Protocol Title: Phase II Study of Atorvastatin, Micro-Dose Methotrexate and Tacrolimus Administered only to Transplant Recipients for the Prophylaxis of Acute Graft-Versus-Host Disease Following Allogeneic Hematopoietic Cell Transplantation

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Principal Investigator:

Mehdi Hamadani, MD
Associate Professor of Medicine,
Division of Hematology and Oncology
Froedtert Hospital and the Medical College of Wisconsin
9200 W. Wisconsin Ave.
Milwaukee, WI 53226

Co-Investigators:

Parameswaran Hari, MD
Mary M. Horowitz, MD
William Drobyski, MD
Timothy S. Fenske
Douglas Rizzo, MD
Marcelo Pasquini, MD

Wael Saber, MD
Michael Craig, MD
Abraham Kanate, MD
William Petros, PharmD
Aaron Cumpston, PharmD
Scot C. Remick, MD

Statistician:

Alexis Visotcky, MS
Biostatistician II
Division of Biostatistics
Institute for Health and Society
avisotcky@mcw.edu

Study Coordinator:
For Medical College of Wisconsin:
Barbara Davies
Division of Hematology and Oncology
Froedtert Hospital and the Medical College of Wisconsin
9200 W. Wisconsin Ave.
Milwaukee, WI 53226
Phone: 414-805-8926
Fax: 414-805-0596
Email: bdavies@mcw.edu

For West Virginia University:
Pam Bunner, MT, CRCC
Clinical Research Specialist
Osborn Hematopoietic Malignancy and Bone Marrow Transplant Service
Mary Babb Randolph Cancer Center
PO Box 8110
One Medical Center Drive
Morgantown, WV 26506
Phone: (304) 598-4511
Fax: (304) 598-4519
Email: bunnerp@wvuhealthcare.com

Pharmacists

Angela Urmanski, Pharm D.
Froedtert Hospital
Inpatient Pharmacy
Attn: Senad Novic
9200 W Wisconsin Ave
Milwaukee, WI 53226

Lisa-Giblin Sutton, Pharm D.,
Investigational Drug Pharmacist
West Virginia University Hospitals Pharmaceutical Services
PO Box 8045
One Medical Center Drive
Morgantown, WV 26506
Phone: (304) 598-4000 x73760
Fax: (304) 598-4073
Email: giblinl@wvuhealthcare.com

Schema

Phase II study of Atorvastatin, micro-dose methotrexate and tacrolimus administered only to transplant recipients for the prophylaxis of acute graft-versus-host disease following allogeneic hematopoietic cell transplantation.

PRIMARY OBJECTIVES:

1. Determine the efficacy of an atorvastatin/tacrolimus/methotrexate regimen in preventing grade II-IV acute GVHD in patients undergoing allogeneic hematopoietic stem cell transplantation (HSCT).
2. Assess the safety of an atorvastatin/tacrolimus/methotrexate regimen in patients undergoing allogeneic HSCT.

SECONDARY OBJECTIVES:

1. To assess rates of chronic GVHD.
2. To assess rates of late onset acute GVHD at days +180 and +365.
3. To assess rates of grade III-IV acute GVHD at days +100, +180 and +365.
4. To assess non-relapse mortality at 100 days post-HSCT.
5. To assess time to successful neutrophil engraftment.
6. To assess time to successful platelet engraftment.
7. To assess incidence of primary and secondary graft failure.
8. To assess incidence of primary and secondary graft rejection.
9. To assess relapse rate of the primary hematological malignancy.
10. To assess lineage specific chimerism kinetics in patients at days +28, +100, +180 and +365.
11. To assess immune reconstitution following transplantation at days +28, +100, +180, and +365.
12. To assess 1 year progression free survival (PFS) and overall survival (OS) following transplantation.
13. To evaluate biologic and genomic markers potentially associated with GVHD and/or atorvastatin.

STUDY DESIGN:

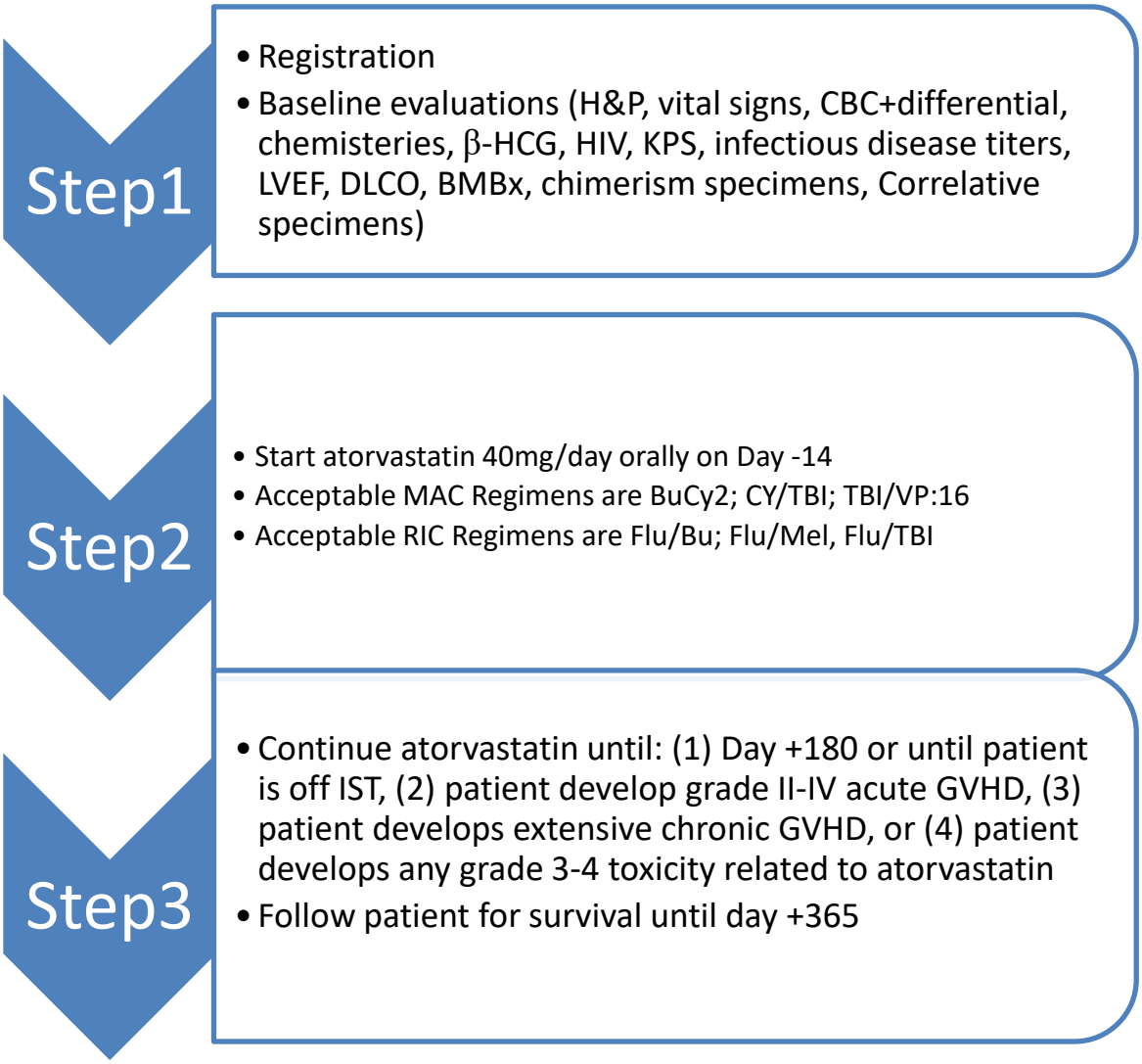
This is a phase II study of atorvastatin for the prophylaxis of acute GVHD in patients undergoing allogeneic HSCT. The study will have two separate arms: (a) Matched sibling transplants, and (b) Matched unrelated donors transplants. Both arms will be enrolled, and analyzed separately.

