

# Commentary

## How Stress Induces Intestinal Hypersensitivity

Andre G. Buret

*From the Department of Biological Sciences, Mucosal Inflammation Research Group, University of Calgary, Calgary, Alberta, Canada*

Chronic disorders of the gastrointestinal tract such as food allergies, inflammatory bowel disease (IBD), and irritable bowel syndrome (IBS) continue to impose considerable personal and social burdens worldwide. However, a detailed understanding of what causes the symptoms associated with these disorders is lacking. These diseases appear to share an exaggerated inflammatory response to an otherwise benign external stimulus, or one against which tolerance should have been developed.<sup>1</sup> The mechanisms through which "initial sensitization" occurs may shed new light on the pathophysiology of a variety of chronic gastrointestinal disorders and pave the way toward therapeutic strategies for patients with allergic disorders.

The role of external factors, such as psychological stress, in triggering inflammatory reactions has become a topic of intense research activity. Stress may trigger allergic reactions in the gut and other organs, and depression or anxiety may worsen symptoms in inflammatory disorders of the intestine.<sup>2-4</sup> The cascade of biological events leading to stress-induced enteropathy remains poorly understood. One of the many challenging tasks of the gastrointestinal tract is to mount an aggressive response against enteric microbes while maintaining tolerance to food antigens and commensal bacteria. Homeostasis of intestinal epithelial barrier function represents a critical determinant of this ability.<sup>1</sup> In an elegant series of experiments presented in this issue of *The American Journal of Pathology*, Yang and colleagues demonstrate that, in a genetically susceptible host, stress contributes to the development of food allergies by increasing transepithelial permeability in a corticotropin-releasing hormone (CRH)-dependent fashion.<sup>5</sup> Their report offers convincing data to support their earlier groundbreaking observations that stress may alter epithelial function. Using a model of water-avoidance-stress in rats, the authors observed that during chronic stress intestinal antigen uptake is increased, which in turn sensitizes the host to mast cell-induced enteropathy on secondary exposure to the same antigen. Treatment with a

CRH antagonist prevented the increased antigen uptake and the subsequent anaphylactic response of the intestine.

The mechanisms responsible for the CRH-induced augmentation of epithelial permeability warrant further investigation. It has been known for some time that the paracellular permeability offered by tight junctions can be altered in response to physiological and pathological stimuli.<sup>6-8</sup> In recent years, it has been established that paracellular permeability of the gastrointestinal epithelium may be increased by dephosphorylation or degradation of transmembrane tight junctional proteins.<sup>9</sup> In addition, phosphorylation of myosin light chain by myosin light chain kinase (MLCK) or by Rho kinases may also physiologically regulate paracellular permeability by placing tension on the tight junctional complexes.<sup>10</sup> An ever-increasing number of reports suggest that gastrointestinal pathogens may cause disease, at least in part, by increasing transepithelial permeability. For example, *Helicobacter pylori* has the ability to increase the passage of food antigens across the gastric epithelium,<sup>11</sup> and infection with this gastric pathogen may be associated with the development of food allergies.<sup>12</sup> In view of the findings by Yang and colleagues,<sup>5</sup> investigations now must address how stress, and CRH in particular, affects tight junctional protein structure and function. As inhibition of MLCK has been found to inhibit the tight junctional disruptions induced by microbes or pro-inflammatory mediators,<sup>13</sup> such studies may provide fertile ground from which to develop a new class of therapeutic agents targeting the loss of gastrointestinal barrier function.

The findings of Yang and colleagues that stress may contribute to food allergies by increasing transepithelial permeability bears particular interest in view of the protective role played by endogenous bacteria in modulating the development of allergies.<sup>5</sup> Several studies have established an association between normal gut flora and the development of allergies. Children with allergies are more likely to harbor aerobic bacteria and less likely to be colonized with lactobacilli, when compared to nonallergic

---

Accepted for publication September 27, 2005.

This commentary relates to Yang et al, *Am J Pathol* 2006, 168:104-114, published in this issue.

Address reprint requests to Dr. André Buret, Dept. of Biological Sciences, University of Calgary, 2500 University Dr., NW, Calgary, AB, T2N 1N4, Canada. E-mail: aburet@ucalgary.ca.

children.<sup>14</sup> A recent study found that children delivered by cesarean section were at particularly high risk of developing food allergies.<sup>15</sup> Together with other observations, this supports a role for vaginal birth in helping to establish normal gut flora. Finally, in keeping with the "hygiene hypothesis," early colonization with bifidobacteria seems to protect against allergies,<sup>16</sup> and administration of probiotic bacteria after birth significantly reduces the development of allergies in the lungs as well as in the gut.<sup>17,18</sup> Previous observations by the same group of investigators found that chronic psychological stress induced mast cell-dependent bacterial attachment to the epithelium and increased bacterial translocation.<sup>19</sup> The present findings by Yang and colleagues<sup>5</sup> underscore the need for more research into how inadequate responses to bacteria that favor the development of allergy may be modulated by stress.

