

# Chronic fatigue syndrome: lactic acid bacteria may be of therapeutic value

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**Summary** Chronic fatigue syndrome (CFS) is complex illness with unknown aetiology. Recent research shows that patients with CFS have marked alterations in microbial flora, including lowered levels of bifidobacteria and small intestinal bacterial overgrowth (SIBO). Research also indicates that CFS patients are under increased oxidative stress, have a type 2 helper cell dominate cytokine profile, frequently report allergies, have altered essential fatty acid (EFA) status and may have malabsorption of certain micronutrients. Lactic acid bacteria (LAB) have the potential to influence the immune system in CFS patients by supporting T helper cell 1 driven cellular immunity and may decrease allergies. In addition LAB are strong antioxidants, may improve EFA status, can enhance absorption of micronutrients by protecting the intestinal epithelial barrier, and have been used to treat SIBO. It is our contention that LAB may have a therapeutic role in the treatment of CFS.

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## INTRODUCTION

Chronic fatigue syndrome (CFS) is a medically unexplained illness, characterized by persistent and relapsing fatigue. In addition to cognitive dysfunction, headaches, joint pains, central nervous system disturbances, and a variety of other symptoms, many CFS patients complain of gastrointestinal disturbances (1,2). Patients with CFS are more likely to report a previous diagnosis of Irritable bowel syndrome (IBS), meet diagnostic criteria for IBS and experience IBS-related symptoms (3). In a one-year retrospective evaluation, over 70% of patients with chronic fatigue met the Manning criteria for IBS (4).

While the etiology of CFS remains unknown, several causative factors have been suggested, including viral infection, altered immune function and pre-illness stressors (5). Recent research indicates that there are

marked alterations in the intestinal microflora of CFS patients (6,7). Although the gastrointestinal (GI) microflora is itself far from being completely understood, it is clear that it plays an important role in the health of the human body. The goal of this report is to integrate various branches of research in order to support our hypothesis, that lactic acid bacteria may be of therapeutic value in the treatment of CFS.

## NORMAL INTESTINAL FLORA

The bacteria that make up the GI microflora are part of a complex and delicately balanced ecosystem. It is estimated that the human intestine contains close to 500 bacterial species, belonging to about 200 genera (8). The variety and location of bacteria in the intestines is dependent on a number of factors, including stomach acid secretion and small bowel motility. The proximal small bowel of healthy individuals normally contains relatively small numbers of microorganisms in the range of  $10^2$ – $10^5$  colony forming units (cfu)/ml of intestinal content (9). Anaerobic bacteria normally outnumber aerobic bacteria in the small intestine by about 100:1 (10).

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In the colon there is a dramatic increase in bacterial counts, rising to  $10^{10}$ – $10^{12}$  cfu/g. The most prevalent microorganisms in the adult colon are obligate anaerobes, outnumbering aerobes by up to 1000:1 (9). The most common anaerobic microorganisms are bacteroides, bifidobacteria, lactobacilli and anaerobic cocci (11). Pathogenic and opportunistic bacteria also make up a small part of the normal GI flora.

### ROLE OF NORMAL MICROFLORA

Normal GI microflora is considered beneficial in the human body for a number of reasons. Bacteria that are normally found in high numbers, such as bifidobacteria and lactobacilli (lactic acid bacteria) are involved in vitamin synthesis, stimulation of the immune response, prevention of pathogenic and opportunistic bacterial colonization, protection of the intestinal barrier defense system, production of short chain fatty acids for enterocyte energy, and metabolism of carcinogenic substances (8,10–12). Lactobacilli and bifidobacteria can lower levels of potentially neurotoxic compounds such as ammonia, amines and indoles (13).

