World Journal of Gastroenterology

Online Submissions: http://www.wjgnet.com/1007-9327office
wjg@wjgnet.com
doi:10.3748/wjg.v16.i21.2591

World J Gastroenterol 2010 June 7; 16(21): 2591-2599
ISSN 1007-9327 (print)

TOPIC HIGHLIGHT

Peter L Lakatos, MD, PhD, Assistant Professor, *Series Editor*

Is the disease course predictable in inflammatory bowel diseases?

Peter Laszlo Lakatos, Lajos S Kiss

Peter Laszlo Lakatos, Lajos S Kiss, 1st Department of Medicine, Semmelweis University, H-1083 Budapest, Koranyi S 2A, Hungary
Author contributions: Lakatos PL and Kiss LS contributed equally to this paper.
Correspondence to: Dr. Peter Laszlo Lakatos, MD, PhD, 1st Department of Medicine, Semmelweis University, H-1083 Budapest, Koranyi S 2A, Hungary. kislakpet@bel1.sote.hu
Telephone: +36-1-2100278 Fax: +36-1-3130250
Received: November 21, 2009 Revised: December 16, 2009
Accepted: December 23, 2009
Published online: June 7, 2010

Abstract

During the course of the disease, most patients with Crohn's disease (CD) may eventually develop a stricturing or a perforating complication, and a significant number of patients with both CD and ulcerative colitis will undergo surgery. In recent years, research has focused on the determination of factors important in the prediction of disease course in inflammatory bowel diseases to improve stratification of patients, identify individual patient profiles, including clinical, laboratory and molecular markers, which hopefully will allow physicians to choose the most appropriate management in terms of therapy and intensity of follow-up. This review summarizes the available evidence on clinical, endoscopic variables and biomarkers in the prediction of short and long-term outcome in patients with inflammatory bowel diseases.

Key words: Inflammatory bowel disease; Crohn's disease; Ulcerative colitis; Disease course; Predictive markers; Clinical; Serology; Genetics

Peer reviewer: Cesare Ruffolo, MD, PhD, IV Unit of Surgery, Regional Hospital Cà Foncello, Piazza Ospedale 1, Treviso, 31100, Italy

Lakatos PL, Kiss LS. Is the disease course predictable in inflammatory bowel diseases? *World J Gastroenterol* 2010; 16(21): 2591-2599 Available from: URL: http://www.wjgnet.com/1007-9327/full/v16/i21/2591.htm DOI: http://dx.doi.org/10.3748/wjg.v16.i21.2591

INTRODUCTION

Inflammatory bowel disease (IBD) is a multifactorial disease with probable genetic heterogeneity[1]. In addition, several environmental risk factors (e.g. diet, smoking, measles or appendectomy) may contribute to its pathogenesis. During the past decades, the incidence pattern of various forms of IBD has changed significantly[2], showing some common but also quite distinct characteristics for the different variations of the disorder.

The phenotypic classification of Crohn's disease (CD) plays an important role in determining the treatment, and may assist in predicting the likely clinical course of disease[3]. In 2005, the Montreal revision of the Vienna classification system was introduced[4]. Using the Vienna classification system, it has been shown in clinic-based cohorts that there can be a significant change in disease behavior over time, whereas disease location remains relatively stable[3,5]. Since the early introduction of immunomodulators and/or biologicals might be justified in patients at risk for disease progression, so it is important to identify these patients as soon as possible. Much emphasis has been placed in recent years on the determination of important predictive factors. In this review, the authors try to highlight some of the clinical, endoscopic variables and biomarkers that are important in the prediction of disease course in IBD (Table 1).

CLINICAL, ENVIRONMENTAL, AND ENDOSCOPIC FACTORS

Clinical and endoscopic variables are important medium

Table 1 Possible use of predictors for long and short term disease course in inflammatory bowel diseases

	Marker name	Clinical outcome
Clinical markers	Young age at onset (pediatric or < 40 yr)	Disabling disease
	Small bowel disease	
	Stricturing	
	Perianal disease weight loss > 5 kg	
	Steroid need for the first flare at diagnosis	
	Early immunosupression and/or biological therapy (protective?)	
	Young age at onset (pediatric or < 40 yr)	Surgery
	Small bowel disease	
	Stricturing or penetrating at diagnosis	
	Early immunosupression and/or biological therapy (protective?)	
	Extensive disease	Colectomy in UC
	Younger age at onset (< 50 yr)	
	Smoking (protective)	
Genetic markers	NOD2/CARD15	Disease location/behavior/surgery in CD
	rs1363670 near IL12B	Disease behavior
	TPMT	Azathioprine toxicity
Serology markers	ASCA, glycans	Complicated disease, surgeries
Biomarkers	CRP	Clinical flares
	ESR	
	Calprotectin	
Endoscopy markers	Complete or partial mucosal healing (protective)	Clinical flares and surgery

NOD2/CARD15: Nucleotide oligomerization domain2/caspase activation recruitment domain15; TPMT: Thiopurine S-methyltransferase; ASCA: Anti-*Saccharomyces cerevisiae* antibodies; CRP: C reactive protein; ESR: Erythrocyte sedimentation rate; CD: Crohn's disease; UC: Ulcerative colitis.

and long-term markers in the prediction of complicated disease course in IBD. A young age at onset or pediatric presentation is an important risk factor for developing complicated disease behavior, and even disease extension in a large proportion of patients, as reported in a recent French population based-cohort study[6,7].

In CD, an initial need for steroid use (OR: 3.1, 95% CI: 2.2-4.4), an age below 40 years (OR: 2.1, 95% CI: 1.3-3.6), and the presence of perianal disease (OR: 1.8, 95% CI: 1.2-2.8) were associated with the development of disabling disease in the study by Beaugerie *et al*[8]. The positive predictive value of disabling disease in patients with 2 and 3 predictive factors of disabling disease was 0.91 and 0.93, respectively. More recently, authors from New Zealand[3] have shown that > 70% percent of CD patients had inflammatory disease at diagnosis, with the proportion of patients with complicated disease increasing over time. Progression towards complicated disease was more rapid in those with small bowel than colonic disease location, ($P < 0.001$), and perianal disease was a significant predictor of change in CD behavior [hazard ratio (HR): 1.62, $P < 0.001$]. Similarly, perianal lesions, the need for steroids to treat the first flare and ileocolonic location, but not age below 40 years were confirmed as predictive markers for developing disabling disease (according to the predefined criteria) at 5 years[9]. In the same study, stricturing behaviour (HR: 2.11, 95% CI: 1.39-3.20) and weight loss (> 5 kg) (HR: 1.67, 95% CI: 1.14-2.45) at diagnosis were independently associated with the time to development of severe disease. In addition, small bowel location and stricturing disease were predictors for surgery in a long term follow-up study[10]. Finally, terminal ileal location ($P < 0.001$), stricturing ($P = 0.004$), penetrating behavior ($P < 0.001$), and age younger than 40 years ($P = 0.03$) at diagnosis were independent risk factors for subsequent surgery in a prospective 10-year follow-up study by the IBSEN group[11].