PATIENT ELIGIBILITY CRITERIA:

1. Patients with a history of a hematological malignancy or bone marrow failure syndrome suitable for allogeneic stem cell transplantation in the opinion of treating transplant physician.
2. Patients aged 18-75 years of age are eligible. Patients with age ≥ 18 and ≤ 50 years will be eligible for myeloablative conditioning (MAC), while patients > 50 years of age, or those with previous history of autologous transplantation, high hematopoietic cell transplant comorbidity index (HCT-CI) score (>2), and baseline diagnosis of hodgkin's lymphoma, chronic lymphocytic leukemia and follicular lymphoma will be suitable for reduced intensity conditioning (RIC) transplantation (however intensity of conditioning regimen will remain at the discretion of treating physician).
3. All patients must have at least one suitable HLA-matched sibling or unrelated donor according to transplant center's guidelines (for selection of appropriate donor).
4. Patient must provide informed consent.
5. Left ventricular ejection fraction ≥ 40%. No uncontrolled arrhythmias or uncontrolled New York Heart Association class III-IV heart failure.
6. Bilirubin ≤ 2 x the ULN and AST, and ALT ≤ 3 x ULN; and absence of hepatic cirrhosis. For patients with Gilbert's syndrome, bilirubin ≤ 3 x ULN is permitted.
7. Adequate renal function as defined by a serum creatinine clearance of ≥ 40% of normal calculated by Cockcroft-Gault equation.
8. DLCOcor (corrected for hemoglobin) or FEV1 or DL/VA ≥ 40% of predicted.
9. Karnofsky performance status ≥ 70.
10. A negative pregnancy test will be required for all women of child bearing potential. Breast feeding is not permitted.
11. Patients with positive HIV serology **are** eligible.
12. No evidence of active bacterial, viral or fungal infection at the time of transplant conditioning.
13. Patients with history of intolerance or allergic reactions with atorvastatin will not be eligible.

14. Patients who have previously been taking atorvastatin or any other statin drug will be eligible as long as there is no contraindication to switch to atorvastatin (40mg/day) in the opinion of the treating physician.
15. Patients undergoing a T-cell depleted allogeneic transplantation will not be eligible.
16. Patients receiving conditioning regimens containing antithymocyte globulin, and/or campath will not be eligible.
17. Method of stem-cell collection from the donor will be at the discretion of the treating physician. Although it is anticipated that majority of donors will undergo G-CSF induced stem cell mobilization; however donors undergoing bone marrow harvest or stem cell mobilization with experimental agents (e.g. plerixafor) will remain eligible for the study.

Patient Treatment Plan



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1. Introduction:

1.1. Graft versus Host Disease:

Acute graft-versus-host disease (GVHD) is one of the most frequent complications after allogeneic hematopoietic stem cell transplantation (HSCT) (1). It develops in 30-75% of recipients of allogeneic HSCT depending on the degree of histocompatibility between the donor and the recipient, number of T-cells in the graft, recipient's age and GVHD prophylactic regimen used (2-4). Novel strategies designed to effectively prevent the development of this life threatening complication of allogeneic transplantation are urgently needed.

1.2. HMG-CoA Reductase Inhibitors:

HMG-CoA reductase inhibitors (statins), the cholesterol-lowering drugs that are prescribed worldwide to prevent and treat atherosclerosis (5), are increasingly being recognized to exhibit a variety of immunomodulatory properties (6). The mechanism of action of statins involves binding to HMG-CoA reductase and displacement of its natural substrate, HMG-CoA, leading to inhibition of mevalonate and cholesterol biosynthesis (5).

1.3. Immunomodulatory effects of Statins:

In addition to lowering lipid concentrations, statins have recently been shown to possess potent immunomodulatory and anti-inflammatory properties that are relevant not only in the context of atherosclerosis-associated inflammation but also in autoimmune diseases such as multiple sclerosis and rheumatoid arthritis (7-9). Reduced mevalonate production by statins, not only inhibits the synthesis of cholesterol but also that of key isoprenoid intermediate molecules required for the isoprenylation of GTP-binding cell signaling proteins such as Ras, Rho and Rac (5). By inhibiting this posttranslational modification, statins affect intracellular signaling pathways and cellular functions such as differentiation, motility, secretion and proliferation (7,10). Inhibiting Ras leads to a bias towards T helper type 2 (Th-2) cell development, and inhibition of pro-inflammatory Th-1 driven responses (7,11). This change in intracellular signaling can also affect the development of regulatory T (Treg) cells, as exposure of human CD4+ T cells to atorvastatin *in vitro* increases expression of forkhead box P3 (FOXP3) (12). In fact increased numbers of circulating FOXP3+ Treg cells are present in dyslipidemic patients

taking statins for 4-6 weeks (12). Mira et al. demonstrated statin mediated increases in the migration of Treg cells to inflammatory foci in skin by increased the expression of CCL1, the ligand for CCR8, a chemokine receptor that is highly expressed on Th-2 cells and FOXP3+ Treg cells (13). In addition to direct effects on T cells, statins can also indirectly decrease T-cell activation by inhibiting interferon-γ–induced expression of major histocompatibility complex class (MHC) II and by blocking up-regulation of a variety of co-stimulatory molecules and cytokines in antigen presenting cells (APCs) (7). Hence statins appear to suppress T cell–dependent immune responses at many levels and hold promise as a new class of immunomodulatory drugs, with potential role in preventing acute GVHD.

1.4. Pathophysiology of GVHD:

Acute GVHD is initiated when donor T cells encounter host APCs in lymphoid tissues and become activated (14). This T-cell activation process requires co-stimulation via CD80 and CD86, which are up-regulated on APCs during the early phase of acute GVHD. Activation is enhanced by local tissue damage due to the conditioning regimen. Local proinflammatory cytokines such as tumor necrosis factor (TNF), interleukin 1 (IL-1), and interferon (IFN)-gamma, promote Th-1 differentiation of donor-derived T cells and enhance their proliferation and reactivity against host tissues (15). Massive cytokine release related to the Th-1 phenotype predicts the incidence and severity of acute GVHD in both murine models and humans, whereas patients with high IL-10 production are less at risk for GVHD development (16-18).

1.5. Statins and Murine Acute GVHD:

Zeiser and colleagues (19) reported protective effects of atorvastatin against acute GVHD in a MHC mismatched mouse model. In a series of elegant experiments, the authors demonstrated reduced risk of lethal acute GVHD by pre-treating the donor mice with atorvastatin. T-cells derived from these atorvastatin pre-treated mice not only showed reduced proliferation potential, but also displayed (both in-vivo and in-vitro) reduced Th-1 cytokine (TNF, IFN-gamma) and increased Th-2 cytokine (IL-10, IL-4) production. Zeiser and colleagues also investigated the impact of atorvastatin administration to recipient mice. Atorvastatin when given to recipient

mice only translated into acute GVHD protection with a long-term survival of 50%, compared with 0% survival in control animals. Recipient APCs derived from liver, spleen, and lymph nodes of recipient mice getting atorvastatin or PBS were analyzed at different time points after transplantation. Surface expression of CD80, CD86, CD40, and MHC class II was decreased in animals treated with the atorvastatin as compared with PBS-treated recipients. Interestingly pre-treating both donor and recipient mice with atorvastatin produced synergistic protective effects, when compare with survival of only donor or recipient pre-treated groups. Perhaps the most significant finding was that atorvastatin pretreatment did not interfere with perforin-mediated cytolysis or Fas–Fas-ligand interaction–mediated killing. In summary these key experiments highlight the potential of atorvastatin in protecting against acute GVHD, while preserving the graft-versus-leukemia (GVL) effects.

These landmark experiments hold great translational potential for clinical benefit in the field of allogeneic SCT. In humans, elevated IL-10 production by peripheral blood mononuclear cells before allogeneic SCT is associated with a subsequent low incidence of acute GVHD and transplant-related mortality (20,21), whereas low IL-10 promoter activity predicts an increased risk for GVHD (19). Shimada and colleagues have shown Th-2 polarization in adult patients with acute coronary syndrome after treatment with atorvastatin (22). A number of other publications in humans have reported similar findings (23-26)., which highlight the fact that the reduced GVHD reported by Zeiser et al (19) by pre-treating donors with atorvastatin in murine models, can be translated into real clinical benefit in allogeneic transplant recipients. In humans and murine models statins also inhibit the expression of T-bet, a transcription factor critically involved in Th-1 cell differentiation (25,27,28). Lastly the down regulation of co-stimulatory molecules and MHC-II expression in recipient mice demonstrated by Zeiser et al (19), has also been described in humans treated with statin drugs (25,29)

1.6. Statins and Clinical GVHD:

Prompted by these tantalizing data, we therefore asked whether statin use at the time of allogeneic SCT would result in reduced acute GVHD while sparing GVL activity (30). Sixty-seven consecutive patients with acute leukemia underwent T-cell replete allogeneic SCT between June 2002 and October 2006 at The Ohio State University. Patients taking statins

(defined as any HMG-CoA reductase inhibitor) at ≥40mg/day for at least 1month before and 3months after allogeneic SCT (n=10) were compared to those without a history of statin use (n=57). The median age was 43 years (range 20-73yrs). Diagnosis included acute myeloid leukemia (AML) (n=49) and acute lymphoblastic leukemia (ALL) (n=18). The two groups had similar baseline characteristics. Acute GVHD was scored according to modified consensus criteria (31). The rate of grade II-IV acute GVHD was 10% (n=1) in the statin group compared to 40% (n=23) in the no-statin group (p-value=0.08). 56 patients were evaluable for chronic GVHD. No difference in the incidence of chronic GVHD was seen in patients using statins (55%) compared to those in no-statin group (57%) (p-value=0.9). No patient in either group experience primary or secondary engraftment failure. On subgroup analysis of patients with AML only (n=49), a significantly reduced incidence of grade II-IV acute GVHD was seen in statin group (0%) compared to 43% (n=18) in the no-statin group (p-value=0.02). Rates of chronic GVHD were 43% and 58% in similar order (p-value=0.68). We further explored to see if statin use, while reducing acute GVHD, mitigated the GVL effect in patients with AML. Kaplan-Meier estimates of progression free survival (PFS) at 3years in AML patients with or without statin use were 54% and 28% respectively (p-value=0.17) respectively (Figure 1). This non significant trend of improved PFS indicates that the GVL is preserved in patients using statins at the time of allografting.

