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INTESTINAL IMMUNE REGULATION EXEMPLIFIED BY COELIAC DISEASE

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SUMMARY

Polymeric IgA is produced by 80-90% of the plasma cells in normal intestinal mucosa. It is transported externally by the epithelial secretory component and acts in a first line of mucosal defence by blocking antigen penetration. IgG, IgE and T cell mediated responses against soluble non-replicating luminal antigens are normally downregulated, perhaps by mechanisms involving interactions between the gut epithelium, T cells and macrophages leading to anergy or suppression collectively termed "oral tolerance". It has been claimed that IgA-producing cells escape suppression due to a specific effect of contrasupressor cells but the mechanisms remain unclear. Abrogation of oral tolerance may lead to increased activation of mucosal T lymphocytes with elevated lymphokine production. An altered lymphokine profile may induce aberrant epithelial HLA class II expression, increased epithelial permeability, crypt hyperplasia and disproportionate local overproduction of IgG. Expansion of a particular TCRγ/δ^+ intraepithelial T cell subset may also be involved in break of oral tolerance. Immunecomplex-mediated subepithelial complement activation together with local production of tumour necrosis factor and interferon-γ may cause epithelial damage and increased epithelial desquamation. Coeliac disease appears to be an example of break in oral tolerance to gluten peptides where the immunogenetics is at least partially defined.

INTRODUCTION

The human gastrointestinal tract is exposed to an enormous amount of antigens from food and micro-organisms. Despite this persistent bombardment of the mucosal membrane, adverse immunological responses to dietary antigens are relatively uncommon while effective immunity to infectious agents is usually elicited. The homeostatic mechanisms that explain a state of intestinal hyporesponsiveness to soluble non-replicating agents is known as "oral tolerance" and involve both humoral and cell-mediated immunity.

THE NORMAL STATE

Humoral immunity

There is a striking preponderance (80-90%) of IgA-producing blasts and plasma cells in the intestinal mucosa, and it has been estimated that at least 80% of all Ig-producing cells of the body are located in the gut (*Brandtzaeg* et al., 1989). The IgA cells produce

mainly dimers and larger polymers of IgA containing a disulphide-linked polypeptide called the J ("joining") chain (*Brandtzaeg*, 1985; *Mestecky* and *McGhee*, 1987). Polymeric IgA is transported externally along with J chain containing pentameric IgM by an epithelial glycoprotein called secretory component (SC) or poly-Ig receptor (*Brandtzaeg*, 1985). The secretory immunoglobulins (SIgA and SIgM) act in a first line of mucosal defence (*Brandtzaeg* et al., 1989). IgA is probably crucial for immunologic homeostasis also within the lamina propria because it lacks potent effector functions such as classical complement activation and hence may block triggering of non-specific biological amplification mechanisms (*Brandtzaeg* et al., 1987). At the same time antigen binding to SIgA blocks antigen penetration and thereby dampens potentially phlogistic IgG and IgE reactions and T cell mediated hypersensitivity against harmless luminal antigens (*Brandtzaeg* et al., 1987).

Cellular immunity

The normal intestinal mucosa contains numerous intraepithelial lymphocytes (IEL). Human IEL are mainly T lymphocytes ($CD3^+$) and 80-90% express CD8 whereas the lamina propria T cells are mainly $CD4^+$ (*Brandtzaeg* et al., 1989). IEL express the marker HML-1 that originally was thought to be unique for mucosal lymphocytes (*Cerf-Bensussan* et al., 1987) but later shown to be an activation antigen (*Schieferdecker* et al., 1990). However, IEL are negative for markers of recent activation such as MHC class II and CD25 (p55 IL-2 receptor) (*Selby* et al., 1983; *Trejdosiewicz* et al., 1987). Human IEL show no spontaneous cytotoxicity (*Cerf-Bensussan* et al., 1985) and they are negative for the H366 antigen which is a putative marker of MHC restricted cytotoxic $CD8^+$ T cells (*Trejdosiewicz* et al., 1987). These results and data from *in vitro* studies suggest that human IEL are mainly suppresser cells although this concept remains quite controversial as discussed later. In the normal state, intestinal IEL are found mainly along the basement membrane, apparently crossing it in either direction (*Marsh*, 1975b). Thus, after immune stimulation they may return to the lamina propria where their immunoregulatory function(s) perhaps are exerted.

Much interest is focused on intraepithelial T cells that express the γ/δ T cell receptor (TCR) (discussed in more detail later). In humans, 0-39% (median 2%) of the intraepithelial T cells express this alternative antigen receptor while few $TCR\gamma/\delta^+$ cells are seen in the lamina propria (*Halstensen* et al., 1989). About 75% of the intraepithelial $TCR\gamma/\delta^+$ lymphocytes are $CD8^-$ (*Halstensen* et al., 1989) and a remarkably large fraction (median 67%) express the Vδ1/Jδ1 encoded epitope revealed by monoclonal antibody δTCS1 (*Spencer* et al., 1989; *Halstensen* et al., 1989).

It has been claimed that the intestinal IgA production escapes suppression because of antisuppressor or contrasuppressor cells that preferentially support IgA responses (*Suzuki* et al., 1988; *Ernst* et al., 1988; *Green* et al., 1988), but the mechanisms remains unclear.

Migration of mucosal lymphocytes

Regulation and effector mechanisms of mucosal immunity depend on the migration of specifically primed T and B lymphocytes from organised gut associated lymphoid tissue (GALT) through lymph and peripheral blood primarily to the intestinal lamina propria. Extravasation of lymphoid cells appears to be mediated by receptors for endothelial

determinants ("vascular addressins") which to some extent are tissue specific (*Jalkanen* et al., 1986, 1988; *Streeter* et al., 1988). Additional adhesion molecules and other local factors are also involved (*Duijvestijn* and *Hamann*, 1989; *Bienenstock* et al., 1983). While B cells localise in the lamina propria and proliferate and differentiate there, many T cells find their way into the intestinal epithelium. The latter phenomenon is partially antigen-independent because intraepithelial lymphocytes are observed even before birth (*Spencer* et al., 1986), but luminal antigens clearly determine the magnitude of the intraepithelial migration (*Ferguson* and *Parrott*, 1972). The follicle associated epithelium covering the human PP contains particularly many T cells, especially near the antigen-transporting M ("membrane") cells (*Bjerke* et al., 1988).

COELIAC DISEASE

Coeliac disease is a small intestinal disorder characterised clinically by malabsorption, histologically by villous atrophy and crypt hyperplasia (*McNeish* et al., 1979), and genetically by a strong association with certain HLA class II allotypes (*Sollid* et al., 1989). It is precipitated by ingestion of gluten in wheat and some other grains and may be regarded a model for a break of oral tolerance.

Immunopathology of coeliac disease

T lymphocytes and macrophages. After gluten challenge in treated coeliac patients, there is a rapid and dose dependent migration of lymphocytes into the jejunal epithelium (*Rosenkrans* et al., 1981; *Leigh* et al., 1985). The proportion of $CD3^+CD45R0^+$ IEL is significantly raised and the density of such putative memory T cells is strikingly increased both in the surface and crypt epithelium of untreated patients compared with normal controls (*Scott* et al., 1987). There is also an elevated mitotic activity and blastoid transformation of many IEL (*Marsh*, 1975; 1988). However, the CD4 to CD8 ratio is unchanged compared with normal (*Selby* et al., 1983; *Malizia* et al., 1985). The proportion and absolute numbers of IEL $TCR\gamma/\delta^+$ cells are significantly increased both in untreated and partially treated coeliac patients (*Halstensen* et al., 1989; *Savilathi* et al., 1990). In untreated patients an increased fraction of the lamina propria $CD4^+TCR\alpha/\beta^+$ cells express IL-2 receptors (*Halstensen* et al., 1991) and there are raised circulating levels of soluble IL-2 receptor (*Crabtree* et al., 1989). An increase of IL-2R positive T cells is also detected after gluten stimulation *in vitro* for 24 h of small intestinal biopsy specimens from coeliac patients (*Scott* et al., 1991). Moreover, in such specimens cultivation with gluten induces IL-2R expression by macrophages (Mø) in significantly higher numbers than in controls (*Scott* et al., 1991). This strongly suggests that mucosal Mø play an important role in the pathogenesis of coeliac disease, perhaps by being a key link between gluten and genetic (HLA class II determined) disease susceptibility.

