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HLA Genetics and Clinical Features of Self-Treated Patients on a Gluten-Free Diet

John A. Coburn, BS, Jennifer L. Vande Voort, MD, Brian D. Lahr, MS, Carol T. Van Dyke, SoCRA, Cynthia M. Kroning, MLT, ASCP, Tsung-Teh Wu, MD, PhD, Manish J. Gandhi, MD, and Joseph A. Murray, MD

Department of Psychiatry and Psychology (Dr Vande Voort), Division of Biostatistics and Informatics (Mr Lahr), Division of Gastroenterology and Hepatology (Ms Van Dyke and Dr Murray), Division of Transfusion Medicine (Ms Kroning and Dr Gandhi), Division of Anatomic Pathology (Dr Wu), and the Department of Immunology (Dr Murray), Mayo Clinic, Rochester, Minnesota. Mr Coburn is a student, Mayo Medical School, College of Medicine, Mayo Clinic, Rochester, Minnesota

Abstract

Background and Aims—Increasingly, persons start a gluten-free diet (GFD) without a clear celiac disease (CD) diagnosis. Human leukocyte antigen (HLA) genotyping is useful in ruling out CD in patients with equivocal results of serologic testing or small-bowel biopsy (SBB), but its utility and the clinical features of patients on self-treated GFD (ST-GFD) are largely unknown.

Methods—Retrospective study of single tertiary care center cohort compared 137 patients on ST-GFD and 443 patients with well-defined CD. We compared HLA genotype, symptoms, serologic and SBB results, and response to GFD between the 2 groups. Analysis used univariate logistic regression modeling, adjusted for age and sex.

Results—Patients with ST-GFD presented more often with diarrhea ($P<.001$), abdominal distention ($P<.001$), flatulence ($P=.002$), cramping ($P=.02$), itchy skin ($P=.02$), oral inflammation ($P=.04$), and constipation ($P=.01$) and less often with anemia ($P<.001$) or malaise ($P=.02$) than CD patients. In addition, 41% did not carry DQ2.5 and DQ8 vs 6% of CD patients ($P<.001$). Only 2% of ST-GFD patients had SBB clearly consistent with CD. Family history of CD showed no difference between groups ($P=.77$). Although CD patients had a statistically higher rate of GFD benefit, both groups had a high responsiveness rate (98% vs 94%; $P=.03$).

Conclusions—HLA genotyping is useful in evaluating patients on a ST-GFD. Although confirmed CD is rare in self-treated patients, most still report benefit from GFD regardless of DQ2 and DQ8 status. Nonceliac gluten sensitivity may play a role.

Keywords

celiac disease; diet; food intolerance; gluten-sensitive enteropathy

Introduction

The gluten-free diet (GFD) is the essential treatment of celiac disease (CD)¹. Gluten-free foods are growing in popularity^{2–4}, with a projected US market exceeding \$5 billion by

Correspondence to: Joseph A. Murray, MD, Division of Gastroenterology and Hepatology, Mayo Clinic, 200 First St SW, Rochester, MN 55905. Tel: 507-255-5713. Fax: 507-255-6318. murray.joseph@mayo.edu..

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2015⁵. Many people without CD try a GFD, including patients with chronic gastrointestinal symptoms. Patients may start a GFD because of a family history of CD, equivocal serologic testing, or small-bowel biopsy (SBB) or as an empirical trial. Patients trying a GFD who have persistent symptoms may wonder about the true nature of their diagnosis and seek medical evaluation only after starting a GFD.

Diagnosing CD in patients on a GFD is challenging because CD-related serologic tests and SBB results may be normal in CD patients adhering to GFD^{6–8}. Although CD affects approximately 1% to 1.5% of people in the United States and Europe⁹, many more are likely on a self-treated GFD (ST-GFD)^{10–11}. Even among patients with high genetic risk of CD on an ST-GFD, few have evidence of CD¹⁰.

Human leukocyte antigen (HLA) genotyping may be useful in ruling out CD when the diagnosis cannot be ruled out by other tests^{1,12,13}. HLA DQ2.5 and DQ8 antigens are present in 90% to 95% of CD patients, and CD patients who do not have these antigens are rare^{14,15}. The usefulness of HLA typing has been demonstrated in studies where SBB or serologic testing are equivocal^{12,13,16}. However, most of these studies had few patients on GFD before evaluation for CD; some lacked a comparison group of CD patients. We retrospectively compared the HLA genotype, presenting symptoms, serologic test results, SBB, and response to GFD of a large group of patients on an ST-GFD to patients with well-defined CD.