More recent research suggests that species from these genera act as potent antioxidants, both in vitro and in vivo (14–17). Lactic acid bacteria (LAB) may also play a role in the prevention of allergies by balancing normal cytokine profiles that regulate T helper cells. Th1 cells are responsible for directing cell-mediated immunity, while Th2 cells direct humoral immunity (18). A number of recent studies have shown that LAB can have a significant impact on the cytokines that both support Th1 immunity and regulate the over expression of the Th2 immunity common in those with atopy (19,20). Indeed children with allergies are more likely to have greater counts of aerobic bacteria along with lower counts of anaerobes, particularly lactobacilli (21). The administration of certain strains of lactobacilli and bifidobacteria has led to improvements in natural killer cell activity (22–24), Interferon gamma (25,26), Il-2 (27,28) and Il-12 (29–32) release and at the same time, lowering levels of Il-4, Il-5 and Il-10 cytokines that continue the Th2 predominance (27,33,34). It is also interesting to note that in atopic infants, the administration of bifidobacteria can increase omega-3 fatty acid concentrations in serum phospholipids (35). This is a significant finding, given the connections between atopy and low levels of omega-3 fatty acids (36,37).

### FACTORS AFFECTING NORMAL FLORA

Although the intestinal microflora is quite stable over time, a number of factors can disturb the normal balance. The GI microflora can be altered by immune

mechanisms of the host, redox state, adrenal function, intestinal pH, peristalsis, diet, aging, drugs, exogenous organisms, climate and emotional stress (10,11). It is clear that many of these factors are capable of causing more than a transient alteration in the flora of patients with CFS.

### INTESTINAL FLORA AND CFS

Henry Butt and colleagues (6) from the Department of Biological and Chemical Sciences, University of Newcastle, Australia have been examining the microbial flora of CFS patients for a number of years. In 1998 they presented the first research showing an altered fecal microbial flora in CFS patients ( $n = 27$ ) vs. controls. The mean distribution of *Escherichia coli* as a percentage of the total aerobic flora of the control subjects was 92.3% compared to 49% in CFS patients. The mean percentage distribution of *Bifidobacterium* spp. was 7.1% in controls and 2% in CFS patients. The incidence of CFS patients with fecal *E. coli* greater than the percentage mean of control subjects was only 7 compared to 21 CFS patients with greater *Bacteroides* spp. ( $p = 0.0001$ ).

Butt and colleagues (7) describe what they call 'Bacterial colonosis' (BC) as a condition involving numerous GI symptoms that is characterized by lack of GI inflammation and a marked alteration of GI microflora. They report BC among patients with persistent fatigue, including CFS, IBS and the related disorder of fibromyalgia (FM). In a very large study ( $n = 1390$ ) involving patients with persistent fatigue, they report patients with BC as having more severe fatigue ( $p < 0.0015$ ) and muscular pain ( $p < 0.01$ ) compared to patients without marked microflora alteration. Once again, in this study they found lower levels of *E. coli* (mean percentage distribution was 36.6% of total aerobic flora vs. 70–95% in healthy subjects  $p < 0.001$ ). While *E. coli* decreased, there was a significant rise in *Enterococcus* spp. ( $p < 0.001$ , 28.7% of total aerobic flora) among patients with persistent fatigue compared to healthy subjects (3–5%). Once again, patients with chronic fatigue had marked reductions in bifidobacteria, making up less than 0.35% of the total anaerobic flora. Finally, in addition to the relationship between BC, pain and fatigue, the researchers found some interesting correlates. The higher the aerobic enterococcal count, the more severe the neurological and cognitive deficits; nervousness ( $p < 0.05$ ), memory loss ( $p < 0.01$ ), forgetfulness ( $p < 0.01$ ) and confusion ( $p < 0.05$ ). They also found that the higher the aerobe to anaerobe ratio, the more severe the deficits in GI functional status.

Research indicates that patients with chronic enteritis also have a decrease in the content of both *E. coli* and bifidobacteria with a concomitant rise in enterococci

(38). Infants with allergies are more likely to have high levels of enterococci, leading researchers to suggest that because some species of enterococci have virulence factors that can compromise the gut barrier, they may be directly involved in atopic sensitization (39). According to animal research, *Enterococcus faecalis* can cause inflammation of the gastrointestinal mucosa (40). Previous research has indicated that IBS patients have alterations of microbial flora, specifically decreased coliforms, lactobacilli and bifidobacteria (41), as well as increased aerobic flora and a reduction in the normal anaerobe to aerobic ratio (42–44). Alterations in gut flora, specifically increased numbers of aerobes, are thought to be involved in the food sensitivities often reported by IBS patients. It is postulated that pharmacologically active metabolites derived from food residues by an altered flora may be involved (44). It is interesting to note that CFS and FM patients also report worsening of symptoms provoked by foods (45,46).

