ISSN: 0067-2904

The Association of Gluten-Free Diet and Liver Function in Iraqi Patients with Celiac Disease

Zahraa Amer Mohammed*, Nada A. Kadhim
Department of Chemistry, College of Science, University of Baghdad, Baghdad, Iraq

Received: 26/3/2023 Accepted: 7/6/2023 Published: 30/6/2024

Abstract

Measurement of anti-TTG, liver manifestation, and BMI in Iraqi patients with celiac disease (CD) with and without a gluten-free diet (GFD) in order to validate their role in the exacerbation of CD symptoms. The study was carried out on CD patients with a duration of disease ranging from 1 to 10 years (n = 52, age range: 20-55). The patients' group was subdivided into the PI group (n = 29), which includes patients with GFD, and the PII group (n = 23), which includes patients without GFD. The studied groups were matched with 51 healthy individuals as the control group (C) (age range: 20-55). The results obtained from the present study indicate a highly significant increase (P = 0.000) in serum anti-TTG in the PI and PII groups as compared with the C group. BMI values showed a highly significant decrease (P = 0.007) in the PI and PII groups as compared with the C group. The activity of GGT, ALT, AST, and GOT/GPT ratio showed no significant difference in the P, PI, and PII groups as compared with the C group, while a highly significant increase in GGT was noticed upon the comparison of PI with PII (P = 0.004). It was concluded from the current study that high levels of TTG are associated with CD patients regardless of adherence to the GFD or not, while the high activity of GGT is associated with long-term adherence to the GFD.

Keywords: Anti-Tissue transglutaminase, Body mass index, Celiac disease, Gluten-free diet, Liver enzymes.

ارتباط النظام الغذائي الخالي من الغلوتين بوظائف الكبد عند المرضى العراقيين المصابين بمرض Celiac disease

زهراء عامر محمد*، ندى عبد الكريم كاظم
[1] قسم علوم الكيمياء، كلية العلوم، جامعة بغداد، العراق.

الخلاصة

قياس مضاد TTG ، وظائف الكبد ، ومؤشر كتلة الجسم ، في المرضى العراقيين المصابين بمرض Celiac diseaseمع أوبدون GFD. أجريت الدراسة على مرضى CD الذين تتراوح مدة المرض لديهم لمدى من 1 الى 10 سنوات (ن=52، العمر 55−20). تم تقسيم مجموعة المرضى إلى PI (ن=29) والتي تشمل المرضى الملتزمين بالحمية الخالية من الكلوتين، ومجموعة PII (ن=23) والتي تشمل المرضى الغير ملتزمين بالحمية الغذائية. تم مطابقة مجاميع الدراسة مع 51 فردًا سليمًا كمجموعة التحكم (C) (العمر55−20) النتائج التي تم الحصول عليها من هذه الدراسة تشير إلى زيادة معنوية عالية (*p = 0.000*) في مضاد TTG في

*Email: zahraa.aamer1205m@sc.uobaghdad.edu.iq

مجموعات PI، PII بالمقارنة مع المجموعة C. أظهرت قيم مؤشر كتلة الجسم انخفاضًا معنويًا عاليًا ($P = 0.007$) في مجموعات و PI و PII مقارنة بالمجموعة C. لم يُظهر نشاط GGT ، ALT ، AST ،ونسبة GOT / GPT أي فرق معنوي في مجموعات P، PI ، و PII مقارنة بالمجموعة C بينما لوحظ زيادة كبيرة في فعالية GGT عند مقارنة ($P = 0.004$) PI مع PII. استنتج من الدراسة الحالية أن المستويات العالية من مضادات TTG مرتبطة بمرضCD ، في حين أن زيادة فعالية GGT يرتبط بإزدياد فترة المرض للمرضى الملتزمين بالحمية GFD.