Methods

The present study was approved by the Mayo Clinic Institutional Review Board.

Retrospective Cohort Design

We abstracted the age, sex, presenting symptoms, class II HLA typing, CD-related serologic testing, SBB results, and response to GFD (subjective improvement of symptoms while on GFD) of patients on ST-GFD who underwent systematic evaluation. We then compared these features with those of patients with CD.

Study Population

Patients with an unclear diagnosis of CD were identified from the Medical Diagnostic Index, Celiac Disease Clinic, and Mayo Clinic Life Sciences System. Patients were recruited from July 1997 through November 2010¹⁶.

Inclusion and Exclusion Criteria

Patients were included who were on a GFD without a physician recommendation before having testing for CD. Patients were also included if they started a GFD by recommendation of a physician or an alternative medicine practitioner and did not have clear evidence of CD. *Clear evidence* was defined as having at least one of the following: test result positive for anti-immunoglobulin (Ig) A, endomysial antibody (EMA), or anti-tissue transglutaminase (TTG) antibodies or SBB results clearly consistent with CD¹⁶ with no other explanation for enteropathy.

Patients with clear evidence of CD before initiation of a GFD were excluded from our ST-GFD group. Presence of antigliadin antibodies alone was not considered clear evidence of CD^{17,18}. Patients placed on a GFD because of testing positive for antigliadin antibodies alone were included in our ST-GFD group. The comparison group consisted of patients seen at Mayo Clinic in Rochester, Minnesota, with biopsy-proven CD¹⁶. HLA genotyping was not used as part of the diagnostic criteria.

HLA Genotyping, Serology, and Small-Bowel Biopsy

Class II HLA typing, serologic testing, and SBB were performed as previously described¹⁶. With few exceptions, gliadin antibody testing was performed by enzyme-linked immunosorbent assay using both IgA and IgG isotype directed against native gliadin (Scimedix NJ). In accordance with current World Health Organization nomenclature, *DQ2.5* represents HLA-DQA1*05:xx,DQB1*02:01; *DQ2.2*, HLA-DQA1*02:01,DQB1*02:02; and *DQ8*, HLA-DQA1*03:xx,DQB1*03:02 antigens. Negative serologic test results were considered valid if the serologic testing was performed while the patient was on a gluten-containing diet; positive results of serologic testing were considered valid regardless of the patient's dietary status. Similarly, SBB results were invalid if the patient was on a GFD for longer than 2 months when undergoing the biopsy. In most patients, outside specimens were reviewed by an in-house gastrointestinal pathologist.

Statistical Analysis

Descriptive statistics were used to summarize the cohort, which consisted of patients either on a ST-GFD or with well-defined CD. Demographic characteristics, clinical characteristics, and HLA genotype profiles were tested for an association with the 2 groups through logistic regression. Since these groups were not randomized or matched, age and sex were considered potential confounders and were included as adjusting covariates in the logistic models. A (2-sided) level of .05 was used to assess statistical significance.

Results

Overall, 137 patients on ST-GFD were compared with 443 patients who had well-defined CD. Both groups were predominately female (69% vs 71%), with a comparable mean (SD) age of 42.4 (16.5) years and 44.8 (18.6) years. Sixty ST-GFD patients (44%) had no diagnostic testing for CD before starting GFD; 66 (48%) had some documented previous diagnostic testing but did not have clear evidence of CD. In 12 self-treated patients (9%), it was unclear whether CD testing had been performed before starting the GFD.

Small-Bowel Biopsy

By definition, all patients with well-defined CD had SBB results consistent with CD¹⁶. Among 137 ST-GFD patients, 109 (80%) underwent SBB. Of these 109 patients, biopsies were obtained in 56 patients on a gluten-containing diet; the other patients underwent biopsy on a GFD. Of these 56 self-treated patients with valid SBB, 1 patient had villous atrophy with intraepithelial lymphocytosis, suggestive of CD, with a positive response to GFD. In contrast, 44 (79%) had a normal SBB result, whereas 11 (20%) had intraepithelial lymphocytosis with preserved villous architecture¹⁹. All biopsies performed in patients who had been on an ST-GFD longer than 2 months (n=53) showed normal results.

