A Randomized, Placebo-Controlled Trial of Granulocyte Colony-Stimulating Factor (G-CSF) to Reduce Incidence of Nucrobial Infection in Neutropenic Patients with AIDS

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A. Study Purpose and Rationale

a. Background

A deficiency of neutrophils, the white blood cells that prevent bacterial infection, is common in patients infected with HIV. An estimated 50-75% of AIDS patients have at least one episode of neutrophil deficiency (neutropenia) during the course of their disease [1]. HIV-related neutropenia may be caused by direct viral infection of bone marrow cells, derangement of growth factors and cytokines, myelosuppressive medications, opportunistic infection, and infiltration of the marrow by neoplastic cells [1].

Hematopoiesis, the process of blood cell production in the bone marrow, is often suppressed by HIV infection. White blood cell production is dependent on the interaction of precursor cells and a network of cytokines (chemicals that are secreted by cells in the bone marrow). Granulocyte colony--stimulating factor (G-CSF) is a cytokine growth factor that is important for neutrophil development.

Neutropenia has been shown to be an independent risk factor for bacterial infection in patients with AIDS [2-3]. In addition, many antivirals, antimicrobials, and chemotheraputic agents used to treat complications of HIV infection cause further myelosuppression, which may prevent administration of these medicines and hinder therapy of HIV. The most common medicines that cause myelosuppression in AIDS patients are dideoxynucleoside analogs, zidovudine, pentamidine, trimethoprim, sulfonamides, ganciclovir, ayclovir, and pyrimethamine [4].

Clinical data from studies of immunosuppressed populations (transplant, cancer, and AIDS patients) indicate that vulnerability to infection increases when neutrophil levels fall below 1000 cells/ml. When the absolute neutrophil count (ANQ is less than 500, the patient is susceptible to infection by endogenous flora [4-6].

G-CSF and granulocyte-macrophage colony-stimulating factor (GM-CSF) were first described in the 1960s, then developed as pharmaceuticals in the mid-80s using recombinant DNA technology. Clinical trials of these agents in cancer patients began in the late 1980s. The FDA approved G-CSF (Filgrastim) in 1991 for treatment of neutropenia in cancer patients getting chemotherapy [5].

There have been several large clinical trials in cancer patients that show G-CSF decreases the duration of post-chemotherapy neutropenia and reduces the incidence of febrile neutropenia in some groups of patients. It has also been shown to reduce infectious complications in patients getting highdose chemotherapy with stem cell rescue. In one trial, G-CSF administration resulted in a decrease in antibiotic use and hospitalization. None of the clinical trials have shown that G-CSF reduces infectious mortality or increases survival [7-8].

AIDS researchers hope to extrapolate the use of G-CSF to neutropenic patients infected with HIV. This research is based on epidemiological studies that indicate an association between neutropenia and bacterial infection in AIDS patients [2-3]. Investigators hypothesize that treatment with G-CSF will reduce infectious mortality and increase survival these patients.

b. Literature review

In vitro studies of blood drawn from HIV-itifected subjects have demonstrated abnormalities of neutrophil function: impairment of chemotaxis, ineffective phagocytosis, and abnormal cellular adhesion molecules. In addition, HIV-infected neutrophils undergo accelerated apoptosis. Other studies indicate

that neutropenic AIDS patients may have abnormal cytokine regulation. Neutropenic AIDS patients have decreased levels of G-CSF compared to neutropenic cancer patients matched for ANC and non-neutropenic controls [1].

Observational studies of fieutropenic patients with AIDS have shown that neutropenia is an independent risk factor for bacterial infection and hospitalization. A matched cohort from Johns Hopkins observed neutropenic AIDS patients from an urban HIV clinic within a four year study period. Patients with ANC<1000 were selected from the cohort (n=118) and matched to nonneutropenic patients (n=118) using CD4 count and several other criteria. Complete medical records were reviewed for "risk window" (duration of neutropenia) (exposure variable) and evidence of bacterial infection (outcome variable). Neutropenic patients were stratified by ANC for the analysis. The incidence of bacterial infection was calculated based on number of infections per person-months of neutropenia. Results showed 2-3 infections per 100 person-months of neutropenia for patients with ANC less than 1000 and 3-5 infections per 100 person-months of neutropenia for patients with ANC less than 500. This is a relative risk of 2.33 for the occurrence of bacterial infection for patients with an ANC less than 1000 (CI 1.00-5.40, p=.05), and a RR of 7.92 for patients with an ANC of less than 500 (CI 1.18-53.2, p=.03) [2].

