A randomized, double-blind, placebo-controlled trial of intravenous cyclophosphamide plus oral thalidomide versus cyclophosphamide alone in patients with relapsed or refractory multiple myeloma.

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A. Study Purpose and Rationale

Multiple myeloma (MM) is a disorder characterized by the accumulation of malignant plasma cells mainly in the bone marrow. It accounts for about 10% of hematologic malignancies and 2% of cancer-related deaths, with a twofold greater incidence in blacks than whites. Its clinical manifestations are heterogeneous, but common complications include recurrent bacterial infections, anemia, hypercalcemia, renal insufficiency and osteolytic bone lesions. The pathogenesis of these features depends on the aberrant phenotype and genotype of MM cells and monoclonal immunoglobulin (M-protein) which is produced by the tumor cells. The extent of bone marrow infiltration together with the presence of the above complications and the characteristics of the M-protein determine tumor load, as reflected in the Durie-Salmon risk stratification. [1,2]

On a biological level, MM is characterized by multiple chromosomal and genetic abnormalities that can affect disease outcome. Underscoring the significance of cell interactions with the marrow microenvironment, several adhesion and signaling molecules have been implicated in the pathogenesis of this disease. Most notably, IL-6 and to a lesser extent IL-1b, TNF-α, G-CSF, IFN-αamong others have been implicated in the survival and growth of MM cells as well as the clinical manifestations of the disease. As a matter of fact, IL-6 and CRP, produced by hepatocytes in response to the former, together with beta2-microglobulin, probably produced by the neoplastic cells, can be used to predict clinical outcome. Several immunotherapeutic approaches targetin cell surface antigens and cytokines have shown promise in animal and human studies. [1,2,3]

With the introduction of melphalan in the 1950s, the median survival of myeloma patients was extended from 7 months to 3 years. Melphalan and prednisone soon became the accepted first line treatment and remain so today, especially for the elderly or those with a reduced performance status at presentation. It wasn't until the 1980s that more aggressive chemotherapy regimens were developed with the hope of effecting more marked and rapid cytoreduction with a curative intent. Among them, VAD has produced the best results and thus became an alternative first-line combination in younger patients who may be candidates for bone marrow transplantation down the line. With any regimen, most patients fail to achieve a complete disappearance of their M-protein but rather reach a stable reduction and thus attain a plateau. Today, relapse following initial treatment of MM is universal. Younger patients are candidates for high-dose chemotherapy with autologous stem cell support while older people find their options to be limited, especially given the significant toxicity associated with cytotoxic chemotherapy.[1,4]

Cyclophosphamide, an alkylating agent with decidedly less hematological. toxicity than melphalan, has been utilized in the treatment of this disease since the 1960's. Early on, in prospective clinical trials, low dose cyclophosphamide therapy was associated with a median survival of 24 months in chemotherapy naïve patients. Its effectiveness in a melphalan resistant mouse myeloma model prompted investigators to use low dose therapy in melphalan refractory patients with initially mixed results. It was Lenhard et al, [5] as part of the ECOG in 1984, that first utilized high dose cyclophosphamide (2.4g /M^2) given over 4 days in monthly cycles and demonstrated a 43% objective and 62% subjective response in treatment refractory patients with a median survival of 8 mos. Subsequent studies by the same group confirmed the effectiveness of high dose therapy alone, with a) no additional benefit afforded by the

addition of prednisone and with b) a significantly increased toxicity and questionable efficacy of giving the cumulative dose in a single day. [6,7]

It has been postulated that cytotoxic chemotherapy decimates the dominant, more mature tumor cell compartment but has little effect on the MM precursor cell pool. The latter is protected via autocrine and paracrine IL-6 production, which confers an important antiapoptotic signal that interferes with tumor-cell kill.[8] Therefore, drugs that affect the myeloma cell microenvironment may augment the activity of cytotoxic therapy.