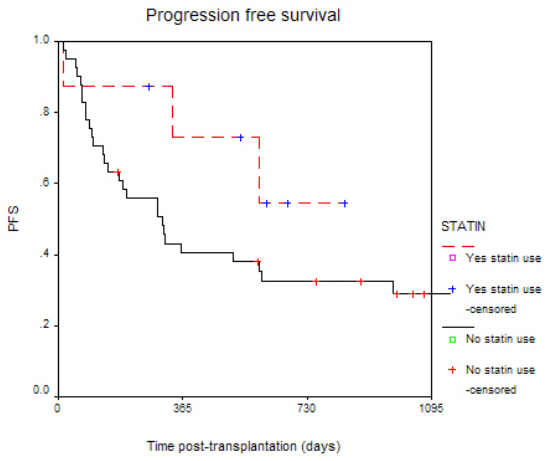


Figure 1. KM estimates of PFS following allogeneic SCT in patients with AML.

Rotta et al. retrospectively analyzed outcomes among 567 patients with hematologic malignancies who had hematopoietic cell transplantation from human leukocyte antigen-identical sibling donors between 2001 and 2007 for a correlation between statin use and risk of GVHD. Compared to allografts where neither the donor nor recipient was treated with a statin at the time of transplantation (n = 464), statin use by the donor was associated with a decreased risk of grade 3-4 acute GVHD (multivariate hazard ratio, 0.28; 95% confidence interval, 0.1-0.9). Statin use by both donor and recipient (n = 12) was suggestively associated with a decreased risk of grade 3 or 4 acute GVHD (multivariate hazard ratio, 0.00; 95% confidence interval, undefined). Risks of chronic GVHD, recurrent malignancy, nonrelapse mortality, and overall mortality were not significantly affected by donor or recipient statin exposure. Statin-associated GVHD protection was restricted to recipients with cyclosporine-based postgrafting immunosuppression and was not observed among those given tacrolimus (P = .009). These results suggest that donor statin treatment may be a promising strategy to prevent severe acute GVHD without compromising immunologic control of the underlying malignancy.

In summary these limited but encouraging retrospective studies, and the extensive pre-clinical and clinical data suggest that the observations made by Zeiser and colleagues (19) in a murine model may have clinical relevance and hints at the potential of statins in reducing acute GVHD.

1.7. Clinical Trial Data for Statin use for preventing acute GVHD:

Based on these data our group, opened a single-arm phase-II study evaluating the safety and efficacy of atorvastatin for the prophylaxis of acute graft-versus-host disease in patients with hematological malignancies undergoing HLA-matched sibling donor hematopoietic SCT (WVU protocol #11010; study registered www.clinicaltrials.gov NCT01175148). To date (September 26, 2013) 30 donor/patient pairs have been enrolled on this protocol WVU 11010. All healthy donors have tolerated atorvastatin with no significant side effects. No statin related grade 3-5 toxicity in donors was seen. Similarly all patients so far have tolerated atorvastatin well, with no study drug related grade 3-5 toxicity. No liver function abnormalities or cases of myopathy were seen. In terms of the primary outcome of WVU11010 (i.e. rates of grade II-IV acute GVHD at day +100 post transplantation), only one patient experienced grade II acute GVHD before day 100. In the current protocol, we will evaluate the role of atorvastatin in addition to methotrexate

and tacrolimus for the prophylaxis of acute GVHD when administered to transplant recipient alone, to assess if routine administration of atorvastatin to healthy sibling donors is warranted. This protocol will also include patients undergoing unrelated donor transplantation to see if atorvastatin can maintain GVHD rates, seen in transplants using ATG. While some retrospective studies (31), but not all (30), suggest that statin-mediated benefit for preventing acute GVHD is limited to patients receiving cyclosporine-based GVHD prophylaxis, in our study use of cyclosporine prophylaxis is not permitted for two reasons: 1) Our prior study has shown excellent safety and efficacy profile of atorvastatin/tacrolimus/methotrexate combination [Hamadani M et al. Journal of Clinical Oncology. Accepted manuscript in press], and 2) strong drug interaction between cyclosporine and atorvastatin (please see section 8.1.5).

1.8. Inclusion of Women and Minorities:

It is the policy of the Mary Babb Randolph Cancer Center to strive for gender and minority patient participation that represents the population of West Virginia in all clinical investigations. Between January 1st 2009 and December 31st 2009, 125 patients were enrolled onto Phase I/II/III trials at the Mary Babb Randolph Cancer Center. Of these patients 73% percent were female (n=92) and 2.4 percent were members of minority ethnic groups. It is anticipated that a similar or greater proportion of patients on this study will be female and/or members of ethnic minorities.

2. Objectives:

2.1. Primary Objectives:

1. Determine the efficacy of an atorvastatin/tacrolimus/methotrexate regimen in preventing grade II-IV acute GVHD in patients undergoing allogeneic hematopoietic stem cell transplantation (HSCT).
2. Assess the safety of an atorvastatin/tacrolimus/methotrexate regimen in patients undergoing allogeneic HSCT.

2.2. Secondary Objectives:

1. To assess rates of chronic GVHD.
2. To assess rates of late onset acute GVHD at days +180 and +365.
3. To assess rates of grade III-IV acute GVHD at days +100, +180 and +365.
4. To assess non-relapse mortality at 100 days post-HSCT.
5. To assess time to successful neutrophil engraftment.
6. To assess time to successful platelet engraftment.
7. To assess incidence of primary and secondary graft failure.
8. To assess incidence of primary and secondary graft rejection.
9. To assess relapse rate of the primary hematological malignancy.
10. To assess lineage specific chimerism kinetics in patients at days +28, +100, +180 and +365.
11. To assess immune reconstitution following transplantation at days +28, +100, +180, and +365.
12. To assess 1 year progression free survival (PFS) and overall survival (OS) following transplantation.
13. To evaluate biologic and genomic markers potentially associated with GVHD and/or atorvastatin.

3. Eligibility Criteria:

3.1. Patient Eligibility Criteria:

1. Patients with a history of a hematological malignancy or bone marrow failure syndrome suitable for allogeneic stem cell transplantation in the opinion of treating transplant physician.
2. Patients aged 18-75 years of age are eligible. Patients with age $\geq$ 18 and $\leq$ 50 years will be eligible for myeloablative conditioning (MAC), while patients > 50 years of age, or those with previous history of autologous transplantation, high hematopoietic cell transplant comorbidity index (HCT-CI) score (>2), and baseline diagnosis of hodgkin's

lymphoma, chronic lymphocytic leukemia and follicular lymphoma will be suitable for reduced intensity conditioning (RIC) transplantation (however intensity of conditioning regimen will remain at the discretion of treating physician).

3. All patients must have at least one suitable HLA-matched sibling or unrelated donor according to transplant center's guidelines (for selection of appropriate donor).
4. Patient must provide informed consent.
5. Left ventricular ejection fraction ≥ 40%. No uncontrolled arrhythmias or uncontrolled New York Heart Association class III-IV heart failure.
6. Bilirubin ≤ 2 x the ULN and AST, and ALT ≤ 3 x ULN; and absence of hepatic cirrhosis. For patients with Gilbert's syndrome, bilirubin ≤ 3 x ULN is permitted.
7. Adequate renal function as defined by a serum creatinine clearance of ≥ 40% of normal calculated by Cockcroft-Gault equation.
8. DLCOcor (corrected for hemoglobin) or FEV1 or DL/VA ≥ 40% of predicted.
9. Karnofsky performance status ≥ 70.
10. A negative pregnancy test will be required for all women of child bearing potential. Breast feeding is not permitted.
11. Patients with positive HIV serology **are** eligible.
12. No evidence of active bacterial, viral or fungal infection at the time of transplant conditioning.
13. Patients with history of intolerance or allergic reactions with atorvastatin will not be eligible.
14. Patients who have previously been taking atorvastatin or any other statin drug will be eligible as long as there is no contraindication to switch to atorvastatin (40mg/day) in the opinion of the treating physician.
15. Patients undergoing a T-cell depleted allogeneic transplantation will not be eligible.
16. Patients receiving conditioning regimens containing antithymocyte globulin, and/or campath will not be eligible.
17. Method of stem-cell collection from the donor will be at the discretion of the treating physician. Although it is anticipated that majority of donors will undergo G-CSF induced stem cell mobilization; however donors undergoing bone marrow harvest or

stem cell mobilization with experimental agents (e.g. plerixafor) will remain eligible for the study.

