[Frontiers in Bioscience S4, 685-698, January 1, 2012]

Innate and adaptive immunity in host-microbiota mutualism

Andrew J. Macpherson, Markus B. Geuking, Kathy D. McCoy

Maurice Muller Laboratories, University Clinic for Visceral Surgery and Medicine, Murtenstrasse 35, University of Bern, CH-3010, Bern, Switzerland

TABLE OF CONTENTS

1. Abstract
2. Introduction
3. Adaptation of the host to the colonisation of the intestine with a commensal microbiota
4. Uncoupled mucosal and systemic immune responses to commensal intestinal microbes
5. The mucosal immune firewall
6. The role of innate immunity in mutualism with intestinal microbes
7. Conclusions
8. Acknowledgement
7. References

1. ABSTRACT

Healthy individuals live in peaceful co-existence with an immense load of intestinal bacteria. This symbiosis is advantageous for both the host and the bacteria. For the host it provides access to otherwise undigestible nutrients and colonization resistance against pathogens. In return the bacteria receive an excellent nutrient habitat. The mucosal immune adaptations to the presence of this commensal intestinal microflora are manifold. Although bacterial colonization has clear systemic consequences, such as maturation of the immune system, it is striking that the mutualistic adaptive (T and B cells) and innate immune responses are precisely compartmentalized to the mucosal immune system. Here we summarize the mechanisms of mucosal immune compartmentalization and its importance for a healthy host-microbiota mutualism.

2. INTRODUCTION

The lower intestine is a habitat for one of the highest densities of microbial consortia on the planet (1). An enduring question is exactly how this load of microbes (reaching 10^{12} organisms/g of intestinal contents) can be accommodated without damaging the host, given that the barrier between the lumen of the intestine and host tissues is composed of a simple epithelial layer, one cell thick. It is clear that although commensal microbes generally lack pathogenicity genes that characterise pathogens, which encode for proteins that facilitate adhesion, penetration of the epithelium, facultative persistence within host cells or toxin formation, they do share prokaryotic molecular patterns that are powerful activators of the innate immune system.

Table 1. Comparison of Ulcerative Colitis and Crohn's disease[1]

Characteristic	Ulcerative Colitis	Crohn's disease
Site of disease	Colon (proctosigmoiditis, left-sided colitis, pancolitis, backwash colitis)	Colon (2/3), Ileum (2/3), infrequent in jejunum and duodenum
Clinical Features	Fever, abdominal pain, bloody diarrhea, weight loss	Fever, abdominal pain, diarrhea, weight loss, fatigue
Intestinal Complications	Strictures absent, fistulas absent, punctate ulcers, pseudopolyps, shortened colon, rectum involved, perianal disease absent	Strictures present (fibrotic and stenotic), fissures and fistulas common, deep ulcers, cobblestone appearance, perianal disease
Inflammation	Primarily mucosal, cryptitis, crypt abscess, granulomas absent, associated with IL-13	Transmural, crypt abscess, granulomas, associated with IL-12/IL-23 and IFN-gamma/IL-17 production

[1]This table was created based on references (39, 40).

This question of how commensal microbial mutualism with the host is induced and maintained is also medically important for several reasons. First, about 2 in every 1000 people in industrialised countries suffer from inflammatory bowel disease (IBD) (2). This can manifest in two forms – ulcerative colitis (UC) and Crohn's disease. Table 1 provides an overview of the major differences in immunity between UC and Crohn's disease. Both probably depend on abnormal mucosal immune responsiveness to commensal intestinal microbes: the evidence for a dysregulated handling of intestinal commensals in Crohn's disease is particularly strong on the basis of clinical data (3), the genes that give a genetic susceptibility in the human population (4-6) and animal models of the disease (Table 2). Secondly, many infections in immunocompromised patients, such as in individuals treated with chemotherapy, are from commensal intestinal bacteria.

In this review we will focus on the compartmentalisation of the immune system in responding and adapting to the presence of commensal intestinal microbes with an emphasis on the induction of IgA against commensals as a method of dissecting host-microbial immune mechanisms. Because of the ability to precisely control and manipulate the intestinal microflora in mouse models, the majority of the data discussed here is derived from animal studies. However, where data is available we have added evidence from human studies.

3. ADAPTATION OF THE HOST TO THE COLONISATION OF THE INTESTINE WITH A COMMENSAL MICROBIOTA

The gut is a tube from mouth to anus, so luminal bacteria are not, strictly speaking, inside the body tissues. The evidence that the host has to adapt to the presence of the commensal intestinal microbes comes from studies that have compared germ-free animals with the same animal strain colonised with intestinal bacteria. An alternative approach is to follow the changes in host immunity and other body systems, as intestinal bacteria are introduced to germ-free animals (7).

Germ-free rodents were first derived nearly a century ago, stimulated by the question of whether animal life was possible in the absence of commensal microbes (8). Initially rodents were delivered aseptically and hand reared for short periods: later it became possible to interbreed the germ-free adults, and this is now standard technology (7, 9-11). Because *in vivo* experiments are easy with mice and many different genetic backgrounds and targeted genetic lesions are available, most germ-free experimentation is with this species. Today, it is convenient to maintain strains in plastic flexible film isolators, introducing sterile food, water and bedding from a sterilised drum connected to the isolator with a plastic sleeve. The inside of the sleeve is sterilised with a 2% peracetic acid mist before the inner door of the isolator is opened and the seal of the drum is broken to access the sterilised contents (7). Although some units still use aseptic Caesarian section to re-derive different genetic strains of mice germ-free, this is a cumbersome and unreliable procedure compared with aseptic embryo transfer into germ-free pseudopregnant females, which then deliver and foster the pups (7).

There are enormous differences between germ-free mice and their colonised counterparts in almost every body system (7). Immunity of the gut is shaped by the introduction of intestinal commensal bacteria through increase in the cellularity and organisation of secondary lymphoid structure of the gut (Peyer's patches and isolated lymphoid follicles) and systemic immune system (spleen, lymph nodes), the induction and secretion of IgA from intestinal lamina propria plasma cells, increase in the content of lamina propria CD4 T cells, and general increase in serum immunoglobulin levels, especially the IgG isotypes (12-14). Paradoxically, IgE levels are abnormally high in germ-free mice and are decreased when the animals become colonised with commensal intestinal bacteria (15).

It is clear from the manifold changes in immunity and other body systems as the intestines of germ-free animals become colonised, that mammals adapt to the presence of commensal intestinal bacteria (14). We assume that these adaptations are functionally relevant, although this is still a developing area as discussed below.

4. UNCOUPLED MUCOSAL AND SYSTEMIC IMMUNE RESPONSES TO COMMENSAL INTESTINAL MICROBES

If pathogens are eliminated from colonies of mice (so they are designated 'specific pathogen free' or SPF) they still have a commensal intestinal microbiota. Such SPF mice mount a strong mucosal response to their intestinal commensals, manifested by the secretion of specific intestinal IgA across the mucosa, but the systemic immune system remains ignorant of these intestinal microbes and no specific serum antibodies or T cells are induced (16, 17). This 'ignorance' of the systemic immune system can easily be broken experimentally by injecting a

Table 2. Animal models of IBD versus human IBD

Defect	Mouse Models	Area involved	Abnormality	Flora-dependence	Relationship to human IBD
Epithelial integrity/enteric glia	Trefoil factor deficiency (41)	Colon	Colitis induced with DSS[1]	Unknown	Role in mucosal healing (42)
	Ablation of enteric Glia (43)	Fulminant Jejuno-Ileitis	Severe inflammation and intraluminal hemorrhage	Unknown	Potential role of glial-derived neurotrophic factor in human IBD (44)
	Multiple drug resistance gene 1a (Mdr1a)-deficient mice (45, 46)	Colon	Spontaneous colitis. Dysfunctional intestinal epithelium rather than altered lymphocyte function.	Colitis absent in germ-free Mdr1a$^{-/-}$ mice (47)	Association between polymorphism in MDR1 gene and ulcerative colitis (48)
	Galphai2-deficient mice (49, 50)	Entire colon	Increased Th1-driven inflammation	Unknown	Potential model for Ulcerative colitis
	Dominant negative N-cadherin mutant Tg mice(51)	Small bowel cecum	Epithelial barrier defect	Unknown	Potential model for Crohn's disease
	DSS administration (52)	Colon	Direct effect on the gut epithelium. Colitis induced in T- and B-cell deficient mice (53). Model for innate immune mechanisms	Germ-free mice susceptible to DSS damage (54, 55) Alleviated intestinal inflammation after antibiotic treatment.	Potential model for Ulcerative colitis
	TNBS[2] administration (56)	Colon (small intestine?)	Th1-mediated colitis	Alleviated intestinal inflammation after antibiotic treatment.	Potential model for Crohn's disease
	Muc2 deficient mice (57-59)	Colon	Microscopic evidence of colitis as early as 5 weeks of age. More susceptible to DSS colitis	Unknown	Muc2 is prominent colonic mucin expressed in UC (60) and expression may be altered in IBD (61-63)
	Chloride channel CLC-5 (Clcn5)-deficient mice (64)	Colon	More susceptible to DSS colitis. Heightened Th1-Th17 cytokine profile after DSS	Unknown	Clc-5 is down-regulated in sigmoid mucosal biopsies of most patients with active UC (65)
	Keratin-8-deficient mice (66, 67)	Cecum, colon	Chronic spontaneous Th2 colitis	Amenable to antibiotic treatment	*KRT8* variants were not found to be overexpressed in familial IBD but potential role in sporadic IBD was not ruled out (68)
Innate immune cell function Antigen presentation	Stat3 deficient macrophages and neutrophils (69)	Cecum, colon	Spontaneous enterocolitis, dependent on IL-12p40, defective IL-10 production	Reduced inflammation in TLR-4$^{-/-}$ or MyD88$^{-/-}$ (70)	Model of Crohn's disease
	A20 deficiency (71)	Multiple organs colon	T- and B-cell independent	Unknown	Butyrate -mediated downregulation of IL-8 in human intestinal epithelial cells may be mediated through A20 (72)
	T-bet deficiency in the innate immune system (73)	Colon	T-bet$^{-/-}$xRag2$^{-/-}$ develop spontaneous colitis, colonic dendritic cells over produce TNF-alpha, anti-TNFalpha therapy prevented colitis	Transfer of microbiota from afflicted mice causes intestinal inflammation in wild-type mice	Anti-TNF-alpha is a therapy to treat IBD
	TLR5 deficient mice (74)	Colon	Spontaneous colitis, increased proinflammatory cytokines	Alterations in gut microflora observed, colitis dependent on TLR-4 and ameliorated by antibiotic treatment	Dominant-negative TLR5 polymorphism negatively associates with Crohn's disease (75)
	NOD2 deficiency (exon 1) (76)	No spontaneous colitis	Th1-mediated colitis induced to microbial antigen (77, 78)	TLR-2-dependent	Mutations in CARD15, the gene encoding NOD2, found to be a major susceptibility factor in Crohn's disease (4-6)
	NOD2 deficiency (exon 3) (79)	No spontaneous colitis	More susceptible to bacterial infection via oral route. Not more susceptible to DSS	Reduced cryptadins in NOD2$^{-/-}$ mice. Altered regulation of commensal flora in NOD2$^{-/-}$ mice and decreased expression of NOD2 under germ-free conditions (80)	
	NOD2^{2939} mutant (81)	No spontaneous colitis	Increased susceptibility to DSS, elevated IkB kinase and caspase-1 in response to muramyl dipeptide	Unknown	

