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ORIGINAL ARTICLE

Diagnostic Yield of Isolated Deamidated Gliadin Peptide Antibody Elevation for Celiac Disease

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Abstract
Background Serologic testing for celiac disease includes tissue transglutaminase and endomysial antibodies. In addition to these tools, assays for deamidated gliadin peptide antibodies have been shown to have sensitivity and specificity that are comparable to tissue transglutaminase testing, and are increasingly being used for celiac disease testing.
Aims The goal of this study is to evaluate the utility of deamidated gliadin peptide (DGP) testing in the setting of a negative tissue transglutaminase (TTG) IgA test.
Methods We reviewed the records of all patients seen at two U.S. celiac disease referral centers and identified those who had an elevated DGP IgA and/or IgG in the setting of a negative TTG IgA. Of these patients, those who underwent duodenal biopsy while on a gluten-containing diet were included. Patients with prior biopsy-proven celiac disease or prior TTG IgA positivity were excluded. The results of the biopsy were used as the gold standard for celiac disease diagnosis, and patients with villous atrophy (Marsh class 3) on duodenal biopsy were considered to have celiac disease.
Results Between the two institutions, 84 patients were identified with negative TTG IgA and positive DGP IgA or IgG who also had duodenal biopsies performed while maintaining a gluten-containing diet. Of these patients, 13 patients (15.5%; 95% CI 8.5–25.0%) were found to have celiac disease on duodenal biopsy.
Conclusions DGP antibody testing can identify cases of celiac disease in TTG-negative individuals, although the low positive predictive value suggests that the yield may be low.

Keywords Celiac disease · Gliadin · Tissue transglutaminase · Serologic tests

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Introduction

Though the diagnosis of celiac disease (CD) requires intestinal biopsy demonstrating villous atrophy, serological testing has an important role in the identification of patients at low or intermediate risk of CD who would benefit from esophagogastroduodenoscopy and duodenal biopsy [1, 2]. Available serologies include endomysial IgA antibodies (EMA), anti-tissue transglutaminase IgA antibodies (TTG), and antigliadin antibodies (AGA). Given the lower sensitivity and specificity of AGA tests for CD, the EMA and TTG tests have replaced AGA serologic testing [3].

More recently, the deamidated gliadin peptide IgA antibody (DGP IgA) has been reported to have 94.6% sensitivity and 99.1% specificity for CD, and the DGP IgG antibody has been reported to have 92.4% sensitivity and 100% specificity for CD [4]. That initial study showed that both DGP IgA and IgG had better sensitivity and specificity than the conventional AGA test and comparable results to the TTG test. It also found that in false-negative TTG tests, DGP was helpful in detecting new CD patients in 3 of 4 cases. However, the positive predictive value of isolated elevated DGP IgA or IgG has not been adequately studied in large numbers of patients.

Some studies have suggested that combining results of TTG and DGP testing could improve test characteristics when compared to either test in isolation [5, 6]. This practice raises the question of what to do with discordant results, such as in the case of negative TTG and positive DGP testing. The discordant result can be challenging for clinicians to interpret. In one study, DGP testing identified 3 out of 12 TTG-negative patients with confirmed Marsh III duodenal biopsy findings [7]. In another study, discordant serologies were found in a similar minority of patients with CD and without CD [8]. This suggests that there may be low diagnostic utility in sending DGP serologies after obtaining a negative TTG result for CD screening because it still may not reliably identify CD patients.

In this study we aimed to evaluate the utility of DGP IgA and IgG antibodies in diagnosing CD when compared to the standard TTG test. Specifically, we evaluated the positive predictive value of a positive DGP IgA or IgG test in patients with negative TTG IgA testing who then underwent duodenal biopsy to test for CD.

