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Histopathological Spectrum of Duodenal Biopsies in Seropositive Pediatric Celiac Disease: A Retrospective Study from Yazd, Iran

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ABSTRACT

Background: Celiac disease (CD) is an immune-mediated enteropathy triggered by gluten, with a global prevalence of approximately 1%. It is strongly associated with HLA-DQ2 and HLA-DQ8 haplotypes. Serological markers, particularly anti-tissue transglutaminase (tTG) immunoglobulin A (IgA), play a pivotal role in screening and diagnosis. This study aimed to evaluate the clinicopathological characteristics of duodenal biopsy specimens from patients with elevated serum tTG IgA levels.

Methods: This retrospective cross-sectional study analyzed 213 pediatric patients (age ≤16 years) with elevated anti-tTG IgA levels who underwent duodenal biopsy at Shahid Sadoughi Hospital, Iran (2016-2020). Demographic, clinical, and histopathological data were collected from medical records. Duodenal biopsies were classified using the Marsh-Oberhuber system. Statistical analysis was performed using SPSS version 22.

Results: The cohort Included 131 females (61.5%) and 82 males (38.5%), with a mean age of 5.9 years. Abdominal pain (58.7%) and failure to thrive (39%) were the most common clinical manifestations. Histopathological analysis revealed Marsh 3 lesions in 77% of cases (3a: 25.8%, 3b: 39.9%, 3c: 11.3%), while mild changes (Marsh 1-2) were observed in 23%. No significant association was found between Marsh classification and gender (P = 0.36).

Conclusion: Elevated anti-tTG IgA levels strongly predict severe mucosal damage in pediatric celiac disease, indicating that serology-based diagnosis could potentially reduce the need for invasive biopsies. Establishing validated antibody thresholds may allow for less invasive diagnostic approaches.

Introduction

Celiac disease (CD) is a chronic, immune-mediated enteropathy of the small intestine that develops in genetically predisposed individuals following exposure to dietary gluten. Since celiac disease is an autoimmune disorder with a strong genetic predisposition, and its onset during childhood can significantly affect growth, nutrition, and neonatal health, the histopathological evaluation of duodenal biopsies in affected children is particularly relevant to is particularly relevant to the fields of perinatology and neonatology [1, 2]. The condition is strongly associated with specific human leukocyte antigen (HLA) haplotypes, namely HLA-DQ2 (present in 90-95% of patients) and HLA-DQ8 (found in 5-10% of patients) [3].

CD is not confined to the gastrointestinal tract; it is a systemic disorder with a wide range of clinical manifestations[1, 4.] Presentations range from a classic malabsorption syndrome to more atypical or non-classical forms, such as iron-deficiency anemia, which is now frequently recognized [5].

A definitive diagnosis of CD requires a histological examination of duodenal biopsies while the patient remains on a gluten-containing diet. Current guidelines recommend collecting four to six biopsy samples, including those from the duodenal bulb, to maximize diagnostic accuracy. The histopathological lesions are commonly classified using the Marsh, Marsh-Oberhuber, or Corazza grading systems[1]

Serological screening is a key component in the diagnostic process. The detection of anti-endomysial antibodies (EMA) and anti-tissue transglutaminase antibodies (TTG) has markedly improved the efficacy of both CD screening programs and diagnostic protocols [1]. These serological markers are well recognized for their high specificity and sensitivity in detecting the disease [6].

Recent studies have investigated the correlation between tissue transglutaminase (tTG) antibody levels and the extent of mucosal damage to assess whether this relationship has a sufficiently high positive predictive value (PPV) for diagnosing celiac disease. This approach is considered advantageous due to its less invasive nature and lower associated costs. Recent evidence suggests that the severity of duodenal histological changes correlates strongly with tTG antibody titers. Consequently, it has been proposed that duodenal biopsies could potentially be omitted in patients whose clinical history and manifestations strongly suggest celiac disease [1].

Celiac disease is estimated to affect approximately 1% of the global population[2]. Notably, prevalence in Iran is reported to be relatively high, affecting about one in every 104 individuals. Given the widespread availability and use of the tissue transglutaminase (tTG) antibody test for screening in this context, serological testing offers a cost-effective diagnostic strategy, especially for certain patient subgroups[1]. However, local data regarding the relationship between serological findings and histopathological features remain limited.

