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ABSTRACT

Background/aim: 5-Hydroxytryptamine (5-HT) released from enterochromaffin cells influences intestinal homeostasis by altering gut physiology and is implicated in the pathophysiology of various gut disorders. The mechanisms regulating 5-HT production in the gut remain unclear. This study investigated the T helper (Th) 1/Th2-based immunoregulation of enterochromaffin cell function and 5-HT production in a model of enteric infection.

Methods and results: Trichuris muris-infected AKR (susceptible to infection and generates Th1 response), BALB/c (resistant to infection and generates Th2 response), Stat4−/− (impaired in Th1 response) and Stat6−/− (impaired in Th2 response) mice were investigated to assess enterochromaffin cells, 5-HT and cytokines. In association with the generation of a Th2 response we observed higher enterochromaffin cell numbers and 5-HT content in the colon of BALB/c mice compared with AKR mice. Numbers of enterochromaffin cells and amount of 5-HT were significantly lower in Stat6−/− mice after infection compared with Stat4−/− mice. In addition, enterochromaffin cell numbers and 5-HT content were significantly higher after reconstitution of severe combined immunodeficient mice with in-vitro polarised Th2 cells.

Conclusion: The study demonstrated that enterochromaffin cell and 5-HT responses to the same infectious agent are influenced by Th1 or Th2 cytokine predominance and suggests that the immunological profile of the inflammatory response is important in the regulation of enterochromaffin cell biology in the gut. In addition to new data on enterochromaffin cell function in enteric infection and inflammation, this study provides important information on the immuno–endocrine axis in the gut, which may ultimately lead to improved strategies against gut disorders.

Enteric endocrine cells are dispersed throughout the gastrointestinal (GI) mucosa. The best characterised subset of enteric endocrine cells is the enterochromaffin cell, which is the main source of the biogenic amine serotonin (5-hydroxytryptamine, 5-HT) in the GI tract. 5-HT influences intestinal homeostasis by altering gut physiology (motility and secretory function) and has been implicated in the pathophysiology of various GI disorders, which include inflammatory bowel disease and functional disorders such as irritable bowel syndrome (IBS). In addition, alteration in enterochromaffin cells is also observed in a number of bacterial, viral and parasitic infections of the GI tract. 5-HT is released from enterochromaffin cells into the blood, into the surrounding tissue and into the gut lumen in a regulated and calcium-dependent manner in response to various mechanical and chemical stimuli including bacterial toxins and participates in gut physiology. The association between alteration in enterochromaffin cells/5-HT production and various GI diseases greatly emphasises the significance of 5-HT in intestinal homeostasis. The precise mechanisms regulating the alteration of enterochromaffin cell function and 5-HT production in the gut during infection and inflammation remain to be determined.

Considering the strategic location of enterochromaffin cells in the GI mucosa, changes in 5-HT signalling and secretory motor function in enteric infection and inflammation are likely to be modulated by the immune system, and we have recently shown evidence that CD4 T cells play an important role in the development of enterochromaffin cell hyperplasia and in the upregulation of 5-HT production in the colon in intestinal nematode infection. There is also evidence of low-grade inflammation and/or immune activation (an increase in CD3-positive T cells) in patients with postinfectious IBS in whom increased numbers of enterochromaffin cells have been reported and this is reflected in an animal model of postinfectious IBS. It is therefore very likely that immune activation may increase enterochromaffin cell numbers in the clinically relevant context of functional bowel disease such as IBS. The reduced number of enterochromaffin cells in the colon of mice with targeted disruption of IL2 and T-cell receptor alpha and the presence of enterochromaffin cells in contact with, or in very close proximity to, lymphocytes further support the presence of an immunological control of enterochromaffin cell function and 5-HT production in the GI tract.

