Original Article

Laboratory Findings of Patients Diagnosed with Celiac Disease

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ABSTRACT

Background:
Celiac disease (CD) is an autoimmune disorder of the small intestine with a genetic predisposition. The gold standard for CD diagnosis is evaluating small bowel biopsy samples. As biopsy sampling is an invasive procedure, researchers focus on finding an alternative non-invasive test that can detect CD at an earlier stage.
We aimed to investigate the laboratory finding of patients with CD to introduce new diagnostic biomarkers for this disorder.

Materials and Methods:
In this cross-sectional study laboratory results of 175 patients with celiac were evaluated. Subjects were analyzed through following variables: age, sex, Marsh classification, hemoglobin level, white blood cell count, platelets, eosinophil, neutrophil, lymphocytes, ferritin, liver enzymes, fat profile, iron profile, calcium, and phosphorus. SPSS software, version 22, was used for analysis.

Results:
69 (39.4%) patients were men and 106 (60.6%) were women with a mean age of 34.4 years. The extraintestinal symptoms were predominant which anemia (28.6%), microcytic erythrocytes (20%) and hypochromic RBCs (26.9%) were the most laboratory findings. Hypertransaminasemia, low ferritin, hypocalcemia and leukemia was reported in 23.4%, 17.1%, 5.7% and 4.7% of patients respectively. According to the pathological classification: 23 (13.1%) patients were Marsh I, 29 (16.6%) patients Marsh II, and 118 (67.4%) patients Marsh III.

Conclusion:
The lack of food, vitamins and minerals can be evaluated through laboratory studies and may open the door to early detection of the disease.

Keywords*:* Celiac disease; Laboratory study; Diagnostic test

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INTRODUCTION

Celiac disease (CD) is a serious autoimmune illness that affects genetically susceptible individuals and damages the small intestine as a result of gluten consumption (1-4). It also causes malabsorption syndrome and gastrointestinal mucosa atrophy. Moreover, CD is one of the most common genetically determined diseases that runs in families. People who have a first-degree family (parent, child, or sibling) with CD have a one-in-ten chance of having CD (5,6). Anemia, osteopenia, miscarriage, liver issues, refractory CD, and lymphoma can all occur if CD is left untreated.

It is thought to impact one out of every 100 people worldwide (7). Approximately 2.5 million undiagnosed Americans are put at risk for long-term health problems (8). CD has been reported to have prevalence rates of 1.4% and 1.6% throughout the world and Asia, respectively. The prevalence of CD was 1% in Iran's general population. However, many CD cases have been left undiagnosed around the world (7,8,9).

CD manifests as steatorrhea, weight loss, developmental problems, abdominal pain, and nutritional symptoms (e.g. vitamin shortage) (10). So far, A stringent, lifelong gluten-free diet (GFD), excluding gluten proteins in wheat (gliadins and glutenins), barley (hordein), rye (secalin), and other related cereals is the only effective treatment for CD (11). Malabsorption and subsequent nutrient shortages can result from the immune-mediated inflammatory response, which can be restored with a GFD.

Among individuals without classic symptoms of CD and in the general population, screening for CD is still a serious problem (12). Bone fractures, lung infections, hyposplenism, intestinal ulcers, collagenous enteritis, and intestinal cancer are some of the difficulties that patients with CD face.

CD is associated with a wide range of gastrointestinal and extraintestinal or is sometimes asymptomatic (1,5,7).

The first step in diagnosing CD is to perform serological tests to measure specific antibodies such as anti- tissue transglutaminase antibody. However, the accurate and gold standard diagnosis of CD is a small bowel biopsy, observing loss of the villous structure, crypt hyperplasia and intraepithelial lymphocytosis (5-8). As biopsy sampling is an invasive procedure and needs certain level of expertise and skill, researchers focus on finding an alternative non-invasive test that can detect CD at an earlier stage (5-7).

We aimed to investigate the laboratory finding of patients with CD referred to Taleghani Hospital Gastrointestinal and Liver Institute to introduce novel biomarkers for CD diagnosis.

MATERIALS AND METHODS

This cross-sectional descriptive study was done on 175 patients with CD who were referred to the Research Center for Gastroenterology and Liver Diseases at Taleghani Hospital in 2016-2017. The CD diagnosis was based on positive serologic tests confirmed by the presence of characteristic small intestinal lesions.

Patients with underlying diseases including diabetes mellitus, kidney and liver disease, thyroid disease, metabolic disease, cancer, history of supplement consumption in the last 6 months, pregnancy, and lactation were excluded from the study. Patients' status after disease confirmation was collected in a separate questionnaire and they underwent a gluten-free diet to improve symptoms.

The patients were analyzed with respect to the following variables: age, sex, pathological level of the disease (according to Marsh criteria), hemoglobin level, white blood cell count, platelets, eosinophil, neutrophil, lymphocytes, ferritin, liver enzymes, fat profile, iron profile, calcium, and phosphorus. Patients were sampled at 8 AM while they were fasting and 10 CC of venous blood was collected for further analysis.