Epithelial SC and HLA class II expression. Increased epithelial expression of both SC and HLA class II determinants is seen in the active coeliac lesion (*Scott* et al., 1981). Bright immunohistochemical staining for SC extends from the crypts to the surface epithelium, whereas DR positivity extends from the surface deep into the hyperplastic crypts (*Scott* et al., 1981). In

addition, there is an increased expression of HLA-DP by the surface epithelium (*Scott* et al., 1987; *Marley* et al., 1987). HLA-DQ is only slightly or not at all expressed by epithelial cells in coeliac disease but strongly by Mø situated just beneath the surface epithelium (*Scott* et al., 1987).

The increased epithelial HLA class II expression in coeliac patients takes place quite rapidly after gluten challenge (*Ciclitira* et al., 1986), appears to be related to the number of intraepithelial $CD45R0^+$ T cells (*Scott* et al., 1987), and is most likely caused by cytokines released from primed T cells and perhaps activated Mø (*Cerf-Bensussan* et al., 1984). This possibility is supported by studies of the intestinal HT29 epithelial cell line which shows differential HLA class II expression (DR>DP>DQ) in response to interferon-γ (*Sollid* et al., 1987) with an additive stimulatory effect of tumour necrosis factor-α (*Kvale* et al., 1988). Local stimulation of T cells may therefore be an early event in the coeliac lesion. A consequence of this is probably also cytokine-induced crypt hyperplasia (*MacDonald* and *Spencer*, 1988) and increased epithelial permeability (*Madara* et al., 1988) with ensuing overstimulation of the B cell system.

Immunoglobulin production. In untreated coeliac disease the average numbers of jejunal IgA, IgM and IgG immunocytes per mucosal tissue unit are raised 3, 3.3 and 4.7 times, respectively (Figure 1). These results are supported by tissue culture studies (*Wood* et al., 1987). The local immune response is thus dominated by a typical SIgA and SIgM response with preserved J chain expression by mucosal plasma cells (about 90% positive) and a relatively high proportion of the IgA2 subclass (*Kett* et al., 1990). SC expression is also enhanced, and immunohistochemical staining indicates increased epithelial IgA and IgM transport (*Brandtzaeg* and *Baklien*, 1977).

Only trace amounts of IgG antibodies to gluten and other food antigens are detected in intestinal juice in contrast to IgA and IgM antibodies (*Labrooy* et al., 1986). However, untreated coeliac patients have substantial amounts of IgG antibodies to gluten and other food antigens in peripheral blood (*Scott* et al., 1984). An imbalanced humoral immune response to gluten is likewise suggested by the finding that a much larger percentage of jejunal IgG than IgA cells produce antibodies to gluten (*Brandtzaeg* and *Baklien*, 1976), and there is a significant inverse correlation between time to clinical relapse and the number of IgG producing cells in jejunal mucosa of coeliac children after gluten challenge (*Scott* et al., 1980). The gluten antibodies disappear more slowly from gut fluid than from serum during gluten restriction and persist for prolonged periods within jejunal IgM in treated adults (*O'Mahoney* et al., 1991).

Complement activation. Activated complement has recently been detected beneath the surface epithelium in untreated coeliac lesions (*Halstensen* et al., 1991a), being well correlated with the numbers of mucosal IgG cells and the serum level of gluten specific IgG and IgM. Furthermore, following gluten challenge, degranulation of mast cells is observed (*Horvath* et al., 1990) and also release of histamine and increased mucosal permeability (*Lavo* et al., 1989; 1990), phenomenona that may be initiated by split products of the complement cascade (especially C3a and C5a). These findings support the putative immunopathological role of IgG and suggest that gluten antigens are part of the complement activating immune complexes. Such complexes containing dimeric IgA may contribute

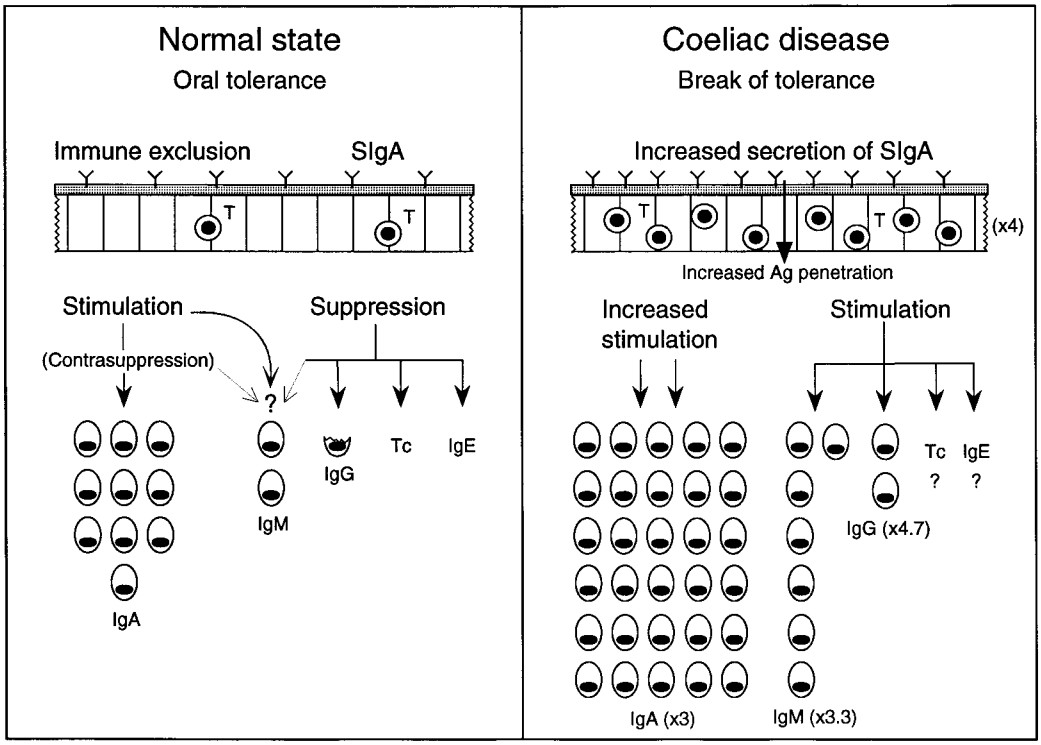

Figure 1: Schematical depiction of the intestinal mucosal immune system in coeliac disease compared with normal. Left: In the normal state a first line of defence is mediated by secretory antibodies, mainly SIgA. Oral tolerance is induced by suppression of systemic (phlogistic) types of immunity - that is, IgG and IgE antibody responses and T cell mediated delayed type hypersensitivity against non-adherent soluble luminal antigens. Contrasuppressor cells may release IgA-promoting helper cells from suppression so that a prominent mucosal IgA response can develop. Right: Coeliac disease represents a break in oral tolerance. Both secretory antibodies, IgG production and mucosal T cells are stimulated, the median number of IgA, IgM and IgG cells per mucosal tissue unit being raised 3, 3.3 and 4.7 times, respectively. The median number of T cells per mm of surface epithelium is raised 4 times and there is increased antigen penetration.

to degranulation of eosinophils since SIgA has been shown to exert this effect by itself *in vitro* (*Abu-Gazaleh* et al., 1989).

