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Prior Misdiagnosis of Celiac Disease Is Common Among Patients Referred to a Tertiary Care Center: A Prospective Cohort Study

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OBJECTIVES: Interest of patients and physicians in celiac disease is growing worldwide, but without a corresponding increase in the awareness of the disease. Many patients are diagnosed as celiacs even without completing the whole diagnostic process, with consequent risk of misdiagnosis and delay in the evaluation of other diseases. The objective of this study was to assess the rates of prior celiac disease misdiagnosis among patients referred to a tertiary care center.

METHODS: From June 2013 to December 2014, we prospectively recruited patients referred for the first time to our Celiac Disease Center. Patients with a previous diagnosis of celiac disease underwent a diagnostic revaluation by second reading of duodenal tissue slides, dosage of specific antibodies, and/or duodenal biopsy sampling; HLA status was investigated in pertinent cases.

RESULTS: A total of 198 subjects were recruited. Of these, 91 "naïve" patients (46%) started the diagnostic screening for celiac disease; 58 of them (64–29% of the whole sample) were diagnosed as celiacs. The remaining 107 patients (54%) came with a previous diagnosis of celiac disease: of these, 52 (49–26% of the whole sample) presented with confirmed diagnosis of celiac disease, whereas 55 (51–28% of the whole sample) underwent diagnostic revaluation. After the reassessment, diagnosis was rejected in 43 cases (78–22% of the whole sample) and confirmed in the remaining 12 (22–6% of the whole sample). Overall, diagnosis was confirmed in only 64 of the 107 subjects with a previous diagnosis (60–32% of the whole sample). Diagnosis of celiac disease was more frequently confirmed in "naïve" patients compared those with a questionable previous diagnosis (64% vs. 22%; $P < 0.0001$).

CONCLUSIONS: A considerable number of patients referred to a tertiary care center are inaccurately diagnosed with celiac disease. Although we cannot exclude that uncertain diagnosis was a reason for the referral, we suggest greater adherence to guidelines to minimize the burden of celiac disease misdiagnosis.

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INTRODUCTION

Celiac disease (CD) is a chronic autoimmune disorder of the small bowel that develops in genetically predisposed subjects. CD is induced by the ingestion of gluten and triggered by environmental factors.[1] According to the Current Guidelines by the American College of Gastroenterology[2] and by the British Society of Gastroenterology[3] for the diagnosis and management of CD in adult patients, CD is usually suspected by the positivity of specific antibody testings (endomysium antibodies (EMAs), transglutaminase antibodies (tTGs), deamidated gliadin antibodies), and the diagnosis is confirmed by the presence of typical histological features of the duodenal mucosa. The whole diagnostic process should be carried on a gluten-containing diet.

Formerly CD was considered a rare condition, with overall prevalence of 0.03% around 1970s.[4] The prevalence of CD has often been compared with an iceberg,[5] whose visible surface is composed of symptomatic subjects, representing a small part of the total. Actually, CD is considered a underdiagnosed disease.[6]

CD was gradually recognized more often over time, and currently the estimated mean overall prevalence in western countries is 1%.[7] In the recent past, interest in CD has gradually increased, not only among clinicians interested in digestive diseases but also among other specialists—such as dermatologists and gynecologists—as well as general practitioners, probably because CD is a systemic disease, and may involve multiple organs.

Moreover, the excitement of media—especially Internet—towards the harmful properties of gluten and towards gluten-related disorders (which includes, beyond CD, wheat allergy and non-celiac gluten sensitivity)[8] is growing worldwide. Therefore, a huge number of individuals exclude gluten from their diet even in the absence of CD, and gluten-free diet (GFD) is advocated by several Web forums, patients, and clinicians as part of a healthy lifestyle, even without any reliable medical reasons.[9] One of the consequences of such a

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trend is that several subjects receive or self-report a diagnosis of CD, consequently starting a GFD, without completing the proper diagnostic process recommended by current guidelines, and often without any prior medical consultation. Such a behavior might lead to a considerable waste of resources and to a significant diagnostic delay both in celiacs and in subjects without CD.