	HLA-B27/human beta2microglobulin transgenic rats (82-84)	Entire intestine Other organs	Increased macrophage and lymphocyte infiltration in colon, increased pro-inflammatory cytokines and myeloperoxidase	No colitis in germ-free conditions	HLA-B27 association with Crohn's disease (85, 86)
Cytokines/immune regulation	TNF D^{ARE} mice (TNF over production) (87, 88)	Multiple organs, terminal ileum, proximal colon	Ileitis depends on presence of lymphocytes, milder disease in the absence of IL-12p40 or IFN-gamma	Unknown	Blockade of TNF-alpha therapeutic in human IBD
	IL-7 Tg mice (89)	Colon	$CD4^+$ T cell-mediated. Long-lived colitogenic $CD4^+$ memory T cells express IL-7R	$IL\text{-}7R^+$ $CD4^+$ colitogenic T cells transferred in germ-free mice do not drive colitis unless commensal bacteria introduced (90)	IL-7 receptor has been identified as a risk loci for ulcerative colitis (91)
	TGF-beta1 deficient mice (92)	Multiple organs	Aberrant immunoregulation, CD4 T cell-mediated	Absence of colitis (and cancer) in germ-free TGF-beta1/$Rag2^{-/-}$-deficient mice (93)	TGF-beta negatively controls inflammatory pathways in the human gut (94), expression is increased in human IBD but Smad signaling pathway impaired (95)
	CD4-dnTGFbeta RII transgenic mice (CD4 T cell-specific expression of dominant negative TGF-beta1 receptor II) (96)	Multiple organs including colon	Aberrant immunoregulation when $CD4^+$ T cells cannot respond to TGF-beta	Activated $CD4^+$ T cells in T cell transfer model of colitis escape control by regulatory T cells (97)	As above
	Intestinal Trefoil factor (ITF)-dnTGFbeta RII transgenic mice (Intestine-specific expression of dominant negative TGF-beta1 receptor II) (98)	No spontaneous colitis	Increased susceptibility to DSS-induced colitis	Unknown	As above
	IL-10 deficiency (99)	Cecum, colon (small intestine?)	Aberrant immunoregulation, Th1 mediated disease, T cell-derived IL-10 important	Attenuated by antibiotic treatment (100) Absent in germ-free (101) Absent in IL-10/MyD88-double-deficient mice (102)	IL-10 decreased in severe cases of UC and CD (103, 104). No consistent association of *IL-10* promoter polymorphisms and IBD susceptibility, strong association of polymorphism downstream of *IL-10* gene and susceptibility to UC (105) and CD (106, 107). Loss-of-function mutations in *IL-10R* results in CD (108)
	IL-2 deficiency (109)	Cecum, colon	Aberrant immuno-regulation (Th1)	Reduced inflammation in germ-free (110, 111)	Polymorphism in *IL-2* associated with UC (91).
	IL-2Ralpha deficient (112)	Cecum, colon	Aberrant immuno-regulation (Th1)	Probably	Polymorphism in *IL-2RA* identified as susceptibility loci in CD (107).
	NFkB p50 deficiency (113)	Cecum and colon	Increased Th1 cytokines	Inflammation dependent on presence of Helicobacter	NFkappaB activation likely involved in human IBD (114)
T cells/immune regulation	Gp39 (CD40L) transgenic mice (115)	Colon, small bowel, other organs	Aberrant immunoregulation	Unknown	Increased soluble CD40L in Crohn's disease, potential marker for intestinal inflammation (116)
	TCRalpha chain deficient (117, 118)	Cecum, colon	Aberrant Th2-type T cells.	No inflammation in germ-free mice or mice with a limited intestinal flora (119)	Some characteristics similar to human ulcerative colitis
	Tg 26e mice (human CD3epsilon transgenic) (120)	Cecum, colon	Abnormal T cell development	Dependent on continuous stimulation by luminal bacteria (121)	Some characteristics similar to human ulcerative colitis
	Stat4 tg mice (122)	Colon, Ileum	Th1-mediated	Unknown	Stat4 is involved in IL-12/IL-23 receptor signaling
	CD4 $CD45RB^{hi}$ T cell transfer into lymphopenic host (123)	Cecum, colon	T cell-mediated inflammation (Th1)	No or severely reduced inflammation in antibiotic treated or germ-free recipient SCID mice	Altered immunity to enteric flora
	Oxazalone administration (124)	Colon	Th2-mediated		Potential model for ulcerative colitis

	Wiskott-Aldrich Syndrome protein (WASP) deficiency (125)	Colon	CD4+ T cell-mediated, Th2 cytokine predominance, and abnormal T regulatory induction (126-128)	Spontaneous colitis requires presence of Helicobacter (129)	Some patients with Wiskott-Aldrich syndrome develop IBD (130, 131)
	Altered B-7-mediated costimulation (132)	Colon	Soluble B7-2 Fc transgenic mice develop chronic colitis, altered activation of CD4 T cells.	Unknown	CTLA-4 blockade as treatment for melanoma has led to development of colitis in some patients (133)
Undefined genetic models	C3H/HeJBir mice (134)	Cecum, colon	$CD4^+$ T cells transfer colitis to scid recipients (135)	Serum IgG reactivity (136) and $CD4^+$ T cells specificity to antigens from intestinal bacteria (135). Reduced colitis in SPF conditions (137)	Illustrates genetic risk factors in humans
	Samp1/Yit mice (138, 139)	Ileum	One of the few models with severe inflammation in the terminal ileum. Th-1-mediated	Reduced (but not absent) inflammation in germ-free	Peroxisome proliferator-activated receptor gamma (PPAR-gamma) is a susceptibility gene in both Samp1/Yit mice and human Crohn's disease (140)

Abbreviations: Dextran sulfate sodium[1], trinitrobenzene sulfonic acid[2]

single dose of a commensal bacteria prepared from pure culture into the tail vein and measuring the appearance of a specific antibody response approximately 14 days later (16).

This shows experimentally that, in unmanipulated SPF animals containing a microbiota, the mucosal immune system mounts responses against commensal microbes quite independently of the systemic immune system (16, 17). As one might expect, the systemic immune system can very easily mount a response provided that the microbes reach systemic secondary lymphoid structures in sufficient numbers (16, 18). In other words immune compartmentalisation is responsible for mutualism with commensal microbes, and there is little or no evidence for systemic immune tolerance (19).

We have recently been able to refine the way in which antibacterial antibodies are detected. Earlier methods of using Western blots of bacterial lysates or binding whole bacteria to plastic and then using enzyme linked (ELISA) methods to detect bound antibodies detect non species-specific binding (16). Although these antibodies are probably functionally relevant, they are presumably of relatively low affinity against bacterial epitopes. Using a flow-cytometric (FACS) assay, we have been able to detect antibodies bound to bacteria with high specificity, for example although Salmonella and Escherichia coli are closely related bacterial species, antibodies raised by priming with the different organisms *in vivo* do not cross-react in the FACS assay (18).

A further advance has been the ability to use genetically modified bacteria containing auxotrophic mutations for synthesis pathways of bacterial compounds that are not found in eukaryotes. These bacteria (HA107) can be grown in culture in the lab (with media containing the appropriate chemical supplements), but they cannot survive in animals, so germ-free mice can be exposed to these bacteria and become germ-free again after about 72 hours (20). This has allowed us to study induction of commensal intestinal immune responses in germ-free animals uncoupled from colonisation.

Germ-free mice treated with *E.coli* HA107, that become germ-free again, develop a specific intestinal IgA response against HA107 that is extremely long lasting (>16 weeks) even though the exposure to live bacteria is very short (72 hours). This shows, in a clean experimental system, that the anti-commensal IgA response in the intestine is very specific (20).

The HA107 tool has also allowed us to show that the threshold for an IgA response in the intestine is very high ($>10^9$ organisms are required for experimental induction) and does not show the prime-boost memory effect that is characteristic of systemic immune priming. Indeed the overall anti-commensal IgA response is rather an integral of the total amount of bacterial exposure. This suggests that in this system memory seems to be achieved by persistence of the IgA response, which will eventually be displaced when a new IgA response is induced to a different intestinal commensal (20).