PUTATIVE MECHANISMS INVOLVED IN INTESTINAL IMMUNE REGULATION

In addition to immune exclusion performed by SIgA antibodies, animal experiments suggest that suppressive mechanisms protect the intestinal mucosa against potentially harmful systemic types of immune responses elicited by IgG, IgE or T-cell-mediated delayed type hypersensitivity. Such hyporesponsiveness in the gut may involve clonal anergy, suppressor T lymphocytes, cytotoxic T lymphocytes or suppressor Mø, but may also to some

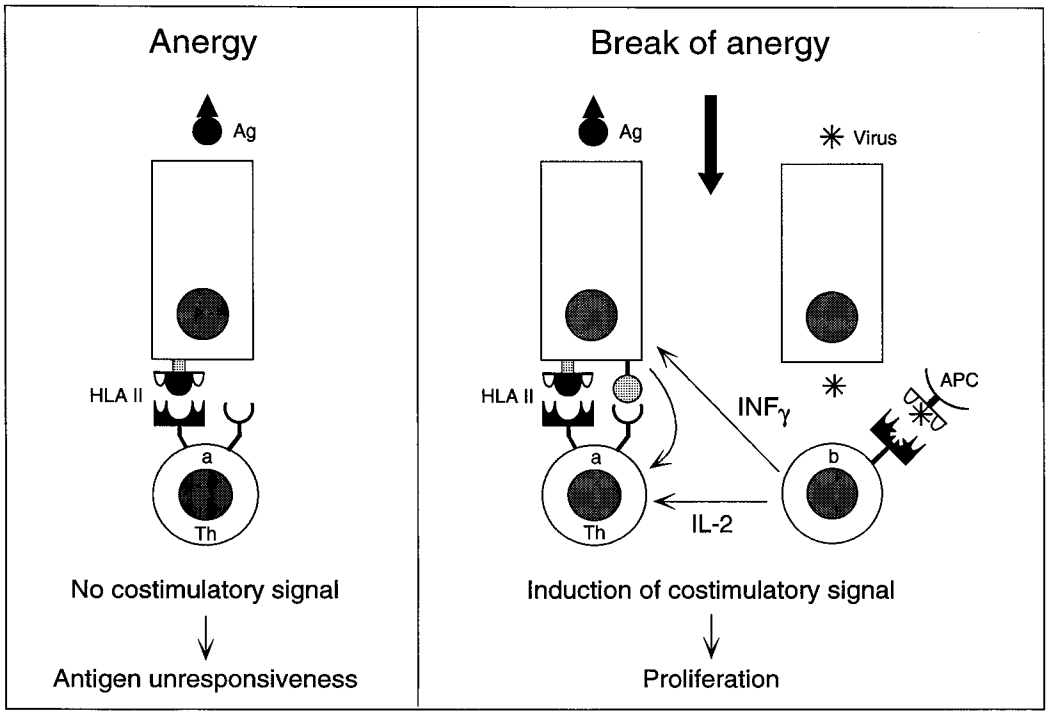

Figure 2: Hypothetical scheme for induction and abrogation of locally induced tolerance. Left: Antigen taken up by the intestinal epithelial cell is degraded and presented in the context of HLA class II to antigen specific $CD4^+$ Th cell in lamina propria. Lack of costimulation (IL-1 and/or adhesion molecules) results in antigen unresponsiveness (anergy). Right: Virusinfection induces activation of T cell (b) that secretes IL-2 and INF-γ. The latter cytokine induces increased expression of HLA determinants and adhesion molecules on epithelial cells, enabeling them together with IL-2 to provide sufficient costimulatory signals for induction of antigen-specific response of T cell (a).

extent depend on the liver (*Brandtzaeg* et al., 1989). Antigen handling by an intact gut epithelium seems to be critical (*Nicklin* and *Miller*, 1983) and direct injections of antigen into Peyer's patches is incompatible with the induction of oral tolerance (*Dunkley* and *Husband*, 1987).

Epithelial presentation of luminal antigens

Small intestinal epithelial cells express HLA class II determinants apically and basolaterally (*Scott* et al., 1980; *Selby* et al., 1983; *Scott* et al., 1987); they may therefore in theory be able to present immunogenic peptides to T lymphocytes which respond to stimuli both by producing effector molecules (lymphokines) and by proliferation (*Schwartz*, 1990). However, complete T cell activation requires two signalling events, one through the antigen specific receptor (TCR) and one through the receptor for a costimulatory molecule (*Schwartz*, 1990). In the absence of the latter signal (Figure 2), the T cells mount only a partial response and, more importantly, are subjected to clonal anergy in which state they do not produce their own growth factor interleukin 2 (IL2) on restimulation (*Schwartz*,

1990). In some cases costimulation can be provided by soluble mediators such as IL-1 (*Durum* et al., 1985). In other cases, cellular interactions are required (*Jenkins* et al., 1988). The two possibilities are difficult to distinguish because a close proximity is sometimes required to deliver high concentrations of labile mediators and such soluble ligands can act indirectly to increase the activity of the interacting cells (*Koide* et al., 1987). Activated keratinocytes induce anergy in T cell clones *in vitro*, presumably because they lack the ability to deliver a costimulatory signal (*Gaspari* et al., 1988). Murine intestinal epithelial cells expressing a relatively low level of MHC class II molecules, were unable to present bovine serum albumin (BSA) to antigen-specific T helper (Th) cells (*Zhang* and *Michael*, 1990). However, INF-γ treatment of the animals strongly enhanced epithelial MHC expression thus enabling antigen presentation to Th cells (*Zhang* and *Michael*, 1990).

Relatively low expression of MHC class II on intestinal epithelial cells may thus contribute to immunosuppression/anergy induced by oral administration of a protein antigen. Down-regulation of TCR and CD8 molecules on the antigen-specific T cells have also been suggested as a mechanism by which peripheral tolerance may occur (*Schonrich* et al., 1991; *Rocha* and *von Boehmer*, 1991). Nevertheless, anergic cells expressing IL-2R may be triggered *in vivo* by IL-2 from adjacent T cells responding to other antigens (*Schwartz*, 1990).

Virus infection may cause T cell activation with increased lymphokine production and increased epithelial MHC class II expression (Figure 2). Under these circumstances epithelial cells may be able to activate T cells because sufficient costimulatory signals are available (Figure 2). A virus infection may therefore be important in the initiation of coeliac disease by enabling epithelial cells in the presence of costimulatory signals to activate antigen specific T cells with gluten peptides.

Suppressor lymphocytes

The numerous intraepithelial $CD8^+$ T cells may be involved in oral tolerance. Bland and *Warren* (1986a) found that MHC class II positive columnar cells from rat villous epithelium presented ovalbumin *in vitro* to primed lymph node T cells, which thereby were induced to proliferate. This apparently class II restricted immune response led to antigen specific suppression and involved a $CD8^+$ subset (*Bland* and *Warren*, 1986b). The presence of T suppressor inducer (TSI) cells in the test system was not excluded. It has recently been shown that one chain of TCR of a particular $CD4^+$ clone can be secreted and contribute to induction of $CD8^+$ suppressor cells (*Zheng* et al., 1988). This finding suggests that $CD4^+$ TSI cells may be derived from $CD4^+$ Th cells and thus have identical specificity. *Mayer* and *Shlien* (1987) applied human colon epithelial cells in autologous or allogeneic mixed lymphocyte responses and included also macrophages pulsed with tetanus toxoid. They observed preferential stimulation of $CD8^+$ T lymphocytes. This might be due to crosslinking of CD8 molecules by CD1d (*Blumberg* et al., 1991a; 1991b) or another unique MHC class I-like epithelial molecule (*L. Mayer*, personal communication). The responding cells did not express a putative cytotoxicity marker (9.3) and showed no cytotoxic effect but caused nonspecific suppression (*Mayer* and *Shlien*, 1987).

