**Using Probiotics to Lessen the Impact of Fatigue in Teens (UPLIFT) Pilot Study**

**Study Protocol**

**Overall Summary**

Chronic Fatigue Syndrome (CFS) is a debilitating disorder characterised by fatigue that does not improve with rest, physical symptoms of pain, nausea, fatigue, dizziness, headaches and post exertional malaise which is a worsening of CFS symptoms following minimal physical or mental effort. In young people, CFS can disrupt typical adolescent development by impacting a young person’s ability to attend school, socialise with peers, and maintain physical activity. Rates of depression and anxiety are also higher in those with CFS. Currently there is no treatment for CFS and patients are encouraged to manage symptoms. Gut microbiota are the colony of bacteria, fungi and viruses that live in the gastrointestinal tract and play a role in physiological processes in the body. The microbiota-gut-brain-axis refers to multiple bi-directional relationships between the microbes in the gut and the brain. Evidence suggests that disruption to the balance of beneficial versus pathological microbes in the gut (dysbiosis) is present in chronic fatigue, depression and anxiety. Probiotics, defined as live microorganisms that confer a benefit to the host when consumed, are one way in which the healthy balance of microbes can be restored in the gut. Preliminary studies have suggested that supplementation with probiotics can improve psychological well-being in people with CFS. To date there have not been any studies of the effect of probiotics in adolescents with CFS. This study aims to determine whether probiotic supplementation improves the psychological wellbeing of adolescents with CFS using a randomised, double blind placebo controlled intervention that comprises a 10-week supplementation period with a combination of the probiotics *Lactobacillus rhamnosus* HN001 and *Bifidobacterium animalis* HN019.

**Background**

Chronic fatigue syndrome (CFS), sometimes called myalgic encephalomyelitis (ME) or fibromyalgia, is a clinical diagnosis characterised by chronic debilitating fatigue not explained by other medical or psychological diagnoses. Common symptoms in children and adolescents include nausea, dizziness, headaches, pain, sleep problems, difficulties with concentration and post-exertion malaise which is a worsening of CFS symptoms following minimal physical or mental effort. In a study of the epidemiology of CFS in Australian children and adolescents, female gender, history of an infectious illness, Caucasian ethnicity and gradual onset of symptoms were common presenting factors [1]. Both psychological and physical stress have also been reported as triggers for CFS [2]. CFS in adolescence is associated with significant disruptions to adolescent development and functioning including educational disruption, social isolation from peers, reduced physical activity and poor emotional wellbeing. Rates of anxiety and low mood are higher in adolescents with CFS [3, 4]. Currently there is no treatment for chronic fatigue syndrome.

**The microbiota gut brain axis, inflammation and health**

The human gastrointestinal tract contains a colony of microbes that outnumber the number of cells in the human body. These microbes which include bacteria, fungi and viruses have an important role in physiology and beiochemistry in the body and their function has been linked to health and behaviour. Dysbiosis is the term used to broadly refer to negative changes in the microbial balance in the gut. Dysbiosis influences brain chemistry via the microbiota gut brain axis. The gut brain axis refers to multiple bi-directional pathways that link the microbial balance in the gut with brain chemistry These pathways are also involved in depression and anxiety and include the immune system, neuroendocrine system (particularly the hypothalamic pituitary adrenal axis), parasympathetic and sympathetic arms of the autonomic nervous system and the vagus nerve [5, 6]. Evidence has reported dysbiosis in patients with depression and anxiety although it is not yet clear if there is a consistent microbial composition associated with depression [7].

The link between dysbiosis and psychological symptoms is also demonstrated via the link between dysbiosis and inflammation, and in turn between inflammation and depression. Dysbiosis has been linked to immune system functioning, inflammation and depressive symptoms in patient populations including those with irritable bowel conditions, diabetes and obesity [8-10]. Inflammation, particularly systemic inflammation, has been shown to increase permeability of the gut which allows the increased circulation of detrimental chemicals around the body (leaky gut), and is associated with increased circulation of pro-inflammatory cytokines and other biomarkers of inflammation [11, 12]. Chronic low grade inflammation has been associated with symptoms of depression [11-13]. Furthermore, inflammation has been shown to be higher in CFS patients [14]. However, it remains unknown whether addressing inflammation by improving gut microbiota balance will have an impact on psychological wellbeing, inflammation and fatigue in people with CFS [15].

