

Histopathological Evaluation of Duodenal Biopsies In Celiac Disease On The Basis of Marsh Classification And Its Correlation With IgA tTG Titer

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Abstract

Background: Celiac disease is also known as Celiac sprue or gluten sensitive enteropathy. Celiac disease is a genetically determined autoimmune like disorder induced by gluten, the storage protein of wheat and by similar protein found in barley and rye. It has a strong genetic background, as suggested clearly by studies on first degree relatives of Celiac patients, on twins and on HLA genes associated with the susceptibility for the disease.

Methods: This study was hospital based study on 102 duodenal biopsy specimen coming to the Department of Pathology Sardar Patel Medical College & Associated group of Hospitals, Bikaner during the study period. The present study was undertaken to histopathological evaluation of duodenal biopsies in Celiac disease on the basis of Marsh classification and its correlation with IgA tTG titre

Results: Mean tTG titer in grade I was 5.71±2.21, in grade II was 7.0±0, in grade IIIa was 56.40±38.17, in grade IIIb was 85.20±52.12 and in grade IIIc was 175.83±66.56. The tTG titer was increased with Marsh grading I to IIIc. The correlation between Marsh grading

and tTG titer was found statistically significant (p-value 0.001).

Conclusion- tTG titer was increased with higher Marsh grading i.e. from grade I to IIIc, so biopsy could be avoided in patients specially in children.

Keywords: Celiac Disease, Small Intestine, Histopathology.

Introduction

Celiac disease is also known as Celiac sprue or gluten sensitive enteropathy.¹ Celiac disease is a genetically determined autoimmune like disorder induced by gluten, the storage protein of wheat and by similar protein found in barley and rye.^{2,3} It has a strong genetic background, as suggested clearly by studies on first degree relatives of Celiac patients, on twins and on HLA genes associated with the susceptibility for the disease.^{4,5}

Patients known to have Celiac disease have to undergo a life-long gluten free diet (GFD), and gluten withdrawal from the diet generally leads to complete recovery of the morphological changes.⁶ Several serologic tests have been developed to detect patients with Celiac disease: antigliadin IgG antibodies have a poor specificity, antigliadin IgA antibodies a poor sensitivity. The

detection of antiendomysial IgA antibodies (EMA) by immunofluorescence, although considered as the “gold standard” of serological Celiac disease markers.⁷ The enzyme tissue transglutaminase (tTG) was recently identified as the major autoantigen in Celiac disease and the antigenic target recognized by EMA. A human recombinant form of tTG was used to develop an ELISA to measure anti tTG serum antibodies for the diagnosis of Celiac disease. Preliminary retrospective reports suggest that the human tTG based ELISA could identify celiac patients missed by the IgA EMA test. Systematic review of the available studies revealed that the IgA tTG antibody test has greater than 90% sensitivity and specificity for Celiac disease.⁸⁻¹⁰

Marsh classification for histopathology of Celiac disease; Type I or infiltrative lesion include villi architecturally within normal morphological limits (normal villi/crypt ratio 3:1) and increased number of intraepithelial lymphocytes (greater than 25-30 per 100 epithelial cells). Type II or hyperplastic lesion include villi architecture like type I, increased number of intraepithelial lymphocytes like type I and hyperplasia of glandular elements (regenerative aspect of the glandular elements highlighted by the reduced muciferous activity and increased number of mitoses). Type III or destructive lesion include various degree of villous atrophy associated with hyperplasia of glandular crypts; reduced surface enterocyte height, with irregular brush border and sometimes cytoplasmic vacuoles; and increased number of intraepithelial lymphocytes (like type I and II lesions).¹¹

Materials & Methods

This study was carried out in department of Pathology, Sardar Patel Medical College & Associated group of Hospitals, Bikaner. This study was hospital based study

on duodenal biopsy specimen received in the department of Pathology during the study period.

Inclusion Criteria

Duodenal biopsy specimen in clinically suspected patients of Celiac disease.

Exclusion Criteria

- 1) Specimen without clinical detail
- 2) Autolysed specimen.