An important environmental factor that may possibly be important in determining changes in disease behavior in CD is smoking. In CD, smoking was reported to be associated with disease location: most, but not all, studies report a higher prevalence of ileal disease and a lower prevalence of colonic involvement in smokers[12,13]. A recent review[13] and previous data have demonstrated that smoking, when measured up to the time-point of disease behavior classification, was associated more frequently with complicated disease, penetrating intestinal complications[12,14,15], and greater likelihood to progress to complicated disease, as defined by the development of strictures or fistulae[13], and a higher relapse rate[16]. In addition, the risk of surgery as well as the risk for further resections during disease course was also noted to be higher in smokers in some studies[12,17] and in a recent meta-analysis[18]. The need for steroids and immunosuppressants was found to be higher in smokers compared to non-smokers[19]. In a recent paper by Aldhous *et al*[20] using the Montreal classification, the harmful effect of smoking was only partially confirmed. Although current smoking was associated with less colonic disease, smoking habits at diagnosis were not associated with time to development of stricturing disease, internal penetrating disease, perianal penetrating disease, or time to first surgery.

In one study by Cosnes *et al*[21], immunosuppressive therapy was found to neutralize the effect of smoking on the need for surgery. The importance of an early aggressive medical strategy in determining long term disease phenotype is further supported by a recent Hungarian study[22,23] where the authors have reported that early aza-

thioprine (AZA)/biological therapy reduced the risk for disease behavior change and delayed the time to the first operation in CD in both smokers and non-smokers. The most convincing data to support a benefit from early use of AZA however, comes from the pediatric literature[24], where in a randomized controlled trial in 55 children, the early use of 6-mercaptopurine (6-MP) was associated with a significantly lower relapse rate (only 9%) compared with 47% in controls ($P = 0.007$). Moreover, the duration of steroid use was shorter ($P < 0.001$) and the cumulative steroid dose was lower at 6, 12, and 18 mo ($P < 0.01$).

In addition, in a recent withdrawal study by the Groupe D'Etude Thérapeutique Des Affections Inflammatoires Du Tube Digestif (GETAID) group[25], authors have provided evidence for a benefit of long term AZA therapy beyond 5 years in patients with prolonged clinical remission. Finally, in a recent controlled randomized prospective trial[26], AZA administered for 12 mo together with metronidazole for 3 mo was more effective in preventing endoscopic postoperative recurrence assessed at 12 mo, compared to metronidazole alone in patients previously only minimally exposed to AZA.

Fewer factors are available in ulcerative colitis (UC). One of the important factors is disease extent, identified by previous studies and also a recent 10 years population-based inception cohort study[27]. In the same study, an age at the time of diagnosis > 50 years was associated with reduced hazard ratio (0.28) for subsequent colectomy. In addition, one-fifth (69/288) of patients with proctitis or left-sided colitis had progressed to extensive colitis. In contrast, pediatric UC was more aggressive. Disease course was characterized by disease extension in 49% of patients during in a 6.5 years population-based cohort study[7]. A delay in diagnosis of more than 6 mo and a family history of IBD were associated with an increased risk of disease extension, with odds ratios of 5.0 (1.2-21.5) and 11.8 (1.3-111.3), respectively. The cumulative rate of colectomy was 8% at 1 year, 15% at 3 years, and 20% at 5 years. Of note, however, surgical rates have shown a wide geographical variation with significantly lower rates reported from the Mediterranean region and Eastern Europe in earlier studies[22,28].

Furthermore, in contrast to CD, smokers with adult UC have been reported to run a more benign disease course compared to non-smokers[29]. Flare-ups, hospitalization rates, the need for oral steroids, and colectomy rates, were reported to be lower, while age at onset was older in smokers compared to non-smokers, though not in all studies. Relapse rates were lower in patients who began smoking after the diagnosis of UC was made. In concordance, in a recent Europe-wide population-based cohort[30], the relapse rate was lower (HR: 0.8, 95% CI: 0.6-0.9) in smokers compared to non-smokers, while being higher in women. In addition, in a meta-analysis of several large series with a total of 1489 UC patients, the risk for colectomy was lower (OR: 0.57, 95% CI: 0.38-0.85) in current smokers compared to non-smokers[31].

Finally, it is still unclear whether mucosal healing prevents the development of complications, although its importance was investigated in several studies using heterogeneous definitions. A well-known example is the study by Allez *et al*[32] where severe endoscopic activity was associated with an increased risk for colectomy in CD after 1, 3 and 8 years of follow-up. This was only partially confirmed in a more recent population-based follow-up study by the IBSEN group, where mucosal healing at 1 year in UC was associated with a lower subsequent colectomy risk, but mucosal healing in CD was not a significant predictor for resective surgery at 5 years, but the risk of future steroid use was decreased[33]. Similarly, in a very recent follow-up report the beneficial effect of early complete mucosal healing was confirmed in early CD. Complete mucosal healing in patients with early-stage CD was associated with significantly higher steroid-free remission rates 4 years after the start of the therapy[34]. Similarly, mucosal healing at 6 mo following the start of scheduled infliximab maintenance therapy was associated with lower need for major abdominal surgery during follow-up in a study from the Leuven group[35].

BIOMARKERS

Biomarkers can be divided into long-term markers (e.g. genetics and serology) influencing the clinical phenotype and short-term markers that may be used for the prediction of flares.

In 2001, the identification of nucleotide oligomerization domain2 (NOD2)/caspase activation recruitment domain15 (CARD15) on chromosome 16 in the IBD1 region, as a candidate gene for CD[36,37], has stimulated further research focusing on novel genetic determinants of susceptibility and phenotype in IBD. The presence of a NOD2/CARD15 mutation increases the risk for CD by 1.4-4.3-fold in heterozygous patients and 17.6-44.0-fold in homozygous and compound heterozygous patients. Of importance, reports exist of homozygous individuals who are disease-free. However, various geographical differences were noted. A much lower prevalence of these mutations was reported in, for example African American, Chinese and Japanese[38] populations. NOD2/CARD15 is a good example for case-control phenotype-genotype correlations, since in earlier studies the three common NOD2/CARD15 mutations were associated with ileal disease and fibrostenosing behavior. On the contrary, in colonic and fistulizing disease, they were relatively less frequent[39,40]. There are many controversial findings, such as the association between the OCTN TC risk haplotype and penetrating disease (OR: 1.474, 95% CI: 1.028-2.114, $P = 0.035$) in one study[41] or non-fistulizing non-fibrostenotic disease (OR: 1.57), colonic disease (OR: 1.31), an earlier age at the onset of disease, and reduced need for surgery (OR: 1.38) in another[42], or the association between the presence of DLG5 R30Q variant and steroid refractory disease that could not be confirmed yet[43]. Finally, in a recent study by Weersma *et al*[44] the same type of statistical analysis was performed in a little more complex genetic dataset by using both novel markers (e.g. ATG16L1,

IL23R) and earlier (NOD2, IBD5 and DLG5) genetic markers. In this study, patients with CD with a more severe disease course, surgeries or an age of onset below 40 years had more risk alleles compared to non-stricturing, nonpenetrating behaviour (P = 0.0008), no operations (P = 0.02) or age at onset greater then 40 years (P = 0.028). However, the association became insignificant after a longer disease duration, confirming that many other factors (e.g. clinical variables, disease phenotype at diagnosis, medical therapy) contribute to the long term evolution of disease phenotype.