Serologic Testing

Overall, 128 (93%) of 137 ST-GFD patients had some type of serologic testing. Because participants on an ST-GFD were often on GFD at our evaluation, only a subgroup (n=56) had serologic testing done while on a gluten-containing diet; the other patients had serologic testing performed while on a GFD. Among patients with valid serologic testing on a gluten-containing diet, 9 (20%) of 45 tested positive to anti-TTG and 1 (3%) of 31 tested positive to anti-EMA IgA. On the basis of a composite serologic result of either positive TTG or positive EMA, 10 (18%) of 56 ST-GFD participants were seropositive on a gluten-containing diet. Only 2 (3%) of 64 patients on GFD at the time of testing had a TTG-positive result, while none (0%) of 47 had an EMA-positive result. Not surprisingly, the rate

of positivity for antigliadin antibody was high in self-treated participants (37/49; 76%) (Table 1).

Clinical Features

Gastrointestinal tract symptoms tended to be more common in patients on an ST-GFD than in CD patients. After control for age and sex, ST-GFD patients more frequently had diarrhea (73% vs 55%; $P<.001$), abdominal distention (47% vs 28%; $P<.001$), flatulence (28% vs 16%; $P=.002$), abdominal cramping (49% vs 37%; $P=.02$), itchy skin (16% vs 9%; $P=.02$), inflamed mouth (9% vs 4%; $P=.04$), and constipation (28% vs 18%; $P=.01$). The ST-GFD patients were less likely than CD patients to have anemia (12% vs 32%; $P<.001$) or malaise (6% vs 14%; $P=.02$) (Table 2).

HLA Genotype

Fewer ST-GFD patients carried DQ2.5 than patients with well-defined CD (37% vs 87%; $P<.001$), adjusted for age and sex. The ST-GFD patients carried DQ8 more frequently than CD patients (24% vs 15%; $P=.016$) but were also more likely to carry DQ8 in absence of DQ2.5 (22% vs 7%; $P<.001$) and to not have both DQ2.5 and DQ8 (41% vs 6%; $P<.001$). The carrier rate of DQ2.2 did not differ significantly between the groups (17% vs 23%; $P=.16$) (Table 3).

Family History of CD and Response to GFD

There was no significant association between group and having a family history of CD (23% ST-GFD vs 22% CD; $P=.77$), adjusted for age and sex. ST-GFD participants were statistically less likely than CD patients to respond to a GFD (94% vs 98%; $P=.03$), although both groups had high responsiveness rates.

Self-Treated Patients With CD-Associated vs Non-CD-Associated Antigens

A subgroup analysis compared ST-GFD patients who had CD-associated HLA type (DQ2.5 or DQ8, or both) with ST-GFD patients who did not have DQ2.5 and DQ8, controlling for age and sex. The ST-GFD patients with CD-associated antigens had a CD family history more often than those without these antigens (31% vs 13%; $P=.01$). However, no other significant differences were found between these groups. Notably, both groups responded favorably to GFD (94% vs 96% in those with and without DQ2.5 and DQ8, respectively; $P=.34$).

Discussion

HLA genotyping is useful when a CD diagnosis is unclear due to equivocal serologic testing or SBB^{1,12,13,16,20,21}. One study addressing HLA genotyping in ST-GFD patients found that 11 (65%) of 17 patients adhering to a GFD before SBB did not have DQ2 and DQ8 antigens¹².

We found that 41% of ST-GFD patients, but only 6% of CD patients, did not have DQ2.5 and DQ8. Therefore, in a substantial proportion (41%) of ST-GFD patients, CD can be essentially ruled out with HLA genotyping, without need for gluten challenge or SBB. Although a large proportion of ST-GFD patients did not have DQ2.5 and DQ8, only marginal differences existed between the groups in their reported response to GFD (98% CD patients and 94% ST-GFD patients responded positively ($P=.03$). So although HLA typing is useful in ruling out CD, it does not exclude the possibility of benefits from a GFD. These findings in ST-GFD patients support the existence of nonceliac gluten sensitivity (NCGS), a condition for which evidence is growing^{22–33}. Two broad categories of gluten intolerance are identifiable in the medical literature: celiac-lite and NCGS.