The Policlinico "Agostino Gemelli" is an academic tertiary care center, and it is a referral center for gastroenterology and especially for CD. It is located in Rome, a metropolis with 3 million people. At this hospital, CD is diagnosed, cared for and followed up, and we accomplish also the certification of the disease (that, in Italy, is needed to receive gluten-free food vouchers and health-care tax exemption, and can be performed only by few empowered centers), as well as the evaluation of subjects with suspected CD and the management of challenging situations, including the reassessment of previous unclear diagnoses.

During our clinical practice, we had the feeling that the number of previous diagnoses of CD, which we rejected after re-evaluation was considerable. Misdiagnosis of CD represents an incoming but burdensome issue, as already assessed by retrospective studies performed in tertiary referral centers.[10] To date, however, prospective data on CD misdiagnosis are still not available.

Our aim was therefore to assess prospectively the rates of prior CD misdiagnosis among patients referred to a tertiary care center.

METHODS

Study design. We designed a prospective study aimed to investigate the rate of prior CD misdiagnosis among patients referred to our tertiary care center for the diagnosis and management of CD. Because of the intrinsic aim of the study, only patients referred to us for the first time were considered for inclusion, whereas subjects returning at our observation for follow-up visits were excluded. Only patients older than 18 years were enrolled. After obtaining their informed consent, enrolled subjects were subdivided into two categories: (a) "naïve" patients, who came without a prior diagnosis of CD; (b) patients with a previous diagnosis of CD. The latter group of patients was critically re-evaluated, to distinguish subjects with a proper diagnosis from those with a questionable diagnosis, and to assess the rates of final CD misdiagnosis among them.

The number of patients diagnosed with CD was assessed in both study groups. Furthermore, we gathered together all patients referred to us without a proper diagnosis of CD, which were, respectively, naïve patients without a prior diagnosis of CD and patients with a questionable previous diagnosis of CD, and compared the rates of final CD diagnosis between these two subgroups.

Criteria for defining proper diagnosis/questionable diagnosis/misdiagnosis of CD. According to the main international guidelines,[2,3] we considered as criteria to diagnose CD the positivity of EMA and/or tTG associated with any of the different degrees of Marsh–Oberhuber classification (villous atrophy, crypt hyperplasia, intraepithelial lymphocytosis).[11]

Although it shows a good diagnostic accuracy, we did not include deamidated gliadin antibodies among antibodies taken into account, as it was proven to be outperformed by tTG in a recent metanalysis.[12] Every diagnosis of CD, which met these criteria, was considered as a proper diagnosis, whereas diagnoses that did not meet these criteria were considered as questionable diagnoses. All previous diagnoses that were finally rejected after our evaluation were considered as misdiagnoses of CD.

Criteria for rejecting CD diagnosis. We rejected previous diagnoses of CD in the presence of at least one of the following reasons: absence or negativity of EMA/tTG dosage on gluten-containing diet; absence of histological assessment; unclear histology report; and negativity of HLA-DQ2 or -DQ8. Data regarding those who diagnosed the disease (physicians or patients itself) and about the gluten exposure status (gluten consumption or gluten avoidance) of the patients at the time of the diagnostic process were also collected.

Procedures to evaluate or to re-evaluate CD. Naïve patients referred to us without a prior diagnosis of CD underwent one or more of the following exams to diagnose CD: serology assessment (total serum immunoglobulin A blood test, EMA, tTG); upper endoscopy with duodenal biopsies. Exams were requested only if they had not been performed before, were not available at the time of our examination, or had been performed on GFD.

Among patients with a previous CD diagnosis, when the diagnosis was questionable, its revaluation was carried out through one or more of the following procedures: repetition of serology assessment (total serum immunoglobulin A blood test, EMA, tTG); second reading of duodenal tissue slides by two experienced histopathologists (V.A., R.R.); repetition of duodenal biopsy and subsequent histological evaluation by the same histopathologists; and assessment of HLA status.

