

Original Article

Prevalence of Celiac Disease among Blood Donors in Sistan and Balouchestan Province, Southeastern Iran

Ali Bahari MD¹, Mehrbod Karimi MD², Ismail Sanei-Moghaddam MD³, Farzad Firouzi MD⁴,
Mohammad Hashemi PhD⁵

Abstract:

Background: The prevalence of celiac disease is common in Iran. The aim of the present study was to determine the prevalence of celiac disease in apparently healthy blood donors of Sistan and Balouchestan Province, southeastern Iran.

Methods: Serum samples of 1600 consecutive apparently healthy blood donors at Zahedan Blood Donation Center were assayed for anti-tissue transglutaminase (tTG) antibody. The levels of IgG antibodies against tTG were screened for all subjects with IgA deficiency. All subjects with positive anti-tTG IgA or IgG were offered upper gastrointestinal endoscopy and duodenal mucosal biopsies.

Results: IgA deficiency was found in 28:1600 (1.8%) of the subjects, among whom 4 cases were positive for IgG-class tTG antibody. Meanwhile, 10 blood donors were positive for anti-tTG IgA antibody. With the exception of 2 subjects who had normal small bowel biopsies, the remainder of the subjects' biopsy findings were compatible with celiac disease. The prevalence of celiac disease was found to be 0.88% (1/114) based on tTGA positivity.

Conclusion: The prevalence of celiac disease among the southeastern Iranian population is high and comparable with other parts of Iran as well as many other countries.

Keywords: Celiac disease, Iran, prevalence, tissue transglutaminase antibody

Introduction

Celiac disease (CD) reflects intolerance to gliadin (a component of gluten) from wheat and related proteins from rye and barley. CD is characterized

Authors' affiliations: ¹Department of Gastroenterology, School of Medicine, Zahedan University of Medical Sciences, Zahedan, ²Department of Pathology, School of Medicine, Zahedan University of Medical Sciences, Zahedan, ³Zahedan Blood Bank Center, Zahedan, ⁴Research Center for Gastroenterology and Liver Diseases, Shahid Beheshti University of Medical Sciences, Tehran, ⁵Department of Clinical Biochemistry, School of Medicine, Zahedan University of Medical Sciences, Zahedan, Iran.

Corresponding author and reprints: Mohammad Hashemi PhD, Department of Clinical Biochemistry, School of Medicine, Zahedan University of Medical Sciences, Zahedan, Iran. Tel: +98-541-244-2881, E-mail: hashemim@zdmu.ac.ir; mhd.hashemi@gmail.com
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by abnormal small intestinal mucosa, signs of immunological activation in the lamina propria of the small bowel, and full recovery from signs and symptoms when placed on a gluten-free diet.¹ CD patients produce antibodies that recognize gliadin, endomysium (an intermyofibril substance found in primate smooth-muscle connective tissue) and transglutaminase (the major autoantigenic component of endomysium).² Serological methods of detecting antibodies to gliadin (AGA), endomysium (EMA) and tissue transglutaminase (tTG antibody) have become the preferred methods of diagnosing and screening both symptomatic and asymptomatic patients for CD.³ However, biopsy of the small intestine remains the "gold standard" for the diagnosis

of CD.^{3,4}

The majority of CD cases are either asymptomatic or have minor/minimal symptoms, suggesting that a considerable number of cases remain undiagnosed and that the true prevalence of the disease is unknown.³ CD is a worldwide health problem and its prevalence varies among different races and nations.⁵⁻¹¹

There are few reports regarding the prevalence of CD in some parts of Iran.^{10,12,13} We screened the blood donors in Sistan and Baluchestan Province, one of the largest provinces in Iran, for presence of CD by serological tests and histological evaluation of the small intestine.

Materials and Methods

This study was performed on 1600 apparently healthy blood donors at Zahedan Blood Donation Center during a four month period (October 2006 to February 2007). The project was approved by the local Ethics Committee of Zahedan University of Medical Sciences and written informed consent was obtained from each individual participating in this study.