It remains unclear how $CD8^+$ cells mediate suppression. T suppressor cells that recognise B or T cell idiotypes have been described (*Nisonoff* et al., 1977; *Lynch* et al., 1979; *Mohagheghpour* et

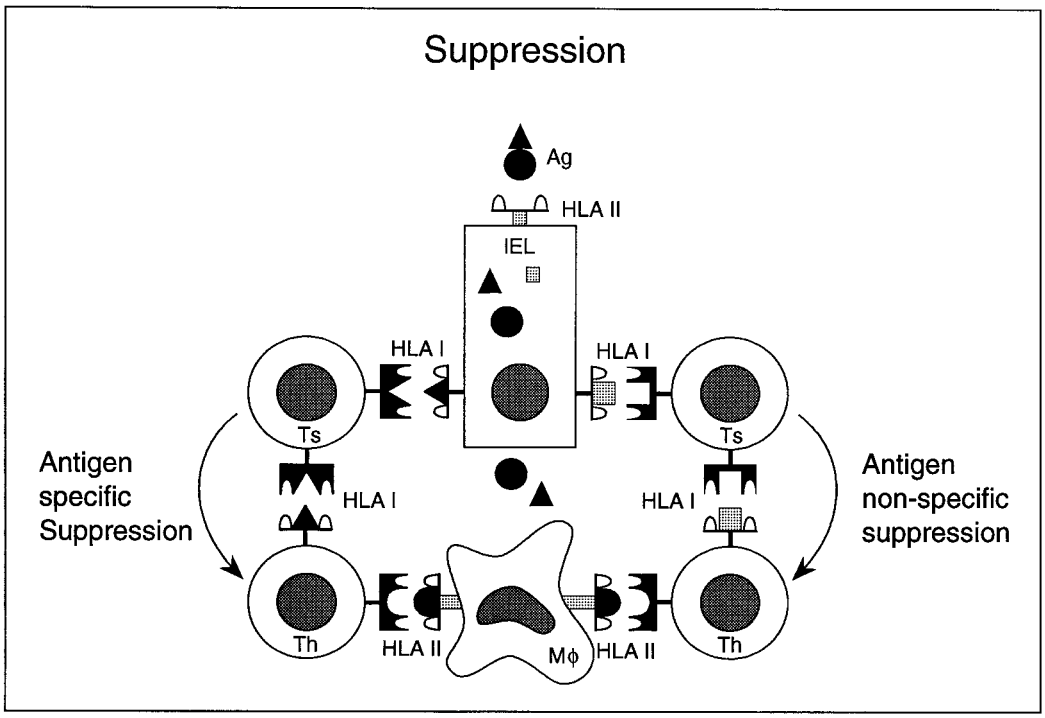

Figure 3: Putative mechanisms for T cell mediated suppression in small intestinal mucosa. Antigen is taken up by epithelial cell bound to apical HLA class II molecule. Antigen is further degraded to peptides with helper determinant (J) and suppressor determinant (H). The latter is presented to antigen specific $CD8^+$ T suppressor cell in the antigen binding grove of HLA class I molecule. The stimulated T suppressor cell returns to lamina propria where it interacts with $CD4^+$ Th cell expressing the suppressor determinant in the context of HLA class I (left). Alternatively, T suppressor cell is activated by fragment of HLA class II molecule presented in the context of HLA class I. T suppressor cell then returns to lamina propria and downregulates $CD4^+$ Th cell expressing the same HLA class II peptide in the antigen binding grove of a similar HLA class I molecule (right side of figure).

al., 1986; *Ben-Nun* et al., 1981; *Batchelor* et al., 1989; *Janeway*, 1989). It has also been found that antigens contain helper determinants and suppressor determinants which generally do not overlap (*Sercartz* and *Krzych*, 1991). Perhaps peptide binding to TCR is followed by endocytosis of the complex, processing and then loading of MHC class I molecules, which appear in the T cell endosomes (*Tse* and *Pernis*, 1984). T suppressor cells may also respond to MHC-class II peptides (*Sercartz* and *Krzych*, 1991) in the antigen binding grove of MHC class I molecules on Th cells (Figure 3). Moreover, soluble CD8 is released in response to lymphocyte activation (*Tomkinson* et al., 1989) and may function as an inhibitory ligand, thereby mediating the immunoregulatory activity of $CD8^+$ cells (*Hambor* et al., 1990). However, only cells undergoing primary but not secondary antigen stimulation through their TCR are susceptible to CD8 dependent inhibition (*Hambor* et al., 1990).

In mixed lymphocyte cultures with

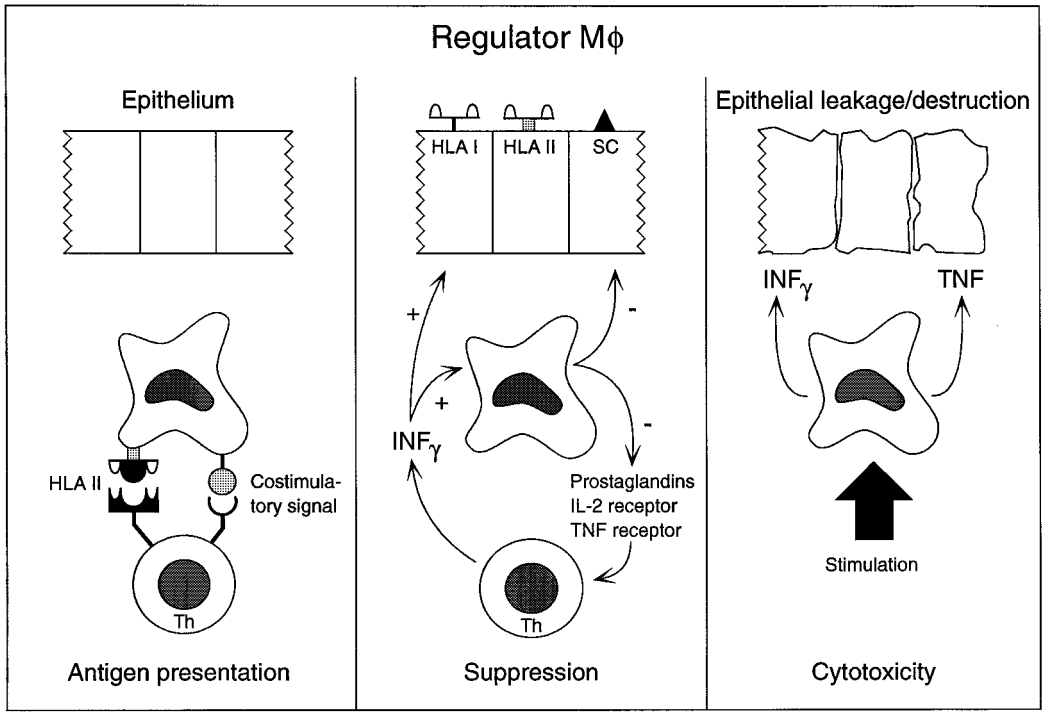

Figure 4: The subepithelial macrophage may have an important role in intestinal immune regulation: it may act as a professional antigen presenting cell (right); it may function as a suppressor cell by production of prostaglandins and soluble receptors for IL-2 and TNF (center); and it may also be a cytotoxic cell by producing TNF and INF-γ that synergistically cause epithelial damage (left).

peripheral blood mononuclear cells and intestinal epithelial cells from patients with inflammatory bowel disease, preferential stimulation of $CD4^+$ T cells is observed (*Mayer* and *Eisenhardt*, 1990). This difference from normal appears independent of the density of epithelial MHC class II molecules as shown by the fact that intestinal epithelial cells from other inflammatory disorders lead preferentially to stimulation of $CD8^+$ cells. There may thus be an intrinsic defect of certain surface molecules on epithelial cells from patients with inflammatory bowel disease, perhaps rendering crosslinking of CD8 molecules impossible. Similar experiments have not been performed in coeliac disease, but it would be very interesting if a preferential activation of $CD4^+$ cells takes place also in this disorder.

