LEADING ARTICLE

Endocannabinoid overactivity and intestinal inflammation

V Di Marzo, A A Izzo

Gut 2006;**55**:1373–1376. doi: 10.1136/gut.2005.090472

Cannabinoid receptors of type 1 and 2 (CB_1 and CB_2), endogenous ligands that activate them (endocannabinoids), and mechanisms for endocannabinoid biosynthesis and inactivation have been identified in the gastrointestinal system. Activation of CB_1 receptors by endocannabinoids produces relaxation of the lower oesophageal sphincter and inhibition of gastric acid secretion, intestinal motility, and fluid stimulated secretion. However, stimulation of cannabinoid receptors impacts on gastrointestinal functions in several other ways. Recent data indicate that the endocannabinoid system in the small intestine and colon becomes over stimulated during inflammation in both animal models and human inflammatory disorders. The pathological significance of this ''endocannabinoid overactivity'' and its possible exploitation for therapeutic purposes are discussed here.

See end of article for authors' affiliations

Correspondence to: Dr V Di Marzo, Endocannabinoid Research Group, Institute of Biomolecular Chemistry, Consiglio Nazionale delle Ricerche, via Campi Flegrei 34, 80078 Pozzuoli, Napoli, Italy; vdimarzo@icmib.na.cnr.it

Revised version received 10 April 2006
Accepted for publication 9 May 2006

The main psychotropic constituent of the plant *Cannabis sativa* and marijuana, Δ^9-tetrahydrocannabinol, exerts its pharmacological effects by activating two G protein coupled cannabinoid receptors.[1] These are the CB_1 receptor, present in central and peripheral nerves (including the human enteric nervous system), and the CB_2 receptor, expressed abundantly in immune cells. In rodents, CB_1 receptor immunoreactivity has been detected in discrete nuclei of the dorsovagal complex (involved in emesis), and in efferents from the vagal ganglia and in enteric (myenteric and submucosal) nerve terminals where they inhibit excitatory (mainly cholinergic) neurotransmission.[2–5] In vivo pharmacological studies have shown that activation of CB_1 receptors reduces emesis,[6 7] produces inhibition of gastric acid secretion[8] and relaxation of the lower oesophageal sphincter [9] (two effects that might be beneficial in the treatment of gastro-oesophageal reflux disease), and inhibits intestinal motility and secretion.[10 11] Consistent with immunohistochemical data showing that CB_2 receptors are particularly evident in colonic tissues from patients with inflammatory bowel diseases (IBD),[12] evidence suggests that CB_2 inhibits intestinal motility during certain pathological states.[13]

The endocannabinoid system of the gastrointestinal tract includes not only cannabinoid receptors but also endogenous agonists of these receptors (that is, the endocannabinoids anandamide and 2-arachidonoylglycerol (2-AG)), as well as mechanisms for their biosynthesis and inactivation. The latter occurs via cellular reuptake, which is facilitated by a putative membrane transporter, and enzymatic degradation by fatty acid amide hydrolase (FAAH) for both anandamide and 2-AG, and by monoacylglycerol lipase for 2-AG. The endocannabinoids have been detected in the digestive tract and there is evidence that at least anandamide is a physiological regulator of colonic propulsion in mice.[10] This is consistent with data from phase III clinical trials that highlighted diarrhoea as one of the initial adverse events associated with administration of the antiobesity drug rimonabant, a selective CB_1 receptor antagonist.[14] Intestinal anandamide levels have been found to be increased after noxious stimuli, food deprivation, or clinically diagnosed colorectal cancer, thus suggesting a possible physiopathological role.[11 15–17] In rodents, endocannabinoids convey protection from enteric hypersecretory states (for example, cholera toxin induced diarrhoea), which is in agreement with anecdotal reports from folk medicine on the use of *Cannabis sativa* in the treatment of diarrhoea.[11]

> ''The endocannabinoid system of the gastrointestinal tract includes not only cannabinoid receptors but also endogenous agonists of these receptors, as well as mechanisms for their biosynthesis and inactivation''