1. Introduction

Celiac disease (CD) is a persistent illness characterized by a lifelong sensitivity to gluten, which results in immunologically induced inflammatory injury to the small intestinal mucosa [1]. Individuals who are genetically prone and have a variable mix of gluten-dependent clinical symptoms, celiac disease CD-specific antibodies, HLA-DQ2 or HLA-DQ8 haplotypes, and enteropathy. The CD has a global incidence of 1.4% and is more prevalent among children than adults [2]. The classical celiac disease is characterized by a broad variety of gastrointestinal and extra-intestinal symptoms, including diarrhea, weight loss, stomach distention, failure to flourish, malabsorption, and iron deficiency anemia [2, 3]. However, some individuals experience extra-intestinal signs such as liver abnormalities [4]. For screening and confirmation of CD, serologic assays such as serum immunoglobulin G (IgG) and/or immunoglobulin A (IgA) tissue transglutaminase antibodies, deamidated gliadin peptide antibodies (IgG class), and IgA endomysial antibodies must be carried. Furthermore, in many countries, a biopsy of the small intestine is still required to prove the diagnosis [4]. The best diagnostic test for patients with CD on GFD has been found to be the anti-tissue transglutaminase (anti-TTG) IgA antibody assay [5]. The time it takes for anti-TTG titres to normalize after starting a GFD is clinically important, and it is a frequent question and source of concern for CD individuals and their families [6-7]. The therapy for CD is a gluten-free diet that must be followed for the rest of one's life (GFD). However, accidental gluten exposure in a GFD is widespread and sporadic [6]. According to recent research, most CD patients can only achieve a gluten-reduced diet rather than the suggested rigid GFD [6]. Gluten exposure may be more prevalent than previously thought, and it differs from slips in an otherwise strictly adhered-to GFD [7]. Gluten's pervasiveness, food cross-contamination, and the restrictions and socio-emotional toll are all major reasons for gluten exposure in a GFD [8]. Gamma-glutamyl transferase (GGT), aspartate aminotransferase (AST), and alanine aminotransferase (ALT) are enzymes that are mostly found in the liver but also in the heart, erythrocytes, pancreas, muscle cells, and kidneys [11]. When an organ or body cell, such as the heart or liver, is damaged, ALT is secreted from the tissues into the bloodstream, producing an increase in enzyme level [11,12]. Asymptomatic liver enzyme increases, vague hepatitis, or chronic liver disease characterize hepatic dysfunction in CD. Many studies look at the proportion of people who have hypertransaminasemia or the relationship between CD and liver diseases, yet they do not look at other abnormal hepatic alterations [9,10]. Extra-intestinal manifestations and celiac disease are linked to liver damage [13]. This study aimed to evaluate liver function in CD patients and study the effect of a gluten-free diet on the levels of anti-TTG and the activity of liver enzymes. In addition, investigate if there is any correlation between anti-TTG and other biochemical parameters for all studied groups.

2. Materials and methods

The collection of blood from patients went off at the medical city, Baghdad hospital (The digestive system and liver department) in Baghdad, Iraq, from October 2021 to March 2022. Patients with a formal history of celiac disease (P, n = 52) (male = 24, female = 28) (aged range 20-55 years) and healthy individuals (C, n = 51) (male = 25, female = 26) (aged range 20-55 years) were included in the present study. The duration of the disease in celiac patients

was 1-10 years. Each patient was characterized by either a biopsy or clinical tests. In addition, a second division was applied to the P group subject to adherence to gluten-free food (PI = 29 with a gluten-free diet and PII = 23 with a non-gluten-free diet).

2.1. Inclusions and exclusions criteria

Patients and controls with obesity, cancer, hepatitis, liver diseases, active inflammatory conditions, irritable bowel disease, and alcohol consumption that may interfere with this study were excluded. Otherwise, CD patients with and without GFD were selected to be studied. The CD patients were diagnosed by laboratory tests or a small intestine biopsy.

2.2. Blood samples collection

Venepuncture was used to take 10 mL of blood from a research participant who had been fasting for at least 8-10 hours. The blood sample was allotted to a gel tube and then left to clot at room temperature. All gel tubes were centrifuged at 3000 × g for ten minutes to collect serum, which is used for the evaluation of anti-tissue transglutaminase and liver enzymes.