**Probiotics, mood and chronic fatigue syndrome**

Probiotic supplementation is one way in which the composition of the gut microbiota can be altered and the effects of dysbiosis reversed hence it has been suggested as a potential treatment for conditions in which dysbiosis and inflammation have been observed [16]. Probiotics are defined as live micro-organisms that, when consumed in adequate quantities, confer a health benefit to the host. Previous trials have demonstrated improvements in depressive symptoms associated with probiotic supplementation [17-19]. In adults with CFS a pilot randomised, double blind, placebo controlled trial of the *Lactobacillus* *casei* Shirota in 38 participants found that both depression and anxiety symptoms were significantly reduced in those taking the probiotic for the 8 week trial when compared with those in the placebo group [20]. However, there have been very few further studies of the effects of probiotics in people with CFS. An open pilot trial in which 15 adults with CFS were given a multispecies probiotic found that it had no effect on self-reported fatigue or levels of physical activity. Those in the study did report a general improvement in quality of life on a single question. While this study encourages further research, it is significantly limited by the small sample size and lack of randomisation or use of a placebo [21]. In a different trial of the effect of probiotics in three patient groups including CFS and a comparison group of healthy controls, those with CFS had higher inflammatory biomarkers including interleukin-6 (IL6), pro-inflammatory cytokines and C-reactive protein (an indicator of systemic inflammation) at baseline compared to healthy controls. Each of these inflammatory markers were reduced in CFS patients who received the probiotic *Bifidobacterium* *infantis* 35624 for 6-8 weeks [22]. To date there have not been any trials of probiotic supplementation conducted in adolescents with CFS

**AIMS**

The overall aim of this study is to run a randomized, double-blind, placebo-controlled pilot trial of probiotic intervention in young people with chronic fatigue with the following specific aims:

* Test the feasibility of recruitment and data collection procedures
* Test the acceptability of a probiotic intervention trial in young people with CFS
* Establish whether there is preliminary evidence that probiotic supplementation may improve psychological outcomes in young people with CFS
* Inform an accurate power calculation for a future full trial

**METHODS**

**Study Design:** Randomised double-blind, placebo controlled pilot trial

**Sample Size:** The UPLIFT pilot study will recruit 40 participants.

**Recruitment:** Young people aged 13-18 years with a diagnosis of chronic fatigue syndrome made by a doctor will be recruited from three sources:

1. General paediatric outpatient clinics at Starship Children’s Hospital and Waitemata Paediatric Services.
2. Via General Practice, specifically, through Dr Ros Vallings, a General Practitioner with a special interest in CFS
3. Chronic Fatigue Syndrome support networks for young people.

The Principal Investigator Dr Rebecca Slykerman has previously worked with adolescents with CFS and has existing relationships with several of the General Paediatricians from the Auckland and Waitemata District Health Boards who provide care for these young people. Dr Raewyn Gavin is a Starship General Paediatrician with a particular interest in CFS and a collaborator on this project. In addition to the patients that Dr Gavin cares for she is also in contact with both paediatric and general practice colleagues who also care for young people with CFS which will assist in recruitment of participants for this study.

**Procedures:** For Paediatricians and interested General Practitioners both written and electronic copies of the Participant Information Sheet (PIS) will be given to them to provide by email or face to face to young people (and for those aged 13-16, their parents/caregivers) who have a diagnosis of CFS. The PIS will contain information about the study and a link to the website where parents and young people can consent and register to participate. An electronic copy of the PIS will be sent to support network coordinators to circulate to young people who may be interested in participating in the study.

*Exclusion criteria:*

1. Taking probiotics regularly
2. On immunosuppressant therapy
3. Participation in another intervention trial for CFS
4. Children under the age of 13 years

**Consent:** For young people aged 13-16 consent will be sought from both the young person and their parent/caregiver. Participation will be dependent on consent from both the young person and a parent/caregiver.For young people aged over 16 consent will be sought from the young person and assent will be sought (but not required) from parent/caregiver.

