Artykuł przeglądowy *Review*

Diagnosis, tests and difficulties in canine and feline inflammatory bowel disease

MICHAŁ WDOWIAK, ANDRZEJ RYCHLIK, RENATA NIERADKA, MAŁGORZATA KANDER, MARCIN NOWICKI, MARTA SZWEDA, ANDRZEJ DEPTA

Department of Clinical Diagnostics, Faculty of Veterinary Medicine, University of Warmia and Mazury, Oczapowskiego 14, 10-719, Olsztyn

Received 07.11.2013 *Accepted 10.02.2014*

Wdowiak M., Rychlik A., Nieradka R., Kander M., Nowicki M., Szweda M., Depta A.

Diagnosis, tests and difficulties in canine and feline inflammatory bowel disease

Summary

Canine and feline inflammatory bowel disease (IBD) is a group of idiopathic alimentary tract disorders diagnosed by the differential diagnosis method. Similarly to humans, the etiopathogenesis of IBD in small animals remains unknown. Blood tests, fecal examinations, intestinal permeability tests, medical imaging, and histopathological screening should be performed to eliminate other diseases and confirm IBD. The required tests prolong the time to diagnosis and increase costs. This article reviews numerous studies and findings relating to IBD in small animals during the diagnostic procedure. Unfortunately, despite a wide range of medical tests, no single diagnostic procedure can confirm the diagnosis. Thus, a clinical analysis should encompass the results of all tests, including the clinical exam, additional tests, histopathological screening, and responses to treatment. Novel biomarkers have been studied, but none of them is useful when applied alone. It seems that the key to finding a specific test or marker lies in the understanding of the etiopathogenesis of IBD.

Keywords: canine, feline, inflammatory bowel disease, diagnostics

Inflammatory bowel disease (IBD) in cats and dogs is a group of idiopathic, chronic disorders of the alimentary tract characterized by permanent or recurrent gastrointestinal (GI) symptoms and inflammation of mucous membranes lining the digestive tract. Inflammations are accompanied by structural changes and mucosal cell infiltration, but the cause of these disorders remains unknown, which is why IBD is classified as an idiopathic disease.

Therefore IBD is a diagnosis of exclusion, where the clinician has to eliminate other primary gastrointestinal disorders mimicking IBD, as well as other systemic diseases that lead to a secondary dysfunction of the alimentary tract and chronic GI symptoms. The diagnostic process is time-consuming, costly, and it requires close co-operation between the owner and the veterinarian. The idiopathic and chronic/recurrent character of the disease makes IBD a "universal" diagnosis in most GI disorders.

Etiology and pathogenesis

The etiology of IBD in small animals remains unknown, but it is presumed to be multifactorial, involving environmental factors (bacterial and food antigens), abnormalities of gastric and intestinal structural and immune dysregulation in genetically susceptible individuals (19). An improvement in medical symptoms after a change in the diet of IBD patients could suggest that dietary factors play a potential role in IBD, while intestinal dysbiosis (qualitative and quantitative changes in intestinal flora) has also been noted and could take part in pathogenesis (57). The composition and number of mucosa-associated bacteria correlate with the presence and severity of inflammatory bowel disease in cats (27). Abnormalities of the intestinal mucosa include quantitative and qualitative mucus changes, increased permeability, regeneration defects in the mucosal barrier function and cell autophagia. A deregulated immune system response is related to the loss of antigen tolerance, increased cell activity and disruption of cell apoptosis.

Genetic factors presumably play a role in the pathogenesis of IBD where the host's immune system misinterprets or over-interprets environmental antigens in the intestinal lumen. Single nucleotide polymorphism (SNP) of toll-like receptors (TLR4 and TLR5) may contribute to the pathogenesis in German Shepherds (34). The influence of genetics has not been confirmed

in cats and dogs, but some breeds (Basenji, Shar-Pei, German Shepherds, Boxers, Rottweilers and Siamese cats) seem to be in a high-risk group.

Idiopathic IBD classification

The classification of IBD is based on the type of cellular infiltration (predominant infiltrating inflammatory cells) and the affected region of the GI tract. Lymphoplasmacytic enteritis (LPE – most common type) and colitis, eosinophilic enteritis and colitis, granulomatous enterocolitis, and histiocytic ulcerative colitis (HUC) in Boxers have been noted in canines (24). Similarly to dogs, the most prevalent disorders in cats are lymphoplasmacytic inflammation, followed by the less frequently observed eosinophilic and purulent gastroenteritis, and neutrophilic and granulomatous types that are very rarely diagnosed. Triaditis, the concurrent presence of IBD, lymphocytic cholangitis, and chronic pancreatitis, has also been noted in feline patients. According to some authors, hepatic involvement is far less common that the concurrent presence of IBD and pancreatitis in feline subjects (4).

Clinical presentation

There are no pathognomonic signs or symptoms of IBD. Emesis and diarrhea are most frequently observed, but other clinical symptoms are also noted (Tab. 1). The clinical presentation is associated with the inflamed region of the GI tract: gastric symptoms are related to the inflammation of the stomach and proximal intestines, whereas diarrhea is caused by inflammations of the large intestine or chronic small intestinal diarrhea. The presence of blood in vomit or stool indicates a more advanced process, and it is frequently observed in eosinophilic inflammations. In colitis, rectal examinations elicit pain. Mild inflammations do not always lead to the loss of appetite, but severe inflammations are accompanied by anorexia. Polyphagia is a rare but possible symptom (12).

Small intestinal diarrhea with vomiting and weight loss is most frequently noted in canines. The mean patient age is 4.9 years (12). Breed predispositions to lymphoplasmacytic inflammation have been noted in Shar-Peis, German Shepherds and Yorkshire Terriers (20). Immunoproliferative enteropathies are diagnosed in Basenjis, whereas HUC affects Boxers. Golden Retrievers, West Highland White Terriers, and Labradors are also more frequently diagnosed with IBD (12).