The interaction between each cytokine and its receptor leads to activation of signalling molecules including signal transducer and activator of transcription (Stat) proteins. Stat6 is activated by both IL4 and IL13 and is essential in T helper (Th) 2 development, whereas Stat4 is activated by IL12 and is important for a Th1 type response. In our previous studies using infection with the small intestinal nematode Trichinella spiralis, we have demonstrated that Th2 cytokines, IL4 and IL13, acting via Stat6, play an essential role in the alteration of intestinal physiology and host defence in this infection. We have also shown that a shift to a Th1 response by gene transfer and over-
expression of IL12 (a critical cytokine in the development of Th1 immune response) significantly altered intestinal physiology and inhibited host protective immunity in this Th2-biased enteric infection. These observations, along with our recent findings on CD4 T-cell-regulated control of 5-HT production in the gut, show an immunological basis for an alteration in gut physiology and prompted further investigations on the precise mechanisms of alteration in enterochromaffin cell function and 5-HT production in the gut.

In a colonic parasitic infection with *Trichuris muris*, resistant strains (BALB/c, C57BL/6, NIH) expel the parasites through the generation of a Th2 response, whereas susceptible strains (AKR, BALB/c, C57BL/6, NIH) expel the parasites through mechanisms of alteration in enterochromaffin cell function and physiology and prompted further investigations on the precise mechanisms of alteration in enterochromaffin cells and 5-HT production in the gut.

Materials and Methods

Animals

Male BALB/c and AKR mice were obtained from Harlan (Indianapolis, Indiana, USA) and C57BL/6 and severe combined immunodeficient (SCID; C57BL/6 background) mice were obtained from Jackson Laboratories (Bar Harbor, Maine, USA). Stat4-deficient (Stat4/−) and Stat6-deficient (Stat6/−) mice on C57BL/6 background were generated as described previously. Breeding pairs of Stat6/− and Stat4/− mice and their wild-type littermates were obtained from the John Curtin School of Medical Research, Australian National University, Canberra, Australia and Indiana University School of Medicine, Indianapolis, Indiana, USA, respectively. All mice were kept in sterilised, filter-topped cages and fed autoclaved food in the animal facilities of McMaster University. Only 8–10-week-old male mice were used. The protocols employed were in direct accordance with guidelines drafted by the McMaster University Animal Care Committee and the Canadian Council on the Use of Laboratory Animals.

Infection Technique

The techniques used for *T. muris* maintenance and infection were described previously. Mice were infected with approximately 300 eggs and worm burdens were assessed as described previously.

Immunohistochemistry

Immunohistochemical studies on 5-HT-expressing enterochromaffin cells and CD3-positive cells were performed on formalin-fixed, paraffin-embedded samples. Sections were deparaffinised in CitriSolv (Fisher Scientific, Ontario, Canada) and rehydrated through a graded series of ethanol and phosphate-buffered saline (PBS). Endogenous peroxidase was blocked by incubation in peroxidase-blocking reagent (DakoCytomation, Ontario, Canada) for 15 minutes. After washing, sections were subjected to antigen retrieval in citrate buffer after heating in a microwave for one minute. After blocking of non-specific binding with 1% bovine serum albumin in PBS, the sections were incubated with rabbit anti-5-HT antibody (ImmuNoStar, Inc, Hudson, Wisconsin, USA; 1:5000 for 1 h at room temperature) or with polyclonal rabbit anti-CD3 antibody (DakoCytomation Inc; 1:500 for 1 h at room temperature). After washing, sections were incubated with Envision (horseradish peroxidase-coupled anti-rabbit secondary reagent; DakoCytomation) for 30 minutes. The sections were developed with 3,3’-diaminobenzidine and counterstained with Meyer’s haematoxylin. The numbers of CD3-positive and 5-HT-expressing enterochromaffin cells were counted by an investigator blinded to conditions and the numbers of CD3-positive and enterochromaffin cells were expressed per high power field and per 10 glands, respectively.

