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Is mean platelet volume a good predictor of sustained response to one year infliximab therapy in pediatric patients with Crohn's disease?

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Abstract: Over the past years, there is a growing number of newly diagnosed pediatric patients with Crohn's disease (CD). Severe course of CD often requires biological treatment with Infliximab (IFX). Loss of response to biological treatment is a major problem. Mean platelet volume (MPV) was reported as a good marker of sustained response to IFX therapy in adults. This study is to determine whether MPV measured prior to IFX therapy and after its third dose can be used as a predictive marker of sustained response to biological therapy in children with severe course of CD. 43 pediatric patients with CD who underwent IFX therapy were enrolled into this study. The clinical response was evaluated after the third dose and after one year of IFX treatment (sustained response). The MPV values at baseline and week 14 were compared to the patients with good response to IFX to those with loss of the response. During 52-week IFX therapy, 2 out of 43 patients enrolled in the study did not achieve primary response after the third dose, another 18 children lost their response to the above therapy after one year. There was no significant association between baseline and 14th week values of MPV between patients with the sustained response to those with loss of response. In opposite to adult patients, MPV cannot be regarded as predictive factor of sustained response to IFX treatment in pediatric patients.

Key words: Pediatric Crohn's disease, mean platelet volume, Infliximab treatment.

Introduction

Inflammatory bowel disease (IBD) is a chronic inflammatory process of the gastrointestinal tract, characterized by periods of exacerbations and remissions, and leading to multiple health complications. Crohn's disease (CD) is a predominant IBD in pediatric patients. The pathogenesis of CD is complex, and still not entirely understood. Over the last few years, a growing number of newly diagnosed pediatric patients with extensive CD can be observed [1]. The recommended treatment in the early onset and severe course of the disease requires anti-TNF-α antibodies therapy with Infliximab (IFX) or Adalimumab (ADA) to support long-term remission and to prevent chronic complications. Loss of response to IFX in CD is a major clinical problem. It affects 23–46% of adult patients after 12-month therapy [2]. Such inhibition process is also observed in children, and often precludes further use of IFX [3]. In the latest research, it is proposed that the CD patients develop antibodies blocking anti-TNF-α agents causing loss of treatment response [3, 4]. There is also suspected role of other factors involved in development of CD, especially those associated with rise of the pro-inflammatory cytokines, such as IL-12, IL-17, IL-23 [5, 6]. There are several blood biomarkers used to reveal, monitor and evaluate both exacerbation of CD and response to IFX, which include erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), white blood count (WBC), platelet count (PLT) and albumin level [7]. Recent studies suggest that platelets can have important role in the pathogenesis of CD [8]. There are also reports which list mean platelet volume (MPV) as a good predictor of a sustained response to infliximab therapy in adults [9]. This study was designed to assess whether MPV can also be used as a valid marker of a sustained response to anti-TNF-α therapy in pediatric patients.

Materials and methods

Patients

This report presents a retrospective pilot study. A population of 62 pediatric patients diagnosed with severe CD was treated with anti-TNF-α agents in Department of Pediatrics Gastroenterology and Nutrition, Jagiellonian University Medical College in Krakow, Poland, between January 2012 and March 2016. The group of 43 children, which included 23 girls (54%) and 20 boys (46%) with mean age 15.0 years [10.5; 16.0] met our inclusion criteria, and were enrolled into the study (Figure 1). The control group consisted of 43 age and sex matched children hospitalized at our Clinic, diagnosed with dyspepsia, irritable bowel syndrome (IBS) or small intestinal bacterial overgrowth (SIBO). The inclusion criteria for CD pediatric patients were: Caucasian, age between 2–17 years old, diagnosed with severe CD (Pediatric Crohn's Disease Activity Index, PCDAI >50 points), qualified for infliximab therapy, complete medical

