INVESTIGATOR'S BROCHURE
MultiStem®

PERIOD COVERED BY THIS REPORT:
26 Nov 2019 through 25 Nov 2020

Version Date: 22 Jan 2021
Edition No.: 8.0

INVESTIGATOR'S BROCHURE RECEIPT

Herewith I acknowledge the receipt of the Investigator's Brochure for MultiStem®.

Investigator's Signature Date

Investigator's Name

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LIST OF ABBREVIATIONS AND DEFINITION OF TERMS

Abbreviation	Definition
AE	Adverse Event
AKI	Acute Kidney Injury
ALL	Acute Lymphocytic Leukemia
AMI	Acute Myocardial Infarction
AML	Acute Myelocytic Leukemia
ARDS	Acute Respiratory Distress Syndrome
ASA	Aminosalicylic Acid
ATP	Adenosine Triphosphate
AZA	Azathioprine
CD	Cluster of Differentiation
CHF	Chronic Heart Failure
CFR	Code of Federal Regulations
CHMP	Committee for Medicinal Products for Human Use
CLL	Chronic Lymphocytic Leukemia
CML	Chronic Myelocytic Leukemia
CNI	Calcineurin Inhibitor
CNS	Central Nervous System
CRM	Continual Reassessment Method
CT	Computed Tomography
DLT	Dose Limiting Toxicity
DMSO	Dimethyl Sulfoxide
DSMB	Data and Safety Monitoring Board
EC	Eriochrome cyanine
EMA	European Medicines Agency
EU	European Union
EudraCT	European Union Drug Regulating Authorities Clinical Trials Database
FDA	Food and Drug Administration
FPI	First Patient In
GFP	Green Fluorescent Protein
GI	Gastrointestinal
GLP	Good Laboratory Practice
GvHD	Graft versus Host Disease
HCT/Ps	Human Cells, Tissues, and Cellular and Tissue Based Products
HLA	Human Leukocyte Antigen
HSA	Human Serum Albumin
HSCT	Hematopoietic Stem Cell Transplantation
HTS	HypoThermosol
IB	Investigator Brochure
IFN	Interferon
IL	Interleukin
IND	Investigational New Drug
IP	Investigational Product
IV	Intravenous
KC	Keratinocyte-Derived Cytokine
LAD	Left Anterior Descending Artery
LPO	Last Patient Out
LPS	Lipopolysaccharide

Abbreviation	Definition
LV	Left Ventricular
LVEF	Left Ventricular Ejection Fraction
MAPC	Multipotent Adult Progenitor Cells
MASTERS	MultiStem Administration for Stroke Treatment and Enhanced Recovery Study
MCAL	Middle Cerebral Artery Ligation
MCAO	Middle Cerebral Artery Occlusion
MDS	Myelodysplastic Syndrome
MHC	Major Histocompatibility Complex
MI	Myocardial Infarction
MiSOT	Mesenchymal Stem Cells in Solid Organ Transplantation
MLR	Mixed Lymphocyte Reactions
6-MP	6-Mercaptopurine
MRI	Magnetic Resonance Imaging
mRS	Modified Rankin Scale
MSC	Mesenchymal Stem Cells
mTOR	Mammalian Target of Rapamycin
N/A	Not Applicable
NHLBI	National Heart, Lung and Blood Institute
NIHSS	National Institutes of Health Stroke Scale
NMP	Normothermic machine perfusion
NOAEL	No-Observable-Adverse-Effect Level
NOD/SCID	Nonobese Diabetic / Severe Combined Immunodeficiency
NSTEMI	Non-ST Elevation Acute Myocardial Infarction
PAI	Plasminogen activator inhibitor
PaO2	Partial Pressure of Arterial Oxygen
PBMC	Peripheral Blood Mononuclear Cells
PBS	Phosphate Buffered Saline
PCI	Percutaneous Coronary Intervention
PD	Population Doublings
PET	Positron Emission Tomography
PMDA	Pharmaceuticals and Medical Devices Agency
PROPPR	Pragmatic Randomized Optimal Platelet and Plasma Ratios
PT	Preferred Term
qPCR	Quantitative Polymerase Chain Reaction
RBC	Red Blood Cell
RMAT	Regenerative Medicine Advanced Therapy
RR	Respiratory Rate
rtPA	Recombinant tissues plasminogen activator
SAE	Serious Adverse Event
SC	Subcutaneous
SCI	Spinal Cord Injury
SDF	Stromal Cell-Derived Factor
SIRS	Systemic Inflammatory Response Syndrome
SNP	Single Nucleotide Polymorphism
SOC	System Organ Class
SOT	Solid Organ Transplant
STEMI	ST Elevation Myocardial Infarction
SUSAR	Suspected Unexpected Serious Adverse Reaction

Abbreviation	Definition
TBD	To Be Determined
TBI	Traumatic Brain Injury
TNF	Tumor Necrosis Factor
tPA	Tissue Plasminogen Activator
TREASURE	Treatment Evaluation of Acute Stroke for Using in Regenerative Cell Elements
UC	Ulcerative Colitis
UK	United Kingdom
UKR	Universitätsklinikum Regensburg
US	United States
VEGF	Vascular Endothelial Growth Factor
WBC	White Blood Cell
WHO	World Health Organization

1. REGULATORY UPDATE

This harmonized Investigator Brochure (IB) is for the single active substance MultiStem®, covering the reporting period 26 Nov 2019 through 25 Nov 2020, it is an update since the last version (7.3) released 15 May 2020.

The Development International Birth Date for MultiStem is 26 Nov 2008.

This harmonized MultiStem IB contains relevant clinical development information for MultiStem in the following indications: prophylaxis and treatment of graft versus host disease (GvHD), IND 13507; treatment of acute myocardial infarction (AMI), IND 13554; treatment of ischemic stroke (Stroke), IND 13852; treatment of moderate and severe acute respiratory distress syndrome (ARDS), IND 16460; treatment of trauma, IND 19272; and treatment of acute spinal cord injury (SCI), expanded access, single patient emergency sponsored under Stroke IND 13852.

In addition, this harmonized IB contains updated information regarding 5 additional completed or terminated MultiStem clinical trials conducted by sponsors other than Athersys [treatment of ulcerative colitis, IND 14512/EudraCT 2010-022766-27/Canada CTA 9427-P0006\7-237C, sponsor Pfizer; treatment of GvHD, expanded access, single patient emergency Investigator (Dr Laura Newell) sponsored IND 16128; and Mesenchymal stem cells in solid organ transplant, EudraCT 2009-017795-25, sponsor Freistaat Bayern/Universitätsklinikum Regensburg (UKR)]. There are also 2 ongoing clinical trials (Ischemic stroke and ARDS) being conducted in Japan by HEALIOS K.K.

Table 1-1 below summarizes the regulatory status of MultiStem ongoing and completed/terminated clinical studies, and the corresponding IND and/or EudraCT numbers.

Table 1-1. Regulatory Status of MultiStem Clinical Trials

	US IND No.	Status of Clinical Study as of 25 Nov 2020	Clinical Study Disposition	Europe EudraCT No.
Graft versus Host Disease	13507	Phase 1 – prophylaxis - completed in November 2011	Safe to proceed (10/2007)	Study was conducted in Belgium (EudraCT No. 2010-018760-16)
		Phase 2/3 – prophylaxis – (registration) planned	SPA agreement granted (12/2015)	TBD
Acute Myocardial Infarction	13554	Phase 1 – STEMI – completed in February 2012	Safe to proceed (12/2007)	N/A
		Phase 2 – NSTEMI – terminated February 2020	Protocol accepted (01/2015)	N/A
Ischemic Stroke	13852	MASTERS-1 Phase 2 – completed December 2016	Safe to proceed (12/2008)	Study is completed in the UK (EudraCT No. 2012-005749-18)
		MASTERS-2 Phase 3 – treatment – (registration) - ongoing	SPA agreement granted (09/2016) Fast Track designation granted (05/2017) RMAT designation granted (08/2017)	Study not initiated yet in EU (EudraCT No. 2019-001680-69)
		*TREASURE Phase 2/3 – treatment – ongoing	Safe to proceed (08/2016) PMDA	N/A (Study conducted in Japan)
Acute Respiratory Distress Syndrome	16460	Phase 1/2 – Moderate/Severe ARDS – completed July 2019	Safe to proceed (07/2015) Fast Track designation granted (04/2019)	Study is completed in the UK (EudraCT No. 2015-001586-96)
	Non-IND	*ONE-BRIDGE Phase 2 – ARDS caused by pneumonia or COVID-19 - ongoing	Safe to proceed (10/2018) PMDA	N/A (Study conducted in Japan)
	16460	MACoVIA Phase 2/3 COVID-19 induced ARDS-ongoing	Safe to proceed (4/2020)	N/A
*Ulcerative Colitis (Partner /Study Sponsor - Pfizer)	14512	Phase 2 – completed November 2014	Safe to proceed (11/2011)	Study is completed in the EU (EudraCT No. 2010-022766-27) and Canada (CTA No. 9427-P0006\7-237C)

Table 1-1. Regulatory Status of MultiStem Clinical Trials

	US IND No.	Status of Clinical Study as of 25 Nov 2020	Clinical Study Disposition	Europe EudraCT No.
*Treatment of GvHD	16128	Phase 1b – expanded access use, single patient emergency IND - terminated	Safe to proceed (08/2014)	N/A
Treatment of SCI	13852	Phase 1b – expanded access use, single patient emergency under Stroke IND - terminated	Safe to proceed (02/2016)	N/A
*MiSOT (Study Sponsor – UKR)	Non-IND	Phase 1/2 – Liver transplant - terminated	Safe to proceed (11/2011)	Study was terminated in Germany (EudraCT No. 2009-017795-25)
Trauma	19272	Phase 2 – Trauma- ongoing	Safe to proceed (4/2020)	N/A

* = additional ongoing or completed MultiStem clinical trials conducted by sponsors other than Athersys
EU = European Union; N/A = not applicable; MiSOT = mesenchymal stem cells in solid organ transplantation; RMAT = Regenerative Medicine Advanced Therapy designation; SPA = Special Protocol Assessment designation; TBD = to be determined.

As of 25 Nov 2020, MultiStem product MultiStem product is being used or has been used in clinical trials as listed above in Table 1-1.

- Five studies with MultiStem product have been completed:
  1. AMI treatment (AMI 07-001): Phase 1 dose-escalation, open-label, safety study in 25 patients including 6 registry patients after acute myocardial infarction.
  2. GvHD prophylaxis (GVHD-2007-001): Phase 1 dose-escalation, open-label, safety study in 36 patients undergoing hematopoietic stem cell transplant.
  3. Ulcerative colitis (B3041001): a double-blind, placebo-controlled Phase 2 clinical trial sponsored by Athersys' partner, Pfizer. This study enrolled 105 patients.
  4. Ischemic stroke (B01-02): a double-blind, placebo-controlled Phase 2 clinical trial in 134 subjects who suffered an acute ischemic stroke.
  5. ARDS (B04-01): a Phase 1/2 clinical trial sponsored by Athersys that included open-label, dose-escalation cohorts and double-blind, placebo-controlled cohorts. This study enrolled 36 subjects.
- Five studies with MultiStem product are ongoing:
  1. Ischemic stroke (B01-03): a double-blind, placebo-controlled Phase 2/3 clinical trial in approximately 220 subjects who suffered an acute ischemic stroke in Japan. This study has enrolled 189 subjects as of this document's cutoff date.
  2. Ischemic stroke (B01-04): a double-blind, placebo-controlled Phase 3 clinical trial in approximately 300 subjects who suffered an acute ischemic stroke in North America, Europe, and Asia Pacific. This study has enrolled 56 subjects in the US as of this document's cutoff date.
  3. ARDS (B04-02): an open-label, standard treatment-controlled Phase 2 clinical trial in approximately 30 subjects who suffered ARDS caused by pneumonia and 5 subjects with ARDS caused by Covid-19 in Japan. This study has enrolled 33 subjects (28 ARDS pneumonia subjects and 5 ARDS Covid-19 subjects) in Japan as of this document's cutoff date.
  4. ARDS (B04-03): a Phase 2/3 clinical trial in approximately 300 to 400 subjects who suffered ARDS caused by Covid-19 in the US. This study has enrolled 10 subjects as of this document's cutoff date.
  5. Trauma (B06-01): a double-blind, placebo-controlled Phase 2 clinical trial in approximately 156 subjects with trauma induced multiple organ failure/systemic inflammatory response syndrome in the US. This study has enrolled zero subjects as of this document's cutoff date.
- Two studies and 2, single patient expanded access cases were terminated:
  1. Treatment of GvHD: an expanded access use, single patient emergency Investigator-sponsored IND was opened in August 2014. The patient was steroid refractory gastrointestinal (GI) and Liver Grade 4, received 4 of 6 MultiStem scheduled treatment doses over a period of 3 months (21 Aug 2014 through 24 Oct 14). The patient died 05 Nov 2014. Death not related to MultiStem treatment. The study was terminated.
  2. Treatment of acute SCI: an expanded access use, single patient emergency sponsored under the Athersys Stroke IND was granted in February 2016. The patient had a C4/C5 fracture and dislocation from an ice hockey accident and received 1.2 billion cells of MultiStem on 23 Feb 2016, which was infused within 36 hours of initial injury. The patient is a quadriplegic and was released from the hospital to continue their recovery. The study was terminated.
  3. Liver Transplant (MiSOT-I): a non-IND investigator-initiated trial for the indication of liver transplantation, which was a Phase 1, open-label study, evaluating safety and feasibility of MultiStem for immunomodulation therapy after liver transplantation sponsored by Freistaat Bayern in Germany. Three subjects were enrolled, and the study was terminated due to poor enrollment.

4. AMI (B02-02): a Phase 2 – study sponsored by Athersys. Thirty-four subjects were enrolled, and the study was terminated due to poor enrollment.

- MultiStem product (adult adherent bone marrow-derived multipotent stem cells) has been granted:
  - Orphan drug designation for the indication "prophylaxis of graft versus host disease" (15 Sep 2010) by the United States Food and Drug Administration (FDA)
  - Orphan drug designation for the prevention of GvHD by the European Commission (16 Jan 2014, EU/3/13/1233)
  - The GvHD program has also received "Fast Track" designation by the FDA (February 2015)
  - The registration pivotal Phase 2/3 adoptive, double blind study design for prevention of GvHD in adults and children older than or equal to 13 years of age following hematopoietic cell transplant for hematological malignancies has been approved by (European Medicines Agency (EMA) and a positive opinion was adopted in February 2015 by Committee for Medicinal Products for Human Use (CHMP)
  - Similarly, the FDA granted Athersys an agreement to Special Protocol Assessment (SPA) for the single pivotal Phase 2/3 registration GvHD study (December 2015)
  - FDA has also granted the pivotal Phase 3 Ischemic Stroke study MASTERS-2 the SPA designation to support registration (September 2016)
  - The registration pivotal Phase 3, double blind study design (MASTERS-2) for treatment of Ischemic Stroke in Adults within 36 hours of event, has been approved by EMA and a positive opinion was adopted in June 2017 by CHMP
  - FDA has granted Ischemic Stroke Fast Track designation (May 2017)
  - EMA has granted the MultiStem product the Advanced Therapy Medicinal Product Certificate for Quality Data (May 2020)
  - FDA has granted Ischemic Stroke Regenerative Medicine Advanced Therapy (RMAT) designation (August 2017)
  - FDA has granted Acute Respiratory Distress Syndrome Fast Track designation (April 2019)
  - FDA has granted the MultiStem program for treatment of Acute Respiratory Distress Syndrome, the Regenerative Medicine Advanced Therapy (RMAT) designation (Sept 2020)
- MultiStem has been well tolerated in approximately 413 subjects exposed to MultiStem as of this document's cut-off date across a range of acute and chronic disorders, doses, dose regimens, and routes of delivery
- This product has not been commercially approved anywhere in the world

2. SUMMARY

MultiStem is an adult adherent cell product derived from the bone marrow of a nonrelated donor and expanded *ex vivo*. MultiStem cell therapy represents a mechanistically novel approach for numerous indications, such as treatment following AMI, prevention of GvHD, treatment of acute ischemic stroke, treatment of UC, support during and after system organ transplant (SOT), treatment of ARDS, and prevention of inflammatory complications following traumatic injury.

The evaluation of MultiStem for clinical use is based on nonclinical pharmacological, biodistribution and safety studies in animal models that support the investigational use of MultiStem cells in these indications. *In vitro* pharmacology studies of rat and human MultiStem cells have shown that MultiStem can inhibit T-cell proliferation, indicating that the cells are immunomodulatory. This conclusion was further supported by *in vivo* pharmacology studies conducted in pig AMI models, mouse and rat GvHD and ischemic injury models, and a rat cardiac transplant model.

MultiStem cells initially localize to the lungs after intravenous (IV) injection. Pulmonary and cardiovascular safety pharmacology studies have demonstrated that after single and multiple dose IV infusions of rat or human MultiStem cells, there were no adverse effects on pulmonary function and no effects on cardiovascular safety measurements.

The nonclinical assessment of the pharmacokinetics of MultiStem cells focused on biodistribution and residual presence in tissue following dosing. The biodistribution of MultiStem following IV infusion of cells in a mouse model of GvHD has been evaluated through *in vivo* imaging of cells labeled with a luciferase reporter gene. Imaging of animals in this model indicated an initial accumulation of MultiStem cells in the lungs in the hours immediately following administration, followed by re-distribution to the GI tract over the following 24 to 48 hours. In the majority of samples measured, the bioluminescence reporter signal was below the limit of detection at 10 days post infusion, suggesting that the majority of the administered cells were cleared by this time point.

Biodistribution and persistence of multi-potent adult progenitor cell (MAPC®)/MultiStem cells have also been evaluated in nonobese diabetic/severe combined immunodeficiency (NOD/SCID) mice and in other rodent and pig disease models. There was no evidence of tumorigenicity in subcutaneous (SC) and IV nude mouse tumorigenicity studies or in any other nonclinical studies where tissues were evaluated. The majority of detectable allogeneic rat MAPC cells were cleared from tissues within a few days of administration in rat studies. In addition, 2- and 4-week biodistribution studies in the brains of stroke injured rats were conducted, and no detectable signal was found by quantitative Polymerase Chain Reaction (qPCR) for human cells in the animal brains at either time point.