Although in some of these studies the authors corrected their findings for disease duration using logistic regression, these are all static correlations between phenotype and genotype. In 2009, we finally observed new examples of genotype-phenotype association scans, where the authors have performed a complete follow-up of the patients and so the association between time-dependent clinical outcomes and genotype could be more preciously evaluated in multivariate analysis using Cox regression. Moreover, the authors realized that clinical parameters should also be included in the complex testing. A fine example of how this should be performed was recently published by the Leuven group[45]. Of course, the presentation of the results is now much more complex. For example, homozygosity for the rs1363670 G-allele in a gene encoding a hypothetical protein near the *IL12B* gene was independently associated with stricturing disease behavior and with shorter time to strictures, especially in patients with ileal involvement, or alternatively male patients carrying at least one rs12704036 T-allele in a gene desert had the shortest time to non-perianal fistula. This landmark study with its complex analysis is, however, bringing genetics closer to being used in routine clinical practice.

Genetic markers may also be used to predict the efficacy and/or side effects of medical therapy, that is pharmacogenetics. This use is also expected to be complex and include certain clinical predictors. The importance of using conventional genetic testing might be highlighted by the well-known thiopurine S-methyltransferase (TPMT) genotype/activity testing. TPMT limits the production of 6-TGNs by converting 6-MP to 6-thioruric acid and 6-methylmercaptopurine[46], which may lead to myelotoxicity. Population studies have shown that the distribution of TPMT activity is trimodal: 0.3%-0.5% of the population has low to absent activity (TPMTL/TPMTL), around 10% have intermediate activity (TPMTL/TPMTH), and approximately 90% inherit normal to high enzyme activity (TPMTH/TPMTH)[46]. In this regard, a correlation between the TPMT genotype and enzyme activity has been proven. Approximately 5% of the white population carries one or more variant TPMT alleles, with more than ten variant alleles reported[47]. The functional consequences of alleles *2, *2A, *3B, and *3C, accounting together for more than 90% of mutant alleles, have been extensively characterized. Nevertheless, it is clear that there are many other causes of myelotoxicity. This was accurately demonstrated by Colombel *et al*[48], who found that only 27% of patients had a documented low TPMT activity. Other confounding genetic and environmental factors include, for instance, the patient's age, renal function, AZA formulation, coadministration of mesalazine (a reversible TPMT inhibitor) and allopurinol (XO inhibitor). Thus, the determination of TPMT activity is not an exclusive test to rely on. It may only be helpful in identifying a certain group of high risk patients but as the negative predictive value is rather low, it is not beneficial in ruling out possible side effects. Also, as the prevalence of double carriage of variant TPMT alleles is as low as 1/300, continuous monitoring of red blood cell counts remains mandatory in clinical practice. A different approach was recently reported by the Leuven group[49]. The authors analysed gene expression signatures in the mucosa of patients with UC and were able to identify predictive panels of genes for (non-)response to infliximab. Further studies of the pathways involved should allow a better understanding of the mechanisms of resistance to infliximab therapy in UC.

Similarly, serological markers represent medium to long term markers of disease phenotype and progression. The phenotype and disease course of CD are associated with the presence and extent of the serologic response targeted against various microbial antigens. In patients with conventional anti-*Saccharomyces cerevisiae* antibodies (ASCA$^+$, IgG and/or IgA) or atypical perinuclear antineutrophil cytoplasmic antibodies (P-ANCA) phenotype, small bowel involvement (with or without colonic disease) is more typical than the pure colonic disease (60%-80% *vs* 30%-45%)[50,51]. ASCA positivity predicts a more aggressive disease course with a higher rate of complications. ASCAs have been associated with stricturing and penetrating disease and a higher risk of small bowel resection[51,52]. Some studies also suggest that ASCA positivity is associated with an earlier onset of disease. The presence of anti-OmpC in adult CD patients is associated with an increased prevalence of the penetrating form only[53], while in children, both the penetrating and stenosing forms[54] are more frequent. Moreover, antibody positivity may lead to a more aggressive course of disease and a higher risk for surgical interventions. Like ASCA and anti-OmpC, anti-I2 also appears to be associated with an increased risk for complications in adult CD patients. It is an independent risk factor for the development of the stenosing form and the need for surgical interventions[54]. Recent research has shown that the anti-CBir1 antibody is associated with ileal involvement in adult CD patients independently from other serological markers, and it predisposes for the development of both stenosing and penetrating forms[55]. Interestingly anti-Cbir1 was more frequent in young children with CD (age: 0-7 years)[56]. In a very recent study, an association was also reported between the presence of pancreatic autoantibodies and perforating perianal disease, arthritis, ocular, and cutaneous manifestations[57]. In addition to qualitative correlations, quantitative correlations with serological responses are also present. The number of antibodies produced against microbial anti-

gens in CD also showed a positive correlation with the severity of the disease course[51].

Similarly, the presence and magnitude of the more newly developed antiglycan antibodies (including the most recent anti-laminarin and anti-chitin markers) in CD was associated with a more aggressive disease phenotype in CD, as well as a younger age at onset, ileal involvement, stricturing/penetrating disease behavior, and risk for surgery[58-60]. In the latter study, gASCA IgG and ASCA IgG were comparable for the diagnosis and determination of disease phenotype in CD; however, each test recognized an additional patient population that was missed by the other, indicating a partially complementary role. The differentiation between colon only CD and UC was also better[58,61].

In prospective pediatric cohorts Dubinsky *et al*[54], found that the presence and magnitude of immune responses to microbial antigens (ASCA, anti-OmpC, anti-I2 and anti-CBir1) are significantly associated with more aggressive disease phenotype. The risk of developing penetrating and/or stricturing CD was increased in those individuals with immune response against all four microbial antigens compared to seronegative cases. Moreover, they demonstrated that the time to develop a disease complication during the 18 mo of follow-up period is significantly faster in those children who have a serologic response against at least one antigen, recently confirmed by the same group in a much larger IBD population[62].

In addition, the occurrence of atypical P-ANCA in UC is associated with a characteristic clinical appearance and represents a distinct subgroup, which is often characterized by specific human leucocyte antigen markers. These patients have a higher probability of developing a severe left-sided UC that is more resistant to treatment than the usual case. The disease has a more aggressive course requiring surgery earlier in the course of the disease[63]. The presence of atypical P-ANCA identifies a subgroup of CD patients characterized by "UC-like" colitis; the inflammation usually involves the left side of the colon and the response to therapy is generally good. The atypical P-ANCA in CD patients is associated with a later age of onset and a relatively smaller incidence of complications, such as stricture and/or perforation[64].

Of note however, one should be aware of the fact that the serology profile is not a stable phenotype; seroreactivity may change during the course of follow-up. This is at least suggested by studies done in patients with celiac disease where seropositivity was lost several months after the introduction of a gluten free diet[65].