All patients showing positivity of EMA and TTG and typical histological signs of CD were not requested to perform HLA testing. Measurement of tTG IgA antibodies was performed by ELISA (Enzyme-Linked Immunosorbent Assay, automated system; Delta Biologicals SRL, Rome, Italy), through human recombinant tTG as antigen; the required threshold for positive tTG was 10 AU/ml. Assessment of EMA IgA was performed by experienced employees, through immunofluorescence analysis on a tissue slide of monkey esophagus (Delta Biologicals SRL). The required threshold for positive EMA was 1:20. All exams were performed at our center.

All patients who were following a GFD at the time of our examination were requested to undergo a minimum 8-week gluten challenge (with at least 3 g of gluten daily) before the diagnostic revaluation, as recommended by the American College of Gastroenterology.[2] We collected at least four biopsies in the second duodenal portion and two biopsies in the duodenal bulb.[2] We took one biopsy per each forcep pass through the working channel of the endoscope, and immediately oriented them for the histological assessment. Both pathologists were blinded to each other during second reading of duodenal tissue slides as well as during the histological evaluation of newly repeated duodenal biopsies.

Statistical analysis. Data were analyzed using the GraphPad Quickcalcs online software (©2014 GraphPad Software, La Jolla, CA, www.graphpad.com; http://www.graphpad.com/quickcalcs). We compared categorical data through a 2 × 2 contingency table, with Fisher's exact test. Interobserver agreement between the two histopathologists (R.R., V.A.) dealing with second reading of duodenal tissue slides was calculated with Cohen's κ-statistics.

RESULTS

From June 2013 to December 2014, a total of 523 patients attended our outpatient clinic for the diagnosis and management of CD. Among these, we recruited for this study 198 patients (males = 50; females = 148; mean age = 39 years old) (38%) at their first examination at our center. All enrolled patients gave their informed consent to participate in the study.

Table 1 Baseline demographic and clinical characteristics of (1) patients at their first examination for CD ("naïve" patients) and (2) patients with a previous diagnosis of CD

Characteristic	Patients
(1) "Naïve" patients	
No. of patients/mean age (s.d.)	91/34 (±15) years old
Females/males	29/62 (32%/68%)
Motivations for suspect of CD (no. of cases—multiple motivations were possibile in a single patient)	
Iron deficiency anemia	75
Fatigue	35
Gastrointestinal symptoms (bloating, diarrhea, abdominal pain)	83
Family history of CD	33
EMA and tTG positivity (%)	58 (64%)
IgA deficiency (%)	0 (0%)
Typical duodenal histology pattern (%)	58 (64%)
	Marsh 1 = 7
	Marsh 2 = 10
	Marsh 3 = 41
Final diagnosis of CD (%)	58 (64%)
(2) Patients with previous diagnosis of CD	
No. of patients/mean age (s.d.)	107/40 (±12) years old
Females/males	40/67 (37%/63%)
Motivations for suspect of CD (no. of cases—multiple motivations were possibile in a single patient)	
Iron deficiency anemia	85
Fatigue	51
Gastrointestinal symptoms (bloating, diarrhea, abdominal pain)	100
Family history of CD	29
EMA and tTG positivity (%)	64 (60%)
IgA deficiency (%)	0 (0%)
Typical duodenal histology pattern (%)	64 (60%)
	Marsh 1 = 12
	Marsh 2 = 12
	Marsh 3 = 40
Final diagnosis of CD (%)	64 (60%)

CD, celiac disease; EMA, endomysium antibody; IgG, immunoglobulin G; s.d., standard deviation; tTG, transglutaminase antibody.

The remaining 325 patients (62%) referred us for follow-up visits of CD, so they were not considered for inclusion.