A retrospective analysis from UCSF analyzed risk of hospitalization for bacterial infection in 2047 patients with HIV based on ANC stratum. The analysis calculated incidence of bacterial infection per days at risk for each ANC stratum. 320 patients (16%) were hospitalized for bacterial infection during the study period. The incidence of infection per 10,000 days at risk doubled for each stratum below ANC=1000. The results indicate that severity and duration of neutropenia are associated with increased incidence of hospitalization for bacterial infection [3].

A retrospective cohort of I52 neutropenic AIDS patients (ANC<1000) from Parkland Hospital in Texas compared 71 patients treated with G-CSF (ANC nadir 355) to 81 neutropenic controls never treated with G-CSF (ANC nadir 531). Medical records were reviewed for incidence of bacteremia. The results showed decreased rates of bacteremia in the G-CSF group compared to the controls (0.54 bacteremias/100 patient months versus 2.2 bacteremias/100 patient months; RR 0.25, p=0.06) [9].

A number of small pilot studies demonstrate increase in neutrophil count in neutropenic AIDS patients given G-CSF; dose 1 ug/kg/day- 10 ug/kg/day; median time to ANC>2000 was 2 days [1].

An open-label non-comparative study of 200 neutropenic HIV infected patients (ANC<1000) showed G-CSF reversed neutropenia in 98% of study patients with median time to reversal two days. The study also looked at changes in dosing of myelosuppresive drugs in each group. The number myelosuppressive drugs taken by each patient in the treatment group increased by more than 20% during the study period. Investigators concluded that G-CSF allows greater use of myelosuppressive medicine [10].

There is one published randomized controlled clinical trial where G-CSF or placebo was given to 258 neutropenic patients (ANC 750-1000) with AIDS. Patients with CD4 counts less than 200 were randomized into daily G-CSF, intermittent G-CSF, or placebo. The primary outcome measured was incidence of severe neutropenia (<500). Secondary endpoints included incidence of bacterial infection, duration of hospitalization, and IV antibiotic use. The study found that severe neutropenia was 34% less common in patients getting G-CSF. Also, G-CSF patients developed 31% fewer bacterial infections and 54% fewer severe bacterial infections than controls, and that these patients required 45% fewer hospital days for bacterial infections [11].

c. Hypothesis

The summary of previous literature indicates that neutropenia is an independent risk factor for bacterial infection in AIDS patients. In addition, treatment of neutropenia with G-CSF prevents severe neutropenia and may reduce the risk of infectious morbidity.

The goal of this study is to demonstrate that administration of G-CSF to neutropenic AIDS patients will reduce infectious morbidity. The primary hypothesis is that a smaller proportion of GCSF treated patients will get bacterial infections than controls. The secondary hypothesis that there will be increased time to infection in the test group.

B. Study Design and Statistical Analysis

This study is a 24-week, multi-center, randomized, placebo-controlled, single-blind clinical trial. The patients will be recruited from the CPMC Harkness-6 HIV clinic and similar urban HIV clinics in New York City.

a. Eligibility

Study patients must be at least I8 years old and have documented HIV infection via EUSA and confirmed by Western blot. To be included in the study the patients must have a CD4 count of less than 200 and ANC less than 1000. In addition, doses of anti-retroviral medications, TMP-SMX, amphotericin B, and gancyclovir must remain constant within 14 days of randomization. Patients will be excluded from the study if they have a known malignancy for which treatment with chemotherapy is standard of care. Patients will also be excluded if they are pregnant or breast-feeding or have received treatment with growth factors within 14 days of randomization.

