Factor (implicated in von Willebrand disease, a hemorrhagic diathesis) also strongly stained endothelium in all tissue types, whereas ICAM-1 (ligand for CD11 and CD18) was slightly less prevalent. The strong staining of EP2 and EP4 suggests that these receptors may play a role in Cox-2's influence in colon cancer, and may imitate the expression patterns of CD34 and/or Von Willebrand Factor, which have known pathologies in other disease states. Further investigation may reveal distinct expression patterns of the PGE2 receptors by endothelial cells in ulcerative colitis or cancer and may help elucidate the role of Cox-2 in cancer initiation and progression.

P4-11

Connexin 43 expression of the intestinal macrophage in patients with Crohn's disease

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Background: The pathogenesis of Crohn's disease (CD) involves excessive activation of macrophages and the presence of microaggregate of macrophages within the lamina propria is a characteristic finding of CD. The gap junction is known to be formed between bone marrow-derived macrophages aggregated in vitro, and proposed to be associated with the development of various pathophysiological conditions. Aberrant macrophage expressions of some adhesion molecules are considered to be involved in the pathogenesis of the disease. However, a role of gap junctional formation for macrophage microaggregation in CD remains unclear. The aim of this study is to clarify whether Connexin 43 (Cx43), a component of gap junction, is expressed on intestinal macrophages and is implicated in the microaggregate of macrophages in CD.
Methods: Biopsied specimens of the large intestine were obtained from active CD (n=6), and from intact sites of patients with colorectal polyp (n=6; normal controls). Using specific antibodies against Cx43 and macrophage marker CD68 (KP1), correlation between localization of Cx43 and distribution of macrophages was investigated by double immunofluorescence staining. Serial sections of the tissues were individually stained with CD68 and Cx43 to confirm the presence of CD68 and Cx43 double positive cells. We also determined the level of Cx43 mRNA expression using RT-PCR.
Results: In normal controls, only a few macrophages stained with CD68 were observed within the lamina propria. Those cells were rarely positive for Cx43. In contrast, Cx43-positive macrophages were abundantly observed within the lamina propria, especially lower half of the mucosa, in CD. In addition, Cx43 expression was detected in aggregated macrophages as well as isolated macrophages. The ratio of Cx43-positive cells against CD68-positive cells was higher in CD than in normal controls. Cx43 mRNA was substantially detected in 4 of 6 samples from CD patients and slightly detected 2 of 6 samples from normal controls. The level of Cx43 mRNA expression in patients with CD was correlated with the severity of the disease determined by endoscopy.
Conclusion: These results suggest that Cx43 is expressed on macrophages within the lamina propria of large intestine. Cx43 might be involved in the macrophage aggregation observed in patients with CD and contribute to the pathogenesis of the disease.

P4-12

Immunohistochemical study of gamma-butyrate in colonic neoplasmas

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Objective: The expression of gamma-aminobutyrate (GABA) is elevated in human colorectal carcinoma tissues compared to adjacent normal mucosa. In this study, we investigated the relation between degree of atypia and the expression level of GABA in colonic intramucosal neoplasmas.
Methods: Fifty-six endoscopically resected human colon neoplasmas were classified into 1. adenocarcinoma (AC), 2. adenoma with high-grade atypia (AH), 3. adenoma with low-grade atypia (AL) according to modified Ajioka fs pathological classification. Expression of GABA was examined by indirect immunofluorescent technique and the results were compared with staining patterns of carcinoembryonic antigen (CEA) and cancer-associated antigen (CA)19-9. The staining patterns of CEA and CA19-9 were classified into three categories (pattern1, 2, and 3) according to modified Taguchi fs classification.
Results: Strong GABA immunoreactivity was observed in 73.7% of AC, 54.6% of AH, 13.3% of AL, and 5.4% of adjacent normal mucosa (NM). Kendall fs correlation coefficient between GABA immunoreactivity and degree of atypia was 0.447. Strongly positive CEA staining (pattern 3) was observed in 57.9% of AC, 36.3% of AH, 13.3% of AL, and 0% of NM. Pattern 3 in CA19-9 staining was observed in 26.3% of AC, 9.1% of AH, and 0% of AL and NM. In AC and AH, the rate of strongly GABA positive glands was greater than that of pattern 3 staining for CA19-9 ($p<0.05$).
Conclusion: Staining of GABA in addition to CEA and CA19-9 is useful to diagnose the malignant potential of colonic intramucosal neoplasmas.