Clinical data was obtained from hospital record and requisition submitted along with tissue specimen received in the department. Tissue bits was routinely processed. Sections was made from paraffin blocks and was stained with H&E stain. Special stains were done whenever necessary. Specimen obtained from eligible study population was examined microscopically.

Data Analysis

To collect required information from eligible patients a pre-structured pre-tested proforma was used. For data analysis Microsoft excel and statistical software SPSS was used and data was analyzed with the help of frequencies, figures, proportions, measures of central tendency and appropriate statistical test.

Observations

This study was a hospital based study on 102 duodenal biopsy specimen coming to the Department of Pathology Sardar Patel Medical College & Associated group of Hospitals, Bikaner during the study period. Duodenal biopsy specimens were undertaken to histopathological evaluation of in Celiac disease on the basis of Marsh classification and its correlation with IgA tTG titres was done.

Maximum 38 (37.25%) patients were of 0-10 yrs age group followed by 35 (34.13%) patients were of 11-20 yrs age, 12 (11.76%) patients were of 21-30 yrs age group, 7 (6.86%) patients were of 41-50 yrs age group and 4

(3.92%) patients were of more than 50 yrs age. Maximum 55 (53.92%) patients were males and 47 (46.08%) patients were females.

Table no. 1. tTG titer wise distribution

tTG titer	No. of patients (n=102)	Percentage (%)
<15 IU/mL	7	6.87
≥15 IU/mL	95	93.13
Total	102	100.00
Level tTG titer (mean±SD)	86.63±79.63 IU/ml	

Table no.1 shows that 95 (93.13%) patient’s tTG titer was more than 15 IU/ml and 7 (6.87%) patient’s tTG titer was less than 15 IU/ml. The mean titer was 86.63±79.63 IU/ml.

Table no. 2. Marsh grading wise distribution

Marsh grading	No. of patients (n=102)	Percentage (%)
I	7	6.86
II	1	0.98
IIIa	61	59.80
IIIb	5	4.90
IIIc	28	27.45
Total	102	100.00

Table no.2 shows that in our study maximum 61 (59.80%) patients were with Marsh grading IIIa followed by 28 (27.45%) patients with Marsh grading IIIc, while patients with Marsh grading I, IIIb & II were 7 (6.86%), 5 (4.90%) & 1 (0.98%) respectively.

Table no. 3. Correlation between Marsh grading and tTG titer

tTG titer / Marsh grading	I	II	IIIa	IIIb	IIIc	p-value
Mean	5.71	7.0	56.40	85.20	175.83	0.001
SD	2.21	0	38.17	52.12	66.56	

Table no.3 shows that mean tTG titer in grade I was 5.71±2.21, in grade II was 7.0±0, in grade IIIa was 56.40±38.17, in grade IIIb was 85.20±52.12 and in grade IIIc was 175.83±66.56. The tTG titer was increased with Marsh grading I to IIIc. The correlation between Marsh grading and tTG titer was found statistically significant (p-value 0.001).

Discussion

This study was hospital based study on 102 duodenal biopsy specimen coming to the Department of Pathology Sardar Patel Medical College & Associated group of Hospitals, Bikaner during the study period. The present study was undertaken to histopathological evaluation of duodenal biopsies in Celiac disease on the basis of Marsh classification and its correlation with IgA tTG titre.

Celiac disease is an under-diagnosed gluten-sensitive enteropathy, often presenting with atypical/extraintestinal features and even as a latent or silent disease, thereby contributing to the 'Celiac iceberg'. The ratio of diagnosed versus undiagnosed cases is as high as 1 : 7.¹² The prevalence among school children in India is 1%.¹³ Though population-based studies among adults in India are not available, Celiac disease is one of the most common causes of small bowel diarrhea.¹⁴ An apparent regional variation of occurrence in India has been seen, possibly due to differences in genetic predisposition, differences in consumption of wheat or both.¹⁵⁻¹⁶

In our study mean tTG titer in grade I was 5.71±2.21, in grade II was 7.0±0, in grade IIIa was 56.40±38.17, in grade IIIb was 85.20±52.12 and in grade IIIc was 175.83±66.56. The tTG titer was increased with Marsh grading I to IIIc. The correlation between Marsh grading and tTG titer was found statistically significant (p-value 0.001).