All eligible volunteers were consecutively included in the study, in accordance with the following criteria established at baseline: good health, aged between 17 and 65 years and weight greater than 50 kg. The study also included volunteers who would have been excluded by reason of anemia (a hematocrit level lower than 36% for women and lower than 38% for men). The exclusion of these volunteers could well reduce the CD prevalence in the sample of blood donors when compared with the general population, as anemia is one of the clinical symptoms of CD. Among 1600 blood donor volunteers, 7 anemic individuals were included.

Sera of the donors were kept at -80°C until analysis. Sera were screened for tissue transglutaminase (tTG) immunoglobulin A (IgA) and total IgA antibodies. The level of IgG antibodies against tTG was screened for all subjects with IgA deficiency. Donors with positive tTG antibodies (IgA or IgG) were contacted and questioned about symptoms and signs of CD with the use of a semi-structured questionnaire and were offered upper endoscopy and small intestinal biopsy to confirm the diagnosis.

Antibodies against tTG IgG and IgA were tested

with an enzyme-linked immunosorbent assay (ELISA) kit (Genesis Diagnostics Ltd., Cambridgeshire, UK) based on recombinant human tTG as an antigen. Quantitative determination of IgA was done using a commercial kit (Dade Behring Marburg GmbH, Germany). The reference value for IgA was 0.7 – 4 g/L.

Upper gastrointestinal endoscopy was performed with an Olympus endoscope (GIF-V 70, Olympus, Japan) at the Department of Gastroenterology. During the procedure, three duodenal biopsy samples were obtained for routine histological analysis.

Formalin-fixed biopsy specimens stained with hematoxylin and eosin were studied with the use of light microscopy and morphometric techniques. A single expert pathologist, unaware of clinical information, assessed the biopsy specimen according to the scoring system described by Marsh. Marsh 0: normal mucosa, Marsh I: increased number of intra-epithelial lymphocytes, usually exceeding 20 per 100 enterocytes, Marsh II: proliferation of the crypts, Marsh III: partial or complete villous atrophy, and Marsh IV: hypoplasia of the small bowel architecture.¹⁴

Results

Serum samples collected from 1600 apparently healthy blood donors (1418 males and 182 females) with the mean age of 33.2 years (range: 17 – 65 years) were investigated. More men than women were enrolled (8:1) because the blood donors were mostly males in our blood donation center.

There were 28 cases (1.8%) revealed to have IgA deficiency, among which 25 were males and 3 were females. Thus, the prevalence of IgA deficiency was 1.8% (25:1418) among males and 1.6% (3:182) among females.

Of the 28 IgA-deficient subjects, 4 cases were positive for IgG-class tTG antibody. On the other hand, 10 healthy blood donors were positive for tTG IgA antibody. Therefore, a total of 14 subjects were positive for tTG antibodies (IgA- or IgG-class). All 14 positive cases for tTG antibodies were male. The results showed that the prevalence of CD was 0.88% (1/114) based on tTG antibodies' positivity. These subjects were offered upper endoscopy and biopsy for definitive diagnosis of CD, of which 2 donors refused the procedure. Among those biopsied, path-

Table 1. Characteristics of blood donors with positive serological antibodies for celiac disease in Sistan and Baluchestan Province, southeastern Iran

Patient No.	Age (year)	Sex	Symptoms	Anti-tissue transglutaminase (tTG)-antibody	Duodenal biopsy
1	38	Male	Abdominal pain, diarrhea	IgA	Refused
2	52	Male	Asymptomatic	IgA	Marsh 0
3	26	Male	Constipation, abdominal pain	IgA	Marsh III
4	32	Male	Abdominal pain, diarrhea	IgA	Marsh I
5	23	Male	Abdominal pain, diarrhea	IgA	Marsh III
6	29	Male	Constipation	IgA	Marsh II
7	62	Male	Anemia, abdominal pain	IgA	Marsh IV
8	20	Male	Asymptomatic	IgA	Marsh I
9	23	Male	Asymptomatic	IgA	Marsh I
10	32	Male	Abdominal pain, diarrhea	IgA	Marsh III
11	24	Male	Recurrent aphthous stomatitis	IgG	Marsh 0
12	37	Male	Asymptomatic	IgG	Refused
13	21	Male	Short stature	IgG	Marsh III
14	32	Male	Asymptomatic	IgG	Marsh II

ological examination revealed changes compatible with Marsh 0 in two, Marsh I in three, Marsh II in two, Marsh III in four and Marsh IV in one (Table 1).

