CORRELATION OF VILLOUS ALTERATIONS IN CELIAC DISEASE PEDIATRIC PATIENTS WITH RISK FACTORS ANALYZE

Oana Belei[1], Ioan Simedrea[1], Monica Marazan[1], Tamara Marcovici[1], Camelia Daescu[1], Rodica Ilie[2], Florina Antonie[1], Georgiana Brad[1]
[1]First Pediatric Clinic, University of Medicine and Pharmacy "Victor Babes" Timisoara
[2]Pathology Department, Emergency Children's Hospital "Louis Turcanu" Timisoara

Abstract

Objectives: There are many studies concerning the risk factors involved in celiac disease ethiopathogeny. The aim of this research was to establish a correlation between presence of several risk factors and villous alteration severity by introducing a risk score. *Material and methods:* The present study was performed on a group of 25 pediatric patients with celiac disease diagnosed between September 2005 and November 2008; celiac disease severity was classified using Marsh criteria. Five celiac risk factors have been analyzed: gluten administration before age of 5 month in artificially nourished infants, presence of first and/ or second degree relatives diagnosed with celiac disease, presence of several autoimmune conditions (type I mellitus diabetes, autoimmune thyroiditis, rheumatoid arthritis, polyendocrine autoimmune conditions), Down syndrome and viral infections in patient's medical history (adeno, herpes or rubella virus). Odds ratio (OR) and relative risk (RR) have been calculated for each of them using an original formula, and the risk score was computed for each patient. The calculated score was compared with the intestinal morphological result. *Results:* We found a strong correlation between the computed score and the villous alteration's degree (r=0, 94). Finally, we estimated the score parameters: sensitivity, specificity and positive predictive value, which validated our score. *Conclusions:* We consider very useful an assessment of risk differentiation in celiac patients with positive serology, knowing that the majority of gluten enteropathy subjects present the latent form of illness, without typical symptoms, according to celiac ice-berg described in 1991.

Key words: celiac disease, villous atrophy, antiendomisium antibodies, antitransglutaminase antibodies

Introduction

Celiac disease is an immune-mediated enteropathy caused by a permanent sensitivity to gluten in genetically susceptible individuals (DQ2 and/or DQ8 HLA haplotype). It occurs in symptomatic subjects with gastrointestinal and non-gastrointestinal symptoms, and in some asymptomatic individuals, including subjects affected by: type I diabetes, Turner syndrome, Williams syndrome, IgA deficiency and first degree relatives of individuals with celiac disease (1). Recent studies incriminates as potential factors involved in celiac disease pathogeny: viral infection with adenovirus (based on structural similarity of 206 – 217 amino-acids gliadine sequence and E1b protein elaborated during infection), rubella, herpes virus infection and/or Plasmodium Yoelli parasite infection (2).

Objectives

In this study we proposed to establish several risk factors contribution in celiac disease appearance and disease's manifestation form. It is well-known that classic risk factors (early gluten exposure – under 5 month of life in formulas fed infants, first degree relatives with celiac disease, several infectious factors, autoimmune or genetic diseases association) , play an important role in celiac disease appearance. The aim of this research was to establish a relation between presence of five risk factors and the severity of intestinal morphological alteration by introducing a risk score. We also assessed this score by establishing its specificity, sensibility and accuracy.

Material and Methods

The study developed between September 2005 and November 2008. The lot of study consisted in 25 patients, aged 7 month – 18 years, sex ratio G/B 16/9. Before including each patient in the lot of study we obtained written informed consent from their parents. IgA selective deficiency was considered exclusion criteria. Celiac disease diagnosis was based on clinical and biological assessment of malabsorption syndrome, positive serological tests and small intestinal biopsy followed by histological exam of biopsy sample. Serological tests consisted in assessment of IgA anti endomysial antibodies (EMA) using indirecte immunofluorescence technique on smooth muscle of monkey esophagus. ImmuGlo^{TM}Anti-Endomysial Antibody test kits were provided by Immco Diagnostics. Assessment of IgA human tissue transglutaminase antibodies (hu-tTG) was made using ELISA technique. ImmuLisa^{TM} anti-hu tTG antibody IgA ELISA kits were provided also by Immco Diagnostics. For intestinal biopsy we used Watson Capsule for children aged less than 6 years old and superior digestive endoscopy followed by controlled biopsy taken in patients aged more than 6 years old.