Patients referred to as *celiac-lite* carry genetic susceptibility for CD and respond to GFD. These include patients with CD-associated HLA genotypes with gluten-responsive chronic diarrhea or diarrhea-predominant irritable bowel syndrome (IBS)^{23,24}. Celiac-lite gluten sensitivity may share pathophysiologic mechanisms with CD on the basis of similar HLA genotype and other features. Celiac-lite patients carry CD-associated antigens²²⁻²⁴ and may have minor enteropathy^{26,34,35}, increased CD-associated antibodies in duodenal aspirate²⁷, increased gluten-specific T-cells^{10,36}, presence of TTG 2-specific IgA deposits in small bowel mucosa³⁷, or positive CD-related serologic findings with normal SBB results²⁸. However, they do not clearly meet criteria for CD because they lack small bowel villous flattening, and may lack specific serologic evidence for CD^{22,28}. Evidence exists for putative mechanisms of symptom generation in these patients²². In contrast, evidence for mechanisms of NCGS is lacking^{29,30,33}.

Patients with NCGS do not have CD-associated antigens, yet they have substantial symptoms on gluten ingestion and improvement after adopting a GFD. In a double-blind, randomized controlled trial, patients whose IBS was symptomatically controlled on GFD were given either gluten or placebo with a GFD for up to 6 weeks³⁰. Thirteen (68%) of 19 patients in the gluten group reported symptoms not adequately controlled compared with 6 (40%) of 15 patients in the placebo group. Of note, "symptomatic responses to gluten did not significantly differ in those expressing HLA-DQ2 and/or HLA-DQ8... compared with those who did not"³⁰. Therefore, symptoms with gluten ingestion could not simply be attributed to celiac-spectrum gluten sensitivity (celiac-lite, potential or latent CD). However, no evidence for mechanisms was identified.

Our study supports the existence of NCGS. The ST-GFD patients differed substantially in DQ2.5 and DQ8 status from patients with CD (59% ST-GFD patients vs 94% CD patients carried DQ2.5 and DQ8); yet, self-treated patients were only marginally less likely to benefit from GFD (94% ST-GFD patients vs 98% CD patients). In subgroup analysis, ST-GFD patients carrying DQ2.5 or DQ8 did not benefit more often from GFD than self-treated patients who did not have DQ2.5 and DQ8 antigens (92% vs 96%; $P=.34$). These results suggest that mechanisms of NCGS are different than CD, with DQ2.5 and DQ8 antigens not required for NCGS. Yet, the possibility remains that symptom improvement in self-treated patients was a "nonspecific gastrointestinal response to ... the low-residue nature of a gluten-free diet"³¹. Self-treated patients tended to be more symptomatic than CD patients which may reflect their reason for seeking treatment and care in the first place. The apparently high responsiveness rate to gluten exclusion in self-treated patients could be due to an anticipated or Nocebo effect of gluten and may not be specific to the absence of gluten in the diet. In the present study, we did not measure the sustainability of patients' responses to GFD. Therefore, it is possible that these benefits were temporary. Further studies are needed to measure the sustainability of responses to a GFD in patients with apparent NCGS.

The fact that both CD patients and NCGS patients respond to gluten exclusion begs the question of why it is important to distinguish these entities. Several reasons exist. CD has known complications, including iron deficiency anemia³⁸, osteoporosis³⁹, malignancy⁴⁰, and others^{41,42}. Our data and the absence of actual damage in the intestine suggest that NCGS patients are not at increased risk for these complications. CD is associated with particular genotypes, and family members carry higher risk of having the disease; thus, there are implications for screening family members⁴³. It is unknown whether NCGS shows considerable heritability. In CD, strict adherence to a GFD is necessary to avoid complications¹. Whether a strict GFD is needed in NCGS is unknown. Currently, CD requires SBB for diagnosis¹; patients with NCGS may not need SBB, especially when they lack celiac-susceptibility HLA genotypes. Proper exclusion of CD and other diagnoses is

necessary before patients are placed on a GFD for indication of NCGS, because hasty placement may delay treatment of potential alternative diagnoses³².