Determination of colonic 5-HT content

Segments of colon were homogenised in 0.5 ml 0.2 mol perchloric acid and centrifuged at 10 000g for five minutes. The supernatants were neutralised with 0.5 ml 1.0 mol borate buffer (pH 9.25) and centrifuged at 10 000g for one minute. The 5-HT content in the supernatant was analyzed by enzyme immunoassay with a commercially available kit (Beckman Coulter, California, USA). The 5-HT content of the tissue was expressed as a function of wet weight (in mg).

Measurement of cytokines in colonic tissues

Frozen colonic tissues were homogenised in lysis buffer containing protease inhibitor cocktail (Sigma, St Louis, Missouri, USA). The homogenates were freeze-thawed three times and centrifuged and the supernatants were collected and stored at −20°C until analyzed.

The homogenised colonic samples were analyzed using a mouse IL13 and IFN-γ ELISA kit (R&D Systems, Minneapolis, Minnesota, USA) according to the manufacturer’s instructions. Results are corrected for protein concentration, which was measured by DC Protein Assay kit (Bio-Rad Laboratories, Hercules, California, USA).

Evaluation of in-vitro cytokine production from splenocytes

Single-cell suspensions of spleen were prepared in RPMI 1640 containing 10% fetal calf serum, 5 mmol L-glutamine, 100 U/ml penicillin, 100 μg/ml streptomycin, 25 mmol Hepes, 0.05 mmol 2-ME (all Gibco BRL, Gaithersburg, Maryland, USA). Cells (107) were incubated in the presence of 5 μg/ml concanavalin A (Con A). IL4 and IL13 levels in the supernatant were measured by enzyme immunoassay using a commercially available kit (R&D Systems; Minneapolis, Minnesota, USA).

In-vitro Th1 and Th2 cell polarisation

Splenocytes from non-infected C57BL/6 mice were collected in Hank’s balanced salt solution containing 10% fetal bovine serum and 1% antibiotic/antimycotic. CD4 T cells were isolated from the mixed splenocytes by negative selection using EasySep mouse CD4 T-cell enrichment cocktail with magnetic nanoparticles (Stem Cell Technologies, Vancouver, BC, Canada). Cell purity, as determined by flow cytometry using antimouse CD4 (LT34) monoclonal antibody (BD Pharmingen, California, USA), was 90%. CD4 T cells (2.5 × 106 cells/ml) were resuspended in RPMI 1640 containing 10% fetal bovine serum and 1% antibiotic/antimycotic and activated with plate-bound 10 μg/ml anti-CD3 and 2 μg/ml anti-CD28 for 24 h. After 24 h, cells were incubated with 5 ng/ml recombinant IL12, 10 μg/ml anti-IL4 and 50 U/ml rIL2 for Th1 differentiation, whereas for Th2 differentiation cells were incubated with 10 ng/ml rIL4 and 10 μg/ml anti-IL12 in the presence of 50 U/ml of rIL2. On day 7, cells were harvested, washed and re-stimulated with
Figure 1  Resistant BALB/c mice expelled most worms by day 21 postinfection with the generation of a T helper (Th) 2 type immune response, whereas susceptible AKR mice failed to clear the infection and generate a Th1 type immune response. Both BALB/c and AKR mice were infected by mouth with 300 eggs of T muris and were killed to investigate the numbers of worms and cytokines (IFN-γ and IL13 were used as candidate Th1 and Th2 cytokines, respectively). (A) worm burden on day 21 postinfection; (B) IL13 levels in colonic tissues and (C) IFN-γ levels in colonic tissues. Day 0 indicates non-infected control mice. *Significant difference (p<0.05) between BALB/c and AKR mice. Each bar represents mean ± SEM from five mice.

plate-bound 10 μg/ml anti-CD3 and 2 μg/ml anti-CD28 for 24 h. After 24 h supernatants were collected and frozen for analyses of cytokines, the cells were harvested and re-suspended in PBS and each SCID mouse received 2.5 × 10^6 cells intraperitoneally.