P4-13

Correlation between Immunohistochemical Cell Kinetics and Macroscopic Phenotype of Colorectal Serrated Adenoma

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(Aim and Background) We investigated the correlation between cell kinetics of colorectal serrated adenoma and its histopathological parameters including macroscopic phenotype.
(Materials and Methods) Samples were taken from 24 patients with colorectal serrated adenoma, and the followings were examined.
(1) Distribution of cell proliferation in colorectal serrated adenoma by immunohistochemical staining using monoclonal antibody against Ki-67 antigen (Immunotech Marseille, France)
(2) Histopathological parameters of colorectal serrated adenoma; the size of the lesion, the mean length of the ducts and the incidence of macroscopic phenotype
(3) Correlation between (1) and (2)
(Results) The distribution of cell proliferation of colorectal serrated adenoma was classified into three patterns; (1) Decreasing pattern showing a gradual decrease of Ki-67 labeling index (Ki-67 LI) from the base to the superficial layer (9 cases). (2) Wave pattern showing alternating increases and decreases of Ki-67 LI with a final decrease, from the base to the superficial layer. Ki-67 briefly LI decreases from the base to the superficial layer but increases intermittently (12 cases). (3) Convex pattern showing an increase of Ki-67 LI from the base to the middle of the ducts and then a decrease toward the superficial layer, that is, Ki-67 positive cells are recognized primarily in the middle of the ducts (3 cases). Size (mm), length (μm) and incidence

of macroscopic phenotype for each distribution pattern of cell proliferation were the followings: Decreasing pattern: 10.6 mm, 887 μm, protruding: 33.3%, sessile: 22.2%, LST: 11.2%. Wave pattern: 10.3 mm, 1110 μm, protruding: 63.6%, sessile: 9.1%. Convex pattern: 9.5 mm, 731 μm, protruding: 33.3%, sessile: 66.7%.
(Discussion) Immunohistochemical cell kinetics of colorectal serrated adenoma correlated with its macroscopic phenotypes as follows:
(1) Wave pattern was correlated with the protruding type.
(2) Convex pattern was correlated with the sessile type which have ducts with short length.
(Conclusion)
(1) Colorectal serrated adenoma showing Decreasing and Wave pattern may originate from hyperplastic polyp. And changes of cell proliferation pattern, Decreasing to the Wave pattern, could occur in the histogenesis of colorectal serrated adenoma.
(2) Serrated adenomas showing Convex pattern may originate de novo.

P4-14

Apoptotic imbalance of infiltrating lymphocytes between tumor and non-tumor tissue in the development of colorectal cancer