IBD follows an alternating disease course and both CD and UC are characterized by periods of remission and relapse. However, disease flare-ups occur in a random way and are often unpredictable. Based on the half-life and changes over time serum laboratory and fecal markers may be best used for the prediction of short or medium term relapses. Erythrocyte sedimentation rate (ESR) and C reactive protein (CRP) are traditional non-specific markers of inflammation. In addition, a number of neutrophil-derived proteins present in stool samples have been studied, including fecal lactoferrin, lysozyme, elastase, myeloperoxidase, and calprotectin.

ESR is defined as the rate at which erythrocytes migrate through the plasma. Inevitably, ESR will depend on the plasma concentration and on the number and size of the erythrocytes. Conditions such as anemia, polycythemia, and thalassemia affect ESR. Compared with CRP, ESR will peak much less rapidly and may also take several days to decrease, even if the patient's clinical condition or the inflammation is ameliorated. Although CRP is non-specifically upregulated in most inflammatory diseases, including IBD, there is remarkable heterogeneity in the CRP response between CD and UC. Whereas CD is associated with a strong CRP response, UC has only a modest or no CRP response in both adult and children IBD populations[66]. This is an important fact to keep in mind when using CRP in clinical practice. There is no good explanation for this heterogeneity.

In general, patients with severe disease more often have abnormal inflammatory markers, compared with patients without or with only a low-grade inflammation. This has been shown in one of the early studies by Fagan *et al*[67], both CRP and ESR correlated well with disease activity, yet the correlation was better for CRP. In addition, the correlation of laboratory markers with disease activity has been shown to be much stronger for CD than for UC. However, a wide range of CRP values was observed and no clear cut-off values exist between mild to moderate (10-50 mg/L), moderate to severe (50-80 mg/L), and severe disease (> 80 mg/L). Therefore, the comparison of individual CRP values with previous values in any given patient is of great clinical importance. In addition, CRP showed acceptable correlation with endoscopic and histological activity in CD[68]. For UC, again, this correlation was weaker. In contrast, more recent data from Australia suggested that a subgroup of patients with active ileal disease, low body mass index, and previous ileocecal resection was characterized by low CRP[69].

In CD, a number of studies have investigated a panel of laboratory markers in predicting clinical relapse. Later, Boirivant *et al*[70] prospectively followed 101 outpatients with CD. Half of the patients had an elevated CRP value and this correlated well with clinical activity. Approximately one-third of CD patients presented with active disease despite normal CRP and one-third had high CRP levels but clinically inactive disease. The likelihood of relapse after 2 years was higher in the patients with an increased CRP compared with those who had a normal CRP value. More recently, the GETAID group[71] proposed a simple biological score for predicting short-term relapse in CD. Multivariate analysis selected two markers predictive of relapse: CRP > 20 mg/L and an ESR > 15 mm/h. The relative risk of short-term relapse for patients with a positive score compared to those with a negative score was 8.0 (95% CI: 2.8-22.9). The score's sensitivity and specificity was 89% and 43%, respectively, with a negative predictive value of 97%, suggesting that normal CRP and ESR could almost certainly rule out a relapse in the next 6 wk. Much less data exist on the value of laboratory mark-

ers in assessing disease course and outcome in UC. The well known, prospective study from Oxford evaluated 49 severe UC patients treated with hydrocortisone and/or cyclosporine (n = 49). On day 3, a daily stool frequency of > 8 or 3-8 with increased CRP (> 45 mg/L) predicted with 85% certainty the need for colectomy[72]. Only one relatively small study investigated the role of hs-CRP in IBD[73]. Ninety CD, 70 UC patients, and 160 controls were investigated. The coefficient of correlation between hs-CRP and the disease activity score was weak but similar in both UC (0.26) and CD (0.36).

Finally, a recent population based study by the IBSEN group has demonstrated[74] that CRP at diagnosis or during follow-up was able to predict medium term outcome as measured by the need for colectomy or resective surgery. In patients with UC with extensive colitis, CRP levels above 23 mg/L at diagnosis predicted an increased risk of surgery (OR: 4.8, 95% CI: 1.5-15.1) during the first 5 years of follow-up. In patients with UC, CRP levels above 10 mg/L after one year predicted an increased risk of surgery during the subsequent 4 years (OR 3.0, 95% CI: 1.1-7.8). A significant association between CRP levels at diagnosis and risk of surgery was also found in patients with CD with terminal ileitis (L1), where the risk increased when CRP levels were above 53 mg/L in this subgroup (OR 6.0, 95% CI: 1.1-31.9).

A change in CRP following therapy serves as a good parameter in assessing the effectiveness of the drug on the underlying inflammation. A decrease in CRP, in response to therapy, is objective evidence that the drug has a beneficial effect on intestinal inflammation, even in patients with small changes in their symptoms. On the other hand, persistently raised CRP indicates therapeutic failure in controlling mucosal inflammation. This was clearly demonstrated by the different response rates to anti-TNF antibodies in patients with CD. In some studies, a high baseline (5-10 mg/L) CRP value before the start of therapy was associated with a higher response compared to patients with lower CRP[75,76]. These findings raise the question of whether CRP should be included in patient selection for future clinical trials, at least for selected drugs. Furthermore, high CRP (> 20 mg/L) was found to be an independent predictor for relapse after AZA withdrawal in patients on AZA therapy for longer than 42 and 63 mo[25,77].

Fecal calprotectin was also shown to predict relapse in CD. Calprotectin, a 36 kDa calcium- and zinc-binding protein, is probably the most promising marker for various reasons. In contrast with other neutrophil markers, calprotectin represents 60% of cytosolic proteins in granulocytes. The presence of calprotectin in feces can therefore be seen as directly proportional to neutrophil migration to the gastrointestinal tract. Fecal calprotectin is a very stable marker (stable for more than 1 wk at room temperature) and is resistant to degradation, making it an attractive option. Other neutrophil protein markers, include fecal lactoferrin, polymorphonuclear-elastase, and S100 A12. They are also more specific to intestinal rather than systemic inflammation, but their clinical role has been less extensively evaluated in IBD.

In the study by Tibble *et al*[78], calprotectin levels of 50 μg/g or more predicted a 13-fold increased risk for relapse. In another study[79], 38 CD and 41 UC patients were investigated. All patients were in remission for a mean duration of 5 mo. A baseline level of calprotectin of 150 μg/g or more was predictive for a relapse in the next year. Although sensitivity was high for both CD (87%) and UC (89%), specificity was much lower in the case of CD (43%) compared to UC (82%). Similarly, in a recent study, fecal calprotectin and lactoferrin were able to differentiate organic colorectal disease and were associated with active disease at endoscopy, both in UC (78% and 75%, respectively) and in CD (87% and 82%, respectively)[80]; however, specificity was low overall. It is thus difficult to define cut-off values based on these studies. In addition to serum markers, fecal calprotectin also correlates well with endoscopic and histological activity in patients with UC and CD, and elevated calprotectin levels normalize once the inflammation has resolved in both adults and children[81,82].

Lastly, calprotectin, at a cut-off value of 200 mg/L, at 3 mo post-surgery, was useful in predicting endoscopic post-surgical recurrence in asymptomatic patients with a sensitivity of 63% and a specificity of 75%[83].