**Data collection:** All consent and data collection will be managed via a secure online website/database. The online consent and data collection procedure has been successfully used in three previous trials in nurses and university students in New Zealand. This means that young people can participate in the study from any location in New Zealand and are not required to attend appointments in order to participate. This significantly reduces the burden to participants. Furthermore, the study can be run under any COVID19 alert level.

Once consent has been obtained participants will be directed to complete online questionnaires. On completion of questionnaires participants will be fully registered for the study and randomised to the intervention or placebo group.

**Measures for young people**

*Mood and Feelings Questionnaire – Short Version (SMFQ): This is the primary outcome measure.* The short version of the MFQ is a 13-item questionnaire that asks young people about recent mood. Scores range from 0-26 with higher scores indicating higher levels of emotional difficulties. The SMFQ has been validated in New Zealand adolescents [24].

*State Trait Anxiety Inventory – six item (STAI-6):* The STAI6 is a short six item version of the longer State Trait Anxiety Inventory used to assess symptoms of anxiety.

*Pediatric Quality of Life Multidimensional Fatigue Scale (PedsQL MDF):* This is an 18 item questionnaire that asks young people aged 13-18 years about symptoms of fatigue in three areas: General Fatigue, Sleep/Rest Fatigue and Cognitive Fatigue. Each subscale contains six questions.

*Post-exertion malaise and gastrointestinal symptoms:* Post-exertion malaise is one of the key features of CFS; however, existing questionnaires are not suitable for an adolescent population. Based on the DePaul Questionnaire, Cotter and colleagues devised a brief questionnaire to assess the frequency and severity of post-exertion malaise [25]. Participants rate each of 5 symptoms for frequency and then severity thus answering 10 questions in total.

**Measures for Parents**

Similar to the procedure for young people, consent and data collection from parents will be done through the same secure online database. At the time of registering for the study parents will complete demographic information about the family and their child’s diagnosis. Parents will also complete questionnaires at baseline and post-intervention as follows:

*Perceived Stress Scale:* Parents/caregivers will complete the Perceived Stress Scale to rate their own levels of stress. The PSS is a 10 item questionnaire that asks people to rate how often in the preceding month they have felt stressed.

*Mood and Feelings Questionnaire – Short version Parent Form:* Like the SMFQ self-report for young people, the parent-form asks parents to rate their child’s recent mood. The 13-item scale gives scores ranging from 0-26 with higher scores indicating increasing emotional difficulties.

*Parent rated Pediatric Quality of Life Multidimensional Fatigue Scale- Teen:* Similar to the self-report version, the parent report version of the Pediatric Quality of life Inventory Multidimensional Fatigue Scale is an 18 item questionnaire that asks parents to report about their young person’s level of general fatigue, sleep/rest and cognitive fatigue.

*Parent rated post-exertional malaise:* Similar to the questions completed by young people, parents will be asked to rate the frequency of five post-exertional malaise symptoms in their children.

**Intervention:** Participants will be randomised to either the intervention group or placebo. The intervention group will receive probiotic capsules containing a combination of the probiotic *Lactobacillus* *rhamnosus* HN001 (6X109 cfu) and Bifidobacterium animalis susp. Lactis HN019 (9×109 cfu) and asked to take one capsule per day. The placebo capsules are identical in appearance, smell and taste and contain corn-derived malto-dextrin and participants in this group will be asked to take one capsule a day. The intervention will last 10 weeks.Participants will be randomized by a computer system to either the treatment or placebo group. Neither the researchers nor the participant will know which group they are in. Randomising participants in this way removes bias from the study because any differences between participants will be randomly distributed between the two groups. Because participants will not know whether they are taking the probiotic or placebo their answers will not be influenced by this knowledge. Adherence will be assessed by providing participants with more capsules than they require and ascertaining the number remaining at the end of the study. This measure of adherence has been used in previous probiotic studies.