5. THE MUCOSAL IMMUNE FIREWALL

Since there is a separation of mucosal and systemic immune responses to commensals, the question of the mechanism of how a mucosal response to commensals can be induced in the absence of a systemic response arises. We, and others, have shown that intestinal dendritic cells (DC) sample commensal bacteria at the mucosal surface (21, 22). The small intestine is thought to contain two main types of DC distinguished by the expression of CD103 (also known as $alpha_E$-integrin) (reviewed in 23). The $CD103^+$ DC subset is thought to be conditioned by factors in the intestinal microenvironment, such as TGF-beta, retinoic acid, microbial antigens, and IL-10, and then migrate via C-C chemokine receptor type 7 (CCR7) to the mesenteric lymph nodes (MLN) where they can promote regulatory T cell development and induction of CCR9 and alpha4beta7 expression on T cells (24-26). This population also seems to be conserved between mouse and man (27). In contrast, the $CD103^-$ DC population has been implicated in T helper 17 (Th17) cell induction (28) and expresses CX3C chemokine recptor 1 (CX_3CR1), which allows them to extend dendrites between the tight junctions of the intestinal epithelial cells and sample microorganisms (21,

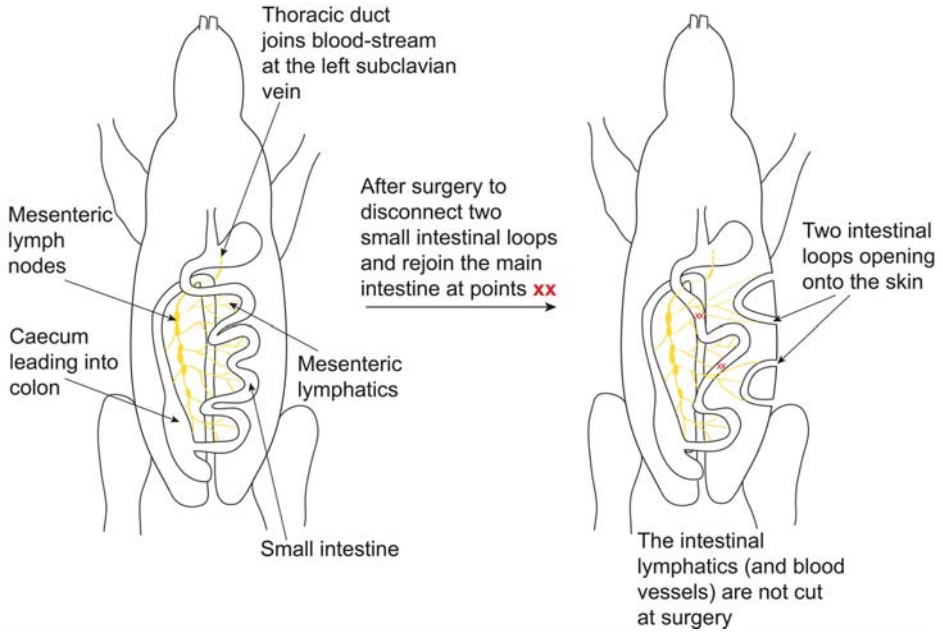

Figure 1. Isolated intestinal loops retain mesenteric lymphatics and blood vessels but are surgically disconnected from the rest of the small intestine. The anatomy of the intestine is shown on the left, with the mesenteric lymphatics and lymph nodes in yellow. Two segments of the small intestine are surgically disconnected from the main small intestine and brought out to the skin with external stoma, as shown on the right. The main small intestine is reanastomosed at the two sections indicated by the red XX.

29). DC can also capture microorganisms that transcytose through specialized M cells in the follicle-associated epithelium of the Peyer's patches (reviewed in 30). Sampling of the commensal microflora occurs across the length of the small intestine under steady-state conditions but may increase, especially in the terminal ileum, following infection with pathogenic bacteria (29, 31). Compared with other host phagocytes, DC have rather poor biocidal activity so the live bacteria are retained for up to several days. The bacterially-loaded DC are capable of inducing IgA B cells and can migrate to reach the MLN but do not penetrate beyond the MLN to reach central body tissues. This means that induction of mucosal immunity by commensals is largely restricted to mucosal inductive sites in animals, provided that the immune system is functioning normally (22).

Of course the restriction of immune induction against commensals to mucosal secondary lymphoid structures does not preclude dissemination of the response across the mucosal immune system, because induced B and T cells recirculate through the lymph to the thoracic duct, where they join the blood stream and home back to mucosal tissues (32, 33). It is important for this recirculation that the induced lymphocytes are programmed to express the necessary homing receptors. Induction of CCR9 and alpha4beta7 on intestinal B and T cells is triggered through constitutive expression of retinoic acid by intestinal dendritic cells (34).

Commensal bacteria (and presumably other elements of the intestinal commensal microbiota) are efficiently phagocytosed. There are several lines of evidence for this.

1. It has been shown that intestinal bacterial pathogens (such as Salmonella, Shigella and Yersinia) frequently employ mechanisms to subvert entry into, or they trigger mechanisms of biocidal compartments of phagocytes, in order to maintain a facultative intracellular existence (35, 36). Since such subversion is required for pathogenicity, it follows that non-pathogenic commensals that penetrate mucosal defences allow themselves to be killed by phagocytes as a part of host-microbial mutualism.

2. When biocidal mechanisms of phagocytes are experimentally rendered deficient as a result of genetic manipulation in mice, the consequence is a severe phenotype with a susceptibility to fatal sepsis from intestinal commensals that starts soon after weaning (37).

3. In experiments with isolated intestinal loops, we asked whether commensal bacteria could penetrate to the mesenteric lymph nodes in a free state, or whether they had to be carried there contained within intestinal dendritic cells (22). The experimental setup is shown in Figure 1. Two segments of small intestine were disconnected surgically, without disturbing the lymphatic or vascular supply, and the main small intestine was reanastomosed at the positions marked "xx" to restore continuity. The disconnected small intestinal loops were brought out onto the skin with external stomas (although the loops are shown close together on the diagram for convenience, in fact they were constructed on separate sides of the abdominal wall). We then pulsed one loop with a commensal (*Enterobacter cloacae*) carrying a naladixic acid antibiotic resistance, and the other loops with *Enterobacter cloacae* carrying rifampicin resistance. After 18 hours the dendritic cells

were plated out at single cell density from disaggregated mesenteric leukocytes. Our reasoning was that if the free bacteria penetrated through the lymphatics some DCs would contain bacteria carrying both resistances. On the other hand, if the bacteria were exclusively taken up by DC at the mucosal surface and then carried to the mesenteric lymph nodes, because the bacterial exposure is in separate loops each with a different bacterial resistance, DC in the mesenteric lymph nodes would only carry *Enterobacter cloacae* containing one antibiotic resistance. This latter situation turned out to be the case. As a control the antibiotic resistant preparations were mixed and put into one or both loops, when mesenteric DC then contained bacteria with both antibiotic markers (22).

In one sense, therefore, the mesenteric lymph nodes are an immunological firewall that protects the systemic immune system from unnecessary exposure to commensals. The system works because microbes are very efficiently phagocytosed: in most cases they will be killed by macrophages or neutrophils, but some can persist in intestinal DC to assist in the induction of anti-commensal immunity. These DC have a relatively short lifespan, and are arrested within the mesenteric lymph nodes: presumably the live commensal bacteria that are released from effete DC in the mesenteric lymph nodes are rapidly destroyed by biocidal activity from the abundant numbers of macrophages present. Since sepsis from commensal bacteria would be potentially serious (for example mastitis in breast-feeding mothers) it is better to have mucosal immune responses that are characterised by distinct immune geography, rather than any system of immune tolerance to these organisms.

6. THE ROLE OF INNATE IMMUNITY IN MUTUALISM WITH INTESTINAL MICROBES

We have described how the normal immune system is able to compartmentalise responses to commensal intestinal microbes with induction within secondary lymphoid tissues of the gut. Three levels of indirect evidence were cited above to show that biocidal activity of the innate immune system is an important part of this compartmentalization.

In fact, it is experimentally possible to allow breaches in the mesenteric lymph node firewall, by surgically removing the mesenteric lymph nodes. Following this operation, the mesenteric lymphatics spontaneously heal and reanastomose, so that the continuity of lymphatic draining is restored within a few weeks: this can be shown by gavaging the animal with olive oil, following which lymphatics appear a (continuous) brilliant white from the micelle emulsion of fat digestion. Whereas central secondary lymphoid structures, such as the spleen, remain sterile when wild-type mice are treated with intestinal doses of commensal bacteria, mice without mesenteric lymph nodes lose this protection and live commensals can be cultured from the spleen. Breaking the firewall also has functional consequences. Mice without mesenteric lymph nodes develop extreme splenomegaly, lymphadenopathy and skin acanthosis during these experiments (22).

Another approach to looking at the importance of innate immunity in providing the immune mutualism with commensal intestinal microbes was to study the effect of mice deficient in both myeloid differentiation primary response gene 88 (MyD88) and TIR-domain-containing adapter-inducing interferon-beta (TRIF) adaptor molecules, which in turn results in deficient Toll-like receptor (TLR) signaling (38). These MyD88/TRIF double deficient mice are hard to breed in normal animal vivaria, but we found that they were very stable once re-derived germ-free (18). We found that when these MyD88/TRIF deficient mice were colonised with a limited ("modified Schaedler") microbiota, containing only 8 culturable bacteria, unlike heterozygous littermates, they spontaneously developed serum IgG antibody responses to the organisms in their commensal microbiota (18). This could be reproduced with a monocolonisation, so it was not a function of particular bacterial species present.

The reason for an abnormal systemic immune response in the face of a MyD88/TRIF double lesion, is that immune phagocytes are poor at killing commensals. This was shown by persistence of commensals following intravenous injection. Moreover, when doses of commensals were gavaged into MyD88/TRIF double deficient mice culturable bacteria can be recovered from the spleen: this is not due to a barrier defect, as measurements of intestinal permeability *ex vivo* in Ussing chambers or the extent of serum protein loss into the intestinal lumen *in vivo* were normal. Indeed when we directly studied mice deficient in the Phox enzyme required to produce superoxide radicals as part of the lysosome biocidal mechanism, the same systemic immune response to the commensal microbiota was seen (18).