2.3. Laboratory tests

Anti-tissue transglutaminase was measured using an ELISA enzyme-linked immunosorbent assay using the commercially available ELISA (My BioSource) kit (U.S.A). Gamma-glutamyl transferase (GGT), alanine aminotransferase (ALT), and aspartate aminotransferase (AST) were determined using the HUMAN, Germany kit. The procedures listed in the manufacturer's instructions were carried out in this study.

2.4. Statistical analysis

SPSS software version 22 was used to conduct the statistical analysis of the statistics. The variables are described as means ± standard deviation. One-way ANOVA and the post hoc Tukey test were used to compare groups. A statistically significant difference was considered with a *P* value of < 0.05. While high significance was considered whenever the *P* value was less than 0.01 and very high significance was considered whenever the *P* value was less than 0.001. The correlation between parameters was determined by Pearson correlation coefficients (r-values).

3. Results and discussion

The results listed in Table 1 revealed that no significant differences were found in age between the P group and the C group. While anti-TTG levels have a highly significant increase ($P = 0.000$) in the P (10.84 ± 5.67 U/L) group as compared with the C group (1.90 ± 0.95 U/L). Moreover, BMI showed a highly significant decrease ($P = 0.000$) in the P group (20.03 ± 2.65 Kg/m^2) as compared with the C group (22.45 ± 2.36 Kg/m^2).

Table 1: The mean ±SD values of the anti-TTG, age, and BMI of the P group compared to the C group

Parameters	Control (C) (Mean ± SD) (N = 51)	Patients (P) (Mean ± SD) (N = 52)	P-value
Anti-TTG (U/L)	1.90±0.95	10.84±5.67	0.000
Age (Year)	37.43±9.72	35.85±10.28	0.688
BMI (Kg/m^2)	22.45±2.36	20.03±2.65	0.000

Significant:*P<0.01; **P<0.001; no Significant: P>0.05.

In addition, Table 2 showed that the activity of liver enzymes (GGT, ALT, and AST) and the ratio of AST/ALT have no significant differences as compared between the P groups and the C groups.

Table 2: The mean ±SD of the activity of GGT, ALT, AST, and the ratio of AST/ALT for the P group compared to the C group

Parameters	Control (C) (Mean± SD) (N = 51)	Patient (P) (Mean± SD) (N = 52)	P-value
GGT (U/L)	23.83±7.06	22.93±8.50	0.829
ALT(U/L)	17.37±4.50	17.79±6.72	0.938
AST (U/L)	15.47±7.18	14.17±6.64	0.607
AST/ALT	0.91±0.52	0.76±0.08	0.053

Significant:*P<0.05; **P<0.001; no Significant: P>0.05.

Upon comparison between the studied parameters depending on diet adherence, the results showed no significant difference in age among all studied groups (Table 3).

Table 3: The mean ±SD values of the anti-TTG, age, and BMI of the PI and PII groups compared to the C group

Parameters	Control (C) (Mean ± SD) (N = 51)	PI with GFD (Mean ± SD) (N = 29)	PII without GFD (Mean ± SD) (N = 23)	P-value	
Anti -TTG(U/L)	1.90±0.95	11.12±6.45	10.99±6.16	C&PI	0.000
				C&PII	0.000
				PI&PII	0.995
Age (Year)	37.43±9.72	37.65±10.06	33.95±8.47	C&PI	0.994
				C&PII	0.321
				PI&PII	0.353
BMI (kg/m^2)	22.45±2.36	20.00±2.98	19.74±2.63	C&PI	0.000
				C&PII	0.000
				PI&PII	0.929

Significant:*P<0.05; **P<0.001; no Significant: P>0.05.

Anti-TTG levels showed a highly significant increase (P = 0.000) in the PI (11.12 ± 6.45 U/L) and PII (10.99 ± 6.16 U/L) groups as compared with the C (1.90 ± 0.95 U/L) group, while no significant difference was found between the PI and PII groups. In addition, BMI showed a highly significant decrease (P = 0.000) in PI (20.00 ± 2.98 kg/m^2) and PII (19.74 ± 2.63 kg/m^2) as compared with the C group (22.45 ± 2.36 kg/m^2**)**. Table 4 revealed a variation in liver function, especially in patients who adhered to GFD. The GGT activity showed no significant difference in PI (28.172 ± 10.388 U/L) and PII (20.756 ± 6.302 U/L) as compared with the C group (23.835 ± 7.069 U/L). While a highly significant increase in GGT activity (P = 0.004) was found between the PI and PII groups. In addition, the results in Table 4 showed no significant differences in the other parameters between the studied groups.