**Safety:** The probiotics *Lactobacillus rhamnosus* HN001 and *Bifidobacterium animalis* HN019have been safely used in both pregnant women and infants in New Zealand studies [26, 27]. Similar to other probiotic supplements, they can be purchased over the counter at health food stores and pharmacies.

**Non-university collaborators**

**Dr Raewyn Gavin** is a General Paediatrician at Starship Children’s Hospital with a particular interest in young people with CFS. Dr Gavin has been involved in the selection of questionnaires and design of the study.

**Fonterra:** This is a researcher initiated project, Fonterra Cooperative Limited who manufacture the probiotics *L. rhmnosus* HN001 and *B. animalis* HN019 will supply the probiotics and placebo capsules for this trial.   Both the probiotics and placebo are manufactured to pharmaceutical grade and undergo testing to ensure they comply with safety requirements. As part of the supply agreement, Fonterra will also undertake the randomisation of participants

**Cultural consultation and considerations**

CFS is more common in Caucasian ethnic groups and less common in Māori and Pacific rangatahi, the reasons for which are unclear. With respect to equity of outcomes for rangatahi with CFS this has not previously been investigated and it is possible that inequities in health outcomes may be present.

**Cultural Considerations**

As researchers we take our responsibilities under Te Tiriti o Waitangi seriously. This study has been designed according to the guidelines set out in Te Ara Tika: Guidelines for Māori Research Ethics . More specifically the following principles have been addressed

Whakapapa- relationships

Ideally the relationship between researchers and participants would be established using some kanohi-ki-te-kanohi (face to face) contact. The online consent and data collection methodology in this study means that face to face contact between the researchers and research participants is not possible at the present time. As researchers we acknowledge that this is not an ideal methodology for building a collaborative relationship between the researchers and participants. In our previous studies that have utilised a similar method of online consent and data collection we have had contact with participants in other ways to try and foster a relationship. In this study we aim to consider the importance of relationship by :

Ensuring all participants receive an email when they register for the study with information about where they can seek help with feelings of stress or worry.

Ensuring that the contact details of the researcher are presented in multiple places and encouraging participants to make contact with the researchers if they have questions.

All emails or text messages from participants are responded to personally by the Principal Investigator.

The potential advantage of online consent and data collection is that it reduces the burden of participation by allowing participants to register and take part from any location at a time convenient for them. The study does not require attendance at any clinic visits and can also be safely run under any COVID19 alert level.

Tika

Considering the importance of Tika this study will collect ethnicity data according to the recommended prioritisation ethnicity framework. While we do not anticipate having a sample size big enough to analyse results stratified by ethnicity we recognise the importance of collecting this information so that we can 1) describe our sample and evaluate its relevance to the wider group of nurses working in Aotearoa New Zealand and 2) help to inform future studies collecting ethnicity data.

We hope that the online consent and data collection methodology will reduce barriers to participation in our study.

Mana

The right of participants to be fully informed and to give consent. The Participant Information Sheet fully lays out what participation in the study involves. Parents/caregivers are also included in this study and will need to give their consent for rangatahi aged 13-15 years to participate. In addition for older rangatahi aged 16 years or older, the assent of their parent/caregivers will also be sought. Potential participants are encouraged to discuss participation with whānau. As researchers we also acknowledge our responsibility to treat all participants with respect and aroha and to respect privacy and confidentiality. All data is kept securely and no identifying data is shared with others. Participants have the right to withdraw their data if they wish to.

**Ethical approvals and trial registration**

An application for ethical approval for the study will be made to the Health and Disability Ethics Committee. Locality approvals from ADHB and WDHB will be sought. The trial will be prospectively registered with the Australia and New Zealand Clinical Trials Registry.

**Significance of the Research:** Although there has been a great deal of research interest in the gut microbiota and health, to date, there have been very few studies examining the potential benefit of probiotic supplementation in CFS patients. Those that have been done have been conducted in adults and are limited by very small sample size and lack of a control group. The current study therefore addresses a gap in the field by conducting a study in adolescents and utilizing a randomized, placebo controlled design. If probiotics are found to benefit the psychological wellbeing of adolescents with CFS this may provide a future benefit to patients with CFS particularly given there is no treatment for this condition.

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