These abnormalities in host-intestinal microbial mutualism in the face of severe innate immune defects show us two things.

1. Innate immunity is an essential requirement for the handling of commensal organisms. It is inevitable that some of these microbes should penetrate the extremely thin epithelial layer; indeed they need to do so in order to induce host mucosal immunity as part of the mutualism process. Biocidal mechanisms of macrophages are very likely to be crucial in mopping up commensal microbes that penetrate the barrier, or in eliminating microbes that are released live from effete dendritic cells.

2. The fact that there is a systemic antibody response suggested that the innate and adaptive immune systems work as a continuum in host microbial mutualism. This point was addressed experimentally by breeding a MyD88 deficient murine strain that also carries a deletion of the J segments of the immunoglobulin heavy chain locus ($J_H^{-/-}$), so it cannot produce antibody of any isotype. The MyD88 lesion was chosen in these experiments as it was known to exert most of the phenotype of the MyD88/TRIF double deficient strain (18). Under germ-free conditions the MyD88, $J_H^{-/-}$ mice bred well and matured normally, but when colonised with an altered Schaedler microbiota the offspring commonly died in the neonatal period, and those

that survived to weaning had a severe growth defect indicating that specific serum anticommensal antibodies can partially compensate for defective innate immune mechanisms.

7. CONCLUSIONS

This review has concentrated on the different compartments in which antibodies to commensal intestinal microbes are induced, as a means of exploring the immune geography of host-microbial mutualism. Of course this is a (small) part of the story and mutualism at the level of the mucosal immune system is a multilayered process with innate responses by epithelial cells and an important component of effector and regulatory T cells. We commonly read that mammals must be 'tolerant' of their commensal intestinal microbes: of course semantically this is so, but in an immunological sense we would like to persuade our readers that the evidence for compartmentalisation which allows generation of a selective mucosal immune response without unnecessarily involving systemic immunity is far better than the concept that systemic immunity is in fact downregulated to these organisms. There would be a significant disadvantage of such downregulation as we would be left liable to serious sepsis from our commensals.

8. ACKNOWLEDGEMENT

This work was supported by grants from the Swiss National Science Foundation.

7. REFERENCES

1. W. B. Whitman, D. C. Coleman and W. J. Wiebe: Prokaryotes: The unseen majority. *PNAS* 95(12), 6578-6583 (1998)

2. R. B. Sartor: Pathogenesis and immune mechanisms of chronic inflammatory bowel diseases. *Am J Gastroenterol* 92(12 Suppl), 5S-11S. (1997)

3. P. Rutgeerts, K. Goboes, M. Peeters, M. Hiele, F. Penninckx, R. Aerts, R. Kerremans and G. Vantrappen: Effect of faecal stream diversion on recurrence of Crohn's disease in the neoterminal ileum. *Lancet* 338, 771-774 (1991)

4. J. Hampe, A. Cuthbert, P. J. P. Croucher, M. M. Mirza, S. Mascheretti, S. Fisher, H. Frenzel, K. King, A. Hasselmeyer, A. J. Macpherson, S. Bridger, S. van Deventer, A. Forbes, S. Nikolaus, J. Lennard-Jones, U. R. Foelsch, M. Krawczak, C. Lewis, S. Schreiber and C. G. Mathew: Association between insertion mutation in NOD2 gene and Crohn's disease in German and British populations. *Lancet* 357, 1925-1928 (2001)

5. J. P. Hugot, M. Chamaillard, H. Zouali, S. Lesage, J. P. Cezard, J. Belaiche, S. Almer, C. Tysk, C. A. O'Morain, M. Gassull, V. Binder, Y. Finkel, A. Cortot, R. Modigliani, P. Laurent-Puig, C. Gower-Rousseau, J. Macry, J. F. Colombel, M. Sahbatou and G. Thomas: Association of NOD2 leucine-rich repeat variants with susceptibility to Crohn's disease. *Nature* 411(6837), 599-603. (2001)

6. Y. Ogura, D. K. Bonen, N. Inohara, D. L. Nicolae, F. F. Chen, R. Ramos, H. Britton, T. Moran, R. Karaliuskas, R. H. Duerr, J. P. Achkar, S. R. Brant, T. M. Bayless, B. S. Kirschner, S. B. Hanauer, G. Nunez and J. H. Cho: A frameshift mutation in NOD2 associated with susceptibility to Crohn's disease. *Nature* 411(6837), 603-606. (2001)

7. K. Smith, K. D. McCoy and A. J. Macpherson: Use of axenic animals in studying the adaptation of mammals to their commensal intestinal microbiota. *Semin Immunol* 19(2), 59-69 (2007)

8. L. Pasteur: Observations relatives a la note de M. Duclaux. *Compt rend* 100, 69 (1885)

9. M. Cohendy: Experiences sur la vie sans microbes. *Comp rendt* 154, 533-536 (1912)

10. M. Cohendy: Experiences sur la vie sans microbes, élevage aseptique de cobages. *Compt. rend* 158, 1283-1284 (1914)

11. G. Glimstedt: Bakterienfrei Meerschweinchen. Aufzucht, Lebensfähigkeit und Wachstum, nebst Untersuchung über das lymphatische Gewebe. *Acta Pathol. Microbiol. Scand. Suppl* 30, 1-295 (1936)

12. A. J. Macpherson, M. B. Geuking and K. D. McCoy: Immune responses that adapt the intestinal mucosa to commensal intestinal bacteria. *Immunology* 115(2), 153-162 (2005)

13. A. J. Macpherson and N. Harris: Interactions between commensal intestinal bacteria and the immune system. *Nature Reviews Immunology* 4, 478-485 (2004)

14. L. V. Hooper and A. J. Macpherson: Immune adaptations that maintain homeostasis with the intestinal microbiota. *Nat Rev Immunol* 10(3), 159-169 (2010)

15. K. D. McCoy, N. L. Harris, P. Diener, S. Hatak, B. Odermatt, L. Hangartner, B. M. Senn, B. J. Marsland, M. B. Geuking, H. Hengartner, A. J. Macpherson and R. M. Zinkernagel: Natural IgE Production in the Absence of MHC Class II Cognate Help. *Immunity* 24(3), 329-339 (2006)

16. A. J. Macpherson, D. Gatto, E. Sainsbury, G. R. Harriman, H. Hengartner and R. M. Zinkernagel: A primitive T cell-independent mechanism of intestinal mucosal IgA responses to commensal bacteria. *Science* 288(5474), 2222-2226 (2000)

17. A. Konrad, Y. Cong, W. Duck, R. Borlaza and C. O. Elson: Tight mucosal compartmentation of the murine immune response to antigens of the enteric microbiota. *Gastroenterology* 130(7), 2050-2059 (2006)

18. E. Slack, S. Hapfelmeier, B. Stecher, Y. Velykoredko, M. Stoel, M. A. Lawson, M. B. Geuking, B. Beutler, T. F.

Tedder, W. D. Hardt, P. Bercik, E. F. Verdu, K. D. McCoy and A. J. Macpherson: Innate and adaptive immunity cooperate flexibly to maintain host-microbiota mutualism. *Science* 325(5940), 617-620 (2009)

19. A. J. Macpherson and T. Uhr: Compartmentalization of the mucosal immune responses to commensal intestinal bacteria. *Ann N Y Acad Sci* 1029, 36-43 (2004)

20. S. Hapfelmeier, M. A. Lawson, E. Slack, J. K. Kirundi, M. Stoel, M. Heikenwalder, J. Cahenzli, Y. Velykoredko, M. L. Balmer, K. Endt, M. B. Geuking, R. Curtiss, 3rd, K. D. McCoy and A. J. Macpherson: Reversible microbial colonization of germ-free mice reveals the dynamics of IgA immune responses. *Science* 328(5986), 1705-1709 (2010)

21. M. Rescigno, M. Urbano, B. Valzasina, M. Francolini, G. Rotta, R. Bonasio, F. Granucci, J. P. Kraehenbuhl and P. Ricciardi-Castagnoli: Dendritic cells express tight junction proteins and penetrate gut epithelial monolayers to sample bacteria. *Nat Immunol* 2(4), 361-367. (2001)

22. A. J. Macpherson and T. Uhr: Induction of protective IgA by intestinal dendritic cells carrying commensal bacteria. *Science* 303, 1662-1665 (2004)

23. J. L. Coombes and F. Powrie: Dendritic cells in intestinal immune regulation. *Nat Rev Immunol* 8(6), 435-446 (2008)

24. J. L. Coombes, K. R. Siddiqui, C. V. Arancibia-Carcamo, J. Hall, C. M. Sun, Y. Belkaid and F. Powrie: A functionally specialized population of mucosal CD103+ DCs induces Foxp3+ regulatory T cells via a TGF-beta and retinoic acid-dependent mechanism. *J Exp Med* 204(8), 1757-1764 (2007)

25. C. M. Sun, J. A. Hall, R. B. Blank, N. Bouladoux, M. Oukka, J. R. Mora and Y. Belkaid: Small intestine lamina propria dendritic cells promote de novo generation of Foxp3 T reg cells via retinoic acid. *J Exp Med* 204(8), 1775-1785 (2007)

26. B. Johansson-Lindbom, M. Svensson, O. Pabst, C. Palmqvist, G. Marquez, R. Forster and W. W. Agace: Functional specialization of gut CD103+ dendritic cells in the regulation of tissue-selective T cell homing. *J Exp Med* 202(8), 1063-1073 (2005)

27. E. Jaensson, H. Uronen-Hansson, O. Pabst, B. Eksteen, J. Tian, J. L. Coombes, P. L. Berg, T. Davidsson, F. Powrie, B. Johansson-Lindbom and W. W. Agace: Small intestinal CD103+ dendritic cells display unique functional properties that are conserved between mice and humans. *J Exp Med* 205(9), 2139-2149 (2008)

