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Original Article

Comparison of the Performance of Bioresonance, Electrophoresis and RT-PCR in the Diagnosis of Feline Infectious Peritonitis

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Abstract

Feline infectious peritonitis (FIP) continues to be one of the most researched infectious diseases of cats. The diagnosis of FIP is challenging, and diverse techniques have been developed for its accurate diagnosis. However, they have some limitations. The present study was conducted to investigate the efficacy of specific modulation frequency (SMF), compared to other routine diagnostic methods for detecting *feline coronavirus*. Blood samples were collected from 30 diseased cats suspected of having FIP based on clinical signs. Electrophoresis, polymerase chain reaction (PCR), and SMF tests were performed for each sample. The sensitivity and specificity of each test, as well as the agreement between the tests and the gold standard (the combination of PCR, electrophoresis, and bioresonance results), were calculated using the Kappa coefficient method. The sensitivity and specificity of electrophoresis, PCR, and SMF for the diagnosis of FIP were 70.6%, 70.6%, 100%, and 100%, 72.7%, 81.8%, respectively. According to the findings of the present study, SMF is effective and safe in FIP diagnosis, which is a challenge in veterinary medicine diagnosis.

Keywords: Cat, Electrophoresis, Feline infectious peritonitis, Polymerase chain reaction, Specific modulation frequency

1. Introduction

Feline infectious peritonitis (FIP) is a fatal, immune-mediated disease triggered by infection with a *feline coronavirus (FCoV)*. *FCoV* belongs to the family *Coronaviridae*, a group of enveloped, positive-stranded RNA viruses frequently found in cats. It causes an enteric infection that leads to a fatal, systemic disease named FIP, an immune-mediated, progressive polyserositis and pyogranulomatous. It occurs worldwide, affecting both domestic and wild felids, and is extremely common in crowded environments (1).

FCoV strains are subdivided into two distinct biotypes: *Feline Enteric Coronavirus* and *Feline Infectious Peritonitis virus (FIPV)*. Serotype 1 *FCoV* includes unique feline strains, while serotype 2 appears to have arisen from the recombination between type 1 *FCoV* and *canine coronavirus*. Although serotype 1 is the most prevalent worldwide, both serotypes 1 and 2 can cause FIP. In cats with FIP, the virus replicates to high titers in monocytes and can be found in many organs. In asymptomatic cats, on the other hand, *FCoV* is mainly confined to the intestine. However, a low level of monocyte-associated viremia can sometimes be detected in healthy animals by reverse-transcriptase polymerase chain reaction (RT-PCR) (2).

Many times, FIP general clinical signs (such as chronic fever, weight loss, anorexia, and malaise) are nonspecific. That is why FIP is a fatal disease that causes several challenges for veterinarians. Difficulties in definitively diagnosing FIP arise from its nonspecific clinical signs, lack of pathognomonic hematological and biochemical abnormalities, as well as the low sensitivity and specificity of tests routinely used in practice. There are several diagnostic techniques for the clinical detection of FIP. Clinicopathologic changes in FIP (including lymphopenia, neutrophilia, anemia, hyperproteinemia, and hypergammaglobulinemia) are not pathognomonic (3).

Liver enzymes, urea, and creatinine can all be elevated depending on the degree and site of organ damage but are generally not useful in establishing a diagnosis (4).

Serum protein electrophoresis (SPE) is a classic indirect test used to support the diagnosis of FIP. One of the most supporting findings for diagnosing FIP is an increased total protein concentration in serum, along with a decreased albumin-to-globulin ratio (5). The FIP electrophoretic pattern is typically characterized by an increase in the alpha $(\alpha)_2$ and gamma (γ)-globulin fractions (6).

Although RT-PCR can often provide false positive and false negative results in healthy cats, it could be a specific tool for diagnosing FIP when applied in a clinical setting (7).

MORA bioresonance therapy (traditional bioresonance therapy) was developed by the physician Franz Morell and the electrical engineering technician Erich Rasche in the 1970s as a result of medical testing in electroacupuncture (8).

Electromagnetic waves are used for making diagnoses and treatments in bioresonance therapy (also called bioinformation therapy). Bioresonance diagnostic equipment detects and modulates healthy and disruptive waves in the human body, as well as medications and other substances by their vibrations. Several clinical studies have shown that deficient and safe levels of amplitude-modulated electromagnetic fields applied by an intrabuccal spoon-shaped probe could provoke diagnostic and remedial properties (9).