Table 4: The mean ±SD of the activity of GGT, ALT, AST, and the ratio of AST/ALT for the PI and PII groups compared to the C group

Parameters	Control (C) (Mean ± SD) (N = 51)	PI with GFD (Mean ± SD) (N = 29)	PII without GFD (Mean ± SD) (N = 23)	P-value	
GGT (U/L)	23.83±7.06	28.17±10.38	20.75±6.30	C&PI	0.056
				C&PII	0.280
				PI&PII	0.004
ALT (U/L)	17.37±4.50	19.45±8.78	16.59±6.57	C&PI	0.349
				C&PII	0.879
				PI&PII	0.252
AST (U/L)	15.47±7.18	15.89±8.78	12.96±6.48	C&PI	0.969
				C&PII	0.385
				PI&PII	0.349
AST/ALT	0.91±0.52	0.77± 0.09	0.74± 0.08	C&PI	0.258
				C&PII	0.181
				PI&PII	0.956

Significant:*P<0.05; **P<0.001; no Significant: P>0.05

Celiac disease has a worldwide incidence of 1%, but there are significant differences between countries [14]. In this disease, the ratio of identified to undiagnosed instances can reach 1:7 [15]. The correlation of clinical, serologic, and histological characteristics is required for the accurate identification of this disease. There are no CD monitoring standards or parameters to assess on a regular basis during follow-up. Anti-TTG titres, on the other hand, are presently used to assess adherence and reaction to a GFD and are typically examined annually in everyday clinical practice. It is well understood that adhering to a GFD will result in disease management for the vast majority of CD patients, lowering the risk of complications and death [16]. Because it is made in the small intestine, which is where gluten causes inflammation in people who are sensitive to it, the amount of anti-TTG IgA in the blood is a better way to diagnose a disease [17]. Some researchers suggest that if serology remains aberrant after one year of GFD, further studies should be conducted [18]. Several studies have found that anti-transglutaminase and/or anti-endomysium (TTG/EMA) positivity after the commencement of a GFD is more commonly linked with intestinal damage [19,20]. Furthermore, antibody negativity is not always linked with histological healing and may be associated with false-negative findings [21,22]. Although the duration of the disease is different, anti-TTG in the current study showed high levels in all the studied groups as compared to the C group, regardless of whether the patient is on a gluten-free diet or not. In addition, upon compression between P1 and PII, no significant difference in anti-TTG levels between the two groups was found, which means GFD does not affect the level of anti-TTG. Previous studies thought that the rise in non-classical CD could be because people are changing the way they eat and eating more processed foods, which exposes people to the gliadin antigen more [23]. The percentage of patients suffering from typical gastrointestinal symptoms, such as weight loss, has been declining in recent decades. More individuals are now identified in screening trials who have extra-intestinal symptoms or have no symptoms [24]. Contrary to popular perception, many adult individuals with celiac disease have a high or average body mass index (BMI) at the time of diagnosis [25,26]. According to some research, those who follow a gluten-free diet religiously have a higher BMI [27]. Other studies documenting BMI in celiac disease patients at diagnosis and/or after a gluten-free diet have produced contradictory findings. Few studies on BMI and other developmental