Generation of an immune response against MultiStem cells, as measured by alloantibody or T-cell activation responses, has not been observed *in vitro* or *in vivo* in single- or multiple-dose cell administration rat studies.

Single-dose toxicity studies in mice and rats have been conducted with both human and rat MultiStem cells. The cells were well-tolerated up to 10 million cells/dose (500 million cells/kg; human MultiStem delivered SC in mice) and up to 40 million cells/dose (200 million cells/kg; rat MAPC delivered IV in rats). Allogeneic rat MultiStem cells have been administered to rats in an IV study up to 5 weeks in duration (once weekly dosing). The no-observed-adverse-effect level (NOAEL) in this study was 2.5 million cells/dose (12.5 million cells/kg). Two IV doses of 10 million allogeneic rat cells/dose (50 million cells/kg) have been given 1 week apart to rats with no adverse effects observed. Additional studies have demonstrated that direct delivery of 200 million MultiStem cells into the adventitia of the pig coronary artery was well tolerated.

The utility of MultiStem is being explored in a number of clinical trials, including completed or terminated trials in patients following AMI and acute ischemic stroke, as a prophylactic treatment for GvHD in patients undergoing hematopoietic stem cell transplantation (HSCT), in patients with UC,

ARDS, and following liver transplantation, and during ongoing trials in patients following an acute ischemic stroke, or in patients with ARDS or trauma-induced multiple organ failure/systemic inflammatory response syndrome (SIRS). These studies use different MultiStem formulations and concentrations, but identical cellular constituents. While MultiStem was delivered locally to the heart in the AMI trials, it was infused as single or multiple IV infusions in the completed GvHD, ischemic stroke, liver transplantation, and UC trials. As of 25 Nov 2020, approximately 413 patients have received MultiStem in these ongoing and completed or terminated trials. From completed or terminated trials, this included 36 AMI patients receiving transarterial injection of MultiStem; 36 GvHD patients receiving IV infusion of MultiStem through a central line; 84 UC patients, 71 acute ischemic stroke patients, 26 ARDS patients, and 1 SCI and 1 GvHD treatment expanded access use patients receiving an IV infusion of MultiStem through a peripheral line, and 3 liver transplant patients receiving MultiStem via the portal circulation and through a peripheral line. From ongoing trials, this included an estimated 123 ischemic stroke patients and 32 ARDS patients receiving an IV infusion of MultiStem through a peripheral line. As of data cut-off on 25 Nov 2020, there have been no infusional or allergic reactions reported per protocol definitions and adverse events have been consistent with the disease states being studied in completed or ongoing trials. The ARDS trial in Japan had a report of a serious adverse event of chills possibly related to MultiStem. In the UC trial, 2 separate serious adverse events of hypersensitivity and pancytopenia were reported and considered possibly related to MultiStem or the product used to dilute the stem cells.

3. INTRODUCTION

MultiStem is an allogeneic bone-marrow derived MAPC-based medicinal product. Accordingly, MultiStem is a proprietary subset of the allogeneic bone marrow-derived stem cells. MultiStem refers to the registered (or trademarked), clinical grade GMP MAPC product, whereas MAPC is used to refer to non-GMP product used in nonclinical studies. In scientific publications and in several instances in this document, the 2 nomenclatures, MultiStem and MAPC are used interchangeably. The product consists of expanded stem cells isolated from allogeneic bone marrow. Cells are expanded *ex vivo* from a single donor. MultiStem cells are well characterized and distinguishable from bone marrow mononuclear cells and classical mesenchymal stem cells (MSCs) based on phenotype, size, transcriptome, secretome, miRNA profile, differentiation, and expansion capacity (Ulloa-Montoya, 2007). MultiStem cells fall within the formal criteria for designation of cells as mesenchymal lineage defined in the position statement of the International Society for Cellular Therapy. The potential benefit of MultiStem in various indications is summarized below.

3.1. Stem Cell Treatment and Potential Indications

3.1.1. Acute Myocardial Infarction

Cardiovascular disease is the leading cause of death globally. Despite important advances in the last twenty years, cardiovascular diseases killed an estimated 17.8 million people in 2017. Of these, an estimated 7.3 million were due to coronary artery disease and 6.2 million were due to stroke (Roth, 2018). Additionally, AMI frequently leads to congestive heart disease, which itself is often characterized by a significant decline in quality of life and high morbidity and mortality rates. Despite many advances over the last two decades, AMI and other diseases caused by ischemic heart disease continue to remain a major cause of morbidity and mortality.

The most frequent cause of AMI is a blockage of blood flow to the heart caused by rupture of an atherosclerotic plaque and formation of an occluding blood clot. Arrhythmia occurs in some form in > 90% of patients. Primary care is aimed at relieving distress, reversing ischemia, limiting the infarct size, reducing cardiac work, and preventing and treating complications (Merck, 2005). Triage is important in determining what further treatments are required for the individual patient.

Aspirin, if not contraindicated, should be used to treat patients with AMI and continued indefinitely to reduce vascular death, nonfatal myocardial infarction (MI), and nonfatal stroke. Oxygen and morphine are used as needed. Thrombolytic therapy is most effective in the first few minutes and hours of AMI. Drugs available in the US include streptokinase, streptokinase-urokinase, and tissue plasminogen activator (tPA). Additional medications, including aspirin, clopidogrel, and dipyridamole, can reduce the risk of AMI in patients with known risk factors such as acute ST-elevation (STEMI) or non-ST elevation (NSTEMI) MI, unstable angina, prior MI, ischemic stroke, and peripheral arterial disease (Aronow, 2007).

During AMI, ischemia causes myocardial cell death and progressive loss of contractile tissue. Subsequently, structural changes lead to left ventricular remodeling, finally resulting in the development of heart failure (Assmus, 2006). Cardiac cells possess a limited ability to regenerate. Therefore, AMI also results in replacement of "perished" cardiomyocytes by scar tissue. The consequence of this is lowering of myocardial contractile function and the development of heart failure.

The mortality rates associated with AMI have significantly decreased over the past 2 decades (Rogers, 2000; Rogers, 2007). Beginning first with thrombolytic therapy for AMI, and more recently with growing acceptance and availability of primary percutaneous coronary intervention (PCI) for ST-elevation AMI, the mortality rates of this devastating ischemic event have decreased from almost 15% in clinical trials in the late 1980s to < 5% in recent primary percutaneous coronary intervention trials (Montalescot, 2001; Grines, 2002; Stone, 2002). Accompanying this decrease in mortality has been a significant increase in

patients with chronic heart failure (CHF). It is estimated that 22% of men and 46% of women who have experienced MI will be disabled with heart failure (Thom, 2006).

3.1.1.1. Stem Cell Treatment in Acute Myocardial Infarction

Cellular therapy for cardiac disease is a burgeoning field of clinical research, aimed at developing potential treatments for patients with ischemic heart disease and/or congestive heart failure. Cell therapeutics hold the promise of treating cardiovascular disease through the protection or support of ischemic tissue, thereby improving function, and preventing or ameliorating cardiovascular remodeling and decline in function. Ischemic injury to the heart can result in significant myocyte cell death and impaired ventricular function. Scar formation and compensatory remodeling can lead to long term consequences associated with congestive heart failure. Preclinical and early clinical data suggests that cell therapy can provide therapeutic benefit through multiple potential biological mechanisms and paracrine effects including preventing apoptosis and limiting inflammatory damage, stimulating angiogenesis, and producing homing factors that may mobilize patient stem cells or progenitors thereby improving cardiac recovery.

The goal of cell therapy for patients with AMI is to improve cardiac function and reduce the development and onset of CHF. MultiStem has the potential for achieving this goal by protecting cardiomyocytes from cell death and stimulating new angiogenesis. Nonclinical studies with small and large animals using both autologous and allogeneic MultiStem at the time of AMI demonstrated significant improvements in cardiac function. MultiStem can be delivered to the peri-infarct zone surrounding the ischemic scar via catheter-based delivery that leads to improved cardiac function. Trophic factors, such as vascular endothelial growth factor (VEGF) and others secreted by MultiStem in response to the injury microenvironment are likely pathway mediators (Van't Hof, 2007; Zeng, 2007). Significant neo-angiogenesis and improved heart bioenergetics are associated with sustained improvement in ventricular function.

A Phase 1 clinical study with MultiStem, for patients with first-time STEMI demonstrated that MultiStem was well tolerated (Penn, 2012). After 4 months, a trend towards an increase in cardiac function was observed as measured by left ventricular ejection fraction (LVEF). Based on these results, it would appear likely that cell therapy will play a role in the prevention and treatment of cardiac dysfunction in the ensuing years.

3.1.2. Graft versus Host Disease

The leukemias are cancers of the white blood cells (WBCs) involving bone marrow, circulating WBCs, and organs such as the spleen and lymph nodes. Malignant transformation usually occurs at the hematopoietic stem cell level, although it sometimes involves a committed progenitor cell with more limited capacity for differentiation. Abnormal proliferation, clonal expansion, and diminished apoptosis (programmed cell death) lead to replacement of normal blood elements with malignant cells. Manifestations of leukemia are due to suppression of normal blood cell formation and organ infiltration by leukemic cells. Inhibitory factors produced by leukemic cells and replacement of marrow space may suppress normal hematopoiesis, with ensuing anemia, thrombocytopenia, and granulocytopenia. Organ infiltration results in enlargement of the liver, spleen, and lymph nodes, with occasional kidney and gonadal involvement.

Leukemias were originally termed acute or chronic based on life expectancy but now are classified according to cellular maturity. Acute leukemias consist of predominantly immature, poorly differentiated cells (usually blast forms); chronic leukemias have more mature cells. Acute and chronic leukemias are divided into lymphocytic (ALL and CLL) and myelocytic (AML and CML). The most common leukemias are ALL in children (peak incidence ages 2-10), AML in any age group, CLL in mid to old age, and CML in young adults. Myelodysplastic syndromes (MDS) involve progressive bone marrow failure but with an insufficient proportion of blast cells (< 30%) for making a definite diagnosis of AML;

40% to 60% of cases evolve into AML (Merck, 2005). Survival in untreated acute leukemia generally is 3 to 6 months.

The goal of treatment is complete remission, including resolution of abnormal clinical features, restoration of normal blood counts and normal hematopoiesis with < 5% blast cells, and elimination of the leukemic clone. Although basic principles in treating ALL and AML are similar, the drug regimens differ. The 4 general phases of treatment for ALL include remission induction, central nervous system (CNS) prophylaxis, post-remission consolidation or intensification, and maintenance. Several regimens emphasize early introduction of an intensive multi-drug regimen. Therapy duration is usually 2.5 to 3 years but may be shorter with regimens that are more intensive in earlier phases and for B cell cases. For a patient in continuous complete remission for 2.5 years, the risk of relapse after therapy cessation is about 20%, usually within 1 year. Thus, when therapy can be stopped, most patients are cured. Leukemic cells may reappear in the bone marrow, the CNS, or the testes. Bone marrow relapse is of particular concern. Although a new round of chemotherapy may induce a second remission in 80% to 90% of children (30% to 40% of adults), subsequent remissions tend to be brief. Only a few patients with late bone marrow relapses achieve long disease-free second remissions or cure. If a human leukocyte antigen (HLA)-matched sibling is available, stem cell transplantation offers the greatest hope of long-term remission or cure. For AML, treatment includes induction chemotherapy to achieve remission and post-remission chemotherapy (with or without stem cell transplantation) to avoid relapse.

For MDS, prognosis depends greatly on classification and on any associated disease. Patients with refractory anemia or refractory anemia with sideroblasts are less likely to progress to the more aggressive forms and may die of unrelated causes. Azacytidine and decitabine improve symptoms, decrease the rate of transformation to leukemia and the need for transfusions, and probably improve survival. Other therapy is supportive, including red blood cell (RBC) transfusions as indicated, platelet transfusions for bleeding, and antibiotic therapy for infection. In some patients, erythropoietin to support RBC needs, granulocyte colony-stimulating factor to manage severe symptomatic granulocytopenia, and, when available, thrombopoietin for severe thrombocytopenia can serve as important hematopoietic support but have not proved to increase survival. Allogeneic stem cell transplantation is useful, and non-ablative allogeneic bone marrow transplantations are now being studied for patients > 50 years of age, for whom myeloablation is too risky.

Stem cell transplantation is an option for some patients with recurrence of ALL, AML, CML, and MDS. A limitation is the availability of a matched donor and the patient's age and disease status. Myeloablation of the existing bone marrow can lead to many complications, including serious infections. Matched related donor transplantation is more effective than non-related. For the patient in leukemic relapse, HSCT is often a last option. There are complications of such transplantation, some related to the myeloablative drugs and others to the conditioning regimen used to try to prevent rejection.

Acute GvHD is one of the major limitations of allogeneic HSCT. This complication is thought to be initiated by activation of adoptively transferred, mature donor T-cells through recognition of target antigens presented on major histocompatibility complex (MHC) molecules expressed on antigen-presenting cells that reside within host tissues. Moderate to severe GvHD Grades II-IV occurs in 30% to 50% of matched related HSCTs (Gale, 1989; Martin, 1991; Weisdorf, 2007) and 50% to 70% of unrelated donor recipients (Hansen, 1997; Nash, 2000), and is a major cause of morbidity and mortality. Although the incidence of GvHD is influenced by many recipient and donor factors, alterations of prophylactic regimens have had only limited evolution over the last fifteen years. It is recognized that innovations in this area are needed which would further reduce the incidence of GvHD without increasing relapse or risk of infection in HSCT patients.

3.1.2.1. Stem Cell Treatment in Graft versus Host Disease

Based on its immunological properties, MultiStem could provide therapeutic benefit following myeloablation and HSCT in hematological malignancy patients as described below.

MultiStem cells are minimally immunogenic and have the potential to diminish GvHD in allogeneic bone marrow recipients. MultiStem cells do not activate allo-T cells *in vitro* in mixed lymphocyte reaction assays (MLR) nor do they generate an allo-immune response when administered *in vivo*. MultiStem cells have been shown to suppress immune responses between allo-reactive T-cells from 2 unrelated individuals as measured in MLR assays. Based upon these properties, MultiStem has the potential to reduce the incidence and/or severity of GvHD in leukemia patients.

Finally, MultiStem may enhance engraftment of allogeneic bone marrow cells by providing an improved micro-environment through the secretion of cytokines and homing factors. A number of publications document enhanced hematopoietic recovery with co-administration of MSC and allogeneic or xenogeneic bone marrow grafts. Nonclinical studies performed using human MSC and human (cluster of differentiation) CD34+ HSC populations show increased chimerism and support the accelerated hematopoietic recovery shown clinically (Auletta, 2010). This is likely achieved via several pathways, including production of "homing factors" such as stromal cell-derived factor-1 (SDF-1) that recruit HSC to bone marrow, and production of cytokines supportive of hematopoiesis such as interleukin (IL)-6 (Klyushnenkova, 2005).

3.1.3. Stroke

Globally, stroke is one of the leading causes of death and disability (Roth, 2018; Katan, 2018). There are over 13.7 million new strokes worldwide each year. One in 4 people over age 25 will have a stroke in their lifetime. Over 5 million people die each year due to stroke and over 116 million cumulative years of healthy life is lost each year due to stroke-related death and disabilities. There are approximately 67.5 million people currently living who have experienced at least 1 ischemic stroke (WSO, 2019). Ischemic stroke involves a blockage of a blood vessel(s) in the brain, resulting in a lack of oxygen and nutrients to the underlying parenchymal tissue and subsequent cell and tissue death (Roger, 2011).

While there can be variability among ischemic stroke victims, the pathology is generally described by a number of defined stages. During the ischemic stage, which may last hours, a clot blocks a vessel, causing loss or reduction of blood flow to cells and tissue downstream of the infarct. This initiates a chain of cellular and inflammatory responses. Within hours of the ischemic event, there is an acute cellular response whereby a complex series of cellular metabolic events can lead to loss of neuronal function and cell death. These cellular responses affect the core ischemic zone and can have impact on the tissues surrounding the infarct. Following ischemia, inflammatory cytokines are up-regulated initiating a multi-stage inflammatory response to the stroke. Neutrophils infiltrate the ischemic area secreting additional inflammatory mediators resulting in the destruction of necrotic and neighboring viable tissue. Macrophages and astrocytes are activated stimulating further inflammatory factors, while activated glial cells isolate the area of damage. Reperfusion of the occluded vessel, whether through intervention, spontaneous resolution, or compensation of collateral circulation, can result in a reinforcing cycle/cascade of inflammatory response and damage. The acute inflammatory process remains active for days and may reach its completion within 1-2 weeks after the initial ischemic event.

Current therapy for stroke is limited. Other than one recombinant protein therapy, recombinant tPA, which is directed at the dissolution of the clot in affected blood vessels in adults following stroke, there are no drugs or biologic therapies available for effectively treating ischemic stroke or the damage associated with the ischemic event approved throughout the world. Three additional small molecule drugs, edaravone, a free radical scavenger, ozagrel, a thromboxane A2 inhibitor, and argatroban, a direct thrombin inhibitor, have been approved for stroke treatment only in Japan. Small molecule therapies such as anti-platelet drugs, anti-coagulants, and statins act as prophylactics and have no immediate benefit following an acute attack. Only 5% to 10% of Americans suffering ischemic stroke receive tPA due to delayed recognition of the symptoms coupled with the limited window for receiving treatment following the stroke. Patients often arrive at hospitals too late for such intervention. The numbers of affected

individuals, the costs necessary to facilitate their care and rehabilitation, coupled with the lack of current therapies reiterate that stroke represents a significant unmet medical need.