Despite the positive effects of various electric and electromagnetic fields on medical sciences, the application of this technology has been limited to specific fields (10, 11).

The present study aimed to compare the results of the specific modulation frequency (SMF) technique, compared to electrophoresis and RT-PCR in detecting *FCoV* in cats.

2. Materials and Methods

2.1. Samples

Blood samples were collected from 30 diseased cats (11 females and 19 males, aged between 2-24 months) suspected of having FIP based on clinical symptoms (Table 1). These samples were obtained randomly from different veterinary clinics in the west of Tehran from 2020 to 2022. Serum samples were harvested by centrifugation (10 mins at 2,500 g) and kept at -20°C until analysis.

2.2. Electrophoresis

Electrophoresis was performed on agarose gels using the Major Science (CA, USA) system, according to the manufacturer's instructions. Briefly, 10 µl of the serum sample was electrophoresed with TBE buffer at pH 8.6, 100 V for 35 min on a 1% agarose gel. Gels were analyzed on a Bio-Rad GS-700 imaging densitometer using the manufacturer's Multi-Analyst®/PC (Bio-Rad, Hemel Hempstead, UK). The concentration of individual fractions was determined by multiplying the percentage volume of the fraction by the sample total protein concentration measured using the biuret method.

2.3. Sample Preparation for RT-PCR

A maximum of 1 ml of non-coagulated ethylenediamine tetraacetic acid (EDTA) blood was centrifuged for 10 min at 2,500 rpm. Plasma was discarded from the cell pellet, and RNA extraction was performed on the cell pellet.

Table1. Specifications of the studied cats

Case	Sex	Age	Breed	Chief complaint	Clinical signs				
					Fever	Ataxia	Ascites	Uveitis	Dehydration
1	Male	8 month	DSH	Anorexia	+	-	-	+	+
2	Male	4 month	DSH	weakness	+	-	-	-	+
3	Male	12 month	DSH	Not alert	-	+	-	-	+
4	Female	4 month	DSH	Weakness	-	+	+	-	+
5	Male	4 month	DSH	Anorexia	+	-	+	-	+
6	Female	2 month	Persian	Anorexia	-	-	+	-	+
7	Male	24 month	DSH	Weakness	-	+	-	+	+
8	Male	8 month	DSH	Anorexi & weakness	+	-	+	+	+
9	Male	4 month	Persian	Not alert	+	-	+	-	+
10	Male	8 month	DSH	Anorexia	-	-	-	+	+
11	Female	12 month	DSH	Abdominal pain	+	-	+	-	+
12	Male	3 month	Persian	Weakness	-	+	-	-	+
13	Male	8 month	DSH	Anorexia	-	-	+	-	+
14	Male	12 month	DSH	Anorexia	+	-	+	+	+
15	Male	4 month	DSH	Abdominal pain	-	-	-	-	+
16	Female	6 month	DSH	Weakness	+	+	-	-	-
17	Female	6 month	DSH	Abdominal pain	+	-	+	-	+
18	Male	7 month	DSH	Weakness	-	+	+	-	-
19	Male	24 month	Persian	Anorexia	-	-	-	+	+
20	Male	23 month	DSH	Anorexia	+	-	-	+	-
21	Male	13 month	DSH	Abdominal pain	+	-	-	+	+
22	Female	5 month	DSH	Anorexia	+	+	-	+	+
23	Female	12 month	DSH	Weakness	-	+	-	+	+
24	Male	10 month	Persian	Weakness	+	-	+	-	-
25	Female	11 month	Persian	Weakness	+	+	+	-	-
26	Male	6 month	DSH	Weakness	+	-	-	-	+
27	Female	10 month	Persian	Anorexia Weakness	+	-	+	-	-
28	Male	4 month	DSH	Wakness	+	+	-	-	-
29	Female	5 month	DSH	Anorexia	+	+	+	+	+
30	Female	7 month	DSH	Anorexia	-	-	-	+	+

2.4. Reverse Transcriptase-Polymerase Chain Reaction

2.4.1. RNA Extraction and Purification

Viral RNA was extracted from blood specimens following the YTzol Pure RNA Kit, based on the manufacturer's protocol (Yekta TajhizAzma, Iran).