parameters in adolescents have been conducted, and the results of those studies have been unclear [27,28]. The results of our study are in disagreement with Diamanti *et al*. [27] and Ukkola *et al*. [29]. They all found that gluten-free diets raise BMI in celiac disease patients due to their high caloric fat and protein amounts. Although CD mainly affects the gut, the disorder's clinical symptoms are broad, affecting many extraintestinal systems and organs, including the liver [30]. Hepatic dysfunction in CD can be anything from elevated liver enzymes or nonspecific hepatitis to chronic liver disease [31]. There is mounting evidence that "cryptogenic-raised transaminases," a largely asymptomatic chronic liver abnormality, is commonly linked with untreated celiac disease in both children and adults [32]. Liver changes in CD patients have been documented since 1977 by Hagander *et al*., who discovered that 40% of adults with incipient CD had elevated serum transaminase activities, which reverted to normal in the vast majority of patients after a gluten-free diet (GFD) [33]. Pals, Myleus, and Norstrom [32] revealed that serum aminotransferase elevations have been observed in approximately one-third of paediatric CD cases [31]. In the present study, no significant difference was found in liver GOT and GPT in any group with or without a gluten-free diet. The present study disagrees with the studies reported in the literature [31-33], and this may be due to the different eating habits of Iraqi society or to the gluten-free food products in the commercial markets, most of which are not subject to health supervision, standardization, or quality control institutions. In addition, the significant increase in GGT activity in the PI group who adhered to a gluten-free diet indicates that patients may have an increased risk of liver damage. The study of multiple stages of patients with CD on GFD provides a clear image of the states of the liver and their impact on the diet of these patients [26]. According to the results, the GFD does not generally affect the liver, but over the long term, the diet may cause liver damage. The majority of research studies look at the frequency of hypertransaminasemia in CD or the relationship between CD and liver diseases, yet they do not look at other pathological alterations in recently identified CD patients [31]. The aetiology of hypertransaminasemia and liver injury in CD is still unknown. They are most likely caused by greater intestinal permeability and changes in gut flora, chronic inflammation of the intestines, and genetic susceptibility [32]. Within one year of therapy (GFD), the majority of paediatric and adult patients have normalized their liver tests [35,36]. If the transaminases normalize on the GFD, a yearly check-up is advised [37]. Another study found that in CD patients, the formation of autoimmune and cryptogenic liver abnormalities (having a favourable reaction to GFD) occurs. Typically, aminotransferase elevations are modest to intermediate, with a low AST/ALT ratio and a wide range of manifestation variations [38]. Other researchers have found that following the implementation of a GFD, liver functions improve. The process is likely complex, and gluten-induced damage to the gut mucosa may result in increased liver aminotransferases in celiac disease [39]. Serum aminotransferase increases will return to normal after gluten is removed from the diet, implying a link between gluten consumption, intestinal impact, and liver injury. Celiac disease causes greater intestinal permeability, which is caused by either inflammation or the stimulation of zonulin release, a tight junction regulator. Individuals with CD and hypertransaminasemia have significantly higher gut permeability than those with regular liver values. Toxins, antigens, and inflammatory chemicals (cytokines and/or autoantibodies) may enter the portal circulation more easily in the context of Celiac disease, and such mediators may play a role in the liver involvement observed in CD patients. Autoantibodies directed against the so-called celiac antigen (tissue transglutaminase) are found in the liver and other extra-intestinal tissues in Celiac disease, suggesting the hypothesis that humoral-mediated immune reactions play a pathogenic function in the liver injury seen in CD [40,41]. A correlation study was carried out between anti-TTG and the other parameters in the PI and PII groups to explore the relationship of GFD with these parameters in CD patients. The results in Table 5 showed that

there was no significant correlation between anti-TTG and the studied parameters in the study groups, whether they adhered to the GFD or not.

Table 5: Correlation values with anti-TTG in the PI and PII groups

Parameters	Anti-TTG					
	PI (N=29)			PII (N=23)		
	r	P	Sig.	r	P	Sig.
BMI	-0.086	0.658	NS	0.037	0.866	NS
GGT	-0.023	0.906	NS	0.250	0.249	NS
GPT	0.204	0.289	NS	0.293	0.175	NS
GOT	0.196	0.309	NS	0.279	0.198	NS
GOT/GPT	0.139	0.472	NS	0.274	0.206	NS

HS: $P < 0.01$, S: $P < 0.05$, NS: $P > 0.05$.

The limitation of this study is that a lot of CD patients develop fatty livers, hence abnormal liver enzymes, which can lead to a false conclusion, which was the most difficult challenge we faced.

