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Reticulin antibodies in coeliac disease

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The diagnostic value of demonstrating reticulin antibodies in children affected by coeliac disease is discussed. The antibodies were shown by immunofluorescence in 201 serum samples of 82 patients during the initial phase, during gluten provocation after several months of gluten-free diet. Demonstration of reticulin antibodies is not useful in screening for coeliac disease in patients suspect of the condition since both false positive and negative results occur in spite of the high sensitivity of the test. Determination of the antibodies is, however, a useful guide in dietary control. The antibodies discriminate coeliac disease from cow's milk protein intolerance.

The jejunal mucosa fulfils two principal tasks, viz. absorption of nutrients and defence against macromolecules and microorganisms. Normally, products of intraluminal digestion are either absorbed, sometimes after further cleavage occurring in the microvilli, or repelled by the jejunal mucosal barrier. Therefore, any mucosal damage may be accompanied by absorption of nutritional antigens and production of antibodies against them. In children, cow's milk protein, gliadin and reticulin are the most frequent nutritional antigens. Governa et al. [5] have shown that reticulin antibodies are directed more against nutritional reticulin rather than the reticulin of the own body. The most frequent cause of severe jejunal mucosal damage of children is coeliac disease.

Since the appearance, course and disappearance of antibodies against nutritional antigens depend much on the condition of the jejunal mucosa, it may be anticipated that determination of these antibodies might be helpful in controlling the disorder by diet. In previous studies [6, 17] we could show that this was particularly true for reticulin antibodies. Production of antibodies against nutritional antigens in coeliac disease is an epiphenomenon, it has thus nothing to do with the ultimate pathogenesis of the disorder.

In this paper we report on the results obtained in a large number of patients.

MATERIALS AND METHODS

Reticulin antibodies were determined in 201 serum samples taken from 82 children aged 7 months to 17 years, in various phases of coeliac disease (Tables I, II).

The diagnosis of coeliac disease was based upon the clinical and jejunal biopsy criteria of the European Society for Paediatric Gastroenterology and Nutrition [8], i.e. a typical history, laboratory findings and a subtotal villous atrophy in the jejunal biopsy specimen. Serial jejunal biopsies

TABLE I

Number of patients and sera examined for reticulin antibodies

	No. of patients	No. of sera
Children with coeliac disease	82	201
Children with acute or chronic intestinal disease	96	96
Children without intestinal disease	32	32

TABLE II

Reticulin antibody titres in children with coeliac disease under various nutritional conditions

Titre	Group				
	I n=31	II n=143	III n=27	IV n=96	V n=32
1280	2	—	—	—	—
640	1	—	1	—	—
320	10	—	—	—	—
160	2	—	1	—	—
80	4	—	4	—	—
40	2	4	12	3	—
20	7	6	3	4	—
10	—	6	2	4	—
less than 10	3	127	4	85	32
Percentage of positive sera	90.3	11.2	85.2	11.5	0
Percentage of negative sera	9.7	88.8	14.8	88.5	100

Group I: no diet (initial phase)
Group II: at least three months gluten-free diet
Group III: at least three months gluten provocation
Group IV: controls with aspecific intestinal symptoms
Group V: controls without intestinal disease

were not considered necessary for the initial diagnosis. For histological stereomicroscopic classification Shmerling's typing [13] was used: Type I (normal), Type II (partial villous atrophy), Type III (subtotal or total villous atrophy).

The control group consisted of 96 children aged 5 months to 14 years, admitted for acute or chronic intestinal disease, in whom coeliac disease was excluded by the subsequent course of the disease. In some of these patients jejunal biopsy was also done; normal or slightly altered jejunal mucosa was the finding in each case. Only 5 children with confirmed cow's milk protein intolerance exhibited moder-

ate or severe mucosal changes of Type II or III. Another control group comprised 32 children aged 6 months to 12 years: with no intestinal symptoms or complaints.

Sera were deep-frozen immediately after taking the blood sample and kept at −20 °C until determination. The person performing the serological work was not aware of the diagnosis.

For reticulin antibody determination frozen sections of guinea-pig kidneys were incubated with the sera diluted 1:10 and stained with FITC labelled goat antihuman globulin produced by the State Institute for Immune Preparations and Culture Media, Berlin. The preparations were then examined in a microscope Fluoval I, of Carl Zeiss, Jena, GDR. Sera exhibiting positive fluorescence in the tubular basal membrane and Bowman's capsule were subsequently titrated.

Results

Results are shown in Table II. Reticulin antibodies could be demonstrated in 90.3% of children with recent, untreated coeliac disease while a positive test was obtained in 11.2% of children affected by coeliac disease and treated for at least three months. In children in whom after at least three months gluten provocation was performed, positivity was again as high as 85.2%. Reticulin antibodies were present in 11.2% of children affected by aspecific intestinal diseases, the highest titre was 1:40. In 6 out of the children with a positive test for reticulin antibody a jejunal biopsy was carried out; in two children the histology was normal, two had moderate cellular infiltration accompanying normal villous findings and again two exhibited partial villous atrophy.

In none of the 5 children with proven cow's milk protein intolerance was a reticulin antibody demonstrable although the blood samples were obtained in the acute phase of the disease and jejunal changes of Type II or III were seen in all the 5 patients.

In the healthy children without any intestinal symptom or complaint no reticulin antibodies were found.

In a number of sera gluten antibodies were determined for comparison of the sensitivity and specificity of the two tests; the results will be published elsewhere.

