When is a coeliac a coeliac?
Report of a working group of the United European Gastroenterology Week in Amsterdam, 2001

It should be recognized that these guidelines should not be deemed inclusive of all proper methods of care or exclusive of methods of care reasonably directed to obtaining the same results. The ultimate judgement regarding the propriety of any specific guideline must be made by the physician in light of all the circumstances presented by the individual patient. *Eur J Gastroenterol Hepatol* 13:1123–1128 © 2001 Lippincott Williams & Wilkins

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**Introduction**

It is now more than 30 years since the diagnostic criteria for coeliac disease were proposed at the Interlaken Meeting of the European Society of Paediatric Gastroenterology and Nutrition (ESPGAN) in 1969 [1]. These criteria were further enunciated at the Second International Symposium on Coeliac Disease in 1974, and are as follows: (1) structurally abnormal jejunal mucosa when taking a diet containing gluten; (2) clear improvement of villous structure when taking a gluten-free diet (GFD); (3) deterioration of the mucosa during gluten challenge [2].

The ‘Interlaken criteria’ were reviewed by ESPGAN in 1990 [3]. It was stated that the ESPGAN criteria might not invariably be required for diagnosis of all coeliacs, especially the deterioration of the mucosa during gluten challenge. Challenge was no longer required except for children under 2 years of age. However, the criteria for the diagnosis of coeliac disease in adults have not been discussed or defined by the major World Societies for Gastroenterology. In September 2000, the Dutch Coeliac Society celebrated its 25th anniversary and organized a workshop in Noordwijkerhout to prepare guidelines on coeliac disease, in anticipation of the 2001 United European Gastroenterology Week (UEGW) in Amsterdam.

Coeliac disease is an intestinal disorder with a multifactorial aetiology. Human leucocyte antigen (HLA) and probably non-HLA genes, together with gluten and possibly additional environmental factors, are involved in the disease development. T cells are central in controlling an immune response to gluten that causes the immunopathology. The actual mechanisms responsible for the tissue damage are as yet only partly understood.

The interest in the pathophysiology, pathology and clinical features of coeliac disease has expanded rapidly in the last 10 years. As our understanding of gluten-sensitive enteropathy deepens, old assumptions are being questioned.

The ESPGAN criteria may need discussion and revision, while the lack of consensus on adult coeliac disease calls out for the attention of the United European and American Gastroenterology Associations.

**Diagnosis of coeliac disease**

During the last decade, better knowledge of coeliac disease has permitted the identification not only of the typical forms, characterized by gastrointestinal symptoms, but also the atypical forms. Coeliac disease may present with non-specific features and may in general be a subclinical condition. A considerable number of coeliacs do not show demonstrable clinical or functional alterations while consuming gluten, despite significant histological abnormalities [4].

The golden standard for diagnosis is the small-bowel biopsy. In addition to the classic criteria for the evaluation of the histological features of coeliac disease, an alternative terminology has been proposed by Marsh to describe the different patterns of mucosal changes in the variable spectrum of gluten sensitivity [4]. Historically, the typical histological lesion of a coeliac patient eating gluten is a total or subtotal villous atrophy with elongated and hyperplastic crypts and a chronic infiltration of lymphocytes. This destructive lesion was thought to be typical of coeliac disease.

The Marsh classification describes a spectrum of consecutive mucosal abnormalities that can be seen in gluten sensitivity. Type I comprises normal mucosal
architecture with a marked infiltration of villous epithelium by lymphocytes – arbitrarily, more than 30 lymphocytes per 100 enterocytes. Type II includes intraepithelial lymphocytosis and also enlargement of crypts in which there is an increased mitotic rate. In interpreting advanced mucosal lesions with destruction of villi (type III), a modification of the original Marsh classification has been introduced [5]. Three distinct stages of villous atrophy have been recognized. In type IIIA, partial villous atrophy, the villi are blunt and shortened, accompanied by lymphocytic infiltration of the epithelium and crypt hyperplasia. Arbitrarily, partial villous atrophy can be diagnosed if the villous–crypt ratio is below 1:1. In type IIIB lesions, sub-total villous atrophy, the mucosa is rather hypertrophic but separate villi are still recognizable. Crypts are enlarged and show strongly increased mitotic rates and intraepithelial lymphocytosis is present. Finally, in type IIIC, total villous atrophy, the mucosa resembles colon mucosa – villi are absent or rudimentary, again alongside crypt hyperplasia, and there is heavy lymphocytosis. Marsh also suggested a class IV lesion, characterized by total villous atrophy with no significant inflammation and a lack of cryptohyperplasia.