Cytotoxic "suppressor" cells

A "suppressor" cell may in theory downregulate an immune response by killing antigen presenting cells, B cells or Th cells. MHC class II restricted $CD8^+$ T suppressor cells efficiently inhibit specific antibody responses by lysing B cells with low levels of internalised and processed antigen (*Shinohara* et al., 1988). In addition, human T cells can process and present antigenic peptides in association with endogenous HLA class II molecules (*Lazavecchia* et al., 1988). Antigen specific Th cell may hence become the

target for cytotoxic T cells of the same specificity. However, studies of antigen specific unresponsiveness in humans with lepromatous lepra as model, suggest that specific $CD8^+$ T suppressor clones do not function by killing of antigen reactive Th clones (*Salgame* et al., 1989) and such cytotoxic cells may not operate *in vivo* (*Lanzavecchia*, 1989).

Regulatory macrophages

In the normal small intestine a population of large pleomorphic macrophages (Mø), positive for the myelomonocytic lysosomal antigen identified by Mab KPI but negative for the 36 KDa myelomonocytic L1 cytosol antigen or "calprotectin" (*Bjerke* et al., 1991) can be seen just below the surface epithelium. This subset is strongly HLA class II positive and also expressed constitutively HLA-DQ in contrast to the surface epithelial cells (*Scott* et al., 1987). Coeliac disease is notably associated with a particular HLA-DQ α/β heterodimer (*Sollid* et al., 1989). Because of the strategic location of these Mø, they may have an important role in intestinal immune regulation (Figure 4): A) They may act as "professional" antigen presenting cells, having the ability both to process antigens and give the necessary costimulatory signals for T lymphocytes. B) They may function as suppressor cells by production of prostaglandins (*Bray* et al., 1978; *Demenkoff* et al., 1980), and by release of blocking factors such as IL-2 receptors (*Loughnan* and *Nossal*, 1990) and tumour necrosis factor (TNF) receptors. In coeliac disease a large number of Mø express IL-2 receptors and this is seen to an even greater extent in small intestinal biopsy specimens from coeliac patients exposed for gluten *in vitro* (*Scott* et al., 1991). C) Activated Mø are effective killer cells; they may eliminate parasites and tumour cells by producing TNF and other toxic cytokines but may also contribute to the damage and increased turnover of surface epithelial cells seen in coeliac disease.

The function of the subepithelial Mø probably depends on the local cytokine profile. Increased antigen load may cause stimulation of T lymphocytes with enhanced interferon-γ (INF-γ) production. INF-γ may activate Mø to produce suppressive factors in a negative feed back system. However, on excessive stimulation of the intestinal immune system, mucosal Mø may change to killer cells with a predominant production of TNF (Figure 4).

T lymphocytes expressing TCRγ/δ^+ receptor

$TCR\gamma/\delta^+$ lymphocytes comprise a separate sublineage of T cells with mostly unknown specificity and restriction elements (*Bluestone* and *Matis*, 1989). Intestinal $TCR\gamma/\delta^+$ cells show a remarkable tropism for the epithelium and employ the variable $\delta 1$ gene ($V\delta 1/J\delta 1$) much more frequently than their counterparts in peripheral blood (*Spencer* et al., 1989; *Halstensen* et al., 1989; *Trejdosiewiz* et al., 1989). Intestinal $TCR\gamma/\delta^+$ IEL may primarily be directed against intestinal microorganisms or are perhaps specialised for elimination of transformed, infected or otherwise stressed autologous cells (*Janeway*, 1988; *Janeway* et al., 1988; *Lefrancois* and *Goodman*, 1989). The $TCR\gamma/\delta^+$ subset is also claimed to mediate contrasuppression in mice (*Fujihashi* et al., 1990).

The lymphokine profile of stimulated $TCR\gamma/\delta^+$ cells is at yet unknown. Both $TcR\alpha/\beta^+$ and $TCR\gamma/\delta^+$ cells may produce haemopoietic growth factors and granulocyte macrophage colony stimulating factor after stimulation, but IL-2 and IL-4 seem to be primarily produced by the $TcR\alpha/\beta^+$ cells (*Warren* et al., 1989). Stimulated $TCR\gamma/\delta^+$ IEL may

produce a different selection of lymphokines, perhaps depending on their actual CD45 isoform (*Halstensen* et al., 1990). Whether human TCRγ/δ IEL in coeliac disease are cytotoxic remains to be shown.

PUTATIVE IMMUNOPATHOLOGICAL MECHANISMS IN COELIAC DISEASE

Coeliac disease may be caused by a break in oral tolerance. A genetic predisposition may involve both HLA and the T cell receptor. Adenovirus 12 has been proposed as an additional environmental factor because of its cross-reactivity with A-gliadin (*Kagnoff* et al., 1987) but other studies have not supported this view (*Howdle* et al., 1989; *Carter* et al., 1989). However, a virus infection, perhaps combined with a high gluten intake, may supply the intestinal mucosa with sufficient costimulatory signals to change the local homeostasis from anergy/suppression to immunostimulation. HLA-DQ positive subepithelial Mø may present gluten peptides to gluten reactive T cells. Activated T cells may produce lymphokines that directly or indirectly increase the proliferation of epithelial crypt cells, increase intestinal permeability, activate B cells, transform B cells to plasma cells, and induce cytokine production in macrophages. Cytokines such as TNF, in addition to local complement activation, may exert a direct toxic effect on epithelial cells leading to increased desquamation and secondary crypt hyperplasia.

LITERATURE

Abu-Ghazaleh, R.I., Fujisawa, T., Mestecky, J., Kyle, R.A., and Gleich, G.J.: IgA-induced eosinophil degranulation. J. Immunol. 142, 2393-2400 (1989).

Batchelor, J.R., Lombardi, G., and Lechler, R.I.: Speculation on the specificity of suppression. Immunol. Today 10, 37-40 (1989).

Ben-Nun, A., Wekerle, H., and Cohen, I.: Vaccination against autoimmune encephalomyelitis with T-lymphocyte line cells reactive against myelin basic protein. Nature 292, 60-61 (1981).

Bienenstock, J., Befus, D., McDermott, M., Mirski, G., and Rosenthal, K.: Regulation of lymphoblast traffic and localization in mucosal tissues, with emphasis on IgA. Fed. Proc. 42, 3213-3217 (1983).

Bjerke, K., Brandtzaeg, P. and Fausa, O.: T cell distribution is different in follicle-associated epithelium of human Peyer's patches and villous epithelium. Clin. Exp. Immunol. 74, 270-275 (1988).

Bjerke, K., Halstensen, T.S., Mason, D.Y., and Brandtzaeg, P.: Distribution and HLA class II expression of three phenotypically distinct subpopulations of the monocyte/macrophage family in human ileal mucosa and Peyer's patches. Submitted for publication, 1991.

