RDW AS A NOVEL MARKER OF DISEASE ACTIVITY IN CROHN'S DISEASE: TESTING A HYPOTHESIS IN AN IBD TERTIARY CARE CENTER IN ROMANIA

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ABSTRACT

BACKGROUND: ONE OF THE MOST COMMON COMPLICATIONS IN PATIENTS WITH INFLAMMATORY BOWEL DISEASE (IBD), EITHER CROHN'S DISEASE (CD) OR ULCERATIVE COLITIS (UC), IS ANEMIA. RED BLOOD CELL DISTRIBUTION WIDTH (RDW) QUANTITATIVELY MEASURES THE SIZE VARIABILITY OF THE RED BLOOD CELL POPULATION AND MIGHT INCREASE EVEN BEFORE ANEMIA MANIFESTS ITSELF. THE AIM OF THIS STUDY WAS TO INVESTIGATE THE USEFULNESS OF RDW IN THE EVALUATION OF DISEASE ACTIVITY IN CD BY COMPARISON WITH ALREADY ACCEPTED MARKERS OF ACTIVITY SCH AS ERYTHROCYTE SEDIMENTATION RATE (ESR), C-REACTIVE PROTEIN (CRP) AND FIBRINOGEN LEVELS. WE ENROLLED 148 PATIENTS WITH CROHN'S DISEASE, AND 51 PATIENTS WITH IRRITABLE BOWEL SYNDROME (IBS) AS THE CONTROL GROUP.

RESULTS: RDW WAS SIGNIFICANTLY HIGHER ONLY FOR PATIENTS WITH SEVERE DISEASE ACTIVITY (P=0.02). RDW HAD SIGNIFICANTLY HIGHER VALUES FOR THE PATIENTS WITH CD (P=0.004). RDW WAS THE MOST SENSITIVE AND SPECIFIC MARKER FOR CD WITH INFLAMMATORY PATTERN (P<0.001). ADDITIONALLY, RDW SHOWED A CORRELATION WITH THE EXTRAINTESTINAL MANIFESTATIONS OF CD (P<0.05).

CONCLUSIONS: RDW IS NOT SUFFICIENTLY SENSITIVE OR SPECIFIC TO DIAGNOSE INDEPENDENTLY THE FLARES OF ACTIVITY IN IBD. RDW CAN BE AN ADDITIONAL MARKER IN DIFFERENTIATING CD ACTIVITY FLARES FROM OVERIMPOSED FUNCTIONAL DISORDERS (IBS). RDW CORRELATES SIGNIFICANTLY WITH EXTRAINTESTINAL MANIFESTATIONS AND INFLAMMATORY PATTERN OF CD.

KEYWORDS: RDW, INFLAMMATORY BOWEL DISEASE, ACTIVITY, CRP, ESR, EXTRAINTESTINAL MANIFESTATIONS

INTRODUCTION

Inflammatory bowel diseases(IBD) are chronic inflammatory diseases of unknown origin and affect different segments of the gut. There are two mainly defined diseases included in the group, Crohn's Disease(CD) and Ulcerative colitis(UC). They are characterized by recurring remission and exacerbation periods. CD is a lifelong disease resulting from a complex interaction between genetic and environmental factors, however

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observed predominantly in developed countries of the world[1]. There is a distinct North-South gradient within Europe, with a recent increase of incidence in Southern countries.

To diagnose and determine disease activity of CD there are noninvasive laboratory markers, endoscopic procedures (such as ileocolonoscopy) and imaging techniques: MR or CT enterography/enteroclysis has the highest diagnostic accuracy for the detection of intestinal involvement of CD including extramural complications, whereas small bowel capsule endoscopy is reserved for patients with a high clinical suspicion of CD despite negative investigation by the previously mentioned techniques[1]. However, because of the invasiveness and cost of endoscopic procedures and the cost of advanced imaging techniques there are several laboratory markers used to determine inflammation and disease activity such as: C-reactive protein (CRP), erythrocyte sedimentation rate (ESR), hemoglobin (Hgb), platelet counts(PLT), as well as new investigations such as: fecal calprotectin[3], VEGF[4], TREM[5], micro-RNA[6], factor XIII[7], Feritin and transferin saturation[8], ANCA and ASCA[9], interleukins 6 and 1, soluble interleukin-2 receptor and soluble intercellular adhesion molecule-1(ICAM-1) [10]. None of these tests, however, have a high specificity or sensibility for monitoring disease activity in CD, nor a high usability as diagnostic markers. Recent studies have revealed that many of the mentioned markers, especially those with higher specificity and sensibility than the rest, are expensive and not widely available. This entails the need for a new way to test IBD disease activity inexpensively and non-invasive as well as not potentially harmful to the patients. Several studies have evaluated and demonstrated the potential of red cell distribution width(RDW) as a valuable marker for the diagnostic and monitoring of disease activity in IBD[11-14].