4. Registration Procedure:

4.1. Registration

- All source documents that support eligibility including a signed informed consent/HIPAA and signed eligibility checklist, should be available review and eligibility verification.
- At the point of registration, the research nurse or data manager will register the patient in the electronic database, including demographic, consent and on-study information. The patient will be assigned a unique sequence number for the study.

4.1.1. **Registration for collaborating institution(s):**

- The collaborating institution (e.g. WVU) must contact Study Coordinator at MCW prior to registering a patient. After speaking with the Study Coordinator, the following documents should be faxed to 414-805-9025 to begin the registration process:
    - The dated and signed informed consent
    - A physician signed eligibility checklist
    - All source documents that validate eligibility
    - The demographic form
- Written confirmation (via email to study coordinator at WVU) will be sent to the site once the patient has been enrolled into the study to issue the unique patient identifier and cohort as applicable.

5. Treatment Plan:

This is a phase II study of atorvastatin for the prophylaxis of acute GVHD in patients undergoing allogeneic HSCT. The study will have two separate arms: (a): Matched sibling transplants, and (b): Matched unrelated donors transplants. Both arms will be enrolled and analyzed separately.

5.1. Administration schedule to Patients (Transplant recipients)

1. Atorvastatin will be administered at dose of 40mg orally daily starting on day -14, to permit an approximately 1-week observation period to rule out any acute atorvastatin-induced side effects before the initiation of transplant conditioning.
2. Patients who have previously been taking atorvastatin or any other statin drug will be eligible as long as there is no contraindication to switch to atorvastatin (40mg/day) in the opinion of the treating physician.
3. Patients undergoing myeloablative conditioning (MAC) matched sibling or unrelated donor allogeneic transplantation can receive one of following three conditioning regimens:
    a) Busulfan ≥0.8 mg/kg/dose (16 doses given every 6 hours, IV on days –7 to –4), and cyclophosphamide 60 mg/kg/dose (2 doses given every 24hours, IV on days –3 and –2). Monitoring busulfan levels with this regimen, per institutional practice, is recommended. Oral busulfan is not permitted.
    b) Total body irradiation (>500cGy) and cyclophosphamide as above.
    c) Total body irradiation (≥1200cGy) and etoposide.
    d) Other ablative regimens will be permitted after consultation with study PI.
4. Patients undergoing reduced-intensity conditioning (RIC) matched sibling or unrelated donor allogeneic transplantation will receive one of following three regimens:
    a) Fludarabine and IV busulfan-based regimen (Flu/Bu2 or Flu/Bu4).
    b) Fludarabine and low dose total body irradiation (200-400cGy)
    c) Fludarabine and melphalan (≤140mg/m^2)
    d) Other RIC will be permitted after consultation with study PI.

In addition to atorvastatin, standard GVHD prophylaxis will consist of:**T**acrolimus (0.015 mg/kg I.V **every 12 hours** or 0.03 mg/kg/day PO; starting day -2) and mini-dose methotrexate (5mg/m^2 on days +1, +3, +6 and +11). Tacrolimus will be monitored to keep levels between 5-12 ng/ml for matched sibling and between 8-12 ng/ml for unrelated donor HCT. In absence of acute GVHD, renal insufficiency and disease relapse/progression it is recommended that tacrolimus taper **SHOULD NOT** commence before day +100. The goal of tapering is the complete discontinuation of immunosuppressive medications by (but not before) day +180. Tacrolimus should be tapered every two to three weeks. Attempt should be

made to apply similar reductions in dosages at each tapering event, to allow complete discontinuation of immunosuppression by day +180. It is recommended by not required that tacrolimus dose be reduced by no more than 25% at each tapering event.

5. Administration of cyclosporine (because of associated risk of myopathy with atorvastatin), mycophenolate mofetil, monoclonal (e.g. campath) or polyclonal antibodies (e.g. antithymocyte globulin) will not be permitted for GVHD prophylaxis.
6. Antibiotic prophylaxis will be given according to institutional guidelines.
7. Temporary interruption (≤10-14days) of atorvastatin is permitted in patients unable to tolerate oral intake secondary to severe nausea, vomiting, mucositis etc. Administration of atorvastatin via feeding tubes is permitted. In case atorvastatin therapy is interrupted, the time interval should be recorded.
8. Toxicity will be evaluated according to CTCAE v4.0.
9. Progression free survival (PFS) and overall survival (OS) following transplantation will be recorded.

5.2. Dose modification

1. Patients experiencing a grade 3-4 hematological or non-hematological toxicity (as specified in CTCAE v4.0) thought to be related to atorvastatin will be removed from the study permanently. No dose modifications for atorvastatin are permitted.
2. Decision to hold atorvastatin in patients developing grade 3-4 liver function abnormalities (bilirubin, AST and ALT) post-HSCT will be at the discretion of treating physician. In such patients if liver function abnormalities are thought not be related to the study drug, then atorvastatin prophylaxis can be restarted at time of resolution of hepatic transaminases (to grade 1 or less) after consulting with principal investigator.

5.3. Duration of therapy

Atorvastatin will be continued until one of the following criteria is met:

- Patient is no longer on immunosuppressive medications or until day +180 after transplantation, whichever occurs first.
- Patient develops grade II-IV acute GVHD.
- Patient develops severe chronic GVHD.

- Patient develops any grade 3-4 toxicity related to atorvastatin use.

Patients experiencing a disease relapse (before day +180) can continue atorvastatin at the discretion of treating physician, provided the patient has not been tapered off of tacrolimus.

5.4. Data Safety Monitoring Plan

At West Virginia University: This protocol will adhere to the policies of the Mary Babb Randolph Cancer Center Data and Safety Monitoring Plan, guidelines in accordance with NCI regulations. The Data and Safety Toxicity Committee (DSTC) will review all serious adverse events and toxicity reports as well as continuing reviews. This study is designated as a **moderate risk level** and requires **quarterly** summary reporting to the DSTC.

At Medical College of Wisconsin: The Medical College of Wisconsin Cancer Center Data Safety Monitoring Committee (DSMC) is responsible for monitoring data quality and subject safety for all cancer center investigator initiated clinical trials. A six to eight member Data and Safety Monitoring Committee will complete a review of protocol-specific data safety monitoring reports, to provide recommendations on trial continuation, suspension or termination. The DSMC will review these reports no less than bi-annually. A summary of the DSMC activities are as follows

- Review the clinical trials for data integrity and safety
- Review all adverse events requiring expedited reporting as defined per protocol
- Review all DSM reports
- Submit a summary of any recommendations related to study conduct
- Terminate the study if deemed unsafe for patients

6. Measurement of Effect:

6.1. Visit Schedule:

The schedule for the study is shown in the table below.

Study Visit	Target Day Post-Transplant
1 week	7 ± 3 days
2 week	14 ± 3 days

3 week	21 ± 7 days
4 week	28 ± 7 days
5 week	35 ± 7 days
6 week	42 ± 7 days
7 week	49 ± 7 days
8 week	56 ± 7 days
9 week	63 ± 7 days
10 week	70 ± 7 days
11 week	77 ± 7 days
12 week	84 ± 7 days
100 day	100± 7 days
6 month	180 ± 28 days
12 month*	365 ± 28 days

*After 12months, patients will be followed at least yearly for survival, relapse and cause of death for 5 years post transplantation.

6.2. GVHD Assessment:

Following neutrophil engraftment, patients will be monitored for development of acute and chronic GVHD at once a week until day +100. After Day 100, patients will be assessed at each study visit for the presence of GVHD. Diagnosis of acute GVHD will require biopsy confirmation in at least one involved organ. When more than one organ is involved, biopsy confirmation of all involved organs is recommended but not necessary. Liver-only GVHD must be confirmed by biopsy. Acute GVHD will be assessed by consensus criteria (Appendix A) (32) and graded on BMT CTN MOP suggested grading sheets (Appendix A). Acute GVHD assessment and grading will be performed by **treating physicians.** Chronic GVHD diagnosis and grading will be according to NIH Criteria. Please see Appendix B (33-34).