Endocannabinoids may also have targets other than cannabinoid receptors.[18] The best characterised is the transient receptor potential vanilloid type 1 (TRPV1) receptor (the molecular target for the pungent component of hot chilli, capsaicin), which is mostly expressed by primary afferent neurones but also detected in myenteric and submucosal nerves.[19] TRPV1 can be activated by anandamide, thus resulting in enteritis in the rat in vivo[20] and enhanced acetylcholine release from myenteric guinea pig nerves.[21] However, under physiological conditions, anandamide reduces mouse intestinal transit in vivo through activation of CB_1, but not TRPV1, receptors.[22]

A high affinity binding site potentially involved in cellular reuptake of endocannabinoids has only recently been characterised in rat basophilic cells.[23] There is no direct evidence for the existence of this putative protein in the gut as yet, although functional studies performed in mice using specific inhibitors of anandamide

Abbreviations: 2-AG, 2-arachidonoylglycerol; CB_1 and CB_2, cannabinoid receptors of type 1 and 2; DNBS, dinitrobenzene sulphonic acid; FAAH, fatty acid amide hydrolase; IBD, inflammatory bowel diseases; LPS, lipopolysaccharide; TNF-α, tumour necrosis factor α; TRPV1, transient receptor potential vanilloid type 1 channel

reuptake suggest that this process might be involved in the control of motility changes associated with experimental ileus[16] and in the secretory diarrhoea evoked by cholera toxin.[11] FAAH mRNA and activity have been detected in different regions of the rodent intestinal tract and functional studies performed using selective inhibitors suggest that this enzyme is physiologically involved in the control of intestinal motility.[24 25]

ENDOCANNABINOID OVERACTIVITY DURING INTESTINAL INFLAMMATORY CONDITIONS

Evidence is accumulating to suggest that during inflammatory conditions affecting the intestine, as with other disorders,[1 26] the tone of the endocannabinoid system is increased because of either increased expression of cannabinoid receptors or upregulation of endocannabinoid levels, or both. During croton oil induced inflammation and subsequent increase in upper gastrointestinal transit, expression of CB_1 receptors in the mouse small intestine is enhanced and so is the inhibitory effect on motility observed following activation of these receptors.[27] Also, the activity of the enzyme responsible for anandamide degradation, FAAH,[28] was found to be increased, suggesting that enhanced turnover of anandamide occurs during croton oil induced inflammation. Massa and colleagues[29] showed that CB_1 receptor expression is increased in the colon of mice treated with intrarectal dinitrobenzene sulphonic acid (DNBS), an experimental model of colitis, and that genetic or pharmacological blockade of CB_1 receptors causes worsening, whereas genetic ablation of FAAH causes amelioration, of the colon inflammatory score of these animals. More recently, D'Argenio and colleagues[30] found that in the colon of DNBS treated mice and trinitrobenzene sulphonic acid treated rats, levels of anandamide, but not 2-AG, were significantly increased. More importantly, further elevation of the amounts of this endocannabinoid, obtained by systemic administration of an inhibitor of anandamide cellular reuptake, was accompanied by complete reversal of histological and biochemical inflammatory parameters in the colon of DNBS treated mice.[30] These findings, taken together, indicate that:

- endocannabinoids and CB_1 receptors are upregulated during intestinal inflammation;
- enhanced endocannabinoid tone, by acting at least in part through CB_1 receptors, affords protection against both epithelial damage and increased motility occurring during intestinal inflammation.

Depending on the type of inflammatory stimulus, CB_1 receptors may not be the only molecular targets involved in the protective functions of endocannabinoids. Mathison and colleagues[13] showed that the increase in gastrointestinal transit caused by lipopolysaccharide (LPS) induced inflammation in rats can be selectively counteracted by agonists of CB_2, but not CB_1, receptors. The effect was independent of nitric oxide but appeared to be mediated by cyclooxygenase derived products as it was attenuated by indomethacin. No experiment was performed by the authors to investigate whether CB_2 agonists exert any direct anti-inflammatory effect, as would be expected from the fact that activation of CB_2 receptors causes inhibition of proinflammatory cytokines.[31] Indeed, CB_2 receptor agonists inhibit tumour necrosis factor α (TNF-α) induced interleukin 8 release in human colonic epithelial cells, which are recognised to exert a major influence on maintenance of intestinal immune homeostasis.[32] On the other hand, in a different study, the CB_1 receptor antagonist rimonabant was found to inhibit the LPS induced increase in plasma levels of TNF-α in rats and wild-type mice, but not in CB_1 receptor null mice.[33] This paradoxical effect of CB_1 blockade might be due to the unmasking of CB_2 mediated anti-inflammatory effects exerted by enhanced endocannabinoid levels when CB_1 receptors are blocked, although this possibility has not yet been investigated.