Felines are more likely to be affected by vomiting than diarrhea. Vomiting is associated with anorexia due to nausea (37), and vomit may contain fur and bile. Some cats cease to defecate in the litter box. In general, middle-aged animals (between 5 to 10 years of age) of both sexes are equally affected, and the mean age of diseased animals is 6.8 years (14). Higher morbidity is noted in Siamese and exotic cats (unconfirmed reports), but breed predisposition has not been proven.

Tab. 1. Clinical symptoms associated with IBD

Vomiting	Hematemesis Bile with/without hair/grass
Small intestinal diarrhea	Watery Large-volume Melena
Large intestinal diarrhea	Hematochezia Mucus in stool High stool frequency Rectal tenesmus
Thickened intestinal loops	
Abdominal pain	
Excessive borborygmus and flatulence	
Change in appetite	Polyphagia Loss of appetite/anorexia Grass eating
Hypoproteinemia/ascites	

No specific abnormalities are found during the physical examination. Some patients are underweight, have thickened intestines and display signs of mesenteric lymphadenopathy, which is more often diagnosed in cats than in dogs. According to some clinicians, intestinal palpation is subjective, and the thickening of the intestines may be attributed to increased turgidity because the difference in the thickness of healthy and afflicted intestines is only 0.5 mm.

Diagnostic procedures

The clinical symptoms of IBD (emesis, diarrhea, weight loss, change of appetite) are linked with the cellular infiltration of the mucosal membrane, inflammation mediators, and enterocyte dysfunction, whereas intestinal contractility disorders are associated with inflammation. IBD is diagnosed on the basis of the following factors (31):

– chronic or recurrent GI symptoms (lasting more than 3 weeks),
– histopathological confirmation of lesions,
– absence of other apparent causes of GI disorders,
– no response to diet change, antibiotic or antiparasitic therapy,
– clinical response to anti-inflammatory or immunosuppressive therapy.

A histopathological evaluation of intestinal biopsies is necessary for a definitive diagnosis. The term „idiopathic IBD" is reserved for cases in which histopathological evidence of inflammation exists, and no other causes can be indicated. Other etiologies, such as infectious and systemic disease, food-responsive diarrhea (FRD), and antibiotic-responsive diarrhea (ARD), have to be ruled out. Therefore, laboratory and imaging tests are required before obtaining a biopsy to rule out other factors, such as anatomical disorders of the intestines (masses, invagination) and non-GI

Tab. 2. Differential diagnostic tests

Laboratory tests	CBC Biochemistry Urinalysis
Fecal examination	Giardia Rectal cytology Salmonella Clostridium perfringens Clostridium difficile E. coli
Empirical deworming Exclusion parasitic diseases	Fenbendazole
Additional blood serum tests	TLI (tripsin-like immunoreactivity) Cortisol concentrations Specific pancreatic lipase immunoreactivity FeLV, FIV, Coronavirus
Medical imaging	X-ray USG Endoscopy
Intestinal biopsy	Endoscopic Surgical
Dietary trials Exclusion FRE	Elimination/hypoallergenic diet Digestible diets for GI disorders High fiber diets (Large intestinal diarrhea)
Antibiotic trials Exclusion ARE	Tylosin Oxyteracycline
Neurological examination	Dysautonomia

diseases (pancreatitis). The most common abnormalities are discussed further in the text, and a summary of additional tests is presented in Table 2.

Hematology. Neutrophilia, with or without a left shift, is sometimes observed (22). Eosinophilia may be indicative of eosinophilic enteritis, but it is not pathognomic and is not always present. Anemia points to chronic inflammation or chronic hemorrhage. Normochromic-normocytic anemia is generally observed (12). Platelet disorders, including thrombocytosis and thrombocytopenia, are also noted. A speculation exists that thrombocytopenia is related to canine IBD due to secondary stimulation of the immune system by bacterial antigens in the intestinal lumen, deregulation of the immune system or both (48). Normal hematological values may be noted in diseased animals.

Serum chemistry. IBD is not associated with pathognomic changes in blood biochemistry, but testing is necessary to exclude other diseases. Hypoproteinemia is observed in protein-losing enteropathy (PLE), whereas hypocholesterolemia is related to malabsorption. A concurrent diagnosis of IBD and hypoproteinemia is associated with poor prognosis (3). Inflammatory processes may also affect the liver, pancreas (28) (particularly in cats – triaditis or tri-tis) and liver enzymes (ALT, ALP – reactive liver), and pancreatic lipase levels may be elevated (59). Hyperglobulinemia may accompany chronic inflammations. In dogs diagnosed with IBD and hypoalbuminemia, lower levels of vitamin D (25-hydroxyvitamin D), ionized hypocalcemia, and high levels of parathyroid hormones have been noted (21). In dogs with lymphoplasmacytic enteritis, higher serum gastrin levels have been observed in comparison with the control group, and they were correlated with the severity of stomach lesions (16). Hypercholesterolemia is reported in cats, and other electrolyte disorders, such as hypocalcemia, hypomagnesemia, and hypophosphatemia (caused by anorexia and malabsorption), may also be present. Feline pancreatic lipase (fPLI – feline pancreatic lipase immunoreactivity) levels were found to be correlated with hypoalbuminemia, and a decrease in serum cobalamin concentrations was also noted. If fPLI $\geq$ 12.0 µg/l, than Hypoalbuminemia and low cobalamin are more frequently diagnosed (6).

Serological markers. The modern diagnostic process of IBD has many disadvantages – it is time-consuming, costly, invasive and characterized by low specificity. Serum biomarkers useful for IBD diagnosis, patient monitoring, and disease outcomes are being studied to reduce these limitations. In small-animal medicine, the main markers in the serological panel are antibodies (pANCA and ACSA) and acute phase proteins (APPs).