CONCLUSION

In conclusion, the search for clinical, laboratory, and molecular markers to define and predict disease outcome in common diseases has moved rapidly forward with the help of modern technologies and vigorous collection of clinical data. However, many questions remain unanswered. Nevertheless, research should be encouraged because, as physicians treating patients with IBD, our ultimate goal is to have at our disposal a complex individual patient profile, including clinical, laboratory and molecular markers around the time of diagnosis which will hopefully allow us to choose the most appropriate management in terms of therapy, intensity of follow-up and frequency of various investigations.

REFERENCES

1 **Lakatos PL**, Fischer S, Lakatos L, Gal I, Papp J. Current concept on the pathogenesis of inflammatory bowel disease-crosstalk between genetic and microbial factors: pathogenic bacteria and altered bacterial sensing or changes in mucosal integrity take "toll" ? *World J Gastroenterol* 2006; **12**: 1829-1841
2 **Lakatos PL**. Recent trends in the epidemiology of inflammatory bowel diseases: up or down? *World J Gastroenterol* 2006; **12**: 6102-6108
3 **Tarrant KM**, Barclay ML, Frampton CM, Gearry RB. Perianal disease predicts changes in Crohn's disease phenotype-results of a population-based study of inflammatory bowel disease phenotype. *Am J Gastroenterol* 2008; **103**: 3082-3093
4 **Silverberg MS**, Satsangi J, Ahmad T, Arnott ID, Bernstein CN, Brant SR, Caprilli R, Colombel JF, Gasche C, Geboes K, Jewell DP, Karban A, Loftus Jr EV, Peña AS, Riddell RH, Sachar DB, Schreiber S, Steinhart AH, Targan SR, Vermeire S, Warren BF. Toward an integrated clinical, molecular and serological classification of inflammatory bowel disease:

Report of a Working Party of the 2005 Montreal World Congress of Gastroenterology. *Can J Gastroenterol* 2005; **19** Suppl A: 5-36
5 **Louis E**, Collard A, Oger AF, Degroote E, Aboul Nasr El Yafi FA, Belaiche J. Behaviour of Crohn's disease according to the Vienna classification: changing pattern over the course of the disease. *Gut* 2001; **49**: 777-782
6 **Vernier-Massouille G**, Balde M, Salleron J, Turck D, Dupas JL, Mouterde O, Merle V, Salomez JL, Branche J, Marti R, Lerebours E, Cortot A, Gower-Rousseau C, Colombel JF. Natural history of pediatric Crohn's disease: a population-based cohort study. *Gastroenterology* 2008; **135**: 1106-1113
7 **Gower-Rousseau C**, Dauchet L, Vernier-Massouille G, Tilloy E, Brazier F, Merle V, Dupas JL, Savoye G, Baldé M, Marti R, Lerebours E, Cortot A, Salomez JL, Turck D, Colombel JF. The natural history of pediatric ulcerative colitis: a population-based cohort study. *Am J Gastroenterol* 2009; **104**: 2080-2088
8 **Beaugerie L**, Seksik P, Nion-Larmurier I, Gendre JP, Cosnes J. Predictors of Crohn's disease. *Gastroenterology* 2006; **130**: 650-656
9 **Loly C**, Belaiche J, Louis E. Predictors of severe Crohn's disease. *Scand J Gastroenterol* 2008; **43**: 948-954
10 **Romberg-Camps MJ**, Dagnelie PC, Kester AD, Hesselink-van de Kruijs MA, Cilissen M, Engels LG, Van Deursen C, Hameeteman WH, Wolters FL, Russel MG, Stockbrügger RW. Influence of phenotype at diagnosis and of other potential prognostic factors on the course of inflammatory bowel disease. *Am J Gastroenterol* 2009; **104**: 371-383
11 **Solberg IC**, Vatn MH, Høie O, Stray N, Sauar J, Jahnsen J, Moum B, Lygren I. Clinical course in Crohn's disease: results of a Norwegian population-based ten-year follow-up study. *Clin Gastroenterol Hepatol* 2007; **5**: 1430-1438
12 **Lindberg E**, Järnerot G, Huitfeldt B. Smoking in Crohn's disease: effect on localisation and clinical course. *Gut* 1992; **33**: 779-782
13 **Mahid SS**, Minor KS, Stevens PL, Galandiuk S. The role of smoking in Crohn's disease as defined by clinical variables. *Dig Dis Sci* 2007; **52**: 2897-2903
14 **Picco MF**, Bayless TM. Tobacco consumption and disease duration are associated with fistulizing and stricturing behaviors in the first 8 years of Crohn's disease. *Am J Gastroenterol* 2003; **98**: 363-368
15 **Louis E**, Michel V, Hugot JP, Reenaers C, Fontaine F, Delforge M, El Yafi F, Colombel JF, Belaiche J. Early development of stricturing or penetrating pattern in Crohn's disease is influenced by disease location, number of flares, and smoking but not by NOD2/CARD15 genotype. *Gut* 2003; **52**: 552-557
16 **Cosnes J**, Carbonnel F, Carrat F, Beaugerie L, Cattan S, Gendre J. Effects of current and former cigarette smoking on the clinical course of Crohn's disease. *Aliment Pharmacol Ther* 1999; **13**: 1403-1411
17 **Sutherland LR**, Ramcharan S, Bryant H, Fick G. Effect of cigarette smoking on recurrence of Crohn's disease. *Gastroenterology* 1990; **98**: 1123-1128