Therefore, this study aimed to assess the clinicopathological characteristics of duodenal biopsy specimens from pediatric patients with elevated serum tTG antibody levels in Yazd, Iran.

Materials and Methods

This retrospective, cross-sectional study was conducted in the pathology department of Shahid Sadoughi Hospital, a tertiary care center in Yazd, Iran. Data from January 2016 to December 2020 were utilized in this study. The study population included all pediatric patients referred to the department for duodenal biopsy who presented with elevated serum tTG IgA levels. Patients with incomplete medical records or insufficient biopsy material were excluded from the analysis.

Data were collected from pathology reports and clinical records. Variables included:

- Demographic data: age and gender.
- Clinical symptoms: failure to thrive, abdominal distension, nausea, vomiting, fatigue, loss of appetite, vertigo, constipation, weight loss, abdominal pain, anemia, diarrhea, and flatulence.
- Histopathological data: Duodenal biopsy findings were classified using the Marsh-Oberhuber system (Type 0 to Type 3c).

Results

This study analyzed 213 duodenal biopsy specimens from patients with elevated anti-tissue transglutaminase (tTG) immunoglobulin A (IgA) levels. The cohort included 82 males (38.5%) and 131 females (61.5%). Participants had a mean age of 5.9 years (range: 1–16 years). Patients exhibited a broad range of clinical manifestations. Abdominal pain was the most common symptom, reported in 125 patients (58.7%). The second most common clinical manifestation was failure to thrive, observed in 83 patients (39%). In contrast, fever and bloating were the least frequent symptoms, each reported in only 3 patients (1.4%).

Histopathological analysis of biopsy findings from patients with elevated anti-tissue transglutaminase (tTG) immunoglobulin A (IgA) levels, classified using the Marsh-Oberhuber criteria, revealed the following distribution: 23 cases (10.7%) Marsh 1, 27 cases (12.4%) Marsh 2, 55 cases (25.8%) Marsh 3a, 85 cases (39.9%) Marsh 3b, and 23 cases (11.2%) Marsh 3c.

Analysis of Marsh classification distribution by gender revealed no statistically significant association between sex and histopathological severity. Among female patients (n = 131), the distribution was as follows: Marsh 1: 17 cases (12.7%), Marsh 2: 12 cases (9.1%), Marsh 3a: 37 cases (28.2%), Marsh 3b: 51 cases (39.1%), and Marsh 3c: 14 cases (10.9%). Among male patients (n = 82), the distribution was: Marsh 1: 5 cases (7.4%), Marsh 2: 14 cases (17.6%), Marsh 3a: 18 cases (22.1%), Marsh 3b: 34 cases (41.2%), and Marsh 3c: 10 cases (11.8%). Chi-square analysis confirmed that the association between gender and Marsh classification was not statistically significant (P = 0.36).

Discussion

Before the introduction of anti-tissue transglutaminase (tTG) antibody testing, serological diagnosis and screening for celiac disease primarily depended on anti-gliadin antibodies (AGA) and anti-endomysial antibodies (EMA). Although EMA showed exceptionally high specificity, the inadequate sensitivity of these assays led to a considerable number of false-negative results, limiting their utility in clinical and diagnostic practice. In 1997, Dietrich et al. identified tTG as the primary autoantigen recognized by endomysial antibodies. This discovery enabled the development of a sensitive and specific enzyme-linked immunosorbent assay (ELISA) for tTG antibody detection, which has since become a cornerstone of celiac disease diagnostics. Subsequent Over the past decade, subsequent studies have shown that serum tTG antibody titers vary with the extent of intestinal mucosal damage and correlate significantly with the histopathological severity defined by the Marsh classification system [7-9].

In 2005, prior to the widespread clinical adoption of anti-tissue transglutaminase (tTG) immunoglobulin A (IgA) testing, Barker and colleagues suggested that intestinal biopsy might not be necessary for the diagnosis of celiac disease in pediatric populations exhibiting markedly elevated tTG antibody titers[10]. Subsequently, Vivas et al. Confirmed these findings, showing that duodenal biopsy could potentially be omitted in children with strongly positive tTG serology[11]. Reflecting this evolving evidence, the European Society for Paediatric Gastroenterology, Hepatology and Nutrition (ESPGHAN) updated its celiac disease diagnostic guidelines after two decades, formally acknowledging that duodenal biopsy may be omitted in a specific subset of patients meeting stringent serological and clinical criteria[12]