Statistical analysis
Data were analyzed using unpaired Student’s t test, one-way analysis of variance with Dunnett’s post-hoc test for comparisons with a control, two-way one-way analysis of variance followed by Tukey’s test and Pearson’s correlation analysis as appropriate, when p<0.05 was considered significant. All results are expressed as the mean ± SEM.

RESULTS
Numbers of enterochromaffin cells and 5-HT content in the colon differ in resistant and susceptible mice after T muris infection
BALB/c mice are resistant to T muris infection and expelled almost all worms by day 21 postinfection, whereas AKR mice failed to clear the infection and we recovered substantial numbers of worms from AKR mice on day 21 postinfection (fig 1A). This was associated with significantly higher levels of IL13 in colonic tissues of BALB/c mice compared with AKR mice (fig 1B), whereas tissue IFN-γ levels were significantly higher in AKR mice compared with those in BALB/c mice (fig 1C). We also observed significantly higher levels of IL4 and IL13 from in-vitro ConA-stimulated spleen cells from resistant BALB/c mice (fig 2).

Th1 and Th2 environments in AKR and BALB/c mice, respectively, in T muris infection had a significant influence on enterochromaffin cells and 5-HT content in the colon. We observed significantly higher numbers of enterochromaffin cells in the colon of BALB/c mice compared with those in AKR mice after infection (fig 3A). Consistent with increased enterochromaffin cell numbers, we also observed significantly higher amounts of 5-HT in the colon of BALB/c mice compared with those in AKR mice after infection (fig 3B). There were significant correlations between the ability to expel the parasite (as measured by worm burden) and both the enterochromaffin cell numbers and amount of 5-HT (Pearson’s correlation coefficient −0.93 and −0.80, respectively; p<0.0001 and p<0.01, respectively).

Figure 2  IL4 and IL13 levels were significantly higher in the supernatant of in-vitro concanavalin A (conA)-stimulated spleen cells of infected BALB/c mice compared with those of infected AKR mice. BALB/c and AKR mice were infected by mouth with 300 eggs of T muris and were killed on day 14 postinfection to investigate the cytokine response in spleen cells. Spleen cells were stimulated with ConA for 24 h and the levels of IL4 (A) and IL13 (B) present in the supernatant were investigated by ELISA. *Significantly (p<0.05) higher in BALB/c mice compared with AKR mice. Each value (pg/ml) represents the mean ± SEM from four mice.

Numbers of enterochromaffin cells and 5-HT content in colon differ in Stat4−/− and Stat6−/− mice after T muris infection
We next examined enterochromaffin cells and 5-HT content in the colon of Stat4−/− and Stat6−/− mice, which are impaired in Th1 and Th2 responses, respectively. As shown in fig 4A the enterochromaffin cell numbers were significantly lower in Stat6−/− mice compared with those in Stat4−/− mice after infection with T muris. We also observed a significantly lower amount of 5-HT in colon tissues of Stat6−/− mice compared with Stat4−/− mice after infection (fig 4B). The reduction in enterochromaffin cell numbers and amount of 5-HT after infection was associated with significant impairment in Th2 cytokines in the colon of Stat6−/− mice. In contrast, the higher enterochromaffin cell numbers and amount of 5-HT in Stat4−/− mice after infection was associated with an upregulation of Th2 cytokines in the colon of Stat6−/− mice. We also observed a significantly lower amount of 5-HT in Stat4−/− mice compared with Stat6−/− mice, with wild-type control mice, Stat4−/− mice and Stat6−/− mice, respectively; IL15 levels (in pg/mg protein): 0 (SD 0), 61.2 (SD 13.6) and 0 (SD 0), in wild-type control mice, Stat4−/− mice and Stat6−/− mice, respectively, on
Figure 3  BALB/c mice exhibited higher numbers of enterochromaffin (EC) cells and 5-hydroxytryptamine (5-HT) compared with those in AKR mice after T. muris infection. BALB/c and AKR mice were infected by mouth with 300 eggs of T. muris and were killed on different days postinfection to investigate enterochromaffin cells and 5-HT in the colon. (A) Enterochromaffin cell numbers in the colon of BALB/c and AKR mice. (B) Amount of 5-HT in the colon of BALB/c and AKR mice. (C) Representative micrograph showing enterochromaffin cells in the colon of AKR mice on day 14 postinfection. (D) Representative micrograph showing enterochromaffin cells in the colon of BALB/c mice on day 14 postinfection. Day 0 indicates non-infected control mice. *Significant difference (p<0.05) between BALB/c and AKR mice. Each bar represents mean ± SEM from five mice.