Bland, P.W., and Warren, L.G.: Antigen presentation by epithelial cells of the rat small intestine. I.Kinetics,antigen specificity and blocking by anti-Ia antisera. Immunology 58, 1-7 (1986a).

Band, P.W., and Warren L.G.: Antigen presentation by epithelial cells of the rat small intestine. II. Selective induction of suppressor T cells. Immunology 58, 9-14 (1986b).

Bluestone, J.A., and Matis, L.A.: TCRγ/δ-cells minor redundant T cell subset or specialized immune system component? J. Immunol. 142, 1785-1788 (1989).

Blumberg, R.S., Allan, C., Bleicher, P., et al.: Expression of CD1D by human gastrointestinal epithelial cells. A 1597, Digestive Disease Week, New Orleans, USA (1991a).

Blumberg,R.S., Landau, S., Ebert, E. and Balk, S.P.: T cell receptor (TCR)-α expression and recognition of CD1 by a human

jejunal intraepithelial lymphocyte (IEL) line. A 982, Digestive Disease Week, New Orleans, USA (1991b).

Brandtzaeg, P.: Role of J chain and secretory component in receptor mediated glandular and hepatic transport of immunoglobulins in man. Scand. J. Immunol. 22, 111-146 (1985).

Brandtzaeg, P. and Baklien, K.: Immunohistochemical studies of the formation and epithelial transport of immunoglobulins in normal and diseased human intestinal mucosa. Scand. J. Gastroenterol. 11 (Suppl. 36), 1-45 (1976).

Brandtzaeg, P. and Baklien, K.: Intestinal secretion of IgA and IgM: a hypothetical model. In:Immunology of the gut. Ciba Foundation Symposium 46. Elsevier, Amsterdam, 77-108 (1977).

Brandtzaeg, P., Baklien, K., Bjerke, K., et al.: Nature and properties of the human gastrointestinal immune system. In: Immunology of the gastrointestinal tract (Eds.: Miller, K., and Nicklin, S.). Boca Raton, Fla. 1-85 (1987).

Brandtzaeg, P., Halstensen, T.S., Kett, K., Krajci, P., Krale, D., Rognum, T.O., Scott, H., and Sollid, L.M.: Immunobiology and Immunopathology of human gut mucosa: humoral immunity and intraepithelial lymphocytes. Gastroenterology 97, 1562-1584 (1989).

Bray, M.A., Gordon, D., and Morley, J.: Prostaglandins as regulators in cellular immunity. Prostagland. Med. 1, 183-199 (1978).

Carter, M.J., Willcocks, M.M., Mitchinson, H.D., Record, C.O., and Madeley, C.R.: Is a persistant adenovirus infection involved in coeliac disease? Gut 30, 1563-1567 (1989).

Cerf-Bensussan, N., Quaroni, A., Kurnick, J.T., and Bhan, A.K.: Intraepithelial lymphocytes modulate Ia expression by intestinal epithelial cells. J. Immunol. 132, 2244-2252 (1984).

Cerf-Bensussan, N., Jarry, A., Brousse, N., Lisowska-Grospierre, B., Guy-Grand, D., and Griscelli, C.: A monoclonal antibody (HML-1) defining a novel membrane molecule present on human intestinal lymphocytes. Eur. J. Immunol 17, 1279-1286 (1987).

Ciclitira, P.J., Ellis, H.J., Wood, G.M., Howdle, P.D., and Losowsky, M.S.: Secretion of gliadin antibody by coeliac jejunal mucosa biopsies cultured *in vitro*. Clin. Exp. Immunol. 64, 119-124 (1986).

Crabtree, J.E., Heatley, R.V., Juby, L.D., Howdle, P.D., and Losowsky, M.S.: Serum interleukin-2-receptor in coeliac disease: response to treatment and gluten challenge. Clin. Exp. Immunol. 77, 345-348 (1989).

Demenkoff, J.H., Ansfield, M.J., Kaltreider, H.B., and Adam, E.: Alveolar macrophage suppression of canine bronchoalveolar lymphocytes: the role of prostaglandin E_2 in the inhibition of mitogen-responses. J. Immunol. 124, 1365-1370 (1980).

Duijvestijn, A., and Hamann, A.: Mechanisms and regulation of lymphocyte migration. Immunol. Today, 10, 23-28 (1989).

Dunkley, M.L., and Husband, A.J.: Distribution and functional characteristics of antigen-specific helper T cells arising after Peyer's patch immunization. Immunology, 61, 475-482 (1987).

Durum, S.K., Schmidt, J.A., and Oppenheim, J.J.: Interleukin 1: an immunological perspective. Annu. Rev. Immunol. 3, 263-287 (1985).

Ernst, P.B., Maeba, J., Lee, S.I., and Paraskevas, F.: A novel mechanism for the selection of isotype-specific antibody responses: the role of intestinal T cells in the regulation of IgA synthesis by the antisuppressor circuit. Immunology 65, 59-66 (1988).

Ferguson, A., and Parrott, D.M.V.: The effect on antigen deprivation on thymus-dependent and thymus-independent lymphocytes in the small intestine of the mouse. Clin. Exp. Immunol. 12, 477-488 (1972).

Fujihashi, K., Taguchi, T., McGhee, J.R., Eldridge, J.H., Bruce, M.G., Green, D.R., Singh, B., and Kiyono, H.: Regulatory function for murine intraepithelial lymphocytes. Two subsets of $CD3^+$, T cell receptor-1^+ intraepithelial lymphocyte T cells abrogate oral tolerance. J. Immunol. 145, 2010-2019 (1990).

Gaspari, A.A., Jenkins, M.K. and Katz, S.I.: Class II MHC-bearing keratinocytes induce antigen-specific unresponsiveness in hapten-specific TH1 clones. J. Immunol. 141, 2216-2220 (1988).

Green, D.R., McGhee, J., and Ptak, W.: Contrasuppression and the control of microenvironmental immune responses. Monogr. Al-