> "Depending on the type of inflammatory stimulus, CB_1 receptors may not be the only molecular targets involved in the protective functions of endocannabinoids"

More recently, the involvement in colon inflammation of another possible target of anandamide, the TRPV1 receptor, was also evaluated.[34] Previous studies had shown that acute activation of this cation channel can contribute to intestinal inflammation,[35–37] and that when TRPV1 agonists cause anti-inflammatory effects they likely do so by causing desensitisation of these receptors.[38] It was also shown that, following toxin A induced inflammation of the rat small intestine, anandamide levels are upregulated in this tissue and contribute towards worsening of the inflammatory score by activating TRPV1 receptors.[20] Therefore, the finding of Massa *et al* that infusion of DNBS induced increased inflammation in $TRPV1^{-/-}$ mice compared with wild-type littermates ($TRPV1^{+/+}$) was quite unexpected. Electrophysiological recordings from circular smooth muscle cells, performed 8 and 24 hours after DNBS treatment, revealed strong spontaneous oscillatory action potentials in $TRPV1^{-/-}$ but not in $TRPV1^{+/+}$ colons, indicating an early TRPV1 mediated control of inflammation induced irritation of smooth muscle activities rather than of epithelial cell damage. These results suggest that TRPV1 receptors, possibly by being activated by elevated anandamide levels observed in the colon following DNBS treatment,[30] may also afford endogenous protection against colonic inflammation induced experimentally. Overall, these studies in experimental models of intestinal inflammation indicate that:

- targets other than CB_1 receptors (that is, TRPV1 and CB_2 receptors) participate in endocannabinoid induced anti-inflammatory effects in the gastrointestinal tract;
- the same endocannabinoid target (for example, the TRPV1 receptor) may play protective or counterprotective roles in intestinal inflammation depending on the intestine section under study and the type of experimental animal model used (that is, chemically induced *v* bacteria induced inflammation, respectively).

> "Overactivity of the endocannabinoid system is becoming a well established concept in human intestinal conditions with an inflammatory component"

Even with these sometimes discrepant results from animal studies, overactivity of the endocannabinoid system is also becoming a well established concept in human intestinal conditions with an inflammatory component. Significantly elevated CB_2 receptor expression and anandamide levels were reported in colon biopsies from patients with ulcerative colitis,[12 30] and elevated anandamide concentrations have been observed in intestinal samples from patients with diverticulosis,[39] and in biopsies from patients with coeliac disease in the atrophic phase (Di Marzo V, Gianfrani C, Mazzarella G, and Sorrentini I, unpublished data). In the latter case, anandamide levels were found to return to normal following remission, thus suggesting that in humans, elevation of endocannabinoid (usually anandamide) intestinal levels represents an adaptive response aimed at providing

protection from inflammation. How such protection is obtained and through which of the many endocannabinoid targets is still a matter of speculation. Importantly, activation of CB_1 receptors might limit the effects of intestinal inflammation not only by regulating the activity of myenteric neurones[27 34 39] but also by inducing wound closure in human colon epithelium in vitro, which is consistent with the presence of CB_1 receptors in human colonic epithelial cells.[12 17] Finally, human TRPV1 immunoreactivity is increased in the hypertrophic extrinsic nerve bundles in Hirschsprung's disease,[40] which is important in the light of the observation that anandamide activates TRPV1 receptors more efficaciously when such receptors are overexpressed.[18]