Methods

Participants

We performed a cross-sectional study of patients attending two celiac disease referral centers during the years spanning 2009–2015: the Celiac Disease Center at Columbia University in New York, and The Celiac Center at Beth Israel Deaconess Medical Center in Boston. Although DGP is usually part of celiac disease testing along with TTG IgA at both institutions, DGP serologies are performed at the discretion of the ordering physician. Among patients with an elevated DGP result referred for further evaluation, TTG testing is performed if it has not already been done. Patients were identified by querying the electronic medical record for adult patients ≥18 years old who had TTG IgA and DGP serologies performed. Of these patients, only patients with negative TTG IgA, positive DGP IgA or IgG, and duodenal biopsy results were included. Patients were excluded if they had a previous diagnosis of CD, if they had a positive TTG IgA at any time, or if they were on a gluten-free diet at the time of duodenal biopsy.

Serologic Testing

At both institutions, TTG IgA testing was performed by Inova QUANTA Lite TTG IgA (Inova Diagnostics, San Diego, California). DGP testing methods differed by institution. At Columbia University, DGP testing was performed by Inova QUANTA Lite Gliadin IgA II and QUANTA Lite Gliadin IgG II kits. At Beth Israel Deaconess, DGP testing was performed via the Inova QUANTA Lite Celiac DGP Screen, which does not differentiate between IgA and IgG. All of the testing kits use a quantitative enzyme-linked immunosorbent assay. Values greater than or equal to 20 are considered positive.

Histology

The diagnosis of CD was confirmed by duodenal biopsy from the second portion of the duodenum and/or the duodenal bulb. Marsh grade III was considered to be diagnostic of CD [1].

Statistical Analysis

We calculated the positive predictive value (PPV) of isolated DGP antibodies by dividing the number of patients with CD by the number of patients with isolated DGP elevation. We stratified the population based on whether the DGP elevation was IgA only, IgG only, or unspecified. We report 95% confidence intervals for each calculated PPV. We compared proportions and mean serology values using the Chi-square test and unpaired student t test, respectively. All reported p values are two-sided. The Institutional Review Boards of Columbia University Medical Center and Beth Israel Deaconess Medical Center approved this study.

Results

Patient Characteristics

A total of 84 patients were included in the study, with 39 at Columbia University and 45 at Beth Israel Deaconess. The characteristics of the patients are shown in Table 1. By virtue of the inclusion criteria, all patients tested negative for TTG IgA and positive for DGP, either for IgA or IgG, or both subtypes. Overall 13 out of 84 patients were found to have CD on duodenal biopsy, yielding a positive predictive value (PPV) of 15.5% (95% CI 8.5–25.0%).

In addition to the 13 patients with Marsh III on duodenal biopsy, 12 out of 84 patients had Marsh I or II findings, which was not considered diagnostic of CD in this study (Table 1). Patients with CD presented with more classical symptoms of malabsorption such as diarrhea and weight loss than patients without CD, though this did not meet statistical significance ($p = 0.075$, Table 2). A minority of patients had endomysial antibody (EMA) testing. All of the patients in the isolated DGP IgA or IgG group who had EMA testing were negative, including one patient with CD in the isolated IgG group. One of the unspecified (combined) IgA/IgG DGP patients with CD had EMA testing and was positive; no other patients in the unspecified (combined) IgA/IgG DGP group had EMA testing. Higher DGP antibody titers were not associated with an increased probability of CD (Fig. 1). Mean DGP IgG titers and unspecified DGP IgA/IgG titers were higher in the CD group, although this was not statistically significant ($p = 0.345$ and 0.230, respectively).

Predictive Value Stratified by Immunoglobulin Subtype and by Center

Isolated DGP IgA antibodies in this study had the lowest positive predictive value at 3.7% (95% CI 0.1–19%, Table 3). The highest positive predictive value for CD was observed among those with unspecified (combined) IgA/IgG DGP elevations (22.2%; 95% CI 11.2–37.1%), which is the test used at Beth Israel Deaconess. This value is higher than the positive predictive value of either of the separate DGP IgA or IgG subclasses, as well as the positive predictive value of both of the separate tests combined (7.5%; 95% CI 1.57–20.4%). The isolated DGP IgA and DGP IgG subclass tests are used at Columbia University.

