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Deamidated gliadin peptide in pediatric patients with moderately increased tissue transglutaminase; does it help?

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ABSTRACT

Background: Deamidated gliadin peptide (DGP) is a relatively new serologic assay used in diagnosis and monitoring of celiac disease. DGP IgG is recommended by some in pediatric patients < 2 y. Use in other pediatric populations is not well established. The utility of the DGP screen (IgG + IgA) in patients with moderate increase of tissue transglutaminase (TTG) IgA has not been studied.
Methods: Cases between January 2015 and October 2017 in which a patient had TTG IgA greater > 19 and < 100, DGP screen, and biopsy were collected. Indication for biopsy and diabetes diagnosis were recorded. Of 495 patients screened, 31 met criteria.
Results: The sensitivity and specificity of DGP screen were calculated, and were 87.4% and 56%, respectively; though lower in patients with diabetes.
Conclusions: The study suggests in patients with moderately increased TTG-IgA, DGP screen lacks specificity and does not provide additional information about whether or not to biopsy.

1. Introduction

Celiac disease (CD) is a common medical condition with prevalence in both children and adults of nearly 1% of the population [1]. Screening serologic testing is utilized to identify individuals at high risk of celiac disease, and upper endoscopy with evaluation of duodenal biopsy specimens is the gold standard for diagnosis. In children, accurate screening helps to guide the decision to move towards confirmatory upper endoscopy. Because endoscopy carries risk, and time under general anesthesia may impact the developing brain, screening is of particular importance in children [2]. Accurate serologic tests can minimize unnecessary procedures, and for select children, the European Society for Pediatric Gastroenterology, Hepatology, and Nutrition (ESPGHAN) has suggested that serologic testing without biopsy is an appropriate modality to diagnose celiac disease [3].

Tissue transglutaminase (TTG) IgA is a serology with high sensitivity and specificity for the detection of celiac disease and has been recommended as the single screening test for celiac disease in individuals > 2 y [4]. Whereas anti-gliadin antibodies do not have high sensitivity or specificity for celiac disease, testing antibodies to deamidated gliadan peptide (DGP) does have a role in children < 2 y, individuals with IgA deficiency, and potentially in combination with other serologic tests [4,5]. While numerous studies have suggested the sensitivity and specificity of TTG IgA to be > 95%, high quality studies evaluating the utility of DGP serologic testing are less clear [6]. Wolf et al. evaluated antibody-based approaches to diagnose CD without biopsy and observed significant scatter in correlating DGP IgG and TTG IgA when mild-to-moderately increased [7].

In pediatric patients it is unclear if all patients should proceed to biopsy when mild-to-moderate increases of TTG IgA antibodies are found. Patient's with Type 1 diabetes mellitus are of particular interest because they are frequently screened for celiac disease rather than presenting with symptomatology suggestive of the diagnosis. The goal of this study was to evaluate the utility of DGP antibody screen (IgA and IgG) in the setting of mild-to-moderate increases of TTG IgA antibodies.

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2. Methods

2.1. Patients

This study was approved by the Seattle Children's Hospital Institutional Review Board.

A search of the Seattle Children's Hospital laboratory information system from January 2015 to October 2017 identified all patients < 21 y with a mild to moderate increase of TTG-IgA (20–99 units), a measured DGP screen (combined IgA + IgG), and a duodenal biopsy performed. Only cases in which serology was completed before the biopsy and biopsy was completed within 1 y of both laboratory tests were included. The patient's age, sex, TTG-IgA concentration, and DGP screen concentration were recorded, and the DGP screen was dichotomized to positive (≥20 U/ml) or negative (< 20 U/ml).

The electronic medical record was reviewed to determine the indication for screening. Additionally the chart was searched to determine whether or not the patient had a prior diagnosis of celiac disease, was on a gluten-free diet at the time of biopsy, had type 1 diabetes mellitus (T1DM), or had a diagnosis of Trisomy 21. Patients with a prior diagnosis of CD or on a gluten-free diet were excluded.

Once all cases had been screened and pathology reviewed, the clinical sensitivity and specificity for testing was calculated along with the negative and positive predictive value. For analysis, biopsy findings were used as the gold standard for diagnosis with a positive biopsy showing changes associated with Marsh 1,2, 3, or 4 pathology. Marsh 1 histology was considered positive because children with this histology combined with increased serologies and clinical suspicion for celiac disease are started on a gluten-free diet at our institution and closely followed [8]. Normal duodenal mucosa (Marsh 0) was categorized as negative for celiac disease.

2.2. DGP IgA + IgG screen and anti-TTG IgA assays

At Seattle Children's Hospital, TTG-IgA and DGP screen were performed using Inova QuantaLite reagents. For both TTG-IgA and DGP screen, the normal cut-off was < 20 U/ml and the upper end of the analytical measurement range was 100 U/ml.