Drug development efforts to date have focused on stroke prevention (eg, anti-hypertensives, cholesterol-lowering drugs, and anti-coagulants), thrombolytics, and neuroprotectants. Most recently, results from preclinical and clinical studies suggest the potential for stem cell therapy as a treatment for ischemic stroke.

3.1.3.1. Stem Cell Treatment in Ischemic Stroke

Stem cells have demonstrated efficacy when transplanted into animal models of stroke, although the mechanisms through which cells provide benefit in these studies has not been definitively established.

Direct transplantation experiments in the brain have utilized cells derived from bone marrow. Fresh bone marrow transplanted directly into the ischemic boundary zone of rat brain improved functional recovery from middle cerebral artery occlusion (MCAO) (Chen, 2000). Similarly, MSC implanted into the striatum of mice after stroke improved functional recovery (Li, 2000). Cerebral grafts of mouse bone marrow also facilitated restoration of cerebral blood flow and blood-brain barrier after stroke in rats (Borlongan, 2004).

Systemic administration of stem cells via IV or intra-arterial injection has also been shown to have positive effects in animal models of stroke. Intra-carotid artery administration of MSC following MCAO in rats improved functional outcome (Li, 2001). Similarly, IV administration of umbilical cord blood stem cells ameliorated motor and neurological deficits after stroke in rats (Chen, 2001). It has also been reported that IV administration of cord blood was more effective than intra-striatal administration in producing functional benefit following stroke in rats (Willing, 2003). IV administration of MSC has been found to induce angiogenesis in the ischemic boundary zone following stroke in rats (Chen et al, 2003). Collectively, these studies illustrate the diversity of cell types, routes of delivery, and potential mechanisms of benefit which may contribute to recovery in animal models of stroke.

MultiStem has been shown to improve motor function when transplanted directly into the brain of adult rats (Zhao, 2002) or into the hippocampus of neonatal rats following induction of hypoxic-ischemic injury (Yasuhara, 2008; Yasuhara, 2006a; Yasuhara, 2006b). In addition, allogeneic and human MultiStem have shown sustained, statistically significant, dose-dependent benefit when administered IV (without immunosuppressive agents) in rat models of ischemic stroke (Mays, 2010; Yang, 2017).

These data suggest that MultiStem can provide therapeutic benefit through multiple potential biological mechanisms, including preventing apoptosis, limiting inflammatory damage in the brain, and producing homing factors that may mobilize endogenous stem cells or progenitors thereby further improving CNS function and recovery. Based on these supportive data, the effects of MultiStem are being investigated in patients who have sustained an ischemic stroke.

3.1.4. Ulcerative Colitis

Ulcerative colitis is a chronic, relapsing inflammatory bowel disease involving all or a portion of the colon. This disease is the most common form of inflammatory bowel disease worldwide with an estimated incidence of 1.2 to 20.3 cases per 100,000 person-years and a prevalence of 3.6 to 214.0 cases per 100,000 per year (Danese and Fiocchi, 2011). Patients with UC most commonly present with diarrhea, urgency, rectal bleeding, and abdominal pain. Patients may also experience fatigue, fevers, weight loss, and dehydration. The symptoms can be incapacitating. The sub-mucosa of the colon becomes progressively dominated by lymphocytic infiltration causing further damage. Many patients will typically suffer a "flare" of disease activity at least once per year followed by a period of remission (Loftus, 2004).

The primary goal of therapy for UC is to induce and maintain remission. Treatment has traditionally involved a step-wise approach of medical therapy. 5-Aminosalicylic acid (5-ASA) agents are the cornerstone of therapy for mild to moderately active UC and are usually used as the first-line therapy

(Kornbluth, 1997). Although they are generally safe and well tolerated, they only induce remission in approximately 50% of patients (Sutherland, 1993). When UC is refractory to 5-ASA agents, corticosteroids are usually the second-line therapy (Kornbluth et al, 1997). However, these medications are associated with numerous side effects, and many patients develop steroid dependency. Therefore, corticosteroids are used only for inducing remission and are not recommended as maintenance therapy. Alternative therapies to corticosteroids for refractory disease include immunosuppressants such as azathioprine (AZA) or 6-mercaptopurine (6-MP), or in severe cases, cyclosporine (Kornbluth et al, 1997). AZA and 6-MP are most effective in steroid sparing and maintaining remission in UC. They may also have a role in the induction of remission, although the time required before a therapeutic response can be prolonged to 2 to 6 months (Sandborn, 1998). Side effects of these therapies include pancreatitis, infection, myelosuppression, hepatotoxicity, and lymphoma. The most recent advance in medical therapy for UC is the introduction of biological therapy. Infliximab, an anti-tumor necrosis factor (TNF) therapy, induces response following a three-dose induction regimen in approximately two-thirds of patients with moderately to severely active UC, and approximately one third of patients will be in clinical remission at 1 year (Rutgeerts, 2005). As with other immunosuppressants, anti-TNF therapies are associated with a significant risk of side effects, including acute and delayed infusion reactions, development of autoantibodies, infections, lymphoma, neurological disease, and hepatotoxicity.

3.1.4.1. Stem Cell Treatment in Ulcerative Colitis

MultiStem represents a mechanistically novel approach for the treatment of UC that has the potential to provide efficacy with a reduced side-effect profile compared to existing treatments. MultiStem appears capable of delivering a therapeutic benefit through more than one mechanism of action. Factors expressed by MultiStem are believed to reduce inflammation and regulate immune system function, protect damaged or injured cells and tissue, promote formation of new blood vessels, and augment tissue repair and healing in other ways (Auletta, 2010). MultiStem has been used in adoptive T-cell models of GvHD in rats and mice that involve intestinal injury with disease pathology akin to that occurring in UC. MultiStem reverses this disease pathology by tempering the inflammatory response and intestinal organ damage via mechanisms that include down regulation of TNF-α cytokine networks and effector T-cells. MultiStem provided survival benefit in lethal acute GvHD models in part by reducing intestinal pathology while preserving intestinal function which allows the animals to regain weight. Thus, the nonclinical studies conducted in GvHD rodent models support the clinical investigation of MultiStem in UC patients.

3.1.5. Solid Organ Transplant

End-stage organ failure is a public health concern with few treatment alternatives, with transplantation often being the best option. The field of SOT has advanced over the 4 last decades with significant developments in the fields of surgery, immunology, drug development, and general standards of care. Over 1 million patients worldwide have undergone successful organ transplantation. In the United States and Europe, the most commonly transplanted organs are kidneys, livers, hearts, lungs, and pancreases (Bloom, 2005; EMA, 2008). Organ graft rejection continues to be an issue with SOT, and, as the demand for organs continues to exceed the supply, the acceptance of extended criteria donors continues with increased risk of unfavorable transplantation outcomes (EMA, 2008).

The goal of immunosuppression in SOT is to control an undesirable immune response while avoiding the complications of immunodeficiency, especially increased risk of infection and malignancy. The use of immunosuppressants has drastically improved short-term patient and graft survival; however, long-term graft survival has not much improved (van Sandwijk, 2013). Current immunosuppressive therapies include glucocorticosteroids; calcineurin inhibitors (CNIs), mammalian target of rapamycin (mTOR) inhibitors, antimetabolites, and antibodies. These immunosuppressants are associated with often severe side effects requiring routine therapeutic drug monitoring due to their narrow therapeutic indices (Johnston, 2013; van Sandwijk et al, 2013). More recent developments aim to promote tolerance through induction or infusion of regulatory T-cells, specialized leukocyte populations that are either selected to

have regulatory function during their development or acquire immunosuppressive properties in the local microenvironment of the allograft (van Sandwijk, 2013).

The first solid organ transplant indication for MultiStem was liver transplantation. End-stage liver disease can be caused by a variety of hepatotoxic agents (hepatotrophic viruses, alcohol, hepatotoxins), autoimmune processes (autoimmune hepatitis, primary biliary cirrhosis, viral hepatitis), or metabolic defects (α-antitrypsin deficiency and many others). In patients in whom conservative medical management fails or is prone to failure, liver transplantation is the current gold standard of treatment for end-stage liver disease (Adam, 2000; Fulginiti, 1968).

The clinical success of liver transplantation is evident, with patient and graft survival rates exceeding 75% after 5 years (Lee, 2007; Northup, 2006). However, next to a continuous shortage of donor organs, the problem of life-long immunosuppression continues to be a major obstacle in transplant medicine, especially considering that further improvements in overall survival may be expected in the years to come. All transplant patients on pharmacological immunosuppression suffer from an increased risk of opportunistic infections, mainly in the early post-transplant period, and from a gradual increase in the incidence of malignant diseases and nonimmunological side effects in the later phases after transplantation (Vajdic and van Leeuwen, 2009; Watt, 2009).

3.1.5.1. Stem Cell Treatment in Solid Organ Transplant

One strategy to reduce the need for immunosuppressive pharmacotherapy in SOT is cell-based immunoregulation using stem cells. MSCs have emerged as promising candidates for cell-based immunomodulatory therapy promoting operational tolerance of solid organ transplantation in a variety of animal models (Ge, 2009; Casiraghi, 2008; Popp, 2008; Inoue, 2006; Bartholomew, 2002). In addition, MSCs possess regenerative potential and may thus participate in the regeneration of marginal organs after transplantation (Hematti, 2008). Thus, MSCs have been shown to be capable of inducing allograft acceptance in rodent models. So far, this phenomenon has only been shown for donor-derived (Ge et al, 2009; Popp et al, 2008; Bartholomew et al, 2002) or haploidentical (Casiraghi et al, 2008) MSCs. However, injecting donor-antigen–bearing cells in a clinical setting carries the risk of recipient sensitization (Dhanireddy, 2009; Flye, 1995). Also, outside the limited field of living related organ transplantation, making donor-type MSCs available for a clinical study is almost impossible. Recipient-derived cells, on the other hand, carry an increased risk of malignant transformation (Kidd, 2009; Worthley, 2009; Tolar, 2007), and their immunological benefit is unproven. Also, MSCs are characterized by limited in-culture expansion potential and are thus of limited clinical use.

Recent nonclinical studies have shown that MultiStem effectively prolongs allograft survival *in vivo*. Based on these results, it is believed that MultiStem can be a valuable addition to current immunosuppressive protocols and contribute to reducing the long-term pharmacological side effects and inducing operational transplant tolerance (Orlando, 2009). The multipotent and immunologic properties of MultiStem could make it a useful adjunct in SOT by inducing immunologic tolerance or at least enabling the reduction of immunosuppressive drug therapies. Given the clinical need for improved anti-rejection and pro-regeneration treatment after liver transplantation, the risk-benefit equation is appropriate for evaluating MultiStem after allogeneic liver transplantation.

3.1.6. Acute Respiratory Distress Syndrome

ARDS is a common clinical entity and a major cause of morbidity and mortality in the critical care setting. Historically, ARDS has been associated with mortality ranging from 25% to 40%, with worse outcomes in the elderly population (Walkey, 2012). According to the NHLBI in 2013 and the ARDS foundation, the annual incidence of ARDS is 190,000 in the US. The NHLBI estimated that in 2009 the annual cost of providing healthcare related to all respiratory conditions, excluding lung cancer, was $113 billion.

ARDS is defined as the sudden failure of the respiratory system and can occur in anyone who is critically ill. This condition can be life-threatening because normal gas exchange does not take place due to severe fluid buildup in both lungs. ARDS is caused mainly by extensive lung inflammation and small blood vessel injury due to sepsis, trauma and/or severe pulmonary infection such as pneumonia. Onset of disease typically occurs within 24-72 hours of the original illness or injury.

3.1.7. Stem Cell Treatment in Acute Respiratory Distress Syndrome

MultiStem, is a cell-based, biological therapy under development for the treatment of ARDS. There is currently no effective drug treatment for ARDS, and this project is intended to demonstrate that an allogeneic cell therapy has high potential for treating the critical and highly expensive ARDS – which has not been achieved to date. MultiStem therapy has been shown to be safe and effective in other indications, and its multifactorial immune modulation capacity suggests real potential in ARDS, making this innovation achievable.

MultiStem is believed to reduce inflammation and regulate immune system function, protect damaged or injured cells and tissue, promote formation of new blood vessels, and augment tissue repair and healing in other ways (Auletta, 2010). MultiStem demonstrates a dose dependent inhibition of allogeneic cell or antibody mediated T-cell proliferation *in vitro*. MultiStem is also capable of modulating immune cells that would be responding to viral infection which could ultimately lead to ARDS. For example, during viral infection, macrophages migrate to affected tissues, engulf infected cells, recruit other immune cells to injured tissues, secrete pro-inflammatory cytokines, act as a potential viral reservoir and clear debris. In multiple animal models of injury, MultiStem administration results in shifting macrophages from a pro- to an anti-inflammatory phenotype. This includes decreasing secretion of pro-inflammatory cytokines while simultaneously inducing an anti-inflammatory phenotype in macrophages.

In animal models, MultiStem cells have demonstrated an ability to reduce the severity of pulmonary distress, reduce alveolar edema and return lung endothelial permeability to normal. Intravenous MultiStem treatment early following the onset of the condition may ameliorate the initial hyper-inflammation and reduce the fibrotic activity that follows, thereby speeding the return to and improving the likelihood of more normal lung function and helping patient recovery. Administration of MultiStem has been shown to have meaningful benefit in multiple animal models of the inflammatory sequalae experienced by patients experiencing the impact of a systemic inflammatory response due to insult, trauma or other precipitating event or condition, including ARDS and Acute Kidney Injury (AKI) – a common comorbidity of ARDS.

3.1.8. Trauma

Trauma is the leading cause of death for individuals between the ages of 1 to 44 and the third leading cause of death in the US overall, accounting for approximately 180,000 fatalities each year, of which up to 20% are potentially preventable (NCHS, 2016). Seventy-five percent of traumatic deaths occur during the first 3 days after injury and are primarily due to uncontrolled bleeding and traumatic brain injury (TBI) (Tisherman, 2015; Holcomb, 2015; Holcomb, 2013). After 3 days, the remaining 25% of deaths occur at a low but steady rate and result from inflammation, blood vessel damage, and poor clotting associated with the initial injury, shock and resuscitation (Tisherman et al, 2015; Holcomb et al, 2015; Holcomb et al, 2013; Langan, 2014). These inflammatory-related complications include acute kidney injury (AKI), ARDS, venous thromboembolic disease, multiple organ failure, and swelling and tissue death after TBI (Langan et al, 2014). Preliminary data from the Pragmatic Randomized Optimal Platelet and Plasma Ratios (PROPPR) trial show that these inflammatory complications occur in 64% of patients who survive the initial injury, and nearly 70% of enrolled trauma patients in PROPPR suffered SIRS, which is associated with a range of serious complications following trauma. Current treatments for these inflammatory complications are supportive only, with no effective interventions (Holcomb, 2015).

3.1.9. Stem Cell Treatment in Trauma

Human and animal data support a beneficial role of early adult stem cell treatment in the prevention of ARDS, TBI secondary injury and AKI. The preponderance of animal data suggests that early (within hours of the injury) infusion of cellular treatment results in decreased organ injury, while delayed treatment does not.

Specifically, MultiStem administration reduced severity of disease, improved clearance of alveolar edema and returned lung endothelial permeability to normal in an ex vivo perfused swine lung model of ARDS (Rojas, unpublished data). In a sheep model of lipopolysaccharide (LPS) induced ARDS, vascular pressure was reduced, partial pressure arterial oxygen (PaO2) levels quickly returned to normal and pulmonary edema cleared in MultiStem-treated animals compared to control animals (Rojas, 2014, Cardenes, 2019).

In TBI and other acute central nervous system injury models, including ischemic stroke and spinal cord injury, early IV administration of MultiStem cells results in a decrease in inflammatory systemic immune system cells in and around the site of injury, decreased presence of inflammatory cytokines in the blood stream, and improved long-term locomotor and neurological outcomes in cell treated animals when compared to saline treated injured animals (Mays and Savitz, 2018).

Similarly, rodent studies have consistently demonstrated a protective effect of mesenchymal stem cells (MSCs), an adult cell type distinctive from MultiStem that share some common immunomodulatory biology, in both acute and chronic kidney injury models. In a systematic analysis of 21 of these studies, a consistent reduction in serum creatinine was observed in MSC-treated animals versus controls. Westenfelder and colleagues demonstrated that administration of MSCs to the kidneys of rats with ischemia/reperfusion-induced AKI ameliorated renal function, provided reno-protection, decreased apoptosis, and resulted in anti-inflammatory effects (Togel, 2005). Further work in this area revealed that, after 1-3 months, animals treated with MSCs had normal renal function, an absence of interstitial fibrosis, and low expression of pro-fibrotic genes like Plasminogen activator inhibitor (PAI)-1 compared to vehicle and fibroblast-treated control animals (Togel, 2009).

In addition to the rodent studies, the results with MultiStem from 2 large animal models of acute inflammation in the lungs and in the *ex vivo* human kidney ischemia/reperfusion injury models highlight the potential efficacy and safety of MultiStem for the treatment of patients following traumatic injury and critical care conditions and their related downstream inflammatory sequalae.

4. PHYSICAL, CHEMICAL, PHARMACEUTICAL PROPERTIES, & FORMULATION

MultiStem is a cell therapy medicinal product originating from adherent adult stem cells taken from the bone marrow of a non-related donor and expanded *ex vivo*.

Laboratory Code	MultiStem
Cell Type	Allogeneic
Tissue Source	Adherent stem cells taken from an adult human bone marrow aspirate

MAPC have been isolated from adult tissues including bone marrow and other nonembryonic sources, have the potential to express proteins indicative of cells representing each of the three germ layers and can be expanded *ex vivo* while retaining their multi-potent differentiation potential. MultiStem is a proprietary subset of the allogeneic bone marrow-derived stem cells. MultiStem refers to the registered (or, trademarked), clinical grade MAPC product. MultiStem cells are derived from a bone marrow aspirate acquired and tested in accordance with 21 CFR Part 1271 Human Cells, Tissues, and Cellular and Tissue Based Products (HCT/Ps) and in compliance with EU regulations on Cells and Tissues; 2004/23/EC, 2006/17/EC and 2006/86/EC.