In total, 300 µl cell pellet and 300 µl Trizol were added into a microtube, mixed vigorously, and left at room temperature for 5 min. Afterward, 0.2 ml of chloroform per 1 ml of YTzol Reagent was added to the mixture (60 µl). Tubes were shaked vigorously for 15 sec and incubated for 2 min at room temperature. Samples were centrifuged at 12,000 g at 4°C. Following centrifugation, the mixture was separated into a lower yellow phenol-chloroform phase, an interphase, and a colorless upper aqueous phase. RNA remained exclusively in the aqueous phase and precipitated from the aqueous phase using 150 µl of isopropyl alcohol. Samples were incubated at room temperature for 10 min and centrifuged at 12,000 g for 5 min at 4°C. The alcohol was thrown away, and then 1 ml ethanol 75% was added to the microtubes and mixed gently. Subsequently, the mixture was centrifuged at 12,000 g for 2 min at 4°C. Microtubes were kept at room temperature for 6 min after the alcohol was discarded. Finally, 30 µl sterile distilled water was added and kept at a temperature of 55°C for 10 min and then stored at -70°C.

2.4.2. Primer Selection

The oligonucleotide primers were chosen from the highly conserved M gene sequence (primer 212) of the *FCoV* genome combined with a primer aiming at the leader sequence of the *FCoV* genome (primer 1179). Primer sequences are shown in table 2 (12).

Table 2. List of primer sequences, size and positions used for detection of *FCoV* in blood samples

Oligo	Sequence(5′-3′)	Size	Position	Orientation
212[a]	TAATGCCATACACGAACCAGCT	22	26440-26461	Antisense
1179[a]	GTGCTAGATTTGTCTTCGGACACC	25	60-38	Sense
1181[b]	CAAAGTTGTCATGGATGACC	20	-	Antisense
1180[b]	CCATGGAGAAGGCTGGGG	18	-	Sense

[a] Numerical position on the genome of FIPV79-1179 as determined from the 5′ ATG start codon; M-gene (de Groot et al., 1988)
[b] Feline GAPDH gene
Adopted from Simons, Vennema (12)

2.4.3. Reverse Transcription

A cDNA Synthesis Kit (Vivantis, Selangor, Malaysia) was used to provide the reliable synthesis of full-length cDNA. The procedure was performed in accordance with the guidelines provided by the kit manufacturer. For the RT reactions, 3 µl of the RNA solution and 3 µl of the reverse primer 212 were mixed, incubated for 2 min at 95°C, and immediately cooled on ice. The cDNA Synthesis Mix was prepared as follows: 1 µl dNTPs, 0.5 µl MMLV reverse, 1 µl MMLV buffer, and 7.5 µl nuclease-free water. The reaction mixture was spun down and incubated for 60 min at 37°C. The enzyme was inactivated by incubation at 95°C for 5 min. Afterward, 280 µl nuclease-free water was added to the samples, and they were stored at -20°C before using in the mRNA PCR assay.

2.4.4. Polymerase Chain Reaction

Following RT, the Ampliqon (Odense M, Denmark) master mix was used for the PCR assay, including 12.5 µl Ampliqon master mix, 1 µl primer 212, 1 µl primer 1179, 5 µl primer cDNA, and 5.5 µl nuclease-free water. The temperature cycling protocol consisted of a 10-min incubation at 95°C followed by 30 cycles of 1-min denaturation at 95°C, 1-min primer annealing at 62°C, and 30-sec primer extension at 72°C. The 30 cycles were followed by 5 min at 72°C, and finally, the reaction mixture was cooled to 4°C.

2.4.5. Analysis of Amplified Products

A total of 10 µl of each PCR sample was analyzed by electrophoresis using a 1.5% tris-acetate-EDTA agarose gel (CinnaGen Co., Karaj, Iran) for 40 min at 100 V. A 100 bp molecular weight marker (Sinaclon, Karaj, Iran) was used to control the size of the amplified PCR product. Following electrophoresis, the gel was immersed in 10 mg/ml of Safe Stain solution (Sinaclon, Karaj, Iran). After staining for 15 min, the gel was photographed under UV illumination. Amplification products were photographed using the Bio-Rad Gel Doc 1000 (Bio-Rad Laboratories, Inc).