The 5 risk factors that have been present before celiac disease in the lot of study are represented by: 1.gluten

administration before age of 5 month in artificially nourished infants; 2. presence of first and/or second degree relatives diagnosed with celiac disease; 3. several autoimmune conditions (type I mellitus diabetes, autoimmune thyroiditis, rheumatoid arthritis, polyendocrine autoimmune conditions); 4. Down syndrome and 5. adeno/herpes or rubella virus infection in patient's medical history.

Odds Ratio (OR) and relative risk (RR) have been calculated for each of these risk factors, according to statistical formulas for transversal studies (3). Using an original formula, the risk score was computed for each patient:

S = £ ORi x FRi (ORi = Odds Ratio for each of 5 risk factors, FRi symbolic represented by values of 0/1, indicates risk factor presence or absence and £ = sum).

The calculated score was compared with intestinal morphological result. Finally, we estimated the score parameters: sensitivity, specificity, positive predictive value and accuracy.

Statistic data processing was made by using SPSS12 application.

The lot of study was divided based on intestinal villous alteration degree, using Marsh classification (1992), modified by Oberhuber (1997) as following: type II hyperplastic (infiltrative lympho-plasmocitic lesions in villous corion, associated by glandular crypt enlargement) and type III destructive (including partial and subtotal villous atrophy – type
IIIa, IIIb respectively and total villous atrophy – type IIIc). (4)

Results and discussions

Distribution of villous alteration type in studied lot indicated 36% of patients with hyperplastic villous lesions (Marsh II type) and 64% of patients presented different degree of villous atrophy (Marsh III type).

Lot characteristics are shown in table I.

Table I - Lot characteristics.

	Risk factors					Type II Marsh	Type III Marsh
No of patients	**Early gluten exposure < 5 month**	**Ist and/or IInd degree relatives**	**Autoimmune conditions**	**Down syndrome**	**Adeno/ herpes/ rubella virus**		
25	**17**	**16**	**15**	**4**	**13**	**9 36%**	**16 64%**

Risk factors weight can be assessed by calculating OR and RR.

OR represents a quotes ratio - probability of having an exposure to certain risk factors in subjects that are now presenting un effect (celiac disease in our case) reported to probability of having the same exposure in subjects that are not presenting un effect now. (3)

RR represents a risk ratio – risk of certain effect appearance (celiac disease in our case) in subjects who have been exposed to a risk factor, reported to the risk of the same effect appearance in subject not exposed to the same risk factor. (3)

OR and RR hierarchy for analyzed risk factors are showed in table II.

Table II – OR and RR for celiac disease's risk factors.

Risk factor	**Early gluten exposure <5 month**	**Down syndrome**	**Autoimmune conditions**	**Adeno/ Herpes/ rubella virus**	**Ist and/or IInd degree relatives**
OR	**2.70**	**2.43**	**2.05**	**1.39**	**1.29**
RR	**1.46**	**1.29**	**1.30**	**1.12**	**1.09**

These results indicate values grater than 1. That confirms the hypothesis that all analyzed factors represent risk factors for celiac disease. OR value for early gluten administration before 5 month old in formula fed infants, Down syndrome and autoimmune conditions association are greater than 2. The results indicate that these 3 factors have a greater weight in influencing the progression of celiac disease to more severe forms of villous injury.

We calculated the risk score values for each patient using the formula mentioned above (S = £ ORi x Fri). Then, we statistically analyzed the medium score value for the lot of patients with Marsh II – hyperplastic villous injury, for the lot of patients with Marsh IIIa and Marsh IIIb together - partial and subtotal villous atrophy - and finally, for the lot of patients with total villous atrophy - Marsh IIIc type.