Selective IgA Deficiency

In the group of DGP IgG positive patients, two patients were found to have selective IgA deficiency, one of which had concomitant CD. In the group of unspecified (combined) IgA/IgG DGP positive patients, three patients were found to have selective IgA deficiency, one of which had concomitant CD. Total IgA data were missing for three patients in the DGP IgG positive group and four patients in the unspecified (combined) IgA/IgG group. Excluding selective IgA-deficient patients and patients with missing total IgA data from the analysis, the overall positive predictive value of DGP serologic testing was relatively unchanged (15.3%; 95% CI 7.9–25.7%).

Discussion

Despite its reported high level of sensitivity and specificity, the role of DGP serologies in diagnostic testing algorithms has remained uncertain, partly because the data for DGP testing is not as robust as the data for TTG testing. Recent literature calls into question the validity of DGP tests in clinical practice. In a prospective study by Volta et al. [9], the sensitivity of DGP IgA and IgG were found to be only 84.3 and 82.3%, respectively. Another study by Naiyer

Table 1 Presenting symptoms, serologies, and pathology results of patients from each institution

	CUMC ($n = 39$)	BIDMC ($n = 45$)	Total ($n = 84$)
Number of males	6 (15%)	13 (29%)	19 (23%)
Mean age (years)	49	45	47
Diarrhea	6 (15%)	23 (51%)	29 (35%)
Weight loss	7 (18%)	7 (16%)	14 (17%)
Vitamin deficiency or anemia	6 (15%)	9 (20%)	15 (18%)
Other (abdominal pain, neuropathy, family history, arthralgias, etc.)	21 (54%)	11 (24%)	32 (38%)
TTG negative	39 (100%)	45 (100%)	84 (100%)
DGP IgA or IgG positive	39 (100%)	45 (100%)	84 (100%)
Isolated DGP IgA positive	27 (69%)	N/A	N/A
Isolated DGP IgG positive	12 (31%)	N/A	N/A
Both IgG and IgA positive	1 (3%)	N/A	N/A
Unspecified (assay does not differentiate)	N/A	45 (100%)	N/A
Marsh score			
0	31 (79%)	28 (62%)	60 (71%)
1	4 (10%)	7 (16%)	11 (13%)
2	1 (3%)	0 (0%)	1 (1%)
3a/b/c	3 (8%)	10 (22%)	13 (15.5%)

BIDMC Beth Israel Deaconess Medical Center, *CUMC* Columbia University Medical Center, *N/A* not applicable

Table 2 Number of celiac disease (CD) and non-celiac disease patients (No CD) with classical malabsorption symptoms (diarrhea or weight loss) and with each serologic result

	CD ($n = 13$)	No CD ($n = 71$)
Classical symptoms	9 (69%)	30 (42%)
Isolated DGP IgA positive	1	26
Isolated DGP IgG positive	2	9
Unspecified (assay does not differentiate)	10	35

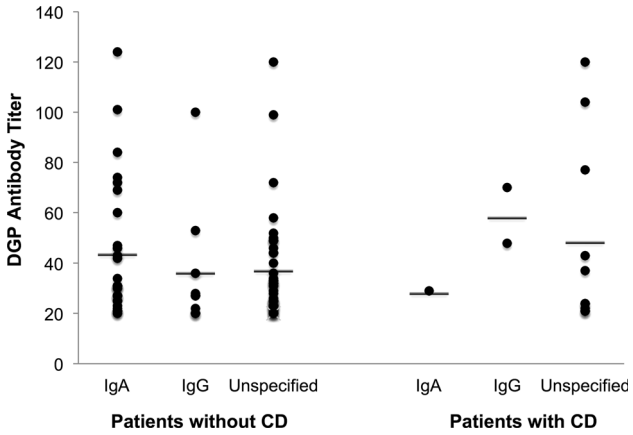

Fig. 1 Comparison of DGP antibody titers for IgA subtype, IgG subtype, and IgA/IgG unspecified subtype (assay does not differentiate) for patients with celiac disease (CD) and patients without celiac disease. *Bars* represent mean values. Values greater than or equal to 20 are considered positive