Perinuclear antineutrophil cytoplasmic antibodies (pANCA) are a group of autoantibodies that produce a staining pattern around the granulocyte nucleus (9). The antibodies are directed mainly to myeloperoxidase, but they also target alternative antigens such as lactoferrin and elastase. Anti-*Saccharomyces cerevsiae* antibodies (ASCA) respond to mannans, a component of yeast cell walls. Both pANCA and ASCA have diagnostic value in human medicine, and they are used to differentiate Crohn's disease from ulcerative colitis (33).

The pANCA status may be useful for diagnosing canine food-responsive diarrhea (FRD) and IBD. In one study, 62% of dogs with FRD were pANCA-positive, whereas only 23% of the animals affected by IBD were pANCA-positive (38). The sensitivity of pANCA and ASCA assays was determined at 51% and 44%, respectively, and their specificity ranged from 82% to 95% for pANCA, and from 56% to 79% for ASCA (1).

APPs are synthesized in the liver during the acute phase response to local or systemic disturbances of various nature (infectious, neoplastic, rheumatoid disorders, trauma, tissue injury, surgery, pregnancy). The main drawback of APPs is their low specificity, which could limit their applicability for patient monitoring. The usefulness of C-reactive protein (CRP), α1-acid glycoprotein (AGP), serum amyloid A (SAA), and haptoglobin as IBD activity markers in canine patients has been extensively researched.

CRP is a useful marker of intestinal inflammation in IBD when the canine inflammatory bowel disease activity index (CIBDAI) > 5 (31). In another study, no correlations were reported between CRP, CIBDAI or histology scores of intestinal biopsies (41). The main

disadvantage of CRP is its low specificity because elevated CRP levels may accompany other inflammatory states, pregnancy, neoplastic diseases, necrosis or trauma.

Studies investigating the usefulness of AGP in IBD diagnosis often deliver contradictory results. While some researchers reported correlations between AGP, histology and CIBDAI scores, others found no differences between healthy and diseased dogs (10, 31). In canine patients, HAP concentrations were higher before treatment than after therapy. HAP and SAA were found to be poor markers of IBD (31).

Fecal and urine examination. The main purpose of fecal tests is to eliminate parasitic (*Giardia*, helminthes) and bacterial (*Salmonella*, *Campylobacter*, *Clostridium*) intestinal inflammations. The Giardia protozoan parasite is not always identified, and empirical antiparasitic therapy may be considered.

The gold standard in PLE diagnosis is the quantitative estimation of albumin loss demonstrated by radioactive chromium-51 (^{51}Cr). This test has limited use because of technical and safety considerations. The analysis of α1-antitrypsin (α_1-PI) is also an effective indirect quantitative method of evaluating intestinal protein loss in enteropathies (42). IBD is accompanied by a deficiency of α_1-PI and intestinal protein, even before the development of hypoproteinemia.

The usefulness of fecal elastase has been evaluated in canine diarrhea with histopathologically confirmed intestinal inflammation. No statistical differences were found between dogs with diarrhea and intestinal inflammation, dogs with non-inflammatory diarrhea, and healthy dogs (8). An analysis of nitrate and IgG levels in intestinal lavage revealed that these indicators are useful in differentiating healthy dogs from dogs with IBD (23).

New inflammatory markers, including fecal calgranulin C (S100A12), calprotectin (S100A8/S100A9), urinary N-methylhistamine (NMH)(9) and leukotriene E4 (LTE4), have been studied in recent years (26). Calgranulin C and calprotectin belong to the family of calcium-binding proteins that are found in neutrophils, macrophages, and monocytes secreted in various inflammatory states. Medical research involving human subjects revealed higher calgranulin C levels in active than in inactive IBD, as well as a correlation with CRP and SAA. Calgranulin C may also be used to distinguish IBS (irritable bowel syndrome) from IBD (40). Calprotectin is correlated with CRP, CDAI (Crohn's Disease Activity Index), CDEIS (Crohn's Disease Endoscopic Index of Severity) and MDAI (Mayo Disease Activity Index) scores (58). Serum calprotectin may be a useful biomarker in canine IBD, with sensitivity of 82.4% and specificity of 68.4% (25). In dogs with IBD, IgA concentrations were significantly decreased in fecal samples, but no changes were noted in serum IgA levels. These findings suggest that canine IBD is associated with IgA deficiency specifically in the gut. Fecal and serum IgA concentrations were similar in healthy dogs and in dogs with intestinal lymphoma (39).

NMH, a histamine metabolite, is a marker of mast cell degranulation and GI inflammation. An increased excretion of urinary NMH was noted in human IBD, and NMH levels could play a possible role in the monitoring of clinical and endoscopic IBD activity (63). LTE4, a proinflammatory derivative of lipooxygenase, is yet another urinary marker investigated in veterinary medicine, and its concentrations were found to be higher in dogs with IBD than in healthy animals (26).

Non-invasive, indirect intestinal evaluation methods.

– Folic acid and cobalamin. Intestinal absorption influences folic acid and cobalamin levels. A decrease in folic acid concentrations could be indicative of proximal intestinal inflammation, whereas a drop in cobalamin levels could point to an inflammation of distal intestine. A combined decline in both parameters suggests a diffuse disease. Changes in concentration levels are not specific for IBD, but should be included in the patient's therapeutic plan.

When other diseases are considered in a differential diagnosis, a decrease in folic acid levels could suggest food intolerance because the proximal intestines are exposed to food allergens, whereas an increase in the above parameters could indicate small intestinal bacterial overgrowth (SIBO) because folic acid is synthesized by bacteria and absorbed. High levels of cobalamin suggest a high supply of dietary B_{12} or coprophagia.

Cats are highly susceptible to cobalamin deficiency, which has been implicated in IBD, intestinal lymphoma, cholangiohepatitis, cholangitis, pancreatic inflammation or a combination of these diseases. Extremely low serum concentrations of cobalamin have been reported in intestinal lymphoma (54).

