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Grinding Halt Celiac Disease by Gluten Free Diet

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Abstract: The aim of this study is to assess the presence of autoantibodies in the studied groups and their significance in the detection of celiac disease (CD), and its relationship with gluten free diet (GFD). This study included 90 subjects who were divided into 60 CD and 30 control groups based on the age and gender. Also, the patients group further into three groups according to period of GFD : (13) without GFD patient , (30) with GFD for period <1year and (17) with GFD for period >1year . All serological tests of autoantibodies were conducted by the Enzyme linked Immunosorbent Assay (ELISA) technology. The findings revealed that all patients were seropositive for autoantibodies profile when compared to healthy controls, with a highly significant difference (p = 0.000). Relative remission of serological tests was seen in (30) patients treated for <1 year, while (17) patients treated for >1 year showed a clear reduction in autoantibodies levels when compared to (13) patients without GFD. In conclusion, immunological tests play an important role in the prognosis of CD. Repeating the immunologic screen 1 year after diagnosis and starting a GFD supports the routine measurement of using it as a gold standard test to confirm recovery of Iraqi CD patients.

Keywords: Celiac disease; GFD; Serological tests; Autoantibodies.

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Introduction

Celiac disease (CD) is a common autoimmune disorder triggered by the ingestion of gluten in genetically susceptible individuals(1). It is an immune response to dietary gluten leads to inflammation and subsequent atrophy of small intestinal villi, causing severe bowel discomfort and malabsorption of nutrients. The major instigating factor for the immune response in celiac disease is the activation of gluten‑specific CD4+ T cells expressing T cell receptors that recognize gluten peptides presented in the context of HLA‑DQ2 and DQ8(2). Celiac disease has become more common in recent decades, with a global prevalence of about 1-1.5%, thanks in part to improved diagnostic tools and a shift in the environment's response to dietary gluten(3). The prevalence of CD is roughly equal in Arabian countries, with Saudi Arabia having the highest rate of up to 3.2 % (4). Gluten is an ethanol-soluble protein that is resistant to digestion. It is composed of two types of proteins, gliadin and glutenin, which are complex proteins high in glutamines and prolines that bind to form a network that supports dough and allows bread to be light and fluffy. Both of these components are resistant to intestinal digestive enzymes, which can result in a dangerous inflammatory process with long-term consequences. Gluten can be

found in grains such as wheat, kamut, and barley(5). Gliadin in food cannot be digested by intestinal enzymes in genetically susceptible people. This could result in an inflammatory response in the intestine(6).

A gluten-free diet may aid in the prevention of small bowel adenocarcinoma as a late sequel(7). Early detection of CD may help to avoid serious complications like iron deficiency anemia, bone growth problems, and infertility (8). Currently, in adult patients a diagnosis of celiac disease is usually made through a combination of serological testing and endoscopic biopsy. A common approach is to firstly screen for serum IgA antibodies to transglutaminase (anti-tTG IgA) and IgG antibodies to deamidated gliadin peptides (anti-DGP IgG) (9). The diagnosis is confirmed by histological examination of the small intestinal mucosa, where intraepithelial lymphocytosis crypt hyperplasia and villous atrophy, are the pathological hallmarks of the disease(10).

Materials and methods

This research was carried out between the 15th of November 2021 and the 15th of February 2022 on sixty celiac disease Iraqi patients (17 males and 43 females) with age rannged 7-76 years who were clinically diagnosed by a consultant medical staff in the Hilla Teaching Hospital Bablon/ Iraq, Central Public Health Laboratory (CPHL) Baghdad/ Iraq and thirty apparently healthy individuals as control who had been randomly selected to be matched with the patients regarding to age and gender. Informed consent was obtained from all the study participants. Blood samples were collected from sixty celiac disease patients and from control group to obtain sera. All the samples subjected to immunological test as Anti-tissue-Transglutaminase (tTg), Anti-giliadin antibody (AGA), Anti-Deamidated giliadin peptide (DGP) and Anti-endomysial antibody (AEA) Serum Autoantibodies profile IgA and IgG was determine by using ELISA Kit (MyBioSource, USA).