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Immunological cells play an important role in neoplastic development. Among immunological cells, lymphocytes exert a diversity of mechanisms against tumor growth. In solid tumors, the presence of tumor infiltrating lymphocytes, natural killer cells and macrophages, provides an effective antitumor response. The number of tumor infiltrating lymphocytes in resected tumor specimens negatively correlates with the size of tumor but positively with survival. The mechanism leading to the dysfunction of infiltrating lymphocytes in the tumor remains unclear. The aim of the study was to investigate whether apoptosis contributed to the loss of infiltrating lymphocytes and the inactivation of their antitumor functions in colorectal cancer. We first demonstrated a significant increase in apoptosis in infiltrating lymphocytes of colorectal cancer tissues, compared to those in non-cancerous tissues from the same patient. Furthermore, our result suggested that disturbance of apoptosis resulted from an imbalance of decreased antiapoptotic molecules and increased proapoptotic ones, reflected by reduction of Bcl-2 level and elevation of Bax level. The shift of balance of Bcl-2 and Bax in favor of the latter accelerated the activity of caspase-3, as Bcl-2 expression led to functional inhibition of caspase-3 while Bax promoted caspase-3 activity. Therefore, caspase-3 could be considered as an executer of apoptosis in Bcl-2 and Bax pathways. In line with the role of caspase-3 in apoptosis caused by a decrease in Bcl-2 and an increase in Bax, the expression of caspase-3 was found to be significantly elevated in infiltrating mononuclear cells of colorectal cancer but not those in non-cancerous tissues. The apoptosis or the activity of caspase-3 may also be associated with inducible nitric oxide synthase (iNOS), whose level increased in the present study. The inhibition of nitric oxide production may revoke immunosuppression and benefit progressively growing malignant tumors. Therefore, increase in iNOS expression in infiltrating mononuclear cells is thought to down-regulate the immune response against colorectal cancer cells and thus may contribute to the loss of lymphocyte functions in the tumor. In conclusion, this study reveals that apoptotic imbalance occurs in infiltrating lymphocytes between tumor and non-tumor tissues. This imbalance may attenuate the function of infiltrating lymphocytes in the tumor tissue and thus contribute to the development of colorectal cancer.

P4-15

Shift from high H6PDH to high G6PDH activity during carcinogenesis induced by environmental pollutants in flatfish liver

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During the past 10 years, we found frequencies of liver cancers up to 34% in flatfish (Platichthys flesus) living in highly polluted areas due to industrial and agricultural effluents. Unfortunately, the class of cancer-initiating and -promoting compounds cannot be identified because over 100 000 different chemicals are released into the sea. The pollution-induced liver lesions and progression from early eosinophilic foci to persistent basophilic foci (FAH) and hepatocellular carcinoma (HCC) show similarities with lesions in chemically-induced liver cancer in mammals. High levels of glucose-6-phosphate dehydrogenase (G6PDH) activity are the most sensitive marker at present for the detection of early preneoplastic foci of altered hepatocytes that are larger than 20 cells (Köhler and Van Noorden (1998). Aquat Toxicol 40: 233–252). G6PDH as the key enzyme of the pentose phosphate pathway is the main source of NADPH in cells and produces riboses for biosynthesis. However, we found recently that the NADPH-producing hexose-6-phosphate dehydrogenase (H6PDH) activity is upregulated in single hepatocytes and microfoci (approx. 1–5 cells). H6PDH activity was elevated in 82% of single altered hepatocytes or microfoci whereas G6PDH activity was not yet altered as compared with extrafocal liver tissue. During cell proliferation and growth of foci, G6PDH activity increased and 67% of (pre)neoplastic lesions, 15.5% adenomas and 18.2% carcinomas showed increased G6PDH activity. H6PDH activity appeared to decrease during cancer progression and was detectable only in 13% of foci, 3.6% adenomas and 1% carcinomas. H6PDH is closely linked to the polycyclic hydrocarbon metabolising system in the smooth endoplasmic reticulum and is inducible by phenobarbital. We hypothesize that environmental chemicals which initiate single hepatocytes to proliferate belong to the phenobarbital type of inducers (PCB congeners 153, 180, 194, HCB, OCS, HCH-isomers; Safe (1994). CRC Crit. Rev. Toxicol. 24: 87–149) which are also found in highest concentrations in flatfish liver.

P4-16

A Case of Clear Cell Hepatocellular Carcinoma

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A case of clear cell hepatocellular carcinoma, a rare malignancy in the liver, in a 53 years old male is documented with immunohistochemical study. The tumor was measured in 8.5×7.0×6.5 cm in the left lobe of the liver with two small daughter lesions. Microscopically, these lesions consisted of uniform large tumor cells with clear cytoplasm containing abundant glycogen. The histological features had a striking resemblance to clear cell tumors that originated in other organs, especially in kidney. The diagnosis was confirmed by focal presence of non-clear tumor cells usually seen in the hepatocellular carcinoma, occasional Mallory bodies and no tumorous lesion in other organs. A brief review of the literature about the prognosis was also done.