– Intestinal permeability. Intestinal permeability to macromolecules is a potential route for antigens, bacterial products, and endogenic proteins normally found in the intestinal lumen across the mucosal membrane. This is significant in the etiopathogenesis of focal and diffuse diseases, such as IBD, or in autoimmune disorders. Dual sugar absorption tests (DSA) evaluate the degree of intestinal damage and the absorption capability of the intestines. The lactulose to ramnose ratio (L/R) is estimated in blood or urine, and the test overlooks problems associated with gastric emptying. Other sugars are also analyzed, including xylose (X), saccharose (S), and methyl glucose (M). This test is useful for diagnosing and monitoring antibiotic-responsive diarrhea (ARD, formerly SIBO), and a lack of improvement with persistent intestinal permeability after antibiotic therapy may indicate IBD as the primary cause of malabsorption (50). In cats, intestinal

permeability is higher than in other species and less susceptible to intestinal flora (32). Differences have also been reported between species and even breeds, subject to environmental, dietary, and sexual factors (47).

The L/R ratio is higher in dogs with LPE than in healthy animals, and significantly higher in dogs with IBD and hypoalbuminemia than in dogs with IBD and normoalbuminemia (36). No correlations were noted between the results of DSA tests and CIBDAI scores, and the L/R, X/M, and S/M ratios cannot be used to monitor IBD (2).

In the test, a sugar solution (lactulose and ramnose, xylose and methyl glucose) is administered orally, and the ratio is evaluated in serum (after 2 hours) or in urine (after 5 hours). A significant difference in the L/R and X/G ratios between a healthy control group and a group consisting of 12 dogs with IBD and 8 dogs with SIBO was noted (55). DSA tests are popular in human medicine on account of their non-invasive character, but their usefulness in veterinary medicine is limited because of urine sampling problems (particularly in cats). Therefore, the serum method is indicated.

In recent studies, iohexol has been evaluated as a potential marker of intestinal permeability. A clear linear association and strong correlations were found between iohexol and ^{51}Cr-EDTA, which suggests that iohexol could be a useful indicator of intestinal permeability (15).

Medical imaging. Medical imaging provides additional information about the severity of disease (focal or diffuse) and its effect on other organs. An ultrasound examination of the biopsy spot supplies information about the thickness of the intestinal wall and the presence of mesenteric lymphadenopathy. X-ray images of the intestines may reveal thickened and gas-filled loops, but the above symptoms often accompany other diseases.

The thickness, echogenicity, structure, and arrangement of the intestinal wall should be evaluated during an ultrasound examination. An increase in wall thickness is a non-specific abnormality. It is generally assumed that abnormal layering of the intestinal wall is more specific for neoplastic diseases than for inflammations. Dogs with abnormal layers of the intestinal wall were 50.9 times more likely to develop tumors than enteritis (46). Granulomatous inflammations can mimic neoplasms in ultrasound exams, and a definitive diagnosis requires biopsy. Full-thickness intestinal biopsy still remains the gold standard for differentiating inflammations from neoplastic diseases (17). An increase in intestinal wall thickness does not confirm IBD, but it eliminates a false positive diagnosis (49). A "grey zone" (observed in both normal physiological and pathological conditions) of 4 to 6 mm was established for dogs, and it is recommended that the maximum thickness of the duodenum (max Ø 6) and the jejunum (max Ø 4,7 mm) is set separately. Similar results were reported in a different study (13). It has also been postulated that villous atrophy in inflammations may contribute to a decrease in thickness.

The mean thickness of the feline intestinal wall is 2.4 mm for the duodenum and 2.1 mm for the jejunum (46). Several intestinal abnormalities have been described in cats with IBD, including weakly differentiated intestinal, focal wall thickening and large mesenteric lymph nodes with hypoechoic changes (5). The thickening of the small intestinal muscularis propria is more likely to occur in cats (particularly in older animals) with alimentary lymphoma (T-cell) than in healthy felines or animals with IBD (65). Lymphadenopathy is associated with lymphoma or IBD, but it is not reported in healthy cats.

The spectral Doppler technique has also been used to diagnose canine chronic enteropathies. Changes in the shape of the spectral line, a decrease in the resistance index (RI), and pulsation index (PI) were noted during digestion in afflicted dogs (18).

Micronodular ultrasound lesions (hypo/anechoic lesions of 1-3 mm in diameter) have been recently described in the colonic submucosa of cats and dogs (11). The results of endoscopic biopsies confirmed lymphocytic/plasmacytic colitis. Micronodular ultrasound lesions could be indicative of colonic inflammatory disease in dogs and cats, but further research is required to determine the sensitivity and specificity of lesions in IBD.

Endoscopic examination and histopathological evaluation of biopsy specimens. Biopsy is required for effective diagnosis of intestinal inflammations. Endoscopy is the simplest, non-invasive method of obtaining bioptates, but it has certain limitations – tissue is sampled only from the surface of the intestines, and the middle sections of the GI tract are not accessible. Full-thickness biopsies may be obtained by laparotomy, but this technique is more invasive and may cause complications in patients with hypoproteinemia.

New endoscopic imaging techniques have been recently introduced, and their utility is still being assessed. Chromoendoscopy that relies on staining techniques (dye-based and virtual staining – NBI, FICE, and i-SCAN), zoom endoscopy (150 × magnification), spectroscopy, endocytoscopy, and capsule endoscopy improve the visibility of mucosal and submucosal structures. These techniques enhance mucosal detail and submucosal vascular patterns, they produce higher-quality biopsies, and minimize the risk of missing lesions (44). The advancements in gastroenterological endoscopy support evaluations of the entire small intestine and produce more comprehensive results than panendoscopy (examination of the proximal and distal duodenum) or retrograde ileocolonoscopy (ileum exam). New modalities, including capsule endoscopy (CE), balloon-assisted endoscopy (BAE), and spiral

endoscopy, support direct examinations of the small bowel and have been shown to be effective in the diagnosis of Crohn's disease in humans. CE is considered a safer, pain-free procedure that does not require anesthesia, and BAE/spiral endoscopy can be used for real-time inspection of the bowel and the collection of biopsy specimens (43, 64). Further work is needed to evaluate the usefulness of these modern techniques, whose popularity is limited because of high cost. For this reason, standard endoscopy with random, multiple biopsies remains the gold standard in IBD diagnosis.