We found no difference among the ST-GFD and CD patients in the carriage of DQ2.2, a version of DQ2 not usually associated with CD⁴⁴. The ST-GFD patients presented with functional bowel disorder-type symptoms more often than the CD patients. Functional bowel disorders, such as IBS, may be more common in ST-GFD patients. The ST-GFD patients presented more often with inflamed mouth or itchy skin than CD patients. These findings may reflect increasing awareness that CD is associated with dermatitis herpetiformis. The CD patients did not have a family history of CD more often than ST-GFD patients. Therefore, patients with family history of CD who have chronic gastrointestinal or other symptoms seem more likely to try a GFD themselves.

Our study suggests that SBB and serologic testing in ST-GFD patients have fairly low diagnostic yield, especially when performed on patients who have had a GFD longer than 2 months at the time of testing. Unfortunately, many patients declined a formal gluten challenge; HLA typing may prove helpful in these situations.

Limitations to our study include its retrospective design, making subjectivity in interpreting patient records possible and follow-up variable. We attempted to control for subjectivity by strictly defining the characteristics measured. Retrospective studies carry risk of confounding factors, which are difficult to control for and could account for some observed effects. It is possible that there may have been subtle changes in the intestinal mucosa detectable by such techniques as immunofluorescence for intraepithelial lymphocytes or IgA deposits in the mucosa or perilesional skin. However, appropriate samples were not available. The history of itchy skin disorders was based on history and rarely did patients have active skin lesions deemed suitable for skin biopsy at evaluation. This could be done in future studies. Another limitation is that this was a tertiary care center study, which may limit applicability of results.

Many questions remain, including who has NCGS; what are the risk factors, natural history, and heritability; and how can NCGS patients be identified. Furthermore, some studies have shown different mucosal responses to dietary gluten in CD and gluten sensitivity³³; more studies are needed to determine NCGS mechanisms.

Conclusions

Functional bowel and mucocutaneous symptoms and a family history of CD are as common in ST-GFD patients as in CD patients. HLA testing is useful to rule out CD in a substantial proportion of patients on an ST-GFD, particularly when valid SBB is not easily obtainable or a patient is unwilling to undergo formal gluten challenge. The diagnostic yield of SBB and serologic testing is low in self-treated patients on a GFD at the time of testing, and these tests should be avoided in ST-GFD patients without a formal gluten challenge because the tests are often uninformative without it. Finally, clinicians may not want to argue with success after alternative diagnoses have been ruled out. Most patients with self-identified NCGS seem to benefit from a GFD at a rate not much less than patients with CD.