Thalidomide was used widely in Europe and Australia in the late fifties as a sedative and potent anti-emetic in the first trimester of pregnancy but was subsequently withdrawn in 1961 after a rapid rise of catastrophic fetal abnormalities. However, even before its withdrawal, it was recognized as a highly effective regimen in the treatment of erythema nodosurn leprosum (ENL), a use that has recently gained FDA approval in the US. Clinical evidence suggests that thalidomide has powerful immunomodulatory and steroid-sparing immunosuppressant properties that can be harnessed in the treatment of diseases such as vasculitis, HIV, RA, sarcoidosis and GVHD. At the molecular level, thalidomide exhibits potent TNF-oc inhibition, with its synthetic derivative MA-3 having an even more pronounced effect on monocyte proliferation and production of TNF-α, GM-CSF, IL-6 and IL-10, as well as, to a lesser extent, IL-2 and IFN-,γ. [9,10] These biological properties make it a promising candidate in the treatment of MM, especially in association with a potent proven cytotoxic agent.

Furthermore, thalidomide may be effective in this disease through its antiangiogenic properties, felt to be largely responsible for its teratogenic potential, its effect on oxidative damage to DNA or the modulation of cell surface adhesion molecules in the marrow microenvironment.[11] Recently, Durie et al [12] used low-dose thalidomide (50-400 mg) in the treatment of relapsing or progressive MM with a >50% response in 6 out of 20 patients and a well-tolerated side effect profile with only 5/20 discontinuing the drug secondary to toxicity. In a phase 2 trial, Singhal et al [13] used thalidomide as a single agent in doses ranging from 200 to 800 mg in refractory MM with a 32% overall response, defined as the reduction of serum or urine M-protein by at least 25%. Most adverse effects reported where mild or moderate and 9/84 patients discontinued the drug secondary to toxicity.

B. Study Design and Statistical Analysis

This is a randomized, double-blind, placebo-controlled clinical trial of thalidomide in addition to cyclophosphamide for the treatment of relapsed or refractory multiple myeloma. Following a pretreatment evaluation for determination of eligibility, subjects will be randomly assigned to receive either cyclophosphamide with thalidomide or cyclophosphamide with placebo, by coin toss. The serum and urine M-protein levels along with additional biochemical parameters, as detailed in section C, will be determined at baseline and monthly intervals thereafter. A ' bone marrow aspiration and biopsy will be performed at baseline and 8 weeks later to determine the extent of plasmacytosis. The results will be evaluated on an intention-to-treat basis. Toxicity will be ascertained at each clinical follow-up and scored according to the NCI toxicity criteria.

The primary endpoint will be reduction of the serum M-protein by at least 50% or, in patients lacking a serum M-protein, a reduction in the urine M-protein by at least 90% at two consecutive measurements 4 weeks apart. Patients with an increase of the serum or urine M-protein by at least 25% from baseline or subsequent treatment nadir will be considered to have progressive disease. Secondary endpoints will include the biochemical marker response to treatment ' the bone marrow response as well as event-free and overall survival. A biochemical marker response will be defined as a decrease in CRP or beta2-microglobulin of at least 50% from baseline sustained over 4 weeks. A bone marrow response will be defined as a reduction in plasma cell content by at least 50% on repeat examination, compared to pretreatment values. Event-free survival will be calculated from the start of therapy to the time of disease progression, removal from the study, death or last follow-up, whichever occurs first. Overall survival will be calculated from the start of therapy to the time of death from any cause or last follow-up.

As the observed response to cyclophosphamide alone is about 30% and preliminary studies suggest a comparable response rate to thalidomide, the desired outcome would be a 60% response rate in the combined treatment arm. It was thus calculated that 47 patients per arm are required to show a 60% response with 80% power and a two-tailed alpha set at 5%. Response will be treated as a categorical variable and comparisons will be assessed via the chi-square test or Fisher's exact test, as applicable. Event-free and overall survival will be computed via the Kaplan-Meier method and compared with the log-rank test.

C. Study Procedures

After signing a consent form, patients will, be evaluated with a complete medical history and physical examination including screening for peripheral neuropathy. The pretreatment evaluation will also include a complete blood count, a serum BUN and creatinine to assess renal function, a serum AST/ALT level for hepatic function, quantitative serum and 24-hour urine immunofixation electrophoresis for M-protein determination, a urinalysis, a serum immunoglobulin panel, beta2-microglobulin, CRP, uric acid and a calcium/albumin level. A baseline bone marrow aspirate and biopsy will be performed to assess the extent of plasmacytosis. Along with the above diagnostic tests that constitute the standard of care for this disease, a peripheral nerve conduction study will also be obtained at baseline and repeated every six months thereafter to assess the neurological toxicity of thalidomide.