Clinical Grading of Chronic GVHD (According to Appendix B)

None

Mild chronic GVHD involves only 1 or 2 organs or sites (except the lung: see below ‡), with no clinically significant functional impairment (maximum of score 1 in all affected organs or sites)

Moderate chronic GVHD involves: (1) at least 1 organ or site with clinically significant but no major disability (maximum score of 2 in any affected organ or site) or (2) 3 or more organs or sites with no clinically significant functional impairment (maximum score of 1 in all affected organs or sites). ‡A lung score of 1 will also be considered moderate chronic GVHD.

Severe chronic GVHD indicates major disability caused by chronic GVHD (score of 3 in any organ or site). ‡A lung score of 2 or greater will also be considered severe chronic GVHD.

6.3. Engraftment Assessment:

Neutrophil engraftment will be defined as first of three consecutive days with ANC ≥ 500 x 10^9/L post-conditioning regimen induced nadir. Similarly platelet engraftment is defined as first day of platelet count ≥ 20,000 x 10^9/L, without transfusion for 7 consecutive days. Lineage specific chimerism analysis (CD3 and CD33 subsets), quantitative immunoglobulins and immune reconstitution will be performed on days +28, +100, +180 and +365.

7. Study Parameters (Study Calendar):

7.1. Patient Study Calendar:

The table below summarizes the patient clinical assessments over the course of the study.

Study Assessment	Baseline[1]	Day-14	Day0	Days Post-Transplantation													
				7	14	21	28	35	42	49	56	63	70	77	84	100	180	365[15]
Informed consent and H & PE[2]	X						X					X				X	X	X
Vital signs[3]	X	X	X				X					X				X	X	X
Karnofsky performance status	X						X					X				X	X	X
CBC/differential & platelet count	X		X	X[4]	X[4]	X[4]	X[4]					X				X	X	X
Serum chemistries panel[5]	X		X	X	X	X	X					X				X	X	X
Infectious disease titer[6]	X																	
LVEF[7]	X																	
DLCO	X																	
CMV quantitative PCR						X	X	X	X	X	X	X	X	X	X	X		
Bone marrow aspirate/biopsy[8]	X															X	X	X
B-HCG serum pregnancy test [9]	X																	
Quantitative immunoglobulins[10]							X									X	X	X
Peripheral blood for chimerism	X[13]						X									X	X	X
Immune reconstitution panel[11]							X									X	X	X
CD34/CD3 cell dose infused			X															
Acute/chronic GVHD assessment				X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Toxicity assessment			X	X	X	X	X	X	X	X	X	X	X	X	X	X	X	X
Research specimens (voluntary)[12]	X		X		X		X					X				X	X	X
ANC and platelet recovery[14]																		

Notes

H & PE= history and physical examination

[1]Baseline refers to the period prior to conditioning. Assessments should be made within 4 weeks prior to day of conditioning.

[2]History is only required at baseline.

[3]Vital signs: blood pressure, pulse rate, respiratory rate and temperature.

[4]Daily during the post-infusion period while hospitalized (recommended) and once weekly after discharge until Day 28.

[5]Serum chemistries panel: electrolytes, BUN, ALT, AST, creatinine, bilirubin, alkaline phosphatase, LDH, albumin. Electrolytes to include sodium, potassium, chloride, carbon dioxide, and calcium.

[6]Infectious disease titers: Cytomegalovirus (CMV) antibody test, hepatitis panel (Hepatitis B including HBsAg, HBcAb; Hepatitis C Ab), HIV including HIV testing, syphilis, Epstein-Barr Virus (EBV) IgG and IgM, herpes simplex virus (HSV) IgG and IgM

[7]To be determined by MUGA or echocardiogram.

[8]Required at baseline. On Days +100, +180 and +365, bone marrow biopsy will be performed if clinically indicated. Other tests and/or imaging studies for response assessment will be performed as clinically indicated.

[9]Females of reproductive potential only.

[10]IgG, IgM and IgA.

[11]Peripheral blood flow cytometry to assess recipient immune reconstitution.

[12]Peripheral blood specimens: draw 15 mL in purple top (EDTA containing) tubes AND 5 mL in blue top (citrate containing) tubes at indicated time points and send to the Biospecimen Processing Core at WVU. At MCW voluntary research specimens will not be collected.

[13]Chimersim testing to be performed prior to study from BOTH donor and recipient at any time point. Chimerism testing should be sorted (lineage specific) to assess myeloid and lymphoid cell chimerism.

[14]Record time to neutrophil engraftment defined as first of three consecutive days with ANC $\geq 500 \times 10^9$/L, and platelet engraftment defined as first day of platelet count $\geq 20{,}000 \times 10^9$/L, without transfusion for 7 consecutive days.

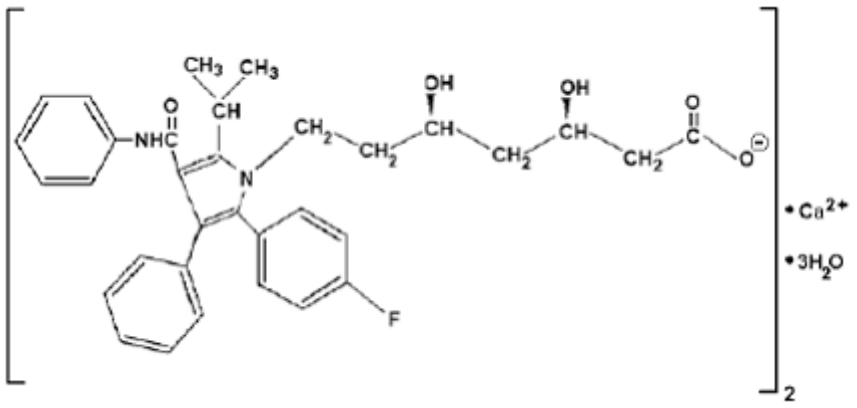
[15]After day 365, patients will be followed at least yearly for survival, relapse and cause of death for a total of 5 years post transplantation.

8. Drug Formulation and Procurement

8.1. Atorvastatin (Lipitor® Pfizer)

8.1.1. Drug description

LIPITOR is a synthetic lipid-lowering agent. Atorvastatin is an inhibitor of 3-hydroxy-3-methylglutaryl-coenzyme A (HMG-CoA) reductase. This enzyme catalyzes the conversion of HMG-CoA to mevalonate, an early and rate-limiting step in cholesterol biosynthesis. Atorvastatin calcium is [R-(R*, R*)]-2-(4-fluorophenyl)-β, δ-dihydroxy-5-(1-methylethyl)-3-phenyl-4-[(phenylamino)carbonyl]-1H-pyrrole-1-heptanoic acid, calcium salt (2:1) trihydrate. The empirical formula of atorvastatin calcium is (C33H34FN2O5)2Ca•3H2O and its molecular weight is 1209.42. Its structural formula is: Atorvastatin calcium is a white to off-white crystalline powder that is insoluble in aqueous solutions of pH 4 and below. Atorvastatin calcium is very slightly soluble in distilled water, pH 7.4 phosphate buffer, and acetonitrile; slightly soluble in ethanol; and freely soluble in methanol. LIPITOR Tablets for oral administration contain 10, 20, 40, or 80 mg atorvastatin and the following inactive ingredients: calcium carbonate, USP; candelilla wax, FCC; croscarmellose sodium, NF; hydroxypropyl cellulose, cellulose, NF; Opadry White YS-1-7040 (hypromellose, polyethylene glycol, talc, titanium dioxide); polysorbate 80, NF; simethicone emulsion.



8.1.2. Pharmacodynamics

LIPITOR, as well as some of its metabolites, are pharmacologically active in humans. The liver is the primary site of action and the principal site of cholesterol synthesis and

LDL clearance. Drug dosage, rather than systemic drug concentration, correlates better with LDL-C reduction.

8.1.3. Pharmacokinetics

Absorption: LIPITOR is rapidly absorbed after oral administration; maximum plasma concentrations occur within 1 to 2 hours. Extent of absorption increases in proportion to LIPITOR dose. The absolute bioavailability of atorvastatin (parent drug) is approximately 14% and the systemic availability of HMG-CoA reductase inhibitory activity is approximately 30%. The low systemic availability is attributed to presystemic clearance in gastrointestinal mucosa and/or hepatic first-pass metabolism. Although food decreases the rate and extent of drug absorption by approximately 25% and 9%, respectively, as assessed by Cmax and AUC, LDL-C reduction is similar whether LIPITOR is given with or without food.

8.1.4. Contraindications

Active Liver Disease which may include Unexplained Persistent Elevations of Hepatic Transaminase Levels

Hypersensitivity to any Component of this Medication

Pregnancy

Nursing Mothers

8.1.5. Drug Interactions

Drug Interactions Associated with Increased Risk of Myopathy/Rhabdomyolysis include the following.