MATERIALS AND METHODS

1. Selection of study participants

This is a cross sectional observational cohort study performed in the Department of Gastroenterology, Fundeni National Institute for Gastroenterology and Hepatology. A total of 148 IBD patients with ages ranging from 20 to 73 years participated in the study. The patients were diagnosed with CD in various stages of activity that was clinically, imagistically, endoscopically and histopathologically confirmed. Additionally, the clinical and laboratory parameters of the patients were reviewed to confirm the diagnosis. A group of 51 patients diagnosed with irritable bowel syndrome with diarrhea(IBS-d), with no laboratory, imagistic or endoscopic abnormalities was employed as a control group due to the frequent need for differential diagnosis with IBD in this group of patients. These control patients had normal laboratory tests, without any inflammation.

Pacients without a clear diagnosis of CD or without insufficient clinical and laboratory and endoscopic data to support the diagnosis were excluded from the study.

The study group was divided into 2 subgroups, based on the status of disease activity (either “active” or “in remission”) and the active disease subgroup was further divided into activity severity groups based on the CDAI score and clinical criteria (mild, moderate and severe activity respectively).

The CD subgroup was also split into 3 groups groups based on disease pattern, inflammatory, structuring or fistulising, respectively. The clinical evaluation criteria used were CD location, perianal lesions, presence of endoscopic lesions, extraintestinal manifestations.

The laboratory parameters used were Hgb levels (ranging from 12 to 14 g/dl), ESR(with a threshold of 15 mm/hr), CRP(with a threshold of 2 mg/l), fibrinogen(threshold of 400 mg/dl) and RDW-SD (range from 39 to 46 FL).

The age, gender, disease location, pattern and disease activity for each patient were recorded. All enrolled patients were required to have biological tests such as complete blood count(CBC), ESR, CRP. All CBC parameters, ESR and CRP values were assessed. Initial comparisons were made for RDW values between the IBS control group and CD group. Then, the patients were grouped based on their disease activity score in groups of actively/in remission CD and different severity CD, and RDW levels were compared between the active and remission period subgroups, as well as between each activity state. Finally, ESR and CRP levels were compared among the groups and crosswise with the control group.

2. Statistical evaluation

SPSS for Windows version 20.0 was used for the statistical analyses. For the evaluation of the data from the study, descriptive statistical methods (mean±standard deviation), Student's t-test and the Mann-Whitney U Test were used to establish potential differences between the averages of 2 independent groups for parameters with and without normal distributions, respectively. The Kruskal-Wallis test was used for variance analysis of intergroup values. The one-way ANOVA test was used for normal distribution, one-way variance analysis. In order to determine a potential significant correlation of RDW with CRP, ESR, clinical symptoms or disease pattern between multiple groups, Spearman's rank correlation coefficient was used. Receiver operating characteristic(ROC) curve analysis was used to calculate sensitivity and specificity levels. For comparisons of qualitative data, the chi-squared test was used. The results in the 95% confidence interval and p-values of less than 0.05 were considered to be significant.

Table 1. Demographic and clinical characteristics of the controls and the patients with IBD

Characteristic	Control group (n=51)	CD(n=148)	P-value
Age, yr	54.43±15.29	38.8±6.458	<0.001
Sex(F/M)	38/13	79/69	<0.001
Localization of disease, n(%)			-
Ileitis	-	30(20%)	-
Colitis	-	88(60%)	-
Ileocolitis	-	30(20%)	-
Clinical activity, n(%)			-
Active disease	-	100(68%)	-
In remission	-	48(32%)	-

RESULTS

There were significant differences in age between the CD group and its subgroups and the control group with IBS. The median age among the patients with CD was 38.8±6.46, underlining the prevalence of CD occurrence at a young age, whereas the patients in the

control group were significantly older, with a median age of 54.43±15.29 years. There was also a significant gender distribution difference between the IBD group and control, with the distribution being similar (52% women, 48% men respectively) among the patients with CD, and in the favor of women for the IBS-d group (75% women). Table 1 presents the demographic and clinical characteristics of the two groups.