Following this initial evaluation, eligible patients will be randomized to treatment with either cyclophosphamide alone, 24 hours after admission for intravenous hydration, at a cumulative dose of 2.4 g/m' divided over 4 days as a 1-2 hour daily IV infusion with hospital discharge 24 hours later, or cyclophosphamide at the above regimen plus nightly thalidomide starting at a 200 mg (four 50 mg tablets) daily oral dose. The thalidomide dose will subsequently be increased by 200 mg increments over 3 weekly intervals, as tolerated, to a target dose of 800 mg nightly (Sixteen 50 mg tablets). Patients on cyclophosphamide alone will receive an equal number of nightly placebo tablets increased weekly by the same method. A second cycle of cyclophosphamide will be administered to the patients without progressive disease, 4 weeks after randomization in the same manner as the first cycle, with delay as needed to permit return of blood counts to baseline levels. Adjustments in the cumulative dose of the second cycle will be permitted for prior prolonged hematologic toxicity. A monthly ' maintenance cyclophosphamide dose of 750 mg/m' along with the highest tolerable nightly thalidomide dose will be administered thereafter to patients without disease progression by 8 weeks after randomization.

Follow-up office visits will be conducted weekly over the first 4 weeks and monthly thereafter, in order to assess thalidomide tolerance and toxicity. If symptoms or signs of drug-induced neuropathy develop, thalidomide will be discontinued and restarted at the next lower dose only upon complete symptom resolution. In accordance with the accepted standard of care, all serum and urine biochemical measurements will be repeated monthly for the duration of the study. A repeat bone marrow aspiration and biopsy will be performed at 8 weeks or when patients leave the study, whichever is earlier.

D. Study Drugs

Thalidomide was approved by the FDA in 1998 for the treatment of ENL, the acute reactional phase of lepromatous leprosy. The rationale for its utilization in multiple myeloma stems from its immunomodulatory and antiangiogenic properties as well as its hypothesized effect on oxidative damage to DNA or the modulation of cell surface adhesion molecules in the marrow microenvironment. As detailed above, it has been studied in chemotherapy refractory MM patients with a 30% response as a single agent. It will be administered in oral doses ranging from 200 to 800 mg per day, as tolerated. The most serious toxicity associated with thalidomide is its well documented human teratogenicity. It is for that reason that women of childbearing potential will be excluded from our study. As it remains unknown whether thalidomide is present in the ejaculate of men, male patients will be required to register with the STEPS program and use barrier contraception during heterosexual intercourse. The most common side

effects in clinical trials over the past 20 years of experience include somnolence, dizziness and hypersensitivity rash. Other side effects include potentially severe peripheral neuropathy that may be irreversible upon discontinuation of treatment, orthostatic hypotension, bradycardia, neutropenia and increase of the viral load in HIV-seropositive patients.

Cyclophosphamide is an FDA approved alkylating agent with well established efficacy in the treatment of malignantand nonmalignant diseases. It has a long history of clinical use in multiple myeloma based on its antineoplastic properties as a cell-cycle specific drug. It will be administered intravenously in a cumulative dose of 2.4 g/m~ divided over 4 days in two monthly treatment cycles, and at 750mg/m' at monthly intervals thereafter. The second cycle dosage will be adjusted for the occurrence of leukopenia. The most common side effects of therapy include dose-limiting leukopenia. and increased incidence of infection, sterility, alopecia, nausea and vomiting, rarely severe hemorrhagic cystitis and urinary bladder fibrosis and the occurrence of second malignancies.

E. Medical Devices

Non applicable.

F. Study Questionnaires

Non applicable.