Figure 4  Numbers of enterochromaffin (EC) cells, amount of 5-hydroxytryptamine (5-HT) and worm burden differ in Stat4−/− and Stat6−/− mice after T. muris infection. Stat4−/− and Stat6−/− mice were infected by mouth with 300 eggs of T. muris and were killed on day 15 postinfection to investigate worm burden, colonic enterochromaffin cells and colonic 5-HT amount. (A) Enterochromaffin cell numbers in the colon of infected Stat4−/− and Stat6−/− mice. (B) Amount of 5-HT in the colon of infected Stat4−/− and Stat6−/− mice. (C) Representative micrograph showing enterochromaffin cells in the colon of infected Stat4−/− mice. (D) Representative micrograph showing enterochromaffin cells in the colon of infected Stat6−/− mice. (E) Worm burden in Stat4−/− and Stat6−/− mice. Each bar represents mean ± SEM from four mice. *Significant difference (p<0.05) between Stat6−/− and Stat4−/− mice.

day 15 postinfection). In addition, in association with the enhanced Th2 response we observed faster worm expulsion in Stat4−/− mice compared with Stat6−/− mice. Almost all worms were expelled from Stat4−/− mice by day 15 postinfection, whereas we recovered substantial numbers of worms from Stat6−/− mice on day 15 postinfection (fig 4E). There were significant correlations between the ability to expel the parasite (as measured by worm burden) and both the enterochromaffin
Numbers of enterochromaffin cells and amount of 5-HT in colon differ after reconstitution of SCID mice with in-vitro polarised Th1 and Th2 cells

In previous studies we have demonstrated significantly lower colonic enterochromaffin cell numbers and 5-HT content in the immunodeficient SCID mice compared with those in wild-type mice in *T. muris* infection. In the present study we observed a difference in the numbers of enterochromaffin cells and 5-HT content in the colon in a Th1 and Th2 environment in *T. muris*-infected AKR and BALB/c mice, respectively. Therefore, we next investigated the role of in-vitro polarised Th1 and Th2 cells in the regulation of enterochromaffin cell function and 5-HT production in the gut using SCID mice. As shown in figure 5 the numbers of enterochromaffin cells were significantly greater after reconstitution of SCID mice with purified CD4 T cells polarised to the Th2 type compared with those mice reconstituted with cells polarised to the Th1 type. Colonic 5-HT content was also markedly higher in SCID mice reconstituted with Th2 cells compared with that in SCID mice reconstituted with Th1 cells (fig 5B).

To investigate the effectiveness of Th1 and Th2 polarisation we investigated the IL4 and IFN-γ levels in the supernatant of the polarised cells after in-vitro Con A stimulation and observed a higher level of IL4 in the supernatant of cells polarised to the Th2 type and a higher level of IFN-γ in the supernatant of cells polarised to the Th1 type (IFN-γ: 9.5 pg/ml versus 0 pg/ml in the supernatant of Th1 versus Th2 polarised cells, respectively; IL4: 35.8 pg/ml versus 0 pg/ml in the supernatant of Th2 versus Th1 polarised cells, respectively). To evaluate the effectiveness of the reconstitution of SCID mice we investigated tissue cytokine levels in the reconstituted mice and observed significantly greater amounts of IFNγ and IL4 in the colon of SCID mice that received Th1 and Th2 cells, respectively, on day 21 post-reconstitution (fig 6). There were significant correlations between Th2-biased (as measured by the ratio between IL4 and IFN-γ levels) and both the enterochromaffin cell numbers and amount of 5-HT (Pearson’s correlation coefficient −0.76 and −0.89, respectively; p<0.05 and p<0.001, respectively). Immunohistochemical studies on colonic CDS-positive T cells revealed a significant upregulation of colonic CDS-positive T cells in the SCID mice reconstituted with both Th1 and Th2 polarised cells (fig 7).