Antibiotics, invaluable in the treatment of infections, have been shown to cause marked alterations in gut flora (47) and can have long term effects on intestinal bifidobacteria (48). In the case of IBS, prior antibiotic use appears to be a risk factor in the development of the illness (49,50). Interestingly, the administration of certain antimicrobials, particularly cephalosporins can decrease *E. coli*, bifidobacteria and anaerobes, resulting in an overgrowth of enterococci (51,52). It is unknown if pre-illness antibiotic use is high among CFS patients.

Further evidence of an altered GI microflora comes from research by Mark Pimentel and colleagues (53) from the Cedars-Sinai Medical Center in Los Angeles. They found that 77% of CFS patients ( $n = 31$ ) had small intestinal bacterial overgrowth (SIBO) based on objective measurements via the lactulose hydrogen breath test. SIBO has also been found in 78% of FM patients ( $n = 123$ ) (54) and 78% of IBS patients ( $n = 202$ ) (55). In addition, a disorder with significant co-morbidity with both CFS and FM, endometriosis (EM) (56), a condition where levels of lactobacilli may be reduced (57), also has an 80% rate of SIBO (58). Patients within all four of these groups, CFS, FM, IBS and EM, report high levels of allergies and various sensitivities (56,59–61), this connection with SIBO needs further research.

SIBO is defined by any condition in which the proximal part of the small intestine contains  $>10^5$  bacteria per ml of intestinal juice (62). While both aerobes and anaerobes are present in SIBO states, it does not reflect normal anaerobic dominance, and aerobes most often predominate (63). SIBO is often a result of intestinal stasis and/or low stomach acid production (64). Symptoms are similar to those reported by CFS/FM/IBS/EM patients, including abdominal pain, bloating, gas and altered bowel habits (54,58). SIBO can cause malab-

sorption of fat, carbohydrate, protein, vitamin B<sub>12</sub> and other micronutrients (65–67). It should be noted that CFS patients, despite a diet that is no different than the North American average (68), have been found to have lower levels of a number of B vitamins (69) and other micronutrients (70).

## STRESS AND INTESTINAL MICROFLORA

There is some research in both humans and animals showing that emotional stressors can negatively affect microflora. States of stress associated with anger and fear have been shown to be related to increases in bacteroides, specifically the thetaiotaomicron and uniformis group (71). Normally comprising 2–4% of the flora, these bacteroides increase to 20–30% of the flora under conditions of anger or fear (72).

Lizko et al. (73,74) examined the effect of nervous-emotional stress on intestinal microflora within a group of astronauts preparing for flight. In the days leading up to the launch, as nervous-emotional stressors were higher, there were marked decreases in bifidobacterium and lactobacilli observed. After flight, the number of lactobacilli remained low, while the enterobacteria and clostridia were substantially increased. Bifidobacteria in particular appear extremely vulnerable to the effects of pre-flight emotional stress (75). Beyond this pre-flight emotional stress, the flight conditions, including restraint stress and physical demands, lead to further decreases in lactobacilli and bifidobacteria (76–78), decreased cellular immunity and increased sensitivity to allergens (79,80).

In research involving endurance athletes, some microflora findings similar to those of CFS patients have been observed. The composition of fecal microflora in endurance athletes ( $n = 44$ ,  $27.5 \pm 6.1$  years,  $60.2 \pm 6.3$  ml/kg/min VO<sub>2</sub> max, training  $13.4 \pm 3.9$  h/week) were analyzed. The percentage distribution of bifidobacteria showed decreased bacteria counts (91% had less than reference values). In contrast, clostridia levels were over the reference values in 36% of the subjects. Interestingly 77% of the endurance athletes had levels of *E. coli* that were lower than reference values. Outside of bifidobacteria, bacteroides, lactobacilli and clostridia, the researchers stated that 100% of the athletes had below the reference range for other additional anaerobes. One significant difference between these healthy athletes and CFS patients is the enterococcal count. The majority of athletes (59%) had less than the reference range of enterococci (81). Perhaps enterococci is a differentiating factor between states of health and disease. Animals fed a semi-synthetic diet have lowered levels of lactobacilli, increased enterococci and are more susceptible to experimental infection (82). It is obvious that

further research is necessary to more closely examine the role of this aerobic bacteria in chronic illness.