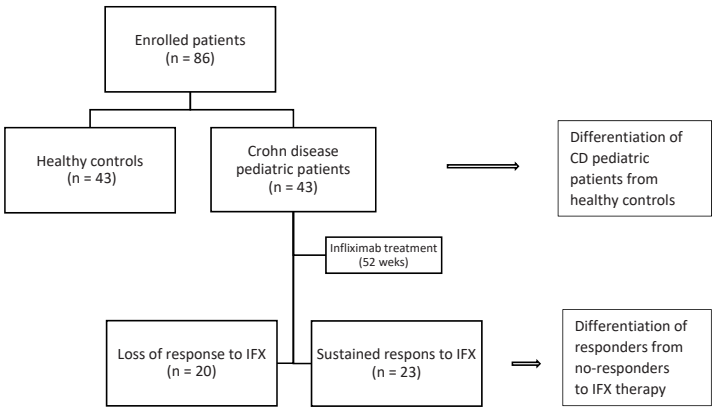

Fig. 1. Study design: A total of 86 subjects were enrolled into this study. 43 healthy controls were compared with CD pediatric patients (Inflammatory markers: CRP, WBC, ESR, PLT, MPV). CD patients were treated during 52 week therapy with anti-TNF-α agent Infliximab. Than laboratory parameters of patients who lost response were compared to the results of patients with sustained response.

documentation. The exclusion criteria were: isolated perianal disease, age under 2 years old and over 17 years old, pregnancy, use of anti-inflammatory drugs (except azathioprine, 6-mercaptopurine, steroids), concomitant conditions such as: atopic dermatitis, diabetes, pulmonary or kidney disease, leukemia. The complete physical examination, laboratory tests and endoscopic examination (gastrodudodenoscopy and ileocolonoscopy) were performed in all the patients as the baseline of the study. CD diagnosis was made based on the clinical, endoscopic, radiological and pathological evaluation according to European Society for Pediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN) revised PORTO criteria for the diagnosis of inflammatory bowel disease in children and adolescents [10]. The activity of disease was checked at designated time points before and during therapy according to Pediatric Crohn's Disease Activity Index (PCDAI) [11]. PCDAI is a complex scoring system covering clinical symptoms (severity of abdominal pain, number and consistency of stools, patient general activity), laboratory tests (ESR, albumin, hematocrit values), growth delay, perianal changes and extra intestinal manifestation of CD. The criteria to initiate biological therapy were PCDAI score over 50 points, lack of effect of the conventional treatment (mesalazine, azathioprine, corticosteroids, clinical nutrition therapy). All the patients enrolled in this biological treatment received an induction treatment: 5 mg/kg IFX infusion at baseline and next 5 mg/kg at the 2nd and 6th week. After the third dose of IFX (14th week), the primary response was evaluated and the patients underwent second endoscopic examination, followed by selection for further biological treatment. A good response to therapy was defined as an improvement of endoscopic changes and reduction in PCDAI score by ≥12.5 points and clinical remission by PCDAI total score ≤30 points. Loss of the response during therapy was

defined as one of following: exacerbation of disease with PCDAI score ≥30 points during at least 2 subsequent visits, CD-related laparotomy, crossover from the scheduled therapy (every 8 weeks) to episodic treatment. Administration of IFX was performed every 8 weeks for the next 38 weeks. After one year of therapy, the patients underwent third endoscopic evaluation.

Laboratory parameters

Blood samples for laboratory analysis were taken twice, first — during qualification for biological treatment procedure (before the first dose of IFX), and then during evaluation of efficacy of induction treatment (after 14th week). Blood was automatically analyzed within 2 hours after its collection. The samples of 2 ml of ethylenediaminetetraacetic acid (EDTA)-anticoagulated venous blood were used to analyze platelet parameters (MPV and PDW), and another 2 ml of venous blood was placed in the serum tubes to assess biochemistry and CRP level. The reference range for MPV in children is 6.5–10 fl, and for CRP <5 mg/L.

Statistical analysis

Statistical analysis was performed using Statistica software, version 12. Data were presented as number of patients (percentage of the group) for categories, and median [lower; upper quartile] for qualitative variables. Chi-squared test, t-test and Mann–Whitney test were used to study differences between groups, according to distribution (as tested with Shapiro–Wilk test). Correlations were assessed with Pearson or Spearman coefficient. The results were considered statistically significant when $p < 0.05$.

Ethical considerations

The study was approved by the Bioethics Committee of Jagiellonian University, Kraków. (No:122.6120.52.2015)

Results

Differences between pediatric patients with CD and healthy controls

Demographic parameters of the pediatric CD patients and the control group are presented in Table 1. The two groups were matched according to their gender and age, however, body mass index (BMI) in the CD children was significantly lower compared to controls. The children with CD demonstrated significantly higher platelet counts, and lower platelet parameters such as platelet distribution width, and medium platelet volume than in the control group (Table 2).