This diagnostic approach was been further validated by Mubarak et al., showing that

small intestinal biopsy may be omitted in symptomatic patients with anti-tissue transglutaminase antibody (tTGA) levels ≥100 U/mL[8]. Similarly, Alessio et al. reported that in adult and pediatric populations, a positive anti-tTG serology result ≥7 times the upper limit of normal (ULN), along with a positive endomysial antibody (EMA) test, indicates a high probability of duodenal damage. Under these specific conditions, duodenal biopsy may be avoided for confirming celiac disease[7]. Most recently, a 2023 study by Wang et al. showed that an anti-tTG level ≥5 times the ULN can diagnose celiac disease with a sensitivity of 83% and specificity of 93%[13].

Furthermore, studies have shown that patients who adhering strictly to a gluten-free diet exhibited significantly lower anti-tTG antibody levels compared to those with poor dietary compliance[14].

In this study, abdominal pain, growth restriction, and diarrhea were the most common clinical manifestations. Rahmati et al. similarly reported abdominal bloating, abdominal pain, and diarrhea as the most common symptoms in patients with elevated tTG levels. except for growth restriction, the two most frequent symptoms in our study align with those reported by Rahmati et al.[15]. Correspondingly, a 2022 Dutch study conducted by Majsiak et al. identified abdominal pain, bloating, and diarrhea as the predominant clinical manifestations in celiac disease patients, consistent with our study's results, despite the lower reported prevalence of bloating in our cohort [16].

A 2023 study by Wang et al. on celiac disease patients with elevated anti-tTG levels reported the following Marsh classification distribution: 6% Marsh 1, 9% Marsh 2, and the remaining 85% Marsh 3a–3c [13]. Our study found comparable frequencies: 10% Marsh 1, 12% Marsh 2, and 78% Marsh 3a–3c, indicating notable consistency in histopathological severity across both cohorts.

Although anemia was infrequent in our study, reported by only six patients, Parveen et al. found significantly lower hemoglobin levels in seropositive celiac disease patients (anti-tTG positive) compared to seronegative patients (anti-tTG negative)[17]. This discrepancy may be due to differences in study populations, diagnostic criteria, or the sensitivity of anemia detection methods.

Kalhan et al. found no significant difference in mean age across Marsh classification categories, consistent with our study. Furthermore, while various clinical manifestations were assessed according to Marsh severity, their frequency distribution likewise demonstrated no statistically significant association. In contrast to our findings, however, Kalhan et al. reported a significant association between the prevalence of anemia and Marsh stage [18]. Considering the pathophysiology of celiac disease, which involves intestinal malabsorption, anemia in these patients is highly plausible. Indeed, numerous studies recognize anemia as one of the most common clinical manifestations of celiac disease[19]. The discrepancy between our study and previous research may be attributed to incomplete medical records, as our data were retrospectively collected from patient files.

This study was conducted on a population with elevated anti-tissue transglutaminase (tTG) immunoglobulin A (IgA) levels. Since previous research has demonstrated that a tTG level exceeding ten times the upper limit of normal (ULN) can diagnose celiac disease with 100% specificity[20], Therefore, future studies should consider the ratio of the patient's tTG level to the ULN to improve diagnostic and analytical precision.

Conclusion

Although duodenal biopsy remains the diagnostic gold standard for celiac disease, anti-tTG IgA titers strongly correlate with histopathological severity and may reliably predict mucosal damage in specific clinical contexts. These findings support the development of serology-based diagnostic protocols that could reduce invasive procedures in pediatric patients with markedly elevated antibody levels. Future studies should

aim to validate precise tTG thresholds to optimize non-invasive diagnostic strategies for pediatric celiac disease.

Conflict of Interest

The authors declare that there is no conflict of interest.

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Ethical Considerations

The study protocol was reviewed and approved by the Ethics Committee of Shahid Sadoughi University of Medical Sciences (Ethics Code: IR.SSU.MEDICINE.REC.1400.156). Patient confidentiality was maintained throughout the study.

Author's Contribution

Conceptualization, M.S., M.V.; methodology, S.E.; formal analysis, S.E.; investigation, S.E., M.S.; original draft preparation, M.V.; review and editing, S.E.; All authors have read and agreed to the published version of the manuscript.

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