2.5. Specific Modulation Frequency

The cases were assessed using the SMF according to the manufacturing company's instructions (MINI-EXPERT-DT, Russia). The current measurement of the device was 6 micro A@ 100 kOhm. The device has the ability to apply electric pulses as positive, negative, and bipolar in the frequency range of 0.1 to 15 KHz and the range of 1 to 20 V. For the codetection, pulse amplitude and pulse frequency were adjusted to 12 V and 793/562 Hz, respectively. The IMIDIS-EXPERT software (IMEDIS-EXPERT, Russia) was used for data analysis.

2.6. Data Analysis

In the present study, the agreement between the tests and the gold standard (the combination of PCR, electrophoresis, bioresonance results) was calculated using the Kappa coefficient method. The Chi-square test and the crosstabs between the tests were also used.

In order to design the prediction model and measure the relationships between the tests and the gold standard, the univariate sensitivity and specificity of each test and the area under the curve (AUC) were also calculated using the receiver operating characteristic (ROC) curve.

The SPSS software (version 27) and GraphPad Prism software (version 9) were used for data analyses, and $P<0.05$ was considered the level of significance.

3. Results

3.1. PCR-Based Detection of *FCoV*

Figure 1 demonstrates the gel electrophoresis results of the PCR method in detecting *FCoV*.

3.2. SMF-Based Detection of *FCoV*

Figure 2 shows the SMF findings for the diagnosis of *FCoV* in cats.

3.3. Results of Three Diagnostic Methods:

The results of the three diagnostic methods for detecting *FCoV* and the merged results considered as the gold standard are shown in table 3.

3.4. Comparison of Diagnostic Methods

3.4.1. Kappa Coefficient

The Kappa coefficient presented in table 4 expresses the correlation between each test and the gold standard.

The AUC of each test was calculated using the ROC curve. The AUCs of electrophoresis, PCR, and bioresonance were calculated as 0.853, 0.717, and 0.909, respectively (Figure 3 and Table 5).

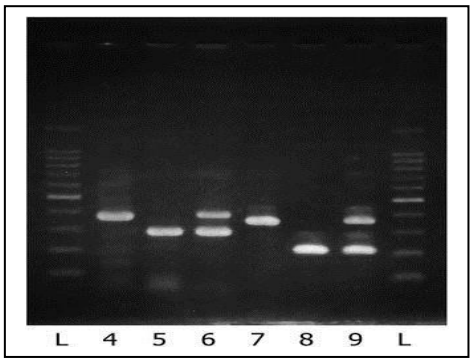

Figure 1. Results of the gel electrophoresis of the PCR used to detect *FcoV* (L:Lader, 4-9: positive samples)

