Christine Chan, PGY1
CRC IRB Proposal

The effect of prophylactic metronidazole treatment on the incidence of Clostridium *difficile*-associated diarrhea: a prospective, randomized, double-blind, multi-center clinical trial

A. Study Purpose and Rationale

Clostridium difficile is an anaerobic gram positive rod, found in commensal bacteria of the human large intestine in up to 5% of healthy adults. This bacteria produces many toxins, of which the most well-characterized are exotoxin A and B: an enterotoxin and cytotoxin respectively, both responsible for causing clinical disease. Spores produced by this bacteria are thought to be responsible for epidemic infections, as they are extremely resistant to traditional methods of destruction and can be present for months to years. C *difficile* infection (CDI) may range from asymptomatic colonization, C *difficile*-associated diarrhea (CDAD), colitis, pseudomembranous colitis, fulminant colitis, and recurrent CDAD [1]. Major risk factors for CDI include hospitalization, age over 65, and antibiotic exposure [2].

The extensive use of antibiotics which disrupt stable microflora and lead to overproduction of C *difficile,* along with inherent environmental contamination have made patients in healthcare settings extremely vulnerable to C *difficile* infection. Nearly all patients who develop C *difficile* associated diarrhea (CDAD) in the healthcare setting have been exposed to antibiotics [2]. The prevalence of C *difficile* colonization in hospitals may be as high as 20% - 50% in hospitals and up to 50% in long term care facilities. Approximately 20% of hospitalized patients become newly colonized with C *difficile* and of these patients about 30% develop C *difficile* infection via CDAD [4]. In large retrospective studies, the incidence of nosocomial CDI increased by up to 2.5-fold in the late 1990s to the early 2000s compared with earlier periods [3].

C *difficile* has proven to be a substantial burden in the healthcare setting; both economically and in regards to rates of morbidity and mortality. Studies have shown that mortality related to CDI has increased by 2.5 fold between 1993-2003 [5], with mortality rates reported to be as high as 16% in studies investigating highly virulent strains of C *difficile* [6]. Economically, the cost associated with CDI is estimated at $1 billion per year in the US as a result of an additional 3-20 extra hospital days per patient [7]. The heath care burden is likely to worsen for a variety of factors including the emergence of highly virulent strains of C *difficile*, increasing number of hospital admissions, increasing length of hospital stays, increasing antibiotic use, an aging population at higher risk for C *difficile* infection, and a rising incidence of community acquired C diff infection [8].

New guidelines regarding prevention of C *difficile* transmission and infection are therefore paramount. Studies investigating prophylactic treatment with probiotics to decrease the incidence of C *difficile* infection have been largely inconclusive offering no clear evidence to support prophylactic treatment with probiotics [9]. Additionally, while studies show that vaccination for C *difficile* toxin A and B are effective in creating immune responses, there is no sufficient evidence via large scale randomized control trials to determine if these efforts have any preventative effects [13]. The use of prophylactic treatment with metronidazole, however, has not been studied thus far and may offer a novel approach to prevention of C *difficile* infection.

Metronidazole is currently considered the drug of choice and first line treatment of mild to moderate CDAD. While metronidazole does not act to prevent colonization, it may help to keep C *difficile* production at a minimum, reducing the incidence of CDAD. Use of metronidazole prophylactically in hospitalized patients receiving empiric antibiotics may therefore decrease the incidence of C *difficile* associated diarrhea and provide a new standard of care for prevention of C *difficile* infection.

B. Study design and statistical analysis

This study will determine the effect of prophylactic metronidazole treatment versus placebo on the incidence of C *difficile* infection amongst patients in the hospital receiving antibiotic treatment. It will be a prospective, multi-center, double-blind, randomized trial. The trial will be set up on the basis of a power calculation which estimates that for an expected incidence rate for CDAD of 30%, approximately 7,200 patients (3,600 patients in each group) would need to be recruited to detect a 10% difference between the metronidazole treatment and placebo groups in the prevention of CDAD with 80% power and a type 1 error rate of α=0.05. Our primary outcome will be the development of CDAD as defined below. The results will be analyzed using chi square testing and intention to treat analysis.

