Anti-Gliadin Antibodies Identify Celiac Patients Overlooked by Tissue Transglutaminase Antibodies

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Abstract

For patients with suspected celiac disease, the American Gastroenterological Association recommends initial screening with anti-tissue transglutaminase antibody (tTG) and confirmation testing with small bowel biopsy. However, at Tripler Army Medical Center we routinely screen patients with both tTG and anti-gliadin antibodies (AGA) in combination. The purpose of this study was to evaluate whether this dual screening method adds to the evaluation of patients with suspected celiac disease or results in more false-positive results than tTG screening alone. A retrospective chart review of all tTG and AGA screening serologies at Tripler Army Medical Center between September 2008 and March 2012 was performed. For patients with positive serologic testing, small bowel biopsy results or reasoning for deferring biopsy were investigated. tTG was found to have a higher positive predictive value for celiac disease than AGA, however AGA identified 5 patients (19% of biopsy confirmed celiac disease) that had a negative tTG and would not have been identified by tTG screening alone. Using AGA in combination with tTG should be considered if the goal of screening is to identify all patients with celiac disease, with the understanding that this strategy will generate more false positive tests and result in additional patients undergoing small bowel biopsy.

Introduction

Celiac disease is a disorder of the small bowel characterized by mucosal inflammation, villous atrophy, and crypt hyperplasia which occurs upon exposure to dietary gluten and has an estimated prevalence in the United States of 1:100.[1-4] Celiac disease has a wide range of clinical presentations. Classically, patients with celiac disease present with gastrointestinal symptoms such as diarrhea, malabsorption, or weight loss. However, celiac disease may be diagnosed in many patients only after they are found to have a nutritional deficiency, such as iron deficiency, or another condition associated with celiac disease, such as delayed puberty, recurrent fetal loss, or premature osteoporosis.

For patients with suspected celiac disease, the American Gastroenterological Association recommends initial serologic testing with anti-tissue tranglutaminase antibody (tTG) and confirmed with a small bowel biopsy.[5] At our facility, we routinely screen with a combination of tTG and anti-gliadin antibody (AGA). The reported sensitivities of tTG, AGA IgA, and AGA IgG are 90 to 98%, 80 to 90%, and 75 to 85% respectively. The reported specificities of tTG, AGA IgA, and AGA IgG are 95 to 97%, 85 to 95%, and 75 to 90% respectively (Table 1).[6,7]

We aimed to test our hypothesis that using AGA and tTG in combination rather than tTG alone would result in more false positive tests while failing to increase identification of patients with celiac disease.

Methods

A retrospective chart review was performed of all celiac serologies at Tripler Army Medical Center between September 2008 and March 2012. No patients were excluded from the study. All celiac serologies were analyzed at the same laboratory and used the same cutoff values for negative, equivocal, and positive results. For the purpose of this study, all equivocal results were treated as positive. In patients with positive serologic testing, medical records were reviewed to determine small bowel biopsy results or the reason for deferring biopsy. A positive biopsy was defined by the presence of any of the following: increased intraepithelial lymphocytes, crypt hyperplasia, and/or villous atrophy; Marsh staging of biopsies was not defined in pathology reports and not included in analysis. Patients that screened positive but did not undergo a small bowel biopsy were categorized into three groups: those that were not referred to the gastroenterology service, those that were seen by the gastroenterology service and not biopsied, and those that were referred to the gastroenterology service but lost to follow-up. Positive predictive values for each of the serologic assays were calculated according to the formula: positive predictive value = true positives / total positive screening. True positives were defined as patients with small bowel histological evidence consistent with celiac disease as defined above. Each screening serology was used for calculation of positive predictive value in patients that underwent small bowel biopsy. Investigators adhered to the policies for protection of human subjects as prescribed in 45 Code of Federal Regulation 46.

Results

During the specified time period, 2,733 patients were evaluated with a total of 5,268 AGA and tTG antibody tests. 232 patients had at least one positive screening serology, including 34 tTG, 120 AGA IgA, and 119 AGA IgG. Of the 232 patients with positive screening serologies, 87 (38%) underwent a small bowl biopsy and 26 were found to have celiac disease (Figure 1). The positive predictive value of tTG was calculated to be 100%, AGA IgA was 36%, and AGA IgG was 16% (Table 2). 5 of 26 patients (20%) with biopsy-proven celiac disease during our specified period had a positive AGA IgA or IgG and a negative tTG. Of patients with at least one positive serologic screening antibody, 145 (62.5%) did not undergo a small bowel biopsy. Of the 145 patients with positive screening serologies who were not biopsied, 73 (50.3%) were never referred to the Tripler gastroenterology service, 49 (33.8%) were seen by the gastroenterology service but never biopsied, and 23 (15.9%) were referred to the gastroenterology service but lost to follow-up.