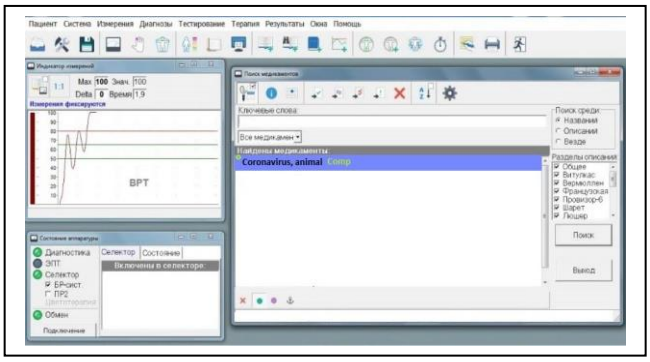

Figure 2. Specificity modulation frequency used for detection of *FCoV*

Table 3. Results of three diagnostic methods for diagnosis *FCoV*

Sample No	Electrophoresis	PCR	Bioresonance	Merged
1	Negative	Positive	Positive	Negative
2	Negative	Negative	Positive	Negative
3	Negative	Negative	Positive	Negative
4	Negative	Positive	Positive	Positive
5	Negative	Negative	Negative	Negative
6	Negative	Negative	Negative	Negative
7	Positive	Positive	Positive	Positive
8	Positive	Negative	Positive	Positive
9	Negative	Positive	Positive	Positive
10	Negative	Negative	Negative	Negative
11	Positive	Positive	Positive	Positive
12	Negative	Positive	Positive	Positive
13	Negative	Positive	Negative	Negative
14	Positive	Negative	Positive	Positive
15	Negative	Negative	Negative	Negative
16	Negative	Positive	Positive	Positive
17	Positive	Positive	Positive	Positive
18	Positive	Positive	Positive	Positive
19	Negative	Negative	Negative	Negative
20	Negative	Positive	Negative	Negative
21	Positive	Positive	Positive	Positive
22	Positive	Positive	Positive	Positive
23	Positive	Positive	Positive	Positive
24	Positive	Negative	Positive	Positive
25	Negative	Positive	Positive	Positive
26	Negative	Negative	Positive	Positive
27	Negative	Negative	Negative	Negative
28	Negative	Negative	Negative	Negative
29	Positive	Positive	Positive	Positive
30	Negative	Negative	Positive	Negative

Table 4. Kappa coefficient between each tests with gold standard

Kappa Coefficient	PCR	Electrophoresis	Bioresonance	Gold Standard
PCR	1	0.267	0.333	0.420
Electrophoresis	0.267	1	0.444	0.653
Bioresonance	0.333	0.444	1	0.845

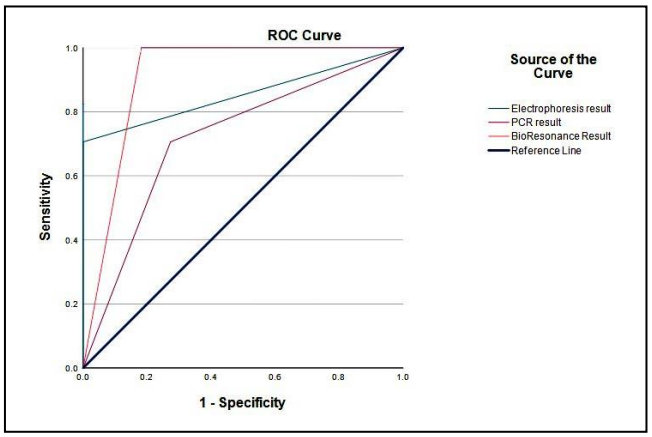

Figure 3. ROC curves of the studied tests for FIP diagnosis

Table 5. Sns, Sps, overall clinical accuracies and AUCs of the studied tests

Tests	Kappa	R Square	Specificity	Sensitivity	Overall	95% CI for OR		AUC
						Lower	Upper	
Electrophoresis	0.653	0.633	100.0	70.6	82.1	-	-	0.853
PCR	0.420	0.229	72.7	70.6	71.4	1.183	34.61	0.717
Bioresonance	0.845	0.795	81.8	100.0	92.9	-	-	0.909

4. Discussion

The FIP diagnosis is a challenge in veterinary medicine. This report describes the bioresonance assay to detect *FCoV* in cats. This study compared the clinical accuracies of electrophoresis, RT-PCR, and bioresonance for detecting *FCoV* in the affected cats.

The FIP electrophoretic pattern is typically characterized by an increase in the alpha $(\alpha)_2$ and the gamma (γ)-globulin fractions. In experimentally induced FIP, α_2-globulin fraction rapidly increases owing to the increased plasma concentration of acute-phase proteins. On the other hand, the γ-globulin fraction increases after about two weeks when clinical signs occur. Abnormal electrophoretic patterns were recently found in 95.1% of cats affected by FIP, the majority of which showed an increased γ-globulin fraction. Serum protein electrophoresis may reveal both polyclonal and monoclonal hypergammaglobulinemia, as well as an increase in acute-phase proteins (13).

Giori, Giordano (14) reported the sensitivity and specificity of SPE for FIP diagnosis as 37.5% and 50%, respectively. In another study, Stranieri, Giordano (15) reported the sensitivity and specificity of SPE as 43% and 90%, respectively. The results of these studies are not consistent with the present results. Thayer, Gogolski (16) reported a sensitivity of 9-77% and a specificity of 88-100% for SPE, which are similar to the present findings.

In the present study, the sensitivity and specificity of SPE were calculated as 70.6% and 100%, respectively. The difference between the results of the present study and that of the others can be attributed to the difference between the gold standards used (immunohistochemistry in the above-mentioned studies

and the merged results of electrophoresis, bioresonance, and PCR in this study).