28. K. Atarashi, J. Nishimura, T. Shima, Y. Umesaki, M. Yamamoto, M. Onoue, H. Yagita, N. Ishii, R. Evans, K. Honda and K. Takeda: ATP drives lamina propria T(H)17 cell differentiation. *Nature* 455(7214), 808-812 (2008)

29. J. H. Niess, S. Brand, X. Gu, L. Landsman, S. Jung, B. A. McCormick, J. M. Vyas, M. Boes, H. L. Ploegh, J. G. Fox, D. R. Littman and H. C. Reinecker: CX3CR1-mediated dendritic cell access to the intestinal lumen and bacterial clearance. *Science* 307(5707), 254-258 (2005)

30. T. Kucharzik, N. Lugering, K. Rautenberg, A. Lugering, M. A. Schmidt, R. Stoll and W. Domschke: Role of M cells in intestinal barrier function. *Ann N Y Acad Sci* 915, 171-183 (2000)

31. M. Chieppa, M. Rescigno, A. Y. Huang and R. N. Germain: Dynamic imaging of dendritic cell extension into the small bowel lumen in response to epithelial cell TLR engagement. *J Exp Med* 203(13), 2841-2852 (2006)

32. A. J. Husband and J. L. Gowans: The origin and antigen-dependent distribution of IgA-containing cells in the intestine. *J Exp Med* 148, 1146-1160 (1978)

33. N. F. Pierce and J. L. Gowans: Cellular kinetics of the intestinal immune response to cholera toxoid in rats. *J Exp Med* 142, 1550-1563 (1975)

34. J. R. Mora, M. Iwata, B. Eksteen, S. Y. Song, T. Junt, B. Senman, K. L. Otipoby, A. Yokota, H. Takeuchi, P. Ricciardi-Castagnoli, K. Rajewsky, D. H. Adams and U. H. von Andrian: Generation of gut-homing IgA-secreting B cells by intestinal dendritic cells. *Science* 314(5802), 1157-1160 (2006)

35. P. Sansonetti: Phagocytosis of bacterial pathogens: implications in the host response. *Semin Immunol* 13(6), 381-390. (2001)

36. P. J. Sansonetti: War and peace at mucosal surfaces. *Nat Rev Immunol* 4(12), 953-964 (2004)

37. M. U. Shiloh, J. D. MacMicking, S. Nicholson, J. E. Brause, S. Potter, M. Marino, F. Fang, M. Dinauer and C. Nathan: Phenotype of mice and macrophages deficient in both phagocyte oxidase and inducible nitric oxide synthase. *Immunity* 10(1), 29-38. (1999)

38. K. Hoebe, X. Du, P. Georgel, E. Janssen, K. Tabeta, S. O. Kim, J. Goode, P. Lin, N. Mann, S. Mudd, K. Crozat, S. Sovath, J. Han and B. Beutler: Identification of Lps2 as a key transducer of MyD88-independent TIR signalling. *Nature* 424(6950), 743-748 (2003)

39. D. K. Podolsky: Inflammatory bowel disease. *N Engl J Med* 347(6), 417-429 (2002)

40. W. Strober, I. Fuss and P. Mannon: The fundamental basis of inflammatory bowel disease. *J Clin Invest* 117(3), 514-521 (2007)

41. H. Mashimo, D. C. Wu, D. K. Podolsky and M. C. Fishman: Impaired defense of intestinal mucosa in mice lacking intestinal trefoil factor. *Science* 274(5285), 262-265 (1996)

42. D. Taupin and D. K. Podolsky: Trefoil factors: initiators of mucosal healing. *Nat Rev Mol Cell Biol* 4(9), 721-732 (2003)

43. T. G. Bush, T. C. Savidge, T. C. Freeman, H. J. Cox, E. A. Campbell, L. Mucke, M. H. Johnson and M. V. Sofroniew: Fulminant jejuno-ileitis following ablation of enteric glia in adult transgenic mice. *Cell* 93(2), 189-201 (1998)

44. M. Steinkamp, I. Geerling, T. Seufferlein, G. von Boyen, B. Egger, J. Grossmann, L. Ludwig, G. Adler and M. Reinshagen: Glial-derived neurotrophic factor regulates apoptosis in colonic epithelial cells. *Gastroenterology* 124(7), 1748-1757 (2003)

45. C. M. Panwala, J. C. Jones and J. L. Viney: A novel model of inflammatory bowel disease: mice deficient for the multiple drug resistance gene, mdr1a, spontaneously develop colitis. *J Immunol* 161(10), 5733-5744 (1998)

46. L. Maggio-Price, D. Shows, K. Waggie, A. Burich, W. Zeng, S. Escobar, P. Morrissey and J. L. Viney: Helicobacter bilis infection accelerates and H. hepaticus infection delays the development of colitis in multiple drug resistance-deficient (mdr1a-/-) mice. *Am J Pathol* 160(2), 739-751 (2002)

47. J. N. Wilk, J. Bilsborough and J. L. Viney: The mdr1a-/- mouse model of spontaneous colitis: a relevant and appropriate animal model to study inflammatory bowel disease. *Immunol Res* 31(2), 151-159 (2005)

48. M. Schwab, E. Schaeffeler, C. Marx, M. F. Fromm, B. Kaskas, J. Metzler, E. Stange, H. Herfarth, J. Schoelmerich, M. Gregor, S. Walker, I. Cascorbi, I. Roots, U. Brinkmann, U. M. Zanger and M. Eichelbaum: Association between the C3435T MDR1 gene polymorphism and susceptibility for ulcerative colitis. *Gastroenterology* 124(1), 26-33 (2003)

49. C. E. Hornquist, X. Lu, P. M. Rogers-Fani, U. Rudolph, S. Shappell, L. Birnbaumer and G. R. Harriman: G(alpha)i2-deficient mice with colitis exhibit a local increase in memory CD4+ T cells and proinflammatory Th1-type cytokines. *J Immunol* 158(3), 1068-1077 (1997)

50. U. Rudolph, M. J. Finegold, S. S. Rich, G. R. Harriman, Y. Srinivasan, P. Brabet, G. Boulay, A. Bradley and L. Birnbaumer: Ulcerative colitis and adenocarcinoma of the colon in G alpha i2-deficient mice. *Nat Genet* 10(2), 143-150 (1995)

51. M. L. Hermiston and J. I. Gordon: Inflammatory bowel disease and adenomas in mice expressing a dominant negative N-cadherin. *Science* 270(5239), 1203-1207 (1995)

52. I. Okayasu, S. Hatakeyama, M. Yamada, T. Ohkusa, Y. Inagaki and R. Nakaya: A novel method in the induction of reliable experimental acute and chronic ulcerative colitis in mice. *Gastroenterology* 98(3), 694-702 (1990)

53. L. A. Dieleman, B. U. Ridwan, G. S. Tennyson, K. W. Beagley, R. P. Bucy and C. O. Elson: Dextran sulfate sodium-induced colitis occurs in severe combined immunodeficient mice. *Gastroenterology* 107(6), 1643-1652 (1994)

54. S. Kitajima, M. Morimoto, E. Sagara, C. Shimizu and Y. Ikeda: Dextran sodium sulfate-induced colitis in germ-free IQI/Jic mice. *Exp Anim* 50(5), 387-395 (2001)

55. K. M. Maslowski, A. T. Vieira, A. Ng, J. Kranich, F. Sierro, D. Yu, H. C. Schilter, M. S. Rolph, F. Mackay, D. Artis, R. J. Xavier, M. M. Teixeira and C. R. Mackay: Regulation of inflammatory responses by gut microbiota and chemoattractant receptor GPR43. *Nature* 461(7268), 1282-1286 (2009)

56. M. F. Neurath, I. Fuss, B. L. Kelsall, E. Stuber and W. Strober: Antibodies to interleukin 12 abrogate established experimental colitis in mice. *J Exp Med* 182(5), 1281-1290 (1995)

57. M. Van der Sluis, B. A. De Koning, A. C. De Bruijn, A. Velcich, J. P. Meijerink, J. B. Van Goudoever, H. A. Buller, J. Dekker, I. Van Seuningen, I. B. Renes and A. W. Einerhand: Muc2-deficient mice spontaneously develop colitis, indicating that MUC2 is critical for colonic protection. *Gastroenterology* 131(1), 117-129 (2006)

58. K. S. Bergstrom, V. Kissoon-Singh, D. L. Gibson, C. Ma, M. Montero, H. P. Sham, N. Ryz, T. Huang, A. Velcich, B. B. Finlay, K. Chadee and B. A. Vallance: Muc2 protects against lethal infectious colitis by disassociating pathogenic and commensal bacteria from the colonic mucosa. *PLoS Pathog* 6(5), e1000902 (2010)

59. A. Velcich, W. Yang, J. Heyer, A. Fragale, C. Nicholas, S. Viani, R. Kucherlapati, M. Lipkin, K. Yang and L. Augenlicht: Colorectal cancer in mice genetically deficient in the mucin Muc2. *Science* 295(5560), 1726-1729 (2002)

60. K. M. Tytgat, F. J. Opdam, A. W. Einerhand, H. A. Buller and J. Dekker: MUC2 is the prominent colonic mucin expressed in ulcerative colitis. *Gut* 38(4), 554-563 (1996)

61. D. K. Podolsky and K. J. Isselbacher: Glycoprotein composition of colonic mucosa. Specific alterations in ulcerative colitis. *Gastroenterology* 87(5), 991-998 (1984)

62. D. K. Podolsky and K. J. Isselbacher: Composition of human colonic mucin. Selective alteration in inflammatory bowel disease. *J Clin Invest* 72(1), 142-153 (1983)

63. J. E. Smithson, A. Campbell, J. M. Andrews, J. D. Milton, R. Pigott and D. P. Jewell: Altered expression of mucins throughout the colon in ulcerative colitis. *Gut* 40(2), 234-240 (1997)