Macroscopic imaging of the stomach and intestines in endoscopy. Endoscopic lesions in IBD are non-specific and sometimes invisible in feline and canine patients. Redness, friability, enhanced granularity, erosion or ulceration, abnormal rigidity and deterioration of submucosal vascularization have been reported (5, 30).

Unfortunately, macroscopic images of the stomach and intestines do not correlate with histopathology results. Dogs with severe macroscopic lesions may not have histopathological changes, and the reverse also applies, in particular in patients with mild and moderate disease activity (51).

Histopathology limitations. The specification of GI inflammations is limited because of the absence of histopathological standards for mucosal biopsies. The type of inflammation is classified based on the predominant type of cells infiltrating the lamina propria (lymphoplasmacytic, eosinophilic, granulomatous). These cell populations may be present in other diseases and combinations. Other limitations of histopathology include variations in the quality of biopsy material (61) and the absence of generally accepted criteria for interpreting lesions. High levels of inconsistency between histopathologists have also been noted, at up to 50% (60, 62). This is a serious problem in routine diagnosis and patient monitoring. Inflammations are also difficult to differentiate from intestinal lymphomas (28). A retrospective study demonstrated that histopathological results from different intestinal segments may vary in the same cat when infiltrative diseases, such as alimentary lymphoma or IBD, are involved (53). Biopsies performed in a single region (duodenum or ileum) may lead to a missed diagnosis. Endoscopic biopsy specimens are regarded as inappropriate for differentiating between IBD and lymphosarcoma in the small intestine. The jejunum and the ileum are the most common sites of alimentary lymphosarcoma in cats, and a full-thickness biopsy specimen should be obtained by laparotomy or laparoscopy (7). A novel diagnostic algorithm has been proposed for differentiating between lymphoma and inflammation in feline small intestinal biopsies. Histomorphologic assessment should be followed by immunophenotyping and PCR to determine the clonality of infiltrating T and B cells (35). A recent study described a significantly higher percentage of anti-apoptotic Bcl-2-positive cells in cats with GI lymphoma than in cats with IBD (56). The overall percentage of Bcl-2-positive cells is high in both diseases, and the significant difference noted in the above study could have greater clinical than diagnostic value.

Tab. 3. FCEAI

Variable	Value
Behavior/activity	0 = normal 1 = mild decrease 2 = moderate decrease 3 = severe decrease
Appetite	0 = normal 1 = mild decrease 2 = moderate decrease 3 = severe decrease
Vomiting	0 = none 1 = mild (1/wk) 2 = moderate (2-3/wk) 3 = severe (> 3/wk)
Diarrhea	0 = well-formed feces; 1 = somewhat soft feces, fecal blood, mucus, or slightly increased stool frequency (2-3 times/d); 2 = very soft feces or moderately increased stool frequency (4-5 times/d); 3 = watery diarrhea or highly increased stool frequency (45 times/d)
Weight loss	0 = none 1 = mild (< 5% loss) 2 = moderate (5-10% loss) 3 = severe (> 10% loss)
Total proteins	0 = norm 1 = increase
ALT	0 = norm 1 = increase
ALP	0 = norm 1 = increase
Phosphorus	0 = norm 1 = decrease
Endoscopic lesions	0 = no 1 = yes

Indices of disease activity

The clinical signs of IBD in canines and felines vary considerably, subject to the affected region and the intensity of the disease. The diversity of clinical symptoms hampers the precise evaluation of anti-inflammatory therapies. IBD is characterized by high individual variability, which makes it difficult to evaluate and compare disease activity between patients.

The canine inflammatory bowel disease activity index (CIBDAI) is a simple scoring system for evaluating the clinical activity of IBD in dogs. The system consists of six fundamental clinical symptoms that are graded by the clinician on a scale of 0 to 3 (0 = normal, 1 = mild change, 2 = moderate change, 3 = severe change). Behavior/activity, appetite, vomiting, stool consistency, stool frequency, and weight loss are evaluated. The six variables are summed to produce the overall CIBDAI score: 0-3 points – clini-

cally insignificant, 4-5 points – mild IBD, 6-8 points – moderate IBD, 9 + points – severe IBD.

CIBDAI is a simple but practical system for evaluating dynamic changes in IBD. CIBDAI is a useful system because: it evaluates all of the major GI symptoms, it relies on symptoms that can be observed only at the clinical level, it accounts for changes in clinical symptoms over time, it is easy to calculate, it correlates with disease activity, it offers prognostic information before and after treatment.

A positive correlation has been reported between CIBDAI and histology scores of the canine small intestinal mucosa (52).

FCEAI (feline chronic enteropathy activity index) is used to assess the degree of enteropathy in cats. Similarly to CIBDAI, FCEAI accounts for clinical symptoms, patient observations, and the results of laboratory tests that reveal changes in the patient's clinical status between visits (29). The cumulative score provides information about the severity of the disease (0-3 clinically insignificant, 4-5 mild IBD, 6-8 moderate IBD, ≥ 9 severe IBD) (Tab. 3).

Conclusions

The clinician needs simple, repeatable, sensitive, and specific tests to effectively diagnose recurrent diseases with diverse and often spontaneously resolved symptoms, such as IBD. None of the diagnostic techniques discussed here fulfills the above description, but they are useful in differential diagnosis and disease/therapy monitoring. Further research is needed to shed more light on the etiopathogenesis of IBD because effective and specific diagnostic methods cannot be developed as long as IBD remains an idiopathic disease.