2.3. Biopsy review

All duodenal biopsies were reviewed and graded using the Marsh classification system by the study pathologist, MCP, based on the area within the biopsy which was most affected [9]. The number of tissue fragments within the biopsy was recorded. Increased intraepithelial lymphocytes was defined as greater than or equal to 30 lymphocytes per 100 epithelial cells.

3. Results

3.1. Demographics

Four hundred and ninety-five patient records were screened and 31 patients met all inclusion criteria (Table 1). The mean age at time of biopsy was 11.8 y (range 2–20 y). Fifty-five percent of patients were male (17) and 45% were female (14). The indications for serologic testing were as follows: T1DM, 13 patients; abdominal pain, 8 patients; and "other," 9 patients. All but one patient who was considered refractory to a gluten-free diet were on a gluten-containing diet at the time of biopsy.

3.2. Primary outcomes

The patient's TTG-IgA and DGP screen results were compared with the Marsh scores. None of the patients had a Marsh score of 4. The patient on a gluten-free diet had normal histology in spite of abnormal serology. Seven patients had a positive TTG-IgA and DGP screen results with negative biopsies. The indication for biopsy in four of these patients was T1DM and the remaining indications were abdominal pain, failure to thrive, and emesis. The average number of tissue fragments within the biopsies was 5.2 and 23 patients had ≥5. Four of the patients with < 5 tissue fragments were scored as Marsh 0.

Table 1
Patients with TTG IgA 20–99 units between January 2015 and October 2017 and screen findings.

Biopsy status	DGP positive	DGP negative	No DGP performed
No biopsy (n = 332)	**94**[a]		**238**
Positive biopsy (n = 102)	**17**	**3**	**82**
[b]T1D (N = 8)	7	1	
[b]Without T1D (N = 7)	6	1	
Negative biopsy (n = 61)	**8**	**11**	**42**
[b]T1D (N = 5)	4	1	
[b]Without T1D (N = 11)	3	8	

[a] DGP Screen results were not categorized as positive or negative – without a paired biopsy, they were excluded from the analysis.

[b] An additional 8 patients with biopsy, DGP, and mild TTG-IgA were excluded due to known celiac disease (n = 3), gluten free at time of biopsy (n = 2), or > 1 year between biopsy and serology n = (4). The patients included in the final analysis are further classified as T1D and Without T1D.

Overall, the sensitivity and specificity of the DGP screen was 86.7% and 56.3%, respectively. For those with T1DM the specificity fell to 20%; whereas those patients without diabetes had increased specificity to 82%. The positive predictive value in all groups was about the same at 65%, but the negative predictive value was particularly low in the T1DM group, 50%, see Table 2.

4. Discussion and conclusions

This retrospective study involved 31 children who had mild to moderate increases in TTG-IgA, who had a DGP screen, and who subsequently underwent endoscopic duodenal biopsy. The clinical sensitivity and specificity of the DGP screen was evaluated in this population, and was 86.7% and 56.3%, respectively. The study population was further categorized into two groups: T1DM (n = 13) and non-diabetic (n = 18). The DGP screen performed poorly in the group with T1DM (specificity 20%); however, in the non-diabetic group, the specificity and negative predictive value improved (82% and 90%, respectively). This suggests that in a symptomatic population, the DGP may be able to reduce false positives from mild to moderate increases in TTG-IgA and avoid unnecessary biopsies. It is possible that the lower specificity in the T1DM group is because this population is screened for celiac disease due to the known association between the diseases. This may result in detection of increased TTG-IgA prior to histologic manifestations of the disease. Alternately, patients with TIDM may have increased false positive serologies. In this case, a false positive means that the increased measurement of TTG-IgA and DGP screen do not correlate with a clinical/biopsy diagnosis of CD.

While there are known groups of asymptomatic children at high risk for developing CD, the value of screening with serology is still not fully understood. TTG-IgA has an excellent diagnostic sensitivity and specificity in a symptomatic population, 100% and 97% respectively [10].

Table 2
Sensitivity and specificity of DGP in patients with TTG IgA 20–99 units.

	Sensitivity	Specificity	Positive predictive value	Negative predictive value
All patients	87%	56%	65%	82%
T1DM	88%	20%	64%	50%
Non-T1DM	86%	82%	67%	90%

Recent reports have suggested that the ESPGHAN algorithm (beginning with a high titer TTG-IgA > 10× upper limit of normal) could also be applied to asymptomatic screening populations [11]; however, these studies only evaluated TTG IgA > 10× upper limit of normal, and did not comment on the frequency or disease-correlation with moderate increases.