Not surprisingly, Yang and colleagues found that stress-induced intestinal-antigen uptake implicated mast cells in the intestinal pathology subsequent to secondary challenge. In their earlier studies, the authors had already established that peripheral administration of CRH alone could mimic the effects of chronic stress on transepithelial macromolecular transport and that a specific CRH antagonist may abolish these effects.<sup>20</sup> A recent report has now provided the first evidence that mast cells express CRH receptors.<sup>21</sup> On binding to CRH, mast cells secrete vascular endothelial growth factor (VEGF), but not tryptase, histamine or any of its typical pro-inflammatory cytokines. This response appears to be mediated, at least in part, by adenylate cyclase and cAMP.<sup>21</sup> The authors postulate that this mechanism may be implicated in the stress-induced worsening of arthritis or psoriasis. Together with these observations, the findings of Yang and colleagues<sup>5</sup> now question whether this pathway may also be involved in stress-induced worsening of chronic inflammatory disorders of the gut, including food allergy.

Activation of epithelial apoptosis by pro-inflammatory TNF $\alpha$  or Fas ligand, by pathogenic microbes, or by proteinase-activated receptor pathways may be responsible for loss of epithelial barrier function.<sup>22–25</sup> Other findings indicate that lactobacilli may maintain the survival of epithelial cells by activating the anti-apoptotic Akt-protein kinase pathway and by blocking pro-apoptotic p38 MAP kinase signals.<sup>26</sup> The enteric flora also activates the cytoprotective proteins hsp25 and hsp72.<sup>27</sup> Research must examine how stress and its mediators may regulate epithelial programmed cell death. Yang and colleagues have paved the way for novel studies into the signal transduction pathways responsible for stress-induced loss of intestinal barrier function and the subsequent development of food allergies.<sup>5</sup>

It remains to be determined if psychological factors are primary causes of food allergies, amplifiers of clinical symptoms, or both. The findings by Yang and colleagues<sup>5</sup> illustrate a new CRH-dependent mechanism through which stress may contribute to either one of these pathways. Additional basic and clinical research is warranted to identify the cellular and molecular cascades leading to such responses and how these mechanisms may apply to humans. The current study represents a

significant step toward these goals. In recent years, a number of studies have demonstrated the important role played by T regulatory cells in generating oral tolerance to food antigens.<sup>28</sup> Whether or not stress affects the adequate development of this process, or that of antigen presentation by dendritic cells, represent other important topics for future investigation. The results in this issue have offered gastroenterologists yet another important conceptual contribution to encourage reevaluation of our current treatment strategies for patients at risk of developing food allergies.