lergy 24, 236-243 (1988).
Halstensen, T.S., Scott, H., and Brandtzaeg, P.: Intraepithelial T cells of the TCRγ/δCD8$^-$ and Vδ1/Jδ1$^+$ phenotypes are increased in coeliac disease. Scand. J. Immunol. 30, 665-672, (1989).
Halstensen, T.S., Farstad, I.N., Scott, H., and Brandtzaeg, P.: Intraepithelial TcRα/β$^+$ lymphocytes express CD45RO more often than the TCRγ/δ$^+$ counterparts in coeliac disease. Immunology 71, 460-466 (1990).
Halstensen, T.S., Hvatum, M., Scott, H., Fausen, O., and Brandtzaeg, P.: Association of subepithelial deposition of activated complement and immunoglobulin G and M response to gluten in celiac disese. Gastroenterology 102, 751-759 (1992).
Halstensen, T.S., Farstad, I.N., Scott, H., et al.: Increased expression of IL-2 receptor on mucosal CD4$^+$ T cells in untreated coeliac disease and after gluten challenge *in vitro*. A 37-5, 11th Meeting in the European Federation of Immunological Societies, Helsinki, Finland (1991b).
Hambor, J.E., Kaplan, D.R., and Tykocinski, M.L.: CD8 functions as an inhibitory ligand in mediating the immunoregulatory activity of CD8$^+$ cells. J. Immunol. 145, 1646-1652 (1990).
Horvath, K., Nagy, L., Horn, G., Simon, K., Csiszar, K., and Bodanszky, H.: Intestinal mast cells and neutrophil chemotactic activity of serum following a single challenge with gluten in coeliac children on a gluten-free diet. J. Pediatr. Gastroenterol. Nutr. 9, 276-280 (1989).
Howdle, P.D., Blair Zajdel, M.E., Smart, C.J., Trejdosiewicz, L.K., Blair, G.E., and Losowsky, M.S.: Lack of a serologic response to an E1B protein of adenovirus 12 in coeliac disease. Scand. J. Gastroenterol. 24, 282-286 (1989).
Jalkanen, S., Reichert, R.A., Gallatin, W.M., Bargatze R.F., Weissman, I.L., and Butcher, E.C.: Homing receptors and the control of lymphocyte migration. Immunol. Rev. 91, 39-60 (1986).
Jalkanen, S., Streeter, P., Lakey, E., Bargatze, R., and Butcher, E.C.: Lymphocyte and endothelial cell recognotion elements that control lymphocyte traffic to mucosa-associated lymphatic tissues. Monogr. Allergy 24, 144-149 (1988).
Janeway, C.A.: Frontiers of the immune system. Nature 333, 804-806 (1988).
Janeway, C.A.: Immunotherapy by peptides? Nature 341, 482-483 (1989).
Janeway, C.A., Jones, B., and Hayday, A.: Specificity and function of T cells bearing γ/δ receptors. Immunol. Today 9, 73-76 (1988).
Jenkins, M.K., Ashwell, J.D., and Schwartz, R.H.: Allogeneic non-T spleen cells restore the responsiveness of normal T cell clones stimulated with antigen and chemically modified antigen-presenting cells. J. Immunol. 140, 3324-3330, (1988).
Kagnoff, M.F., Paterson, Y.J., Kumar, Kasarda, D.D., Carbone, F.R., Unsworth, D.J., and Austin, R.K.: Evidence for the role of a human intestinal adenovirus in the pathogenesis of coeliac disease. Gut 28, 995-1001 (1987).
Kett, K., Scott, H., Fausa, O., and Brandtzaeg, P.: Secretory immunity in celiac disease: cellular expression of immunoglobulin A subclass and joining chain. Gastroenterology 99, 386-392 (1990).
Koide, S.L., Inaba, R.M., and Steinman, R.M.: Interleukin 1 enhances T-dependent immune responses by amplifying the function of dendritic cells. J. Exp. Med. 165, 515-530 (1987).
Kvale, D., Brandtzaeg, P., and Løvhaug, D.: Up-regulation of the expression of secretory component and HLA molecules in a human colonic cell line by tumor necrosis factor-α and gamma interferon. Scand. J. Immunol. 28, 351-357 (1988).
Labrooy, J.T., Hohmann, A.W., Davidson, G.P., Hetzel, P.A., Johnson, R.B., and Shearman, D.J.: Intestinal and serum antibody in coeliac disease: a comparison using ELISA. Clin. Exp. Immunol. 66, 661-668 (1986).
Lanzavecchia, A.: Is suppression a function of class II restricted cytotoxic T cells? Immunol. Today 10, 157-159 (1989).
Lanzavecchia, A., Roosnek, E., Gregory, T., Berman, P., and Abrigani, S.: T cells can present antigens such as HIV gp120 to their own surface molecules. Nature 334, 530-532 (1988).
Lavo, B., Knutson L., Loof, L., Odlind, B., Venge, P., and Hallgren, R.: Challenge with gliadin induces eosinophil and mast cell activation in the jejunum of patients with coeliac disease. Am. J. Med. 87, 655-

660 (1989).
Lavo, B., Knutson, L., Loof, L., and Hallgren, R.: Gliadin challenge-induced jejunal prostaglandin E2 secretion in coeliac disease. Gastroenterology 99, 703-707 (1990).
Lefrancois, L., and Goodman, T.: *In vivo* modulation of cytolytic activity and Thy-1 expression in TCR-γ/δ^+ intraepithelial lymphocytes. Science 243, 1716-1718 (1989).
Leigh, R.J., Marsh, M.N., Crowe, P., Kelly, C., Garner, V., and Gordon, D.: Studies of intestinal lymphoid tissue. IX. Dose-dependent, gluten-induced lymphoid infiltration of coeliac jejunal epithelium. Scand. J. Gastroenterol. 20, 715-719 (1985).
Loughnan, M.S., and Nossal, G.J.V.: A regulatory role for the soluble IL-2 receptor via competition with the p75 cell-surface form of the receptor for IL-2. J. Mol. Cell. Immunol. 4, 307-316 (1990).
Lynch, R.G., Rohrer, J.W., Odermatt, B., et al.: Immunoregulation of murine myeloma cell growth and differentiation: a monoclonal model of B cell differentiation. Immunol. Rev. 48, 45-80 (1979).
MacDonald, T.T., and Spencer, J.: Evidence that activated mucosal T cells play a role in the pathogenesis of enteropathy in human small intestine. J. Exp. Med. 167, 1341-1349 (1988).
Madara, J.L., and Stafford J.: Interferon-γ directly affects barrier function of cultured intestinal epithelial monolayers. J. Clin. Invest. 83, 724-727 (1988).
Malizia, G., Trejdosiewicz, L.K., Wood, G.M., Howdle, P.D., Janossy, G., and Losowsky, M.S.: The microenvironment of coeliac disease. T cell phenotypes and expression of the T2 "T blast" antigen by small bowel lymphocytes. Clin. Exp. Immunol. 60, 437-446 (1985).
Marley, N.J.E., Macartney, J.C., and Ciclitira, P.J.: HLA-DR, DP and DQ expression in the small intestine of patients with coeliac disease. Clin. Exp. Immunol. 70, 386-393 (1987).
Marsh, M.N.: Studies of intestinal lymphoid tissue. I. Electron microscopic evidence of "blast transformation" in epithelial lymphocytes of mouse small intestinal mucosa. Gut 16, 665-674 (1975).
Marsh, M.N.: Studies of intestinal lymphoid tissue. II. Aspects of proliferation and migration of epithelial lymphocytes in the small intestine of mice. Gut 16, 674-682 (1975b).
Marsh, M.N.: Studies of lymphoid tissue. XI. The immunopathology of cell-mediated reactions in gluten sensitivity and other enteropathies. Scanning Microsc. 2, 1663-1684 (1988).
Mayer, L., and Shlien, R.: Evidence for function of Ia molecules on gut epithelial cells in man. J. Exp. Med. 166, 1471-1483 (1987).
Mayer, L., Eisenhardt, D., Salomon, P., Bauer, W., Plous, R., and Piccinini, L.: Expression of class II molecules on intestinal epithelial cells in humans. Gastroenterology 100, 3-12 (1991).
McNeish, A.S., Harms, H.K., Rey, J., et al.: The diagnoses of celiac disease: a commentary on the current prectices of members of the European Society for Paediatric Gastroenterology and Nutrition (ESPGAN). Arch. Dis. Child. 54, 783-786 (1979).
Mestecky, J., and McGhee, J.R.: Immunoglobulin A (IgA): Molecular and cellular interactions involved in IgA biosynthesis and immune response. Adv. Immunol. 40, 153-245 (1987).
Mohagheghpour, N., Damle, N.K., Takada, S., and Engelman, E.G.: Generation of antigen receptor-specific suppressor T cell clones in man. J. Exp. Med. 164, 950-955 (1986).
Nicklin, S. and Miller, K.: Local and systemic immune responses to intestinally presented antigen. Int. Archs. Allergy Appl. Immunol. 72, 87-90 (1983).
Nisonoff, A., Ju, S.T., and Owen, E.L.: Studies of structure and immunosuppression of a cross-reactive idiotype in strain A mice. Immunol. Rev. 34, 89-118 (1977).
O'Mahony, S., Arranz, E., Barton, J.R., and Ferguson, A.: Dissociation between systemic and mucosal humoral immune responses in coeliac disease. Gut 32, 29-35 (1991).
Rocha, B., and von Boehmer, H.: Peripheral selection of the T cell repertoire. Science 251, 1225-1228 (1991).
Rosenkrans, P.C.M., Meijer, C.J.L.M., Polanco, I., et al.: Long-term morphological and immunohistochemical observations on biopsy specimens of small intestine of children with gluten-sensitive enteropathy. J. Clin. Pathol. 34, 138-144 (1981).
Salgame, P., Modlin, R., and Bloom, B.R.:

On the mechanism of human T cell suppression. Int. Immunol. 1, 121-129 (1989).