Statistical analysis

The Statistical Program for Social Science (SPSS) was used to evaluate the effect of different factors in study parameters. T-test was used to significant compare between percentage and least significant difference . Values less than (0.05)* and (0.01)** were considered to be statistically significant and high significant respectively.

Results and discussion

The patients were divided into six groups based on age . It was found that the highest CD rate appeared in the age group (21-30) years (35.0%), among the categories of patients with no significant difference between the six age groups (P = 0.933). However, in terms of gender, females outnumbered males (71.7% vs. 28.3%, respectively, female to male ratio) (2:1). The numerical difference was not statistically significant (p = 0.190). Serum level measurement of anti-tTG IgA and anti-tTG IgG in the patients as compared with control revealed that, the mean ± SD of anti-tTG IgA level was 3.83 ± 2.57 ng/ml in the patient group while in the control group was 1.81 ± 0.48 ng/ml, and anti-tTG IgG level was 0.48 ± 0.24 ng/ml in the group of patients. In the control group was 0.19 ± 0.02 ng/ml, it was clear that the mean serum levels of anti-tTG IgA and anti-tTG IgG were significantly higher in

patients with CD than in control with a significant difference (p value = 0.000) for each of them as shown in table (1).

Table (1): Anti-tissue transglutaminase IgA and IgG serum level

Parameters		Number	Mean	SD	P value
tTG IgA (ng/ml)	Control	30	1.81	0.48	0.000**
	Patient	60	3.83	2.57	
tTG IgG (ng/ml)	Control	30	0.19	0.02	
	Patient	60	0.48	0.24	

Serum level measurement of AGA IgA and AGA IgG in the patients as compared with control clarified that, the mean ± SD of AGA IgA level was 0.59±0.33 ng/ml in the patient group while it was 0.14±0.02 ng/ml in the control group, for AGA IgG the serum level was 0.32±0.04 ng/ml and 0.1±0.02 ng/ml, in patients and control group respectively through the results, it was noted that the mean serum levels of AGA IgA and AGA IgG are significantly higher in CD than in control with a significant difference (p value = 0.000) for each of them as shown in table (2).

Table (2): Anti-gliadin antibody IgA and IgG serum level

Parameters		Number	Mean	SD	P value
AGA IgA (ng/ml)	Control	30	0.14	0.02	0.000**
	Patient	60	0.59	0.33	
AGA IgG (ng/ml)	Control	30	0.02	0.01	
	Patient	60	0.32	0.04	

Other autoantibodies as DGP IgA, DGP IgG and AEA IgA were estimation in the patients in comparison with control, the mean of these three parameters was 0.56±0.36 ng/ml, 0.43± 0.21 ng/ml, 222.41±78.45 ng/ml respectively in patients group corresponding to 0.16 ± 0.02, 0.17 ± 0.02, 125.41 ± 36.18 ng/ml respectively in control group . From these results , it was observed that, the mean serum levels of AGA IgA, AGA IgG and AEA IgA were significantly higher in patients with CD than in control with significant difference P value (p = 0.000) for each of them as shown in Table (3).

Table (3): Anti-deamidated gliadin peptide IgA, IgG and anti-endomysial antibody IgA

Parameters		Number	Mean	SD	P value
DGP IgA	Control	30	0.16	0.02	0.000**
	Patient	60	0.56	0.36	
DGP IgG	Control	30	0.17	0.02	
	Patient	60	0.43	0.21	
END IgA	Control	30	125.41	36.18	
	Patient	60	222.48	78.45	