64. P. Alex, M. Ye, N. C. Zachos, J. Sipes, T. Nguyen, M. Suhodrev, L. Gonzales, Z. Arora, T. Zhang, M. Centola, S. E. Guggino and X. Li: Clcn5 knockout mice exhibit novel immunomodulatory effects and are more susceptible to dextran sulfate sodium-induced colitis. *J Immunol* 184(7), 3988-3996 (2010)

65. S. Sullivan, P. Alex, T. Dassopoulos, N. C. Zachos, C. Iacobuzio-Donahue, M. Donowitz, S. R. Brant, C. Cuffari,

M. L. Harris, L. W. Datta, L. Conklin, Y. Chen and X. Li: Downregulation of sodium transporters and NHERF proteins in IBD patients and mouse colitis models: potential contributors to IBD-associated diarrhea. *Inflamm Bowel Dis* 15(2), 261-274 (2009)

66. H. Baribault, J. Penner, R. V. Iozzo and M. Wilson-Heiner: Colorectal hyperplasia and inflammation in keratin 8-deficient FVB/N mice. *Genes Dev* 8(24), 2964-2973 (1994)

67. A. Habtezion, D. M. Toivola, E. C. Butcher and M. B. Omary: Keratin-8-deficient mice develop chronic spontaneous Th2 colitis amenable to antibiotic treatment. *J Cell Sci* 118(Pt 9), 1971-1980 (2005)

68. G. Z. Tao, P. Strnad, Q. Zhou, A. Kamal, L. Zhang, N. D. Madani, S. Kugathasan, S. R. Brant, J. H. Cho, M. B. Omary and R. H. Duerr: Analysis of keratin polypeptides 8 and 19 variants in inflammatory bowel disease. *Clin Gastroenterol Hepatol* 5(7), 857-864 (2007)

69. K. Takeda, B. E. Clausen, T. Kaisho, T. Tsujimura, N. Terada, I. Forster and S. Akira: Enhanced Th1 activity and development of chronic enterocolitis in mice devoid of Stat3 in macrophages and neutrophils. *Immunity* 10(1), 39-49 (1999)

70. M. Kobayashi, M. N. Kweon, H. Kuwata, R. D. Schreiber, H. Kiyono, K. Takeda and S. Akira: Toll-like receptor-dependent production of IL-12p40 causes chronic enterocolitis in myeloid cell-specific Stat3-deficient mice. *J Clin Invest* 111(9), 1297-1308 (2003)

71. E. G. Lee, D. L. Boone, S. Chai, S. L. Libby, M. Chien, J. P. Lodolce and A. Ma: Failure to regulate TNF-induced NF-kappaB and cell death responses in A20-deficient mice. *Science* 289(5488), 2350-2354 (2000)

72. M. Weng, W. A. Walker and I. R. Sanderson: Butyrate regulates the expression of pathogen-triggered IL-8 in intestinal epithelia. *Pediatr Res* 62(5), 542-546 (2007)

73. W. S. Garrett, G. M. Lord, S. Punit, G. Lugo-Villarino, S. K. Mazmanian, S. Ito, J. N. Glickman and L. H. Glimcher: Communicable ulcerative colitis induced by T-bet deficiency in the innate immune system. *Cell* 131(1), 33-45 (2007)

74. M. Vijay-Kumar, C. J. Sanders, R. T. Taylor, A. Kumar, J. D. Aitken, S. V. Sitaraman, A. S. Neish, S. Uematsu, S. Akira, I. R. Williams and A. T. Gewirtz: Deletion of TLR5 results in spontaneous colitis in mice. *J Clin Invest* 117(12), 3909-3921 (2007)

75. A. T. Gewirtz, M. Vijay-Kumar, S. R. Brant, R. H. Duerr, D. L. Nicolae and J. H. Cho: Dominant-negative TLR5 polymorphism reduces adaptive immune response to flagellin and negatively associates with Crohn's disease. *Am J Physiol Gastrointest Liver Physiol* 290(6), G1157-1163 (2006)

76. A. L. Pauleau and P. J. Murray: Role of Nod2 in the response of macrophages to toll-like receptor agonists. *Mol Cell Biol* 23(21), 7531-7539 (2003)

77. T. Watanabe, A. Kitani, P. Murray and W. Strober: NOD2 is a negative regulator of Toll-like receptor 2-mediated T helper type-1 responses. *Nature Immunology* 5, 800-808 (2004)

78. T. Watanabe, A. Kitani, P. J. Murray, Y. Wakatsuki, I. J. Fuss and W. Strober: Nucleotide binding oligomerization domain 2 deficiency leads to dysregulated TLR2 signaling and induction of antigen-specific colitis. *Immunity* 25(3), 473-485 (2006)

79. K. S. Kobayashi, M. Chamaillard, Y. Ogura, O. Henegariu, N. Inohara, G. Nunez and R. A. Flavell: Nod2-dependent regulation of innate and adaptive immunity in the intestinal tract. *Science* 307(5710), 731-734 (2005)

80. T. Petnicki-Ocwieja, T. Hrncir, Y. J. Liu, A. Biswas, T. Hudcovic, H. Tlaskalova-Hogenova and K. S. Kobayashi: Nod2 is required for the regulation of commensal microbiota in the intestine. *Proc Natl Acad Sci U S A* 106(37), 15813-15818 (2009)

81. S. Maeda, L. C. Hsu, H. Liu, L. A. Bankston, M. Iimura, M. F. Kagnoff, L. Eckmann and M. Karin: Nod2 mutation in Crohn's disease potentiates NF-kappaB activity and IL-1beta processing. *Science* 307(5710), 734-738 (2005)

82. R. E. Hammer, S. D. Maika, J. A. Richardson, J. P. Tang and J. D. Taurog: Spontaneous inflammatory disease in transgenic rats expressing HLA-B27 and human beta 2m: an animal model of HLA-B27-associated human disorders. *Cell* 63(5), 1099-1112 (1990)

83. H. C. Rath, H. H. Herfarth, J. S. Ikeda, W. B. Grenther, T. E. Hamm, Jr., E. Balish, J. D. Taurog, R. E. Hammer, K. H. Wilson and R. B. Sartor: Normal luminal bacteria, especially Bacteroides species, mediate chronic colitis, gastritis, and arthritis in HLA-B27/human beta2 microglobulin transgenic rats. *J Clin Invest* 98(4), 945-953 (1996)

84. H. C. Rath, M. Schultz, R. Freitag, L. A. Dieleman, F. Li, H. J. Linde, J. Scholmerich and R. B. Sartor: Different subsets of enteric bacteria induce and perpetuate experimental colitis in rats and mice. *Infect Immun* 69(4), 2277-2285 (2001)

85. T. R. Orchard, H. Holt, L. Bradbury, J. Hammersma, E. McNally, D. P. Jewell and B. P. Wordsworth: The prevalence, clinical features and association of HLA-B27 in sacroiliitis associated with established Crohn's disease. *Aliment Pharmacol Ther* 29(2), 193-197 (2009)

86. S. Steer, H. Jones, J. Hibbert, E. Kondeatis, R. Vaughan, J. Sanderson and T. Gibson: Low back pain, sacroiliitis, and the relationship with HLA-B27 in Crohn's disease. *J Rheumatol* 30(3), 518-522 (2003)

87. D. Kontoyiannis, G. Boulougouris, M. Manoloukos, M. Armaka, M. Apostolaki, T. Pizarro, A. Kotlyarov, I. Forster, R. Flavell, M. Gaestel, P. Tsichlis, F. Cominelli

and G. Kollias: Genetic dissection of the cellular pathways and signaling mechanisms in modeled tumor necrosis factor-induced Crohn's-like inflammatory bowel disease. J Exp Med 196(12), 1563-1574 (2002)

88. D. Kontoyiannis, M. Pasparakis, T. T. Pizarro, F. Cominelli and G. Kollias: Impaired on/off regulation of TNF biosynthesis in mice lacking TNF AU-rich elements: implications for joint and gut-associated immunopathologies. Immunity 10(3), 387-398 (1999)

89. M. Watanabe, Y. Ueno, T. Yajima, S. Okamoto, T. Hayashi, M. Yamazaki, Y. Iwao, H. Ishii, S. Habu, M. Uehira, H. Nishimoto, H. Ishikawa, J. Hata and T. Hibi: Interleukin 7 transgenic mice develop chronic colitis with decreased interleukin 7 protein accumulation in the colonic mucosa. J Exp Med 187(3), 389-402 (1998)

90. Y. Nemoto, T. Kanai, K. Kameyama, T. Shinohara, N. Sakamoto, T. Totsuka, R. Okamoto, K. Tsuchiya, T. Nakamura, T. Sudo, S. Matsumoto and M. Watanabe: Long-lived colitogenic CD4+ memory T cells residing outside the intestine participate in the perpetuation of chronic colitis. *J Immunol* 183(8), 5059-5068 (2009)