Processing data with Spearman correlation index for each three values of score averages classified based on villous alteration type, we obtained the value R=0,943. The result indicated a strong correlation and validated the proposed score.

Table III - Risk score values.

Marsh classification of villous alterations		Hyperplastic (type II) N = 9 patients	Partial/subtotal villous atrophy (type IIIa, IIIb) N = 7 patients	Total villous atrophy (type IIIc) N = 9 patients
Score	Average	4.39	5.44	5.70
	SD	1.94	2.39	2.22

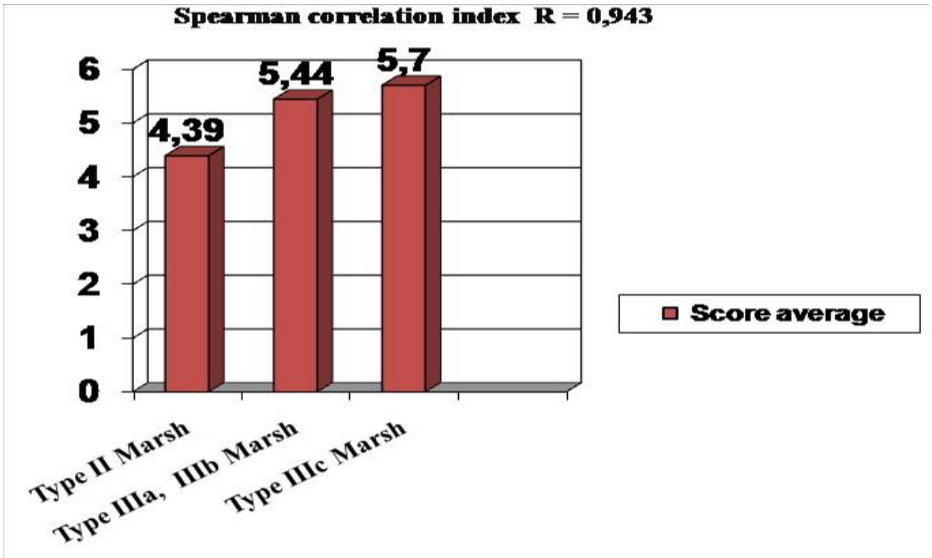

Figure 1 - Score average for different Marsh degree villous alteration.

In order to statistically compare the score averages of type II Marsh lot (N=9 patients) and type III Marsh lot (N=16 patients), we used Wilcoxon test and we obtained the value p=0, 05, indicating the presence of a statistically significant difference.

Classificators quality assessment is realized by sensibility and specificity analyze. So, we assessed sensibility and specificity variation for discrimination limit values between classes in accordance with quartiles between average score of type II Marsh lesions group (4,387) and type III Marsh lot (5,579). Quantitative index values varying with limit values between classes (quartiles) are showed in table IV.

Although the specificity value is not high enough, the sensitivity of the proposed score has a sufficiently high value in order to recommend this score as a useful tool for assessment of celiac disease risk level in certain subjects.