CONCLUSIONS: NEW THERAPIES FOR THE TREATMENT OF IBD FROM THE ENDOCANNABINOID SYSTEM

The inhibitory effects of cannabinoids on intestinal inflammation, as well as on intestinal motility and secretory diarrhoea, observed in preclinical studies, increase the potential for their use in the treatment of IBD. In fact, based on these data in animal studies, a clinical study with *Cannabis* in patients with relapse of chronic intermittent Crohn's disease has been started at the University Hospital of Munich.[41] Particular attention will have to be paid during these studies to potential ''central'' side effects of *Cannabis*, such as tolerance and proconvulsant effects.[42] Regarding the endocannabinoids, although the exact mechanisms of their anti-inflammatory effects remain elusive, it is well established that they might be effective in relieving a number of symptoms experienced by patients with IBD, including nausea, anorexia, cramps, diarrhoea, pain, and inflammation. It appears that endocannabinoids might regulate the intestinal response to inflammation at three levels: (1) reducing the release of neurotransmitters that affect intestinal motility and secretion; (2) directly suppressing the production of proinflammatory mediators such as TNF-α; and (3) promoting epithelial wound healing (fig 1). From our present knowledge, two possible strategies might be envisaged for the endocannabinoid based pharmacological inhibition of bowel inflammation without provoking psychotropic side effects such as those of marijuana, largely mediated by CB_1 receptors in the brain:

- because activation of both CB_1 and CB_2 receptors is expected to elicit protective effects, the design of CB1/CB2 cannabinoid receptor agonists that do not cross the blood-barrier may reduce intestinal inflammation and associated diarrhoea through activation of enteric cannabinoid receptors (see also Kimball and colleagues[43] for a further example of the role played by both CB_1 and CB_2 receptors against chemically induced colitis and diarrhoea);
- the use of inhibitors of endocannabinoid inactivation by increasing levels of anandamide only ''where and when'' this is upregulated (that is, in the intestine during inflammation), will have greater site and time selectivity than drugs directly acting on cannabinoid receptors ''always and everywhere''.

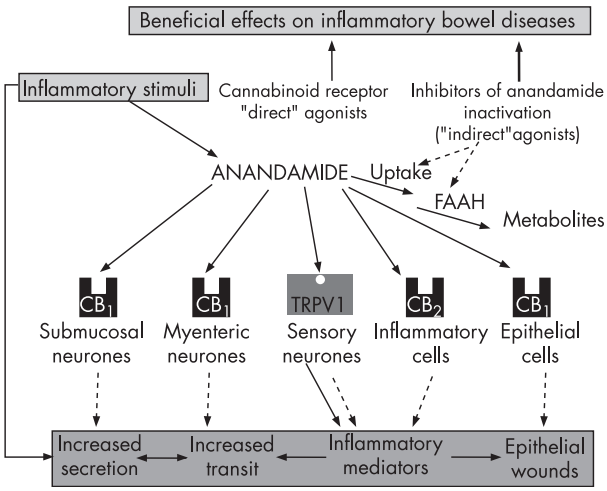

Figure 1 Endocannabinoid control of intestinal inflammation. Inflammatory stimuli upregulate anandamide levels and, in some cases, its major molecular targets, the cannabinoid CB_1 and CB_2 receptors. Depending on their cellular localisation, activation of these receptors by anandamide may cause various effects, in most cases leading to reduction of the consequences of inflammation. Therefore, blockers of anandamide inactivation (for example, inhibitors of anandamide cellular reuptake or intracellular hydrolysis by fatty acid amide hydrolase (FAAH)), by elevating anandamide levels further, and thus ''indirectly'' activating anandamide targets only ''where and when'' there is enhanced anandamide turnover, might also produce therapeutic effects in intestinal inflammatory conditions, possibly more efficaciously and safely than ''direct'' cannabinoid receptor agonists. Anandamide may also activate vanilloid TRPV1 receptors (mostly located on primary afferent neurones), resulting in pro- (following activation) or anti- (after desensitisation) inflammatory effects. Continuous arrows denote stimulation, induction, or processing; broken arrows denote inhibition. TRPV1, transient receptor potential vanilloid type 1 receptor