Table 1. Demographic features of pediatric CD patients and control group. BMI-body mass index.

	Pediatric CD patients n = 43		Control group n = 43		p-value
Gender	male (46.5%)	female (53.5%)	male (46.5%)	female (53.5%)	1.000
Age	15.0 [10.5; 16.0]		14.0 [10.5; 16.2]		0.798
BMI	16.4 [14.5; 18.0]		18.2 [15.9; 21.0]		0.009

Table 2. Comparison of platelet counts (PLT), platelet distribution width (PDW) and mean platelet volume (MPV) between CD patients (at baseline) and control group.

	Pediatric CD patients n = 43	Control group n = 43	p-value
PLT ths/ul	483.0 [346.5; 532.5]	247.0 [219.0; 281.5]	<0.001
PDW fl.	10.2 [9.5; 11.1]	12.1 [11.1; 13.5]	<0.001
MPV fl.	9.4 [8.8; 9.9]	10.4 [9.9; 11.1]	<0.001

Table 3 demonstrates that the main inflammatory markers: erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), white blood count (WBC) were also significantly higher in CD patients. Hemoglobin, hematocrit, red blood count levels were lower in the CD pediatric patients in comparison to control group (Table 4).

Table 3. Differences between CD patients and control group — inflammatory markers: erythrocyte sedimentation rate (ESR), C-reactive protein (CRP), white blood count (WBC).

	Pediatric CD patients	Control group	p-value
ESR mm/h	24.0 [15.0; 44.0]	5.0 [3.0; 7.0]	<0.001
CRP mg/L	15.7 [6.1; 40.5]	5.0 [5.0; 5.0]	<0.001
WBC ths/ul	9.6 [6.7; 12.4]	5.9 [5.0; 7.2]	<0.001

Table 4. Hematocrit (HCT), Hemoglobin (HGB) and red blood count (RBC) in CD patients and control group.

	Pediatric CD patients	Control group	p-value
RBC mln/ul	4.3 [4.0; 4.8]	4.8 [4.6; 5.0]	0.002
HGB g/dl	12.0 [10.8; 12.9]	13.5 [12.9; 14.3]	<0.001
HCT %	38.1 [33.7; 40.8]	40.6 [39.2; 42.2]	<0.001

Correlations

We did not observe any correlations between MPV and BMI or ESR, or WBC value, however, we identified a negative correlation between MPV and CRP (Table 5).

Table 5. Correlations at baseline between MPV (mean platelet volume) and BMI (body mass index) and inflammatory markers in CD pediatric patients (n = 43). WBC (white blood count); ESR (erythrocyte sedimentation rate); CRP (C-reactive protein).

	r-value	p-value
BMI	0.08	0.601
WBC ths/ul	–0.13	0.406
ESR mm/h	–0.13	0.395
CRP mg/L	–0.39	0.010

Comparison of pediatric CD patients with sustained response to IFX (responders) to the ones who lost response to IFX during 52-week therapy (non-responders)

During a yearlong therapy, 20 out of 43 patients (7 boys, 13 girls) gradually lost their IFX response, while the other 23 patients demonstrated sustained response to biological treatment. We compared the results from the group that lost response to IFX with those children, who maintained their response. The significant differences were observed. The activity of the disease measured by PCDAI in responders group was significantly lower than PCDAI in the non-responders group (0.0 [0.0; 5.0] vs. 8.8 [4.4; 15.6]; p <0.001). The decline of PCDAI, analyzed as delta PCDAI measured before the first dose and after the third dose was significantly lower in the non-responders in comparison to the responders (–45.0 [–52.5; –36.2] vs. –52.5 [–52.5; –50.0]; p = 0,007). There were no significant differences regarding change in PLT (Δ PLT) or MPV levels (Δ MPV) between the two groups. Out of analyzed inflammatory markers, only ESR levels were significantly higher in the non-responders than in the responders group (20.5 [9.8; 29.2] vs. 7.0 [4.0; 11.5]; p = 0.001). It was interesting to find out that the children with sustained response to IFX had significantly higher levels of hemoglobin compared to the patients who lost response to IFX (12.9 [12.2; 13.7] vs. 11.2 [10.9; 13.0]; p = 0.009) (Table 6).