References

1. *Allenspach K., Luchschander N., Styner M., Seibold F., Doherr M., Aeschbach D., Gaschen F.*: Evaluation of assays for perinuclear antineutrophilic cytoplasmic antibodies and antibodies to Saccharomyces cerevisiae in dogs with inflammatory bowel disease. Am. J. Vet. Res. 2004, 65, 1279-1283.
2. *Allenspach K., Steiner J. M., Shah B. N., Berghoff N., Ruaux C., Williams D. A., Blum J. W., Gaschen F.*: Evaluation of gastrointestinal permeability and mucosal absorptive capacity in dogs with chronic enteropathy. Am. J. Vet. Res. 2006, 67, 479-483.
3. *Allenspach K., Wieland B., Gröne A., Gaschen F.*: Chronic enteropathies in dogs: Evaluation of risk factors for negative outcome. J. Vet. Intern. Med. 2007, 21, 700-708.
4. *Armstrong P. J., Williams D. A.*: Pancreatitis in cats. Top. Companion Anim. Med. 2012, 27, 140-147.
5. *Baez J. L., Hendrick M. J., Walker L. M., Washabau R. J.*: Radiographic, ultrasonographic and endoscopic findings in cats with inflammatory bowel disease of the stomach and small intestine: 33 cases (1990-1997). J. Am. Vet. Med. Assoc. 1999, 215, 349-354.
6. *Bailey S., Benigni L., Eastwood J., Garden O. A., McMahon L., Smith K., Steiner J. M., Suchodolski J. S., Allenspach K.*: Comparisons between cats with normal and increased fPLI concentrations in cats diagnosed with inflammatory bowel disease. J. Small Anim. Pract. 2012, 51, 484-489.
7. *Barrs V., Beatty J.*: Feline alimentary lymphoma: 1. Classification, risk factors, clinical signs and non-invasive diagnostics. J. Feline Med. Surg. 2012, 14, 182-190.
8. *Battersby I. A., Peters I. R., Day M. J., German A. J., Hall E. J.*: Effect of intestinal inflammation on fecal elastase concentration in dogs. Vet. Clin. Path. 2005, 34, 49-51.
9. *Berghoff N., Steiner J. M.*: Laboratory tests for the diagnosis and management of chronic canine and feline enteropathies. Vet. Clin. North Am. Small Anim. Pract. 2011, 41, 311-328.
10. *Cerón J. J., Eckersall P. D., Martinez-Subiela S.*: Acute phase proteins in dogs and cats: current knowledge and future perspectives. Vet. Clin. Path. 2005, 34, 85-99.
11. *Citi S., Chimenti T., Marchetti V., Millanta F., Mannucci T.*: Micronodular ultrasound lesions in the colonic submucosa of 42 dogs and 14 cats. Vet. Radiol. Ultrasound 2013, doi: 10.1111/vru.12077.
12. *Craven M., Simpson W., Ridyard A. E., Chandler M. L.*: Canine inflammatory bowel disease: retrospective analysis of diagnosis and outcome in 80 cases (1995-2002). J. Small Anim. Pract. 2004, 45, 336-342.
13. *Delaney F., O'Brien R. T., Waller K.*: Ultrasound evaluation of small bowel thickness compared to weight in normal dogs. Vet. Radiol. Ultrasoun. 2003, 5, 577-580.
14. *Dennis J. S., Kruger J. M., Mullaney T. P.*: Lymphocytic/plasmacytic gastroenteritis in cats: 14 cases (1985-1990). J. Am. Vet. Med. Assoc. 1992, 200, 1712-1718.
15. *Frias R., Strube K., Ternes W., Collado M. C., Spillman T., Sankari S., Westermarck E.*: Comparison of ^{51}Chromium-labeled ethylenediamine tetraacetic acid and iohexol as blood markers for intestinal permeability testing in Beagle dogs. Vet. J. 2012, 192, 123-125.
16. *García-Sancho M., Rodríquez-Franco F., Sainz A., Rodríguez A., Silván G., Illera J. C.*: Serum gastrin in canine chronic lymphocytic-plasmacytic enteritis. Can. Vet. J. 2005, 46, 630-634.
17. *Gaschen L.*: Ultrasonography of small intestinal inflammatory and neoplastic diseases in dogs and cats. Vet. Clin. North Am. Small. Anim. Pract. 2011, 41, 329-344.
18. *Gaschen L., Kircher P.*: Two-dimensional grayscale ultrasound and spectral Doppler waveform evaluation of dogs with chronic enteropathies. Clin. Tech. Small. Anim. Pract. 2007, 22, 122-127.
19. *German A. J., Hall E. J., Day M. J.*: Chronic intestinal inflammation and intestinal disease in dogs. J. Vet. Intern. Med. 2003, 17, 8-20.
20. *German A. J., Hall E. J., Day M. J.:* Relative deficiency in IgA production by duodenal explants from German Shepherd dogs with small intestinal disease. Vet. Immunol. Immunopathol. 2000, 76, 25-43.
21. *Gow A. G., Else R., Evans H., Berry J. L., Herrtage M. E., Mellanby R. J.*: Hypovitaminosis D in dogs with inflammatory bowel disease and hypoalbuminaemia. J. Small. Anim. Pract. 2011, 52, 411-418.
22. *Guilford W. G., Center S. A., Strombeck D. R., Williams D. A., Meyer D. J.*: Strombeck's small animal gastroenterology. Saunders W. B. company, USA 1996, pp. 469.
23. *Gunawardana S. C., Jergens A. E., Ahrens F. A., Niyo Y.*: Colonic nitrite and immunoglobulin G concentrations in dogs with inflammatory bowel disease. Am. Vet. Med. Assoc. 1997, 211, 318-321.
24. *Hall E. J., Rutgers H. C., Scholes S. F. E., Middleton D. J., Tennant B. J., King N. M., Kelly D. F.*: Histiocytic ulcerative colitis in Boxer dogs in the UK. J. Small Anim. Pract. 1994, 35, 509-515.
25. *Heilmann R. M., Jergens A. E., Ackermann M. R., Barr J. W., Suchodolski J. S., Steiner J. M.*: Serum calprotectin concentrations in dogs with idiopathic inflammatory bowel disease. Am. J. Vet. Res. 2012, 73, 1900-1907.
26. *Im Hof M., Schnyder M., Hartnack S., Stanke-Labesque F., Luckschander N., Burgener I. A.*: Urinary leukotriene E4 concentrations as a potential marker of inflammation in dogs with inflammatory bowel disease. J. Vet. Intern. Med. 2012, 26, 269-274.
27. *Janeczko S., Atwater D., Bogel E., Greiter-Wilke A., Gerold A., Baumgart M., Bender H., McDonough P. L., McDonough S. P., Goldstein R. E., Simpson K. W.*: The relationship of mucosal bacteria to duodenal histopathology, cytokine mRNA, and clinical disease activity in cats with inflammatory bowel disease. Vet. Microbiol. 2008, 128, 178-193.
28. *Jergens A. E.*: Feline idiopathic inflammatory bowel disease: what we know and what remains to be unraveled. J. Feline Med. Surg. 2012, 14, 445-458.
29. *Jergens A. E., Crandell J. M., Evans R., Ackermann M., Miles K. G., Wang C.*: A clinical index for disease activity in cats with chronic enteropathy. J. Vet. Intern. Med. 2010, 24, 1027-1033.
30. *Jergens A. E., Moore F. M., Haynes J. S., Miles K. G.*: Idiopathic inflammatory bowel disease in dogs and cats. 84 cases (1987-1990). J. Am. Vet. Med. Assoc. 1992, 201, 1603-1612.
31. *Jergens A. E., Schreiner C. A., Frank D. E.*: A scoring index for disease activity in canine inflammatory bowel disease. J. Vet. Intern. Med. 2003, 17, 291-297.
32. *Johnston K. L., Ballèvre O. P., Batt R. M.*: Use of an orally administered combined sugar solution to evaluate intestinal absorption and permeability in cats. Am. J. Vet. Res. 2001, 62, 111-118.