Azita Ganji et al (2016)¹⁷ detected a significant correlation between anti-tTG titer and the degree of GI tract mucosal atrophy. Our study also showed tTG \geq 200 IU/mL which is 100% specific for Marsh III. Atieh Rahmati et al (2014)¹³ observed that 159 patients with tTG titer and pathology reports were enrolled in their study, Mean \pm SD of the patients was 35.6 \pm 15.2 IU/mL which is 100% specific for Marsh III.

Bhattacharya M et al suggested that in symptomatic patients duodenal biopsy can be avoided if tTG level is more than 100 U/mL (kit value of >10 as positive).¹⁸ Value of more than 10 times of normal limit was associated with villous atrophy of the GI tract mucosa and more severe clinical presentations with sensitivity and specificity of 98% and 99% respectively. Atieh Rahmati et al (2014)¹³ observed that mean tTG titer was 35.6 \pm 15.2 U/mL.

In a study by Fernández-Bañares and colleagues, tTG titer of at least 11.4 times of normal had a PPV of 98.6%. In all these studies the researchers found that more than 10 times of normal level for tTG in adults could be diagnostic for villus atrophy as it is diagnostic in children. In our study 93% of the patients with Marsh III had tTG more than 76 IU/ml and 100% of the patients with anti tTG \geq 200 IU/ml (10 times of normal value) had Marsh III.¹⁹

In the year 2011 a study conducted by Shivani Kalhan²⁰ observed that correlation between tTG levels and Marsh grades in adult Celiacs. tTG levels of all (except one) cases were raised. They found a statistically significant increase in tTG values from histologically milder forms of the disease to more severe forms ($P<0.001$). tTG values in grades IIIC were highest amongst all (354.66 \pm 111.43 U/ml). Patients in latent category were diagnosed on the basis of increased tTG levels. They had one case with tTG level of 11 U/ml, who presented with classic GI symptoms

with iron deficiency anemia and had Marsh grade IIIc on biopsy.

Conclusion

Patients with Celiac disease show specific histological changes in the form of atrophy (either partial, subtotal or total) of villi with lymphocytic infiltration, in these patients tTG titer was found raised, so after analyzing the results of present study it was concluded that tTG titer was increased with higher Marsh grading i.e. from grade I to IIIc, so biopsy could be avoided in patients specially in children.

References

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Legends Figure

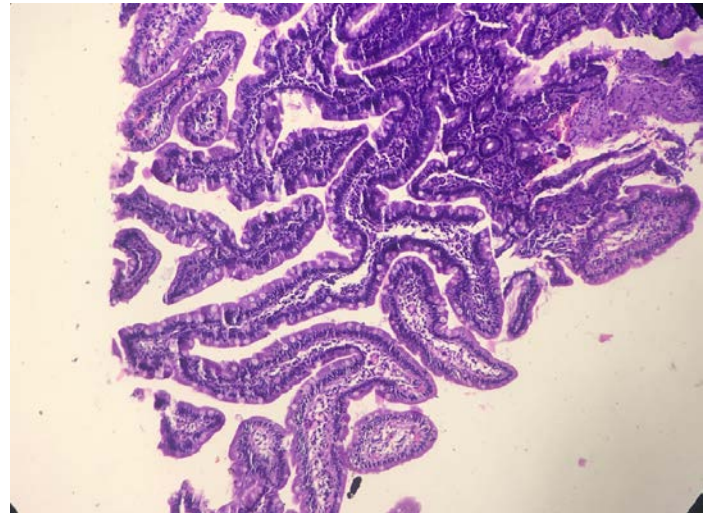


Fig.1. Marsh grade I (Duodenal Biopsy): Histopathological slide of patient showing intra epithelial lymphocytes (H & E 10x)