C. Study procedure

Patients admitted to the hospital and started on antibiotics will be identified. Patients meeting inclusion criteria who are willing to participate in the study will be recruited. Informed consent will be obtained from the patient or health care proxy. Stool samples will be collected on admission, prior to admission. Patients will then be randomized to either the treatment group or placebo group. Patients randomized to the treatment arm will receive metronidazole tablet (250mg PO 4 times a day) while the placebo group will get a similar tablet in appearance to the metronidazole pill at the same dosing intervals. Trial treatments will be initiated within 36 hours of antibiotic administration and continued for 10 days. Patients will be followed for 21 days to observe for the incidence of CDAD as defined by the presence of diarrhea (more than 3 unformed bowel movements or >200ml unformed stools for subjects with rectal collection devices), which tests positive for C *difficile* toxin A, B or both; Or by presence of C *Difficile* toxin without subjective measure of diarrhea.

Bowel habits in regards to consistency and frequency will be documented upon admission and daily via patient interview. In addition, information such as sex, age, comorbid conditions, medications, length of stay, and ICU stay will also be documented for use in possible sub-group analysis.

Patients will be interviewed daily to monitor for diarrhea. Patients will not be allowed to receive any anti-diarrheal medications during the course of the study as this may mask reportable symptoms. Pts will also be asked not to drink alcohol during the course of this study due to possible disulfuram reaction which may unblind patients and investigators to intervention status. Subjects reporting diarrhea during the 21 day trial period will submit a stool sample for analysis. All subjects will submit a final stool sample at the end of the 21 day trial period. Patients discharged from the hospital earlier than 21 days will be called daily at home to monitor for diarrhea until the end of the trial. Stool samples will be submitted to the microbiology lab at

each respective study site and processed within 24 hours. C *difficile* toxin will be detected using EIA assay.

The primary outcome assessed will be the development of CDAD. Intolerance will be defined as the inability or refusal to continue the medication because of adverse reactions. Non-adherence will be defined as missing >3 doses of the study medication during the 10 days of therapy for reasons other than intolerance.

Adverse reactions will be assessed daily by history and physical exam while admitted in the hospital or via patient report over telephone if discharged prior to the end of the 21 day study period.

D. Study Drugs

Metronidazole, a nitroimidazole antibiotic used for treatment against anaerobic bacteria and protozoa, is currently the drug of choice for mild to moderate C *difficile* infection. Common adverse reactions to this drug include nausea, diarrhea, and/or metallic taste in the mouth. More infrequent adverse effects include hypersensitivity reactions, headache, dizziness, vomiting, glossitis, stomatitis, dark urine and parasthesia. Metronidazole has a high rate of efficacy, and is thought to have a lower potential for selection of vancomycin resistant enterococcus (VRE). Furthermore, it is a low cost drug that is readily available [10].

E. Medical Devices

Not applicable

F. Study Questionnaire

Not applicable

G. Study Subjects

Inclusion criteria will include any patient over the age of 18, admitted to the hospital, initiated on antibiotic treatment regardless of the etiology or severity of their underlying illness. Patients will be enrolled if they are willing to complete the study and are able to complete the full course of treatment drugs orally. Patients who test positive for C *difficile* toxin on admission will be excluded from the study. Patients will also be excluded if they receive active treatment with any drug known to act against C *difficile* (including vancomycin, metronidazole, fidaxomicin, andrifaximin) or have received any other antibiotic over the past 14 days prior to admission. Patients who have been hospitalized over the past 14 days prior to admission will also be excluded. Patients on a course of antibiotics lasting longer than the 21 day study period will be withdrawn from the trial with final stool specimen collected. Patients who receive anti-diarrheal agents or consume alcohol during the study period will be withdrawn from the study. Subjects who have a known allergy or hypersensitivity to metronidazole, are pregnant, have a known immunodeficiency, or are receiving chemotherapy will also be excluded. In addition, patients with average of >3 episodes of diarrhea per week at baseline, and known Crohn's disease, Ulcerative colitis, celiac disease or known malabsorption disorder will be excluded.

H. Recruitment of Subjects

Subjects will be recruited for the study upon admission to the hospital once the decision to start antibiotics is made. No advertisements, private practices or clinics will be involved.

I. Confidentiality of Study Data

All participants will receive a unique identifier coded numerically. Any and all personal identifying information including name, address, numbers, medical record numbers will be removed. The information obtained in the study will be stored in a secure database and visible only to study investigators.

J. Conflict of Interest

No study investigators have a financial interest in metrondizole treatment. No conflicts of interest are present.

K. Location of the Study

This is a multi-center study conducted in the US. The study will be subject to review and approval by the IRB at each respective site.