"Naïve" patients. Of 198 recruited patients, 91 "naïve" patients (male = 29; female = 62; mean age = 34 years old) (46%) referred us without a previous diagnosis of CD, for one or more of the following reasons: signs and symptoms possibly related to the disease (such as gastrointestinal symptoms, fatigue, iron deficiency anemia), family history of CD, positivity of EMA and tTG, typical histological pattern of the duodenum (see details in Table 1). All 91 subjects were on gluten-containing diet at the time of our evaluation. Fifty-eight of them (64–29% of the whole sample) fulfilled the diagnostic criteria and were diagnosed as celiacs; in the remaining 33 patients (36–16% of the whole sample), the diagnosis of CD was ruled out. Final diagnoses of patients with rejected diagnosis of CD are shown in Table 2.

Revaluation of previous CD diagnosis. One-hundred and seven of the 198 recruited patients (54%) came to our observation with a previous CD diagnosis, mainly to receive CD certification (that is required in Italy for obtaining exemption from payment of gluten-free food) or for a follow-up visit. Fifty-two of them (49–26% of the whole sample) presented with confirmed diagnosis of CD, and their follow-up was scheduled.

The remaining 55 patients (51–26% of the whole sample) did not meet the standardized diagnostic criteria for CD and the previous diagnosis was considered questionable. All of them were already on GFD at the time of our examination, and underwent gluten challenge before the diagnostic revaluation. Overall, the gluten challenge period ranged from 3 to 6 months (mean = 4 months). Thirty-nine patients (71%) were diagnosed with CD by their trusted doctor (gastroenterologist, gynecologist, dermatologist, or general practitioner), and 16 (29%) patients instead self-diagnosed the disease after an advice from a friend and/or surfing the Internet. Supportive evidence for the previous diagnoses of CD is shown in Table 3a, as well as the reasons we put forward to reject CD diagnoses. The procedures we thought necessary for a proper

Table 2 Final diagnoses of patients with rejected diagnosis of CD

Diagnosis	Overall	Naïve	Previous doubtful diagnosis
Non-celiac gluten sensitivity[a]	20	5	15
Irritable bowel syndrome[b]	15	8	7
Functional dyspepsia[b]	10	5	5
Lactose intolerance	15	7	8
Giardia lamblia infection	2	0	2
Gastritis	14	8	6
Total number	76	33	43

CD, celiac disease; IgE, immunoglobulin E; NCGS, non-celiac gluten sensitivity; WA, wheat allergy.
[a]The diagnosis of NCGS was hypothesized after the exclusion of CD and WA, when the patient reported the occurrence of symptoms after gluten exposure, with their improvement/disappearance after gluten withdrawal, and recurrence after gluten challenge.[21] WA was excluded by dosage of wheat-specific IgE and skin prick tests.[21]
[b]Diagnosed according to the Rome III Criteria.[22,23]

Table 3a Supportive evidence for previous CD diagnoses in the 55 patients with doubtful diagnosis[a]

Supportive evidence for previous CD diagnosis	No. of patients/% of patients
Positivity of serum antibodies	17 (AGA = 13; EMA = 4[b]; tTG = 4[b])/31 %
Suggestive histological features	20/36 %
Positivity of HLA-DQ2 or HLA-DQ8	26/47 %
Amelioration of symptoms after GFD	16/29 %
Supportive evidence for contesting previous CD diagnosis	**No. of patients/% of patients**
Negativity or absence of EMA and/or tTG	51/93 %
Duodenal biopsy not performed	35/64 %
Unclear histology features	20/36 %
Negativity of HLA-DQ2 or -DQ8	2/4 %

AGA, antigliadin antibody; CD, celiac disease; EMA, endomysium antibody; tTG, transglutaminase antibody.
[a]Multiple evidences were possible in a single patient.
[b]Same patients.