PCR-based techniques are gaining importance in diagnosing many infectious diseases, and FIP is no exception. Since clear genetic markers for FIPV are still unknown, results should be interpreted with care.

Stranieri, Giordano (15) reported that 3'-UTR PCR had 75% sensitivity and 100% specificity on blood samples. In another study, Felten, Leutenegger (17) reported the sensitivity, negative predictive value (NPV), overall accuracy, and prevalence of the real-time RT-PCR in serum/plasma as 0-23%, 3.8-43.4%, 3.8-43.4%, 82.4%, respectively.

Hartmann, Binder (18) reported the positive predictive value of serum RT-PCR as 0.90%, NPV: 0.47%, specificity: 0.88%, and sensitivity: 0.53%. These results are different from the present findings.

The difference between the current PCR results and others can be attributed to targeting different genomic regions of *FCoV*, different cDNA synthesis kit materials, and different temperature cycling protocols.

Since the 1970s, MORA bioresonance therapy has been globally applied in the context of complementary medicine for various indications. In recent years, bioresonance therapy has proven to be a feasible treatment in several pathologies, used both complementary to classical therapy or independently (8).

Badtieva, Pavlov (19) reported that bioresonance therapy can be considered a method for the correction of the overtraining syndrome in athletes with enhanced activity of the sympathetic nervous system.

According to the findings obtained by Pihtili, Galle (8), bioresonance therapy is clinically effective in smoking cessation and does not show any adverse side effects.

Muresan, Salcudean (20) reported that bioresonance therapy could be useful in the treatment of recurrent major depressive disorders with moderate depressive episodes, independently or as a complementary therapy to antidepressants.

A quasi-experimental study was conducted by Karakos, Grigorios (21) to examine whether bioresonance had an effect on patients' symptoms. The study included 311 patients from doctors and bio-coordinating laboratories in Athens, Thessaloniki, Volos, and Xanthi. The sample of the study included both men (120 subjects, 38.58%) and women (191 subjects, 61.42%) aged 2-76 years. The patients under treatment came forward with symptoms mostly nasal, followed by eye, respiratory, cutaneous, and gastrointestinal symptoms. Most of them (90%) showed no symptoms at all or showed significant improvement in their symptoms after a period of 12 months of bioresonance treatment. They concluded that bioresonance intervention had a significant effect on the improvement of symptoms.

Regarding the results of the present study, the application of bioresonance in diagnosing FIP can be rated as successful. Bioresonance results (AUC of 0.909, compared to 0.853 for electrophoresis and 0.717 for PCR) seem to be promising in FIP diagnosis.

There are few studies related to the effectiveness of bioresonance in veterinary medicine. To the best of the authors' knowledge, this is the first report on the effectiveness of bioresonane in the diagnosis of FIP. According to the findings of this study, bioresonance is a practical and reliable method of diagnosing FIP, which is a challenge in veterinary medicine diagnosis.

Authors' Contribution

Study concept and design: S. S.
Acquisition of data: F. F.
Analysis and interpretation of data: S. H. Sh. B.
Drafting of the manuscript: S. H. Sh. B.
Critical revision of the manuscript for important intellectual content: S. S.
Statistical analysis: F. F.
Administrative, technical, and material support: F. F.

Ethics

The study design was approved by the ethics committee of Islamic Azad University, Tehran, Iran.

Conflict of Interest

The authors declare that they have no conflict of interest.

