Biol Blood Marrow Transplant 22 (2016) 1829–1835

Biology of Blood and Marrow Transplantation

journal homepage: www.bbmt.org

A Pharmacokinetic and Pharmacodynamic Study of Maraviroc as Acute Graft-versus-Host Disease Prophylaxis in Pediatric Allogeneic Stem Cell Transplant Recipients with Nonmalignant Diagnoses

Pooja Khandelwal [1,2,*], Tsuyoshi Fukuda [2,3], Kana Mizuno [2,3], Ashley Teusink-Cross [4], Parinda A. Mehta [1,2], Rebecca A. Marsh [1,2], Angela D.M. Kashuba [5], Alexander A. Vinks [2,3], Stella M. Davies [1,2]

[1] *Division of Bone Marrow Transplant and Immune Deficiency, Cincinnati Children's Hospital Medical Center, Cincinnati, Ohio*
[2] *Department of Pediatrics, University of Cincinnati College of Medicine, Cincinnati, Ohio*
[3] *Division of Clinical Pharmacology, Cincinnati Children's Hospital Medical Center, Cincinnati, Ohio*
[4] *Division of Pharmacy, Cincinnati Children's Hospital Medical Center, Cincinnati, Ohio*
[5] *Division of Pharmacotherapy and Experimental Therapeutics, University of North Carolina at Chapel Hill, Chapel Hill, North Carolina*

Article history:
Received 26 May 2016
Accepted 1 August 2016

Key Words:
Maraviroc
Chemokine receptor type 5 (CCR5) inhibition
CCR5 blockade
Lymphocyte chemotaxis

A B S T R A C T

Maraviroc is an allosteric small molecule antagonist of chemokine receptor type 5 (CCR5) and has been used in adult allogeneic hematopoietic stem cell transplant (HSCT) recipients to prevent acute graft-versus-host disease (GVHD) of the gastrointestinal (GI) tract and liver. The goal of this study was to establish feasibility and pharmacokinetic and pharmacodynamic profiles of maraviroc in pediatric HSCT recipients. Children ages 2 to 12 years were enrolled and maraviroc was added to standard GVHD prophylaxis, which included a calcineurin inhibitor and either steroids or mycophenolate mofetil. Maraviroc was started on day -3 and administered at a dose of approximately 300 mg/m^2 orally twice daily until day +30 after stem cell infusion. On days 0 and day +10, samples for pharmacokinetic analysis were collected before the dose and 1, 2, 4, 6, 8, and 12 hours after maraviroc administration. Additional trough concentrations were collected on days +7, 14, and 21. Patients were followed until day +100 for acute GVHD. Functional blockade of CCR5 was assessed in a pharmacodynamic assay by flow cytometry. Thirteen patients, median age of 4 years (range, 2 to 11 years), were prospectively enrolled. Underlying diagnoses included a primary immune deficiency (n = 6), hemoglobinopathy (n = 4), metabolic disorder (n = 1), and bone marrow failure syndrome (n = 2). Patients received either a myeloablative preparative regimen (n = 7) or a reduced-intensity conditioning regimen (n = 6). Cyclosporine and methylprednisolone (n = 7) was the predominant GVHD prophylactic regimen, followed by tacrolimus and mycophenolate mofetil (n = 4) and tacrolimus and steroids (n = 2). Two formulations of maraviroc (150-mg tablets and 20-mg/mL solution) were used on study. Mean (± SD) area under the concentration-time curve from 0 to 12 hours was 4805 ± 3265 hour * ng/mL on day 0 and 5917 ± 4048 hour * ng/mL on day +10. Four patients developed grade 1 or 2 acute skin GVHD before day +100 and were successfully treated. Two patients developed grade 3 acute GI GVHD on days +23 and +24 after HSCT and both had discontinued maraviroc before development of GI GVHD. No adverse effects attributable to maraviroc were observed and administration by enteral tubes was well tolerated by children and accepted by parents. All evaluable patients demonstrated functional CCR5 blockade on day 0. Administration of maraviroc is feasible in most pediatric HSCT recipients with good safety and tolerability profile.