Savilath, E., Arato, A., and Verkasalo, M.: Intestinal γ/δ receptor-bearing T lymphocytes in celiac disease and inflammatory bowel diseases in children. Constant increase in celiac disease. Ped. Res. 28, 579-581 (1990).

Schieferdecker, H.L., Ullrich, R., Weiss-Breckwoldt, A.N., Schwarting, R., Stein, M., Riecken, E.O., and Zeitz, M.: The HML-1 antigen of intestinal lymphocytes is an activation antigen. J. Immunol. 144, 2541-2549 (1990).

Schonrich, G., Kalinke, U., Momberg, F., Malissen, M., Schmitt-Verhulst, A.M., Malissen, B., Hammeling, G.J., and Arnold, B.: Down-regulation of T cell receptors on self reactive T cells as a novel mechanism for extrathymic tolerance induction. Cell 65, 293-304 (1991).

Schwartz, R.H.: A cell culture model for T lymphocyte clonal anergy. Science 248, 1349-1356 (1990).

Scott, H., Ek, J., Baklien, K., and Brandtzaeg, P.: Immunoglobulin-producing cells in the jejunal mucosa of children with coeliac disease on a gluten-free diet and after gluten challenge. Scand. J. Gastroenterol. 15, 81-88 (1980).

Scott, H., Solheim, B.G., Brandtzaeg, P., and Thorsby, E.: HLA-DR-like antigens in the epithelium of the human small intestine. Scand. J. Immunol. 12, 77-82, 1980.

Scott, H., Brandtzaeg, P., Solheim, B.G., and Thorsby,E.: Relation between HLA-DR-like antigens and secretory component (SC) in jejunal epithelium of patients with coeliac disease or dermatitis herpetiformis. Clin. Exp. Immunol. 44, 233-238 (1981).

Scott, H., Fausa, O., Ek, J., and Brandtzaeg, P.: Immune response patterns in coeliac disease. Serum antibodies to dietary antigens measured by an enzyme-linked immunosorbent assay (ELISA). Clin. Exp. Immunol. 57, 25-32 (1984).

Scott, H., Sollid, L.M., Fausa, O., Brandtzaeg, P., and Thorsby, E.: Expression of MHC class II subregion products by jejunal epithelium of patients with coeliac disease. Scand. J. Immunol. 26, 563-572 (1987).

Scott, H., Halstensen, T.S., Farstad, I.N., et al.: Interleukin-2 receptor positive T cells and macrophages are increased in coeliac mucosa after gluten stimulation *in vitro*. A 37-5, 11th Meeting in the European Federation of Immunological Societies, Helsinki, Finland (1991).

Selby, W.S., Janossy, G., Bofill, M., and Jewell, D.P.: Lymphocyte subpopulations in human small intestine. The findings in normal mucosa and in the mucosa of patients with coeliac disease. Clin. Exp. Immunol. 52, 219-228 (1983).

Sercarz, E., and Krzych, U.: The distinctive specificity of antigen-specific suppressor T cells. Immunol. Today 12, 111-118 (1991).

Shinohara, N., Watanabe, M., Sachs, D.H., and Hozumi, N.: Killing of antigen reactive B cells by class II-restricted soluble antigen-specific $CD8^+$ cytolytic T lymphocytes. Nature 336, 481-484 (1988).

Sollid, L.M., Gaudernack, G., Markussen, G., Kuale, D., Brandtzaeg, P., and Thorsby, E.: Induction of various HLA class II molecules in a human colonic adenocarcinoma cell line. Scand. J. Immunol. 25, 175-180 (1987).

Sollid, L.M., Markussen, G., Ek, J., Gjerde, H., Vartdal, F., and Thorsby, E.: Evidence for a primary association of coeliac disease to a particular HLA-DQ α/β heterodimer. J. Exp. Med. 169, 345-350 (1989).

Spencer, J., Dillon, S.B., Isaacson, P.G., and MacDonald, T.T.: T cell subclasses in fetal human ileum. Clin. Exp. Immunol. 65, 553-558 (1986).

Spencer, J., Isaacson, P.G., Diss, T.C., and MacDonald, T.T.: Expression of disulfide-linked and non-disulfide-linked forms of the T cell receptor γ/δ heterodimer in human intestinal intraepithelial lymphocytes. Eur. J. Immunol. 19, 1335-1338 (1989).

Streeter, P.R., Berg, E.L., Rouse, B.T.N, Bargatze, R.F., and Butcher, E.C.: A tissue-specific endothelial cell molecule involved in lymphocyte homing. Nature 331, 41-46 (1988).

Suzuki, G., Kawase, Y., Koyasu, S., Yahara, I., Kobayashi, Y., and Schwartz, R.H.: Antigen-induced suppression of the proliferative response of T cell clones. J. Immunol. 140, 1359-1365 (1988).

Tomkinson, B.E., Brown, M.C., Ip, S.H., Carrabis, S., and Sullivan, J.L.: Soluble CD8 during T cell activation. J. Immunol. 142, 2230-2236 (1989).

Trejdosiewicz, L.K., Malizia, G., Badr-el-Din,

S., Smart, C.J., Oakes, D.J., Southgate, J., Howdle, P.D., Janossy, G., Poulter, L.W., and Losowsky, M.S.: T cell and mononuclear phagocyte populations of the human small and large intestine. Adv. Exp. Med. Biol. 216A, 465-473 (1987).

Trejdosiewicz, L.K., Smart, C.J., Oakes, D.J., Howdle, P.D., Malizia, G., Campana, D., and Boylston, A.W.: Expression of T-cell receptors TcR1 (γ/δ) and TcR2 (α/β) in the human intestinal mucosa. Immunology 68, 7-12 (1989).

Tse, D.B., and Pernis, B.: Spontaneous internalization of class I major histocompatibility complex molecules in T lymphoid cells. J. Exp. Med. 159, 193-204 (1984).

Warren, H.S., Bezos, A., McPhun, V., Pembrey, R.G., Pullman, W.E., Elsbury, S., Preston, L., and Hapel, A.J.: Differential cytokine production by natural killer cells and T cells of the α/β and γ/δ t-cell receptor classes. Transpl. Proc. 21, 77 (1989).

Wood, G.M., Howdle, P.D., Trejdosiewicz, L.K., and Losowsky, M.S.: Jejunal plasma cells and *in vitro* immunoglobulin production in adult coeliac disease. Clin. Exp. Immunol. 69, 123-132 (1987).

Zhang, Z., and Michael, J.G.: Orally inducible immune unresponsiveness is abrogated by INF-γ treatment. J. Immunol. 144, 4163-4165 (1990).

Zheng, H., Boyer, M., Fotedar, A., Singh, B., and Green, D.R.: An antigen-specific helper T cell hybridoma produces an antigen-specific suppressor inducer molecule with identical antigen fine specificity: implications for the antigen recognition and function of helper and suppressor inducer cells. J. Immunol. 140, 1351-1358 (1988).