33. *Joossens S., Reinisch W., Vermeire S., Sendid B., Poulain D., Peeters M., Geboes K., Bossuyt X., Vandewalle P., Oberhuber G., Vogelsang H., Rutgeerts P., Colombel J. F.*: The value of serologic markers in indeterminate colitis: a prospective follow-up study. Gastroenterology 2002, 122, 1242-1247.
34. *Kathrani A., House A., Catchpole B., Murphy A., German A., Werling D., Allenspach K.*: Polymorphism in the Tlr4 and Tlr5 gene are significantly associated with inflammatory bowel disease in German shepherd dogs. PLoS One 2010, 5: e15740.
35. *Kiupel M., Smedley R. C., Pfent C., Xie Y., Xue Y., Wise A. G., DeVaul J. M., Maes R. K.*: Diagnostic Algorithm to Differentiate Lymphoma From Inflammation in Feline Small Intestinal Biopsy Samples. Vet. Pathol. 2011, 48, 212-222.
36. *Kobayashi S., Ohno K., Uetsuka K., Nakashima K., Setoguchi A., Fujino Y., Tsujimoto H.*: Measurement of intestinal mucosal permeability in dogs with lymphocytic-plasmacytic enteritis. J. Vet. Med. Sci. 2007, 69, 745-749.
37. *Lecoindre P.*: Chronic inflammatory bowel diseases, etiopathogeny, diagnosis. Bull. Acad. Vét. France 2006, 159, 333-342.
38. *Luckschander N., Allenspach K., Hall J., Seibold F., Gröne A., Doherr M. G., Gaschen F.*: Perinuclear antineutrophilic cytoplasmic antibody and response to treatment in diarrheic dogs with food responsive disease or inflammatory bowel disease. J. Vet. Intern. Med. 2006, 20, 221-227.
39. *Maeda S., Ohno K., Uchida K., Nakashima K., Fukushima K., Tsukamoto A., Nakajima M., Fujino Y., Tsujimoto H.*: Decresed immunoglobulin A concentrations in feces, duodenum, and peripheral blood mononuclear cells of dogs with inflammatory bowel disease. J. Vet. Intern. Med. 2013, 27, 47-55.
40. *Manolakis A. C., Kapsoritakis A. N., Georqoulias P., Tzavara C., Valotassiou V., Kapsoritaki A., Potamianos S. P.*: Moderate performance of serum S100A12, in distinguishing inflammatory bowel disease from irritable bowel syndrome. BMC Gastroenterol. 2010, 10, 188.
41. *McCann T. M., Ridyard A. E., Else R. W., Simpson J. W.*: Evaluation of disease activity markers in dogs with idiopathic inflammatory bowel disease. J. Small Anim. Pract. 2007, 48, 620-625.
42. *Melgarajo T., Williams D. W., Asem E. K.*: Enzyme-linked immunosorbent assay for canine α_1-protease inhibitor. Am. J. Vet. Res. 1998, 59, 127-130.
43. *Naganuma M., Hosoe N., Ogata H.*: Inflammatory bowel disease and novel endoscopic technologies. Dig. Endosc. 2013, 26, doi: 10.1111/den.12161.
44. *Neumann H., Neurath M. F., Mudter J.*: New endoscopic approaches in IBD. World J. Gastroenterol. 2011, 17, 63-68.
45. *Newell S. M., Graham J. P., Roberts G. R., Ginn P. E., Harrison J. M.*: Sonography of the normal feline gastrointestinal tract. Vet. Radiol. Ultrasound 1999, 40, 40-43.
46. *Penninck D., Smyers B., Webster C. R. L., Rand W., Moore A. S.*: Diagnostic value of ultrasonography in differentiating enteritis from intestinal neoplasia in dogs. Vet. Radiol. Ultrasound 2003, 44, 570-575.
47. *Randell S. C., Hill R. C., Scott K. C., Omori M., Burrows C. F.*: Intestinal permeability testing using lactulose and rhamnose: a comparison between clinically normal cats and dogs and between dogs of different breeds. Res. Vet. Sci. 2001, 71, 45-49.
48. *Ridgway J., Jergens A. E., Niyo Y.*: Possible causal association of idiopathic inflammatory bowel disease with thrombocytopenia in the dog. J. Am. Hosp. Assoc. 2001, 37, 65-74.
49. *Rudorf H., van Shaik G., O'Brien R. T., Brown P. J., Barr F. J., Hall E. J.*: Ultrasonographic evaluation of the small intestinal wall in dogs with inflammatory bowel disease. J. Small Anim. Pract. 2005, 46, 322-326.
50. *Rutgers H. C., Batt R. M., Proud F. J.*: Intestinal permeability and function in dogs with small intestinal bacterial overgrowth. J. Small. Anim. Pract. 1996, 37, 428-434.