91. C. A. Anderson, G. Boucher, C. W. Lees, A. Franke, M. D'Amato, K. D. Taylor, J. C. Lee, P. Goyette, M. Imielinski, A. Latiano, C. Lagace, R. Scott, L. Amininejad, S. Bumpstead, L. Baidoo, R. N. Baldassano, M. Barclay, T. M. Bayless, S. Brand, C. Buning, J. F. Colombel, L. A. Denson, M. De Vos, M. Dubinsky, C. Edwards, D. Ellinghaus, R. S. Fehrmann, J. A. Floyd, T. Florin, D. Franchimont, L. Franke, M. Georges, J. Glas, N. L. Glazer, S. L. Guthery, T. Haritunians, N. K. Hayward, J. P. Hugot, G. Jobin, D. Laukens, I. Lawrance, M. Lemann, A. Levine, C. Libioulle, E. Louis, D. P. McGovern, M. Milla, G. W. Montgomery, K. I. Morley, C. Mowat, A. Ng, W. Newman, R. A. Ophoff, L. Papi, O. Palmieri, L. Peyrin-Biroulet, J. Panes, A. Phillips, N. J. Prescott, D. D. Proctor, R. Roberts, R. Russell, P. Rutgeerts, J. Sanderson, M. Sans, P. Schumm, F. Seibold, Y. Sharma, L. A. Simms, M. Seielstad, A. H. Steinhart, S. R. Targan, L. H. van den Berg, M. Vatn, H. Verspaget, T. Walters, C. Wijmenga, D. C. Wilson, H. J. Westra, R. J. Xavier, Z. Z. Zhao, C. Y. Ponsioen, V. Andersen, L. Torkvist, M. Gazouli, N. P. Anagnou, T. H. Karlsen, L. Kupcinskas, J. Sventoraityte, J. C. Mansfield, S. Kugathasan, M. S. Silverberg, J. Halfvarson, J. I. Rotter, C. G. Mathew, A. M. Griffiths, R. Gearry, T. Ahmad, S. R. Brant, M. Chamaillard, J. Satsangi, J. H. Cho, S. Schreiber, M. J. Daly, J. C. Barrett, M. Parkes, V. Annese, H. Hakonarson, G. Radford-Smith, R. H. Duerr, S. Vermeire, R. K. Weersma and J. D. Rioux: Meta-analysis identifies 29 additional ulcerative colitis risk loci, increasing the number of confirmed associations to 47. *Nat Genet* 43(3), 246-252 (2011)

92. M. M. Shull, I. Ormsby, A. B. Kier, S. Pawlowski, R. J. Diebold, M. Yin, R. Allen, C. Sidman, G. Proetzel, D. Calvin and *et al.*: Targeted disruption of the mouse transforming growth factor-beta 1 gene results in multifocal inflammatory disease. *Nature* 359(6397), 693-699 (1992)

93. S. J. Engle, I. Ormsby, S. Pawlowski, G. P. Boivin, J. Croft, E. Balish and T. Doetschman: Elimination of colon cancer in germ-free transforming growth factor beta 1-deficient mice. *Cancer Res* 62(22), 6362-6366 (2002)

94. A. Di Sabatino, K. M. Pickard, D. Rampton, L. Kruidenier, L. Rovedatti, N. A. Leakey, G. R. Corazza, G. Monteleone and T. T. MacDonald: Blockade of transforming growth factor beta upregulates T-box transcription factor T-bet, and increases T helper cell type 1 cytokine and matrix metalloproteinase-3 production in the human gut mucosa. *Gut* 57(5), 605-612 (2008)

95. G. Monteleone, M. Boirivant, F. Pallone and T. T. MacDonald: TGF-beta1 and Smad7 in the regulation of IBD. *Mucosal Immunol* 1 Suppl 1, S50-53 (2008)

96. L. Gorelik and R. A. Flavell: Abrogation of TGFbeta signaling in T cells leads to spontaneous T cell differentiation and autoimmune disease. *Immunity* 12(2), 171-181 (2000)

97. L. Fahlen, S. Read, L. Gorelik, S. D. Hurst, R. L. Coffman, R. A. Flavell and F. Powrie: T cells that cannot respond to TGF-beta escape control by CD4(+)CD25(+) regulatory T cells. *J Exp Med* 201(5), 737-746 (2005)

98. K. B. Hahm, Y. H. Im, T. W. Parks, S. H. Park, S. Markowitz, H. Y. Jung, J. Green and S. J. Kim: Loss of transforming growth factor beta signalling in the intestine contributes to tissue injury in inflammatory bowel disease. *Gut* 49(2), 190-198 (2001)

99. R. Kuhn, J. Lohler, D. Rennick, K. Rajewsky and W. Muller: Interleukin-10-deficient mice develop chronic enterocolitis. *Cell* 75(2), 263-274 (1993)

100. K. L. Madsen, J. S. Doyle, M. M. Tavernini, L. D. Jewell, R. P. Rennie and R. N. Fedorak: Antibiotic therapy attenuates colitis in interleukin 10 gene-deficient mice. *Gastroenterology* 118(6), 1094-1105 (2000)

101. R. K. Sellon, S. Tonkonogy, M. Schultz, L. A. Dieleman, W. Grenther, E. Balish, D. M. Rennick and R. B. Sartor: Resident enteric bacteria are necessary for development of spontaneous colitis and immune system activation in interleukin-10-deficient mice. *Infect Immun* 66(11), 5224-5231 (1998)

102. S. Rakoff-Nahoum, L. Hao and R. Medzhitov: Role of toll-like receptors in spontaneous commensal-dependent colitis. *Immunity* 25(2), 319-329 (2006)

103. I. Correa, M. Veny, M. Esteller, J. M. Pique, J. Yague, J. Panes and A. Salas: Defective IL-10 production in severe phenotypes of Crohn's disease. *J Leukoc Biol* 85(5), 896-903 (2009)

104. S. Schreiber, T. Heinig, H. G. Thiele and A. Raedler: Immunoregulatory role of interleukin 10 in patients with inflammatory bowel disease. *Gastroenterology* 108(5), 1434-1444 (1995)

105. A. Franke, T. Balschun, T. H. Karlsen, J. Sventoraityte, S. Nikolaus, G. Mayr, F. S. Domingues, M.

Albrecht, M. Nothnagel, D. Ellinghaus, C. Sina, C. M. Onnie, R. K. Weersma, P. C. Stokkers, C. Wijmenga, M. Gazouli, D. Strachan, W. L. McArdle, S. Vermeire, P. Rutgeerts, P. Rosenstiel, M. Krawczak, M. H. Vatn, C. G. Mathew and S. Schreiber: Sequence variants in IL10, ARPC2 and multiple other loci contribute to ulcerative colitis susceptibility. *Nat Genet* 40(11), 1319-1323 (2008)

106. V. Andersen, A. Ernst, J. Christensen, M. Ostergaard, B. A. Jacobsen, A. Tjonneland, H. B. Krarup and U. Vogel: The polymorphism rs3024505 proximal to IL-10 is associated with risk of ulcerative colitis and Crohns disease in a Danish case-control study. *BMC Med Genet* 11, 82 (2010)

107. A. Franke, D. P. McGovern, J. C. Barrett, K. Wang, G. L. Radford-Smith, T. Ahmad, C. W. Lees, T. Balschun, J. Lee, R. Roberts, C. A. Anderson, J. C. Bis, S. Bumpstead, D. Ellinghaus, E. M. Festen, M. Georges, T. Green, T. Haritunians, L. Jostins, A. Latiano, C. G. Mathew, G. W. Montgomery, N. J. Prescott, S. Raychaudhuri, J. I. Rotter, P. Schumm, Y. Sharma, L. A. Simms, K. D. Taylor, D. Whiteman, C. Wijmenga, R. N. Baldassano, M. Barclay, T. M. Bayless, S. Brand, C. Buning, A. Cohen, J. F. Colombel, M. Cottone, L. Stronati, T. Denson, M. De Vos, R. D'Inca, M. Dubinsky, C. Edwards, T. Florin, D. Franchimont, R. Gearry, J. Glas, A. Van Gossum, S. L. Guthery, J. Halfvarson, H. W. Verspaget, J. P. Hugot, A. Karban, D. Laukens, I. Lawrance, M. Lemann, A. Levine, C. Libioulle, E. Louis, C. Mowat, W. Newman, J. Panes, A. Phillips, D. D. Proctor, M. Regueiro, R. Russell, P. Rutgeerts, J. Sanderson, M. Sans, F. Seibold, A. H. Steinhart, P. C. Stokkers, L. Torkvist, G. Kullak-Ublick, D. Wilson, T. Walters, S. R. Targan, S. R. Brant, J. D. Rioux, M. D'Amato, R. K. Weersma, S. Kugathasan, A. M. Griffiths, J. C. Mansfield, S. Vermeire, R. H. Duerr, M. S. Silverberg, J. Satsangi, S. Schreiber, J. H. Cho, V. Annese, H. Hakonarson, M. J. Daly and M. Parkes: Genome-wide meta-analysis increases to 71 the number of confirmed Crohn's disease susceptibility loci. *Nat Genet* 42(12), 1118-1125 (2010)

108. E. O. Glocker, D. Kotlarz, K. Boztug, E. M. Gertz, A. A. Schaffer, F. Noyan, M. Perro, J. Diestelhorst, A. Allroth, D. Murugan, N. Hatscher, D. Pfeifer, K. W. Sykora, M. Sauer, H. Kreipe, M. Lacher, R. Nustede, C. Woellner, U. Baumann, U. Salzer, S. Koletzko, N. Shah, A. W. Segal, A. Sauerbrey, S. Buderus, S. B. Snapper, B. Grimbacher and C. Klein: Inflammatory bowel disease and mutations affecting the interleukin-10 receptor. *N Engl J Med* 361(21), 2033-2045 (2009)

109. B. Sadlack, H. Merz, H. Schorle, A. Schimpl, A. C. Feller and I. Horak: Ulcerative colitis-like disease in mice with a disrupted interleukin-2 gene. *Cell* 75(2), 253-261 (1993)

110. N. V. Contractor, H. Bassiri, T. Reya, A. Y. Park, D. C. Baumgart, M. A. Wasik, S. G. Emerson and S. R. Carding: Lymphoid hyperplasia, autoimmunity, and compromised intestinal intraepithelial lymphocyte development in colitis-free gnotobiotic IL-2-deficient mice. *J Immunol* 160(1), 385-394 (1998)

111. M. Schultz, S. L. Tonkonogy, R. K. Sellon, C. Veltkamp, V. L. Godfrey, J. Kwon, W. B. Grenther, E. Balish, I. Horak and R. B. Sartor: IL-2-deficient mice raised under germfree conditions develop delayed mild focal intestinal inflammation. *Am J Physiol* 276(6 Pt 1), G1461-1472 (1999)