18 **Reese GE**, Nanidis T, Borysiewicz C, Yamamoto T, Orchard T, Tekkis PP. The effect of smoking after surgery for Crohn's disease: a meta-analysis of observational studies. *Int J Colorectal Dis* 2008; **23**: 1213-1221
19 **Johnson GJ**, Cosnes J, Mansfield JC. Review article: smoking cessation as primary therapy to modify the course of Crohn's disease. *Aliment Pharmacol Ther* 2005; **21**: 921-931
20 **Aldhous MC**, Drummond HE, Anderson N, Smith LA, Arnott ID, Satsangi J. Does cigarette smoking influence the phenotype of Crohn's disease? Analysis using the Montreal classification. *Am J Gastroenterol* 2007; **102**: 577-588
21 **Cosnes J**, Carbonnel F, Beaugerie L, Le Quintrec Y, Gendre JP. Effects of cigarette smoking on the long-term course of Crohn's disease. *Gastroenterology* 1996; **110**: 424-431
22 **Szamosi T**, Banai J, Lakatos L, Czegledi Z, David G, Zsigmond F, Pandur T, Erdelyi Z, Gemela O, Papp M, Papp J, Lakatos PL. Early azathioprine/biological therapy is associated with decreased risk for first surgery and delays time to surgery but not reoperation in both smokers and nonsmokers with Crohn's disease, while smoking decreases the risk of colectomy in ulcerative colitis. *Eur J Gastroenterol Hepatol* 2009; Epub ahead of print
23 **Lakatos PL**, Czegledi Z, Szamosi T, Banai J, David G, Zsigmond F, Pandur T, Erdelyi Z, Gemela O, Papp J, Lakatos L. Perianal disease, small bowel disease, smoking, prior steroid or early azathioprine/biological therapy are predictors of disease behavior change in patients with Crohn's disease. *World J Gastroenterol* 2009; **15**: 3504-3510
24 **Markowitz J**, Grancher K, Kohn N, Lesser M, Daum F. A multicenter trial of 6-mercaptopurine and prednisone in children with newly diagnosed Crohn's disease. *Gastroenterology* 2000; **119**: 895-902
25 **Treton X**, Bouhnik Y, Mary JY, Colombel JF, Duclos B, Soule JC, Lerebours E, Cosnes J, Lemann M. Azathioprine withdrawal in patients with Crohn's disease maintained on prolonged remission: a high risk of relapse. *Clin Gastroenterol Hepatol* 2009; **7**: 80-85
26 **D'Haens GR**, Vermeire S, Van Assche G, Noman M, Aerden I, Van Olmen G, Rutgeerts P. Therapy of metronidazole with azathioprine to prevent postoperative recurrence of Crohn's disease: a controlled randomized trial. *Gastroenterology* 2008; **135**: 1123-1129
27 **Solberg IC**, Lygren I, Jahnsen J, Aadland E, Høie O, Cvancarova M, Bernklev T, Henriksen M, Sauar J, Vatn MH, Moum B. Clinical course during the first 10 years of ulcerative colitis: results from a population-based inception cohort (IBSEN Study). *Scand J Gastroenterol* 2009; **44**: 431-440
28 **Hoie O**, Wolters FL, Riis L, Bernklev T, Aamodt G, Clofent J, Tsianos E, Beltrami M, Odes S, Munkholm P, Vatn M, Stockbrügger RW, Moum B. Low colectomy rates in ulcerative colitis in an unselected European cohort followed for 10 years. *Gastroenterology* 2007; **132**: 507-515
29 **Lakatos PL**, Szamosi T, Lakatos L. Smoking in inflammatory bowel diseases: good, bad or ugly? *World J Gastroenterol* 2007; **13**: 6134-6139
30 **Höie O**, Wolters F, Riis L, Aamodt G, Solberg C, Bernklev T, Odes S, Mouzas IA, Beltrami M, Langholz E, Stockbrügger R, Vatn M, Moum B. Ulcerative colitis: patient characteristics may predict 10-yr disease recurrence in a European-wide population-based cohort. *Am J Gastroenterol* 2007; **102**: 1692-1701
31 **Cosnes J**. Tobacco and IBD: relevance in the understanding of disease mechanisms and clinical practice. *Best Pract Res Clin Gastroenterol* 2004; **18**: 481-496
32 **Allez M**, Lemann M, Bonnet J, Cattan P, Jian R, Modigliani R. Long term outcome of patients with active Crohn's disease exhibiting extensive and deep ulcerations at colonoscopy. *Am J Gastroenterol* 2002; **97**: 947-953
33 **Frøslie KF**, Jahnsen J, Moum BA, Vatn MH. Mucosal healing in inflammatory bowel disease: results from a Norwegian population-based cohort. *Gastroenterology* 2007; **133**: 412-422
34 **Baert F**, Moortgat L, Van Assche G, Caenepeel P, Vergauwe P, De Vos M, Stokkers P, Hommes D, Rutgeerts P, Vermeire S, D'Haens G. Mucosal healing predicts sustained clinical remission in patients with early-stage Crohn's disease. *Gastroenterology* 2010; **138**: 463-468; quiz e10-e11
35 **Schnitzler F**, Fidder H, Ferrante M, Noman M, Arijs I, Van Assche G, Hoffman I, Van Steen K, Vermeire S, Rutgeerts P. Mucosal healing predicts long-term outcome of maintenance therapy with infliximab in Crohn's disease. *Inflamm Bowel Dis* 2009; **15**: 1295-1301
36 **Ogura Y**, Bonen DK, Inohara N, Nicolae DL, Chen FF, Ramos R, Britton H, Moran T, Karaliuskas R, Duerr RH, Achkar JP, Brant SR, Bayless TM, Kirschner BS, Hanauer SB, Nuñez