Discussion

The first celiac serology, AGA IgA, was developed in the early 1980s and revolutionized the diagnostic process of celiac dis-

Table 1. Reported sensitivities and specificities of celiac screening antibodies			
	tTG IgA	**AGA IgA**	**AGA IgG**
Reported Sensitivity	90-98%	80-90%	75-85%
Reported Specificity	95-97%	85-95%	75-90%

Table 2. Positive predictive value calculations for each screening serology				
Test	**Positive or Equivocal Serology**	**Number Biopsied**	**Positive Biopsy**	**Positive Predictive Value**
tTG IgA	34	21	21	100%
AGA IgA	120	40	18	45%
AGA IgG	119	45	7	16%

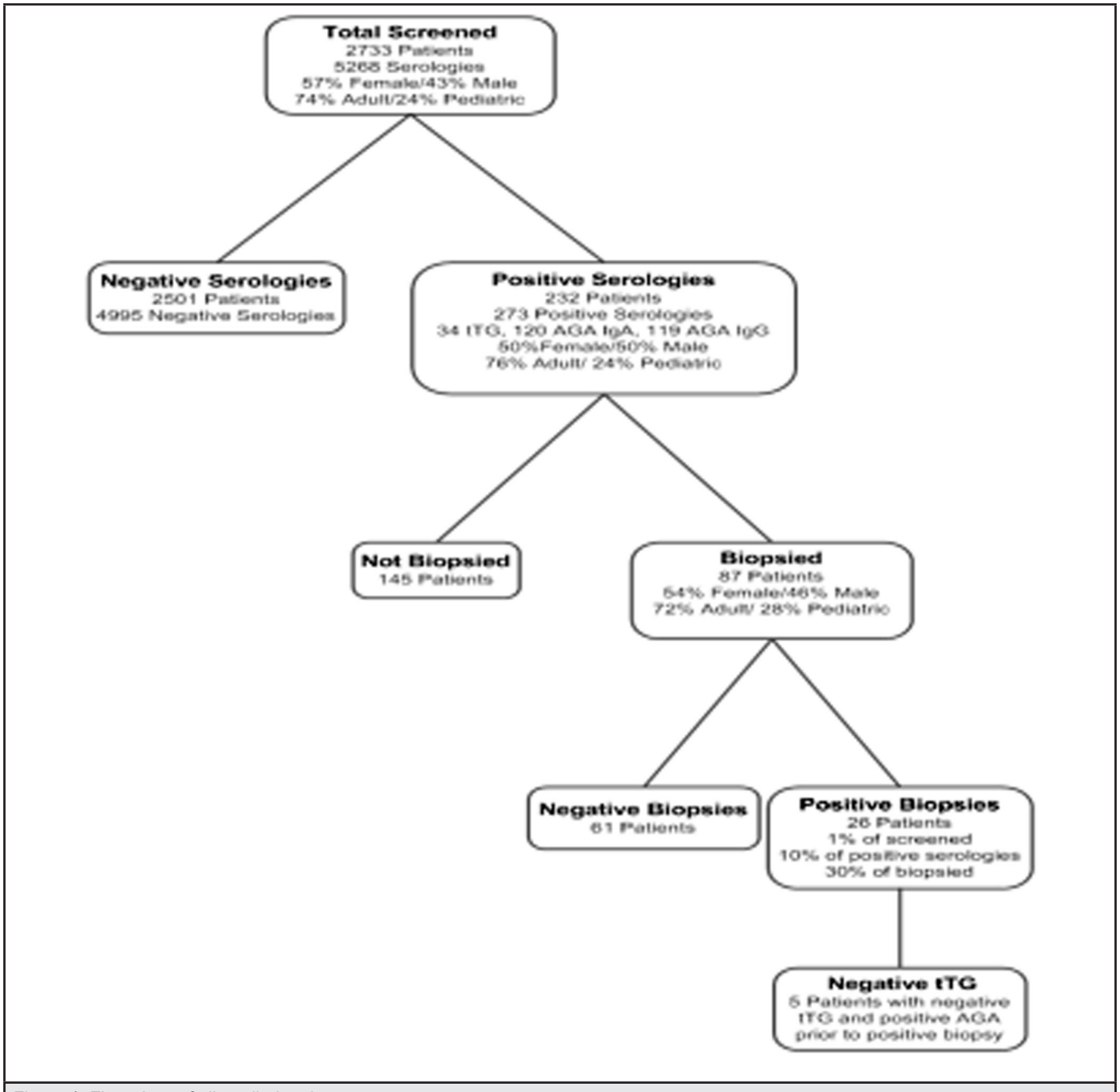

Figure 1. Flow chart of all studied patients

ease.[8] Prior to serologic studies, there was no screening test for celiac disease other than clinical suspicion, which was confirmed by small bowel biopsy. Shortly after the development of AGA, other serologic tests were introduced including tTG, antideaminated gliadin peptide antibodies, and antiendomysial antibodies.[9] Although we recognize the significance of antiendomysial and antideaminated gliadin peptide antibodies, they are not routinely performed at our institution and were not included in the study.