> ''The inhibitory effects of cannabinoids on intestinal inflammation, as well as on intestinal motility and secretory diarrhoea, observed in preclinical studies, increase the potential for their use in the treatment of IBD''

This second approach would be preferable because it may lead to activation of all of the endocannabinoid targets involved in protection from inflammation, and is also expected to minimise some peripheral side effects (for example, tachycardia, hypotension) potentially associated with activation of cardiovascular CB_1 receptors. Indeed, inhibitors of anandamide reuptake entirely abolish DNBS induced colon inflammation in mice without causing the undesirable behavioural side effects of psychoactive cannabinoids.[30] As genetic inactivation of FAAH also affords protection in the same animal model of IBD,[29] pharmacological targeting of this enzyme should also represent a therapeutic strategy. However, a FAAH inhibitor was found to be less efficacious than a reuptake inhibitor in this context, and to be ineffective at elevating anandamide levels.[30] In fact, FAAH also catalyses the metabolism of other bioactive amides, including the anti-inflammatory compound palmitoylethanolamide,[44] which exerts inhibitory effects on intestinal motility[45] and is elevated in patients with ulcerative colitis.[44] It is thus possible that FAAH inhibition causes anti-inflammatory actions by elevating levels of this and/or other bioactive FAAH substrates.

> ''There is great potential for the development of new therapeutic agents against intestinal inflammation from the endocannabinoid system''

In conclusion, there is great potential for the development of new therapeutic agents against intestinal inflammation from the endocannabinoid system. While full understanding of the mechanisms of the anti-inflammatory actions of cannabinoid receptor activation is still to be pursued, ad hoc clinical studies will ascertain whether the promising results obtained in animals can be extrapolated to the clinic.

ACKNOWLEDGEMENTS

VDM would like to thank Epitech Srl. for partly supporting some of the studies reviewed in this article.

Authors' affiliations

V Di Marzo, Endocannabinoid Research Group, Institute of Biomolecular Chemistry, Consiglio Nazionale delle Ricerche, Napoli, Italy
A A Izzo, Department of Experimental Pharmacology, University of Naples ''Federico II'', Naples, Italy

Conflict of interest: None declared.