The laboratory parameters from the patients with CD compared to those of the control group are presented in table 2. When the CD patients were compared based on their disease state, active disease versus remission, we observed the following: 1) regardless of disease state, the RDW level was statistically different between control, active and in remission disease; 2) the RDW values between active and in remission subgroups were not statistically different(p= 0.13); 3) with regards to activity state, RDW was significantly different only for the severe activity stage of CD (ANOVA test p-value=0.01; Mann-Whitney U test p-value <0.001); 4) there was no statistically significant difference in RDW levels between disease patterns (p-value = 0.46);

CRP and ESR levels in patients with active disease were significantly higher than those with CD in remission or control. The Pearson correlation (table 3) indicated that increased inflammatory parameters such as ESR, CRP and Fibrinogen were significantly correlated with an increased RDW level in patients with active disease. The inflammatory markers themselves were also significantly correlated. All these correlations indicate that RDW levels increase in the presence of inflammation similarly to other inflammatory markers, thus giving the possibility of RDW to be considered an indicator of active inflammation.

Table 4 and Fig. 1 show comparisons with the other inflammatory parameters (ESR, CRP, Fibrinogen and Hgb, in patients with inflammatory CD. Only for that particular disease pattern did the ROC curve for RDW-SD show an increased AOC (area under curve of 0.493) compared to the other markers(except CRP) and the largest confidence interval of all the markers used(for a 95% confidence level, the lower bound was 0.35 and the upper bound 0.637). RDW-SD was the second most specific indicator with 63% (after Fibrinogen at 85%) and had the lowest sensitivity with 48%. The most sensitive indicator for the CD patients was CRP with 87.5% but its specificity was 37%. Overall, in CD patients, RDW was not a signifficant indicator for disease activity, in terms of specificity and sensibility, whereas CRP, VSH and Fibrinogen were statistically signifficant (table 5).

Table 2. Comparison of Laboratory Parameters of CD group and Controls (One-way ANOVA test)

Parameter	Control group (n=51)	CD group		P Value
		Active disease (n=100)	In remission (n=48)	
RDW-SD(FL)	42.51±3.1	46±6.5	44.44±4.9	0.001
Hbg(g/dl)	13.63±1.3	12.3±2.3	13.7±1.5	<0.001
ESR(mm/hr)	28.4±15.7	36.6±19.6	22.6±13	0.003
CRP(mg/dl)	5.8±3.7	31.7±50	5±13	0.001
Fibrinogen(mg/dl)	340.3±99	456±140.4	328.6±90.7	<0.001

Table 3. Correlation of RDW with other inflammatory parameters (Pearson Correlation)

Parameter	Pearson Correlation	P value
CRP(mg/dl)	0.15	0.059
ESR(mm/hr)	0.28	0.005
Fibrinogen(mg/dl)	0.23	0.002
Hbg(g/dl)	-0.4	<0.001

Table 4. Statistical data for RDW, CRP, ESR, Hbg and Fibrinogen in the active CD group

Area Under the Curve			
Crohn's disease inflammatory pattern	Area	Asymptotic 95% Confidence Interval	
		Lower Bound	Upper Bound
RDW(FL)	0,493	,350	,637
CRP(mg/l)	0,495	,346	,645
ESR(mm/h)	0,482	,334	,630
Fibrinogen(mg/dl)	0,423	,271	,575

