Progress report

Human intestinal intraepithelial lymphocytes

Of all the various cell types present within the lamina propria, it is remarkable that only lymphocytes predominantly gain access to the epithelium. It seems likely that intestinal intraepithelial lymphocytes have an important immune function, and that this function is distinct from that of the lamina propria lymphocyte, although lymphocytes from both compartments (epithelium and lamina propria) presumably interact in their immune functions. Considerable functional and morphological data have been derived concerning the intraepithelial lymphocytes since this topic was reviewed by Ferguson in 1977. It is my intention to review data in regard to the intraepithelial lymphocytes largely published since 1977, to provide some new data, to compare the function and structure of the intraepithelial lymphocytes with that of the lamina propria lymphocyte, and to compare (when appropriate) the structure and function of both intraepithelial lymphocytes and lamina propria lymphocytes with that of peripheral blood lymphocytes. The role of the human intraepithelial lymphocytes will be compared and contrasted to that of the rodent intraepithelial lymphocytes when pertinent. Intraepithelial lymphocytes have been described since 1847 and have been the subject of many curious and speculative publications. For those interested in details of earlier studies of the intraepithelial lymphocytes, the reviews by Otto in 1973 and by Douglas and Weetman in 1975 are highly recommended.

Morphology

These cells are located within the epithelium, above the basal lamina, and between epithelial cells—that is, interepithelial cell and intraepithelial in location. Presence of lymphocytes within epithelial cells (emperipolesis) has not been demonstrated. Intraepithelial lymphocytes tend to be found largely at the base of epithelial cells, below epithelial nuclei. In the normal intestine these cells are largely medium sized (5–9 μm in diameter), contain sparse cytoplasmic organelles including mitochondria, lysosomal granules, ribosomes (both isolated and polyribosomes), endoplasmic reticulum (rough predominating over smooth by a ratio of 2:1), and a small Golgi complex. The nucleus contains large amounts of heterochromatin and lesser amounts of euchromatin. Nucleoli are often present. The cytoplasm tends to be less dense than that of adjacent epithelial cells, contains organelles indicated above, and endocytic vesicles are often present along the plasma membrane. The intraepithelial lymphocyte tends to be rounded to elongated in shape, and in active inflammatory states often has numerous elongated cytoplasmic projections which make...
intimate contact with adjacent epithelial cell membranes. Morphology of the intraepithelial lymphocytes in diseased intestine will be described in later sections. In the non-inflamed intestine, the intraepithelial lymphocytes tend to be tightly compressed between epithelial cells. Intraepithelial lymphocytes do not form junctional contacts (desmosomes and/or tight junctions) with adjacent epithelial cells.

The nature of intraepithelial lymphocyte ‘granules’ may be of importance in defining intraepithelial lymphocyte function, and thus some background material in regard to lymphocyte granules is reviewed. Rodent intraepithelial lymphocytes have very large granules, some of which contain histamine, and there is conflicting evidence that rodent intraepithelial lymphocytes may be mast cell derived. There is no solid evidence that human intraepithelial lymphocytes are mast cell derived and their granules are small in size. The granules are membrane bound and are clearly lysosomes. Confusion could be avoided if they were called lysosomes, not granules. The custom of calling heterogeneous and homogeneous dense bodies, when found in lymphocytes, as granules rather than lysosomes is so deeply embedded in the scientific literature, however, that both terms must be utilised.

The granules are azurophilic in Giemsa and Wright stained preparations, and are stained with alcian blue (pH 2-2) and are metachromatic with toluidine blue (pH 4). The lysosomal nature of lymphocyte granules was documented in 1965 by showing that these granules contained acid phosphatase. Lymphocyte granules have also been shown to contain beta-glucuronidase and alpha-naphthyl acetate esterase. Structures that are the site of both alpha-naphthyl acetate esterase and acid phosphatase activity represent the Gall body, a cluster of primary lysosomes surrounding a lipid droplet. The distribution of lysosomes in lymphocytes is of limited value in defining their function. The majority of human T cells which possess surface receptors for IgM contain Gall bodies which stain for alpha-naphthyl acetate esterase in a localised or dot-like fashion while the majority of human T cells which possess surface receptors for IgG have lysosomes scattered diffusely throughout the cytoplasm.