The MultiStem product is a light, amber-colored homogenous sterile cell suspension composed of MultiStem cells and formulated in cryopreservation medium including Plasma-Lyte A or equivalent, dimethyl sulfoxide (DMSO) and human serum albumin (HSA). The MultiStem product is stored frozen in a container such as a bag or vial in the vapor phase of liquid nitrogen prior to preparation for administration to a subject and the number of containers will vary depending upon the dose, mode of administration, and clinical indication.

4.1. Acute Myocardial Infarction: Dosage Form and Delivery

Once the MultiStem product is thawed, contrast agent is added to each vial and the material filtered through a 41 μm filter prior to administration directly into the adventitia of the target coronary vessel via micro-infusion catheter. The final diluted product consists of Plasma-Lyte A, DMSO (4%), HSA (4%) and 20% contrast agent. Storage and transport of the thawed product is at room temperature using ambient gel packs until catheter loading.

The biocompatibility of MultiStem cells with the transarterial catheter and filter has been demonstrated *in vitro* (RM-071005-01FR). Infusion of human MultiStem through a transarterial catheter and filter does not affect cell recovery, viability, or potency.

4.2. Graft versus Host Disease: Dosage Form and Delivery

The MultiStem diluted product for infusion was prepared from the MultiStem drug product cryobags by diluting the cryobag contents with Plasma-Lyte A (1:1) and passed through a 200 μm blood filter prior to infusion. The final diluted product consisted of Plasma-Lyte A, DMSO (5%) and HSA (2.5%), to produce a final IV bag for patient administration with the targeted cell dose. Storage of the final diluted product was 2°C to 8°C prior to the infusion.

4.3. Stroke: Dosage Form and Delivery

The MultiStem diluted product for infusion in the MASTERS-1 trial was prepared from the MultiStem drug product cryobags by diluting the cryobag contents with Plasma-Lyte A or equivalent (1:1) and passed through a 200 μm blood filter prior to infusion. The final diluted product consisted of Plasma-Lyte A or equivalent, DMSO (5%) and HSA (2.5%), to produce a final IV bag for patient administration with the targeted cell dose. Storage of the final diluted product was 2°C to 8°C prior to the infusion.

The MultiStem diluted product for infusion in the TREASURE and MASTERS-2 trials is being prepared from the MultiStem drug product vials in 6- and 50-mL vial sizes, respectively. These vials are prepared

by diluting the vial contents with about 250 mL of Plasma-Lyte A or equivalent. The final diluted product is passed through a 200 µm blood filter prior to infusion. The final diluted product consists of Plasma-Lyte A or equivalent, DMSO (1%) and HSA (1%), to produce a final IV bag for patient administration with the targeted cell dose. Storage of the final diluted product is 2°C to 8°C prior to the infusion.

4.4. Ulcerative Colitis: Dosage Form and Delivery

The MultiStem diluted drug product for use in Ulcerative Colitis was prepared from the MultiStem drug product cryobags where the cells were concentrated, and a buffer exchange was performed to formulate the cells into Hypothermosol (HTS) that was the final buffer for the infused product. The infused product contained DMSO (~0.6%), HSA (~0.3) and HTS and was passed through a 200 µm blood filter prior to infusion. The final diluted MultiStem product was stored at 2°C to 8°C until infused.

4.5. Solid Organ Transplant: Dosage Form and Delivery

The MultiStem diluted product for infusion was prepared from the MultiStem drug product cryobags by diluting the cryobag contents with Plasma-Lyte A (1:1). The final diluted product consisted of Plasma-Lyte A or equivalent, DMSO (5%) and HSA (2.5%). The first dose was run through a 200 µm blood filter and administered through the portal vein after the new liver was transplanted and the patient was stable. The product was prepared just prior to administration. The second dose was initiated 24 hours after the first dose from frozen cryobags where the thawed bag was diluted 1:1 with Plasma-Lyte A, run through a 200 µm blood filter before it was delivered intravenously. The dilution and administration was performed at the bedside so storage of the product is at room temperature.

4.6. Acute Respiratory Distress Syndrome: Dosage Form and Delivery

The MultiStem diluted product for infusion during the US/United Kingdom B04-01 ARDS trial was prepared from the MultiStem drug product cryobags by diluting the cryobag contents with Plasma-Lyte A or equivalent (1:1) and passed through a 200 µm blood filter prior to infusion. The final diluted product consisted of Plasma-Lyte A or equivalent, DMSO (5%) and HSA (2.5%), to produce a final IV bag for patient administration with the targeted cell dose. Storage of the final diluted product is 2°C to 8°C prior to the infusion.

The MultiStem diluted product for infusion during the MACoVIA B04-03 trial in the US is prepared from the MultiStem drug product vials by diluting the vial contents with approximately 250 mL of Plasma-Lyte A or equivalent. The final diluted product is passed through a standard blood filter prior to infusion. The final diluted product consists of Plasma-Lyte A or equivalent, DMSO (5%) and HSA (2.5%), to produce a final IV bag for patient administration with the targeted cell dose. Storage of the final diluted product will be 2°C to 8°C prior to the infusion.

The MultiStem diluted product for infusion during the ARDS B04-02 ONE-BRIDGE trial in Japan is prepared from the MultiStem drug product vials by diluting the vial contents with approximately 250 mL of Plasma-Lyte A or equivalent. The final diluted product is passed through a 200 µm blood filter prior to infusion. The final diluted product consists of Plasma-Lyte A or equivalent, DMSO (1%) and HSA (1%), to produce a final IV bag for patient administration with the targeted cell dose. Storage of the final diluted product will be 2°C to 8°C prior to the infusion.

4.7. Trauma: Dosage Form and Delivery

The MultiStem diluted product for infusion in the Trauma B06-01 trial is being prepared from the MultiStem drug product vials by diluting the vial contents with approximately 250 mL of Plasma-Lyte A or equivalent. The final diluted product is passed through a 200 µm blood filter prior to infusion. The final diluted product consists of Plasma-Lyte A or equivalent, DMSO (1%) and HSA (1%), to produce a final IV bag for patient administration with the targeted cell dose. Storage of the final diluted product is 2°C to 8°C prior to the infusion.

5. NONCLINICAL STUDIES

The nonclinical pharmacology, biodistribution and persistence, immunogenicity, and safety profile of MAPC were investigated *in vitro* and/or *in vivo* in numerous species and injury models. Table 5-2 provides a summary overview of the studies, and the sections that follow provide a textual overview of the nonclinical MAPC studies.

Table 5-2. Overview Nonclinical Studies of MultiStem

Study Title Study Number/Reference	Test System	Number of Doses, Route of Administrations, Dosage, Test Article	Study Results	GLP Status/ Section Ref
***In Vitro* Studies**				
Immune-regulatory properties of MultiStem RM-051012-02FR1	*In vitro*	N/A (*In vitro*) rat MAPC	Rat MAPC suppressed ConA-mediated T-cell activation and proliferation in vitro as measured by ^{3}H-thymidine incorporation. Syngeneic and third-party MAPC suppressed T-cell activation by splenocytes from non-matched rats in a dose-dependent manner. ~50% inhibition was observed at low MAPC to T-cell ratios (1:50-1:100).	Non-GLP *In Vitro* Pharmacology
Immunosuppressive activity of Cambrex manufactured MultiStem lots in T-cell activation assays RM-070109-02FR1	*In vitro*	N/A (*In vitro*) human MAPC	There was consistent dose-dependent inhibition of T-cell activation by MAPC from different donors. Three lots of clinical lot-derived human MAPC all showed 50% inhibition at MAPC to PBMC ratios of 1:2 to 1:8 when investigated in two independent experiments.	Non-GLP *In Vitro* Pharmacology
Multipotent adult progenitor cells prevent macrophage-mediated axonal dieback and promote regrowth after spinal cord injury Busch et al, 2011	*In vitro* Rat/Sprague Dawley	N/A (*In vitro*) rat MAPC 1 Dose, Intraspinal, 2×10^{5} rat MAPC	MAPC secreted factors induced a greater than 5-fold increase in the relative ratio of Arg1 to iNos transcript in macrophages, demonstrating that MAPC are able to induce a shift from a pro-inflammatory M1 phenotype toward an anti-inflammatory M2 phenotype. When injected immediately after injury, rat MAPC was present at the transplantation site 2, 4, and 7 days but not 28 days later. This suggests that rat MAPC acts on injury sites for a short period of time, and the risk of residual cells contributing to ectopic tissue formation or aberrant physiology is minimized.	Non-GLP *In Vitro* Pharmacology Pharmacokinetics
Effect of multistem on endothelial cell activation and neutrophil adhesion RM-090113-01FR	*In vitro*	N/A (*In vitro*) human MAPC	Coculture of MAPC with endothelial cells prevented upregulation of E-selectin, V-CAM and to a lesser degree, I-CAM, to the cell surface of endothelial cells following activation with TNF-α or interleukin-1β. The reduction in cell surface adhesion molecule expression resulted in decreased neutrophil binding to endothelial cells.	Non-GLP *In Vitro* Pharmacology

Table 5-2. Overview Nonclinical Studies of MultiStem				
Study Title Study Number/Reference	**Test System**	**Number of Doses, Route of Administrations, Dosage, Test Article**	**Study Results**	**GLP Status/ Section Ref**
Clinical scale expanded adult pluripotent stem cells prevent graft-versus-host disease Kovacsovics-Bankowski et al, 2009	*In vitro*	N/A (*In vitro*) rat MAPC	Rat MAPC did not induce proliferative T-cell sensitization *in vitro*. When added to MLRs, third-party MAPC suppressed T-cell proliferation in a dose-dependent manner as measured by ^{3}H-thymidine incorporation. Inhibition was reversible upon removal of MAPC from culture, was mediated by soluble factors, and was non-MHC restricted.	Non-GLP *In Vitro* Pharmacology
Clinical grade MAPCs durably control pathogenic T-cell responses in human models of transplantation and autoimmunity Reading et al, 2013	*In vitro*	N/A (*In vitro*) human MAPC	MAPC suppressed alloreactive T-cell proliferation in a dose-dependent manner after 4 days in culture, including homeostatic T-cell expansion driven by antigen-driven and antigen-independent stimuli as measured by flow cytometry. MAPC inhibited effector cytokine secretion measured by intracellular flow cytometry. MAPC induced Treg in the presence of effector T-cell suppression. T-cell suppression was mediated by induction of indoleamine dehydroxygenase, as inhibitors of this pathway reversed T-cell suppression.	Non-GLP *In Vitro* Pharmacology
Acute Myocardial Infarction				
Direct delivery of syngeneic and allogeneic large-scale expanded multipotent adult progenitor cells improves cardiac function after myocardial infarct Van't Hof et al, 2007	*In vitro* Rat/Lewis	1 Dose, Myocardial injection, 1×10^{7} rat MAPC	Myocardial injection immediately after infarction with syngeneic or allogeneic MAPC resulted in an 88% and 55% increase in shortening fraction compared to controls, respectively, with no significant differences between the syngeneic and allogeneic groups. Immunostaining demonstrated significant engraftment of MAPC at 1 day after AMI and cell administration, with < 10% of either syngeneic or allogeneic cells remaining at 6 weeks. Myocardial MAPC injection after AMI resulted in increased vascular density within the infarct zone.	Non-GLP *In Vitro* Pharmacology

Table 5-2. Overview Nonclinical Studies of MultiStem

Study Title Study Number/Reference	Test System	Number of Doses, Route of Administrations, Dosage, Test Article	Study Results	GLP Status/ Section Ref
Bioenergetic and functional consequences of bone marrow derived multipotent progenitor cell transplantation in hearts with postinfarction LV remodeling Zeng et al, 2007	Pig/ Yorkshire	1 Dose, Myocardial injection, 5×10^7 pig MAPC	MAPC treatment immediately following infarction resulted in greater LV function, improved phosphocreatine/ATP ratio in the border zone of the infarct and increased phosphocreatine/ATP across the left ventricle. Approximately 0.3% to 0.5% cell engraftment was observed, with cells frequently found as vessel wall components. No difference in cell retention or cardiac performance was observed in the presence or absence of cyclosporine. MAPC was associated with increased vascular density in the infarct and infarct border zones, but not in the remote regions of the heart.	Non-GLP *In Vivo* Pharmacology
MultiStem delivered by transarterial catheter in a pig model of acute myocardial infarction (AMI) RM-070208-01FR2	Pig/ Yorkshire	1 Dose, Transarterial catheter, 5×10^7 pig MAPC	Animals treated with MAPC immediately following infarction showed significant improvement in the ejection fraction from 1 to 4 weeks (33% to 46%) after cell infusion. Four weeks post-infarction the control animals had significantly lower ejection fractions than the MAPC-treated animals (35% vs 46%).	Non-GLP *In Vivo* Pharmacology

Table 5-2. Overview Nonclinical Studies of MultiStem

Study Title Study Number/Reference	Test System	Number of Doses, Route of Administrations, Dosage, Test Article	Study Results	GLP Status/ Section Ref
Evaluation of safety and cell persistence following direct and catheter –based delivery of MultiStem in porcine model of acute myocardial infarction RM-051011-02FR2	Pig/ Yorkshire	Phase 1: 1 Dose, Myocardial injection, 5×10^7 pig MAPC Phase 2: 1 Dose, Transarterial or intracoronary catheter, 5×10^7 or 2×10^8 pig MAPC	Phase 1: Direct injection of MAPC immediately following infarction resulted in cell persistence of < 1% at 2 and 8 weeks. The range of cell persistence, based on the number of βgal positive cells in one ring from each animal, was 0% to 0.4%. However, the total cell persistence might be higher than what was observed in one ring. Phase 2: When Administered immediately after infarction, there was a trend of higher cell persistence by transarterial injection compared to intracoronary injection (not statistically significant). Irrespective of the dose, there was a trend of lower cell persistence in the 8-week animals compared to the 2-week animals. During both Phases 1 and 2, there were no test article-related effects on body weights, hematology, or clinical chemistry parameters. During Phase 1 (direct injection), 3 of 13 animals died of refractory ventricular fibrillation after the initial ligation of the LAD and prior to treatment with the test article. One animal was initially placed on study as an 8-week animal. This animal was found dead on Day 16 and was reported as a 2-week animal. Upon necropsy, death was considered to be related to infarct, not test article. All other Phase 1 animals survived to the scheduled necropsy. During Phase 2 (catheter-based injection), 8 of 22 animals died during the 60-minute occlusion of the LAD or at reperfusion of the LAD and prior to treatment with the test article. All other Phase 2 animals survived to the scheduled necropsy.	Non-GLP *In Vivo* Pharmacology Pharmacokinetics Toxicology

Table 5-2. Overview Nonclinical Studies of MultiStem				
Study Title Study Number/Reference	**Test System**	**Number of Doses, Route of Administrations, Dosage, Test Article**	**Study Results**	**GLP Status/ Section Ref**
A GLP study to evaluate safety and efficacy of allogenic pig MultiStem cells in an acute myocardial infarction model in pigs RM-060523-01FR3	Pig/ American Yorkshire-Landrace-Duroc	1 Dose, Transarterial catheter, 2×10^7 or 2×10^8 pig MAPC	Pig MAPC were administered 2 days after injury. Compared to the control or high-dose group, the low-dose group had higher ejection fraction, smaller increases in end systolic volume, and smaller wall motion scoring index in LAD-related territories. The smaller wall motion scoring index in the low-dose group becomes more significant when compared to control and high-dose scores when the analysis is limited to segments directly affected by the LAD occlusion. Administration of MAPC immediately after infarction was not associated with any adverse clinical observations, bodyweight changes, hematology or coagulation parameters, cardiac markers, alloantibody responses or macroscopic or microscopic findings. Mild changes in clinical chemistry values were evident and were considered to be related to the infarct procedure and tended to resolve between 30- and 90-days post-injections. MAPC cells did not affect cardiac biomarkers. Increases in Troponin I and Creatine Kinase-MB were noted in all groups and were considered to be a result of the infarct procedure. Increases in total Creatine Kinase noted on Day 2 at 6 hours post-injection were considered to be a result of the surgical procedure.	GLP *In Vivo* Pharmacology Toxicology
Graft versus Host Disease				
Evaluation of the role of MultiStem in syngeneic bone marrow engraftment post-lethal irradiation RM-041006-01FR1	Rat/Lewis	1 Dose, IV, 5×10^6 rat MAPC	Administration of syngeneic MAPC cells 24 hours following lethal irradiation and infusion of syngeneic bone marrow cells increased survival rate of Lewis rats by 3 days. However, no long-term benefit was observed, as all rats died by Day 19.	Non-GLP *In Vivo* Pharmacology

Table 5-2. Overview Nonclinical Studies of MultiStem				
Study Title Study Number/Reference	**Test System**	**Number of Doses, Route of Administrations, Dosage, Test Article**	**Study Results**	**GLP Status/ Section Ref**
Evaluation of MultiStem benefit in a rat (P to F1) acute GvHD model RM-060401-01FR1	Rat/Lewis x Buffalo F1	1 or 2 Doses, IV, 2.5×10^6 rat MAPC	In both MAPC treated groups (single dose at Day 1 or 2 doses on Days 1 and 8), a survival advantage was seen and Log-rank survival analysis showed that the survival profile in the group that received two MAPC doses was significantly different from the PBS control group.	Non-GLP *In Vivo* Pharmacology
Stroke				
Phase 3 neonatal rat hypoxic-ischemic injury study: MultiStem IV dose escalation RM-060914-02FR1	Rat/Sprague Dawley	1 Dose, IV, 1×10^4 to 1×10^6 rat MAPC	MAPC were administered IV 7 days after hypoxic-ischemic injury in neonatal rats. Statistically significant sustained recovery in locomotor function was observed at ≥ 0.1 million cells/animal (0.5 million cells/kg) that persisted for ≥ 1-year post-dose. Improvements in viable endogenous neurons in the hippocampus were also detected in the brains of injured animals treated with rat MAPC. There were no MAPC-related clinical observations or changes in body weight, gross pathology, or clinical pathology parameters. There was a small increase in spleen weights of MAPC treated animals as compared to control animals, but the mean spleen weights were within the historical control range and there were no correlating macroscopic findings.	Non-GLP/GLP *In Vivo* Pharmacology Toxicology
Preclinical optimization of immunosuppression, route of administration and window of therapeutic benefit in a rat model of ischemic stroke RM-050903-01FR1	Rat/Sprague Dawley	1 Dose, IV, 1×10^6 human MAPC 1 Dose, Intra-cerebral injection, 4×10^5 rat or human MAPC	MAPC were administered intracranially 7 days post-injury, or administered IV 1-, 2-, or 7-days post-injury. A single IV dose of 1 million human MAPC cells (3 million cells/kg) in rats with ischemic stroke injuries administered any time up to 7 days post-injury resulted in statistically significant improvements in both locomotor and neurological test results out to 56 days post-stroke. The improvement was statistically significant with cyclosporine A or with vehicle. There were no observations of tumor or ectopic tissue formation. Evaluation of endogenous neuronal survival indicated that earlier administration of MAPC cells results in statistically significant protection of endogenous neurons.	Non-GLP *In Vivo* Pharmacology