4. Conclusion

It is concluded from the results of the current study that following a gluten-free diet did not aid in decreasing anti-TTG and that high activity of GGT may increase the risk of liver disease in CD patients who adhere to a gluten-free diet. We also advise CD patients to have periodic checks for liver functions to avoid liver damage. In addition, we recommended prospective studies for the determination of other hepatic parameters in celiac patients, and further studies on larger groups of patients are needed to explore what role GFD may play in the development of the metabolic disorder in CD patients.

Ethics clearance

The research ethical committee at scientific research has the ethical approval of environmental, health, higher education, and scientific research ministries in Iraq

Conflict of interest

The authors declare that they have no conflicts of interest.

Acknowledgement

The authors would like to express their gratitude to all the staff of the Hydrogen Medical Laboratory, Calcium Medial Laboratory, and Baghdad Teaching Hospital. Sincere thanks go to the patients and healthy people who supplied us with blood samples.

References

[1] R. S. Aboud, "The relationship between celiac disease and unexplained infertility", *Iraqi Journal Science*, vol. 57, no. 1, pp. 72-77, 2016.

[2] A. I. S. Al-Assaf, H. M. Ali, and A. H. Ad'hiah, "Gene expression of NLRP3 inflammasome in celiac disease of Iraqi children", *Ibn AL- Haitham Journal for Pure Applied and Science*, vol. 2021, pp. 15-22, 2021.

[3] K. Kwiatek-Średzińska, K. Kondej-Muszyńska, M. Uścinowicz, I. Werpachowska, M. Sobaniec-Łotowska, and D. Lebensztejn, "Liver pathology in children with newly diagnosed celiac disease", *Clinical Expermantal Hepatology*, vol. 5, no. 2, pp. 129-132, 2019.

[4] H. R. Hasan, D. A. White, and R. J. Mayer, "Extensive destruction of newly synthesized casein in mammary explants in organ culture", *Biochemistry Journal*, vol. 202, no. 1, pp. 133-138, 1982.

[5] I. D. Hill, "Guideline for the diagnosis and treatment of celiac disease in children: recommendations of the north american society for pediatric gastroenterology, hepatology and nutrition," *Journal of Pediatric Gastroenterology and Nutrition*, vol. 40, no. 1, pp. 1-19, 2005.

[6] G. Previtali, L. Licini, and A. Marsealia, "Celiac disease diagnosis without biopsy: is a 10 × ULN Antitransglutaminase result suitable for a chemiluminescence method?," *Journal of Pediatric Gastroenterology and Nutrition*, vol. 66, no. 4, pp. 645-650, 2018.

[7] J. A. Silvester, A. Therrien, and C. P. Kelly, "Celiac disease: fallacies and facts", *Am. Journal Gastroenterology*, vol. 116, no. 6, pp. 1148-1155, 2021.

[8] E. Abaas, "Relationship between interleukin-33(IL-33) and C-reactive protein in Iraqi women patients with celiac disease", *Ibn AL-Haitham Journal for Pure and Applied Science*, vol. 33, no. 1, pp. 288-296, 2014.

[9] J. A. Silvester, L. A. Graff, L. Rigaux, J. R. Walker, and D. R. Duerksen, "Symptomatic suspected gluten exposure is common among patients with coeliac disease on a gluten-free diet", *Alimentary Pharmacology and Therapeutics*, vol. 44, no. 6, pp. 612-619, 2016.

[10] J. P. Stefanolo, M. Talamo, S. Dodds, and A. Florencia "Real-world gluten exposure in patients with celiac disease on gluten-free diets, determined from gliadin immunogenic peptides in urine and fecal samples", *Clinical Gastroenterology and Hepatology* , vol. 19, no. 3, pp. 484-491, 2021.

[11] C. Anania, E. De Luca, G. De Castro, C. Chiesa, and L. Pacifico, "Liver involvement in pediatric celiac disease", *World Journal of Gastroenterology*, vol. 21, no. 19, pp. 5813-5822, 2015.