Lymphocytes do not contain peroxidases, an enzyme found in the myelo-monocytic series. All subclasses of T lymphocytes examined have been shown to contain at least, in part, lysosomal granules including Tg and Tm cells; Leu 3a+ cells; Leu 7+ and (Leu 11+) cells; Leu 3+, 7a, M1+ cells; Leu 2a+, T8+ cells; and lymphokine activated killer cells (Leu 1+, T8+, T3+).

B-lymphocytes do not contain granules and have very little acid phosphatase and alpha-naphthyl acetate esterase activity. It is likely that the presence of lysosomal granules (including multivesicular bodies) is necessary for the cytolytic activity of lymphocytes, whether the activity is antibody mediated or natural killer mediated. Indeed the presence of granules may represent a marker for all human cytolytic lymphocytes.

In 1974, Guy-Grand showed in the rat that the vast majority of intraepithelial lymphocytes were T cells and that, while both B cells and T cells home to the gut, only T cells home to the epithelium. In 1976, Meuwissen et al showed in human colon and intestine that the vast majority of intraepithelial lymphocytes were T cells, defined by immunoperoxidase localisation of intraepithelial lymphocytes using an anti-T cell globulin.
This finding has been repeatedly confirmed with immunoperoxidase and immunofluorescent localisation of a variety of monoclonal antibodies to T cells.\textsuperscript{12,20,25-32} \textit{In vitro} data indicating the presence of B cells within the intraepithelial lymphocyte population must be discounted.\textsuperscript{33} The B cells must have been contaminants derived from the lamina propria or from lymphoid follicles.\textsuperscript{26} Plasma cells are not found within the epithelium except possibly in the lymphoepithelial complex found in Peyer's patches.\textsuperscript{34} Macrophages and mast cells are occasionally found within the epithelium while epithelial PMN leucocytes and eosinophils are quite unusual except in inflammatory states.\textsuperscript{5,12,26,34}

Intraepithelial lymphocytes, virtually all of which are T cells, do not express class II histocompatibility antigens (HLA-DR, 1a-like), even in inflammatory states.\textsuperscript{20,26,30-32} Moreover, 80 to 90\% of intraepithelial T cells express the phenotype of suppressor cells (T8, Leu 2a), while only 10 to 20\% express the helper phenotype (T4, Leu 3a).\textsuperscript{20,27,29-32} In contrast, lamina propria lymphocytes are predominantly of the helper phenotype (60-70\% T4\textsuperscript{+}, Leu 3a\textsuperscript{+}).\textsuperscript{20,27,29-32} As expected, intraepithelial lymphocytes express Class I HLA antigens\textsuperscript{29} but do not express T cell activation antigen (Tac),\textsuperscript{30,31} complement receptors (c3b),\textsuperscript{30,31} and rarely express antigens characteristic of natural killer cells (HNK-1, Leu 7, Leu 11, T 10, M1).\textsuperscript{20,30,32}

In summary, intraepithelial lymphocytes are T cells predominantly of the suppressor phenotype. They are not activated, and do not express class II MHC antigens, and even though they often have the morphology of natural killer and/or killer cells (large granular lymphocytes), they do not bear surface antigens characteristic of natural killer cells. Intraepithelial lymphocyte populations do not contain B cells. Macrophages, mast cells, and PMN and eosinophilic leucocytes constitute a distinct minority of this cellular compartment in the normal intestine.

Quantification of intraepithelial lymphocytes is inappropriately a subject of controversy.\textsuperscript{35} Two methods of quantification have been proposed, one is to quantify the number of intraepithelial lymphocytes in relation to the number of epithelial cells\textsuperscript{36} and the other is to quantify the number of intraepithelial lymphocytes in relation to mucosal volume as determined by the length of underlying muscularis mucosae—that is, areal density.\textsuperscript{6} In coeliac sprue there is an increase in number of intraepithelial lymphocytes/100 epithelial cells while there is a decrease in number of intraepithelial lymphocytes per unit area (millimeters of muscularis mucosae) when compared with normal intestine.\textsuperscript{37} The latter (areal density) method would be the most appropriate method for quantification of intraepithelial lymphocytes if the immune response were directed towards all portions of the intestinal mucosa, while the former (intraepithelial lymphocytes/epithelial cells) would be appropriate if the immune response were largely directed towards the epithelium. Morphologically, the immune response in most intestinal diseases of unknown aetiology, or pathogenesis, is directed both towards the epithelium (\textit{vide infra}) and towards the lamina propria. It is my opinion that generally quantification of intraepithelial lymphocytes should be in relation to the epithelium, not in relation to areal density, and that both epithelial and lamina propria be quantified separately. In the final analysis, any method of quantification of intraepithelial lymphocytes at present is arbitrary.
The number of intraepithelial lymphocytes/100 epithelial cells in normal jejunum ranges from 9–39 with a mean of 21.1 with a standard deviation of 7.5.2 We found a mean of 20 intraepithelial lymphocytes/100 epithelial cells in normal jejunum,5 13/100 epithelial cells in normal ileum and 5/100 epithelial cells in normal colon.32