Table 5-2. Overview Nonclinical Studies of MultiStem

Study Title Study Number/Reference	Test System	Number of Doses, Route of Administrations, Dosage, Test Article	Study Results	GLP Status/ Section Ref
Phase 3 rat pre-clinical stroke study: IV dose escalation of human MultiStem RM-060914-03FR5	Rat/Sprague Dawley	1 Dose, IV, 4×10^5 to 2×10^7 human MAPC	MAPC were delivered 2 days after induction of stroke. Statistically significant recovery of locomotor and neurological function was observed from 2 to 12 weeks post-MAPC cell injection, respectively, at doses ≥ 1 million cells. The dose effects reached a plateau at 4 million cells/animal (~12 million/kg). There were no test article-related clinical observations or alterations in hematology, coagulation or serum chemistry parameters, final body weight, organ weights or gross pathology. Microscopic evaluation of multiple tissues from animals in the nonviable cell control, 1 million and 20 million MAPC treatment groups revealed no abnormal tissue or findings.	Non-GLP/GLP *In Vivo* Pharmacology Toxicology
Transplantation of multistem in a rat occlusion model for stroke: proof-of-concept study RM-040916-01FR	Rat/Sprague Dawley	1 Dose, Direct striatal injection, 1, 2, or 4×10^5 human MAPC	Significant improvements in locomotor and neurological tests were observed when compared to vehicle-treated animals. There was a dose-dependent improvement in animals receiving 0.2 and 0.4 million cells, showing statistically significant improvement as early as 7 days post-MAPC injection. No ectopic tissue was observed in the brain at the gross level.	Non-GLP *In Vivo* Pharmacology
Transplantation of MultiStem in an MCA ligation rodent stroke model: proof-of-concept study RM-050822-03FR	Rat/Sprague Dawley	1 Dose, Direct striatal injection, 1, 2, or 4×10^5 human MAPC	MAPC were administered 7-days post-injury. Statistically significant improvements in locomotor and neurological tests were observed when compared to vehicle-treated animals at 14 days post-MAPC dose. There was a trend towards increasing improvement at increasing cell dose. Histological evaluation suggested graft cells transplanted into the striatum migrated to the site of injury in the cortex. No ectopic tissue was observed in any animal.	Non-GLP *In Vivo* Pharmacology

Table 5-2. Overview Nonclinical Studies of MultiStem

Study Title Study Number/Reference	Test System	Number of Doses, Route of Administrations, Dosage, Test Article	Study Results	GLP Status/ Section Ref
Multipotent adult progenitor cells enhance recovery after stroke by modulating the immune response from the spleen Yang et al, 2017	Rat/Long-Evans	1 Dose, IV, 1.2×10^7 /kg human MAPC	MAPC treatment administered within 24 hours in a rat middle cerebral artery occlusion (MCAo) model of ischemic stroke resulted in improved recovery as measured by a composite behavioral score consisting of motor, sensory and balance tests. There was an associated reduction in brain tissue loss. QTracker labeled MAPC was more abundant in the lungs and spleen 1 day after administration as compared with sham operated rats. There were notable decreases in the lungs and spleen on Day 3 when compared with Day 1. On both of Days 1 and 3, MAPC was present in the liver but not in the brain, kidneys, or thymus. MAPC administration significantly reduced the number of activated T-cells in the blood and decreased the genetic signature of T-cells in the brain of stroke injured animals.	Non-GLP *In Vivo* Pharmacology Pharmacokinetics
Intravenous multipotent adult progenitor cell therapy for traumatic brain injury: Preserving the blood brain barrier via an interaction with splenocytes Walker et al, 2010	Rat/Sprague Dawley	2 Doses, IV, 2 or 10×10^6 human MAPC	Two doses of human MAPC labeled with quantum dots were intravenously administered 2 and 24 hours after TBI induction, and 6 hours after the second administration, the spleen was isolated and the amount of MAPC in the spleen was evaluated. A dose-dependent localization was observed. MAPC were present within the white pulp in close approximation with the blood vessels, suggesting interaction with resident splenic lymphocytes.	Non-GLP *In Vivo* Pharmacology Pharmacokinetics
Intravenous multipotent adult progenitor cell therapy attenuates activated microglial/macrophage response and improves spatial learning after traumatic brain injury Bedi et al, 2013	Rat/Sprague Dawley	2 Doses, IV, 2 or 10×10^6 /kg human MAPC	IV MAPC administration at 2 and 24 hours after traumatic brain injury significantly reduced the number of activated microglia and macrophages in the dentate gyrus of the ipsilateral hippocampus. The number of microglia and macrophages in the dentate gyrus of the untreated hippocampus was not different among groups.	Non-GLP *In Vivo* Pharmacology

Table 5-2. Overview Nonclinical Studies of MultiStem				
Study Title Study Number/Reference	**Test System**	**Number of Doses, Route of Administrations, Dosage, Test Article**	**Study Results**	**GLP Status/ Section Ref**
Intravenous multipotent adult progenitor cell treatment decreases inflammation leading to functional recovery following spinal cord injury DePaul et al, 2015	Rat/Sprague Dawley	1 Dose, IV, 1, 4, or 6×10^5, or 1, 4, or 8×10^6 human MAPC	Administration of MAPC one day after spinal cord injury induced the repair of gray matter of the spinal cord as assessed by eriochrome cyanine (EC) staining. Neuroprotection by MAPC induces dose-dependent and statistically significant improvement of neurological and locomotor functions after SCI. Qdot labeled tracking demonstrated that MAPC mainly localized in peripheral organs including the spleen, but were rapidly cleared 48 hours after administration	Non-GLP *In Vivo* Pharmacology Pharmacokinetics
Solid Organ Transplant				
Heart grafts tolerized through third-party multipotent adult progenitor cells can be retransplanted to hosts with no immunosuppression Eggenhofer et al, 2013	*In vitro* Rat/ Lewis (MHC haplotype: $RT1^l$) or ACI (MHC haplotype: $RT1^a$)	N/A (*In vitro*) rat MAPC 1 Dose, IV, 2, 4, or 10×10^6 rat MAPC 2 Doses, intrasplenic, 5×10^6 rat MAPC	In a mixed lymphocyte reaction, stimulator-type and third party MAPCs dose-dependently inhibit T-cell proliferation upon allogeneic stimulation as measured by flow cytometry. *Results with donor-type MAPC:* IV injection of a single dose of donor-type MAPC on Day -4 increased graft survival, but not in a clearly dose-dependent manner. Intrasplenic administration of 5 million rat MAPC on both Day -4 and Day 0, followed by MPA therapy, resulted in 80% heart graft survival, compared to 0% in the control and MPA therapy alone groups. *Results with third-party MAPC:* Intrasplenic administration of 5 million rat MAPC on both Day -4 and Day 0, followed by MPA therapy, resulted in almost 60% heart graft survival, compared to 0% in the control and MPA therapy alone groups.	Non-GLP *In Vitro* Pharmacology *In vivo* Pharmacology

Table 5-2. Overview Nonclinical Studies of MultiStem

Study Title Study Number/Reference	Test System	Number of Doses, Route of Administrations, Dosage, Test Article	Study Results	GLP Status/ Section Ref
Acute Respiratory Distress Syndrome				
Human adult bone marrow-derived stem cells decrease severity of lipopolysaccharide-induced acute respiratory distress syndrome in sheep Rojas et al, 2014	Sheep/ Dorsett Cross	1 Dose, Intrabronchial administration, 4, 10, or 40×10^6 human MAPC	After administration of endotoxin, there was a rapid decline in oxygenation to hypoxemic values, indicative of severe-to-moderate ARDS. Animals were administered MAPC or saline 30 minutes after receiving endotoxin. None of the animals treated with saline solution recovered to normal baseline values during the 6 hours that the animals were followed. In contrast, blood oxygen levels in sheep treated with a dose of 40 million MAPC returned to baseline two hours after the cells were infused. Similarly, improvements in carbon dioxide (CO2) clearance, pulmonary vascular pressures and inflammation were observed and confirmed by histology and by the decrease in lung edema.	Non-GLP *In Vivo* Pharmacology
Cell therapy for ARDS: efficacy of endobronchial versus intravenous administration and biodistribution of MAPCs in a large animal model Cardenes et al, 2019	Sheep/ Dorsett Cross	1 Dose, IV, 1×10^7 human MAPC/kg 1 Dose, Endobronchial, 1×10^6 human MAPC/kg	MAPC were delivered 1 hour after lipopolysaccharide infusion in a sheep model of ARDS. The results demonstrate the safety, efficacy and equivalence of MAPC cells when delivered via IV or endobronchial administration into the lung, improving oxygenation 2 hours after onset of LPS-induced ARDS. To examine biodistribution MAPC cells were labelled with [18F] fluoro-29-deoxy-D-glucose delivered endobronchially or IV. PET/CT images were acquired at 1 and 5 hours of administration. MAPC delivered endobronchially remained at the site of administration and no changes were observed between 1 and 5 hours, whereas with the IV route, the cells had a broad biodistribution to different organs with the lung being the main organ of retention at 1 and 5 hours.	Non-GLP *In Vivo* Pharmacology Pharmacokinetics

Table 5-2. Overview Nonclinical Studies of MultiStem				
Study Title Study Number/Reference	**Test System**	**Number of Doses, Route of Administrations, Dosage, Test Article**	**Study Results**	**GLP Status/ Section Ref**
Multipotent adult progenitor cells decrease cold ischemic injury in *ex vivo* perfused human lungs: an initial pilot and feasibility study La Francesca et al, 2014	*Ex vivo* Human lung	1 Dose, Bronchoscopic installation, 1×10^7 human MAPC	MAPC were bronchoscopically instilled in the lower left lobe of donor lungs, which were then perfused and mechanically ventilated for 4h. All left lower lobes consistently demonstrated a significant decrease in histologic and BALF inflammation compared to vehicle-treated right lower lobes. These studies suggest that use of non-HLA-matched allogeneic MAPC during donor lung processing can decrease markers of cold ischemia-induced lung injury.	Non-GLP *In Vivo* Pharmacology
Trauma				
Therapeutic time window of multipotent adult progenitor therapy after traumatic brain injury Bedi et al, 2018	Rat/Sprague Dawley	2 Doses, IV, 1×10^7 human MAPC/kg	A rat model of TBI was created with moderate to moderately severe controlled cortical impact to the temporoparietal lobe of the brain by impactor. Animals received 2 doses of 10 million cells per kg administered intravenously, 2 and 24 hours, as well as 6 and 24 hours, 12 and 36 hours, and 36 and 72 hours after the injury. The results demonstrate the safety and efficacy of MultiStem cells in providing long term significant locomotor and neurological recovery of the cell treated vs saline treated animals, via modulation of microglial activation. Early dosing windows were superior as only the animals first dosed at 2 or 6 hours had significant neurological recovery compared to animals first dosed at 12 or 36 hours.	Non-GLP *In Vivo* Pharmacology
Novel delivery of cellular therapy to reduce ischemia reperfusion injury in kidney transplantation Thompson et al, 2020	*Ex vivo* human kidneys	1 Dose, via cannulated renal artery, 5×10^7 human MAPC	Reperfusion injury in this model represents the type of injury that would be observed in kidneys, following blood volume stabilization after hemorrhagic trauma. Acute Kidney Injury is the predominant post-trauma inflammation-related organ injury. The results demonstrate the safety and efficacy of MultiStem cells in reducing inflammatory biomarkers including IL-6 and markers of acute kidney injury (NGAL), while simultaneously increasing kidney function (urine output) and organ perfusion.	Non-GLP *In Vivo* Pharmacology

Table 5-2. Overview Nonclinical Studies of MultiStem

Study Title Study Number/Reference	Test System	Number of Doses, Route of Administrations, Dosage, Test Article	Study Results	GLP Status/ Section Ref
Safety Pharmacology				
Safety of repeated administration of allogeneic multistem in rats RM-060401-03FR1	Rat/Buffalo	1 Dose, IV, 2.5×10^6 rat MAPC 5 Doses, IV, 2.5×10^6 rat MAPC	Animals received a single IV dose, or multiple IV doses of vehicle or rat MAPC once a week for 5 weeks starting at 2 days after irradiation. There was no MAPC-related morbidity or mortality. There were no biologically or toxicologically important MAPC-treatment related changes in clinical observations, clinical chemistry parameters or histopathology as compared to PBS-treated control animals. There was no evidence of respiratory distress upon infusion. No major differences were observed in the RR of animals receiving rat allogeneic MAPC cells after single or multiple doses as compared to control animals. There were no consistent treatment-related changes in immunogenicity as measured by alloantibody formation or allo-T-cell sensitization. The NOAEL was 2.5 million cells/dose (12.5 million cells/kg).	Non-GLP Safety Pharmacology Toxicology
Pharmacodynamic Drug Interactions				
Sensitivity of MultiStem to drugs used in the AMI clinical trial protocol RM-070822-01FR1	*In Vitro*	N/A (*in vitro*) human MAPC	Exposure to the drugs aspirin, N-Acetylcysteine, heparin, paclitaxel, metroprolol, clopidogrel, captopril, eptifibatide, atorvastatin, and Abciximab at expected plasma concentrations did not affect MAPC viability or growth. Exposure to peak plasma and 10-fold under peak plasma levels of rapamycin decreased cell proliferation but did not affect cell viability.	Non-GLP Drug Interactions: AMI/Stroke/ARDS
Sensitivity of MultiStem to drugs used in the HM clinical trial protocol RM-060615-01FR	*In Vitro*	N/A (*in vitro*) human MAPC	Exposure to the drugs cyclosporine A, methotrexate, and tacrolimus at physiological dose levels did not affect MAPC viability or plating efficiency.	Non-GLP Drug Interactions: GvHD

Table 5-2. Overview Nonclinical Studies of MultiStem

Study Title Study Number/Reference	Test System	Number of Doses, Route of Administrations, Dosage, Test Article	Study Results	GLP Status/ Section Ref
Evaluation of the drugs used in the HM clinical trial on the immunosuppressive activity of MultiStem RM-070301-01FR	*In Vitro*	N/A (*in vitro*) human MAPC	Exposure to the drugs cyclosporine A, methotrexate, and tacrolimus at physiological dose levels did not affect MAPC immunomodulatory activity in T-cell proliferation assays.	Non-GLP Drug Interactions: GvHD
Sensitivity of MultiStem to drugs Used in the FA clinical trial protocol RM-060301-09FR	*In Vitro*	N/A (*in vitro*) human MAPC	Exposure to the drugs antithymocyte globulin, hydrocortisone, diphenhydramine, cyclosporine A, Filgrastim, and methylprednisolone at expected plasma concentrations did not affect MAPC viability.	Non-GLP Drug Interactions: GvHD/ARDS
In Vitro cytotoxicity study of argatroban and ozagrel hydrochloride on MAPC RM-FR-015-16	*In Vitro*	N/A (*in vitro*) human MAPC	Exposure to the drugs argatroban and ozagrel did not have cytotoxic effects on MAPC viability in a concentration range comparable to clinical usage. Functional tests of MAPC were also unaffected by drug exposure.	Non-GLP Drug Interactions: Stroke
In vitro assessment of MultiStem viability and activity in presence of drugs prescribed as standard of care for inflammatory bowel disease RM-100329-01FR	*In Vitro*	N/A (*in vitro*) human MAPC	Exposure to the drugs budesonide, 5-aminosalicylic acid, and Humira at peak plasma concentrations did not affect MAPC viability or proliferation. Exposure to the drugs 6-mercaptopurine and azathioprine at peak plasma concentrations did not affect MAPC viability but did inhibit proliferation. Exposure to the drugs budesonide, Humira, 6-mercaptopurine, and azathioprine at peak plasma concentrations, or greater, displayed similar immunomodulatory activity in a T-cell proliferation assay, while 5-aminosalicylic acid at peak plasma concentration decreased, but did not abrogate, the ability of MAPC to modulate T-cell proliferation.	Non-GLP Drug Interactions: Ulcerative Colitis/ARDS