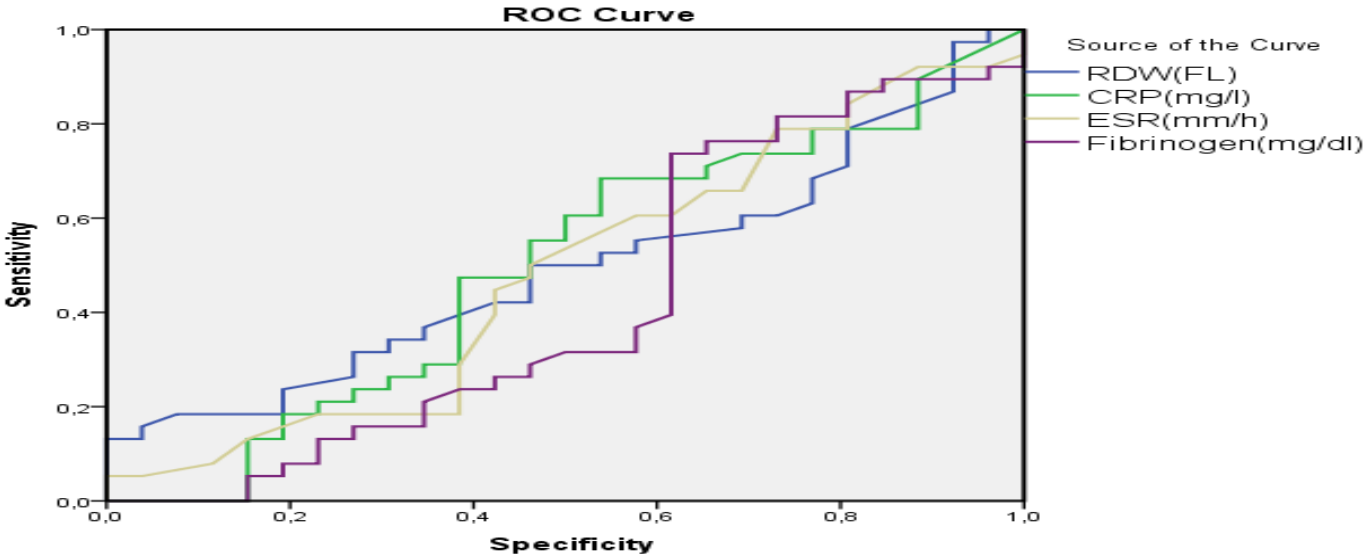

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Fig. 1. ROC curves for CRP, ESR, Fibrinogen and RDW compared to the activity level of inflammatory pattern CD(the area under curve for RDW is 0.493

Table 5. Sensibility and specificity for RDW-SD, CRP, VSH and Fibrinogen as disease activity markers

Parameters	AUC(95% CI)	Highest value	Sensitivity(%)	Specificity(%)	P value
RDW-SD(FL)	0.584(0.448-0.720)	46 46.5 47	48 37 35.4	63 66 66	0.229
CRP(mg/dl)	0.794(0.687-0.901)	0.85 1.05 1.5	87.5 87.5 83	33 41 52	<0.001
VSH(mm/h)	0.694(0.576-0.813)	15 16.5 19	85 83 81	22 30 33	0.005
Fibrinogen(mg/dl)	0.815(0.717-0.913)	400 410 425	58 56 54	85 85 89	<0.001

DISCUSSION

Nowadays, many tests in the field of CD are pointed towards the identification of a marker or test for the detection and assessment of the active disease that are easy to perform, affordable, noninvasive, and doable with the equipment available in the majority of the clinics worldwide. In our study, we investigated the correlation of RDW with ESR, CRP and Fibrinogen. Furthermore, a control group was included in the study to assess alterations in RDW between these controls and the CD patients. There were significant increases observed in RDW-SD, ESR and CRP in all groups compared to the control group. There was also a signifficant increase of all the markers in the active disease periods of CD compared to the control group. This demonstrated the correlation between ESR, CRP, Fibrinogen and RDW-SD in CD.

The evaluation of RDW values in CD patients would be an affordable and easy test to do in most clinics, without any excess expenses.

Our study is concordant with the previous ones[11-14] in terms of correlation between inflammatory markers and RDW-SD for activity flares of CD, and also for differentiating between IBD and IBS-d. We consider that the results obtained for the inflammatory pattern of CD could be explained by the greatest inflammatory burden of these patients compared to other patterns of activity. In these patients, multiple pathophysiological mechanisms of anemia concur thus giving rise to microcytosis and elevation in RDW-SD. The same explanation is acceptable for the association IBD-extraintestinal manifestations. Another explanation for this correlation would be the fact that these patients usually present with altered nutritional status which leads to heterogeneous cell population and increased RDW values[18]. It has been already demonstrated that elevated RDW is more frequently seen in iron deficiency states than in thalassemia or anemia of chronic disease this way explaining

probably the higher RDW in purely inflammatory pattern of CD rather that structuring or fistulising CD. [19]

Finally, is to be mentioned that in the past several years, there have been many studies which have investigated the relation between RDW levels and cardiac and non-cardiac death[15-17], which could be an interesting aspect in the study of cardiac diseases associated to IBD.

CONCLUSIONS

RDW values did not correlate with symptoms or location of CD probably because it is a marker of the general inflammatory burden. It was neither more sensitive nor more specific as compared to the other markers of activity for evaluation of CD. However, the differences in median values between CD in remission and severe activity, and also between active CD and control group, were statistically significant making RDW an useful tool for defining active disease. There was also seen an association between RDW and the other makers of inflammation used in the study, confirming the fact that RDW is in fact correlated with systemic inflammatory burden.