**DISCUSSION**

Our study demonstrates a difference in enterochromaffin cell numbers and colonic 5-HT content in the Th1 and Th2 environment and provides further evidence for the immunological control of endocrine function in the gut in enteric infection and inflammation. We observed greater numbers of enterochromaffin cells and a higher amount of 5-HT in the colon of mice that are resistant to *T. muris* infection and generate a Th2 type immune response compared with those in the colon of mice that are susceptible to infection with the generation of a Th1 type immune response. There were also significantly lower numbers of enterochromaffin cells in mice deficient in Stat6, which are impaired in generating a Th2 response. In contrast, mice deficient in Stat4, which are impaired in Th1 cytokine production, exhibited greater numbers of enterochromaffin cells. In addition, adoptive transfer studies in immunodeficient SCID mice with in-vitro polarised Th1 and Th2 cells show a significantly higher number of enterochromaffin cells and higher 5-HT content in the colon of mice that received Th2 cells compared with those in the colony of mice that received Th1 cells. Taken together, the present study provides evidence for
the first time that enterochromaffin cell response and 5-HT production to the same infectious agent are influenced by Th1 or Th2 cytokine predominance.

Enterochromaffin cells have specialised microvilli that project into the lumen and contain enzymes and transporters known to be present in the apical parts of the entocytes. Enterochromaffin cells function as sensors for the contents of the gut lumen and respond to luminal stimuli directly via these transporters and/or indirectly by mediators from the surrounding cells. 5-HT produced from enterochromaffin cells is implicated in the neuro-immuno-endoctrine networks of both humans and rodents. 5-HT is an important enteric mucosal signalling molecule and by virtue of their structural location enterochromaffin cells are likely to play a strategic role in the maintenance of gut homeostasis. 5-HT has a confounding range of effects in the GI tract and has been implicated in the pathophysiology of a number of GI disorders such as enteritis, IBS, inflammatory bowel disease and colon carcinoma. It has recently been shown that T. spiralis infection-induced upregulation of enterochromaffin cells is attenuated in T-cell receptor (β × 8) knockout mice. The role of the host’s immune response underlying changes in enterochromaffin cells and 5-HT has also been demonstrated in mice infected with the bacterial pathogen Citrobacter rodentium. The number of enterochromaffin cells and amount of 5-HT in the colon was significantly reduced in C. rodentium-infected immunocompetent mice. This C. rodentium infection-induced alteration in enterochromaffin cells and 5-HT was not, however, evident in SCID mice. In our previous study we have shown that CD4 T cells play a critical role in the development of enterochromaffin cell hyperplasia and 5-HT production in nematode infection. In the present study we observed that the T. muris infection-induced upregulation of enterochromaffin cells and 5-HT was significantly higher in the Th2 environment of resistant mice compared with that in the Th1 environment in susceptible mice. The influence of Th1 and Th2 type immune responses on enterochromaffin cell function and 5-HT production was further illustrated by the studies using Stat4−/− and Stat6−/− mice. Infection-induced changes in enterochromaffin cells and 5-HT were significantly lower in Stat6−/− mice compared with those in Stat4−/− mice. This was associated with an attenuated and enhanced Th2 response in infected Stat6−/− mice and Stat4−/− mice, respectively. In addition, transfer experiments with in-vitro polarised Th1 and Th2 cells in SCID mice provide direct evidence on the differential regulation of Th1 and Th2 cells in enterochromaffin cell and 5-HT response in the gut. These observations suggest that infection and inflammation of the GI tract cause significant alteration in enterochromaffin cell function and 5-HT production and the immunological profile of the inflammatory response may be an important determinant of the changes that occur in the 5-HT response in the gut. Our findings on Th1/Th2 regulation of enterochromaffin cell biology corroborate with recent studies on lower numbers of enterochromaffin cells and a lower amount of 5-HT in Th1-biased bacterial infection with C. rodentium and on the inhibitory effect of IFN-γ on the proliferation of BON1 tumour cells (model of human enterochromaffin cells). Related to this it has recently been shown that the rate of epithelial cell turnover in the large intestine was higher in resistant BALB/c mice in T. muris infection compared with that in susceptible AKR mice and that the rate of epithelial cell movement is under immune control by IL13. By laser capture microdissection-based molecular and immunofluorescence techniques we have recently demonstrated the presence of the IL13 receptor on colonic enterochromaffin cells. As enterochromaffin and epithelial cells arise from common totipotent stem cells in the gut it is surmised that Th2 cytokines play an important role in enterochromaffin cell biology in this infection by influencing cell turnover and the release of 5-HT.