b. Randomization

After a patient has been deemed eligible to participate, informed consent will be obtained by the study PI. All patients will be stratified to one of four groups based on their original ANC: 0-300; 301--499,500-749,750-1000. Patients from each ANC stratum will be randomized to either test group or placebo group. Both patients and primary medical doctors will be blinded to group assignment.

c. Enrollment

We expect to enroll approximately 200 patients (100 patients/group). This number was determined using a chi-squared power analysis using estimates of the proportion of patients in each group to reach the primary outcome. We expect 40% of the control group and 20% of the test group to get an infection during the 24 week follow-up period. These estimates are based on the findings of the retrospective studies and the clinical trial described above. [The observational studies showed approximately 40% of neutropenic patients (ANC 0-1000) developed a microbial infection. The clinical trial found that 33% of neutropenic patients (ANC 750-1000) developed an infection; the GCSF group got 31% fewer infections that the control group. Because our study enrolls patients with lower ANCs, we expect to see a larger number of infections in both groups and a greater effect of GCSF treatment.]

d. Study Design

The test group will receive subcutaneous G-CSF at an initial dose of 5 ug/kg/day while the control group will receive subcutaneous placebo injection. Neutrophil counts will be monitored via measurement of CBC with differential three times per week for first two weeks of the study. G-CSF dose will be adjusted (1 ug/kg/day to 10 ug/kg/day) to titrate ANC to 2000-1000. After the first two weeks CBC will be monitored once a week for remaining 22 weeks.

Patients in both groups will receive conventional treatment for HIV and prophylaxis for opportunistic infections at discretion of their primary medical doctor (PMD). Regardless of group assignment, each PMD will adjust myelosuppressive drugs based on clinical scenario. Study participants will be followed for 24 weeks. The presence of microbial infection will be determined by in-hospital physicians (housestaff and attending physicians).

Outcomes to be measured are incidence of microbial infection and length of time until infection. Microbial infections will be assessed according to the following criteria:

i. *Bacteremia*--pathogen growth in one or more peripheral blood cultures
ii. *Line infection*- pathogen growth in one or more peripheral blood cultures and simultaneous growth of >15 colonies of same pathogen from IV line

iii. *Bacterial or fungal pneumonia*- findings on physical exam consistent with pneumonia (crackles, consolidation, egophany) new infiltrate, cavitation, or pleural effusion on CXR and diagnostic sputum gram stain or positive culture from BAL.
iv. *Meningitis*-physical findings c/w meningitis (photophobia, headache, nucal rigidity) and identification of pathogen on gram stain of CSF or growth of pathogen in CSF culture.
v. *Infection of deep tissue site/abscess*-focus of infection seen on imaging procedure and one of following: isolation of organism from culture, or tissue sample with histopathological evidence of infection
vi. *Enducarditis*- at least two positive blood cultures positive and at least one of- new murmur, vegetation seen on ECHO, or embolic evidence of endocarditis or evidence of vegetation seen at surgery or autopsy
vii. *Enterocolitis*- clinical indications of bowel infection (diarrhea, blood stool, abdominal tenderness) with leukocytes documented in stool and culture positive stool sample or toxin positive stool sample

e. Statistical analysis

All analysis will be on an intention to treat basis. The proportion of patients in each group who develop a microbial infection will be analyzed via the chi-squared test. The time until first microbial infection in each group will be analyzed with a Kaplan-Meier analysis.

C. Study Procedures

a. Follow-up and duration

This study will require participants to have an initial visit and several follow-up visits with study investigators. In the first two weeks of the study each patient will be seen for a blood draw and medication dose adjustment three times per week. Participants will receive G-CSF or placebo at these visits. For the remaining 22 weeks of the study, follow-up visits with blood draws will be done weekly.