Table 3 Positive predictive value (PPV) with 95% confidence intervals (95% CI) for each test

	PPV (%)	95% CI (%)
Isolated DGP IgA positive	3.7	0.1–19.0
Isolated DGP IgG positive	18.2	2.3–51.8
Unspecified (assay does not differentiate)	22.2	11.2–37.1
Any DGP IgA or IgG positive	15.5	8.5–25.0

et al. [10] showed that in clinical practice, the sensitivities and specificities of DGP testing were lower than reported in earlier literature. One possible explanation for this finding is that the majority of the studies that evaluate the performance of DGP IgA include patients with elevated TTG and EMA serologies, when in fact its practical utility would be highest among TTG-negative patients with CD.

In this two-center study, we found that the overall positive predictive value of DGP serologic testing in the setting of a negative TTG IgA was 15.5%. This is considerably lower than would be expected based on the test characteristics demonstrated in other studies which tested this assay independent of TTG IgA results [4, 11–14]. There are multiple potential explanations for the low positive predictive value. Celiac antibody testing often does not perform as well as expected in clinical practice [15]. Previous studies have demonstrated poorer performance characteristics for DGP testing in patients with a low pretest probability of disease [14, 16]. In our study, these patients arguably had a lower pretest probability because they already had a negative TTG IgA. Another possibility is that the patients with positive DGP may not have overt CD, but may be in the early stages of developing this condition. In this study, 12 out of 84 patients (14.3%) had Marsh I or II findings. Prior studies have shown that DGP serologies have good sensitivity in patients with Marsh I/II biopsy findings and that DGP titers may correlate with the severity of histologic changes [9, 17]. This could be further studied in a prospective study following DGP positive patients over time to see if they eventually develop overt CD.

Prior studies have shown that higher DGP titers are more predictive of CD [5, 18]. This was not seen in this study, suggesting that the magnitude of DGP elevation is not informative of CD likelihood in the context of a negative TTG IgA.

Selective IgA deficiency is a potential application for DGP IgG testing, as patients with this condition will not develop TTG IgA antibodies, although the sensitivity may be lower in the setting of selective IgA deficiency [19]. In this study, several patients were found to have selective IgA deficiency. With these patients excluded, the overall positive predictive value of DGP serologic testing remained similar, 15.3%.

Limitations of this study include its cross-sectional design and small sample size, though it is the only study to our knowledge to investigate the phenomenon of isolated DGP elevation. The sample size was inherently limited because the number of patients who have TTG and DGP testing and then go on to continue a gluten-containing diet and undergo a duodenal biopsy with this combination of serological results is small. As we did not follow those CD-negative patients over time to see whether they eventually develop CD, it remains possible that an isolated DGP elevation could be a harbinger of future CD in a subset of patients. Our study design introduces the possibility of selection bias as there may have been patients with discordant serologies that did not undergo biopsy due to low pretest probability as estimated by the clinician. This could have overestimated the utility of DGP testing in this setting, meaning that the true positive predictive value is even lower than our calculation.

In conclusion, we found that the overall yield of isolated DGP elevation is low, 15.5%. Since CD remains underdiagnosed, it is reasonable to evaluate patients with isolated DGP elevations, and to include DGP in testing algorithms. Nevertheless, given the low positive predictive value, patients should be informed that the majority of patients with isolated DGP elevations do not have CD, and a duodenal biopsy is necessary to identify the minority of patients in this setting who should be prescribed a gluten-free diet.

Compliance with ethical standards

Conflict of interest All authors declare that they have no conflict of interest and nothing to declare.

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