Limitations to our study include the potential bias introduced by the strict inclusion criteria. The criteria of mild increase in TTG IgA might introduce ascertainment bias, but not all screened subjects with increase in TTG IgA underwent duodenal biopsy. Likewise, not all subjects with TTG IgA screen had DGP screening. Because DGP is not commonly used as a primary screening serology, a significant proportion of individuals in our study did not have DGP screening (Table 1). DGP screen measures combined DGP IgA and DGP IgG, which is less studied in the literature than independent measures of DGP IgA and DGP IgG. Our institution preferred the screen approach because it was cost-effective, and simplified the ordering strategy. Four of our patients with normal biopsies had < 5 biopsy fragments which may have affected the sensitivity of our biopsy results. Our study demonstrates the lack of a strong correlation between mild increase in TTG IgA and DGP positivity, and our findings suggest the performance of DGP screening has less predictive power than initially suggested in the literature. In fact, a recent similar study found that patients with DGP-IgG positive and TTG-IgA negative serologies went on to have negative biopsies, suggesting that DGP-IgG does not effectively distinguish patients with and without CD [12].

This study was designed to assess the performance of DGP when TTG-IgA was mildly to moderately increased (< 10× upper limit of normal), which limited the candidate pool. However, this group of patients is of greatest concern because distinguishing when to proceed with biopsy is unclear. Our study suggests that serologic screening in at risk, asymptomatic Type 1 diabetic patients will result in a higher number of moderate increases of DGP; without adding specificity thereby negating the benefit of testing. In patients without T1DM and with symptomatology presumed suspicious for celiac disease, the performance of DGP testing is somewhat improved but not better than TTG-IgA. Based on the small number of cases studied it seems that in the diagnosis of celiac disease TTG IgA is the best test and sensitivity and specificity are not aided by DGP screen; biopsy remains the gold standard of diagnosis for patients with moderately increased TTG-IgA. However, the question remains whether DGP performs better after diagnosis as a marker of ongoing duodenal inflammation. Future prospective studies are needed to better understand the limitations and advantages of additional serologic screening assays for patients with moderate increases in TTG-IgA.

References

[1] P.H. Green, C. Cellier, Celiac disease, N. Engl. J. Med. 357 (17) (2007) 1731–1743.
[2] D.B. Andropoulos, M.F. Greene, Anesthesia and developing brains - implications of the FDA warning, N. Engl. J. Med. 376 (10) (2017) 905–907.
[3] S. Husby, S. Koletzko, I.R. Korponay-Szabó, et al., European Society for Pediatric Gastroenterology, Hepatology, and Nutrition guidelines for the diagnosis of coeliac disease, J. Pediatr. Gastroenterol. Nutr. 54 (1) (2012) 136–160.
[4] A. Rubio-Tapia, I.D. Hill, C.P. Kelly, A.H. Calderwood, J.A. Murray, ACG clinical guidelines: diagnosis and management of celiac disease, Am. J. Gastroenterol. 108 (5) (2013) 656–657.
[5] K. Kaukinen, P. Collin, K. Laurila, T. Kaartinen, J. Partanen, M. Mäki, Resurrection of gliadin antibodies in coeliac disease. Deamidated gliadin peptide antibody test provides additional diagnostic benefit, Scand. J. Gastro. 42 (12) (2007) 1428–1433.
[6] I. Hill, What are the sensitivity and specificity of serologic tests for celiac disease? Do sensitivity and specificity vary in different populations? Gastroenterology 128 (4 Suppl 1) (2005) S25–S32.
[7] J. Wolf, D. Petroff, T. Richter, et al., Validation of antibody-based strategies for diagnosis of pediatric celiac disease without biopsy, Gastroenterology 153 (2017) 410–419.
[8] M.E. Robert, S.E. Crowe, L. Burgart, R.K. Yantiss, B. Lebwohl, J.K. Greenson, S. Guandalini, J.A. Murray, Statement on best practices in the use of pathology as a diagnostic tool for celiac disease, Am. J. Surg. Pathol. 42 (9) (2018) e44–e58.
[9] M.N. Marsh, Gluten, major histocompatibility complex, and small intestine: a molecular and immunobiologic approach to the spectrum of gluten sensitivity ('celiac sprue'), Gastroenterology 102 (1992) 330–354.
[10] A. Carroccio, G. Vitale, L. Di Prima, et al., Comparison of anti-transglutaminase ELISAs and an anti-endomysial antibody assay in the diagnosis of celiac disease: a prospective study, Clin. Chem. 48 (9) (2002) 1546–1550.
[11] C.M. Trovato, M. Montuori, C. Anania, et al., Are ESPGHAN "biopsy-sparing" guidelines for celiac disease also suitable for asymptomatic patients? Am. J. Gastroenterol. 110 (10) (2015) 1485–1489.
[12] M.J. Gould, H. Brill, M.A. Marcon, N.J. Munn, C.M. Walsh, In screening for celiac disease, deamidated gliadin rarely predicts disease when tissue transglutaminase is normal, J. Pediatr. Gastroenterol. Nutr. 68 (1) (2019) 20–25.