[12] T. U. Mohammed, J. H. Murtadha, I. Hashim, and A. Razzaq, "Measurement and purification of alanine aminotransferase (ALT) enzyme activity in patients with celiac disease", *Baghdad Science Journal*, vol. 14, no. 3, pp. 557-563, 2017.

[13] R. L. Wolf., B. Lebwohi, A. R. Lee, and P. Zybert, "Hypervigilance to a gluten-free diet and decreased quality of life in teenagers and adults with celiac disease", *Digestive Diseases and Science*, vol. 63, no. 6, pp. 1438-1448, 2018.

[14] L. Zhang, Y. Lv, A. Xu, and H. Wang, "The prognostic significance of serum gamma-glutamyltransferase levels and AST/ALT in primary hepatic carcinoma", *BMC cancer*, vol. 19, Article no. 841, 2019.

[15] B. R. Devrajani, A. Rahman, and S. Zulfiquar, "Serum trace metals and enzyme activity in patients with hepatic encephalopathy", *World Application and Science Journal*, vol. 16, no. 8, pp. 1053-1059, 2012.

[16] E. Lionetti and C. Catassi, "New clues in celiac disease epidemiology, pathogenesis, clinical manifestations, and treatment", *International Reviews of Immunology*, vol. 30, no. 4, pp. 219-231, 2011.

[17] A. Tommasini, T. Not, V. Kiran, and V. Baldas, "Mass screening for coeliac disease using antihuman transglutaminase antibody assay", *Archives disease of Childhood.*, vol. 89, no. 6, pp. 512-515, 2004.

[18] E. Farina, L. Roncoroni, V. Lombardo, A. Scricciolo, "Clinical value of tissue transglutaminase antibodies in celiac patients over a long term follow-up", *Nutrients*, vol. 13, no. 9, Article no. 3057, 2021.

[19] M. A. Lodhi, Z. Saleem, A. Ayub, T. Munir, and S. Hassan, "Diagnostic accuracy of IgA anti-tissue transglutaminase antibody in the diagnosis of celiac disease taking histopathology as gold standard", *Pakistan Armed Forces Medical Journal*, vol. 71, no. 2, pp. 418-421, 2021.

[20] N. I. A. Haddad, E. Nori, and S. H. Ali, "The effect of type two diabetes mellitus on superoxide dismutase (SOD) activity and its correlation with HbA1c in Iraqi patients", *International Journal Engeenering Research and Science*, vol. 2, no. 4, pp. 7-15, 2016.

[21] C. L. Jansson-Knodell, J. A. Murray, and A. Rubio-Tapia, "Management of small bowel villous atrophy in patients seronegative for celiac disease", *The American Journal of Gastroenterology*, vol. 115, no. 4, pp. 492-497, 2020.

[22] B. Lebwohl, D. S. Sanders, and P. H. R. Green, "Coeliac disease," *Lancet*, vol. 391, no. 10115, pp. 70-81, 2018.

[23] A. Sadeghi, N. Rad, S. Ashtari, and M. Rostami-Nejad, "The value of a biopsy in celiac disease follow up: Assessment of the small bowel after 6 and 24 months treatment with a gluten free diet", *Revista Espanola Enfermedades Digestive*, vol. 112, no. 2, pp. 101-108, 2020.

[24] M. Nikzamir, M.R. Nejad. A. Sadeghi, and A. Moradi, "The correlation between endoscopy manifestations and pathology outcome for diagnosis of celiac disease", *International Journal of Celiac Disease*, vol. 5, no. 3, pp. 101-103, 2017.

[25] H. J. Freeman, "Evaluation of non-dietary alternatives for treatment of adults with celiac disease", *Frontiers in Nutation Journal*, vol. 7, Article no. 562503, 2020.

[26] L. Elli, F. Ferretti, S. Orlando, and M. Vecchi, "Management of celiac disease in daily clinical practice", *European Journal of Internal Medecine*, vol. 61, pp. 15-24, 2019.

[27] A. Diamanti, T. Capriati, M. S. Basso, and F. Panetta, "Celiac disease and overweight in children: An update", *Nutrients*, vol. 6, no. 1, pp. 207-220, 2014.