References

1. Horzinek MC, Osterhaus AD. Feline infectious peritonitis: a worldwide serosurvey. Am J Vet Res. 1979;40(10):1487-92.
2. Kokkinaki K. Study on the epidemiology and the clinical importance of selected infectious, parasitic and metabolic diseases in cats from Greece. Karditsa: Thessaly; 2021.
3. Paltrinieri S, Grieco V, Comazzi S, Cammarata Parodi M. Laboratory profiles in cats with different pathological and immunohistochemical findings due to feline infectious peritonitis (FIP). J Feline Med Surg. 2001;3(3):149-59.
4. Addie D, Belak S, Boucraut-Baralon C, Egberink H, Frymus T, Gruffydd-Jones T, et al. Feline infectious peritonitis. ABCD guidelines on prevention and management. J Feline Med Surg. 2009;11(7):594-604.
5. Pedersen NC. An update on feline infectious peritonitis: virology and immunopathogenesis. Vet J. 2014;201(2):123-32.
6. Sparkes AH, Gruffydd-Jones TJ, Harbour DA. Feline infectious peritonitis: a review of clinicopathological changes in 65 cases, and a critical assessment of their diagnostic value. Vet Rec. 1991;129(10):209-12.
7. Doenges SJ, Weber K, Dorsch R, Fux R, Hartmann K. Comparison of real-time reverse transcriptase polymerase chain reaction of peripheral blood mononuclear cells, serum and cell-free body cavity effusion for the diagnosis of feline infectious peritonitis. J Feline Med Surg. 2017;19(4):344-50.
8. Pihtili A, Galle M, Cuhadaroglu C, Kilicaslan Z, Issever H, Erkan F, et al. Evidence for the efficacy of a bioresonance method in smoking cessation: a pilot study. Forsch Komplementmed. 2014;21(4):239-45.
9. Barbault A, Costa FP, Bottger B, Munden RF, Bomholt F, Kuster N, et al. Amplitude-modulated electromagnetic fields for the treatment of cancer: discovery of tumor-specific frequencies and assessment of a novel therapeutic approach. J Exp Clin Cancer Res. 2009;28(1):51.
10. Costa FP, de Oliveira AC, Meirelles R, Machado MC, Zanesco T, Surjan R, et al. Treatment of advanced hepatocellular carcinoma with very low levels of amplitude-modulated electromagnetic fields. Br J Cancer. 2011;105(5):640-8.
11. Vargas Luna J, Mayr W, Cortes-Ramirez J. Amplitude Modulation Approach for Real-Time Algorithms of ECG-Derived Respiration. Rev Mex Ing Biomed. 2013;35:53-69.
12. Simons FA, Vennema H, Rofina JE, Pol JM, Horzinek MC, Rottier PJ, et al. A mRNA PCR for the diagnosis of feline infectious peritonitis. J Virol Methods. 2005;124(1-2):111-6.
13. Stranieri A, Giordano A, Bo S, Braghiroli C, Paltrinieri S. Frequency of electrophoretic changes consistent with feline infectious peritonitis in two different time periods (2004-2009 vs 2013-2014). J Feline Med Surg. 2017;19(8):880-7.
14. Giori L, Giordano A, Giudice C, Grieco V, Paltrinieri S. Performances of different diagnostic tests for feline infectious peritonitis in challenging clinical cases. J Small Anim Pract. 2011;52(3):152-7.
15. Stranieri A, Giordano A, Paltrinieri S, Giudice C, Cannito V, Lauzi S. Comparison of the performance of laboratory tests in the diagnosis of feline infectious peritonitis. J Vet Diagn Invest. 2018;30(3):459-63.
16. Thayer V, Gogolski S, Felten S, Hartmann K, Kennedy M, Olah GA. 2022 AAFP/EveryCat Feline Infectious Peritonitis Diagnosis Guidelines. J Feline Med Surg. 2022;24(9):905-33.
17. Felten S, Leutenegger CM, Balzer HJ, Pantchev N, Matiasek K, Wess G, et al. Sensitivity and specificity of a real-time reverse transcriptase polymerase chain reaction detecting feline coronavirus mutations in effusion and serum/plasma of cats to diagnose feline infectious peritonitis. BMC Vet Res. 2017;13(1):228.
18. Hartmann K, Binder C, Hirschberger J, Cole D, Reinacher M, Schroo S, et al. Comparison of different tests to diagnose feline infectious peritonitis. J Vet Intern Med. 2003;17(6):781-90.
19. Badtieva V, Pavlov V, Khokhlova M, Pachina A. The application of bioresonance therapy for the correction of the overtrained athlete syndrome. Vopr Kurortol Fizioter Lech Fiz Kult. 2018;95:51.

20. Muresan D, Salcudean A, Sabau DC, Bodo CR, Gabos Grecu I. Bioresonance therapy may treat depression. J Med Life. 2021;14(2):238-42.

21. Karakos P, Grigorios T, Theodoros K, Theodoros L. The Effectiveness of Bioresonance Method on Human Health. Open J Epidemiol. 2019.