Enterochromaffin cells are located in close proximity to the mucosal sensory nerve endings and interganglionic neurons, which synapse on motor neurons. When secreted from enterochromaffin cells, 5-HT activates intrinsic and extrinsic neural pathways affecting GI motor function. Alteration in GI motor function occurs in diverse clinical settings, including infections by a variety of infectious agents. Data obtained from...
previous studies in nematode models including the *T. muris* model show that Th2 cells play a pivotal role in the regulation of intestinal muscle function and mucin production and this is associated with host resistance in enteric parasitic infection.57, 58

The present study shows that the extent of enterochromaffin cell response and 5-HT production is dependent on the immunological profile (Th1 or Th2) in the same infection. The results of this study corroborate well with the findings on the amount of 5-HT in Crohn’s disease and cholera, which generates predominant Th1 and Th2 responses, respectively.59–61

In addition, it has recently been shown that the reduced expression of phospho-MEK, a downstream target of c-Raf, in neuroendocrine cells in the colon biopsies correlates with clinical responses in Crohn’s disease as a result of treatment with the anti-inflammatory small molecule semapimod, suggesting that neuroendocrine cells that are important regulators of gut physiology might be involved in the pathogenesis of human colonic inflammation.62

Taken together, in addition to providing novel data on the mechanisms of 5-HT production, these observations give new insights into the mechanisms of gut immune–endocrine interaction in relation to gut physiology, which may ultimately lead to improved therapeutic strategies in various GI infections and inflammatory conditions in which alterations of 5-HT signalling are seen in association with immune activation in the GI tract.

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Competing interests: None.

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to combine adult and paediatric practice (eg, the chapters on assessment and on intestinal failure-associated liver disease). I would not be so unkind as to describe it from the perspective of the curate faced with the dodgy egg, but I did find it somewhat unsatisfactory. It is good as it prompts both groups of specialists to consider problems more particular to the other, but these chapters seem to lose focus in the process.

I was disappointed to find no mention of the random urine sodium in assessment of the intestinal failure patient—in my view a hugely helpful simple investigation—but perhaps its absence is symptomatic of its neglect in the literature as a whole.

An important portion of the book revolves around intestinal transplantation (16 of the 42 chapters), and there are many gems therein. It is helpful to have the surgical approaches summarised, but these are of most practical relevance to those in transplant centres, and I think most potential readers will be more concerned with the prior strategies on indications and referral, and with the postoperative aspects in patients who may be seen once they are released from hyperspecialist care. Accordingly I was disappointed with the chapter on long-term management as the opportunity to provide a hit-list of things that must and must not be done has been missed. This screams out for a secure algorithm for the non-specialist centre faced with a sick (or indeed not so sick) transplant patient. Finally there are no transplant outcome data more recent than 2005–6, which is a shame for such a rapidly moving field; I had hoped that Professor Grant might have given a sneak preview of more recent results.