Table 3b Diagnostic procedures performed for the revaluation of previous doubtful diagnoses[a]

Diagnostic procedures	No. of patients
Dosage of EMA and/or tTG ($n = 102$)	EMA = 51; tTG = 51
Upper endoscopy and duodenal biopsies ($n = 35$)	35
Second reading of duodenal tissue slides ($n = 20$)	20
Search of HLA-DQ2 or -DQ8 ($n = 2$)	2

EMA, endomysium antibody; tTG, transglutaminase antibody.
[a]Multiple procedures were possible in a single patient.

diagnostic reassessment of patients are summarized in Table 3b.

After our revaluation, CD diagnosis was confirmed only in 12 of the 55 patients with questionable previous diagnosis (22–6% of the whole sample), whereas it was withdrawn in the remaining 43 patients (78–22% of the whole sample). Strikingly, none of the patients coming with the only positivity of antigliadin antibodies were finally diagnosed with CD. Final diagnoses of patients with a rejected diagnosis of CD are shown in Table 2.

Overall, CD diagnosis was confirmed in 64 of the 107 subjects referred to us with a previous diagnosis (60–32% of the whole sample).

A flow chart of overall patients at different levels of evaluation is available in Figure 1.

After the accomplishment of the complete diagnostic process, CD diagnosis was confirmed in 12 of the 55 patients with a questionable previous diagnosis compared with 58 of the 91 naïve patients without a previous diagnosis of CD, with a statistically significant difference between the two groups (22% vs. 64%; $P < 0.0001$; odds ratio (OR) = 0.16; 95% confidence interval = 0.07–0.34) (Figure 2). Neither gender nor age of patients influenced the rates of final CD diagnosis.

Interobserver agreement between two histopathologists (R.R., V.A.) who reviewed the duodenal tissue slides was excellent ($K = 0.91$).

DISCUSSION

Formerly, CD was considered a rare pediatric disorder, often underdiagnosed or diagnosed with significant delay among adults. A better knowledge of the real prevalence of CD and of its proper diagnosis was advocated <10 years ago.[6] Nevertheless, the increasing awareness of the disease, along with the widespread diffusion of serology antibody screening and with the improvement in diagnostic techniques,[13] moved CD to be recognized as a frequent condition. Therefore, CD started to be commonly investigated among patients with pertinent gastrointestinal and extraintestinal symptoms, as well as among relatives of celiacs. The current estimated prevalence of CD among European adults is about 1%.[7] The disease remains still underdiagnosed in several categories of patients (above all, elderly population and patients presenting with non-classical or asymptomatic CD). Notwithstanding, the overestimation of CD prevalence in general population has been claimed.[14] The number of inconsistent diagnoses is increasing continuously, and represents a real issue for routine clinical practice; furthermore, it is associated with unnecessary expenses related to the exemptions for gluten-free food and to the performance of diagnostic exams.[15] Current Guidelines by the American College of Gastroenterology[2] and the British Society of Gastroenterology[3] recommend that CD should be detected by dosage of specific serum antibodies, and diagnosis should be confirmed by upper endoscopy and duodenal biopsies; patients should keep to a gluten-containing diet during the whole diagnostic process.

In the present study, we found that, among patients referred to us with a questionable previous diagnosis of CD, only 22% ($n = 12$) of them were confirmed to be celiacs, whereas the diagnosis was rejected in 78% of them ($n = 43$). Diagnosis of CD was more frequently confirmed in "naïve" patients without a prior diagnosis of CD compared with those with a questionable previous diagnosis (64% vs. 22%; $P < 0.0001$) (Figure 2).

Overall, CD diagnosis was confirmed only in 64 of the 107 subjects referred to us with a previous diagnosis (60–32% of the whole sample). This is surprising, especially if we consider that rates of CD diagnosis among naïve patients were slightly higher (58 patients –64–29% of the whole sample).

Thirty-nine of 55 patients were first diagnosed with CD by other physicians, including gastroenterologists, gynecologists, dermatologists or general practitioners. We questioned the previous diagnoses for several reasons, including absence or negativity of specific antibodies (EMA and tTG), unclear histological report, and even the absence of duodenal biopsy and negativity of HLA-DQ2/-DQ8.