The cellular immune content of the lamina propria is strikingly different from that of the epithelium, and there are clearly differences in immune function of intraepithelial lymphocytes and of lamina propria lymphocyte.26 Lamina propria lymphocytes consist predominantly of the helper phenotype (T4, Leu 3a) and with a smaller component of the suppressor phenotype (T8, Leu 2a), the ratio of helper to suppression being similar to that of peripheral blood, 2:1.20 27 29–32 Plasma cells are profusely present in the lamina propria with ratios of IgA:IgM:IgG containing cells of 20:3:1. Macrophages, fibroblasts, and occasional mast cells and leucocytes are present throughout the normal lamina propria. T cells found in the lamina propria are occasionally activated on the basis of expression of Class II MHC antigens and Tac antigen,30–32 and NK cells (Leu 7+, and/or Leu 11+) are far more easily detected in the lamina propria than in the epithelium.20 30 32 A detailed description of the function of lamina propria lymphocytes is beyond the scope of this review. A comparison of lamina propria lymphocyte function to that of intraepithelial lymphocytes function will be given when appropriate in the next section.

**In vitro studies of intraepithelial lymphocytes**

Enriched populations of intestinal and/or colonic intraepithelial lymphocytes can be collected and analysed *in vitro*.12 26 33 38 The intestinal epithelium with its associated lymphocytes is separated from the underlying lamina propria by incubation in a calcium-magnesium free medium containing 0-7mM ethylenediaminetetraacetate.26 33 The epithelial cells are then removed by washing through nylon mesh and glass wool columns. The yield of intraepithelial lymphocytes is approximately 10^6 cells/g of tissue with a viability of 88%.26 These isolated cells, when tested *in vitro*, are likely to reflect *in vivo* function although some caution must be exercised in interpretation of the *in vitro* data.38 39 The isolation procedure may deplete the T8+ cell population.31 A mechanical procedure for isolation of intraepithelial lymphocytes12 yields cells with an appearance and function similar to cells collected using chemical methods.26 38 Most of the isolated cells are T cells (E rosette positive, T 11+, t3+), and express the phenotype associated with cytotoxic-suppressor T cells (T8+). Isolated intraepithelial lymphocytes contain few B cells (sIg+, clg+, B1+), macrophages and monocytes (NSE-, M1-), and almost no (<1%) natural killer cells (Leu 7-).26 Approximately 80% of the cells are medium sized lymphocytes and 20% are small sized lymphocytes.12 About 20% of the isolated lymphocytes contain small lysosomal granules which are azurophilic with Giemsa staining.12 26

Isolated intraepithelial lymphocytes have a low rate of spontaneous replication and respond little, or not at all, to a variety of non-specific mitogens.12 26 Addition of autologous macrophages, and addition of interleukin-2 to cultures of intraepithelial lymphocytes and mitogen does not initiate DNA synthesis.26 In contrast, lamina propria lymphocyte and
peripheral blood lymphocytes isolated under similar conditions responded vigorously to such mitogens. They do not mediate natural killer activity, nor do they mediate killer cell activity. Intraepithelial lymphocytes, like lamina propria lymphocyte, can respond to mitogens to be cytotoxic for Chang cells, but in contrast with lamina propria lymphocyte, are not cytotoxic for chicken erythrocytes. Addition of gamma interferon, or of interleukin-2, to the culture medium fails to induce natural killer activity by isolated intraepithelial lymphocytes, nor does addition of antagonists of histamine or prostaglandins.