G. Study Subjects

a. Inclusion Criteria

- Patients must be able to understand and sign an informed consent document.
- Patients must be 18 years of age or older.
- Male patients must be able to comply with the requirements of the STEPS program.
- Female patients must be post-menopausal, i.e. amenorrheic for at least 24 months or a status post hysterectomy.
- Patients must carry the diagnosis of multiple myeloma, as defined by the Leukemia and Myeloma Task Force.
- Patients must have failed prior standard therapy for multiple myeloma, including melphalan/prednisone, intensive chemotherapy with or without autologous hematopoietic stem cell support or allogeneic bone marrow transplantation.
- Patients must have quantifiable disease via either a serum M-protein greater than 1 g/dL or a urine M-protein greater than 200 mg over 24 hours.
- Patients must exhibit evidence of disease progression as defined by an increase in the serum or urine M-protein levels of at least 25% from their nadir on therapy or at least 50% plasma cells in the pretreatment bone marrow biopsy.
- Patients with a concurrent reversible complication of MM, e.g. hyperuricemia, hypercalcemia, spinal cord compression, will be eligible once appropriate therapy for this complication has been initiated.
- Patients must be afebrile while not on any antibiotics during the week prior to randomization.
- Patients must be willing to be admitted to CPMC for the administration of cyclophosphamide and subsequently travel from their home to CPMC for follow-up visits and necessary diagnostic testing.

b. Exclusion Criteria

- Patients receiving antiretroviral therapy for HIV disease will not be eligible.

- Patients with concurrent malignancies, other than nonmalignant skin cancer, or life threatening illnesses will not be eligible.
- Patients that have received corticosteroids, cytotoxic chemotherapy or radiation therapy in the 4 weeks prior to randomization will be excluded.
- Patients who have previously demonstrated hypersensitivity or other severe adverse effect from cyclophosphamide or thalidomide will not be eligible.
- Patients who have had prior treatment with cyclophosphamide at doses of 1g/m' per cycle or more will be excluded.
- Patients with an absolute neu.trophil count of less than 1000/mm' and serum AST/ALT levels greater than 2 times normal at the pretreatment evaluation will not be eligible.
- Patients with NCI/CTEP grade 2 or greater peripheral neuropathy at the pretreatment evaluation will not be eligible for this study.
- Patients receiving sedative /hypnotic agents such as barbiturates, benzodiazepines, chlorpromazine, reserpine or alcohol, as well as anticoagulant or anticonvulsant medications that cannot be discontinued prior to study entry, will be excluded.

H. Recruitment of Subjects

Only patients whose primary attending physician has agreed that they are suitable for the study and has ascertained from the patient that he or she is willing to be approached by the research team, after discussing standard and investigational therapies for this disease, will be contacted by the investigators. While eligibility may be assessed over the phone, solicitation and screening will be performed in the office.

I. Confidentiality of Study Data

All study files will be coded and stored in a secure locked office accessible only by the investigators.

J. Potential Conflict of Interest

No investigator has a proprietary interest in any of the drugs under investigation, or might stand to benefit financially or in any other way from the results of this investigation.

K. Location of the Study

The study will be conducted at the inpatient and outpatient facilities of the department of oncology at the Columbia-Presbyterian campus of the New YorkPresbyterian Hospital.

L. Potential Risks

Participants may not benefit from this study and in addition may experience certain adverse side effects as described in section D above. As the treatment is experimental, there may be further side effects that may have not been seen previously. - Most of the side effects are temporary, reversible and clinically manageable, but they could potentially lead to serious or even fatal outcomes. Other risks are not different from those seen in the patient population with this disease undergoing standard treatment.

M. Potential Benefits

Participants may or may not benefit personally from this study. The possible benefit from this study is a longer period of disease control, if the study treatment is effective. Information derived from this trial will be useful in the treatment of multiple myeloma in the future.

N. Alternative Therapies

No therapy, or supportive care without chemotherapy, in which case the disease will continue to progress.

Off-study treatment with cyclophosphamide or other conventional chemotherapy.

New experimental agents under study.

O. Compensation to Subjects

Participants will receive no compensation for participation in this study.

P. Costs to Subjects

Participants will not be charged for the cost of thalidomide or any tests and procedures performed solely for research purposes. Participants or the appropriate third party, will be charged for cyclophosphamide, routine blood tests, radiologic evaluation and office visits that are considered part of the standard care for this disease.

Q. Minors as Research Subjects

Only adults 18 years of age or older will be included in this study.

R. Radiation or Radioactive Substances

Non applicable.

S. References

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