These findings is consistent with a previous studies such as Velikova *et al*. (11) in Bulgaria , as well as a recent

local study by Alattabi *et al* (12). In both studies, serum levels of anti-tTG IgA and anti-TTG IgG were significantly higher in patients with CD than in control subjects. the ingested of gluten elicits the formation of tissue autoantibodies IgA class in the serum of untreated patients with celiac disease . These antibodies appear to be very specified for celiac disease. In Turkey, Hazar *et al* (13) found that mean serum levels of AGA IgA and AGA IgG were significantly higher in CD patients than in the control group. Because AGA IgA and AGA IgG antibodies are highly accurate in diagnosing celiac disease, antibody concentrations in assay panels correlate with the degree of mucositis. It has been proposed that autoantibodies that bind to receptors in different tissues via immune interaction are responsible for celiac disease's multisystem involvement. In addition to the local study by Elia *et al* (14). The mean serum levels of AGA IgA and AGA IgG were significantly higher in patients with CD than in the control group and patients with gastrointestinal disorders respond to gliadin peptide, which is reflected in the high prevalence of anti-gliadin antibodies. Sensitivity and specificity for IgA and anti-gliadin IgG antibodies are up to 91% and 94% for IgA and up to 88% and 92% for IgG . In the meantime, it was noted in two earlier investigations carried out by Walker *et al* (15) and Volta *et al* (16). In both trials, CD patients had considerably higher mean serum levels of anti-DGP IgA and anti-DGP IgG than did healthy people. However, Singh *et al* (17) found in their investigations that CD patients had significantly higher mean blood levels of anti-AEA IgA than did control subjects, with a significant difference in the p-value.

The patients were divided into three groups to illustrate the possible association of positive serum levels of the immune markers used in this study with the duration of the (GFD). The first group without GFD ,second group with period <1year GFD , and the third group with period >1year GFD. There was a clear rise in the mean±SD serum level of immunological parameters as shown in Table (4). In patients who did not abstain from gluten, then patients who abstained from eating gluten for one year, and they are followed in the last place patients who abstained from eating gluten for more than one year, with significant difference in AGA IgA, AEA IgA (P value 0.008, 0.009) respectively. it was observed that the longer the period of gluten abstinence, the lower the mean±SD serum level for the above mentioned immunological criteria. The results obtained in this study showing that GFD can significantly improve serum levels of autoantibodies thus improve gastrointestinal symptoms and through it, it was found that there is a possible link between autoantibodies, on the one hand, and gluten on the other. This result is consistent with Kurppa *et al* (18), who demonstrated in his study that patients benefit from a GFD as treatment to reduction the clinical symptoms and serum level of immunological parameters and they saw improvement in several objective disease scores with GFD. Furthermore, the findings were consistent with Webb *et al* (19) study in Sweden, which discovered that patients who followed a gluten-free diet for at least one year had significantly fewer symptoms and lower concentrations of serological tests than

those who did not follow the GFD. In addition his observed that patients who avoided gluten had significantly lower serum levels of serological tests for untreated celiac disease patients. It is currently the most popular treatment.

Table (4): Relationship serum levels of autoantibodies with GFD.

Parameters		Number	Mean	SD	*P.* value
tTG IgA	**No GFD**	13	4.67	1.56	0.290
	<1 Year	30	3.84	3.08	
	>1 Year	17	3.17	2.01	
tTG IgG	**No GFD**	13	0.59	0.29	0.200
	<1 Year	30	0.46	0.25	
	>1 Year	17	0.44	0.17	
AGA IgA	**No GFD**	13	0.69	0.36	0.008**
	<1 Year	30	0.67	0.34	
	>1 Year	17	0.38	0.19	
AGA IgG	**No GFD**	13	0.62	0.33	0.302
	<1 Year	30	0.55	0.34	
	>1 Year	17	0.44	0.27	
DGP IgA	**No GFD**	13	0.68	0.41	0.131
	<1 Year	30	0.59	0.36	
	>1 Year	17	0.43	0.27	
DGP IgG	**No GFD**	13	0.53	0.27	0.113
	<1 Year	30	0.42	0.18	
	>1 Year	17	0.36	0.19	
AEA IgA	**No GFD**	13	243.11	86.37	0.009**
	<1 Year	30	240.83	84.47	
	>1 Year	17	174.33	26.96	

In conclusion, autoantibodies are an excellent screening tests for CD Therefore, it can be safely recommended that patients having even fewer clinical features should be screened by tTG, AGA, DGP and AEA to detect CD patients with minimal signs and symptoms. So far, it continues to be true that the only effective and safe treatment for CD is strict GFD for life.

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