REFERENCES

1 **Di Marzo V**, Bifulco M, De Petrocellis L. The endocannabinoid system and its therapeutic exploitation. *Nat Rev Drug Disc* 2004;**3**:771–84.
2 **Duncan M**, Davison JS, Sharkey Ka. Endocannabinoids and their receptors in the enteric nervous system. *Aliment Pharmacol Ther* 2005;**22**:667–83.
3 **Coutts AA**, Izzo AA. The gastrointestinal pharmacology of cannabinoids. An update. *Curr Opin Pharmacol* 2004;**4**:572–9.
4 **Hornby PJ**, Prouty SM. Involvement of cannabinoid receptors in gut motility and visceral perception. *Br J Pharmacol* 2004;**141**:1335–45.
5 **Massa F**, Storr M, Lutz B. The endocannabinoid system in the physiology and pathophysiology of the gastrointestinal tract. *J Mol Med* 2005;**83**:944–54.
6 **Darmani NA**. Delta(9)-tetrahydrocannabinol and synthetic cannabinoids prevent emesis produced by the cannabinoid CB(1) receptor antagonist/inverse agonist SR 141716A. *Neuropsychopharmacology* 2001;**24**:198–203.
7 **Van Sickle MD**, Oland LD, Ho W, *et al*. Cannabinoids inhibit emesis through CB1 receptors in the brainstem of the ferret. *Gastroenterology* 2001;**121**:767–74.
8 **Adami M**, Frati P, Bestini S, *et al*. Gastric antisecretory role and immunohistochemical localization of cannabinoid receptors in the rat stomach. *Br J Pharmacol* 2002;**135**:1598–606.
9 **Lehmann A**, Blackshaw LA, Branden L, *et al*. Cannabinoid receptor agonism inhibits transient lower esophageal sphincter relaxations and reflux in dogs. *Gastroenterology* 2002;**123**:1129–34.
10 **Pinto L**, Izzo AA, Cascio MG, *et al*. Endocannabinoids as physiological regulators of colonic propulsion in mice. *Gastroenterology* 2002;**123**:227–34.
11 **Izzo AA**, Capasso F, Costagliola A, *et al*. An endogenous cannabinoid tone attenuates cholera toxin-induced fluid accumulation in mice. *Gastroenterology* 2003;**125**:765–74.
12 **Wright K**, Rooney N, Feeney M, *et al*. Differential expression of cannabinoid receptors in the human colon: cannabinoids promote epithelial wound healing. *Gastroenterology* 2005;**129**:437–53.
13 **Mathison R**, Ho W, Pittman QJ, *et al*. Effects of cannabinoid receptor-2 activation on accelerated gastrointestinal transit in lipopolysaccharide-treated rats. *Br J Pharmacol* 2004;**142**:1247–54.
14 **Van Gaal LF**, Rissanen AM, Scheen AJ, *et al*. Effects of the cannabinoid-1 receptor blocker rimonabant on weight reduction and cardiovascular risk factors in overweight patients: 1-year experience from the RIO-Europe study. *Lancet* 2005;**365**:1389–97.
15 **Gomez R**, Navarro M, Ferrer B, *et al*. A peripheral mechanism for CB1 cannabinoid receptor-dependent modulation of feeding. *J Neurosci* 2002;**22**:9612–17.
16 **Mascolo N**, Izzo AA, Ligresti A, *et al*. The endocannabinoid system and the molecular basis of paralytic ileus in mice. *FASEB J* 2002;**16**:1973–5.
17 **Ligresti A**, Bisogno T, Matias I, *et al*. Possible endocannabinoid control of colorectal cancer growth. *Gastroenterology* 2003;**125**:677–87.
18 **Di Marzo V**, De Petrocellis L. Non-CB_1, non-CB_2 receptors for endocannabinoids. In: Onaivi ES, Sugiura T, Di Marzo V, eds. *Endocannabinoids. The brains's marijuana and beyond*. Boca Raton: Taylor & Francis-CRC Press, 2006:151–74.
19 **Ward SM**, Bayguinov J, Won KJ, *et al*. Distribution of the vanilloid receptor (VR1) in the gastrointestinal tract. *J Comp Neurol* 2003;**465**:121–35.
20 **McVey DC**, Schmid PC, Schmid HH, *et al*. Endocannabinoids induce ileitis in rats via the capsaicin receptor (VR1). *J Pharmacol Exp Ther* 2003;**304**:713–22.
21 **Mang CF**, Erbelding D, Kilbinger H. Differential effects of anandamide on acetylcholine release in the guinea-pig ileum mediated via vanilloid and non-CB_1 cannabinoid receptor. *Br J Pharmacol*, 2001;**134**:161–7.
22 **Izzo AA**, Capasso R, Pinto L, *et al*. Effect of vanilloid drugs on gastrointestinal transit in mice. *Br J Pharmacol* 2001;**132**:1411–16.
23 **Moore SA**, Nomikos GG, Dickason-Chesterfield AK, *et al*. Identification of a high-affinity binding site involved in the transport of endocannabinoids. *Proc Natl Acad Sci U S A* 2005;**102**:17852–7.