The intracytoplasmic granules stain with alcian blue (for acidic mucopolysaccharides) and can incorporate radioactive sulphate—all features in common with mast cells. Unlike mast cells, they contain no histamine and their granules are quite dissimilar to mast cell granules seen at electron microscopy. Finally, the mast cell yield in isolated intraepithelial lymphocytes collections is minimal compared with lymphocyte yield.

Intraepithelial lymphocytes have been shown to possess immunoregulatory activity when added to co-cultures of peripheral blood lymphocytes in a pokeweed-mitogen driven immunoglobulin synthesis system. The effect of intraepithelial lymphocytes was the same for both autologous or heterologous assay systems, resulting in enhancement of IgA synthesis and of IgM synthesis. When increasing numbers of intraepithelial lymphocytes were added to peripheral blood lymphocytes, enhancement disappeared and suppression of immunoglobulin synthesis was observed. Similar results have been observed (using lamina propria lymphocytes) when increasing numbers of T cells were added to a fixed number of B cells in a similar assay system, although Elson has shown that the predominant lamina propria response is that of help.

The specific role of intraepithelial lymphocytes remains to be defined. The cells are largely T cells of the suppressor phenotype, and in common with other suppressor effector cells (in contrast with cytotoxic effectors) respond poorly to mitogens. They do not mediate cytotoxic functions in vitro, even though one-fourth of intraepithelial lymphocytes have the morphology of large granular lymphocytes, cells that characteristically mediate natural killer function. Even though intraepithelial lymphocyte granules bear some similarity to mast cell granules, intraepithelial lymphocytes are clearly not mast cells, nor do they appear to be mast cell derivatives. Suppressor T cells are reactive with class I (HLA-A, B, C) antigens, antigens strongly expressed by intestinal epithelium. It is conceivable that intraepithelial lymphocytes play an important role in the local immune response by reacting to epithelial cells altered by viral infection or other foreign antigens. Increased numbers of intraepithelial lymphocytes are present in coeliac sprue, cow’s milk intolerance, parasitic diseases, and graft-versus-host disease. The intestinal immune response is predominantly a helper response, particularly when lamina propria lymphocyte are assessed. It is possible that the epithelial suppressor cells dampen this helper response, particularly when exuberant, although there are no data to support this speculation at present.

Cerf-Bensussan et al have recently shown that isolated rat intraepithelial lymphocytes secreted a factor (probably gamma interferon) capable of inducing Ia antigen (HLA-DR in man) expression by an intestinal
epithelial cell line, IEC 17. Importantly, lymphocyte proliferation was not essential for the secretion of the Ia-inducing factor. These data suggest that intraepithelial lymphocytes may be involved in modulating some epithelial cell functions, and specifically Ia antigen expression. This permits the speculation that Ia antigen positive epithelial cells have the ability to present antigen to T cells. Further, growth of the epithelial cell line was inhibited by supernatants from Con A stimulated intraepithelial lymphocytes, indicating that intraepithelial lymphocytes may modulate epithelial cell growth.

**Origin and traffic of intraepithelial lymphocytes**

Lymphocytes isolated either from efferent lymph of gut associated mesenteric nodes, and returned intravenously after radiolabelling to the same animal will preferentially recirculate through those regions from which they were first isolated. There are considerable data available in laboratory animals (but none in man) that provides a reasonable understanding of the mechanisms of this selective migration, or 'homing', of lymphocytes to gut associated lymphoid tissue (GALT).

Peyer's patches are an enriched (probably exclusive) source of precursor's of IgA - secreting plasma cells found in the intestinal lamina propria while the presence of mesenteric nodes is not required for population of the lamina propria. Antigenic stimulation is not essential to homing, but has a profound effect on the number and distribution of cells in the lamina propria. There is a large increment of plasma cells in the gut at the site of locally applied antigen. Not only do Peyer's patch derived non-recirculating large lymphocytes give rise to plasma cells in the lamina propria but Peyer's patch derived recirculating small lymphocytes also contribute. The increase in plasma cells in the lamina propria after antigenic challenge is because of both recruitment from the circulating lymphocyte pool and proliferation in the lamina propria of these newly recruited cells. Cells obtained from mesenteric lymph nodes also localise preferentially in GALT. Guy-Grand, and colleagues showed that both B cells and T cells take part in preferential homing to GALT, and that antigen does not direct homing in that homing to fetal mucosa (unexposed to antigen) paralleled that to antigen exposed mucosa. Peripheral lymph node blast cells, immunised by an antigen also present in the gut, do not home to the gut. In later studies, Guy-Grand also showed that the presence of T cells in the gut are the result of antigenic stimulation in that such cells were absent in germ free mice, but showed that the gut homing mechanism was independent of antigen in that it was the generation of T blasts within Peyer's patches that conferred the gut homing property.