Discussion

Prevalence of reticulin antibodies in coeliac disease is between 33 to 93% [2, 4, 6, 10, 12]. The frequency is higher in children than in adults. In two earlier studies we observed a frequency of 70 and 79%, respectively, in children affected by coeliac disease and exhibiting total villous atrophy [6, 17]. The higher percentage (90.3%) of positive findings in the present material was due to the fact that in the previous studies the titres were correlated to the actual type of mucosal alteration and e.g. in patients with Type III no difference was made whether they had florid coeliac disease or were provoked with gluten after a gluten-free diet. The technique was also more adequate in the present study.

In the course of gluten-free dietary treatment the antibodies disappear or at least show a marked reduction in titre; after at least three months

of treatment only 11.2% of the sera were still positive. The persisting positivity can be explained by the observation that in some cases disappearance ensues only after four months or more. Of course, imperfect adherence to the diet could not be excluded, either.

On the basis of our findings it can be stated that determination of reticulin antibodies is a useful indicator of the quality of treatment. In spite of the fairly high grade of specificity, i.e. only 11.5% of the sera obtained from children affected by non-coeliac intestinal diseases and none of the sera of children having no intestinal symptoms at all were positive for reticulin antibodies, the reticulin antibodies cannot be used for screening cases suspect of coeliac disease in order to find out in which patient should a jejunal biopsy be carried out: there are both false positive and false negative results. In addition, in suspect cases jejunal biopsy cannot be avoided since it may reveal other causes of malabsorption in addition to coeliac disease; thus, the test cannot be replaced by anything in the diagnostics of malabsorption.

In family studies, a search for reticulin antibody positive members may be useful: by this method symptom-free candidates for jejunal biopsy can be selected [7, 14, 15, 16]. It is also noteworthy that reticulin antibodies are present in a fairly high percentage of sera taken from patients affected by other disorders like dermatitis herpetiformis [4, 9, 11], rheumatoid arthritis [9], Crohn's disease [1, 4], Sjögren's syndrome [9] or heroin addiction [9]. However, up to now no case afflicted by cow's milk protein intolerance with demonstrable reticulin antibodies has been described [3, 12, 17]; also in our own 5 patients with this condition no antibodies could be demonstrated although the blood samples were taken during the acute phase of the condition.

References

1. Alp MH, Wright R: Autoantibodies to reticulin in patients with idiopathic steatorrhoea, coeliac disease, and Crohn's disease, and their relation to immunoglobulins and dietary antibodies. Lancet 2:682, 1971
2. Bardare M, Villani R, Giunta A: Anti-reticulin antibodies in malabsorption syndromes. Helv Paediatr Acta 29:203, 1974
3. Essen R von, Savilahti E, Peltonen P: Reticulin antibody in children with malabsorption. Lancet 1:1157, 1972
4. Etermann KP, Feltkamp TEW: Antibodies to gluten and reticulin in gastrointestinal diseases. Clin Exp Immunol 31:92, 1978
5. Governa M, Franchini I, Cella GD, Durand P: Serum anti-reticulin antibody as further evidence of the increased immune response to dietary antigens in coeliac children. Helv Paediatr Acta 28:267, 1973
6. Henker J, Zugehör M, Morenz J: Retikulin-Antikörper bei kindlicher Zöliakie. Dtsch Gesundh Wes 34:83, 1979
7. Mallas EG, Williamson GN, Cooper BT, Cooker WT: IgA class reticulin antibodies in relatives of patients with coeliac disease. Gut 18:647, 1977
8. McNeish AS, Harms HK, Rey J. Shmerling DH, Visakorpi JK, Walker-Smith JA: The diagnosis of coeliac disease. A commentary of the current practices of members of the European Society for Paediatric Gastroenterology and Nutrition (ESPGAN). Arch Dis Child 54:783, 1979
9. Rizzetto M, Doniach D: Types of "reticulin" antibodies detected in human sera by immunofluorescence. J Clin Path 26:841, 1973

10. Rotthauwe HW, Sennekamp J, Emons D, Becker M: Antiretikulin-Antikörper und präzipitierende Antikörper gegen Nahrungsmittelprotein im Serum von Kindern mit Cöliakie. Eur J Pediatr 121:215, 1976
11. Seah PP, Fry L, Hoffbrand AV, Holborow EJ: Tissue antibodies in dermatitis herpetiformis and adult coeliac disease. Lancet 1:834, 1971
12. Seah PP, Fry L, Holborow EJ, Rossiter MA, Doe WF, Magelhaes AF, Hoffbrand AV: Antireticulin antibody: Incidence and diagnostic significance. Gut 14:311, 1973
13. Shmerling DH: Peroral intestinal mucosal biopsies in infants and children. Helv Paediatr Acta Suppl 22:1, 1970
14. Stern MS, Bender SW, Grüttner R, Posselt HG, Strobel S: Serum antibodies against gliadin and reticulin in a family study of coeliac disease. Eur J Pediatr 135:31, 1980
15. Stevens FM, Lloyd R, Egan-Mitchell B, Mylotte MJ, Fotrell PF, Wright R, McNicholl B, MacCarthy CF: Reticulin antibodies in patients with coeliac disease and their relatives. Gut 16:598, 1975
16. Stokes PL, Ferguson R, Holmes GKT, Cooke WT: Familial aspects of coeliac disease. Quart J Med 45:567, 1976
17. Zugehör M, Bannert N, Morrenz J: Immunpathogenese und Immundiagnostik der Zöliakie im Kindesalter. Kinderärztl Prax 46:1, 1978

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