Study methods and their objectives were clearly explained to the patients and they were given enough opportunity to decide to enter the study. The informed consent form was completed by patients and their case was studied after obtaining permission. The protocol of this study was reviewed and approved by the Research Ethics Committee of Shahid Beheshti University of Medical Sciences, Tehran, Iran (IR.SBMU.RETECH.REC).

Statistical analysis was performed using the SPSS software (Version 22.0, SPSS Inc., Chicago, IL, USA). The data were summarized and classified using descriptive

statistics, central indications, and dispersion. One-way analysis of variance (one-way ANOVA) was used to analyze data and test the hypotheses, and the significant level was considered to be 0.05.

RESULTS

The mean ± SD age of the participants was 34.4±13.3 years (range: 3-71 years). Of the 175 participants 106 (60.6%) were women and 69 (39.4%) were men and most of them (67.4 %) were Marsh III degree (Table 1).

The Mean Mean Corpuscular Volume (MCV), hemoglobin (Hgb), white blood cells (WBCs), Cholesterol (Chol), and Sodium (Na), Calcium (Ca) levels were normal, but mean corpuscular hemoglobin (MCH) decreased (Table 2).

50 (28.6%) patients had anemia, 35 (20%) patients had microcytic erythrocytes and 47 (26.9%) had hypochromic RBCs. Moreover, 41 (23.4%) patients had increased liver enzyme levels (including SGPT, SGOT and ALK-P), 30 (17.1%) had low ferritin (less than 12), 13 (4.7%) had leukemia and 10 (5.7%) had hypocalcemia (calcium less than 8.5) (Figure 1).

One-way analysis of variance (one-way ANOVA) showed a significant correlation between the age of a patient with CD and phosphorus levels (Table 3). Also, one way ANOVA showed a significant relationship between hemoglobin, hematocrit, and platelets with sex ($p < 0.05$). And this test also showed that there was no significant relationship between the patient's laboratory and pathology results.

DISCUSSION

Despite advances in diagnostics, there are a number of reasons to continue looking for new biomarkers to improve CD diagnosis and follow-up. In this study, the laboratory results of 175 patients with CD were examined.

Regarding laboratory results and patients' clinical signs, there was a significant relationship between the number of neutrophils in patients with cramps, hemoglobin with bloating, SGOT and ferritin with neurological symptoms, MCV and MCH with cutaneous symptoms, and thus these variables had a higher amount than the average in symptomatic individuals. The relationship between

Table 2: Mean±SD of Laboratory tests

Variables	Mean SD	Max	Min	Change*
MCV	82.7±9.1	109.4	57.3	Normal
MCH	26.9±3.7	38	15	Decreased
Hgb	12.8±2.1	17.6	8.1	Normal
Hct	39.6±6.7	88.5	29.2	Normal
WBC (10^3)	6.2±2.1	17.6	2.1	Normal
Neut	54.2±11.5	76	1.9	Normal
Lym	36.8±9.9	63	1.6	Normal
Eos	2.4±1.9	13.3	0.0	Normal
PLT (10^3)	259.8±90.3	588	30.1	Normal
TIBC	338.6±105.5	518	3.3	Normal
Ferritin	30.5±47.5	310	1	Normal
Fe	75.5±4	176	3	Normal
TG	100.8±40.3	237	52	Normal
Chol	171.1±43.1	283	100	Normal
Bill T	0.9±0.4	2.6	0.2	Normal
Bill D	0.3±0.2	1.3	0.08	Normal
Na	137.8±4.7	144	122	Normal
K	4.5±0.4	4.9	3.3	Normal
Ca	8.9±1.7	15.3	0.8	Normal
P	4.1±1.1	9	2.3	Normal
SGPT	35.8±32.7	249	8	Normal
SGOT	32.1±19.1	118	9	Normal
ALK-P	253.4±165.1	764	65	Normal

*Change compared to normal range

Table 1: Characteristic of participants

Variables	Group	Frequency (Percent)	*P* value
Sex	Female	106 (60.6 %)	0.58
	Male	69 (39.4 %)	
Chief Compliant	Extraintestinal symptoms	128 (73.1 %)	0.04
	Intestinal symptoms	47 (26.9 %)	
Pathological degree	Marsh I	23 (13.1 %)	0.03
	Marsh II	29 (16.6 %)	
	Marsh III	118 (67.4 %)	
	Missing	5 (2.9 %)	