Only T cells enter the epithelium and these cells have a very low mitotic rate. Husband showed that initial appearance of lymphoblasts in the lamina propria is independent of antigen, but that there was a selective antigen associated accumulation of specific plasma cells at a later stage of the local response. He found no specialised site (vide infra) within the lamina propria for lymphocytes to enter the tissue, and that the migratory lymphocytes accumulated around the crypt region in immunised intestine, a portion of the accumulation being accounted for by proliferation within the lamina propria.
In summary, T cells (and B cells) arising as a result of antigenic stimulation in Peyer's patches populate the gut lamina propria and epithelium. These cells are normally processed both in the spleen and in mesenteric lymph nodes, but neither the spleen nor mesenteric lymph nodes are essential to homing of Peyer's patch derived cells to the gut. Antigen is not essential to homing but greatly expands the accumulation and proliferation of cells within the gut, and determines distribution of homing. There is no evidence that intraepithelial lymphocytes (in contrast with lamina propria lymphocyte proliferate in situ.

The localisation of B cells and T cells to Peyer's patches is mediated by interactions between these cells and the endothelial cells lining post-capillary high endothelial venules (HEV). There is preferential adherence of Peyer's patch lymphocytes to high endothelial venules in Peyer's patches over those in peripheral lymph nodes. Further, there is preferential binding of B cells to Peyer's patch high endothelial venules and of T cells to peripheral node high endothelial venules, while B and T cells bind equally well to mesenteric node high endothelial venules. Finally, T cells of the helper phenotype localise more efficiently in Peyer's patches whereas cells of helper phenotype and of suppressor phenotype localise equivalently in peripheral lymph nodes. The specificity of migration of lymphocytes appears to be determined by selective recognition of organ specific determinants on endothelial cells of high endothelial venules, specialised venules that mediate the exit of migrating lymphocytes from the blood, and which appear to be an important factor in determining the character of local immune responses. Selective lymphocyte entry, however, is probably only one of many factors controlling the distribution of lymphocytes, -- that is, this mechanism is operative in Peyer's patches but is absent in the lamina propria.

The factors that determine the highly exclusive entry of T suppressor cells into the epithelium have not been determined. There is clear morphologic evidence of lymphocyte migration from the lamina propria into the epithelium and back again, and virtually no evidence of extrusion of lymphocytes into the gut lumen. Marsh studied proliferation and migration of intraepithelial lymphocytes in mice after intraperitoneal injections of tritiated thymidine. He found labelled lymphocytes crossing the basal lamina for seven days after injection, that they divided at a rate of 1% per hour, and circulated through the epithelium returning to the lamina propria at a rate of three intraepithelial lymphocytes/1000 epithelial cell nuclei per hour. No intraepithelial lymphocytes were observed adjacent to or extending through tight junctions of epithelial cells. Even though lymphocytes do not enter the lumen from normal epithelium, there is an intriguing and well documented report showing in adult rats that lymphocytes placed in the intestinal lumen can traverse the epithelial layer and gain access to host tissues and produce a graft-vs-host response—that is, they are 'naturally transplanted'. It is unlikely that such an event takes place in adult man, but may play a role in lymphocyte transfer from mother to offspring during lactation (breast milk contains both lymphocytes and macrophages) and nursing. For example, there are reports of the transfer of specific reactivity to tuberculin in human infants breast fed by tuberculin positive mothers.
Intraepithelial lymphocytes are markedly decreased in germ free animals.8 When expressed as a ratio to number of epithelial cells (see previous page), intraepithelial lymphocytes are clearly increased in number in several disease states. They are increased in coeliac sprue, tropical sprue, and in the coeliac sprue lesion found in dermatitis herpetiformis.5 7 30 37 Failure to find a clear increase of intraepithelial lymphocytes in an intestinal lesion otherwise compatible with that of untreated coeliac sprue should cast serious doubt upon the diagnosis.2 Intraepithelial lymphocytes are increased, but not consistently so, in other inflammatory states of the intestine such as in the stasis syndromes.5 Intraepithelial lymphocytes are increased in the small intestine, but not in the colon of subjects with acquired immunodeficiency syndrome, both in the presence and absence of diarrhoea and/or of obvious pathogens.58 Intraepithelial lymphocytes are not increased in inflammatory bowel disease.2 24 31 32