INTRODUCTION

Acute graft-versus-host disease (GVHD) is the most significant complication of hematopoietic stem cell transplantation (HSCT), with high rates of morbidity and mortality [1]. Visceral involvement (gastrointestinal [GI] and liver) is particularly challenging to treat and carries a higher mortality risk when compared with acute skin GVHD [1-3]. Rates of acute GVHD continue to range from 20% to 50%, despite

Financial disclosure: See Acknowledgments on page 1834.

* Correspondence and reprint requests: Pooja Khandelwal, MD, Division of Bone Marrow Transplantation and Immune Deficiency, Cincinnati Children's Hospital Medical Center, 3333 Burnet Avenue, Cincinnati, OH 45229.
E-mail address: pooja.khandelwal@cchmc.org (P. Khandelwal).

http://dx.doi.org/10.1016/j.bbmt.2016.08.001
1083-8791/

the established benefit of prophylactic agents, such as calcineurin inhibitors, methotrexate and mycophenolate mofetil [2,4-7].

Chemokines are a group of small proteins that act together with their cell surface receptors to direct cells to specific locations throughout the body and function in T cell trafficking [8,9]. T cell trafficking is required for the development of visceral GVHD [10-12].

Chemokine receptor type 5 (CCR5) is a chemokine receptor, whose natural ligands are macrophage inflammatory protein-1-alpha and macrophage inflammatory protein-1-beta as well as (CCL5) [13,14]. CCR5 is expressed on a subset of T cells with a memory cell phenotype and on macrophages [15]. CCR5 is a G-protein coupled receptor with 7 transmembrane domains and is responsible for migration of T lymphocytes to the liver and the GI tract [16-18]. In murine models and adult human clinical trials, CCR5 inhibition has successfully reduced the incidence of acute visceral GVHD [17,19,20].

Maraviroc is the first Food and Drug Administration–approved drug in its class of CCR5 antagonists [21]. It is approved for second-line treatment of CCR5 tropic human immunodeficiency virus (HIV) infection and is a specific slowly reversible small molecule antagonist of the CCR5 receptor, preventing HIV-1 entry into cells [22,23].

Prevention of acute visceral GVHD by targeting CCR5 was explored in a single-center study in which adult HSCT patients received maraviroc in addition to standard GVHD prophylaxis. Virtually no liver or GI GVHD (0%) were observed until day +100 [20]. Using this preliminary experience, our overall objective was to utilize this lymphocyte trafficking inhibition strategy to reduce the incidence of acute visceral GVHD in pediatric HSCT recipients. This report describes the pharmacokinetic, pharmacodynamic, feasibility, and tolerability profiles of maraviroc in pediatric HSCT recipients.

METHODS

Study Subjects

Patients were prospectively enrolled on our center's institutional review board–approved study. Maraviroc was administered under an investigational new drug number 122997. Inclusion criteria to receive maraviroc on study were ages 2 to 12 years, GVHD prophylaxis of calcineurin inhibitor and either mycophenolate mofetil or corticosteroids, alanine aminotransferase (ALT) <5× upper limit of normal (ULN) on day of maraviroc administration, and no concurrent use of strong Cytochrome P450 3A4 (CYP3A4) inducers or inhibitors (eg, antifungal azoles) while receiving maraviroc. We chose to study the pharmacokinetic profiles of patients ≤12 years of age, as pharmacokinetic data are available for adult dosing in GVHD prophylaxis and it is likely that teenagers will have pharmacokinetic results similar to adults [20,24]. Exclusion criteria were ex vivo T cell depletion, any prior allergic reactions to maraviroc, or any known HIV infection before receiving maraviroc.

Maraviroc Administration

Maraviroc was administered orally, twice daily starting from day -3 and ending on day +30; day 0 was the day of planned stem cell infusion. Initial dosing of maraviroc (~300 mg/m^2) was based on data from study A4001031 [25], the phase 1 study of maraviroc in congenital HIV patients (Table 1), and doses were calculated based on body surface area. The formulations of maraviroc used were either 150-mg tablets or 20-mg/mL solution, provided by Pfizer. Enteral tubes were placed whenever feasible in younger children to ensure the reliability of dosing. Maraviroc administration with food was permitted. If emesis occurred within 30 minutes of administering maraviroc, the dose was repeated. Pharmacy and nursing drug administration records documented compliance and if patients were discharged before day +30, parents were provided with a log to document compliance with maraviroc.