On balance I am very positive about the book, consider it very good value for money and would strongly recommend it to all units dealing with intestinal failure. Some opportunities have been missed, and I am sure these will be dealt with in the future editions that are likely to emerge. It scores over the Nightingale book in its handling of transplantation, but this is mainly the consequence of the 7 years that separate the two volumes. Personally I am pleased to have both!

Alastair Forbes

Correspondence to: Professor A Forbes, Department of Gastroenterology and Nutrition, University College Hospital, 235 Euston Road, London NW1 2BU, UK; a.forbes@ucl.ac.uk

Endoscopy in a new light...


Narrow band imaging (NBI) comes as standard with the latest version of the hardware sold by the UK’s most popular supplier of endoscopy equipment, but how many UK gastroenterologists know the theory of the new modality, how to use it, the appearances of even common lesions and the potential benefits for patients? This book attempts to bring the experience of the (mostly Japanese) front runners in NBI to a more general audience.

NBI uses restricted parts of the spectrum of white light to enhance the vessel pattern and superficial detail of mucosal lesions, allowing the diagnosis of early lesions such as cancers and dysplasia, so poor in current UK endoscopic practice. It is easy to use in narrow tubular organs such as the oesophagus and colon where light intensity is relatively high; light levels are lower in the more capacious stomach, reducing the value of NBI as a screening tool, though it still appears to be of value in detailed study of lesions first suspected by white light endoscopy. It is easy to deploy at the touch of a button, making it more practical than chromoendoscopy. There is a lot to learn about interpretation of the images where haemoglobin is shown in brown or green depending on the depth and size of the containing vessel, and the importance of the microvascular pattern. The common artefacts, due to poor bowel preparation for example where faeces is rendered as red, take some adjustment in the brain of the endoscopist. There are also more related acronyms and more Japanese classifications to take on!

The utility of NBI is not yet well supported by peer-reviewed publications, and as this book makes clear, it is complementary to a variety of other modalities including high-resolution endoscopy (HRE), magnification and chromoendoscopy which are not yet routine in UK endoscopy units.

The chapters explaining the basis of NBI and its potential applications are in quite good English, but some parts have not translated well from the original Japanese articles. The book contains a large series of good-quality endoscopic photos which could be better arranged, however. The illustrations with each chapter are printed at the end of the chapters, rather than integrated in the text where they would be easier to relate. The atlas in the second half of the book presents white light, NBI and magnification and chromoendoscopy images of lesions, but not always on the same page which is slightly irritating. The text and images also come on an included searchable DVD containing good-quality still images from the book and 55 video clips. The perfection of the Japanese clips is remarkable and the videos are very helpful in understanding the NBI appearances, though a number of the sequences are irrelevant to the subject of the book and seem to have been included as “stocking fillers”.

Endoscopy now has to compete with other non-invasive imaging techniques. NBI, HRE and other new intraluminal imaging modalities are likely to give endoscopy a sensitivity unmatched by any other technique, and combined with its therapeutic potential will contribute to the development and survival of endoscopy. The benefits to patients of this increased sensitivity will need to be shown, but serious endoscopists will want to assimilate the contents of this publication, recognising that the study of NBI has much potential.

Peter D Fairclough

Correspondence to: Dr P Fairclough, Endoscopy Unit, The Royal London Hospital, London E1 1BB, UK; peter.f@pobox.com

CORRECTIONS

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Enterochromaffin cell and 5-hydroxytryptamine responses to the same infectious agent differ in Th1 and Th2 dominant environments. Gut 2008;57:475–81. There is an error in the key for figure 5: solid bars represent SCID+ Th2 and open bars represent SCID+ Th1.