Table 6. Comparison of baseline mean platelet volume (MPV), platelet count (PLT) and Δ MPV, Δ PLT (during 14 week therapy) between responders and non-responders to IFX therapy.

	Non-responders n = 20	Responders n = 23	p-value
PLT ths/ul	372.0 [314.5; 431.2]	328.0 [266.0; 410.0]	0.073
MPV fl	9.8 [9.4; 10.8]	9.6 [9.4; 10.3]	0.583
ΔPLT ths/ul	–85.5 [–179.5; –60.5]	–65.0 [–194.5; –33.0]	0.884
ΔMPV fl	0.4 [0.1; 0.7]	0.6 [0.2; 1.1]	0.414

Discussion

Despite significant progress in CD therapy, IFX (and its biosimilars) therapy remains the most important treatment for severe pediatric CD patients. IFX is a chimeric (human-mice) monoclonal antibody biologic drug that works against tumor necrosis factor alpha (TNF-α) and is used to treat inflammatory bowel diseases. It was first approved to use in adult population in 1999. In May 2006, it was approved to use in severe course of CD in pediatric patients. The loss of response to IFX is attributed to production of anti-IFX antibodies by the recipient's immune system [4].

Mean platelet volume is one of the platelet indices, automatically assessed, that adds no extra cost or effort to a full blood count examination. MPV normal value in pediatric patients is estimated between 6.5–10 fl. This is a constant parameter that does not change with age. It is assumed to be influenced by BMI [12], but we did not find this correlation significant in our research. Many studies show a possible association between MPV value and pediatric inflammatory diseases such as rheumatoid arthritis, juvenile idiopathic arthritis, acute appendicitis and viral gastroenteritis [13–15]. It is also assumed that MPV can be used in diagnostic process of IBD patients and as a marker to predict treatment response to IFX in adults [16, 9]. It is hypothesized that larger platelets are cumulated in inflammation sites, such as intestine walls in IBD; therefore, only small platelets with low MPV are circulating in the bloodstream and can be easily detected [16]. The other explanation of this phenomenon is that inflammatory reaction mediated by pro-inflammatory cytokines and acute phase reactants stimulates megakariocytopoiesis and increases production of small size platelets [8]. There are also reports that IFX therapy can ameliorate the hypercoagulable state in adult patients with CD probably due to platelet changes [17].

In this study, we demonstrate that there is no significant correlation between MPV value and BMI, or between MPV and inflammatory markers, such as ESR or WBC in CD patients. However, we identify a negative correlation between MPV and CRP. In contrast to the earlier findings in CD adult patients [9], this study does not confirm MPV value as a significant predictor of response to IFX in children with severe CD. Although MPV is not a good predictive marker of the response to IFX therapy, it might still be used to differentiate between CD patients and healthy children. Loss of response in pediatric CD patients undergoing maintenance therapy with IFX is a warring phenomenon, and we observed this pattern in our study. Therefore, new markers for monitoring response to biological treatment are needed, and they can change existing approach to patients' follow-up.

Implementation of appropriate treatment is crucial for the course of the disease. This study shows that patients with lower initial PCDAI, ESR, and higher hemoglobin level respond better to IFX therapy and can develop sustained response. It is an interesting observation that children with initially lower activity of the disease and

with higher concentration of hemoglobin respond better to this biological treatment. This outcome is in accordance with the latest study, which shows that early IFX induction treatment ("top-down" strategy) is more effective in pediatric CD patients than conventional therapy ("step-up" strategy) [18]. Such studies may result in new recommendations of earlier introduction of infliximab in patients with moderate to severe CD.

Conclusion

We demonstrated that MPV cannot be used as a valid predictive marker of the sustained response to IFX therapy, but it can help to differentiate healthy children from the children suffering from CD. Additionally, it appears that the earlier administration of IFX in pediatric patients with lower activity of the disease (lower ESR and higher concentration of hemoglobin), more likely maintains their response to the biological anti-TNF-α therapy.

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Conflict of interest

None declared.

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