In coeliac sprue, the T4:T8 ratio of intraepithelial lymphocytes is unchanged when compared with normal (0·08 vs 0·19), but the proportion of T8+ intraepithelial lymphocytes expressing Leu 1 (a pan-T cell antigen) is significantly increased when compared with control (56% vs 32%).30 Heterogeneity of Leu 1+ expression was not found in the T8+ lamina propria lymphocyte population. The intraepithelial lymphocytes in coeliac sprue were not activated in that they failed to express HLA-DR and Tac, the latter antigen expressed by T cells after stimulation in culture. Only very rare natural killer (HNK-1+) cells were found.30

Intraepithelial lymphocytes are not increased in number in inflammatory bowel disease (Crohn's colitis, Crohn's ileitis, and ulcerative colitis) nor is there a change in the ratio of T8+ cells to T4+ cells when compared with normal.2 24 31 32 Further, T8+ intraepithelial lymphocytes co-expressed Leu 1+ in a ratio similar to control (one-third positive) and were not activated (HLA-DR−, Tac−).31 Colonic epithelium in active ulcerative colitis and coeliac disease showed variable expression of HLA-DR whereas normal colonic epithelium, non-involved mucosa from diseased specimens, and mucosal epithelium in inactive disease did not.31 32 Both normal and inflammatory bowel disease small intestinal epithelium express HLA-DR.20 59 There is a significant decrease in intraepithelial lymphocytes (when compared with normal ileum) in ileal coeliac disease, the change being largely because of a decrease in T8+ intraepithelial lymphocytes.32 The T4:T8 ratio in ileal coeliac disease was 0·37±0·5, in normal margins of disease tissue, 0·22±1·0, and in control ileum, 0·13±0·1.32 The induction of colonic epithelial cell expression of HLA-DR in inflammatory bowel disease has led to the hypothesis that there is an autoimmune mechanism in inflammatory bowel disease in which the epithelium presents antigen to T4+ intraepithelial lymphocytes and possibly to T4+ lamina propria lymphocyte.60 Both graft-vs-host disease and naturally occurring immunological stimuli can, however, induce the expression of Ia antigen (HLA-DR-like) in rat intestinal epithelium61 indicating that epithelial expression of Ia (DR) is secondary to a variety of immunological stimuli.61 B cells and natural killer cells (Leu 7+) do not enter the epithelium in inflammatory bowel disease.31 32 Chiba et al isolated intraepithelial lymphocytes from diseased specimens of inflammatory bowel disease patients and
found no significant differences in cytotoxic capabilities of disease tissue intraepithelial lymphocytes vs normal tissue intraepithelial lymphocytes.\textsuperscript{38}

In untreated Whipple’s disease, the number of intraepithelial lymphocytes/100 epithelial cells remains in the normal range.\textsuperscript{3, 5} At electron microscopy, Whipple intraepithelial lymphocytes were similar to normal intraepithelial lymphocytes.\textsuperscript{5} Intraepithelial lymphocytes in Whipple’s disease were slightly larger in size, had an increased number of lysosomal granules, and an increased content of ribosomes when compared with normal intraepithelial lymphocytes. There was a clear increase in intraepithelial eosinophils, PMN leucocytes, and macrophages (the latter two cells often containing ingested Whipple bacilli) in untreated Whipple’s disease. Clearly leucocytes other than lymphocytes are involved in the epithelial immune response in Whipple’s disease.\textsuperscript{5} Curiously, there was a four-fold increase in number of intraepithelial lymphocytes in the single patient with Whipple’s disease without intestinal involvement.\textsuperscript{5}