Table 5-2. Overview Nonclinical Studies of MultiStem				
Study Title Study Number/Reference	**Test System**	**Number of Doses, Route of Administrations, Dosage, Test Article**	**Study Results**	**GLP Status/ Section Ref**
Biocompatibility of coated central line catheters and MultiStem RM-FR-023-15	*In Vitro*	N/A (*in vitro*) human MAPC	Clinical MultiStem product released for research use only (MAPC) was passed through uncoated and coated catheters, after which viability, plating efficiency, and activity in a T-cell suppression assay were determined. There were no differences in results between catheter types.	Non-GLP Drug Interactions: ARDS
Sensitivity of MultiStem to drugs used in the ARDS clinical trial protocol RM-141203-01FR	*In Vitro*	N/A (*in vitro*) human MAPC	Exposure to the drugs zosyn, cefepime, vancomycin, fentanyl, versed, cistracurium, omerprazole, and vasopressin approximately at or greater than cMax levels did not negatively affect MAPC viability or cytokine production.	Non-GLP Drug Interactions: ARDS
Study of the in vitro effects of epinephrine, ketamine, midazolam and propofol on MAPC RM-FR-032-18	*In Vitro*	N/A (*in vitro*) human MAPC	Exposure to the drugs epinephrine, ketamine, midazolam, and propofol at cMax concentrations did not inhibit MAPC cell growth or viability. Functional tests of MAPC were also unaffected by drug exposure at this concentration.	Non-GLP Drug Interactions: Trauma
Pharmacokinetics				
Biodistribution and mechanisms of benefit of human MultiStem in the prevention of experimental graft versus host disease in mouse models RM-080714-01FR	Mouse/ B6D2F1(H-2^{bxd})	1 Dose, IV, 1×10^6 rat MAPC	Luciferase-expressing rat MAPC was administered 1 and 4 days after the preparation of the mouse GvHD model, and the biodistribution of rat MAPC was evaluated over time by *in vivo* imaging. After the first administration, rat MAPC promptly accumulated in the lungs, and it was localized in the gastrointestinal tract 24 to 48 hours after administration. Six days after the second administration, bioluminescence signals mostly disappeared, suggesting that most of administered cells are eliminated during this period.	Non-GLP Pharmacokinetics
Biodistribution and homing of MultiStem in a nude mouse model of acute myocardial infarction RM-080603-01FR	Mouse/ Nude mouse	1 Dose, IV, 5×10^5 rat MAPC	Luciferase-expressing rat MAPC were intravenously administered following injury in a mouse AMI model. They migrated to the heart and persisted in the body for 2 to 3 days on average, with cells detected in only one animal on day 7.	Non-GLP Pharmacokinetics

Table 5-2. Overview Nonclinical Studies of MultiStem				
Study Title Study Number/Reference	**Test System**	**Number of Doses, Route of Administrations, Dosage, Test Article**	**Study Results**	**GLP Status/ Section Ref**
Evaluation of Human MultiStem Engraftment and Differentiation in a NOD-SCID Mouse Model RM-050822-01FR1	Mouse/ NOD SCID	1 Dose, IV, 1×10^6 human MAPC	MAPC were administered 1 day after sub-lethal irradiation. MAPC cells did not cause lesions or form tumors/ectopic tissue. Evidence for hematopoietic engraftment of human cells (1% to 2%) was observed in a single MAPC injected animal.	Non-GLP Pharmacokinetics
Toxicology Studies: Single Dose Toxicity				
Evaluation of clinical pathology following MAPC intravenous injection RM-060403-01FR1	Mouse/ NOD SCID Rat/Lewis	Mice: 1 Dose, IV, 1×10^6 human MAPC Rats: 1 Dose, IV, 1×10^7 rat MAPC	IV administration of a single dose of human MAPC cells in NOD/SCID mice, or syngeneic MAPC in Lewis rats was well tolerated. There was no apparent hematology or clinical chemistry effects.	GLP Single-dose Toxicity
28-Day histopathology of porcine coronary arteries after vehicle/contrast, porcine MAPC or human MAPC infusion with MDL09-2040-145-34 RM-110822-01FR	Pig/ Domestic Yorkshire crossbred	1 Dose, transarterial catheter, 5×10^7 human or pig MAPC	MAPC was administered to pigs and the animals were followed for 28 days to evaluate safety. No adverse test or control article related events were observed when evaluating clinical observations, body weight values, clinical pathology parameters or macroscopic observations. At the microscopic level, infusion of the first vehicle control alone (4% DMSO, 4% HSA) resulted in mild chronic inflammatory infiltrate at a lower incidence when compared to the infusion of pig or human MAPC cells delivered in either the first (4% DMSO, 4% HSA) or the second (Plasma-Lyte A) vehicle control. A similar inflammatory pattern was not present at infusion sites receiving the second vehicle control alone (Plasma-Lyte A). The mild to moderate chronic inflammatory infiltrate observed at the infusion sites treated with the pig or human MAPC cells with either the first (4% DMSA, 4% HSA) or the second (Plasma-Lyte A) vehicle control was characterized by formation of many lymphoid nodules admixed with lymphocytes, plasma cells, eosinophils, and macrophages.	GLP Single-dose Toxicity

Table 5-2. Overview Nonclinical Studies of MultiStem

Study Title Study Number/Reference	Test System	Number of Doses, Route of Administrations, Dosage, Test Article	Study Results	GLP Status/ Section Ref
Acute observations and 28-day histopathology of porcine coronary arteries after saline/contrast or MAPC infusion with MDL09-2040-145-34 RM-110308-01FR	Pig/ Domestic Yorkshire crossbred	1 Dose, transarterial catheter, 5×10^7 human MAPC	MAPC was administered to pigs and the animals were followed for a 27-day recovery period. Infusion of the test article using the injection catheters resulted in a severe chronic inflammatory infiltrate, characterized by numerous lymphoid nodules admixed with lymphocytes, plasma cells, macrophages and fewer numbers of eosinophils and pigmented macrophages. Occasionally, mineral deposits surrounded by multinucleated giant cells were observed. Generally, no appreciable difference in the microscopic findings at coronary artery sites 1 cm proximal to the infusion sites was observed between control and treated sites. No difference in microscopic findings was observed between the 2 delivery devices. There were no delivery device, test or vehicle related effects on body weight, clinical chemistry, coagulation times or hematology parameters in either treatment group at the post intervention and recovery intervals compared to pretest.	GLP Single-dose Toxicity

Table 5-2. Overview Nonclinical Studies of MultiStem

Study Title Study Number/Reference	Test System	Number of Doses, Route of Administrations, Dosage, Test Article	Study Results	GLP Status/ Section Ref
Transarterial infusion of pig MAPC in the coronary artery with severe vascular trauma and pre-implanted stent(s) RM-061011-02FR1	Pig/ Domestic farm	1 Dose, transarterial catheter, 1×10^8 pig MAPC	Pig MAPC were administered after injury. Angiography results revealed no findings of significance, no disturbance to the overlapped stents or maintenance of vessel patency. Visual inspection of the catheters after cell injection revealed no significant damage to the catheters after passage through pre-implanted overlapping stents. Analysis of the injury scores determined by histological analysis of the vessels indicated that there was no significant difference in the injury to the control vessels compared to the vessels that had cell injections. X-gal staining of the surrounding tissue at the injection site revealed the presence of positive staining indicating the successful delivery of βgal labeled MAPC cells. Intravascular ultrasound data revealed no significant changes in the stent architecture before or after the transarterial catheter.	Non-GLP Single-dose Toxicity
Pivotal study of human MultiStem cells in a MCAO stroke model in rats RM-061212-01FR3	Rat/Sprague Dawley	1 Dose, IV, 1×10^6 - 2×10^7 human MAPC	MAPC were administered 2 days after induction of stroke and the animals were followed for 14 or 28 days. There were no test article-related clinical observations, effects on body weight, food consumption or hematology and serum chemistry parameters. There were no test article related macroscopic findings or effects on organ weights. A few animals in both the MAPC and vehicle control groups died, but these deaths were considered related to the surgical procedure in view of timing of death and incidence.	GLP Pharmacokinetics Single-dose Toxicity
Toxicology Studies: Repeat-dose Toxicity				
Evaluation of hematopoietic reconstitution in irradiated rats in the presence or absence of MultiStem RM-060401-02FR2	Rat/ Lewis or Sprague Dawley	1 Dose, IV, 2.5×10^6 rat MAPC 2 Doses, IV, 2.5×10^6 rat MAPC	Rat MAPC were injected 1 day, or 1 day and 8 days, after irradiation. MAPC did not interfere with reconstitution of the hematopoietic system, neither after sub-lethal irradiation, nor after lethal irradiation followed by HSCT. The rates and reconstitution levels of neutrophils, white blood cells and/or platelets in MAPC recipients were the same as in control animals.	Non-GLP Repeat-dose Toxicity

Table 5-2. Overview Nonclinical Studies of MultiStem				
Study Title Study Number/Reference	**Test System**	**Number of Doses, Route of Administrations, Dosage, Test Article**	**Study Results**	**GLP Status/ Section Ref**
Safety of administration of human MAPC processed in HypoThermosol in a mouse model of graft versus host disease RM-100730-01FR	Mouse/ B6D2F1	5 Doses, IV, 1×10^6 human MAPC	MAPC were administered 1 day after disease induction and every 3 days thereafter for 5 doses total. Acute infusion-related toxicity was not observed in any group. There were no biologically or toxicologically relevant differences in weight-loss or clinical scores between any of the groups. Gross pathology observations were as expected for GvHD animals and there were no other gross pathology findings. At the end of the in-life portion of the study there were no deaths in the non-GvHD group, 1/8 animals died in the GvHD (no vehicle) and Plasma-Lyte A control groups, and 4/8 and 3/8 animals died in the HTS and MAPC in HTS groups, respectively. The early deaths in all groups were likely related to GvHD model-based radiation injury. Slight increases in the severity of lung pathology (GvHD related lesions of increasing thickness characterized by perivascular lymphocyte infiltration) were observed in HTS and MAPC in HTS groups as compared to the treated and non-treated control groups.	Non-GLP Repeat-dose Toxicity
Toxicology Studies: Immunogenicity				
Evaluation of T-Cell sensitization and alloantibody generation after intravenous injection of allogeneic MultiStem in rats RM-051026-01FR3	Rat/Lewis or Buffalo	1 or 2 Doses, IV, 1 to 10×10^6 rat MAPC. 5 Doses, IV, 2.5×10^6 rat MAPC	Rat MAPC were administered IV to healthy rats on day 1 or days 1 and 35. Alternatively, irradiated rats were administered rat MAPC once a week for 5 doses, beginning on the day of irradiation. Infusion with high doses (62.5 million cells/kg) of allogeneic MAPC did not induce alloantibody formation or allosensitized T-cells. After infusion with allogeneic splenocytes significant allo T-cell sensitization and alloantibody generation was observed. Repeated IV administration (5x) of a high dose (12.5 million cells/kg) of allogeneic MAPC did not result in the generation of detectable levels of alloantibodies, or in significant T-cell sensitization against allogeneic MAPC.	Non-GLP Immunogenicity

Table 5-2. Overview Nonclinical Studies of MultiStem				
Study Title Study Number/Reference	**Test System**	**Number of Doses, Route of Administrations, Dosage, Test Article**	**Study Results**	**GLP Status/ Section Ref**
Toxicology: Tumorigenicity				
Evaluation of human MultiStem tumorigenicity in nude mice under GLP conditions RM-060522-02FR1	Mouse/ nude	1 Dose, IV, 1×10^6 human MAPC	Administration of MultiStem was well tolerated by the mice. There were no adverse clinical observations or effects on body weights associated with MultiStem. In the B6-F10 group, 10 of 10 male and 9 of 10 female mice exhibited metastatic lung tumors. There was no test article-related macroscopic or microscopic pathology in the cellular test article or vehicle control groups.	GLP Pharmacokinetics Tumorigenicity
Evaluation of human MultiStem tumorigenicity in nude mice RM-051017-01FR1	Mouse/ nude	1 Dose, SC injection, 7.4 to 10×10^6 human MAPC	Administration of MAPC test article was well tolerated. There were no adverse clinical observations or effects on body weights associated with MAPC administration and none of the mice formed tumors. In the HT1080 group, 4 of 5 male and 2 of 5 female mice formed tumors. There were no test article-related macroscopic observations or organ weight changes in the cellular test article groups.	Non-GLP Tumorigenicity
Evaluation of tumor formation in nude mice following subcutaneous injection of human MultiStem RM-070117-01FR	Mouse/ athymic nude	1 Dose, SC injection, 1×10^7 human MAPC	All positive control mice were necropsied 13 days post-injection (PI) as all mice (10/10) showed large masses (> 10mm diameter) at the injection site. All mice in the negative control group injected with PBS were necropsied 84 days PI. None of the animals (0/10) showed evidence of solid tumor formation. All mice in the test group injected with MAPC were necropsied 84 days post-dose. None of these animals (0/10) showed evidence of solid tumor formation.	GLP Tumorigenicity

Table 5-2. Overview Nonclinical Studies of MultiStem				
Study Title Study Number/Reference	**Test System**	**Number of Doses, Route of Administrations, Dosage, Test Article**	**Study Results**	**GLP Status/ Section Ref**
GLP tumorigenicity assay study using the IV dosing route with interim timepoint RM-FR-003-15	Mouse/ nude mouse (nu/nu)	1 Dose, IV, 1×10^6 Human MAPC	In the positive control group, metastatic neoplastic foci were macroscopically observed in 19 of 20 animals and microscopically confirmed. In the negative control and MAPC groups, no macroscopically observed or histologically confirmed neoplastic metastases were observed at 90 days after administration (5 animals/sex), nor at 182-183 days after administration (15 animals/sex). No tumorigenicity was observed in athymic mice after single IV administration of MAPC when assessed according to the predefined criteria.	GLP Tumorigenicity
GLP tumorigenicity assay study protocol with IV dosing route RM-FR-004-15	Mouse/ nude mouse (nu/nu)	1 Dose, IV, 1×10^6 Human MAPC	In the positive control group, metastatic tumor lesions were macroscopically observed in 20 of 20 animals and microscopically confirmed. In the negative control and MAPC groups (20 animals each), no macroscopically or microscopically identified tumor metastasis were observed. No tumorigenicity was observed in athymic mice after single IV administration of MAPC when assessed according to the predefined criteria.	GLP Tumorigenicity
Genomic Stability				
Genomic stability and microarray analysis of MultiStem RM-051012-01FR1	*In vitro*	N/A (*In vitro*) human MAPC	Two-independent human MAPC products were evaluated by transcription profiling, chromosome SNP analysis, gene methylation array analysis, and G-banding/karyotyping for normal and equivalent cytogenetics at early and late population doublings. The MAPC products displayed normal karyotype and G-banding, absence of significant chromosomal rearrangement by SNP analysis, and equivalency in transcriptional profiling and gene methylation profile between population doubling ~20 and 40.	Non-GLP Genomic Stability

Table 5-2. Overview Nonclinical Studies of MultiStem				
Study Title Study Number/Reference	**Test System**	**Number of Doses, Route of Administrations, Dosage, Test Article**	**Study Results**	**GLP Status/ Section Ref**

AMI = acute myocardial infarction; ATP = adenosine triphosphate; βgal = β-galactosidase; BALF = bronchoalveolar lavage fluid; ConA = concavalin A; DMSO = dimethyl sulfoxide; FACS = fluorescence-activated cell sorting; GLP = Good Laboratory Practice; GRO/KC = GRO1 oncogene and keratinocyte-derived cytokine; GvHD = graft versus host disease; HLA = human leukocyte antigen; HSA = human serum albumin; HSCT = hematopoietic stem cell transplantation; HTS =- hypothermosol; IFN = interferon; IL = interleukin; IV = intravenous; LAD = left anterior descending artery; MAPC = multipotent adult progenitor cells; MCA= middle cerebral artery; MCP = monocyte chemotactic protein; MHC = major histocompatibility complex; MLR = mixed lymphocyte reaction assay; MPA = mycophenolate; MSC = mesenchymal stem cell; N/A = not applicable; NOAEL = no observed adverse effect level; NOD SCID = Nonobese diabetic/severe combined immunodeficiency; PBMC = peripheral blood mononuclear cell; PBS = phosphate-buffered saline; PET/CT = positron emission tomography/ computed tomography; RANTES = regulated upon activation normal T-cell expressed and presumably secreted; RR = respiratory rate; SC = subcutaneous; SNP = Single Nucleotide Polymorphism; VEGF = vascular endothelial growth factor; X-gal = 5-bromo-4-chloro-3-indolyl-β-D-galactopyranoside.

5.1. Nonclinical Pharmacology

5.1.1. *In Vitro* Pharmacology

A series of *in vitro* studies were performed to evaluate the immunoregulatory properties of rat MAPC and human MAPC cells. A dose-dependent inhibition of T-cell activation was observed with rat cells and was replicated using human MAPC cells isolated from different donors. The *in vitro* pharmacology studies conducted with MAPC are summarized in Table 5-2.

The immunosuppressive properties of human and rat MAPC have been evaluated in a series of *in vitro* T-cell activation assays, as described in the *in vitro* studies section of Table 5-2. The allogeneic potential of these cells was tested using MLR. Both rat and human MAPC had a low immunogenicity profile with immunosuppressive potency *in vitro* and were capable of suppressing T-cell proliferation after exposure to allogeneic cell stimuli (RM-051012-02FR1; RM-070109-02FR1, Kovacsovics-Bankowski, 2009). Inhibition of T-cell proliferation by rat MAPC cells *in vitro* was shown to be non-MHC restricted, reversible, and mediated by soluble factors (Kovacsovics-Bankowski et al, 2009). MAPC cells inhibit T-cell proliferation induced by allogeneic stem cells or other stimuli in a dose-dependent manner through the depletion of tryptophan derived from indoleamine-2,3-dioxygenase (IDO) (Reading, 2013). In another *in vitro* model, MAPC cells decrease pro-inflammatory macrophages and convert them to an anti-inflammatory phenotype (Busch, 2011). Further, MAPC suppresses the expression of adhesion molecules on endothelial cells to reduce neutrophil binding (RM-090113-01FR).

Human MAPC product lots generated in large-scale expansions consistent with the manufacturing protocol for clinical grade material did not elicit immune responses *in vitro* and suppressed active immune responses in a dose-dependent manner (RM-070109-02FR1). These results confirmed that the human MAPC product produced by a large-scale expansion protocol retained their immunosuppressive properties.

5.1.2. *In Vivo* Pharmacology

Numerous *in vivo* pharmacology studies have been conducted with MAPC in support of various indications, including AMI, GvHD, ischemic stroke, UC, SOT, ARDS, and Trauma. These studies are discussed below.