[28] O. Olén, S. M. Montgomery, C. Marcus, A. Ekbom, and J. F. Ludvigsson, "Coeliac disease and body mass index: a study of two Swedish general population-based registers", *Scandinavian Journal of Gastroenterology*, vol. 44, no. 10, pp. 1198-1206, 2009.

[29] A. Ukkola, M. Maki, K. Kurppa, P. Collin, and H. Huhtala, "Changes in body mass index on a gluten-free diet in coeliac disease: a nationwide study", *European Journal of Internal Medecine*, vol. 23, no. 4, pp. 384-388, 2012.

[30] T. A. Kabbani, A. Goldberg, and C. P. Kelly, "Body mass index and the risk of obesity in coeliac disease treated with the gluten-free diet", *Alimentary Pharmacology and Therapeutics.*, vol. 35, no. 6, pp. 723-729, 2012.

[31] E. J. Hoffenberg, L. M. Emery, and K. J. Barrigi, "Clinical features of children with screening-identified evidence of celiac disease", *Pediatrics*, vol. 113, no. 5, pp. 1254-1259, 2004.

[32] M. van der Pals, A. Myleus, and F. Norstrom, "Body mass index is not a reliable tool in predicting celiac disease in children", *BMC Pediatrics*, vol. 14, Article no. 165, 2014.

[33] H. Jericho, N. Sansotta, and S. Guandalini, "Extraintestinal manifestations of celiac disease: effectiveness of the gluten-free diet", *Journal of Pediatrics and Gastroenterology Nutret*, vol. 65, no. 1, pp. 75-79, 2017.

[34] F. Tovoli, G. Negrini, R. Fari, "Increased risk of nonalcoholic fatty liver disease in patients with coeliac disease on a gluten-free diet: beyond traditional metabolic factors", *Alimentenary Pharmacology and Therapeutics*, vol. 48, no. 5, pp. 538-546, 2018.

[35] K. A. Kwiatek-średzińska, K. Kondej-muszyńska, M. Uścinowicz, I. Werpachowska, M. Sobaniec-łotowska, and D. Lebensztejn, "Liver pathology in children with newly diagnosed celiac disease", *Clinical and Experemantel Hepatology*, vol. 5, no. 2, pp. 129-132, 2019.

[36] O. I. Saadah, A. Khayat, and O. Abusharifah, "Liver function changes following the introduction of a gluten-free diet in patients with celiac disease", *Clinical and Experemantal Hepatology*, vol. 7, no. 4, pp. 415-421, 2021.

[37] G. J. Lee, B. Boyle, T. Ediger, and I. Hill, "Hypertransaminasemia in newly diagnosed pediatric patients with celiac disease", *Journal of Pediatric Gastroenterology and Nutrition*, vol. 63, no. 3, pp. 340-343, 2016.

[38] M. Aggarwal, R. Garg, P. Kumar, J. W. Fleming, and C. J. Knodell, "Bi-directional Relationship Between Celiac Disease and Liver Chemistries: A Systematic Review and Meta-Analysis", *Digestive Diseases and Science*, vol. 68, no. 4, pp. 1369-1380, 2022.

[39] A. Jena, P. Kumar-M, A. Kumar, C. Brida, A. Choudhury, N. Kumar, D. Rami, A. Facciorusso, and J. Samanta, "Liver abnormalities in celiac disease and response to gluten free diet: A systematic review and meta-analysis", *Journal Gastroenterology and Hepatology*, vol. 38, no. 1, p. 11-22, 2022.

[40] J. V. Kim and G. Y. Wu, "Celiac disease and elevated liver enzymes: A review", *Journal of Clinical and Translational Hepatology*, vol. 9, no. 1, pp. 116-124, 2021.

[41] R. H. Nafal, H. S. Al-Warid, and H. J. Al-Sultan, "Seroprevalence of toxoplasmosis in patients with chronic liver disease in Baghdad", *Iraqi Journal of Science*, vol. 60, no. 8, pp. 1667-1672, 2019.