Interacting Agents	Prescribing Recommendations
Cyclosporine	Atorvastatin is a substrate of the OATP1B1 transporter. Inhibitors of the OATP1B1 (e.g., cyclosporine) can significantly increase the bioavailability of atorvastatin. Concomitant administration of atorvastatin and cyclosporine is not permitted on the protocol.
Clarithromycin, itraconazole, HIV protease inhibitors	Caution when exceeding doses

(ritonavir plus saquinavir or lopinavir plus ritonavir)	> 20mg atorvastatin daily. The lowest dose necessary should be used.
Atorvastatin is metabolized by cytochrome P450 3A4. Concomitant administration of atorvastatin with inhibitors of cytochrome P450 3A4 can potentially lead to increased plasma atorvastatin concentrations.	Use of strong inhibitors of cytochrome P450 3A4 (erythromycin, clarithromycin, protease inhibitors, diltiazem, or itraconazole), with atorvastatin should be done with caution.
Grapefruit juice	: Contains one or more components that inhibit CYP 3A4 and can increase plasma concentrations of atorvastatin, especially with excessive grapefruit juice consumption (>1.2 liters per day).
Gemfibrozil	Due to an increased risk of myopathy/rhabdomyolysis when HMG-CoA reductase inhibitors are co-administered with gemfibrozil, concomitant administration of LIPITOR with gemfibrozil should be avoided
Digoxin	Digoxin concentrations are unaffected by atorvastatin doses of <80mg/day
Rifampin or other Inducers of Cytochrome P450 3A4:	Concomitant administration of LIPITOR with inducers of cytochrome P450 3A4 (e.g., efavirenz, rifampin) can lead to variable reductions in plasma concentrations of atorvastatin. Due to the dual interaction mechanism of rifampin, simultaneous co-administration of LIPITOR with rifampin is recommended, as delayed administration of LIPITOR after administration of rifampin has been associated with a significant reduction in atorvastatin plasma concentrations.

8.1.6. **Adverse Events:**

Skeletal Muscle

Rare cases of rhabdomyolysis with acute renal failure secondary to myoglobinuria have been reported with LIPITOR and with other drugs in this class. A history of renal impairment may be a risk factor for the development of rhabdomyolysis. Such patients merit closer monitoring for skeletal muscle effects. Atorvastatin, like other statins, occasionally causes myopathy, defined as muscle aches or muscle weakness in conjunction with increases in creatine phosphokinase (CPK) values >10 times ULN. The concomitant use of higher doses of atorvastatin with certain drugs such as cyclosporine and strong CYP3A4 inhibitors (e.g., clarithromycin, itraconazole, and HIV protease inhibitors) increases the risk of myopathy/rhabdomyolysis. Myopathy should be considered in any patient with diffuse myalgias, muscle tenderness or weakness, and/or marked elevation of CPK. Patients should be advised to report promptly unexplained muscle pain, tenderness, or weakness, particularly if accompanied by malaise or fever. LIPITOR therapy should be discontinued if markedly elevated CPK levels occur or myopathy is diagnosed or suspected.

Liver Dysfunction

Statins, like some other lipid-lowering therapies, have been associated with biochemical abnormalities of liver function. Persistent elevations (>3 times the upper limit of normal [ULN] occurring on 2 or more occasions) in serum transaminases occurred in 0.7% of patients who received LIPITOR in clinical trials. The incidence of these abnormalities was 0.2%, 0.2%, 0.6%, and 2.3% for 10, 20, 40, and 80 mg, respectively

Endocrine Function

Statins interfere with cholesterol synthesis and theoretically might blunt adrenal and/or gonadal steroid production. Clinical studies have shown that LIPITOR does not reduce basal plasma cortisol concentration or impair adrenal reserve. The effects of statins on male fertility have not been studied in adequate numbers of patients. The effects, if any, on the pituitary-gonadal axis in premenopausal women are unknown. Caution should be exercised if a statin is administered concomitantly with drugs that may decrease the levels or activity of endogenous steroid hormones, such as ketoconazole, spironolactone, and cimetidine.

CNS Toxicity

Brain hemorrhage was seen in a female dog treated for 3 months at 120 mg/kg/day. Brain hemorrhage and optic nerve vacuolation were seen in another female dog that was sacrificed in moribund condition after 11 weeks of escalating doses up to 280 mg/kg/day. The 120 mg/kg dose resulted in a systemic exposure approximately 16 times the human plasma area-under-the-curve (AUC, 0-24 hours) based on the maximum human dose of 80 mg/day. A single tonic convulsion was seen in each of 2 male dogs (one treated at 10 mg/kg/day and one at 120 mg/kg/day) in a 2-year study. No CNS lesions have been observed in mice after chronic treatment for up to 2 years at doses up to 400 mg/kg/day or in rats at doses up to 100 mg/kg/day. These doses were 6 to 11 times (mouse) and 8 to 16 times (rat) the human AUC (0-24) based on the maximum recommended human dose of 80 mg/day.

Other adverse reactions reported in placebo-controlled studies include: *Body as a whole*: malaise, pyrexia; *Digestive system:* abdominal discomfort, eructation, flatulence, hepatitis, cholestasis; *Musculoskeletal system*: musculoskeletal pain, muscle fatigue, neck pain, joint swelling; *Metabolic and nutritional system*: transaminases increase, liver function test abnormal, blood alkaline phosphatase increase, creatine phosphokinase increase, hyperglycemia; *Nervous system*: nightmare; *Respiratory system:* epistaxis; *Skin and appendages*: urticaria; *Special senses*: vision blurred, tinnitus; *Urogenital system:* white blood cells urine positive.

8.2. How supplied and Storage/Handling

40 mg tablets: coded “PD 157” on one side and “40” on the other.

NDC 0071-0157-23 bottles of 90

NDC 0071-0157-73 bottles of 500

NDC 0071-0157-88 bottles of 2500

NDC 0071-0157-40 10 x 10 unit dose blisters

Stored at controlled room temperature 20 - 25°C (68 - 77°F).

8.3. Methods of procurement

The Medical College of Wisconsin & West Virginia University Hospitals Departments of Pharmacy will order Atorvastatin 40mg and receive drug specifically for this study. The CTRU (at WVU) and CTO (at MCW) will be billed for the cost of the drug which will be paid for by study funds. The drug will be dispensed per patient and labeled according to more stringent state or federal law. Drug accountability and inventory records will be created and maintained within the Investigational Drug Services.

9. Statistical Considerations:

The study is a single-arm phase II trial evaluating the safety and efficacy of atorvastatin for the prophylaxis of acute GVHD in patients with hematological malignancies undergoing HLA-matched HSCT. The primary objective of this study is to evaluate the cumulative incidence of grade II-IV acute GVHD by day +100 in patients receiving acute GVHD prophylaxis with atorvastatin, tacrolimus and methotrexate. The incidence of grade II-IV acute GVHD in patients undergoing MAC or RIC matched sibling allogeneic SCT, and receiving GVHD prophylaxis with tacrolimus and methotrexate is approximately 30-40%. The respective rates for patients undergoing RIC unrelated donor transplantation with ATG are 45-55% and without ATG 50-60%. The sibling and unrelated donor arms of this study will be analyzed separately, with aim for separate publications. At the time of publication the results of unrelated donor arm of the study will be retrospectively compared against patients undergoing unrelated transplantation at our center after conditioning with Flu/Bu2 or Flu/Bu4 and receiving GVHD prophylaxis with tacrolimus, MTX and ATG.

For this phase II study, we will use an open-label, minimax two-stage design as specified by Simon. We will consider the “experimental” acute GVHD prophylactic regimen of atorvastatin/tacrolimus/methotrexate to be no more effective than GVHD prophylaxis with tacrolimus/methotrexate alone for matched sibling allogeneic HCT, if the true probability of developing grade II-IV acute GVHD is more than 35% (p0). We will also assume that the new prophylactic regimen is worthy of further study if the true probability of grade II-IV

acute GVHD is equal or less than 15% (p1). In statistical terms, we are testing the null hypothesis H_0: $p \geq 0.35$, versus the alternate H_1: $p \leq 0.15$, where p is probability of grade II_IV acute GVHD. These figures result in a two-stage design of 25 and 30 patients during stage 1 and 2 respectively, with an alpha of 0.05 and beta of 0.20. In the first stage, we will enter 25 patients undergoing matched sibling transplantation. If 6 or more of these patients develop grade II-IV acute GVHD, we will stop and conclude in favor of H_0: $p \geq 0.35$. If only 5 or less of the first 25 patients develop grade II-IV acute GVHD, we will enter an additional 5 patients, for a total of 30 patients. In the final analysis, if 7 or fewer of the 30 patients develop grade II-IV acute GVHD we will reject null hypothesis.

