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## The Microbiome, Viscerosensory Signaling and Autism



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Throughout this series we have pointed to evidence of an increasingly complex understanding of the relationship between the gut, its commensal bacterial composition, and its link to various pathological states within different organ systems. Paramount in our articles has been a theme of holistic interconnectedness. The relationships between the gut microbiome and the disease states already discussed are not isolated but are inextricably linked. Here we will briefly discuss the emerging research that demonstrates the connection between gut microbes and the nervous and immune systems as well as how disproportional bacterial concentrations may be implicated in Autism and other neuropsychiatric ailments.

Contemporary research has shown that gastrointestinal (GI) infections lead to behavioral changes in mice and that the immune system affects mood and learning.<sup>2,5,12</sup> In turn, depression and stress can cause changes in immune functioning.<sup>5</sup> The former relationship, that of infection influencing

behavior, was observed in a study by Lyte et al. in 1998. Researches noted increased anxiety in mice after the per-oral administration of *Campylobacter jejuni*, as compared to saline treated mice.<sup>11</sup> Also of note was the lack of bacteria or pro-inflammatory cytokines found in the systemic circulation of the *C. jejuni*-treated mice. This finding was a springboard for the investigation of intestinal viscerosensory nerves as a possible conduit between the gut and the brain and behavioral responses. Within the gut, there are two types of viscerosensory nerves, intrinsic and extrinsic. Intrinsic nerves control motility and secretion, but do not directly convey signals to the central nervous system. Extrinsic nerves, including the vagus nerve and the spinal visceral nerves communicate with the CNS, innervate the intrinsic nerves, and contact lymphoid tissue in the subepithelium.<sup>5</sup> To firmly establish the connection between the immune system, infection, and changes in behavior, Goehler et al. and subsequently Lyte et al. demonstrated evidence of c-Fos, an early gene product and marker of cell “activation”, in the vagal neurons of mice after inoculation with *Campylobacter jejuni* or *Citrobacter rodentium*, respectively.<sup>5,6,10</sup> Another intestinal-CNS link was elucidated by Castex et al. in 2005. In this study, researchers noted c-Fos expression in specific brain regions in

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rats in response to intestinal ischemia. Furthermore, they found that intraperitoneal administration of ondansetron or perivagal capsaicin attenuated c-Fos expression, implicating 5-HT<sub>3</sub> in the immune “activation” of the viscerosensory nerves.<sup>1</sup> A number of other proinflammatory mediators/receptors have also been identified as possible players in the intestinal activation of vagal/spinal nerves. These include: Bradykinin, prostaglandins and leukotrienes, ATP and adenosine, vanilloid receptors, proteinase-activated receptors, and nerve growth factor (NGF).<sup>9</sup> Vagal fibers also express toll-like receptors (TLRs), specifically TLR-4 on their surface, which is known to respond to bacterial lipopolysaccharide.<sup>7</sup>

The established paradigm demonstrating a behavioral response to intestinal bacteria, bacterial products, and mechanical stimuli logically suggests that some neuropsychiatric disorders may be, at least in part, due to derangements in the human intestinal microbiome. The relationship between irritable bowel syndrome (IBS) and depression and anxiety are widely known, and one can imagine that other intestinal insults may be responsible for various neurologic and/or psychiatric conditions and vice versa. In fact, there is evidence that predictable gut microbiome derangements may contribute to Autism Spectrum Disorder (ASD), as GI symptoms are increasingly associated with autism.<sup>3,4</sup> Per the most recent CDC statistics, roughly 1 in 59 children are affected by ASD and the worldwide prevalence is somewhere between 1-2%; ASD is about 4 times more likely to be identified in boys as compared to girls.<sup>13</sup> In a very recent study by Finegold et al. published in *Anaerobe*, researchers studied stool specimens from 33 autistic children ages 2-9 with intestinal symptoms and 13 control children without autism or GI upset. Results showed that the intestinal microbiome of autistic children with GI disease was colonized by proportionally higher counts of *Clostridium perfringens*. In addition to the statistically significant higher raw number of *C. perfringens* colony forming units, it was also noted that children with ASD harbor significantly more of the *C. perfringens* responsible for beta2-toxin production, as opposed to colonies producing other *C. perfringens* toxin genes.

Diets including specific probiotics have been shown to inhibit visceral pain caused by colonic

distension.<sup>8</sup> We have discussed the commonality in viscerosensory afferents. If alterations in diet can mediate visceral pain, it is likely that further research will lead to dietary and pharmacologic interventions aimed at the microbial disproportions associated with ASD and other neuropsychiatric illnesses. ■

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