In particular, 13 patients were previously diagnosed as celiacs because of the positivity of native antigliadin antibodies, which have been dismissed long ago for the diagnosis of CD, and are no longer recommended by international guidelines,[2,3] because of the high variability in their diagnostic accuracy.[16]

These results suggest that a considerable number of physicians are not aware of the current diagnostic criteria for CD. As CD is a systemic disease, and can present with gastrointestinal as well as extraintestinal symptoms, different consultants could be involved in the management of the

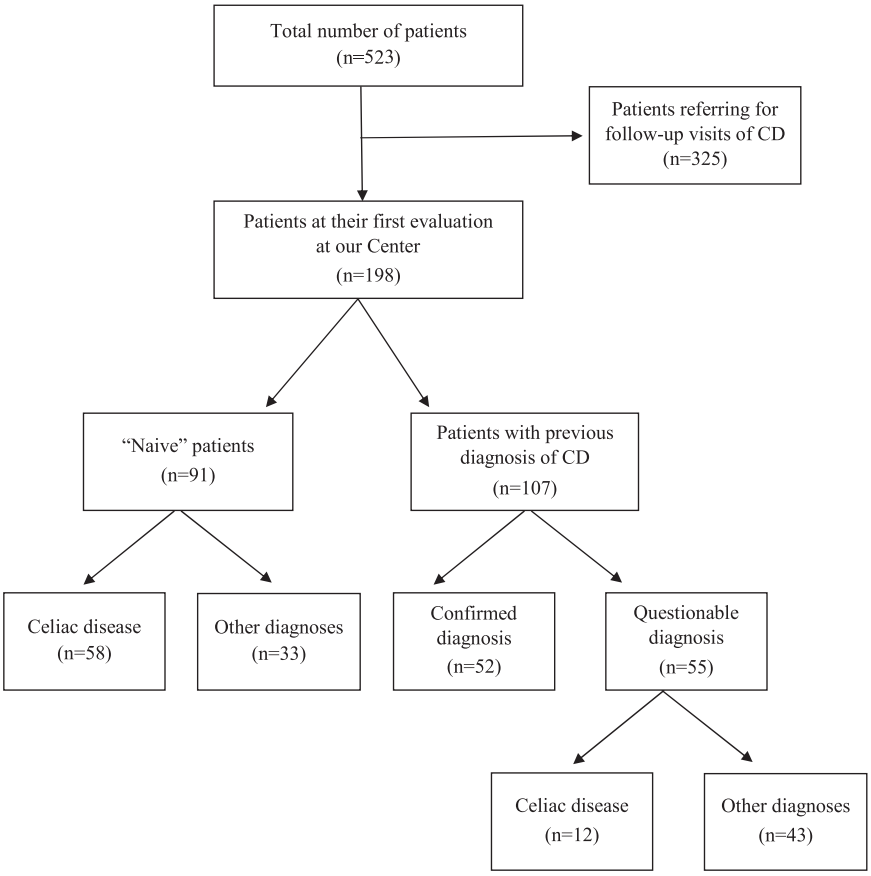

Figure 1 Flow chart of overall patients at different levels of evaluation.

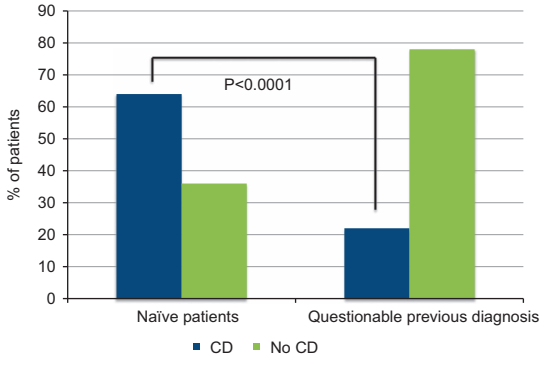

Figure 2 Rates of CD diagnosis confirmation in patients with a questionable diagnosis and in "naïve" patients. CD, celiac disease.