For the unrelated donor HCT arm of this study we will consider the "experimental" acute GVHD prophylactic regimen of atorvastatin/tacrolimus/methotrexate to be no more effective than GVHD prophylaxis with tacrolimus/methotrexate alone, if the true probability of developing grade II-IV acute GVHD is more than 55% (p0). We will also assume that the new prophylactic regimen is worthy of further study if the true probability of grade II-IV acute GVHD is equal or less than 40% (p1). In statistical terms, we are testing the null hypothesis H0: $p \geq 0.55$, versus the alternate H1: $p \leq 0.35$, where p is probability of grade II_IV acute GVHD. These figures result in a two-stage design of 30 and 39 patients during stage 1 and 2 respectively, with an alpha of 0.05 and beta of 0.20. In the first stage, we will enter 30 patients undergoing unrelated donor transplantation. If 14 or more of these patients develop grade II-IV acute GVHD, we will stop and conclude in favor of H0: $p \geq 0.55$. If only 13 or less of the first 30 patients develop grade II-IV acute GVHD, we will enter an additional 9 patients, for a total of 39 patients. In the final analysis, if 17 or fewer of the 39 patients develop grade II-IV acute GVHD we will reject null hypothesis.
We anticipate that a maximum of 30 each, matched sibling and unrelated donor patients will be accrued to the study provided 10 or less patients develop grade II-IV acute GVHD during the first stage of the study. Descriptive statistics (i.e. means, standard deviations, 95% confidence intervals for continuous variables, and frequencies for discrete data) will be computed for all correlative laboratory parameters.

Safety/Stopping Rules:

1. Development of grade 3-4 adverse events deemed to be related to atorvastatin in 6 or more transplant recipients, will mandate halting further patient accrual until review by DSMB.
2. Cumulative incidence of relapse of >50% (with NRM as competing event) observed after enrolling at least 20 patients in each cohort, will mandate halting further patient accrual until review by DSMB.
3. In patients complaining of grade II or worse muscle pain, weakness or myopathy, a serum CPK level will be checked to rule out development of rhabdomyolysis.

Accrual Estimate: 55-69 patients. Patients experiencing mortality within 100 days of transplant due to disease relapse/progression, will be replaced to fully assess late onset acute GVHD rates. However, all patients will be included in toxicity and survival analysis.
Accrual Period : Approximately 24-36 months

Follow-Up Period : One year to evaluate incidence of acute and chronic GVHD and relapse pattern. Five years for survival outcomes, relapse, and mortality.

10. Adverse Event Reporting Requirements

Definitions

The following are definitions of adverse events as defined by 21CFR312.32.

Types of Adverse Events

Adverse Event means any untoward medical occurrence associated with the use of a drug in humans, whether or not consider drug related.

Life-threatening adverse event or life-threatening suspected adverse reaction: An adverse event or suspected adverse reaction is considered "life-threatening" if, in the view of either the investigator or sponsor, its occurrence places the patient or subject at immediate risk of death. It does not include an adverse event or suspected adverse reaction that, had it occurred in a more severe form, might have caused death.

Serious adverse event or serious suspected adverse reaction: An adverse event or suspected adverse reaction is considered "serious" if, in the view of the investigator or sponsor, it results in any of the following outcomes:

- Death,
- A life-threatening prolongation of existing hospitalization,
- A persistent or significant incapacity or substantial disruption of the ability to conduct normal life functions, or
- A congenital anomaly/birth defect.
- Important medical events that may not result in death, be life-threatening, or require hospitalization may be considered serious when, based upon appropriate medical judgment, they may jeopardize the patient or subject and may require medical or surgical intervention to prevent of the outcomes listed above.

Suspected adverse reaction means any adverse event for which there is a reasonable possibility that the drug caused the adverse event. "Reasonable possibility" means there is evidence to suggest a causal relationship between the drug and the adverse event. Suspected adverse reaction implies a lesser degree of certainty about causality than adverse reaction, which means any adverse event caused by a drug.

Unexpected adverse event or unexpected suspected adverse reaction: An adverse event or suspected adverse reaction is considered "unexpected" if it is not listed in the investigator brochure or is not listed at the specificity or severity that has been observed; or, if an investigator brochure is not required or available, it is not consistent with the risk information currently described.

Unexpected also refers to adverse events or suspected adverse reactions that are mentioned in the investigator brochure as occurring with a class of drugs or as anticipated from the pharmacological properties of the drug, but are not specifically mentioned as occurring with the particular drug under investigation.

Adverse Event Grading

Grade	Description
0	**No AE** (or within normal limits).
1	**Mild**; asymptomatic or mild symptoms; clinical or diagnostic observations only; intervention not indicated.
2	**Moderate**; minimal, local or noninvasive intervention (e.g., packing cautery) indicated; limiting age-appropriate instrumental activities of daily living (ADL).
3	**Severe** or medically significant but not immediately life-threatening; hospitalization or prolongation of hospitalization indicated; disabling; limiting self-care ADL.
4	**Life-threatening** consequences; urgent intervention indicated.
5	**Death** related to AE

Adverse Event Attribution

Relationship	Attribution	Description
Unrelated to investigational agent/intervention	Unrelated	The AE *is clearly **NOT** related* to the intervention
	Unlikely	The AE *is **doubtfully related*** to the intervention
Related to investigationl agent/intervention	Possible	The AE ***may be related*** to the intervention
	Probable	The AE ***is likely related*** to the intervention
	Definite	The AE ***is clearly related*** to the intervention

Reporting of Adverse Events

Because all of the study transplant recipients will be receiving potentially toxic preparative therapy, significant regimen related toxicity is expected. These risks are listed in the consent form. A study specific toxicity CRF will be designed to capture information regarding these expected events. Transplant is also related to a degree of mortality and this will also be captured by a study designed CRF. Unexpected adverse events will be reported throughout the study.

The sub-site (i.e. West Virginia University) must report all serious adverse events to the sponsor-investigator (*SAE FAX-414-805-9025 or Froedtert Hospital and the Medical College of Wisconsin Cancer Center Clinical Trials Office 9200 W. Wisconsin Ave. Milwaukee, WI*) so that the sponsor-investigator can meet his/her foregoing reporting obligations to the required regulatory agencies, unless otherwise agreed between the sponsor-investigator and sub-investigator(s).

Unexpected Adverse Events

Grades 3-5 (severe, life threatening, disabling, or fatal) require expedited reporting and will be submitted to the DSTC within 24 hours of discovery for review. If the Grade 3-5 event is determined to be an unanticipated problem, the event will be forwarded to the WVU IRB for review as required by their policy. Unexpected adverse events, regardless of severity, will be reported to the DSTC and reviewed on a quarterly basis.

Expected Adverse Events

All fatal (grade 5) expected adverse events require expedited reporting and will be reported to the DSTC within 24 hours for review. Expected adverse events that are being captured on the study toxicity form will be reported at the time of the form's scheduled due date.

All DSTC reports and recommendations will be submitted to the IRB for their review.

Adverse Events Occurring after the End of the Study

Follow-up of AEs

Any unexpected AEs ongoing at the time of study discontinuation will be followed until resolution or stable for at least 2 months.

FDA Reporting Procedures

Commercial Agents: Commercial agents are those agents not provided under an IND but obtained instead from a commercial source. In some cases an agent obtained commercially may

be used for indications not included in the package label. The following procedures should be followed to determine if an adverse is reportable to the FDA:

Refer to the pharmaceutical section of the protocol to determine if an agent is investigational or commercial.

- **WHAT TO REPORT:** An unexpected, life-threatening (Grade 4) or unexpected, fatal (Grade 5) adverse event with an attribution of possible, probable or definite.

- **WHEN TO REPORT:** These events should be reported within (7) working days.

- **WHERE TO REPORT:** These adverse events with commercial agents must be reported to the FDA using the MedWatch form. A copy of the MedWatch form can be obtained from theFDA's MedWatch web site at www.fda.gov/medwatch. You can mail the reports to the address below or fax it 1-800-332-0178.

  MedWatch
  5600 Fishers Lane
  Rockville, MD 20852-9787

11. Patient Consent and Peer Review Statement

11.1. Subject Information and Informed Consent

Written informed consent must be obtained from the subject prior to study participation.

The informed consent document must be signed and dated by the subject and properly witnessed (if applicable) before initiation of any study procedures including any change in medication or initiation of study drug dosing.

Subjects must be consented in accordance with all local regulatory and legal requirements. This process must include a verbal explanation of the nature, scope, and possible consequences of the study provided in plain language. The information should

be presented by the investigator unless a designee is permitted by local regulations. The potential study subject should be encouraged to ask questions about the study.

The informed consent document must be prepared in accordance with GCP guidelines and with local regulatory and legal requirements. A copy of the signed consent form will be given to the subject and the original document must be safely archived by the investigator so that the forms can be retrieved at any time for monitoring, auditing, and inspection purposes.