## References

1. MacDonald TT, Monteleone G: Immunity, inflammation, and allergy in the gut. *Science* 2005, 307:1920–1925
2. Marshall GD, Agarwal SK: Stress, immune regulation, and immunity: applications for asthma. *Allergy Asthma Proc* 2000, 21:241–246
3. Collins SM: Stress and the gastrointestinal tract IV. Modulation of intestinal inflammation by stress: basic mechanisms and clinical relevance. *Am J Physiol* 2001, 280:G315–G318
4. Dunlop SP, Jenkins D, Neal KR, Spiller RC: Relative importance of enterochromaffin cell hyperplasia, anxiety, and depression in postinfectious IBS. *Gastroenterology* 2003, 125:1651–1659
5. Yang PC, Jury J, Soderholm JD, Sherman PM, McKay DM, Perdue MH: Chronic psychological stress in rats induces intestinal sensitization to luminal antigens. *Am J Pathol* 2005, 168:104–114
6. Madara JL: Tight junction dynamics: is paracellular transport regulated? *Cell* 1988, 53:497–498
7. Nusrat A, Parkos CA, Verkade P, Foley CS, Liang TW, Innis-Whithouse W, Eastburn KK, Madara JL: Tight junctions are membrane microdomains. *J Cell Sci* 2000, 113(Pt. 10):1771–1781
8. Matter K, Balda MS: Signaling to and from tight junctions. *Nature Rev* 2003, 4:225–236
9. Berkes J, Visvanathan VK, Savkovic SD, Hecht G: Intestinal epithelial responses to enteric pathogens: effects on the tight junction barrier, ion transport, and inflammation. *Gut* 2003, 52:439–451
10. Turner JR, Rill BK, Carlson SL, Carnes D, Kerner R, Mrsny RJ, Madara JL: Physiological regulation of epithelial tight junctions is associated with myosin light-chain phosphorylation. *Am J Physiol* 1997, 273:C1378–C1385
11. Matysiak-Budnik T, van Niel G, Megraud F, Mayo K, Bevilacqua C, Gaboriau-Routhiau V, Moreau MC, Heyman M: Gastric *Helicobacter* infection inhibits development of oral tolerance to food antigens in mice. *Infect Immun* 2003, 71:5219–5224
12. Corrado G, Luzzi I, Lucarelli S, Frediani T, Pacchiarotti C, Cavaliere M, Rea P, Cardì E: Positive association between *Helicobacter pylori* infection and food allergy in children. *Scand J Gastroenterol* 1998, 33:1135–1139
13. Wang F, Graham WV, Wang Y, Witkowsky ED, Schwarz BT, Turner JR: Interferon-gamma and tumor necrosis-alpha synergize to induce intestinal epithelial barrier dysfunction by up-regulating myosin light chain kinase expression. *Am J Pathol* 2005, 166:409–419
14. Benn CS, Thorsen P, Jensen JS: Maternal vaginal microflora during pregnancy and the risk of asthma hospitalization and use of anti-asthma medication in early childhood. *J Allergy Clin Immunol* 2002, 110:72–77
15. Eggesbo M, Botten G, Stigum H, Nafstad P, Magnus P: Is delivery by cesarean section a risk factor for food allergy? *J Allergy Clin Immunol* 2003, 112:420–426
16. Hooper LV, Wong MH, Thelin A, Hansson L, Falk PG, Gordon JI: Molecular analysis of commensal host-microbial relationships in the intestine. *Science* 2001, 291:881–884
17. Kalliomaki M, Salminen S, Poussa T, Arvilommi H, Isolauri E: Probiotics and prevention of atopic disease: 4 year follow-up of a randomized placebo-controlled trial. *Lancet* 2003, 361:1869–1871
18. Vanderhoof JA, Young RJ: Role of probiotics in the management of patients with food allergy. *Ann Allergy Asthm Immunol* 2003, 90:99–103
19. Soderholm JD, Yang PC, Ceponis P, Vohra A, Riddell R, Sherman PM,

- Perdue MH: Chronic psychological stress induces mast-cell dependent bacterial adherence to the epithelium and initiates mucosal inflammation in the intestine. *Gastroenterology* 2002, 123:1099–1108
20. Santos J, Saunders PR, Hanssen NP, Yang PC, Yates D, Groot JA, Perdue MH: Corticotropin-releasing hormone mimics stress-induced colonic epithelial pathophysiology in the rat. *Am J Physiol* 1999, 277:G391–G399
  21. Cao J, Papadopoulou N, Kempuraj D, Boucher WS, Sugimoto K, Cetrulo CL, Theoharides TC: Human mast cells express corticotropin-releasing hormone (CRH) receptors and CRH leads to selective secretion of vascular endothelial growth factor. *J Immunol* 2005, 174:7665–7675
  22. Gitter AH, Bendfeldt, Schultzke JD, Fromm M: Leaks in the epithelial barrier caused by spontaneous and TNF-alpha-induced single-cell apoptosis. *FASEB J* 2000, 14:1749–1753
  23. Abreu MT, Palladino AA, Arnold ET, Kwon RS, McRoberts J: Modulation of barrier function during Fas-mediated apoptosis in human intestinal epithelial cells. *Gastroenterology* 2000, 119:1524–1536
  24. Chin AC, Teoh DA, Scott KGE, Meddings JB, Macnaughton WK, Buret AG: Strain-dependent induction of enterocyte apoptosis by *Giardia lamblia* disrupts epithelial barrier function in a caspase-3-dependent manner. *Infect Immun* 2002, 70:3673–3680
  25. Chin AC, Vergnolle N, MacNaughton WK, Wallace JL, Hollenberg MD, Buret AG: Proteinase-activated receptor 1 activation induces epithelial apoptosis and increases intestinal permeability. *Proc Natl Acad Sci USA* 2003, 100:11104–11109
  26. Yan F, Polk DB: Probiotic bacterium prevents cytokine-induced apoptosis in intestinal epithelial cells. *J Biol Chem* 2002, 277:50959–50965
  27. Kojima K, Mush MW, Ren H, Boone DL, Hendrickson BA, Ma A, Chang EB: Enteric flora and lymphocyte-derived cytokines determine expression of heat shock proteins in mouse colonic epithelial cells. *Gastroenterology* 2003, 124:1395–1407
  28. Mowat AM: Anatomical basis of tolerance and immunity to intestinal antigens. *Nat Rev Immunol* 2003, 3:331–341