Further evidence of the role stress plays in GI microbial alterations has been found in animal studies. Bailey and Coe (83) from the Department of Psychology, University of Wisconsin, examined the role of separation stress in the intestinal microflora of infant rhesus monkeys. In addition to examining the GI flora, these investigators wanted to determine if an alteration could result in an internal environment conducive to pathogen colonization and growth. The results showed that maternal separation caused significant reductions in lactobacilli. Animals with the fewest lactobacilli had higher pathogen titers of *Shigella* spp. and *Campylobacter* spp. indicating a protective effect of lactobacilli. While cortisol increased markedly, it did not predict the lactobacilli decrease, suggesting that more than cortisol may be at work. The authors suggest that stress can alter GI flora by altering acidity of gut secretions, gut motility, and that the direct effect of other neurochemicals such as norepinephrine may be involved.

These results support previous findings in a variety of animal species demonstrating that stressors can alter microflora. Environmental stress such as crowding and heat can increase aerobic bacteria (84), and decrease lactobacilli (85). Food deprivation, restraint conditions and acoustic stress have all been shown to alter microflora, lowering lactobacilli in animals (86–88).

### STRESS, CYTOKINES AND CFS

Research suggests that stress plays a significant role in the onset and exacerbations of CFS symptoms. Both physical and psychological stressors appear to contribute to the illness, leading some investigators to refer to CFS as a disorder of stress (89). The majority of CFS patients believe that stress, or a combination of stress and an infection resulted in the condition (90–93). There is some research to support this notion, as those who reported to a doctor's office with a viral infection were more likely to develop CFS if they were under psychological distress at the time (94). In addition, Theorell and colleagues (95) found an almost twofold increase in the prevalence of both infections and negative life events in the three months preceding the onset of CFS. Various studies have found that up to 95% of CFS patients report increased levels of stress prior to illness onset (96–98).

A number of experimental studies have shown that CFS patients have a decreased tolerance for stress and that stress exacerbates the symptoms (99–101). Perhaps the most significant finding in this area is that which shows a direct correlation between closeness to the high impact areas of Hurricane Andrew in South Florida. Closeness to high impact areas and the related post-

hurricane distress response became a strong predictor of the likelihood and severity of CFS relapse (102). Whether cause, effect or both, that stress is involved in the pathogenesis seems clear. It is obvious that among a patient population that was most often high functioning pre-illness (103), dealing with a chronic, debilitating illness places them in a high stress category. It is not surprising that researchers have shown that plasma levels of adrenaline are substantially higher among CFS patients (104).

Various investigative groups have shown that psychological stress can influence Th1:Th2 balance. Specifically, emotional stressors lead to a bias towards Th2 cytokines, increasing humoral immunity and decreasing Th1 driven cellular immunity (105,106). This increased Th2 dominance during stress may at least partially explain the increased incidence of type 2-mediated conditions such as increased viral infections, latent viral expression, allergies and asthma during periods of high or chronic stress (107).

Research indicates that CFS patients have a chronic lymphocyte overactivation, alterations in plasma levels of proinflammatory cytokines and a decrease in the ratio of Th1:Th2 cytokines (5). These observed immune abnormalities are thought to account for many of the symptoms observed in CFS (108). Low natural killer cell cytotoxicity, increased associated allergic conditions and elevated levels of type 2 cytokines indicate that conditions such as CFS and Gulf War syndrome have a bias toward humoral immunity and diminished cellular immunity (109). The decreased Th1:Th2 ratio may be a result of altered glucocorticoid regulation on IL-10 and IL-12, key chemicals in humoral and cellular immunity (110,111). A bias toward Th2 is reflected in the research which shows that as many as 80% of CFS patients have allergies (59,91).