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References

1. AGA Institute. AGA Institute Medical Position Statement on the Diagnosis and Management of Celiac Disease. *Gastroenterology*. 2006; 131:1977–1980. [PubMed: 17087935]
2. Allen, JE. Gluten-free: the low-carb of this decade? [Internet]. ABC News. May 3. 2011 [cited 2011 Aug 24]. Available from: <http://abcnews.go.com/Health/Diet/gluten-free-diet/storSty?id=13482695>
3. Storrs, C. Will a gluten-free diet improve your health? [Internet]. *Health.com*. Health.com[updated 2011 Apr 5; cited 2011 Aug 25]. Available from: http://www.health.com/health/article/0,,20479423_2,00.html
4. Voiland, A. Gluten-free diet: a cure for some, a fad for most. *US News and World Report* [Internet]. Oct 31. 2008 [cited 2011 Aug 24]. Available from: http://health.usnews.com/health-news/family-health/digestive-disorders/articles/2008/10/31/gluten-free-diet-a-cure-for-some-a-fad-for-most_print.html
5. Gluten-free foods and beverages in the U.S. 3rd edition Feb 1. 2011 InternetPackaged Facts; c2012 [cited 2011 Aug 25]. Available from: <http://www.packagedfacts.com/Gluten-Free Foods-2710664>
6. Sugai E, Nachman F, Vaquez H, et al. Dynamics of celiac disease-specific serology after initiation of a gluten-free diet and use in the assessment of compliance with treatment. *Dig Liver Dis*. 2010; 42:352–358. Epub 2009 Aug 12. [PubMed: 19679520]
7. Hutchinson JM, West NP, Robins GG, et al. Long-term histological follow-up of people with coeliac disease in a UK teaching hospital. *QJM*. 2010; 103:511–517. Epub 2010 Jun 2. [PubMed: 20519276]
8. Tursi A, Brandimarte G, Giorgetti GM, et al. Endoscopic and histological findings in the duodenum of adults with celiac disease before and after changing to a gluten-free diet: a 2-year prospective study. *Endoscopy*. 2006; 38:702–707. [PubMed: 16810593]
9. Walker MM, Murray JA, Ronkainen J, et al. Detection of celiac disease and lymphocytic enteropathy by parallel serology and histopathology in a population-based study. *Gastroenterology*. 2010; 139:11211–9. Epub 2010 Apr 13.
10. Brottveit M, Raki M, Bergseng E, et al. Assessing possible celiac disease by an HLA-DQ2-gliadin Tetramer Test. *Am J Gastroenterol*. 2011; 106:1318–1324. Epub 2011 Mar 1. Erratum in: *Am J Gastroenterol* 2012;107:638. [PubMed: 21364548]
11. Kaukinen K, Turjanmaa K, Maki M, et al. Intolerance to cereals is not specific for coeliac disease. *Scand JGastroenterol*. 2000; 35:942–946. [PubMed: 11063153]
12. Kaukinen K, Partanen J, Maki M, et al. HLA-DQ typing in the diagnosis of celiac disease. *Am J Gastroenterol*. 2002; 97:695–699. [PubMed: 11922565]
13. Kapitany A, Toth L, Tumpek J, et al. Diagnostic significance of HLA-DQ typing in patients with previous coeliac disease diagnosis based on histology alone. *Aliment Pharmacol Ther*. 2006; 24:1395–1402. [PubMed: 17059521]
14. Karell K, Louka AS, Moodie SJ, et al. HLA types in celiac disease patients not carrying the DQA1*05-DQB1*02 (DQ2) heterodimer: results from the European Genetics Cluster on Celiac Disease. *Hum Immunol*. 2003; 64:469–477. [PubMed: 12651074]
15. Margaritte-Jeannin P, Babron MC, Bourgey M, et al. HLA-DQ relative risks for coeliac disease in European populations: a study of the European Genetics Cluster on Coeliac Disease. *Tissue Antigens*. 2004; 63:562–567. [PubMed: 15140032]
16. Vande Voort JL, Murray JA, Lahr BD, et al. Lymphocytic duodenitis and the spectrum of celiac disease. *Am J Gastroenterol*. 2009; 104:142–148. [PubMed: 19098862]
17. Rostom A, Dube C, Cranney A, et al. The diagnostic accuracy of serologic tests for celiac disease: a systematic review. *Gastroenterology*. 2005; 128:S38–S46. [PubMed: 15825125]
18. van der Windt DA, Jellema P, Mulder CJ, et al. Diagnostic testing for celiac disease among patients with abdominal symptoms: a systematic review. *JAMA*. 2010; 303:1738–1746. [PubMed: 20442390]
19. Kakar S, Nehra V, Murray JA, et al. Significance of intraepithelial lymphocytosis in small bowel biopsy samples with normal mucosal architecture. *Am J Gastroenterol*. 2003; 98:2027–2033. [PubMed: 14499783]

