, *Escherichia coli*, *Staphylococcus aureus*, *Klebsiella pneumoniae*, *Salmonella enteritidis*, *Salmonella typhimurium*, *Pseudomonas aeruginosa*, *Clostridium difficile*, *Candida albicans*, *Helicobacter pylori*, *Streptococcus mutans* and labeled as a food supplement.
For 10 days he IgY was 10 mg/daily, PO.
Symptomatic and supportive therapies were implemented for side pathologies:
- antidiarrheal-therapy for digestive disorders;
- vitamin complexes;
- antibiotic therapy;
- rehydration solutions;
- diuretics.

The patient survived 3 more days after the second set of assays.
The study was performed according to the in force ethic procedures.

RESULTS AND DISCUSSIONS

The catexpressed a clinical evolution specific of FIP effusive form (Table 2). We evaluated the clinical signs that literature presents as being associated with this form, as follows: the moderate pyrexia, abdominal distension, weight loss, anorexia, dyspnea, pericardial effusions, jaundice, ocular lesions and neurological signs (Cobzariu et al., 2015).
Regardless of therapy, of IgY supplement and of the periodic centesis, the overall status of the patient quickly degraded.

Table 2. Clinical signs prior and after avian immunoglobulin treatment

Clinical sign	Prior treatment	After treatment
General status	Modified	Modified
Appetite for food and water	Present	Present, but reduced
Weight loss	Cachexia	Advanced cachexia
Fatigue	No	Yes
Balance in rest	No	Yes
Balance in walking	No	Yes
Lymphadenopathy	No	No
Diarrhea	Yes - soft stool	Yes - soft stool
Abdominal pain	No	Yes
Abdomen	Enlarged	Enlarged
Appearance	abdomen	abdomen
Fever	Yes	No
Respiratory disorders	No	Difficult breathing
Jaundice	No	Yes
Urinary disorders	No	No

Concerning the hematological parameters (Table 3), we note the following:
- severe progressive anemia;
- the decreasing of white blood cells;
- abnormal values of all parameters.

Table 3.Values of the hematological parameters

Parameter	Normal Range	Before	After	Evolu-tion
HCT (%)	24.0-45.0	15.9	16.1	↗
HCB (g/dL)	8.0-15.0	5.6	5.0	↘
WBC (K/µL)	5.00-18.90	21.00	10.90	↘
GRANS (K/µL)	2.50-12.50	15.60	6.50	↘
L/M (10^9/L)	1.5-7.8	5.4	4.4	↘
PLT (K/µL)	175-500	741	243	↘

Concerning the biochemical parameters (Table 4):
- high blood sugar value (quite possible induced by the administration of supportive treatment);
- the renal functions were responsive until the last day of life, when the increase of the creatinine and urea over the limit suggest the onset of renal failure;
- the liver and pancreas indicators maintained at physiological values throughout the course of the treatment;
- the increased value of the total bilirubin is in correlation with the destruction of the red cells, this being the cause of the severe anemia.

Table 4. Evolution of the biochemical parameters in blood serum

Parameter	Normal Range	Before	After	Evolu-tion
Glucose (mg/dL)	74-159	88	166	↗
Urea (mg/dL)	16-36	20	43	↗
Creatinine (mg/dL)	0.8-2.4	0.5	0.7	↗
Calcium (mg/dL)	7.8-11.3	8.6	7.1	↘
Alanine aminotransferase (U/L)	12-130	68	98	↗
Alkaline phosphatase (U/L)	14-111	42	32	↘
Gamma glutamyl transferase (U/L)	0-1	0	0	→
Total bilirubin (mg/dL)	0.0-0.9	1.7	3.1	↗
Amylase (U/L)	500-1500	1058	915	↘
Lipase (U/L)	100-1400	815	691	↘

The serum protein electrophoresis (Table 5) pattern, in FIP positive cats, is relevant regarding the Albumin/Globulin ratio (ranging < 0.8). In our case the Albumin/Globulin ratio, prior and during the treatment, was in respect to data before.
The Albumin values remained below the normal range, even decreased, despite the supportive therapy set up; for the Globulins, the values wasover the normal range from the start of therapy and increased slightly, respectively for the Alpha1-globulin and Gamma-globulin.
Although investigated parameters in relation to the supportive therapy recorded differences in absolute value, compared to the normal range but also between the levels before and after therapy, the variation wasn't significant ($p>0.05$).
Despite the evolution of the disease was acute, the patient survived for a period of time that was above the average - 13 days since the treatment started, totaling a 6 weeks period from the earliest clinical signs, comparative to the 8-9 days period provided by most of the literature (Kim et al., 2016; Fischer et al., 2011; Ritz et al., 2007).

Table 5. Evolution of the electrophoresis parameters before and after treatment

Parameter	Normal Range	Before	After	Evolution
Total protein (g/L)	57-78	76.1	80	↗
Albumin (g/L)	27-38	13.2	12.1	↘
Globulin (g/L)	23-50	62.9	67.9	↗
Alpha1-globulin (g/L)	0.5-1	3.1	3.9	↗
Alpha2-globulin (g/L)	12-18	13.2	14	In range
Beta-globulin (g/L)	4-6	5.5	6.1	In range
Gamma-globulin (g/L)	11-19	41.1	43.9	↗
Albumin/ globulin ratio	0.8-1	0.21	0.18	↘

CONCLUSIONS

The cat tolerated well the supportive therapy based on egg yolk IgY, without evidences of side effects.
However, due to the pathological stage of the disease when the supportive therapy was set up, we have not recorded relevant data to conclude on the benefits of this therapy. Further studies must be carried, on subjects submitted to the supportive therapy, as soon as confirmed FIP.

AKNOWLEDGEMENTS

We acknowledge and are grateful for the assistance provided by the stuff of the Faculty of Veterinary Medicine in Bucharest.
The producer of chicken yolk egg IgY provided this compound, but did not interfere and will not interfere in any stage of this research.

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