MODULATION OF LYMPHOCYTE SUBSETS IN PEYER'S PATCHES OF MICE TREATED WITH MONOCLONAL

25 ANTIBODY AGAINST HELPER T CELLS. T.H. Ermak, R.L. Owen, and M.F. Heyworth. UCSF and VAMC, San Francisco, CA 94121

Treatment of mice with anti-L3T4, a monoclonal antibody directed against the helper T cell $[T_h]$, prevents clearance of intestinal <u>Giardia muris</u> infection. This study examines the effect of anti-L3T4 treatment on the distribution of T and B cells within Peyer's patches. Female BALB/c mice aged 8 wks were given 7 weekly injections of either anti-L3T4 (1 mg/wk) or phosphatebuffered saline. Cryostat serial sections of Feyer's patches frozen at the end of treatment were incubated with biotinylated monoclonal antibodies reacting with B220 (B cells), Thy-1.2 (all T cells), L3T4, or Lyt-2 (cytotoxic/suppressor T cells $[T_{c/s}]$), followed by avidin-biotin-peroxidase complexes (ABC) and cobalt-In anti-L3T4 treated mice, Peyer's patch DAB. follicles (B-cell areas) were reduced in size. In untreated mice, T_h cells had roughly the same distribution as all T cells, i.e., they were abundant in interfollicular areas, and scattered below the dome epithelium, in follicles, and in germinal centers. $T_{c/s}$ cells were restricted largely to interfollicular areas. In anti-L3T4 treated mice, Peyer's patches were depleted of T_h cells yet the distribution of all T cells was still similar to that in untreated mice. cells were now not only numerous in inter-T_{c/s} cells were now not only numerous in another follicular areas, but also scattered below the dome epithelium, in follicles, and in germinal centers. Thus, $T_{c/8}$ cells had redistributed into areas depleted of T_h cells. These results indicate that treatment with anti-L3T4 monoclonal antibody eliminates helper T cells, secondarily increases the number of $\rm T_{c/s}$ cells in follicles, interfollicular areas, and antigen sampling regions of Peyer's patches, and indirectly decreases the size of B-cell areas.

Immunohistochemical demonstration of lymphocyte markers in formalin-fixed and paraffin-embedded specimens. Y. Tsutsumi T. Ogata & K. Kawai; Dept. of Pathol., Tokai Univ. Sch. of Med. & Div. of Diag.

Pathol., Tokai Univ. Hosp., Isehara 259-11 JAPAN The present study describes immunchistochemi-cal localization of lymphocyte markers in routinely prepared sections with commercial mouse monoclonal antibodies to leucocyte common anti-gen (LCA, DAKO, x10), T or B cell (Bioscience, x10) and LeuMl (Becton-Dickinson, x100). In all, 196 samples were examined by indirect immu-noperoxidase method: These include 155 non-Hodgkin lymphomas (NHLs), 13 Hodgkin lymphomas (HLs), 26 myelomas and 2 myeloid leukemias (MLs) as well as reactive lymph nodes and thymuses. In nontumorous tissues, LCA was positive in most lymphocytes while plasma cells and granulocytes were negative. T cell antigen was localized in paracortical lymphocytes, thymocytes, granulocytes and epithelioid cells. B cell antiger was mainly seen in lymphoid follicles while B cell antigen plasma cells were unreactive. LeuMl was present in granulocytes and some epithelial cells. Results of NHLs were as follows: LCA+T+B+ 6 cases, LCA+T+B- 40 cases, LCA+T-B+ 44 cases, LCA+T-B- 37 cases, LCA-T+B- 8 cases, LCA-T-B- 20 cases and LeuMl+ 6 cases. Admixture of small T and B cells was characteristic of HLs and 6 cases were LeuMl+ in Hodgkin cells. Myelomas were B-LeuMland 5 T+ and 3 LCA+ cases were noted. MLs were The following problems are LCA+T+B-LeuM]+. pointed out: 1) LCA negative NHLs are not rarely encountered. 2) Some B cell tumors are unreac-tive to anti-B cell. 3) Anti-T cell recognizes non-T cells such as granulocytes and some B cell tumors. 4) LeuMl negative Hodkin cells and LeuMl positive NHL cells are occasionally seen.

Immunocytochemical Studies on the Structure of Human Peyer's Patches and Solitary Lymphoid Follicles

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The present investigation is aimed to provide the information on immunohistochemical characteristics of lymphoid cells and vascular endothelial cells in Peyer's patches and solitary lymphoid follicles of the human intestine, and discuss the role of the vascular endothelial cells on the differentiation, proliferation and migration mechanism of the patch cells.

The cells in the germinal center had IgM and J chain in the cytoplasm, and occasilnally IgM or IgA on the surface. Considerable amounts of helper/inducer T cells, Leu 7-positive cells and C3b receptor-, HLA-DR-positive dendritic cells also coexisted, sharing a function for B cell proliferation. The major population of the cell in the T cell zone were helper/inducer T cells, and some expressed IL-2 receptor. Several S100positive dendritic cells were intermingled. Post-capillary venules found in the interfollicular T cell zone shared antigens with peripheral blood monocyte and macrophage subset, capable of presenting soluble antigens and triggering autologous mixed lymphocyte reaction, associated with OK-M5 and HLA-DR, but not OK-M1. This suggests that the germinal center is

This suggests that the germinal center is the site of immune responses and class switching from IgM to IgA of the pach B cells, and the surface antigens on the endothelium could be the factors responsible for the regulation of the lymphocyte passage through the venules and the triggering the lymphocyte maturation, activation and proliferation during the migration.

> Monocyte/Macrophage Antigens in Human Carcinomas. R. Giorno, Allergy and Clinical Immunology, University of Colorado School of Medicine, Denver, CO 80262.

A panel of eight antibodies was used to characterize cells of monocyte/macrophage lineage in frozen sections of ten cases each of moderately differentiated carcinomas of the colon and breast. The antibodies used were LeuM1 (granulocyte, some monocyte), LeuM2 (monocyte, endothelium), LeuM3 (macrophage, follicular dendritic cell, Langerhans cell), LeuM5 (macrophage, interdigitating cell), R4/23 (follicular dendritic cell), Leu6 (Langerhans cell, thymocyte), Leu3a+b (macrophage, Langerhans cell, helper-inducer T cell) and anti-S100 (macrophage, Langerhans cell, interdigitating cell). Frozen sections were incubated with the antibodies which were localized by an avidin-biotin immunoperoxidase method (Giorno, Histochemistry 81:505, 1984). The results indicate that tumors contain two populations of cells of monocyte/macrophage lineage_which can be distinguished by their immunoreactivity. The predominant popu-lation of cells is LeuM3⁺LeuM5⁺Leu3⁺S100⁺, a phenotype consistent with macrophages (Giorno, Histochemistry, in press). These cells surround nests of tumor cells or are localized in adjacent stroma. A second, minor population of cells is Leu6 5100 and is found in close apposition to tumor cells. These cells have the phenotype characteristic of Langerhans cells, which are known to act as antigen presenting cells. An additional finding is that in most cases the malignant epithelial cells are LeuM1⁺LeuM2⁺. Most cells of monocyte/macrophage lineage found in human tumors are macrophages, with a minor popu-lation of Langerhans cells, which may be sufficient to present antigen to the large number of T cells which infiltrate most tumors (Giorno, Arch. Pathol. 107:415 1983). The results further indicate the limited useful-ness of some antibodies, such as LeuM1 and LeuM2 in determining the lineage of certain cells, due to their widespread immunoreactivity with cells of diverse lineage.