Traditionally, clear histological changes in the small-bowel biopsy, such as Marsh type III, had to be found before diagnosis of coeliac disease could be made for the prescription of a GFD. However, future research should elucidate which categories of patients with Marsh types I–II must be included in the diagnosis of coeliac disease.

Normalization of biopsies
Children who adhere to a GFD generally show a normalization of their villous structure. Intraepithelial lymphocytosis in adults, especially γ/δ T-cell receptor bearing cells, may persist for years [6,7]. There have been some suggestions that small-bowel histology in children on a GFD will normalize in a shorter period than in adults on a GFD [8]. Coeliac disease is confirmed when symptoms (if any), previous serology (if abnormal) and small-bowel histology improve in general within 1 year of a GFD. However, symptoms generally improve within a few weeks.

Where to take the biopsy
Originally, the small intestinal mucosal specimen was obtained using the Crosby capsule. With the advent of flexible endoscopes for both adults and children, multiple sampling obtained with forceps during an upper endoscopy became routine. Three to four biopsies from the distal duodenum seem sufficient for diagnosis. Careful orientation of the samples is important to avoid misdiagnosis [9]. Some pathologists favour correct orientation with a stereomicroscope to improve their interpretation. Prospective studies must evaluate these orientation items, as there is a lack of data.

Antibodies
Coeliac disease is common both in Europe and the USA, with population studies suggesting a screening prevalence of the order of 1:150–1:300 [10–12]. All these studies are based on primary antibody screening studies. However, failed or delayed diagnosis in individuals is common [13]. Although small-bowel biopsy remains mandatory for diagnosis, serological testing for antibodies has sufficient sensitivity or specificity in the majority of coeliacs to allow selection of patients for biopsy.

In the late 1980s it became clear that certain circulating antibodies such as anti-gliadin antibody and anti-endomysium antibody had a high degree of sensitivity and specificity for the diagnosis of coeliac disease in childhood. An editorial in the Lancet in 1991 noted the disparity between diagnosis and serology [14]. In most studies, the sensitivities of serological markers have been evaluated in terms of severe (flat) mucosal lesions. Alternatively, a biopsy was performed only when serological markers such as anti-gliadin antibody and/or anti-endomysium antibody were positive. The positive predictive value of these IgA antibodies for coeliac disease came close to 100%.

Recent work in adults has suggested that antibodies fail to identify a subgroup of patients with villous atrophy [5,15]. Most of these patients do not have selective IgA deficiency, which is usually regarded as the main cause of false negative serology. A possibility is that the amount of gluten in the diet of untreated coeliacs may influence antibody positivity on a regional or individual basis, as it is known that a GFD causes the disappearance of anti-endomysium antibody before any villous recovery [16–18]. Another possibility is that serological positivity depends on the severity of the lesion and possibly on the length of intestine involved.

The endomysial antibody test is recognized as the most sensitive and specific standard for the serological part of the coeliac disease diagnosis. However, since the antigen recognized by the anti-endomysium antibody was found to be tissue transglutaminase (tTG), tests have been developed [19,20]. tTG was identified as the main, if not sole, coeliac disease auto-antigen. Anti-tTG testing therefore seems promising. The source of the antigen from guinea pig was a problem but recombinant techniques provide the genuine human tTG and minimize the risk for false positives [21]. Data for sensitivity in partial, total or subtotal villous atrophy have to be awaited.

Where there is a strong clinical suspicion of coeliac
In general, most coeliacs carry the HLA-DQ alleles of the HLA-DQA1 and HLA-DQB1 alleles, because: (1) the majority of coeliacs carry a combination of these alleles, which genes products together form the HLA-DQ2 heterodimer; and (2) gluten-specific HLA-DQ2 restricted T-cell clones have been isolated from coeliacs. tTG can deamidate particular glutamine residues in gluten-peptides into negatively-charged glutamic acid residues, thereby enhancing the binding affinity to HLA-DQ2 or DQ8 and increasing the T-cell response.

The HLA-DQ2 heterodimer, expressed by approximately 90% of coeliacs, seems extremely important in the T-cell response. It is likely that the HLA-DQ8 heterodimer, expressed by approximately 8–10% of coeliacs, works in the same way. However, only a small percentage of HLA-DQ2 and DQ8 carriers develop coeliac disease. Developing coeliac disease in HLA-DQ2nonDQ8nonDQ8 individuals seems rare, probably below 5%, which means that DQ2 DQ8 might be of help in selecting patients for serological testing and/or biopsy.

Coeliac disease is polygenetic. There is definitely a role for HLA genes, probably a major role. However, within this genetic puzzle the genes responsible for susceptibility are unknown.