disease, also at the beginning of the diagnostic process, and should be confident with the proper flow chart for the investigation of CD. More strikingly, almost a third of patients (29%) presented us with a self-diagnosed disease, after an advice from a friend and/or surfing the Web, without consulting any physician. Such a phenomenon is not unfrequent,[17] and probably has been increased by the expanding widespread access to the Web.[18]

All 55 patients with a doubtful diagnosis of CD were already on GFD at the time of the examination. Such results could have been influenced by the increasing diffusion of GFD also among patients without CD. GFD represents the primary treatment for uncomplicated CD, and is mandatory for CD patients. Notwithstanding, a considerable part of individuals embrace GFD in the absence of CD, or even without searching for CD and without any prior medical consultation.[8] As a consequence, US gluten-free market is increasing since 2008, reaching more than US$2.5 billion volume in 2010.[19]

This is not the first study assessing the rate of misdiagnosis among patients referred to a tertiary care center for CD. In 2009, Pinto-Sanchez *et al.*[20] described a considerable rate of overdiagnosis of CD in a setting of community medicine, primarily because of histological misdiagnosis. In a 9-year retrospective study, Biagi *et al.*[10] questioned CD diagnosis in 180 of 614 patients, confirming CD in only 61 of them (29.5%). One hundred and seventeen patients (65%) were on GFD at the time of authors' examination. Compared with previous reports, we found higher percentages of people being on GFD at the time of the examination, but lower rates of confirmed CD diagnoses. These findings may reflect that both patients and physicians increased their interest in CD over the past few years, but that this excitement was not associated with a corresponding increase in the awareness of the proper diagnostic process of the disease.

In conclusion, our study shows that a considerable number of patients referred to a tertiary care center experience previous misdiagnosis and/or overdiagnosis of CD. Greater accuracy in the application of the proper diagnostic process and closer adherence to guidelines are needed to minimize the burden of CD misdiagnosis.

CONFLICT OF INTEREST

Guarantor of the article: Gianluca Ianiro, MD.
Specific author contributions: Gianluca Ianiro and Giovanni Cammarota designed the study and wrote the manuscript. Gianluca Ianiro, Stefano Bibbò, and Giovanni Bruno were involved in the recruitment of patients. Riccardo Ricci and Vincenzo Arena were involved in the second reading of tissue duodenal slides. Gianluca Ianiro, Antonio Gasbarrini, and Giovanni Cammarota contributed to the analysis of data. Gianluca Ianiro, Riccardo Ricci, Vincenzo Arena, Antonio Gasbarrini, and Giovanni Cammarota revised critically the manuscript. All authors approved the final version of the manuscript.
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Study Highlights

WHAT IS CURRENT KNOWLEDGE

- ✓ Currently, misdiagnosis of celiac disease (CD) represents an incoming but burdensome issue.
- ✓ Prospective data on misdiagnosis are still not available.

WHAT IS NEW HERE

- ✓ Our results show that a considerable number of patients referred to a tertiary care center are inaccurately diagnosed with CD.
- ✓ CD was confirmed in only 60% of subjects referred to our center with a previous diagnosis of the disease.
- ✓ Diagnosis of CD was more frequently confirmed in “naïve” patients compared with those with a questionable previous diagnosis.