G, Cho JH. A frameshift mutation in NOD2 associated with susceptibility to Crohn's disease. *Nature* 2001; **411**: 603-606
37 **Hampe J**, Cuthbert A, Croucher PJ, Mirza MM, Mascheretti S, Fisher S, Frenzel H, King K, Hasselmeyer A, MacPherson AJ, Bridger S, van Deventer S, Forbes A, Nikolaus S, Lennard-Jones JE, Foelsch UR, Krawczak M, Lewis C, Schreiber S, Mathew CG. Association between insertion mutation in NOD2 gene and Crohn's disease in German and British populations. *Lancet* 2001; **357**: 1925-1928
38 **Inoue N**, Tamura K, Kinouchi Y, Fukuda Y, Takahashi S, Ogura Y, Inohara N, Núñez G, Kishi Y, Koike Y, Shimosegawa T, Shimoyama T, Hibi T. Lack of common NOD2 variants in Japanese patients with Crohn's disease. *Gastroenterology* 2002; **123**: 86-91
39 **Lakatos PL**, Lakatos L, Szalay F, Willheim-Polli C, Osterreicher C, Tulassay Z, Molnar T, Reinisch W, Papp J, Mozsik G, Ferenci P. Toll-like receptor 4 and NOD2/CARD15 mutations in Hungarian patients with Crohn's disease: phenotype-genotype correlations. *World J Gastroenterol* 2005; **11**: 1489-1495
40 **Ahmad T**, Armuzzi A, Bunce M, Mulcahy-Hawes K, Marshall SE, Orchard TR, Crawshaw J, Large O, de Silva A, Cook JT, Barnardo M, Cullen S, Welsh KI, Jewell DP. The molecular classification of the clinical manifestations of Crohn's disease. *Gastroenterology* 2002; **122**: 854-866
41 **Vermeire S**, Pierik M, Hlavaty T, Claessens G, van Schuerbeeck N, Joossens S, Ferrante M, Henckaerts L, Bueno de Mesquita M, Vlietinck R, Rutgeerts P. Association of organic cation transporter risk haplotype with perianal penetrating Crohn's disease but not with susceptibility to IBD. *Gastroenterology* 2005; **129**: 1845-1853
42 **Török HP**, Glas J, Tonenchi L, Lohse P, Müller-Myhsok B, Limbersky O, Neugebauer C, Schnitzler F, Seiderer J, Tillack C, Brand S, Brünnler G, Jagiello P, Epplen JT, Griga T, Klein W, Schiemann U, Folwaczny M, Ochsenkühn T, Folwaczny C. Polymorphisms in the DLG5 and OCTN cation transporter genes in Crohn's disease. *Gut* 2005; **54**: 1421-1427
43 **Lakatos PL**, Fischer S, Claes K, Kovacs A, Molnar T, Altorjay I, Demeter P, Tulassay Z, Palatka K, Papp M, Rutgeerts P, Szalay F, Papp J, Vermeire S, Lakatos L. DLG5 R30Q is not associated with IBD in Hungarian IBD patients but predicts clinical response to steroids in Crohn's disease. *Inflamm Bowel Dis* 2006; **12**: 362-368
44 **Weersma RK**, Stokkers PC, van Bodegraven AA, van Hogezand RA, Verspaget HW, de Jong DJ, van der Woude CJ, Oldenburg B, Linskens RK, Festen EA, van der Steege G, Hommes DW, Crusius JB, Wijmenga C, Nolte IM, Dijkstra G. Molecular prediction of disease risk and severity in a large Dutch Crohn's disease cohort. *Gut* 2009; **58**: 388-395
45 **Henckaerts L**, Van Steen K, Verstreken I, Cleynen I, Franke A, Schreiber S, Rutgeerts P, Vermeire S. Genetic risk profiling and prediction of disease course in Crohn's disease patients. *Clin Gastroenterol Hepatol* 2009; **7**: 972-980.e2
46 **Weinshilboum RM**, Sladek SL. Mercaptopurine pharmacogenetics: monogenic inheritance of erythrocyte thiopurine methyltransferase activity. *Am J Hum Genet* 1980; **32**: 651-662
47 **Krynetski EY**, Evans WE. Genetic polymorphism of thiopurine S-methyltransferase: molecular mechanisms and clinical importance. *Pharmacology* 2000; **61**: 136-146
48 **Colombel JF**, Ferrari N, Debuysere H, Marteau P, Gendre JP, Bonaz B, Soulé JC, Modigliani R, Touze Y, Catala P, Libersa C, Broly F. Genotypic analysis of thiopurine S-methyltransferase in patients with Crohn's disease and severe myelosuppression during azathioprine therapy. *Gastroenterology* 2000; **118**: 1025-1030
49 **Arijs I**, Li K, Toedter G, Quintens R, Van Lommel L, Van Steen K, Leemans P, De Hertogh G, Lemaire K, Ferrante M, Schnitzler F, Thorrez L, Ma K, Song XY, Marano C, Van Assche G, Vermeire S, Geboes K, Schuit F, Baribaud F, Rutgeerts P. Mucosal gene signatures to predict response to infliximab in patients with ulcerative colitis. *Gut* 2009; **58**: 1612-1619
50 **Vermeire S**, Peeters M, Vlietinck R, Joossens S, Den Hond E, Bulteel V, Bossuyt X, Geypens B, Rutgeerts P. Anti-Saccharomyces cerevisiae antibodies (ASCA), phenotypes of IBD, and intestinal permeability: a study in IBD families. *Inflamm Bowel Dis* 2001; **7**: 8-15
51 **Papp M**, Altorjay I, Norman GL, Shums Z, Palatka K, Vitalis Z, Foldi I, Lakos G, Tumpek J, Udvardy ML, Harsfalvi J, Fischer S, Lakatos L, Kovacs A, Bene L, Molnar T, Tulassay Z, Miheller P, Veres G, Papp J, Lakatos PL. Seroreactivity to microbial components in Crohn's disease is associated with ileal involvement, noninflammatory disease behavior and NOD2/CARD15 genotype, but not with risk for surgery in a Hungarian cohort of IBD patients. *Inflamm Bowel Dis* 2007; **13**: 984-992
52 **Mow WS**, Vasiliauskas EA, Lin YC, Fleshner PR, Papadakis KA, Taylor KD, Landers CJ, Abreu-Martin MT, Rotter JI, Yang H, Targan SR. Association of antibody responses to microbial antigens and complications of small bowel Crohn's disease. *Gastroenterology* 2004; **126**: 414-424
53 **Papp M**, Norman GL, Altorjay I, Lakatos PL. Utility of serological markers in inflammatory bowel diseases: gadget or magic? *World J Gastroenterol* 2007; **13**: 2028-2036
54 **Dubinsky MC**, Lin YC, Dutridge D, Picornell Y, Landers CJ, Farrior S, Wrobel I, Quiros A, Vasiliauskas EA, Grill B, Israel D, Bahar R, Christie D, Wahbeh G, Silber G, Dallazadeh S, Shah P, Thomas D, Kelts D, Hershberg RM, Elson CO, Targan SR, Taylor KD, Rotter JI, Yang H. Serum immune responses predict rapid disease progression among children with Crohn's disease: immune responses predict disease progression. *Am J Gastroenterol* 2006; **101**: 360-367
55 **Papadakis KA**, Yang H, Ippoliti A, Mei L, Elson CO, Hershberg RM, Vasiliauskas EA, Fleshner PR, Abreu MT, Taylor K, Landers CJ, Rotter JI, Targan SR. Anti-flagellin (CBir1) phenotypic and genetic Crohn's disease associations. *Inflamm Bowel Dis* 2007; **13**: 524-530
56 **Markowitz J**, Kugathasan S, Dubinsky M, Mei L, Crandall W, LeLeiko N, Oliva-Hemker M, Rosh J, Evans J, Mack D, Otley A, Pfefferkorn M, Bahar R, Vasiliauskas E, Wahbeh G, Silber G, Quiros JA, Wrobel I, Nebel J, Landers C, Picornell Y, Targan S, Lerer T, Hyams J. Age of diagnosis influences serologic responses in children with Crohn's disease: a possible clue to etiology? *Inflamm Bowel Dis* 2009; **15**: 714-719
57 **Lakatos PL**, Altorjay I, Szamosi T, Palatka K, Vitalis Z, Tumpek J, Sipka S, Udvardy M, Dinya T, Lakatos L, Kovacs A, Molnar T, Tulassay Z, Miheller P, Barta Z, Stocker W, Papp J, Veres G, Papp M. Pancreatic autoantibodies are associated with reactivity to microbial antibodies, penetrating disease behavior, perianal disease, and extraintestinal manifestations, but not with NOD2/CARD15 or TLR4 genotype in a Hungarian IBD cohort. *Inflamm Bowel Dis* 2009; **15**: 365-374
58 **Rieder F**, Schleder S, Wolf A, Dirmeier A, Strauch U, Obermeier F, Lopez R, Spector L, Fire E, Yarden J, Rogler G, Dotan N, Klebl F. Association of the novel serologic antiglycan antibodies anti-laminarin and anti-chitin with complicated Crohn's disease behavior. *Inflamm Bowel Dis* 2010; **16**: 263-274
59 **Ferrante M**, Henckaerts L, Joossens M, Pierik M, Joossens S, Dotan N, Norman GL, Altstock RT, Van Steen K, Rutgeerts P, Van Assche G, Vermeire S. New serological markers in inflammatory bowel disease are associated with complicated disease behaviour. *Gut* 2007; **56**: 1394-1403
60 **Papp M**, Altorjay I, Dotan N, Palatka K, Foldi I, Tumpek J, Sipka S, Udvardy M, Dinya T, Lakatos L, Kovacs A, Molnar T, Tulassay Z, Miheller P, Norman GL, Szamosi T, Papp J, Lakatos PL. New serological markers for inflammatory bowel disease are associated with earlier age at onset, complicated disease behavior, risk for surgery, and NOD2/CARD15 genotype in a Hungarian IBD cohort. *Am J Gastro-*