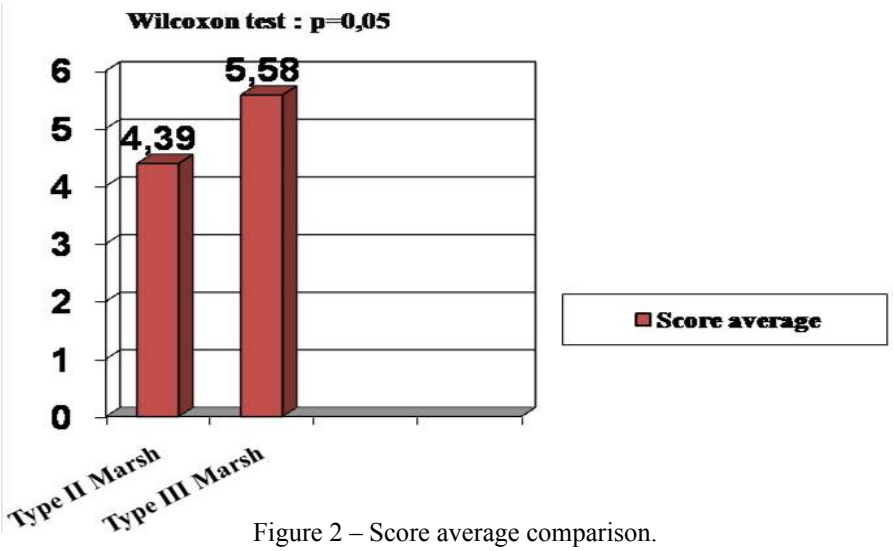

Figure 2 – Score average comparison.

Table IV - Quantitative index of risk score.

Limit	4.387	4.685	4.983	5.281	5.579
Sensibility	0.694	0.690	0.611	0.611	0.528
Specificity	0.474	0.474	0.632	0.684	0.737
Positive predictive value	0.714	0.714	0.759	0.786	0.792
Negative predictive value	0.450	0.451	0.462	0.481	0.452
Accuracy	0.618	0.618	0.618	0.636	0.600

Conclusions

A scoring scale awards the advantage of having a synthetic perspective upon patients possible evolution. Medical literature offers few atempts of computing celiac disease appearance score in accordance with risk factors exposure. Comparing our results with other statistic studies based on celiac disease risk factors identification in genetically susceptibile individuals (DQ2 or DQ8 HLA haplotype), we found a strong concordance (5), (6). Our results emphasize the necessity of serological screening (EMA, hu-tTG antibodies) in certain patients groups (autoimmune conditions, genetical diseases, viral infection) as well as in subjects with first/second degree relatives diagnosed with celiac disease (7). After serological screening, histological confirmation of gluten enteropathy in early stage is indicated, followed by gluten exclusion in order to stop the illness progression to its most feared complication – intestinal lymphoma. We consider very useful a risk factors hierarchy in patients with positive serologic tests, based on the fact that most celiac patients present silent or atypical form of diseases, in accordance with celiac ice-berg described by Richard Logan in 1991. (8)

References

1. Darren Craig; Gerry Robins; Peter D. Howdle, Advances in celiac disease, Curr. Opin. Gastroenterol. 2007;23:142-148
2. Bach J., Infections and autoimmune diseases, J. Autoimmun. 2005;25 Suppl:74 – 80
3. Ramesh C. Gupta, Suzhong Tian, Statistical inference for the risk ratio in 2×2 binomial trials with structural zero, Computational Statistics & Data Analysis, March 2007;Vol 51, Issue 6,1:3070-3084
4. Carlo Catassi; Alessio Fasano, Celiac Disease, Curr Opin Gastroenterol. 2008; 24(6): 687-691

5. Grzegorz Telega, Celiac Disease Patients Now Typically Asymptomatic at Diagnosis, Arch Pediatr Adolesc Med 2008;162:164-168
6. Evelyn Artz, Julia Warren-Ulanch, Dorothy Beckeret al, Seropositivity to celiac antigens in asymptomatic children with type 1 diabetes mellitus: association with weight, height and bone mineralization, Pediatric Diabetes 2008; 9 (Part I):277-284
7. Jeremy Woodward, Celiac disease, Medicine, Apr 2007; Vol 35, Issue 4:226-230
8. Catassi C, Kryszak D, Louis-Jacques O, et al. Detection of Celiac disease in primary care: a multicenter case-finding study in North America, Am J Gastroenterol. Jul 2007; 102(7):1454-60.

Correspondence to:
Dr. Oana Belei
Heinrich Heine Street no 4, ap. 17,
code 300041, Timisoara
Telephone number: 0040723289480
Fax: 0256201976
E-mail: oana22_99@yahoo.com