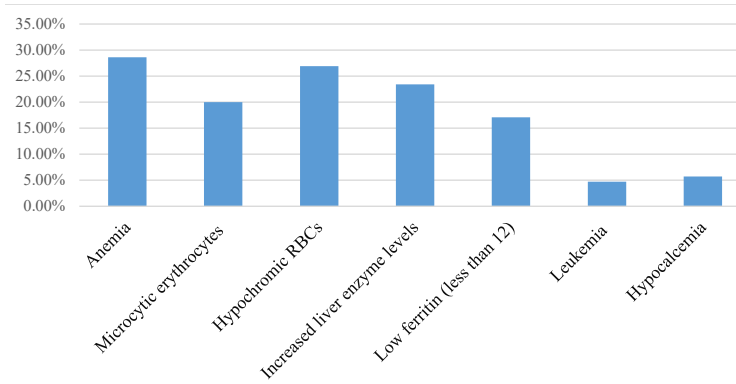

Fig.1: Common abnormalities in patients with CD

Table 3: One-way variance analysis of clinical variables and symptoms

Symptoms	sig Ferritin	sig P	sig WBC	sig Neut	sig Eos	sig Hgb	sig Plt	sig MCV	sig MCH	sig SGOT	sig SGPT	sig ALK
Diarrhea	0.061	0.141	0.991	0.760	0.993	0.916	0.895	0.851	0.880	0.617	0.274	0.383
Bloating	0.058	0.349	0.936	0.051	0.967	**0.034**	0.248	0.270	0.077	0.129	0.244	0.437
Cramp	0.923	0.488	0.466	**0.027**	0.874	0.400	0.269	0.235	0.149	0.361	0.663	0.362
Weight loss	0.358	0.834	0.608	0.093	0.732	0.110	0.508	0.733	0.945	0.129	0.110	0.259
Nausea	0.139	0.705	0.994	0.143	0.707	0.857	0.206	0.335	0.191	0.438	0.086	0.956
Neurological symptoms	**0.026**	0.790	0.874	0.504	0.374	0.272	0.260	0.502	0.486	**0.036**	0.108	0.914
Cutaneous symptoms	0.925	0.684	0.095	0.295	0.240	0.807	0.738	**0.032**	**0.031**	0.365	0.252	0.186

laboratory results and the degree of the pathology of the disease (marsh) was not significant.

According to a previous study, patients with CD on a normal diet often had very low ferritin levels, and ferritin level increased after adherence to a gluten-free diet (13). Also, population research in the United States showed no link between blood hemoglobin and undiagnosed CD; however, no link was found between lower serum ferritin levels and undiagnosed CD. Furthermore, in a study on symptoms and biomarkers associated with undiagnosed celiac seropositivity, a significant association was seen between levels of serum ferritin and CD. In our study also, the level of ferritin reduced in 17% of the patients. Hence, it can be concluded that there is a link between abnormal intestinal changes and low ferritin levels in CD, and both improve following a GFD (13).

Lewis and colleagues (2009) measured the fat profiles of 100 patients with CD referred to Royal Hallamshire at the time of diagnosis and compared them with the Health Survey for England as well as the cholesterol levels of patients after receiving a 12-month GFD (14). The mean±SD cholesterol level was 4.84±2.1 (dL) in adults who had recently been diagnosed with CD (mean age of 51 years) (14). According to their study, in patients with CD, the level of total cholesterol at the time of diagnosis was lower than the general population and did not increase with a year of BFD, while the level of HDL cholesterol increased slightly after treatment (14). In another study, the researchers did not find significant symptoms, but for low serum cholesterol measurements and low blood hemoglobin they discovered a significant association with undiagnosed CD (15). Also, Karhus and co-workers found an association between reduced level of cholesterol and undiagnosed CD (16). On the contrary, in our study no significant change was observed in the fat profile of the patients.

Liver dysfunction in CD ranges from asymptomatic elevated liver enzymes or nonspecific hepatitis to chronic liver disease. Liver enzymes changes in patients with CD were reported by Hagander and co-workers in 1977. They reported that transaminases were often elevated in untreated CD and return to normal with a strict GFD (17). Following that, Bonamico and colleagues discovered elevated liver enzyme levels in 39 of 65 children with CD (60%) (18). Furthermore, Bardella and co-workers (7) examined 158 adult patients with CD for abnormal liver enzyme levels in another key study and reported the same results about the increased liver enzyme levels (19,20). 23.4% of our patients also had increased liver enzyme levels (including SGPT, SGOT and ALK-P). Thus, this study confirms the significant association between elevated liver enzyme levels and CD.

According to a review, CD was a common systemic disorder with multiple hematologic manifestations (21). Iron deficiency anemia is very common in CD and is reported in up to 46% of subclinical cases (12). Iron deficiency anemia usually presents as microcytic anemia, hypochromic, and in patients with markedly low serum iron, the valence of Iron binding and ferritin was high (22). In our study, 35 (20%) patients had microcytic erythrocytes and 47 (26.9%) had hypochromic RBCs.

Thrombocytosis seems to be more common in CD than thrombocytopenia, occurring in up to 60% of patients. An abnormally low white blood cell count has been reported in a few children with CD (12). But in our study white blood cell count was normal in patients with CD.

Since the first description of CD, there has been significant growth in public awareness of the condition. However, CD appears to be on the rise worldwide, although being underdiagnosed. Despite advancements in diagnostics, there are various reasons to continue looking for new biomarkers to improve CD diagnosis and follow-up. Our study showed that low ferritin, anemia, and increased liver enzyme levels were associated with CD. Also, thrombocytosis was common in patients with CD. It must be considered that changes in laboratory variables may have different causes than a specific disease. Further studies with larger sample sizes are needed to confirm/ reject the results of our study.

CONFLICTS OF INTEREST

The authors declare no conflict of interest related to this work.

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