5.1.2.1. Acute Myocardial Infarction

In vivo studies using rat and pig AMI models showed that direct myocardial injection of allogeneic MAPC post-AMI results in significant increases in functional performance compared to vehicle controls. In the rat, myocardial MAPC injection of 10 million cells after AMI resulted in significant increase in vascular density within the infarct zone, with < 10% of MAPC remaining at 6 weeks (Van't Hof, 2007). In pig studies, approximately 0.3% to 0.4% cell engraftment was observed following direct injection into the peri-scar area (5 injections of 10 million cells each), with cells frequently found as vessel wall components (Zeng, 2007). Pig studies also showed increased vascular density around MAPC injection areas (RM-070208-01FR2). Administration of cyclosporine A did not affect cell retention or improvement of cardiac performance, supporting therapeutic benefit of allogeneic MAPC, without need for tissue matching or co-administration of immunosuppressive agents.

Studies in the pig AMI model showed higher cardiac persistence of MAPC upon transarterial injection (Mercator MedSystems catheter) as compared to intracoronary injection (Edwards catheter) (RM-051011-02FR2). Allogeneic pig MAPC (50 million cells) delivered by transarterial catheter also provided statistical benefit to left ventricular heart function at 4 weeks post dose (RM-070208-01FR). It was also shown that a dose of 20 million cells produced significantly less ventricular remodeling compared to a dose of 200 million cells (RM-060523-01FR3). The results from these studies support the therapeutic hypothesis of improvement of heart function following MAPC treatment of acute ischemic injury by enhancement of re-vascularization.

5.1.2.2. Graft versus Host Disease

Studies were conducted to evaluate the efficacy of MAPC cells in rodent models of HSCT and acute GvHD.

The effects of rat MAPC cells were evaluated in a rat model of HSCT in which rats were sublethally or lethally irradiated followed by IV injection of bone marrow mononuclear cells and then IV infusion of MAPC cells 24 hours later (RM-041006-01FR1). These studies showed that rat MAPC cells, at a dose of 2.5 million cells, do not interfere with the autologous or syngeneic hematopoietic reconstitution of either sublethally or lethally irradiated rats. In addition, there was a short-term survival benefit after IV administration of a dose of 5 million MAPC cells in that the 50% mortality rate was extended from 11 days for controls to 14 days for MAPC-treated rats; however, no long-term survival benefit was observed as all rats died by Day 19.

Studies were conducted to determine the efficacy of rat MAPC cells in a rat or mouse acute GvHD model (RM-060401-01FR1). In an acute GvHD rat model, animals receiving IV doses of 2.5 million allogeneic rat MAPC cells (12.5 million cells/kg) on Days 1 and 8 showed a statistically significant dose-dependent survival advantage over control groups (RM-060401-01FR1).

5.1.2.3. Ischemic Stroke

A series of *in vivo* experiments with rat or human MAPC cells were conducted to determine their efficacy in two different rodent models of ischemic stroke.

In a neonatal rat model of hypoxic-ischemic injury, a single dose escalation of MAPC via IV injection showed significant sustained functional recovery in locomotor function at ≥ 0.1 million cells/animal that persisted for 1-year post-dose (RM-060914-02FR1). Improvements in viable endogenous neurons in the hippocampus were also detected in the brains of injured animals treated with rat MAPC cells.

Ischemic stroke was studied in a Middle Cerebral Artery Ligation (MCAL) model in rats (RM-050903-01FR1). Direct stereotactic delivery of a single dose of 0.4 million cells/animal of either allogeneic rat MAPC or xenogeneic human MAPC, into the brains of rats 7 days post-injury, resulted in statistically significant improvements in locomotor and neurological testing when compared to non-viable cell treated control animals. The improvement was statistically significant with and without cyclosporine A, indicating immunosuppression is not required for the cells to exert their benefit in this model. IV injections of 0.4 million human MAPC at 7 days post-injury resulted in statistically significant improvements in locomotor behavior and neurological deficit compared to control, beginning at 2 weeks post-injury, and lasting through the end of the study at 8-weeks post-injury. The timing of the dose was then investigated, with 1 million human MAPC injected IV at 1-, 2-, or 7-days post-injury. Again, statistically significant improvements in locomotor behavior and neurological deficit compared to control, beginning at 2-weeks post-injury and lasting through the end of study at 56 days post-injury. Histological examination of endogenous neuronal protection showed animals dosed 1-day post-injury experienced a statistically significant protection of the ischemic penumbra compared to animals receiving cells 2- and 7-days post-injury. Animals dosed 2-days post-injury showed statistically significant protection of the ischemic penumbra compared to animals receiving cells 7-days post-injury.

Direct stereotactic delivery of human MAPC after either MCAO or MCAL resulted in significant improvements in locomotor and neurological tests when compared to vehicle-treated animals (RM-040916-01FR, RM-050822-03FR). There was a trend towards increasing improvement with increasing cell dose. No ectopic tissue was observed in any animal.

In another study, human MAPC at 0.4, 1, 2, 4, 10, or 20 million cells/dose or irradiated nonviable human MAPC at 10 million cells/dose (negative control) were administered to male and female rats 2 days after the induction of stroke via MCAL as a single IV infusion (RM-060914-03FR5). Locomotor and neurological functions were evaluated every 2 weeks from 2 weeks after the administration of MAPC (for

12 weeks for male rats and 54 weeks for female rats). Toxicity tests (clinical and gross pathology, body and organ weights, and histopathology) were performed for 3 female rats per group 1 year after the administration of MAPC. For 2 weeks post-MAPC cell injection through the end of the study, statistically significant recoveries of locomotor function and neurological function were observed for MAPC doses of 1 million cells/dose or higher. In addition, the effects reached a plateau at 4 million cells/dose. Immunohistochemistry on brain sections of both the males and females at the end of the study showed dose-dependent reduction in endogenous ischemic cell loss.

A study in a rat MCAO model of ischemic stroke demonstrate that administration of MAPC within 24 hours after the onset of injury resulted in improved recovery as measured by a composite behavioral score consisting of motor, sensory and balance tests (Yang, 2017). There was a statistically significant decrease in detectable CD3+ T-cells when comparing isolated splenocytes from MAPC treated vs. placebo treated ischemic stroke injured animals, demonstrating a clear effect of the MAPC cells in minimizing CD3+ T-cell mobilization.

Administration of MAPC 24 hours after onset of an injury to the CNS results in significant changes in the macrophage and microglial activation status in models of ischemic stroke (Yang et al, 2017), traumatic brain injury (Walker, 2010; Bedi, 2013) and spinal cord injury (SCI) (Busch, 2011; DePaul, 2015). MAPC treatment has also been shown to simultaneously increase tissue sparing, resulting in statistically significant improvements in neurological and/or locomotor outcomes.

These cumulative data demonstrated the potential of MAPC to provide therapeutic benefit for treatment following ischemic stroke.

5.1.2.4. Ulcerative Colitis

The nonclinical strategy for the evaluation of MAPC in treatment of UC is to use the data from the nonclinical pharmacology studies that support the clinical investigational use of MAPC cells in the treatment of GvHD. The rationale for using data from the nonclinical GvHD studies to support the use of MAPC cells in the UC setting in humans is based on the significant commonality of pathology between these 2 disorders, both of which are immune system related.

Administration of MAPC cells in nonclinical models of GvHD (Section 5.1.2.2) indicated that these cells distributed to the intestinal tract after IV dosing and provided protection from inflammatory damage. MAPC cells provided survival benefit in lethal acute GvHD models in part by reducing intestinal pathology while preserving intestinal function allowing the animals to regain weight. MAPC cell treatment resulted in significant efficacy in a murine model of GvHD with almost complete reversal of GI tract pathology including observed changes in intestinal crypts and villi. That MAPC cells afford protection from intestinal disease in nonclinical GvHD studies provides the rationale for evaluation of MAPC benefit in UC patients suffering from long-term inflammatory disease.

5.1.2.5. Solid Organ Transplant

In support of the solid organ transplant indications, a heterotopic cardiac transplant model study was conducted with MAPC cells in rats (Eggenhofer, 2013). A mycophenolate-based immunosuppression regimen was used in this study based on literature showing that this regimen prolongs donor-MSC–induced allograft survival (Popp, 2008), whereas CNI-based immunosuppression reduces the positive immunomodulatory effect of MSCs (Inoue, 2006).

A modest increase in graft survival was seen after IV injection of a single dose of donor-type MAPC cells on Day -4 (Eggenhofer et al, 2013). Dose escalation from 2 to up to 10 million cells did not increase the beneficial effect of MAPC. IV injection of 2 doses of donor-type MAPC (5 million cells/dose) on Days -4 and 0 markedly improved graft survival, and a further improvement in graft survival was seen when donor-type MAPC (5 million cells/dose) was applied intrasplenically. IV injection of 2 doses of third-party MAPC (5 million cells/dose) significantly prolonged survival of fully mismatched allogeneic grafts.

As already observed with donor-derived MAPC, intrasplenic injection of third-party MAPC (5 million cells/dose) further improved graft survival, implying that the liver is a prime site of immunological engagement in allograft rejection.

MAPC conferred long term protection from allogeneic immune rejection by inducing regulatory T-cells in the tissue environment subject to immune rejection. Protection can be long lasting (> 100 days), and allogeneic tissues can be re-transplanted to allogeneic recipients at 100% frequency without immunosuppressive drug or cell treatment. MAPC induced a protective immune homeostasis preventing recurrent allogenic recognition and rejection responses. This study demonstrated that MAPC cells mediate long-term acceptance of fully mismatched vascularized heart grafts when administered concurrently with low-dose CNI–free immunosuppression in rats.

5.1.2.6. Acute Respiratory Distress Syndrome

The safety of MAPC cells in lungs (intravascular and intrabronchial) was tested in animal and ex vivo human lung models. These nonclinical studies testing the safety and efficacy of MAPC cells in multiple species, injury models have produced data published in peer reviewed journals by independent academic research collaborators which demonstrate that MAPC cells significantly reduces lung inflammation after injury.

In a lipopolysaccharide (LPS)-induced model of ARDS described by Rojas (2013), IV infused MAPC cells reduced the severity of endotoxin induced lung injury, decreased hypoxemia and improved the clearance of alveolar edema (Rojas, unpublished data). MAPC cells suppressed the acute humoral and physiologic responses induced by endotoxemia by modulating the inflammatory response.

Additional studies have shown that in a sheep model of LPS-induced ARDS, administration of MAPC cells reduced inflammation and decreased time to recovery (Rojas, 2014). After administration of endotoxin, there was a decline in the levels of PaO2, followed by a slow recovery. Animals dosed endobronchially with 40 million MAPC cells at 30 minutes following the injury sustained a fast recovery, reaching baseline levels of PaO2 within 2 hours after cell therapy. LPS-induced increase in vascular pressure was reduced in animals treated with MAPC cells. MAPC treatment induced lower plasma levels of pro-inflammatory cytokine IL-8 and had less inflammation and congestion in the lungs when compared to control animals. Similar improvements in arterial blood oxygenation were observed in the LPS-induced sheep ARDS model when MAPC cells were delivered at 30 minutes following the injury either endobronchially (1 million cells/kg) or intravenously (10 million cells/kg) (Cardenes, 2019).

Studies with *ex vivo* human lungs also demonstrated that MAPC can reduce inflammation due to ischemia/reperfusion injury (La Francesca et al, 2014). *Ex vivo* human lungs not utilized for transplant were perfused and mechanically ventilated following bronchoscopic administration of MAPC cells (La Francesca, 2014). MAPC cells (40×10^6/lung) decreased lung injury and reduced inflammation.

5.1.2.7. Trauma

Human and animal data support a beneficial role of early adult stem cell treatment in the prevention of ARDS, TBI secondary injury and AKI. MAPC administration reduced severity of disease, improved clearance of alveolar edema and returned lung endothelial permeability to normal in an ex vivo perfused swine lung model of ARDS (Rojas, unpublished data). In a sheep model of LPS induced ARDS, vascular pressure was reduced, partial pressure arterial oxygen (PaO2) levels quickly returned to normal and pulmonary edema cleared in MAPC-treated animals compared to control animals (Rojas et al, 2014; Cardenes et al, 2019).

In TBI and other acute central nervous system injury models, including ischemic stroke and spinal cord injury, early IV administration of MAPC cells results in a decrease in inflammatory systemic immune system cells in and around the site of injury, decreased presence of inflammatory cytokines in the blood

stream, and improved long-term locomotor and neurological outcomes in cell treated animals when compared to saline treated injured animals (Mays and Savitz, 2018).

In one specific rodent TBI study, the timing of dose administration and the effects of MAPC cells were examined in the rat TBI model comparing repeat dosing at 2 and 24 hours vs. 6 and 24 hours vs. 12 and 36 hours vs. 36 and 72 hours (Bedi, 2018). Animals receiving MAPC cells at the earlier time points of 2 and 6 hours demonstrated significant improvements in blood brain barrier protection and spatial learning compared to animals that received MAPC cells at 12 and 36 hours initially, suggesting the importance of earlier cell administration on physiological and recovery outcomes.

Recently, the potential benefit of MAPC to treat *ex vivo* human kidney ischemia/reperfusion injury was described (Thompson, 2020). This report describes how donated human kidneys, which were declined for transplant due to a suspicion of non-renal malignancy realized at the time of the retrieval operation, could be maintained via normothermic machine perfusion (NMP) for as long as 7 hours and benefit from initial treatment with MAPC. Five pairs of kidneys were evaluated with each pair randomized so that one kidney would receive, after 1 hour of stable perfusion, a dose of 50 million MAPC cells in 10 mL physiologic red cell perfusate via the cannulated renal artery. The other kidney would get 10 mL of perfusate alone after 1 hour of stable perfusion. The kidney pairs were maintained on NMP another 6 hours after treatment and urine output was measured, as was the expression of pro and anti-inflammatory cytokines and biomarkers associated with AKI in the perfusate. Microvascular perfusion was measured via contrast enhanced ultrasound in the medulla and kidney cortex. MAPC treated kidneys demonstrated improved urine output, decreased expression of the kidney injury biomarker NGAL (urine neutrophil gelatinase-associated lipocalin), improved microvascular perfusion on contrast enhanced ultrasound (cortex and medulla), downregulation of IL-1β and upregulation of IL-10 and indolamine-2, 3-dioxygenase when compared to the matched kidney treated with perfusate only. These *ex vivo* human kidney reperfusion injury model data demonstrate that treatment with MAPC cells to prevent ischemia-reperfusion associated AKI is feasible when undertaken in the time period immediately following the injury event (ie, death and organ harvest) and improves function while simultaneously decreasing inflammation and markers of nephrotoxicity associated with acute kidney injury.

The results with MAPC, from two large animal models for acute inflammation in the lungs and in the *ex vivo* human kidney ischemia/reperfusion injury models, highlight the potential efficacy and safety of MultiStem for the treatment of patients following traumatic injury and critical care conditions and their related downstream inflammatory sequalae.

5.1.3. Safety Pharmacology

Safety pharmacology studies to evaluate MAPC were conducted in rat models (RM-051012-03FR3 and RM-060401-03FR1). The effects of MAPC on the CNS, cardiovascular system, and respiratory system were evaluated as part of separate safety studies and repeat-dose toxicity studies using normal rodent or rodent disease models, described in Section Section 5.3

Rat MAPC cells were assessed in two rat respiratory safety pharmacology studies due to the observed accumulation of cells in the lung following IV administration (RM-051012-03FR3 and RM-060401-03FR1). These studies in healthy rats or a HSCT model demonstrated that IV administration of allogeneic rat MAPC cells as a single dose up to 200 million cells/kg, or repeated doses (5 weekly injections of 2.5 million cells per dose), did not cause pulmonary distress as measured by respiratory rate (RR).

5.1.4. Pharmacodynamic Drug Interactions

5.1.4.1. Acute Myocardial Infarction

In vitro studies were conducted to evaluate the growth, viability, and functionality (as measured by VEGF secretion) of MAPC cells in the presence of drugs potentially used during treatment of patients following an AMI including clopidogrel, atorvastatin, metoprolol, acetyl salicylic acid, abciximab, eptifibatide, N-

acetylcysteine, captopril, heparin, and drugs used to coat stents, rapamycin or paclitaxel (RM-070822-01FR1). Only rapamycin reduced cell growth and VEGF production in MAPC cells. These results confirm that rapamycin was functioning as a cell proliferation inhibitor at the concentrations tested.

5.1.4.2. Graft versus Host Disease

In vitro studies were performed to evaluate whether combinations of immunosuppressive drugs routinely used for prophylactic treatment of GvHD after allogeneic HSCT have any adverse effects on the viability, growth, and immunosuppressive activity of MAPC cells (RM-060615-01FR; RM-070301-01FR). Combinations of methotrexate with tacrolimus or cyclosporine did not have a detrimental effect on short-term MAPC viability or plating efficiency; however, MAPC expansion in culture was entirely inhibited in the long-term (14 days) presence of these drugs at physiological levels, confirming that the drugs act as cell-proliferation inhibitors. These drugs did not impact the immunosuppressive activity of MAPC cells as measured in T-cell activation assays.

Since patients that will be receiving MultiStem cells would also be receiving a HSCT prior to the infusion of MultiStem, the patients would be undergoing a conditioning regimen for this indication. The combinations of immunosuppressive and chemotherapeutic drugs used in the pre-conditioning include fludarabine, cyclophosphamide, anti-thymocyte globulin, and cyclosporine prior to bone marrow transplant and MultiStem infusion. After bone marrow transplant and MultiStem administration, patients will continue on a regimen of drugs including cyclosporine, hydrocortisone, methylprednisolone, and Granulocyte-Colony Stimulating Factor. Fludarabine and cyclophosphamide are stopped 2 days before bone marrow transplant; therefore, these drugs were not tested. The other drugs did not interfere with MAPC cell viability *in vitro* (RM-060301-09FR).