Screening
In general, most coeliacs carry the HLA-DQ alleles associated with coeliac disease. The presence of symptoms does not discriminate between individuals with or without coeliac disease. One screening in a lifetime may not be enough to detect coeliac disease. An accurate, two-step strategy for screening based on selection of individuals with potential coeliac disease in high-risk groups – such as first-degree relatives and patients with Downs syndrome and/or auto-immune diseases – by HLA-DQ typing and on longitudinal serological coeliac disease screening has been suggested in selected groups. Case finding is still preferred above mass screening of the general population.

One screening in a lifetime may be not enough to detect coeliac disease in first-degree relatives or patients with so-called associated diseases. Future research is necessary to establish whether (poly)genetic labelling might help us to identify individuals with a high risk for coeliac disease.

Compliance with a gluten-free diet
William Dicke demonstrated that a GFD in which children avoid foods containing wheat, rye, barley (and oats) can be used to treat coeliac disease. However, sources of unintentional gluten intake by coeliacs are recognized and can be classified as follows: (1) residual gluten in so-called gluten-free wheat starch; (2) contamination of foods which are ‘naturally’ gluten free, e.g. contamination of egg, rice or maize flours with wheat flour; (3) mislabelling of foods and/or drugs, e.g. gluten is present but there are no gluten-containing constituents in the ingredient list; (4) consumption of products of which no unambiguous scientific opinion exists as to whether they are toxic to coeliacs.

Where there is a relapse of symptoms and/or histology and the patient has complied with the diet, an evaluation must be carried out. GFDs vary among coeliacs from very strictly gluten-free to occasional consumption of gluten. However, we advise a regimen which is as strict as possible according to the local regulations.

At present, wheat-starch consumption is not allowed by the coeliac societies in southern Europe; however, in some studies it has not hampered patients’ villous recovery. Additional data are required before definitive conclusions can be made. A subgroup of coeliacs, who are very sensitive to gluten, should use a wheat-starch free diet. In general, however, we accept that gluten-free wheat-starch based products are easier to use in cooking and are more palatable. This probably enhances the patients’ compliance with the GFD.

Oats can be well tolerated by coeliacs. Studies on oat challenge in coeliacs showed no effect on intraepithelial lymphocytes (IEL), villous height or quality of life. However, the oat products on sale in many countries...
Refractory coeliac disease can be divided into those without aberrant T cells (type 1) and those with aberrant T cells (type 2). We consider patients with ulcerative jejunitis to be type 2 refractory coeliacs, since their histology is not yet compatible with small-bowel lymphoma. However, we realize that there is a close correlation between type 2 refractory coeliac disease and enteropathy-associated T-cell lymphoma and more definitive criteria must be established.

Refractory coeliac disease could be primary, when patients fail to respond to a GFD following the initial diagnosis, or secondary, when a documented histological response is not maintained despite adhering to a GFD. For treatment trials to be designed, DQ typing and immunohistology are mandatory to understand and compare results. Treatment strategies for refractory coeliac disease must be based on adequate data.

Conclusion
The diagnosis of coeliac disease does not require further confirmation if it is based on duodenal histology showing villous atrophy, crypt hyperplasia and intraepithelial lymphocytosis while using a gluten-containing diet, which normalizes on a GFD. It is important that pathologists and clinicians become familiar with the pitfalls in diagnosis. Otherwise, every patient with an abnormal biopsy is in danger of being diagnosed with coeliac disease and given a GFD.

The finding of circulating antibodies (IgA, anti-endomysium antibody and/or αTG antibody) before commencing a GFD supports the diagnosis but is not essential, and should not be used for diagnosis without histological confirmation. The finding of HLA-DQ2 or HLA-DQ8 is still considered circumstantial evidence. However, DQ2-DQ8 typing is becoming easy, quick and less expensive.

Whether gluten challenge is required for minor histological abnormalities, such as intraepithelial lymphocytosis with or without crypt hyperplasia, must be investigated.

The definition from the early 1970s of coeliac disease as a permanent gluten intolerance producing a flat small-intestinal mucosa has been very valuable as a firm base for clinical studies and for comparison of data. Nowadays, we prefer an approach that allows for both simplicity and diversity in the diagnosis.

A careful prospective follow-up of patients in whom coeliac disease has been accurately diagnosed will allow more definitive diagnostic and therapeutic criteria to be established, provide better criteria for recognizing coeliacs at risk of becoming refractory and provide guidelines for the management of coeliacs. For instance,
looking for osteopenia in all adult coeliacs seems mandatory [39,40].

ESPGAN has established a diagnostic protocol for children, recognizing that diagnostic procedures such as immunogenetics and/or serology may be useful. Future working groups aiming to improve the diagnosis and management of coeliac disease, based on future research, will refine definitions and therapies in an evolutionary process.

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