Intraepithelial lymphocytes have not been quantified in human graft-vs-host disease but are clearly increased in murine graft-vs-host disease.\textsuperscript{62} The intraepithelial lymphocytes count rose within 24 hours of induction of graft-vs-host disease while increases in crypt length occurred within three days. The data indicated that measurements of mucosal architecture and intraepithelial lymphocytes counts could be used to quantify mucosal cell mediated immune reactions in appropriate experimental conditions.\textsuperscript{62} Intraepithelial lymphocytes are implicated as effectors in the damage to rectal crypt epithelium in human graft-vs-host disease.\textsuperscript{63} In this morphological study, lymphocytes were shown to be the predominant cell infiltrating the epithelium and to have multiple features of cytolytic cells,—that is, indentation of adjacent epithelial cells by narrow cytoplasmic probes, extension of brood pseudopods to the nuclear membranes of epithelial cells, and protoplasmic surrounding of desmosomes.\textsuperscript{63} Such morphologic features suggested that the lymphocyte-to-epithelial cell contacts represented the recognition phase of alloimmune T-lymphocyte cytolysis.\textsuperscript{63} Epithelial injury and lymphocytic infiltration predominated in the bases of crypts in mild graft-vs-host disease and extended to the surface epithelium in severe graft-vs-host disease.

High intraepithelial lymphocytes counts have been reported in children with giardiasis\textsuperscript{2} and in murine giardiasis.\textsuperscript{2} There are limited data in the murine model that intraluminal lymphocytes may promote clearance of giardia.\textsuperscript{64} Finally, we have morphological evidence that intraepithelial lymphocytes are involved in the immune response to microsporidial invasion of intestinal epithelial cells in AIDS.\textsuperscript{65} There is a clear increase in intraepithelial lymphocytes number and many of these intraepithelial lymphocytes contain remnants of microsporidial spores and trophozoites. There are little other data\textsuperscript{2} in regard to intraepithelial lymphocyte role in response to intestinal parasites although it is likely that intraepithelial lymphocytes are actively involved in the immune response to parasites.

Of mice and men

Throughout this paper, frequent reference has been made to intraepithelial
Human intestinal intraepithelial lymphocytes

lymphocytes morphology and function in rodents and other small laboratory animals while the major emphasis has been upon the human intraepithelial lymphocytes. The Table compares the major features of human and rodent intraepithelial lymphocytes, features derived from the references cited above and from several other important publications concerning rodent intraepithelial lymphocytes. The most notable distinction between the intraepithelial lymphocytes of the two species is that rodent intraepithelial lymphocytes are more likely to be granulated and are capable of mediating all cytotoxic functions while human intraepithelial lymphocytes are exceedingly limited in their cytotoxic capabilities. It is suspected that the human intraepithelial lymphocytes will be shown to have cytotoxic capabilities similar to those of the rodent when appropriate experimental conditions are devised.

Conclusion

Intraepithelial lymphocytes are a unique set of T cells that are predominantly of the suppressor phenotype and which have a varied expression of other T cell antigens, at least in coeliac sprue. Intraepithelial lymphocytes are not activated as determined by expression of HLA-DR antigen and Tac antigen and do not respond to non-specific mitogens. They do not have natural killer phenotype or function, although they resemble natural killer cells in that they are often large granular lymphocytes. Intraepithelial lymphocytes in rodents have some similarity to mast cells, but there is little evidence in man that the intraepithelial lymphocytes is related to mast cells. Intraepithelial lymphocytes may play an immuno-regulatory role in man, possibly by suppressing the systemic immune response to antigens that simultaneously promote a mucosal (lamina propria) helper immune response. The epithelium is ideally suited to the function of suppressor cells because of its strong expression of class I major histocompatibility complex antigens. The small population of helper cells within the epithelium may add further control over the functions of suppressor intraepithelial lymphocytes, especially in inflammatory condi-

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ND - not determined, MICC - mitogen induced cellular cytotoxicity, ADCC - antibody dependent cellular cytotoxicity killer (K) cell function, SCMC - spontaneous cell mediated cytotoxicity natural killer (NK) function, MLR - mixed lymphocyte response, MLC - mixed lymphocyte cytotoxicity.

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tions of the colon when the colonic epithelium is induced to express Class II major histocompatibility complex antigens.\(^{20,31,32}\)

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