112. D. M. Willerford, J. Chen, J. A. Ferry, L. Davidson, A. Ma and F. W. Alt: Interleukin-2 receptor alpha chain regulates the size and content of the peripheral lymphoid compartment. *Immunity* 3(4), 521-530 (1995)

113. S. Erdman, J. G. Fox, C. A. Dangler, D. Feldman and B. H. Horwitz: Typhlocolitis in NF-kappa B-deficient mice. *J Immunol* 166(3), 1443-1447 (2001)

114. I. Atreya, R. Atreya and M. F. Neurath: NF-kappaB in inflammatory bowel disease. *J Intern Med* 263(6), 591-596 (2008)

115. C. H. Clegg, J. T. Rulffes, H. S. Haugen, I. H. Hoggatt, A. Aruffo, S. K. Durham, A. G. Farr and D. Hollenbaugh: Thymus dysfunction and chronic inflammatory disease in gp39 transgenic mice. *Int Immunol* 9(8), 1111-1122 (1997)

116. O. Ludwiczek, A. Kaser and H. Tilg: Plasma levels of soluble CD40 ligand are elevated in inflammatory bowel diseases. *Int J Colorectal Dis* 18(2), 142-147 (2003)

117. P. Mombaerts, E. Mizoguchi, M. J. Grusby, L. H. Glimcher, A. K. Bhan and S. Tonegawa: Spontaneous development of inflammatory bowel disease in T cell receptor mutant mice. *Cell*, 75(2), 274-282 (1993)

118. A. K. Bhan, E. Mizoguchi, R. N. Smith and A. Mizoguchi: Spontaneous chronic colitis in TCR alpha-mutant mice; an experimental model of human ulcerative colitis. *Int Rev Immunol* 19(1), 123-138 (2000)

119. L. Dianda, A. M. Hanby, N. A. Wright, A. Sebesteny, A. C. Hayday and M. J. Owen: T cell receptor-alpha beta-deficient mice fail to develop colitis in the absence of a microbial environment. *Am J Pathol* 150(1), 91-97 (1997)

120. G. A. Hollander, S. J. Simpson, E. Mizoguchi, A. Nichogiannopoulou, J. She, J. C. Gutierrez-Ramos, A. K. Bhan, S. J. Burakoff, B. Wang and C. Terhorst: Severe colitis in mice with aberrant thymic selection. *Immunity* 3(1), 27-38 (1995)

121. C. Veltkamp, S. L. Tonkonogy, Y. P. De Jong, C. Albright, W. B. Grenther, E. Balish, C. Terhorst and R. B. Sartor: Continuous stimulation by normal luminal bacteria is essential for the development and perpetuation of colitis in Tg(epsilon26) mice. *Gastroenterology* 120(4), 900-913 (2001)]

122. S. Wirtz, S. Finotto, S. Kanzler, A. W. Lohse, M. Blessing, H. A. Lehr, P. R. Galle and M. F. Neurath:

Cutting edge: chronic intestinal inflammation in STAT-4 transgenic mice: characterization of disease and adoptive transfer by TNF- plus IFN-gamma-producing CD4+ T cells that respond to bacterial antigens. *J Immunol* 162(4), 1884-1888 (1999)

123. F. Powrie, M. W. Leach, S. Mauze, L. B. Caddle and R. L. Coffman: Phenotypically distinct subsets of CD4+ T cells induce or protect from chronic intestinal inflammation in C. B-17 scid mice. *Int Immunol* 5(11), 1461-1471 (1993)

124. M. Boirivant, I. J. Fuss, A. Chu and W. Strober: Oxazolone colitis: A murine model of T helper cell type 2 colitis treatable with antibodies to interleukin 4. *J Exp Med* 188(10), 1929-1939 (1998)

125. S. B. Snapper, F. S. Rosen, E. Mizoguchi, P. Cohen, W. Khan, C. H. Liu, T. L. Hagemann, S. P. Kwan, R. Ferrini, L. Davidson, A. K. Bhan and F. W. Alt: Wiskott-Aldrich syndrome protein-deficient mice reveal a role for WASP in T but not B cell activation. *Immunity* 9(1), 81-91 (1998)

126. D. D. Nguyen, M. H. Maillard, V. Cotta-de-Almeida, E. Mizoguchi, C. Klein, I. Fuss, C. Nagler, A. Mizoguchi, A. K. Bhan and S. B. Snapper: Lymphocyte-dependent and Th2 cytokine-associated colitis in mice deficient in Wiskott-Aldrich syndrome protein. *Gastroenterology* 133(4), 1188-1197 (2007)

127. M. H. Maillard, V. Cotta-de-Almeida, F. Takeshima, D. D. Nguyen, P. Michetti, C. Nagler, A. K. Bhan and S. B. Snapper: The Wiskott-Aldrich syndrome protein is required for the function of CD4(+)CD25(+)Foxp3(+) regulatory T cells. *J Exp Med* 204(2), 381-391 (2007)

128. S. Humblet-Baron, B. Sather, S. Anover, S. Becker-Herman, D. J. Kasprowicz, S. Khim, T. Nguyen, K. Hudkins-Loya, C. E. Alpers, S. F. Ziegler, H. Ochs, T. Torgerson, D. J. Campbell and D. J. Rawlings: Wiskott-Aldrich syndrome protein is required for regulatory T cell homeostasis. *J Clin Invest* 117(2), 407-418 (2007)

129. D. D. Nguyen, M. A. Eston, S. Muthupalani, M. W. Mobley, N. S. Taylor, S. B. Snapper and J. B. Fox: T1792 Helicobacter is Required for Colitis in WASP-Deficient Mice and Induces Colon Cancer. *Gastroenterology* 138(5), S-579-S-578 (2010)

130. K. H. Hsieh, M. H. Chang, C. Y. Lee and C. Y. Wang: Wiskott-Aldrich syndrome and inflammatory bowel disease. *Ann Allergy* 60(5), 429-431 (1988)

131. C. Folwaczny, C. Ruelfs, J. Walther, A. Konig and B. Emmerich: Ulcerative colitis in a patient with Wiskott-Aldrich syndrome. *Endoscopy* 34(10), 840-841 (2002)

132. G. Kim, O. Turovskaya, M. Levin, F. R. Byrne, J. S. Whoriskey, J. G. McCabe and M. Kronenberg: Spontaneous colitis occurrence in transgenic mice with altered B7-mediated costimulation. *J Immunol* 181(8), 5278-5288 (2008)

133. G. Q. Phan, J. C. Yang, R. M. Sherry, P. Hwu, S. L. Topalian, D. J. Schwartzentruber, N. P. Restifo, L. R. Haworth, C. A. Seipp, L. J. Freezer, K. E. Morton, S. A. Mavroukakis, P. H. Duray, S. M. Steinberg, J. P. Allison, T. A. Davis and S. A. Rosenberg: Cancer regression and autoimmunity induced by cytotoxic T lymphocyte-associated antigen 4 blockade in patients with metastatic melanoma. *Proc Natl Acad Sci U S A* 100(14), 8372-8377 (2003)

134. J. P. Sundberg, C. O. Elson, H. Bedigian and E. H. Birkenmeier: Spontaneous, heritable colitis in a new substrain of C3H/HeJ mice. *Gastroenterology* 107(6), 1726-1735 (1994)

135. Y. Cong, S. L. Brandwein, R. P. McCabe, A. Lazenby, E. H. Birkenmeier, J. P. Sundberg and C. O. Elson: CD4+ T cells reactive to enteric bacterial antigens in spontaneously colitic C3H/HeJBir mice: increased T helper cell type 1 response and ability to transfer disease. *J Exp Med* 187(6), 855-864 (1998)

136. S. L. Brandwein, R. P. McCabe, Y. Cong, K. B. Waites, B. U. Ridwan, P. A. Dean, T. Ohkusa, E. H. Birkenmeier, J. P. Sundberg and C. O. Elson: Spontaneously colitic C3H/HeJBir mice demonstrate selective antibody reactivity to antigens of the enteric bacterial flora. *J Immunol* 159(1), 44-52 (1997)

137. H. R. Gaskins, G. L. Vondrak-Juergens, B. A. McCracken and J. H. Woolsey: Specific-pathogen-free conditions enhance inflammatory bowel disease in T-cell receptor knockout, but not C3H/HeJBir mice. *Lab Anim Sci* 47(6), 650-655 (1997)

138. S. Matsumoto, Y. Okabe, H. Setoyama, K. Takayama, J. Ohtsuka, H. Funahashi, A. Imaoka, Y. Okada and Y. Umesaki: Inflammatory bowel disease-like enteritis and caecitis in a senescence accelerated mouse P1/Yit strain. *Gut* 43(1), 71-78 (1998)

139. M. M. Kosiewicz, C. C. Nast, A. Krishnan, J. Rivera-Nieves, C. A. Moskaluk, S. Matsumoto, K. Kozaiwa and F. Cominelli: Th1-type responses mediate spontaneous ileitis in a novel murine model of Crohn's disease. *J Clin Invest* 107(6), 695-702 (2001)

140. K. Sugawara, T. S. Olson, C. A. Moskaluk, B. K. Stevens, S. Hoang, K. Kozaiwa, F. Cominelli, K. F. Ley and M. McDuffie: Linkage to peroxisome proliferator-activated receptor-gamma in SAMP1/YitFc mice and in human Crohn's disease. *Gastroenterology* 128(2), 351-360 (2005)

Key Words: Commensal Microflora, Host-Microbial Mutualism, Innate Immunity, Adaptive Immunity, Compartmentalization, Iga, Mucosal Immune System, Germ-Free, Anti-Commensal Immunity, Review

Send correspondence to: Kathy D. McCoy, Department of Clinical Research, Room C817, Murtenstrasse 35, 3010 Bern, Switzerland, Tel: 41 31 632 0931, Fax: 41 31 632 32 97, E-mail: mccoy@dkf.unibe.ch

http://www.bioscience.org/current/vol4S.htm