L. Potential Risks

Potential risks include development of side effects from treatment with metronidazole as detailed above. Also with administration of any antibiotics, there is a potential risk of creating antibiotic resistance to metronidazole. Currently there is some literature that suggests there may already exist strains of C *difficile* with antibiotic resistance. Administration of prophylactic metronidazole may therefore act to increase antibiotic resistance, however, rates of resistance are noted to be low and appear stable despite increasing rates of treatment failure; suggesting that any increase in resistance created by this study will be minimal. Furthermore, the mechanism by which C *difficile* develops antibiotic resistance is largely unknown and not well characterized, with some skeptical that widespread antibiotic use has any association with antibiotic resistance [11].

M. Potential Benefits

Subjects may or may not benefit from this study. It is possible that subjects in the treatment group will benefit by a protective effect of the treatment drug and effectively prevent the development of C *difficile* associated diarrhea. Nonetheless, all information derived from this study will be used towards the development of research and preventative guidelines for C *difficile* associated diarrhea which may help to benefit other patients in the future.

N. Alternative Therapies

Studies investigating other prophylactic treatment modalities such as probiotics have been conducted with inconclusive results. Currently there is no sufficient evidence to suggest that prophylactic Priobiotic treatment has any significant effect towards reducing the incidence of CDAD amongst hospitalized patients [12]. Additionally, while studies show that vaccination for C *difficile* toxin A and B are effective in creating immune responses, there is no sufficient evidence via large scale randomized control trials to determine if these efforts have any preventative effects[13]. Most other treatment methods against C *difficile* have been largely aimed at treatment against initial disease or recurrence.

O. Compensation to Subjects
No compensation will be provided to subjects participating in this study.

P. Costs to Subjects
There will be no financial costs incurred to each subject during this study.

Q. Minors as Research Subjects
Not applicable. All research subjects will be >18 years of age.

R. Radiation or Radioactive Substances
Not applicable

References:

1. Vaishnavi, C., *Clostridium difficile infection: clinical spectrum and approach to management.* Indian journal of gastroenterology : official journal of the Indian Society of Gastroenterology, 2011. **30**(6): p. 245-54.
2. Bartlett, J.G., *Historical perspectives on studies of Clostridium difficile and C. difficile infection.* Clinical infectious diseases : an official publication of the Infectious Diseases Society of America, 2008. **46 Suppl 1**: p. S4-11.
3. McDonald, L.C., M. Owings, and D.B. Jernigan, *Clostridium difficile infection in patients discharged from US short-stay hospitals, 1996-2003.* Emerging infectious diseases, 2006. **12**(3): p. 409-15.
4. Bartlett, J.G., *Narrative review: the new epidemic of Clostridium difficile-associated enteric disease.* Annals of internal medicine, 2006. **145**(10): p. 758-64.
5. Ricciardi, R., et al., *Increasing prevalence and severity of Clostridium difficile colitis in hospitalized patients in the United States.* Archives of surgery, 2007. **142**(7): p. 624-31; discussion 631.
6. Pepin, J., L. Valiquette, and B. Cossette, *Mortality attributable to nosocomial Clostridium difficile-associated disease during an epidemic caused by a hypervirulent strain in Quebec.* CMAJ, 2005. **173**(9): p. 1037-42.
7. Wilcox, M.H., et al., *Financial burden of hospital-acquired Clostridium difficile infection.* The Journal of hospital infection, 1996. **34**(1): p. 23-30.
8. Khanna, S., et al., *The epidemiology of community-acquired Clostridium difficile infection: a population-based study.* The American journal of gastroenterology, 2012. **107**(1): p. 89-95.
9. Na, X. and C. Kelly, *Probiotics in clostridium difficile Infection.* Journal of clinical gastroenterology, 2011. **45 Suppl**: p. S154-8.
10. Pelaez, T., et al., *Metronidazole resistance in Clostridium difficile is heterogeneous.* Journal of clinical microbiology, 2008. **46**(9): p. 3028-32.
11. McDonald, L.C., et al., *An epidemic, toxin gene-variant strain of Clostridium difficile.* The New England journal of medicine, 2005. **353**(23): p. 2433-41.
12. Pillai, A. and R. Nelson, *Probiotics for treatment of Clostridium difficile-associated colitis in adults.* Cochrane Database Syst Rev, 2008(1): p. CD004611.
13. Kotloff, K.L., et al., *Safety and immunogenicity of oral inactivated whole-cell Helicobacter pylori vaccine with adjuvant among volunteers with or without subclinical infection.* Infect Immun, 2001. **69**(6): p. 3581-90.