1. Gasbarrini GB, Mangiola F, Gerardi V *et al.* Coeliac disease: an old or a new disease? History of a pathology. *Intern Emerg Med* 2014; **9**: 249–256.
2. Rubio-Tapia A, Hill ID, Kelly CP *et al.* American College of Gastroenterology. ACG clinical guidelines: diagnosis and management of celiac disease. *Am J Gastroenterol* 2013; **108**: 656–676.
3. Ludvigsson JF, Bai JC, Biagi F *et al.* BSG Coeliac Disease Guidelines Development Group. British Society of Gastroenterology. Diagnosis and management of adult coeliac disease: guidelines from the British Society of Gastroenterology. *Gut* 2014; **63**: 1210–1228.
4. Lohi S, Mustalahti K, Kaukinen K *et al.* Increasing prevalence of coeliac disease over time. *Aliment Pharmacol Ther* 2007; **26**: 1217–1225.
5. Corazza GR, Andreani ML, Biagi F *et al.* The smaller size of the 'coeliac iceberg' in adults. *Scand J Gastroenterol* 1997; **32**: 917–919.
6. Green PH. Where are all those patients with celiac disease? *Am J Gastroenterol* 2007; **102**: 1461–1463.
7. Dubé C, Rostom A, Sy R *et al.* The prevalence of celiac disease in average-risk and at-risk Western European populations: a systematic review. *Gastroenterology* 2005; **128**: S57–S67.
8. Leonard MM, Vasagar B. US perspective on gluten-related diseases. *Clin Exp Gastroenterol* 2014; **7**: 25–37.
9. Sapone A, Bai JC, Ciacci C *et al.* Spectrum of gluten-related disorders: consensus on new nomenclature and classification. *BMC Med* 2012; **10**: 13.
10. Biagi F, Bianchi PI, Campanella J *et al.* The impact of misdiagnosing celiac disease at a referral centre. *Can J Gastroenterol* 2009; **23**: 543–545.
11. Oberhuber G, Granditsch G, Vogelsang H. The histopathology of coeliac disease: time for a standardized report scheme for pathologists. *Eur J Gastroenterol Hepatol* 1999; **11**: 1185–1194.
12. Lewis NR, Scott BB. Meta-analysis: deamidated gliadin peptide antibody and tissue transglutaminase antibody compared as screening tests for coeliac disease. *Aliment Pharmacol Ther* 2010; **31**: 73–81.
13. Ianiro G, Gasbarrini A, Cammarota G. Endoscopic tools for the diagnosis and evaluation of celiac disease. *World J Gastroenterol* 2013; **19**: 8562–8570.
14. Biagi F, Klersy C, Balduzzi D *et al.* Are we not over-estimating the prevalence of coeliac disease in the general population? *Ann Med* 2010; **42**: 557–561.
15. Gasbarrini G, Miele L, Malandrino N *et al.* Celiac disease in the 21st century: issues of under- and over-diagnosis. *Int J Immunopathol Pharmacol* 2009; **22**: 1–7.
16. Leffler DA, Schuppan D. Update on serologic testing in celiac disease. *Am J Gastroenterol* 2010; **105**: 2520–2524.
17. Campanella J, Biagi F, Bianchi PI *et al.* A clinical response to gluten withdrawal is not an indicator of coeliac disease. *Scand J Gastroenterol* 2008; **43**: 1311–1314.
18. Copelton DA, Valle G. "You don't need a prescription to go gluten-free": the scientific self-diagnosis of celiac disease. *Soc Sci Med* 2009; **69**: 623–631.
19. Volland A. Gluten-free diet: a cure for some, a fad for most. *US News World Rep* 2008; **145**: 66–68.
20. Pinto-Sánchez MI, Smecuol E, Vázquez H *et al.* Very high rate of misdiagnosis of celiac disease in clinical practice. *Acta Gastroenterol Latinoam* 2009; **39**: 250–253.
21. Volta U, Bardella MT, Calabrò A *et al.* Study Group for Non-Celiac Gluten Sensitivity. An Italian prospective multicenter survey on patients suspected of having non-celiac gluten sensitivity. *BMC Med* 2014;**12**:85.
22. Longstreth GF, Thompson WG, Chey WD *et al.* Functional bowel disorders. *Gastroenterology* 2006; **130**: 1480–1491.
23. Tack J, Talley NJ, Camilleri M *et al.* Functional gastroduodenal disorders. *Gastroenterology* 2006; **130**: 1466–1479.