20. Hadithi M, von Blomberg BM, Crusius JB, et al. Accuracy of serologic tests and HLA-DQ typing for diagnosing celiac disease. *Ann Intern Med.* 2007; 147:294–302. [PubMed: 17785484]
21. Rashtak S, Murray JA. Tailored testing for celiac disease. *Ann Intern Med.* 2007; 147:339–341. [PubMed: 17785489]
22. Verdu EF, Armstrong D, Murray JA. Between celiac disease and irritable bowel syndrome: the “no man's land” of gluten sensitivity. *Am J Gastroenterol.* 2009; 104:1587–1594. [PubMed: 19455131]
23. Wahnschaffe U, Schulzke JD, Zeitz M, et al. Predictors of clinical response to gluten-free diet in patients diagnosed with diarrhea-predominant irritable bowel syndrome. *Clin Gastroenterol Hepatol.* 2007; 5:844–850. Epub 2007 Jun 5. [PubMed: 17553753]
24. Wahnschaffe U, Stockmann M, Daum S, et al. Intestinal antibodies against gliadin, tissue-transglutaminase, beta-lactoglobulin, and ovalbumin in patients with irritable bowel syndrome. *Ann N Y Acad Sci.* 1998; 859:280–284. [PubMed: 9928405]
25. Cooper BT, Holmes GK, Ferguson R, et al. Gluten-sensitive diarrhea without evidence of celiac disease. *Gastroenterology.* 1980; 79:801–806. [PubMed: 7419003]
26. Sbarbati A, Valletta E, Bertini M, et al. Gluten sensitivity and ‘normal’ histology: is the intestinal mucosa really normal? *Dig Liver Dis.* 2003; 35:768–773. [PubMed: 14674666]
27. Wahnschaffe U, Ullrich R, Riecken EO, et al. Celiac disease-like abnormalities in a subgroup of patients with irritable bowel syndrome. *Gastroenterology.* 2001; 121:1329–1338. [PubMed: 11729112]
28. Troncone R, Jabri B. Coeliac disease and gluten sensitivity. *J Intern Med.* 2011; 269:582–590. [PubMed: 21481018]
29. Newnham ED. Does gluten cause gastrointestinal symptoms in subjects without coeliac disease? *J Gastroenterol Hepatol.* 2011; 26(Suppl 3):132–4. [PubMed: 21443726]
30. Biesiekierski JR, Newnham ED, Irving PM, et al. Gluten causes gastrointestinal symptoms in subjects without celiac disease: a double-blind randomized placebo-controlled trial. *Am J Gastroenterol.* 2011; 106:508–514. Epub 2011 Jan 11. [PubMed: 21224837]
31. Fan X, Sellin JH. Small intestinal bacterial overgrowth, bile acid malabsorption and gluten intolerance as possible causes of chronic watery diarrhoea. *Aliment Pharmacol Ther.* 2009; 29:1069–1077. Epub 2009 Feb 15. Erratum in: *Aliment Pharmacol Ther* 2009;29:1217. [PubMed: 19222407]
32. Campanella J, Biagi F, Bianchi PI, et al. Clinical response to gluten withdrawal is not an indicator of coeliac disease. *Scand J Gastroenterol.* 2008; 43:1311–1314. [PubMed: 18609173]
33. Sapone A, Lammers KM, Casolaro V, et al. Divergence of gut permeability and mucosal immune gene expression in two gluten-associated conditions: celiac disease and gluten sensitivity. *BMC Med.* 2011; 9:23. [PubMed: 21392369]
34. Kaukinen K, Maki M, Partanen J, et al. Celiac disease without villous atrophy: revision of criteria called for. *Dig Dis Sci.* 2001; 46:879–887. [PubMed: 11330428]
35. Mino M, Lauwers GY. Role of lymphocytic immunophenotyping in the diagnosis of gluten-sensitive enteropathy with preserved villous architecture. *Am J Surg Pathol.* 2003; 27:1237–1242. [PubMed: 12960808]
36. Maki M, Holm K, Collin P, et al. Increase in gamma/delta T cell receptor bearing lymphocytes in normal small bowel mucosa in latent coeliac disease. *Gut.* 1991; 32:1412–1414. [PubMed: 1836446]
37. Kaukinen K, Peraaho M, Collin P, et al. Small-bowel mucosal transglutaminase 2-specific IgA deposits in coeliac disease without villous atrophy: a prospective and randomized clinical study. *Scand J Gastroenterol.* 2005; 40:564–572. [PubMed: 16036509]
38. Zamani F, Mohamadnejad M, Shakeri R, et al. Gluten sensitive enteropathy in patients with iron deficiency anemia of unknown origin. *World J Gastroenterol.* 2008; 14:7381–7385. [PubMed: 19109873]
39. Bianchi ML, Bardella MT. Bone in celiac disease. *Osteoporos Int.* 2008; 19:1705–1716. Epub 2008 Apr 17. [PubMed: 18418638]
40. Rampertab SD, Forde KA, Green PH. Small bowel neoplasia in coeliac disease. *Gut.* 2003; 52:1211–1214. [PubMed: 12865284]

41. Botero-Lopez JE, Araya M, Parada A, et al. Micronutrient deficiencies in patients with typical and atypical celiac disease. *J Pediatr Gastroenterol Nutr.* 2011; 53:265–270. [PubMed: 21865972]
42. Freeman HJ. Neurological disorders in adult celiac disease. *Can J Gastroenterol.* 2008; 22:909–911. [PubMed: 19018335]
43. Goldberg D, Kryszak D, Fasano A, et al. Screening for celiac disease in family members: is follow-up testing necessary? *Dig Dis Sci.* 2007; 52:1082–1086. Epub 2007 Feb 16. [PubMed: 17380406]
44. van de Wal Y, Kooy YM, Drijfhout JW, et al. Unique peptide binding characteristics of the disease-associated DQ(alpha 1*0501, beta 1*0201) vs the non-disease-associated DQ(alpha 1*0201, beta 1*0202) molecule. *Immunogenetics.* 1997; 46:484–492. [PubMed: 9321428]