enterol 2008; **103**: 665-681
61 **Seow CH**, Stempak JM, Xu W, Lan H, Griffiths AM, Greenberg GR, Steinhart AH, Dotan N, Silverberg MS. Novel antiglycan antibodies related to inflammatory bowel disease diagnosis and phenotype. *Am J Gastroenterol* 2009; **104**: 1426-1434
62 **Dubinsky MC**, Kugathasan S, Mei L, Picornell Y, Nebel J, Wrobel I, Quiros A, Silber G, Wahbeh G, Katzir L, Vasiliauskas E, Bahar R, Otley A, Mack D, Evans J, Rosh J, Hemker MO, Leleiko N, Crandall W, Langton C, Landers C, Taylor KD, Targan SR, Rotter JI, Markowitz J, Hyams J. Increased immune reactivity predicts aggressive complicating Crohn's disease in children. *Clin Gastroenterol Hepatol* 2008; **6**: 1105-1111
63 **Sandborn WJ**, Landers CJ, Tremaine WJ, Targan SR. Association of antineutrophil cytoplasmic antibodies with resistance to treatment of left-sided ulcerative colitis: results of a pilot study. *Mayo Clin Proc* 1996; **71**: 431-436
64 **Klebl FH**, Bataille F, Bertea CR, Herfarth H, Hofstädter F, Schölmerich J, Rogler G. Association of perinuclear antineutrophil cytoplasmic antibodies and anti-Saccharomyces cerevisiae antibodies with Vienna classification subtypes of Crohn's disease. *Inflamm Bowel Dis* 2003; **9**: 302-307
65 **Papp M**, Foldi I, Altorjay I, Palyu E, Udvardy M, Tumpek J, Sipka S, Korponay-Szabo IR, Nemes E, Veres G, Dinya T, Tordai A, Andrikovics H, Norman GL, Lakatos PL. Antimicrobial antibodies in celiac disease: trick or treat? *World J Gastroenterol* 2009; **15**: 3891-3900
66 **Beattie RM**, Walker-Smith JA, Murch SH. Indications for investigation of chronic gastrointestinal symptoms. *Arch Dis Child* 1995; **73**: 354-355
67 **Fagan EA**, Dyck RF, Maton PN, Hodgson HJ, Chadwick VS, Petrie A, Pepys MB. Serum levels of C-reactive protein in Crohn's disease and ulcerative colitis. *Eur J Clin Invest* 1982; **12**: 351-359
68 **Solem CA**, Loftus EV Jr, Tremaine WJ, Harmsen WS, Zinsmeister AR, Sandborn WJ. Correlation of C-reactive protein with clinical, endoscopic, histologic, and radiographic activity in inflammatory bowel disease. *Inflamm Bowel Dis* 2005; **11**: 707-712
69 **Florin TH**, Paterson EW, Fowler EV, Radford-Smith GL. Clinically active Crohn's disease in the presence of a low C-reactive protein. *Scand J Gastroenterol* 2006; **41**: 306-311
70 **Boirivant M**, Leoni M, Tariciotti D, Fais S, Squarcia O, Pallone F. The clinical significance of serum C reactive protein levels in Crohn's disease. Results of a prospective longitudinal study. *J Clin Gastroenterol* 1988; **10**: 401-405
71 **Consigny Y**, Modigliani R, Colombel JF, Dupas JL, Lémann M, Mary JY. A simple biological score for predicting low risk of short-term relapse in Crohn's disease. *Inflamm Bowel Dis* 2006; **12**: 551-557
72 **Travis SP**, Farrant JM, Ricketts C, Nolan DJ, Mortensen NM, Kettlewell MG, Jewell DP. Predicting outcome in severe ulcerative colitis. *Gut* 1996; **38**: 905-910
73 **Zilberman L**, Maharshak N, Arbel Y, Rogowski O, Rozenblat M, Shapira I, Berliner S, Arber N, Dotan I. Correlated expression of high-sensitivity C-reactive protein in relation to disease activity in inflammatory bowel disease: lack of differences between Crohn's disease and ulcerative colitis. *Digestion* 2006; **73**: 205-209
74 **Henriksen M**, Jahnsen J, Lygren I, Stray N, Sauar J, Vatn MH, Moum B. C-reactive protein: a predictive factor and marker of inflammation in inflammatory bowel disease. Results from a prospective population-based study. *Gut* 2008; **57**: 1518-1523
75 **Louis E**, Vermeire S, Rutgeerts P, De Vos M, Van Gossum A, Pescatore P, Fiasse R, Pelckmans P, Reynaert H, D'Haens G, Malaise M, Belaiche J. A positive response to infliximab in Crohn disease: association with a higher systemic inflammation before treatment but not with -308 TNF gene polymorphism. *Scand J Gastroenterol* 2002; **37**: 818-824
76 **Schreiber S**, Rutgeerts P, Fedorak RN, Khaliq-Kareemi M, Kamm MA, Boivin M, Bernstein CN, Staun M, Thomsen OØ, Innes A. A randomized, placebo-controlled trial of certolizumab pegol (CDP870) for treatment of Crohn's disease. *Gastroenterology* 2005; **129**: 807-818
77 **Lémann M**, Mary JY, Colombel JF, Duclos B, Soule JC, Lerebours E, Modigliani R, Bouhnik Y. A randomized, double-blind, controlled withdrawal trial in Crohn's disease patients in long-term remission on azathioprine. *Gastroenterology* 2005; **128**: 1812-1818
78 **Tibble JA**, Sigthorsson G, Bridger S, Fagerhol MK, Bjarnason I. Surrogate markers of intestinal inflammation are predictive of relapse in patients with inflammatory bowel disease. *Gastroenterology* 2000; **119**: 15-22
79 **Costa F**, Mumolo MG, Ceccarelli L, Bellini M, Romano MR, Sterpi C, Ricchiuti A, Marchi S, Bottai M. Calprotectin is a stronger predictive marker of relapse in ulcerative colitis than in Crohn's disease. *Gut* 2005; **54**: 364-368
80 **D'Incà R**, Dal Pont E, Di Leo V, Ferronato A, Fries W, Vettorato MG, Martines D, Sturniolo GC. Calprotectin and lactoferrin in the assessment of intestinal inflammation and organic disease. *Int J Colorectal Dis* 2007; **22**: 429-437
81 **Røseth AG**, Aadland E, Grzyb K. Normalization of faecal calprotectin: a predictor of mucosal healing in patients with inflammatory bowel disease. *Scand J Gastroenterol* 2004; **39**: 1017-1020
82 **Canani RB**, Terrin G, Rapacciuolo L, Miele E, Siani MC, Puzone C, Cosenza L, Staiano A, Troncone R. Faecal calprotectin as reliable non-invasive marker to assess the severity of mucosal inflammation in children with inflammatory bowel disease. *Dig Liver Dis* 2008; **40**: 547-553
83 **Orlando A**, Modesto I, Castiglione F, Scala L, Scimeca D, Rispo A, Teresi S, Mocciaro F, Criscuoli V, Marrone C, Platania P, De Falco T, Maisano S, Nicoli N, Cottone M. The role of calprotectin in predicting endoscopic post-surgical recurrence in asymptomatic Crohn's disease: a comparison with ultrasound. *Eur Rev Med Pharmacol Sci* 2006; **10**: 17-22

S- Editor Wang YR **L- Editor** O'Neill M **E- Editor** Zheng XM