Patients will continue to follow-up with their PMD for usual care. The follow-up interval will be determined by the individual PMD. In the event that a patient is hospitalized for an infection, care will not differ from usual hospital care. Patients will continue to self-administer study medication while in the hospital.

b. Drug administration

The study requires G-CSF/placebo to be administered by subcutaneous injection. Study personnel will teach patients to self~administer treatment/placebo. After an initial teaching session of fifteen minutes, self-administration will take approximately five minutes per day. There is minimal discomfort involved in subcutaneous injection. A small amount of bruising surrounding the injection site is sometimes seen.

D. Study Drugs

The FDA approved G-CSF (Filgrastim) in 1991 for treatment of neutropenia in cancer patients getting chemotherapy. The usual G-CSF dose in these patients is 5 ug/kg/day administered subcutaneously. Safety and tolerance in all clinical trials was good; the predominant side effect was been bone pain, occurring in 15-39% of patients in the test groups compared to 0%-21% of controls. Infrequent side effects include rashes, and allergic reactions, and mild alopecia. The laboratory changes seen with G-CSF treatment included increases alkaline phosphatase and uric acid [6-7].

G-CSF has not been approved for treatment of neutropenia in AIDS patients. As describedabove, other studies have suggested that G-CSF may reduce infectious morbidity in neutropenic patients with AIDS. Its use may also allow use of myclosuppressive medications, which may need to be discontinued

in neutropenic patients treated with standard therapy. In our study G-CSF Will bb administered in the same route and similar doses as in cancer patients.

E. Medical Devices

N/A

F. Study Questionnaires

N/A

G. Study Subjects

Study participants must be at least 18 years old and have documented HIV infection via ELISA and confirmed by Western blot. To be included in the study the patients must have a CD4 count of less than 200 and ANC less than 1000. In addition, doses of anti-retroviral medications, TMP-SMX, amphotericin B, and gancyclovir must remain constant within 14 days of randomization. Patients will be excluded from the study if they have a known malignancy for which treatment with chemotherapy is standard of care. Patients will also be excluded if they are pregnant or breast-feeding or have received treatment with growth factors within 14 days of randomization.

H. Recruitment of Subjects

Patients will be recruited from the CPMC HIV clinic (Harkness-6) and similar HIV clinics at NYC medical centers. They will be referred to the study investigators by their PMDs and study flyers will be posted at the clinic.

I. Confidentiality of Study Data

All study data will be confidential.

J. Potential Conflict of Interest

Neither of the study investigators nor the hospital has a proprietary interest in G-CSF.

K. Location of Study

The study will be located within the Harkness-6 clinic and on the HIV inpatient floors at CPMC, as well as within the confines of other NYC HIV clinics and inpatient HIV floors.

L. Potential Risks

As described above, safety and tolerance of G-CSF in all clinical trials has been good. In previous studies involving the same drug dose and mode of administration the predominant side effect was bone pain, occurring in 15-39% of patients in the test groups compared to 0%-2 1 % of controls [6-7].

Although G-CSF is FDA-approved to decrease incidence of febrile neutropenia in cancer patients getting myelosuppressive chemotherapy, it has never been shown to make a difference in infectious mortablity or survival between G-CSF treated patients and placebo patients [7-8]. There has not yet been a study using G-CSF on neutropenic AIDS patients with enough power to show a definitive reduction in infectious morbidity in the test group. Bemuse G-CSF is not considered standard of care this population, the placebo group is not considered to be at additional risk.

M. Potential Benerits

The goal of this study is to demonstrate that administration of G-CSF to neutropenic AIDS patients will reduce infectious morbidity. Subjects treated with G-CSF may or may not get fewer infections than control patients. In addition, patients in the test group may be able to take higher doses of myelosuppressive medicines. If treatment with G-CSF can reduce the proportion of infections in neutropenic AIDS patients, money may be saved on the cost of hospitalization and treatment for this patient population.

N. Alternative Therapies-

N/A

O. Compensation to Subjects

No compensation will be provided to study participants.

P. Costs to Subjects

Patients are required to pay for transportation to the clinic for follow-up visits.

Q. Minors as Research Subjects –

N/A

R. Radiation or Radioactive Substances-

N/A

S. Bibliography

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