Table 1
Dosing Schema for Maraviroc as per A4001031

Body Surface Area, m^2	Dose of Maraviroc to be Administered Orally
<.20	40 mg twice daily
.2-043	100 mg twice daily
.44-.72	200 mg twice daily
≥.73	300 mg twice daily

Pharmacokinetic Analysis

To characterize maraviroc pharmacokinetics in pediatric patients, plasma samples were obtained on day 0 (± 2 days) and day +10 (± 3 days) at the following time points: before dose and 1, 2, 4, 6, 8, and 12 hours after maraviroc administration. In addition, plasma samples were also obtained as troughs on day +7, day +14, and day +21 to collect additional pharmacokinetic data. Plasma concentrations of maraviroc were measured by liquid chromatography/mass spectrometry with the dynamic range of the assay being 1 ng/mL to 1000 ng/mL. Plasma concentration-time profiles were explored graphically and descriptive pharmacokinetic parameters, trapezoidal area under the concentration-time curve from 0 to 12 hours calculated by linear trapezoidal rule (AUC_{0-12}), maximum concentration, time to reach maximum concentration, and apparent oral clearance were obtained by noncompartmental analysis with the software package WinNonlin (Version 6.2, Pharsight Corporation, Palo Alto, CA).

Pharmacodynamic Assay

As previously reported, we observed a significant decrease in CCR5 expression on peripheral blood mononuclear cells once these were isolated by Ficoll-Hypaque density gradient separation [26]. To preserve CCR5 expression for accurate analysis, a pharmacodynamic assay was developed using whole blood with erythrocyte lysis. One hundred microliters of healthy control blood was incubated with 100 μL of patient plasma obtained before transplantation, day 0, or day +14, in the presence of 100 nM of CCL5 at 37°C and 5% CO_2 for 60 minutes. Cells were stained with fluorescent antibodies against CD3, CD8, CCR7, CD45RA, and CCR5. Red cells were lysed using FACSlyse (BD Biosciences, San Jose, CA) for 10 minutes, washed with phosphate-buffered saline twice, and fixed with 1% paraformaldehyde. Surface expression of CCR5 was assessed by a BD FACS Canto II flow cytometer (San Jose, CA). After gating on lymphocytes on forward versus side scatter plots, CD3+CD8+ T cells were gated and effector memory T cells were identified as $CD3^+CD8^+CCR7^-CD45RA^-$. CCR5 expression of these $CD8^+$ effector memory T cells was observed on histogram plots. In the presence of CCL5, CCR5 internalizes and is not detected via surface flow cytometry, but this expression is preserved when CCR5 in inhibited successfully with maraviroc (Figure 1). A concentration of approximately 50 ng/mL of maraviroc inhibits CCR5 internalization on $CD8^+$ effector memory T cells [20].

Monitoring for Acute GVHD

Patients were observed clinically until day +100 for an incidence of acute GVHD of any grade and any organ. Acute GVHD was graded using the modified Glucksberg criteria [27] by the treating physician.

Monitoring for Toxicity

Adverse effect monitoring was performed throughout the study. Toxicity was monitored by Common Terminology Criteria for Adverse Events version 4.03. As maraviroc carries a black box warning for hepatotoxicity, liver function tests were monitored frequently while the drug was administered. Maraviroc was held for grade 3 toxicities attributable to the drug or any grade 3 or higher liver toxicity, even if unrelated to the drug. Additional clinical parameters including infectious complications, whole blood donor chimerism, and survival at day +100 were also monitored.

RESULTS

Study Subjects

Fifteen patients were enrolled on study and 13 patients received maraviroc. Two patients were enrolled but did not receive maraviroc therapy. The first patient had severe autism spectrum disorder and did not tolerate oral medications or placement of enteral tubes during the transplantation course. The second patient had an elevation of her ALT >5 × ULN on day -3 due to her underlying diagnosis of chronic active Epstein-Barr virus (EBV). As the ALT did not normalize during the period of planned maraviroc administration, the drug was not initiated. Demographics and disease characteristics of the 13 patients who received maraviroc are shown in Table 2. The median age of the patients who received maraviroc was 4 years (range, 2 to 11 years). One patient received a maraviroc dose of 150 mg (rounded to nearest tablet size, which was