24 **Katayama K**, Ueda N, Kurahashi Y, *et al*. Distribution of anandamide amidohydrolase in rat tissues with special reference to small intestine. *Biochim Biophys Acta* 1997;**1347**:212–18.
25 **Capasso R**, Matias I, Lutz B, *et al*. Fatty acid amide hydrolase controls mouse intestinal motility in vivo. *Gastroenterology* 2005;**129**:941–51.
26 **Di Marzo V**, De Petrocellis L. Plant, synthetic, and endogenous cannabinoids in medicine. *Annu Rev Med* 2006;**57**:553–74.
27 **Izzo AA**, Fezza F, Capasso R, *et al*. Cannabinoid CB1-receptor mediated regulation of gastrointestinal motility in mice in a model of intestinal inflammation. *Br J Pharmacol* 2001;**134**:563–70.
28 **Cravatt BF**, Giang DK, Mayfield SP, *et al*. Molecular characterization of an enzyme that degrades neuromodulatory fatty-acid amides. *Nature* 1996;**384**:83–7.
29 **Massa F**, Marsicano G, Hermann H, *et al*. The endogenous cannabinoid system protects against colonic inflammation. *J Clin Invest* 2004;**113**:1202–9.
30 **D'Argenio G**, Valenti M, Scaglione G, *et al*. Up-regulation of anandamide levels as an endogenous mechanism and a pharmacological strategy to limit colon inflammation. *FASEB J* 2006;**20**:568–70.
31 **Klein TW**, Lane B, Newton CA, *et al*. The cannabinoid system and cytokine network. *Proc Soc Exp Biol Med* 2000;**225**:1–8.
32 **Ihenetu K**, Molleman A, Parsons ME, *et al*. Inhibition of interleukin-8 release in the human colonic epithelial cell line HT-29 by cannabinoids. *Eur J Pharmacol* 2003;**458**:207–15.
33 **Croci T**, Landi M, Galzin AM, *et al*. Role of cannabinoid CB1 receptors and tumor necrosis factor-alpha in the gut and systemic anti-inflammatory activity of SR 141716 (rimonabant) in rodents. *Br J Pharmacol* 2003;**140**:115–22.
34 **Massa F**, Sibaev A, Marsicano G, *et al*. Vanilloid receptor (TRPV1)-deficient mice show increased susceptibility to dinitrobenzene sulfonic acid induced colitis. *J Mol Med* 2006;**84**:142–6.
35 **Kihara N**, de la Fuente SG, Fujino K, *et al*. Vanilloid receptor-1 containing primary sensory neurones mediate dextran sulphate sodium induced colitis in rats. *Gut* 2003;**52**:713–19.
36 **Geppetti P**, Trevisani M. Activation and sensitisation of the vanilloid receptor: role in gastrointestinal inflammation and function. *Br J Pharmacol* 2004;**141**:1313–20.
37 **Fujino K**, Takami Y, de la Fuente SG, *et al*. Inhibition of the vanilloid receptor subtype-1 attenuates TNBS-colitis. *J Gastrointest Surg* 2004;**8**:842–7.
38 **Goso C**, Evangelista S, Tramontana M, *et al*. Topical capsaicin administration protects against trinitrobenzene sulfonic acid-induced colitis in the rat. *Eur J Pharmacol* 1993;**249**:185–90.
39 **Guagnini F**, Valenti M, Mukenge S, *et al*. Neural contractions in colonic strips from patients with diverticulosis: role of endocannabinoids and substance P. *Gut* 2006;**55**:946–53.
40 **Facer P**, Knowles CH, Tam PK, *et al*. Novel capsaicin (VR1) and purinergic (P2X3) receptors in Hirschsprung's intestine. *J Pediatr Surg* 2001;**36**:1679–84.
41 Press release of the University Hospital of Munich, 3 February 2005 http://idw-online.de/pages/de/news99374 (last accessed 14 July 2006).
42 **Smith PF**. Cannabinoids as potential anti-epileptic drugs. *Curr Opin Investig Drugs* 2005;**6**:680–5.
43 **Kimball ES**, Schneider CR, Wallace NH, *et al*. Agonists of cannabinoid receptor 1 and 2 inhibit experimental colitis induced by oil of mustard and by dextran sulfate sodium. *Am J Physiol Gastrointest Liver Physiol* 2006;**291**:G364–71.
44 **Darmani NA**, Izzo AA, Degenhardt B, *et al*. Involvement of the cannabimimetic compound, N-palmitoyl-ethanolamine, in inflammatory and neuropathic conditions: Review of the available pre-clinical data, and first human studies. *Neuropharmacology* 2005;**48**:1154–63.
45 **Capasso R**, Izzo AA, Fezza F, *et al*. Inhibitory effect of palmitoylethanolamide on gastrointestinal motility in mice. *Br J Pharmacol* 2001;**134**:945–50.