Screening for celiac disease is recommended by the Amercian Gastroenterologic Association only for symptomatic patients. Although the prevalence of celiac disease has been estimated to be 1% in the general population, there is insufficient evidence to recommend celiac screening in the general population.[10] Patients that are at high risk of celiac disease, such as those with first degree relatives of celiac disease, children or adolescents with short stature, patients with dermatitis herpetiformis, delayed puberty, type 1 diabetes mellitus, Down syndrome, persistent iron deficiency anemia, or osteoporosis should also be considered for serologic screening.[11]

There is a clear genetic predisposition for the development of celiac disease. Approximately 97% of patients with celiac disease share the major histocompatibility complex II class human leukocyte antigen DQ2 or DQ8 haplotype. Testing for these antigens may be considered in patients with equivocal small bowel histological findings.[12] Ordering of human leukocyte antigens is not routinely performed and the results of this testing were not evaluated in this study.

The most significant finding in our study was the identification of five patients with biopsy confirmed celiac disease that had negative tTG but positive AGAs. Had these patients been screened using the American Gastroenterology Association's recommendation for tTG alone, they would have tested negative and would not have been referred for small bowel biopsy. However, positive AGA IgA and IgG antibodies with either negative tTG or untested tTG led to 61 negative small bowel biopsies and, therefore, screening with AGA will increase the number of small bowel biopsies performed. It has been reported that AGAs have a higher clinical significance in the pediatric population. Several studies have identified pediatric patients with celiac disease who were found to have positive AGA and negative tTG or antiendomysial antibodies, suggesting AGA may still be appropriate when screening this population.[13,14] Of the five patients identified in our study, only one was under the age of 18 at the time of diagnosis. False negative tTG IgA testing has also been reported due to selective IgA deficiency. 1.7% of patients with celiac disease also has selective IgA deficiency and thus will have negative IgA screening antibodies.[15] Of the five patients our study identified, 3 of them had normal IgA levels and thus their false negative tTG could not be attributed to a selective IgA deficiency; the other two patients were not tested for IgA deficiency.

Patients that had positive serologic testing that did not undergo small bowel biopsy were investigated to better determine the reason for not undergoing small bowel biopsy. We found that only 87 of 232 (38%) patients with positive serologic testing went on to have biopsy. While this percentage seems quite low, similar rate of biopsy have been described at other institutions; one study reported that only 39% of patients that screened positive in serologic testing had a small bowel biopsy.[16] We further analyzed the 145 patients that did not undergo biopsy and determined that a little more than half (50.3%) were not referred to the gastroenterology service. This may represent primary care physicians treating empirically with a trial of gluten-free diet, patient refusal of referral, or failure to follow up on laboratory results. We also found about a third of patients (33.8%) that were seen by gastroenterology did not receive small bowel biopsy. This was due to a variety of reasons including patient refusal of biopsy, current treatment with gluten free diet (which may result in a false negative biopsy), or the sentiment among gastroenterologists that AGAs generate high number of false positive tests; the deferral of biopsy was often recommended in the pediatric population.

We acknowledge the limitations of our study. The retrospective nature of the study limits the information to that contained in patient medical records and does not reflect the number of patients placed on gluten-free diet without small bowel biopsy. Additionally, we are unable to determine whether serologic testing was performed on a gluten-free diet and thus impacted the results of these screening tests. It is also unclear how many primary care physicians and gastroenterologists discussed the benefits of small bowel biopsy but patients refused the procedure and opted for a trial of dietary modification.

Conclusions

While tTG has a significantly higher positive predictive value than AGA antibodies, AGA antibodies do increase the number of patients identified with celiac disease compared to tTG alone. Based on the results of this study, we reject our initial hypothesis. Screening with tTG in combination with AGA appeared beneficial if the goal of serologic testing is to maximize the number of patients discovered with celiac disease. Close follow-up of patients with positive celiac serologies is needed to ensure that they have the opportunity to undergo small bowl biopsy.

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