The informed consent will be updated as appropriate (e.g., due to protocol amendment or if significant new safety information that may be relevant to consent of the subjects becomes available). If the informed consent is revised, it is the investigator's responsibility to ensure that an amended consent form is reviewed and signed by all subjects subsequently entered into the study and those currently in the study.

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13.Appendix A Assessment of Acute GVHD

Clinical Acute GVHD Assessment

Date _____ Patient ID _____ Karnofsky/Lansky _____

	Code						Differential Diagnosis						
	0	1	2	3	4	5		GVHD	Drug Rxn	Cond Reg	TPN	Infect	VOD	Other
Skin	☐	☐	☐	☐	☐		% body rash:____	☐	☐	☐		☐		_____
Lower GI	☐	☐	☐	☐	☐	☐	Vol: _____	☐	☐	☐	☐	☐		_____
Upper GI	☐	☐						☐	☐	☐	☐	☐		_____
Liver	☐	☐	☐	☐	☐		Max bili: _____	☐	☐	☐	☐	☐	☐	_____

Treatment: ☐ CSA ☐ Tacrolimus ☐ Pred ☐ Methylpred ☐ Ontak
☐ Pentostatin ☐ MMF ☐ Etanercept ☐ Other _____

Code Definitions:

Skin:
0 No rash
1 Maculopapular rash, <25% of body surface
2 Maculopapular rash, 25-50% of body surface
3 Generalized erythroderma
4 Generalized erythroderma with bullous formation and desquamation

Lower GI (Diarrhea):
0 None
1 ≤500 mL/day or <280 mL/m^2
2 501-1000 mL/day or 280- 555 mL/m^2
3 1001-1500 mL/day or 556- 833 mL/m^2
4 >1500 mL/day or >833 mL/m^2
5 Severe abdominal pain with or without ileus, or stool with frank blood or melena

Upper GI:
0 No protracted nausea and vomiting
1 Persistent nausea, vomiting or anorexia

Liver (Bilirubin):
0 <2.0 mg/dl
1 2.1-3.0 mg/dl
2 3.1-6.0 mg/dl
3 6.1-15.0 mg/dl
4 >15.1 mg/dl

Signature _____

TABLE 1.3.1 – GVHD STAGING

Stage	Skin	GI	Liver
1	< 25% rash	Diarrhea > 500ml/d or persistent nausea	Bilirubin 2-3mg/dl
2	25-50%	> 1000 ml/d	Bilirubin 3-6 mg/dl
3	> 50%	> 1500 ml/d	Bilirubin 6-15 mg/dl
4	Generalized erythroderma with bullae	Large volume diarrhea and severe abdominal pain ± ileus	Bilirubin > 15 mg/dl

TABLE 1.3.2 – CONSENSUS GVHD GRADING (PRZEPIORKA, ET. AL., 1995)

Grade	Skin	GI	Liver
I	Stage 1-2	0	0
II	Stage 3 or	Stage 1 or	Stage 1
III	---	Stage 2-4	Stage 2-3
IV	Stage 4	---	Stage 4

14. Appendix B Grading of Chronic GVHD (NIH Criteria)

Check all that apply	Score 0 - None	Score 1 - Mild	Score 2 - Moderate	Score 3 - Severe
Skin: *Clinical features:* ❑Maculopapular rash ❑Lichen planus-like features ❑Papulosquamous lesions or ichthyosis ❑Hyperpigmentation ❑Hypopigmentation ❑Keratosis pilaris ❑Erythema ❑Erythroderma ❑Sclerotic features ❑Poikiloderma ❑Pruritus ❑Hair Involvement ❑Nail Involvement % BSA involved __%	❑No symptoms	❑< 18% BSA with disease signs but NO sclerotic features	❑19-50% BSA, ❑Involvement with superficial sclerotic features "not hidebound" (able to pinch)	❑> 50% BSA ❑Deep sclerotic features "hidebound" (unable to pinch) ❑Impaired mobility, ulceration or severe pruritis
Mouth:	❑No symptoms	❑Mild symptoms with disease signs but not limiting oral intake significantly	❑Moderate symptoms with disease signs WITH partial limitation of oral intake	❑Severe symptoms with disease signs WITH **major** limitation of oral intake
Eyes: Mean tear test (mm): ❑> 10 ❑ 6-10 ❑≤ 5 ❑ Not done	❑	❑Mild dry eyes symptoms not affecting ADL (requiring eyedrops ≤ 3 x per day) ❑ Asymptomatic signs of keratoconjunctivitis sicca	❑Moderate dry eyes symptoms partially affecting ADL (requiring eyedrops > 3 x per day or punctual plugs) WITHOUT vision impairment	❑Severe dry eyes symptoms significantly affecting ADL (special eyewear to relieve pain) ❑Unable to work because of ocular symptoms ❑Loss of vision caused by keratoconjuctivitis sicca
Pulmonary **FEV1** ❑Not done **Pulmonary Fibrosis** **Bronchiolitis Obliterans** **Supplemental O2 required?** ❑Yes ❑No	❑No symptoms ❑FEV1> 80% ❑None ❑Not assessed ❑None ❑Yes, clinical ❑Yes, histologic ❑Not assessed	❑Mild symptoms (shortness of breath after climbing one flight of steps) ❑FEV1 60-78% ❑ Minimal radiographic findings	❑Moderate symptoms (shortness of breath after walking on flat ground) ❑FEV1 40-51% ❑Patchy or bi-basilar radiographic findings	❑Severe symptoms (shortness of breath at rest; requiring O_2) ❑FEV1 ≤ 39% ❑Extensive radiographic findings
GI Tract:	❑No symptoms	❑Symptoms such as dysphagia, anorexia, nausea, vomiting, abdominal pain or diarrhea without significant weight loss (< 5%)	❑Symptoms associated with mild to moderate weight loss (5 – 15%)	❑Symptoms associated with significant weight loss > 15% ❑ Requires nutritional supplement for most caloric needs ❑Esophageal dilation
Liver:	❑Normal LFT	❑Elevated Bilirubin, AP, AST or ALT < 2 x ULN	❑Bilirubin 3 – 10 mg/dl; liver enzymes 2-5 x ULN	❑Bilirubin > 10 mg/dL; liver enzymes > 5 x ULN
Genital Tract:	❑No symptoms	❑Symptomatic with mild signs on exam AND no effect on coitus and minimal discomfort with gynecological exam	❑Symptomatic with moderate signs on exam AND with mild dyspareunia or discomfort with gynecological exam	❑Symptomatic WITH advanced signs (stricture, labial agglutination or severe ulceration) AND severe pain with coitus or inability to insert vaginal speculum

Check all that apply	Score 0 - None	Score 1 - Mild	Score 2 - Moderate	Score 3 - Severe
Joints and Fascia:	❑ No symptoms	❑ Mild tightness of arms or legs, normal or mild decreased range of motion (ROM) AND not affecting ADL	❑ Tightness of arms or legs ❑ Joint contractures, erythema thought due to fascitis, moderate decreased ROM AND mild to moderate limitation of ADL	❑ Contractures WITH significant decrease of ROM AND significant limitation of ADLs (unable to tie shoes, button shirts, dress self etc.)

Other indicators, clinical manifestations or complications related to Chronic GvHD (check all that apply).
Assign a score to it's severity based on functional impact, where applicable (0= none, 1=mild, 2 = moderate, 3= severe)

❑ Ascites (serositis) ___	❑ Esophageal stricture or web___	❑ Nephrotic syndrome ___	❑ Pleural effusions ___
❑ Cardiac conduction defects ___	❑ Eosinophilia > 500 µl ___	❑ Pericardial effusion ___	❑ Polymyositis ___
❑ Cardiomyopathy ___	❑ Myasthenia Gravis ___	❑ Peripheral neuropathy ___	❑ Progressive onset___
❑ Coronary artery involvement ___		❑ Platelets < 100,000/µl ___	
❑ Other(s): Specify & score___			

Based on observations checked in the above table, select the severity of chronic GvHD for this assessment (Check only one)

☐ **None**

☐ **Mild chronic GVHD** involves only 1 or 2 organs or sites (except the lung: see below ‡), with no clinically significant functional impairment (maximum of score 1 in all affected organs or sites)

☐ **Moderate chronic GVHD** involves: (1) at least 1 organ or site with clinically significant but no major disability (maximum score of 2 in any affected organ or site) or (2) 3 or more organs or sites with no clinically significant functional impairment (maximum score of 1 in all affected organs or sites). ‡*A lung score of 1 will also be considered moderate chronic GVHD.*

☐ **Severe chronic GVHD** indicates major disability caused by chronic GVHD (score of 3 in any organ or site). ‡*A lung score of 2 or greater will also be considered severe chronic GVHD.*