Discussion

In this study we found that the prevalence of CD is 0.88% (1:114) among the southeastern Iranian population. Although blood donors cannot be considered to be representative of the general population, we selected them because these are apparently healthy individuals, especially regarding nutrition and the absence of serious diseases. In addition, they are easily accessible and the possibility of using blood collected at the blood center makes the study not only less aggressive for the participants, but also more acceptable and feasible.

For screening of CD, we measured IgA and IgG antibodies to human tissue transglutaminase (tTG) as it offers equal or even superior performance to anti-endomysial antibodies (EMA-IgA) using a monkey substrate.¹⁵⁻¹⁸ Meanwhile, total IgA was assessed nephelometrically. Among 14 cases diagnosed as having CD, only 2 had intact intestinal morphology. The remaining subjects had biopsy changes compatible with CD. There are two reasons for this finding: 1) false positive tTG result that is rare but do occur

2) CD enteropathy can be patchy and missed due to sampling error.

All of the 14 cases with positive serological antibodies for CD were males due to the preponderance of male subjects among blood donors at our center (1418 vs. 182). It has been reported that CD occurs in up to 10% of individuals with selective IgA deficiency.¹⁹ On the other hand, selective IgA deficiency occurs in 1 to 2% of patients with CD.²⁰ In this study, IgA deficiency was found in 1.8% of patients with CD.

There are few reports regarding the prevalence of CD in Iran. Akbari et al.¹² reported a prevalence of 0.96% (1:104) in an apparently healthy population in two cities of northern (Sari) and southern (Kerman) Iran. Shahbazkhani et al.¹⁰ reported the prevalence of 0.6% (1/166) of CD in apparently healthy Iranian blood donors at the Tehran Blood Donation Center. In a study performed on 1440 subjects in the city of Shiraz (southern Iran), Saberi-Firouzi et al.¹³ have found a prevalence of 0.56% (1:180) of which they concluded that CD in that region was not as prevalent as other parts of Iran.

The 0.88% (1:114) estimated prevalence for CD found in our study is in agreement with a previous report of the northern and southern parts of Iran,¹² however, it is higher than previously reported on blood donors from Tehran (capital of Iran)¹⁰ and

Shiraz (southern Iran).¹³ This discrepancy in the prevalence of CD in Iran might be associated with ethnic differences between these regions.

Some investigators determined the frequency of CD in diseases such as diabetes mellitus, chronic non-bloody diarrhea and Behcet's Disease in Iran. Shahbazkhani et al. reported a high prevalence of CD (2.4%) in Iranian patients with type I diabetes mellitus²¹ and 19% (19/100) of patients with chronic non-bloody diarrhea.²² In another study Zamani et al.²³ found that the frequency of CD was 1.3% (4/288) among patients with Behcet's Disease.

The prevalence of CD among blood donors shows geographical variation and is approximately 0.74% in Iceland,²⁴ 1.5% in Brazil,²⁵ 2.6% in Mexico,²⁶ 0.14% in Tunisia,⁹ 1.3% in Turkey, and 0.45% in the Czech Republic.²⁷

In the present study histopathological evaluation was performed using the Marsh criteria. The normal upper limit of intraepithelial lymphocytes (IEL) in the distal duodenum may vary in different areas due to ethnic and environmental factors. Nasseri-Moghaddam et al. have reported that IEL levels less than 35/100 epithelial cells in immunohistochemical and 34/100 epithelial cells in hematoxylin-eosin staining are normal.²⁸

In conclusion, the present study detected a 0.88 (1:114) prevalence of CD in apparently healthy blood donors from southeast Iran. Unfortunately, there are only few studies on the prevalence of CD in Iran.^{10,12,13} This is mainly due to lack of a national registry system as well as the idea of rarity of CD in Iran. Furthermore, since Iran is a wide geographic country of different ethnicities, studying the epidemiological aspects of an uncommon disease requires great financial support, which most research centers lack.

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