In addition to the role of psychological stress, there is a growing body of research demonstrating that increased oxidative stress is playing a role in CFS. Although it is uncertain whether oxidative stress in CFS is a cause or effect of the illness, it is becoming clear that CFS patients have increased markers of oxidative stress and an impaired antioxidant capacity (112–115). There is some evidence showing that antioxidant support leads to clinical improvement in CFS and that symptom reductions are correlated with improvements in erythrocyte fragility, a marker of oxidative stress (116).

### CFS AND ESSENTIAL FATTY ACIDS

The suggestion has been made that essential fatty acid (EFA) deficiency, both viral and immune induced and/or through abnormalities of metabolism, is playing a role in the pathogenesis of CFS (117,118). There is research to

support this, as CFS patients have been found to have lower levels of plasma EFA levels. These levels normalized and clinical improvements resulted during a three-month intervention trial with an EFA preparation (119). The administration of an omega-3 fatty EFA preparation has also led to clinical improvements in the related disorder of FM (120).

### A ROLE FOR LACTIC ACID BACTERIA?

Emerging research suggests that the intestinal microflora in CFS is markedly different than that of healthy controls, findings that are not entirely surprising given the research cited on the influence of stressors on microbial flora. We postulate that stress-induced alterations in microbial flora, specifically diminished lactobacilli, bifidobacteria and anaerobes in general, will amplify the domination of Th2 cytokines. Further, it is our contention that the administration of specific strains of lactic acid bacteria can help to regulate the composition of the intestinal flora and may have a significant impact on shifting the cytokine balance back toward Th1 driven cellular immunity. In addition, lactic acid bacteria can protect the intestinal epithelial barrier and enhance the absorption of vitamins and minerals, particularly those that are decreased in CFS patients. Furthermore, lactic acid bacteria have the potential to act as strong antioxidants in a patient population that has been shown to be under increased oxidative stress and one that has a diminished antioxidant capacity. They also have the potential to improve the EFA status in serum phospholipids, and have been used therapeutically in the treatment of SIBO (121–123), a condition common in CFS and the other so-called functional somatic disorders.

Russian research indicates that the administration of bifidobacteria can not only prevent marked alterations in the microflora of those under emotional and physical stress (124), it can also correct an altered microflora and lead to improvements in general health (125). LAB have been used successfully in controlled studies involving IBS patients (126–128) and can prevent rises in enterococci levels among these patients (129). In vitro and animal research indicates that LAB can inhibit the growth of enterococci (130,131).

LAB, particularly bifidobacteria, can decrease amine production (13), an important consideration given the research on the neurotoxic effects of amines such as spermidine (132) and the neurological findings in CFS (133). Recent animal research shows that orally administered *Campylobacter jejuni*, in subclinical doses too low to elicit immune activation, results in anxiety provoking effects. These results suggest that microorganisms, in the absence of an immune response, can directly activate neural pathways (134). This could explain why Butt

and colleagues found relationships between disturbances of the intestinal microbial milieu in CFS patients and cognitive impairment. It may also potentially provide an organic explanation for some of the behavioral disturbances observed in CFS. It is also interesting to note that eradication of *Helicobacter pylori* has led to improvements in intensity, duration and frequency of migraine headaches (135) and that IBS patients report higher levels of pain if they are infected with *H. pylori* (136). LAB have been shown to be inhibitory to *H. pylori* in vitro, in animal and in human studies (137) and while it is unknown if *H. pylori* infection is related to CFS, it is possible that microbial alterations play a role in cytokine-mediated pain mechanisms among these patients.

The findings of low levels of bifidobacteria in CFS are of particular significance, as the available evidence suggests that this specific group of LAB, when reduced or diminished, indicates an unhealthy state (138). The microbes of the gastrointestinal tract appear intricately involved with the systemic immune and nervous systems and perhaps their role in functional conditions such as CFS is currently underestimated. LAB have not been the subject of controlled studies in CFS, however, based on the research reviewed in this paper, it seems plausible that they would be of therapeutic value and certainly warrant further investigation.

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