Taking into consideration the other studies already published on this theme[11-14], we can conclude that RDW could be an useful parameter for evaluating activity in CD, being a cheap investigation as it is found on every CBC. In correlation with the other markers already used, it could be useful in patients where endoscopic investigations are forbidden or non-contributive , and also in cases with difficult diagnosis.

REFERENCES

1. **Van Assche G, Dignass A,** et al. *The second European evidence-based Consensus on the diagnosis and management of Crohn's disease: Definitions and diagnosis. Journal of Crohn's and Colitis 2010; 4: 7-27*
2. **Shivananda S** et al. *Incidence of inflammatory bowel disease across Europe: is there a difference between the north and south? Results of the European Collaborative Study on Inflammatory Bowel Disease (EC-IBD). Gut 1996; 39: 690-7*
3. **Mendoza JL, Abreu MT**. *Biological markers in inflammatory bowel disease: Practical consideration for clinicians. Gastroenterol Clin Biol (2009) 33, Suppl. 3; S158-S173*
4. **Tsiolakidou G** et al. *Increased expression of VEGF and CD146 in patients with inflammatory bowel disease. Dig and Liv Dis (2008) 40; 673-679*
5. **Billioud V, Gibot S, Massin F, Oussalah A,** et al. *Plasma soluble triggering receptor expressed on myeloid cells-1 in Crohn's disease. Dig Liv Dis (2012) 44; 466-470*
6. **Archanioti P, Gazouli M, Theodoropoulos G**, et al. *Micro-RNAs as regulators and possible diagnostic bio-markers in inflammatory bowel disease. Jo Cro Col (2011) 5; 520-524*
7. **Cougard PA, Desjeux A, Vitton V**, et al. *The usefulness of factor XIII levels in Crohn's disease. J Cro Col (2012) 6; 660-664*
8. Oustamanolakis P, Koutroubakis IE, Kouroumalis EA. *Diagnosing anemia in inflammatory bowel disease: Beyond the established markers. J Cro Col (2011) 5; 381-391*
9. **Iskandar HN, Ciorba MA**. *Biomarkers in inflammatory bowel disease: current practices and recent advances. Tran Res (2012) 159; 313-325*
10. **Yeşil A, Şenanteş E, Bayoğlu IV**, et al. *Red Cell Distribution Width: A Novel Marker of Activity in Inflammatory Bowel Disease. Gut 2011; 4 ; 460-467*
11. **Arhan M, Önal IK, Taş A, Kurt M**, et al. *The role of red cell distribution width as a marker in inflammatory bowel disease. Turk J Med Sci (2011) 41(2); 227-234*
12. **Sond CS** et al. *Association between red cell distribution width and disease activity in patients with inflamatory bowel disease. Digestive diseases and sciences, (2012) 577; 1033-1038*
13. **Cakal B** et al. *Red Cell distribution width for assessment of activity of inflammatory bowel disease", Dig dis sci (2009); p 842-847*
14. **Farkas K, Papp M, Nyari T**, et al. *Red Blood Cell Distribution Width in Combination with Serological Markers can Help in the Differentiation between Crohn's Disease and Ulcerative Colitis. The Open Gastroenterology J (2010) 4; 1-4*
15. **Pascual- Figal DA, Bonaque JC, Redondo B**, et al. *Red Blood Cell Distribution width predicts long-term outcome regardless of anaemia status in acute heart failure patients. Eu J of Heart Failure (2009) 11; 840-846*
16. **Horwich TB, Fonarow GC, Hamilton MA**, et al. *Anemia is associated with worse symptoms, greater impairment in functional capacity and a significant increase in mortality in pacients with advanced heart failure. J Am Coll Cardiol (2002) 39; 1780 – 1786*
17. **Groenveld HF, Januzzi JL, Damman K,** et al. *Anemia and mortality in heart failure patients a systematic review and meta-analysis. J Am Coll Cardiol (2008) 52; 818-827*
18. **Mitchell RM, Robinson TJ.** *Monitoring dietary compliance in coeliac disease using red cell distribution width. Int J Clin Pract.2002;56:249–250.*
19. **Johnson MA**. *Iron: nutrition monitoring and nutrition status assessment. J Nutr. 1990;120(Suppl 11):1486–1491.*