51. *Rychlik A., Nieradka R., Kander M., Depta A., Nowicki M., Sarti K.*: Usefulness of endoscopic examination for the diagnosis of inflammatory bowel disease in the dog. A case report. Pol. J. Vet. Sci. 2007, 10, 113-118.
52. *Rychlik A., Nieradka R., Kander M., Nowicki M., Wdowiak M., Kołodziejska-Sawerska A.*: A correlation between the Canine Inflammatory Bowel Disease Activity Index score and the histopathological evaluation of the small intestinal mucosa in canine inflammatory bowel disease. Pol. J. Vet. Sci. 2012, 15, 315--321.
53. *Scott K. D., Zoran D. L., Mansell J., Norby B., Willard M. D.*: Utility of endoscopic biopsies of the duodenum and ileum for diagnosis of inflammatory bowel disease and small cell lymphoma in cats. J. Vet. Intern. Med. 2011, 25, 1253-1257.
54. *Simpson K. W., Fyfe J., Cornetta A., Sachs A., Strauss-Ayali D., Lamb S. V., Reimers T. J.*: Subnormal Concentrations of Serum Cobalamin (Vitamin B12) in Cats with Gastrointestinal Disease. J. Vet. Intern. Med. 2001, 15, 26-32.
55. *Sørensen S. H., Proud F. J., Rutgers H. C.*: A blood test for intestinal permeability and function: a new tool for the diagnosis of chronic intestinal disease. Clin. Chim. Acta. 1997, 262, 103-115.
56. *Swanson C. M., Smedley R. C., Saavedra P. V., Kiupel M., Kitchell B. E.*: Expression of the Bcl-2 apoptotic marker in cats diagnosed with inflammatory bowel disease and gastrointestinal lymphoma. J. Feline Med. Surg. 2012, 14, 741-745.
57. *Tamboli C. P., Neut C., Desreumaux P., Colombel J. F.*: Dysbiosis in inflammatory bowel disease. Gut 2004, 53, 1-4.
58. *Vieira A., Fang C. B., Rolim E. G., Klug W. A., Steinwurz F., Rossini L. G., Candelária P. A.*: Inflammatory bowel disease activity assessed by fecal calprotectin and lactoferrin: correlation with laboratory parameters, clinical, endoscopic and histopathological indexes. BMC Res Notes. 2009, 2, 221.
59. *Weiss D. J., Gagne J. M., Armstrong P. J.*: Relationship between inflammatory hepatic disease and inflammatory bowel disease, pancreatitis and nephritis in cats. J. Am. Vet. Med. Assoc. 1996, 209, 1114-1116.
60. *Willard M. D., Jergens A. E., Duncan R. B., Leis M. S., McCracken M. D., DeNovo R. C., Heilmann R. C., Slater M. R., Harbison J. L.*: Intraobserver variation among histopathologist evaluation of intestinal tissue from dogs and cats. J. Am. Vet. Med. Assoc. 2002, 220, 1172-1182.
61. *Willard M. D., Lovering S. L., Cohen N. D.*: Quality of tissue specimens obtained endoscopically from duodenum of dogs and cats. J. Am. Vet. Med. Assoc. 2001, 219, 474-479.
62. *Willard M. D., Moore G. E., Denton B. D., Day M. J., Mansell J., Bilzer T., Wilcock B., Gualtieri M., Olivero D., Lecoindre P., Twedt D. C., Collett M. G., Hall E. J., Jergens A. E., Simpson J. W., Else R. W., Washabau R. J.*: Effect of tissue processing on assessment of endoscopic intestinal biopsies in dogs and cats. J. Vet. Intern. Med. 2012, 24, 84-88.
63. *Winterkamp S., Weidenhiller M., Otte P., Stolper J., Schwab D., Hahn E. G., Raithel M.*: Urinary excretion of N-methylhistamine as a marker of disease activity in inflammatory bowel disease. Am. J. Gastroenterol. 2002, 97, 3071--3077.
64. *Yamagami H., Watanabe K., Kamata N., Sogawa M., Arakawa T.*: Small bowel endoscopy in inflammatory bowel disease. Clin. Endosc. 2013, 46, 321-326.
65. *Zwingenberger A. L., Marks S. L., Baker T. W., Moore P. F.*: Ultrasonographic evaluation of the muscularis propria in cats with diffuse small intestinal lymphoma or inflammatory bowel disease. J. Vet. Intern. Med. 2010, 24, 289-292.

Corresponding author: Andrzej Rychlik, Assistant Professor, DVM PhD, ul. Oczapowskiego 14, 10-718 Olsztyn; e-mail: rychlik@uwm.edu.pl