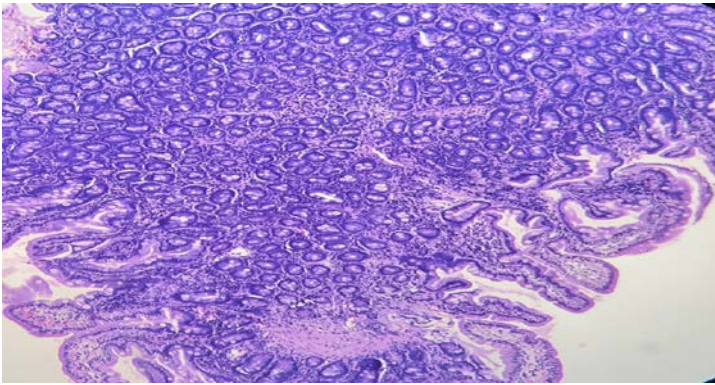


Fig 2. Marsh grade II (Duodenal Biopsy): Histopathological slide of patient showing Crypt Hyperplasia with Intra Epithelial Lymphocytes (H & E 10x).

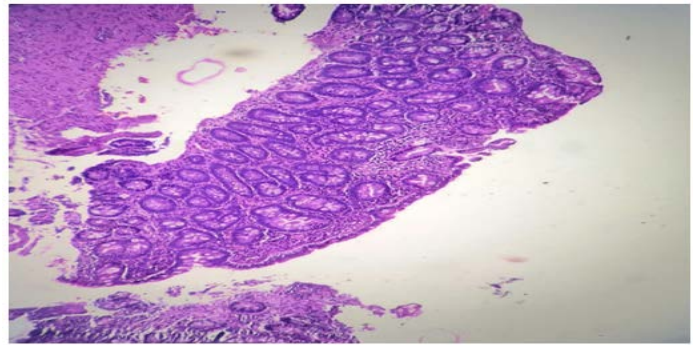


Fig 5. Marsh grade III c (Duodenal Biopsy): Histopathological slide of patient showing total flattening of Villi (H & E 10x)

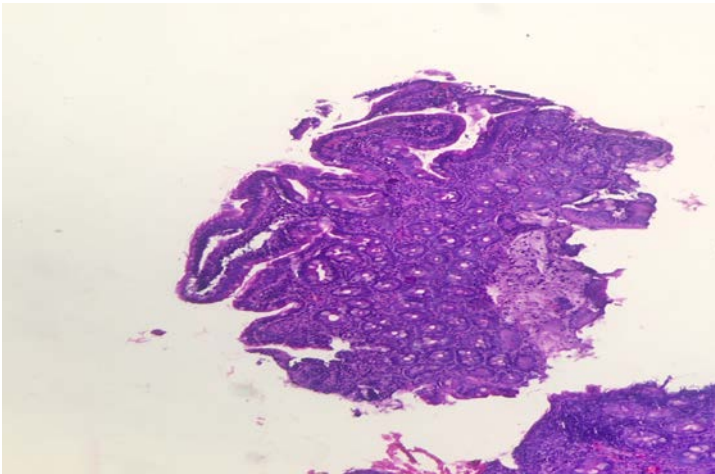


Fig 3. Marsh grade III a (Duodenal Biopsy): Histopathological slide of patient showing partial flattening of Villi (H & E 10x).

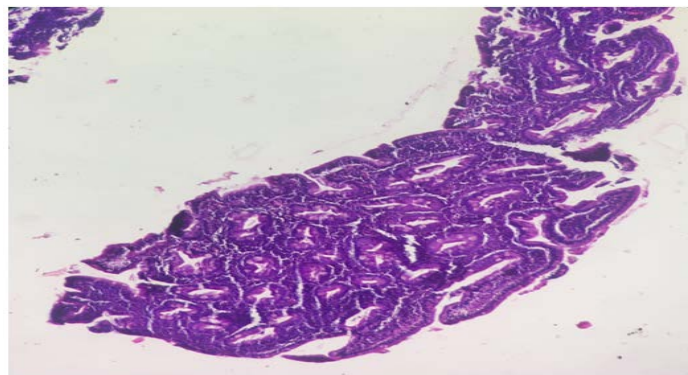


Fig 4. Marsh grade III b (Duodenal Biopsy): Histopathological slide of patient showing moderate (subtotal) flattening of Villi (H & E 10x)