Table 1

Serologic Test Results of Patients on an ST-GFD and With CD

Variable	Observations, No.	Patients With Well-Defined CD, No. (%)	Observations, No.	Patients on ST-GFD, No. (%)
EMA positive at diagnosis	236	206 (87)	31	1 (3)
TTG positive at diagnosis	231	206 (89)	44	9 (20)
Gliadin positive at diagnosis	163	145 (89)	49	37 (76)
Positive serologic results	308	279 (91)	56	10 (18)

Abbreviations: CD, celiac disease; EMA, endomysial antibody; ST-GFD, self-treated gluten-free diet; TTG, tissue transglutaminase.

Table 2

Clinical Features of Patients on ST-GFD vs With CD

Variable	Patients With Well-Defined CD, No. (%) (n=443)	Patients on ST-GFD, No. (%) (n=137)	Age- and Sex-Adjusted Logistic Results, <i>P</i> Value
Diarrhea	243 (55)	100 (73)	<.001 ^a
Greasy stool	52 (12)	15 (11)	.86
Abdominal distention	125 (28)	65 (47)	<.001 ^a
Abdominal cramping	162 (37)	67 (49)	.02 ^b
Flatulence	69 (16)	38 (28)	.002 ^a
Edema	17 (4)	3 (2)	.41
Anorexia	12 (3)	3 (2)	.70
Weight loss	170 (38)	52 (38)	.92
Nausea/vomiting	97 (22)	34 (25)	.53
Paresthesia	15 (3)	5 (4)	.90
Bone pain	26 (6)	2 (1)	.06
Anemia	143 (32)	17 (12)	<.001 ^a
Malaise	60 (14)	8 (6)	.02 ^b
Skin disorder	58 (13)	27 (20)	.05
Inflamed mouth	16 (4)	12 (9)	.04 ^b
Itchy skin	40 (9)	22 (16)	.02 ^b
Headaches	64 (14)	22 (16)	.74
Family history of CD	97 (22)	32 (23)	.77
Failure to thrive	11 (2)	2 (1)	.34
Constipation	78 (18)	38 (28)	.01 ^b

Abbreviations: CD, celiac disease; ST-GFD, self-treated gluten-free diet.

^a*P* value <.01.^b*P* value <.05

Table 3

HLA DQ Type in Patients on an ST-GFD vs Patients With CD

Variable	Patients With Well-Defined CD, No. (%) (n=443)	Patients on ST-GFD, No. (%) (n=137)	Age- and Sex-Adjusted Logistic Results, <i>P</i> Value
DQ2.5 alleles, No.			<.001 ^a
0	59 (13)	86 (63)	
1	302 (68)	46 (34)	
2	82 (19)	5 (4)	
Any ^b DQ2.5 alleles	384 (87)	51 (37)	<.001 ^a
DQ2.2 alleles, No.			.22
0	341 (77)	113 (82)	
1	101 (23)	23 (17)	
2	1 (0)	1 (1)	
Any ^b DQ2.2 alleles	102 (23)	24 (18)	.16
DQ8 alleles, No.			.09
0	378 (85)	104 (76)	
1	55 (12)	32 (23)	
2	10 (2)	1 (1)	
Any ^b DQ8 alleles	65 (15)	33 (24)	.02 ^c
Either DQ2.5 or DQ8	416 (94)	81 (59)	<.001 ^a
DQ8 in absence of DQ2.5	32 (7)	30 (22)	<.001 ^a

Abbreviations: CD, celiac disease; DQ2.2, HLA-DQA1*02:01,DQB1*02:02 antigen; DQ2.5, HLA-DQA1*05:xx,DQB1*02:01 antigen; DQ8, HLA-DQA1*03:xx,DQB1*03:02 antigen; HLA, human leukocyte antigen; ST-GFD, self-treated gluten-free diet.

^a*P*value <.01.

^bProportion of patients that carried at least 1 copy of the specified allele.

^c*P*value <.05.