5.1.4.3. Stroke

The multi-drug regimens used in the acute-onset treatment of ischemic stroke patients include drugs described in Section 5.1.4.1. Medications to stabilize the patient and treat the ischemic attack may include statins, beta-blockers, antiplatelets, and anticoagulants. Select examples from these drugs classes were tested with MAPC and shown to have no impact on cell viability, growth, or activity (RM-FR-015-16, RM-070822-01FR1). In addition, recombinant tissue plasminogen activator (rtPA may be given to patients who have had a stroke within 4.5 hours of presenting in the urgent care facility. With a half-life of 3 to 5 minutes, rtPA and its breakdown products will have cleared systemically at the time of MultiStem administration. Therefore, rtPA was not tested in the *in vitro* studies described in Section 5.1.4.1.

5.1.4.4. Ulcerative Colitis

In vitro studies were conducted to evaluate the potential interaction of MAPC cells with a range of drugs that may be used concomitantly in UC, including 5-aminosalicylic acid (5-ASA), budesonide, azathioprine, 6-mercaptopurine (6-MP), and a TNF inhibitor, adalimumab (RM-100329-01FR1). The studies showed that the concomitant administration of these drugs with MultiStem drug product are unlikely to have a significantly adverse effect on MultiStem cell viability or immunosuppressive activity.

5.1.4.5. Solid Organ Transplant

The possible interaction between MAPC and a CNI-free immunosuppressant (ie, mycophenolate) has been investigated (see Section 5.1.2.5). MAPC mediated long-term acceptance of fully mismatched vascularized heart grafts when administered concurrently with mycophenolate immunosuppression in rats.

5.1.4.6. ARDS

In vitro studies were conducted to investigate possible interactions between MAPC and drugs commonly used in the treatment of ARDS. No effect on growth or viability was shown for budesonide, methylprednisolone, and a TNF inhibitor, adalimumab (RM-100329-01FR, RM-060301-09FR). Viability

was not affected at 24 hours when treated with Zosyn (a combination of piperacillin and tazobactam, a penicillin antibiotic), cefepime (a cephalosporin antibiotic), vancomycin (a glycopeptide antibiotic), fentanyl, Versed, cisatracurium, omeprazole, or vasopressin (RM-141203-01FR1). A macrolide antibiotic, rapamycin (Sirolimus) showed decreased cell growth but did not affect cell viability (RM-070822-01FR1). Long-term exposure of the immunosuppressant, cyclosporine A showed MAPC growth inhibition but had no effect on T-cell activation or viability (RM-060615-01FR, RM-070301-01FR, and RM-060301-09FR). A commonly used drug in shock, hydrocortisone, had no effect on MAPC (RM-060301-09FR).

A central line catheter is the primary venous entry point in patients with ARDS. These catheters are often coated with silver and antibiotics like chlorohexidine. As MultiStem is delivered primarily intravenously in the clinical indications, biocompatibility of the cells with the infusion tubing was tested and showed no negative effects on viability and recovery of MultiStem (RM-FR-023-15).

5.1.4.7. Trauma

In vitro studies were conducted to evaluate the potential interaction of MAPC cells with a range of commonly used drugs that may be administered concomitantly in treatment of trauma patients, including epinephrine, ketamine, midazolam, and propofol. The studies demonstrated that the concomitant administration of these drugs with MultiStem drug product are unlikely to have a significantly adverse effect on MultiStem cell viability or immunomodulatory activity (RM-FR-032-18).

5.2. Pharmacokinetics and Product Metabolism in Animals

5.2.1. Absorption

The product is administered via IV infusion or locally (eg, to the heart for AMI) and therefore there are no relevant absorption processes.

5.2.2. Distribution

The nonclinical assessment of the pharmacokinetics of MAPC cells focused on biodistribution and residual presence in tissue.

The biodistribution of MAPC after transarterial catheter-based administration was examined in a pig model of AMI. Analysis of heart tissue at 2 and 8 weeks after administration of MAPC demonstrated < 1% cell persistence in tissue sections of the heart (RM-051011-02FR2).

The biodistribution of luciferase reporter rat MAPC cells following IV infusion in a mouse model of GvHD has been evaluated. *In vivo* imaging showed an initial accumulation of rat MAPC cells in the lungs in the hours immediately following infusion, followed by re-distribution to the GI tract over the following 24 to 48 hours. In the majority of samples, the bioluminescence signal was below the limit of detection by 10 days post dose, suggesting the majority of the administered cells were cleared in this time frame (RM-080714-01FR).

Distribution to the GI tract was only observed in the circumstance of injury. Similar studies in an ischemic heart injury model showed distribution to the heart with no detectable accumulation in the GI tract (RM-080603-01FR).

The persistence of MAPC cells at 4-, 8-, and 12-weeks post infusion in sublethally irradiated NOD/SCID mice was assessed in blood/bone marrow and selected tissues (RM-050822-01FR1; RM-060522-02FR1). No evidence of engraftment of MAPC was observed in the reproductive organs or gut tissues as examined by immunofluorescence using a human-specific antibody to β2-microglobulin. The blood and bone marrow of these animals were examined for MAPC presence using flow cytometry. One male animal was found to have low levels (1.09% human CD45+, mouse CD45-) in blood and no conclusive staining in bone marrow, but findings in all other animals were negative.

Human MAPC labeled with quantum dots were intravenously administered one day after stroke induction via MCAO or sham surgery in rats (Yang, 2017). MAPC was present in the lungs and spleen 1 and 3 days after administration, but not in the brain, liver, thymus, or kidneys. There were notable decreases in the lungs and spleen on Day 3 when compared with Day 1. On both Days 1 and 3, MAPC was present in the liver but not in the brain, kidneys, or thymus.

In a neonatal rat model of hypoxic-ischemic injury in which rat (green fluorescent protein (GFP) MAPC were administered 7 days after injury, there was a single reported observation of GFP positive cells stained at 52 weeks (RM-060914-02FR1). The GFP+ cells were also positive for MAP-2, a neuronal marker. No control antibody conditions were examined, but it is presumed the staining was artefactual. The dual labeling could also have been due to cell fusion upon initial administration, not long-term persistence.

In a GLP study, 2- and 4-week biodistribution studies in the brains of stroke injured rats were conducted, and no detectable signal was found by qPCR for human cells in the animal brains at either time point (RM-061212-01FR3).

Similar patterns of biodistribution of human quantum dot labeled MAPC have been observed in other models of CNS injury, including spinal cord injury and controlled cortical impact injury (Walker, 2010; DePaul, 2015). In both studies, MAPC in the spleen was present within the white pulp in close approximation with the blood vessel, suggesting interaction with resident splenic lymphocytes. Taken together, the subset of studies of MAPC biodistribution in the CNS suggest that MAPC mainly localizes in peripheral organs including the spleen and is cleared over several days following the insult.

The short-term biodistribution of MAPC cells labelled with [^{18}F] fluoro-29-deoxy-D-glucose and delivered by endobronchial or IV administration was examined in a sheep model of ARDS (Cardenes, 2019). Positron emission tomography/ computed tomography (PET/CT) images were acquired to determine the biodistribution and retention of the cells at 1 and 5 hours of administration. MAPC delivered by EB remained at the site of administration and no changes were observed between 1 and 5 hours, whereas with intravenous route, the cells had a broad biodistribution to different organs with the lung being the main organ of retention at 1 and 5 hours.

Across all nonclinical models examined, the biodistribution of MAPC is dependent on the route of administration, the dose given, and the injury or disease condition. Regardless of the model examined, MAPC cells were mostly cleared over the initial days following administration.

5.2.3. Metabolism

Nonclinical metabolism studies have not been conducted since these are not relevant to a cell-based therapy.

5.2.4. Excretion

Nonclinical excretion studies have not been conducted since these are not relevant to a cell-based therapy.

5.2.5. Pharmacokinetic Drug Interactions

Nonclinical pharmacokinetic drug interaction studies have not been conducted as conventional studies of *in vitro* enzyme inhibition or P450 induction are not relevant to a cell-based therapy.

5.3. Toxicology

5.3.1. Brief Summary

Rat, pig, and human MAPC cells were assessed in a series of nonclinical studies outlined in Table 5-2. The toxicology program focused on areas of particular importance for cell-based therapies, including single- and repeat-dose toxicity, immunogenicity, and tumorigenicity.

- Single-dose toxicity studies in mice and rats have been conducted using the IV and SC routes of administration with both human and rat MAPC cells. No dose-limiting toxicity was identified in general toxicity studies in mice and rats. The cells were well tolerated at up to 10 million cells/dose (500 million cells/kg administered SC) and up to 40 million cells/dose (200 million cells/kg administered IV) in mice and rats, respectively.
- Allogeneic rat MAPC cells have been administered to rats in an IV study for up to 5 weeks in duration (once weekly dosing). The NOAEL in the 5-week study was 2.5 million cells/dose (12.5 million cells/kg). Two IV doses of 10 million cells/dose (50 million cells/kg) have been given 1 week apart to rats with no adverse effects observed.
- In studies using pig cells in AMI pig models, up to 200 million cells were delivered by a transarterial catheter into the pig coronary artery and observed to be well tolerated.
- Immunogenicity, as measured by alloantibody or T-cell responses, has not been observed with allogeneic rat MAPC cells in rats.
- IV and SC tumorigenicity studies have been conducted in nude mice. MultiStem cells were well tolerated and did not induce ectopic tissues or tumors.

5.3.2. Single-Dose Toxicity

Two different human MAPC cell lots or one rat MAPC cell lot were administered as single IV doses to NOD/SCID mice or Lewis rats, respectively (RM-060403-01FR1). All MAPC cells were well tolerated at 1 million cells/dose (50 million cells/kg) in mice and at 10 million cells/dose (50 million cells/kg) in rats. There were no treatment-related effects on clinical signs, body weights or other parameters in either mice or rats during the conduct of the study. There were some changes noted in the clinical pathology parameters; however, none of the findings were determined to be definitely related to MAPC administration, nor were they deemed to be clinically meaningful.

In support of the AMI clinical program, the safety of local administration of pig MAPC cells was evaluated in three single-dose studies in the porcine model of AMI and two studies in uninjured porcine arteries (Zeng, 2007; RM-051011-02FR; RM-060523-01FR). Pig MAPC cells were injected directly into the peri-infarct zone (50 million cells/site × 5 sites) or via intracoronary or transarterial catheters (20-200 million cells). These studies demonstrated the safety of transarterial catheter-based delivery of MAPC. Additionally, this mode of delivery did not increase the injury to traumatized vessels or pre-implanted stents. Administration of pig or human MAPC cells into uninjured porcine arteries did not produce any adverse effects up to 28 days post dose. However, human MAPC infusions in a 4% DMSO and 20% contrast solution administered by transversing a pre-implanted stent resulted in a severe chronic inflammatory infiltrate as a possible immune response to the introduction of a xenogeneic test article into the myocardium of non-immunocompromised animals (RM-110822-01FR). Two additional studies of the safety of local administration of human (RM-110308-01FR) or pig (RM-061011-02FR1) MAPC revealed no MAPC-related effects following transarterial dosing, nor adverse clinical effects.

In support of the clinical program for ischemic stroke, three *in vivo* single dose safety studies were performed to measure the effect of up to 20 million cells/dose of rat or human MAPC transplanted into groups of neonatal or adult rats having undergone surgical induction of hypoxic-ischemic injury or ischemic stroke. In all three studies there were no MAPC-related changes in body weight, food consumption, organ weights, clinical pathology, gross pathology, or histology when rats were necropsied and evaluated between 14 days (RM-061212-01FR3) and 1-year post-dose (RM-060914-02FR1 and RM-060914-03FR5).

The safety of intraportal injection of MAPC was evaluated in support of the SOT clinical program (Eggenhofer, 2013). Administration of 1.5 million cells of allogeneic rat MAPC into the portal vein of rats produced no adverse effects on the hepatic parenchyma.

5.3.3. Repeat-Dose Toxicity

A total of 4 repeat-dose IV toxicity studies of rat and human MAPC were conducted in GvHD model mice and HSCT model rats, including RM-060401-02FR2. The study showed that MAPC cells did not interfere with reconstitution of the hematopoietic system following irradiation and HSCT. A repeat-dose study was conducted with rat MAPC cells in a rat model of HSCT (RM-060401-03FR1). Two females in the rat MAPC cell group died on Days 17 and 18 due to insufficient bone marrow transplant and one male in the rat single dose MAPC cell group was necropsied on Day 14 due to weight loss, and histopathology showed that this animal was debilitated due to radiation-related complication. There was no other morbidity or mortality. There were no biologically or toxicologically important MAPC-treatment related changes in clinical observations, clinical chemistry parameters, or histopathology as compared to control animals. There were no consistent treatment-related changes in immunogenicity as measured by alloantibody formation or allo-T-cell sensitization. The NOAEL for rat MAPC cells following once weekly IV administration for 5 weeks was 2.5 million cells/dose (12.5 million cells/kg).

A repeat-dose study was also conducted in a murine model of GvHD to evaluate the safety and efficacy of human MAPC in HTS when administered by IV injection once every 3 days (Days 1, 4, 7, 10, and 13) for 14 days (RM-100730-01FR). Control groups included untreated, HTS and Plasma-Lyte A. Day 14 was selected for necropsy, as this was deemed optimal for evaluation of animals with progressing GvHD without significant impact of GvHD-related mortality. Early deaths in all groups were likely related to GvHD model-based radiation injury. Slight increases in the severity of lung pathology (GvHD-related lesions) were observed in HTS and MAPC in HTS groups as compared to the Plasma-Lyte A and non-treated control groups. The relationship between the slight increases in the severity of lung pathology to HTS or MAPC in HTS administration could not be determined. In summary, HTS or MAPC in HTS administered IV in mice with early onset GvHD did not elicit any biologically or toxicologically significant effects greater than those observed in control animals induced for GvHD.

5.3.4. Reproductive and Developmental Toxicity

Formal reproductive and developmental toxicity studies have not been conducted with human MAPC cells.

5.3.5. Local Tolerance

No local tolerance studies have been conducted with MAPC. However, no macroscopic changes suggestive of irritation were observed at the administration site in any *in vivo* studies intended for other purposes, and histopathology, which was performed in some studies, showed no abnormal findings. In addition, no abnormal findings at the administration site were observed in any studies using Plasma-Lyte A as a cell diluent or vehicle.

5.3.6. Other Toxicity Studies

5.3.6.1. Antigenicity (Immunogenicity)

In vivo immunogenicity with rat MAPC cells has been assessed by evaluation of alloantibody formation or allo-T-cell sensitization in a stand-alone repeat-dose study in healthy rats and HSCT rat model and as part of a repeat-dose toxicity study in the rat model of HCST (RM-051026-01FR3). Single or repeated IV dosing (5 weekly doses) did not result in the generation of detectable levels of alloantibodies, or in significant T-cell sensitization against allogeneic MAPC cells.

5.3.6.2. Tumorigenicity Studies

Five studies have been conducted in nude mice to evaluate the potential for tumor formation after IV or SC administration of human MAPC/MultiStem cells (RM-060522-02FR1, RM-FR-003-15, RM-FR-004-15, RM-051017-01FR1, and RM-070117-01FR). There was no indication of malignancy or teratomas in the mice that received either MultiStem cells or vehicle (see Table 5-2).

Gross pathology and histopathology evaluations of multiple tissues have been conducted in pharmacology, toxicology, and tumorigenicity studies up to 1 year in duration after single or multiple IV, SC or intra-cranial infusions of rat or human MultiStem cells. There were no MultiStem cell-related alterations in gross pathology or histopathological evaluation of multiple tissues that would be indicative of tumorigenicity identified in any of the studies.

5.3.7. Safety of Diluents

No specific diluent safety studies were conducted. The majority of the nonclinical studies supporting the safety of MAPC cells were conducted with Plasma-Lyte A as the diluent for the cells and/or as a vehicle control.

5.3.7.1. Genomic Stability and Microarray Analysis of MAPC Cells

The long-term culturing of MAPC cells is necessary to provide sufficient numbers of cells for therapeutic treatment. Therefore, the genomic stability of two lots of the cells in long-term culture was examined to confirm that genetically normal and stable cells are provided for clinical use (RM-051012-01FR1). Genomic analysis of early (~20 population doublings [PD]) and late human MAPC cell cultures (~40 PD) demonstrated that little if any changes occurred in the genome with long-term culture (~40 PD). An examination of gene expression data revealed no major differences between the two lots. Karyotype analysis of early and late MAPC cell cultures demonstrated that both cultures had normal phenotype as assessed by G-banding. A more detailed analysis of genomic stability was assessed by Single Nucleotide Polymorphism (SNP) analysis and showed similar results. Finally, the methylation profile analysis demonstrated few differences between early and late human MAPC cell cultures. These results show that human MAPC cells maintain their genomic stability during the culture periods required for manufacture of clinical grade MAPC.

5.3.8. Target Organ Toxicity

Single and multiple IV dose nonclinical studies have been conducted with rat or human MAPC cells in mice and rats. Syngeneic and allogeneic MAPC cells were well tolerated after both single and multiple dose IV administration with no identified adverse effects or organ toxicity. Other routes of administration, including pericardial and intraportal vein, have also been shown to be well tolerated in animal models.

5.3.9. Integrated Overview and Conclusions

Numerous nonclinical studies in various animal models of different diseases and injuries have shown repeatedly that MAPC is safe. The MAPC cell therapy product is non-immunogenic, as demonstrated by suppression of T-cell proliferation *in vitro,* lack of alloreactive antibody formation *in vivo,* and equivalent efficacy in the absence of immunosuppressant drugs.

The MultiStem product is also non-tumorigenic, as illustrated by multiple GLP histopathology studies lasting as long as 1 year. After IV infusion, MAPC initially accumulates in the lungs, but causes no pulmonary effects, followed by efficient cell homing to sites of activity. MAPC cells are generally undetectable within a few days after infusion.

Single doses of 40 million MAPC cells/dose (200 million cells/kg administered IV) or 10 million MAPC cells/dose (500 million cells/kg administered SC) have been shown to be well tolerated in rats or mice, respectively. MAPC cell safety was evaluated in a repeat-dose IV study in a rat HSCT model and there were no adverse effects and no target organ toxicity identified. The NOAEL for rat MAPC cells in the rat HSCT model following once weekly IV administration for 5 weeks was 2.5 million cells/dose (12.5 million cells/kg).

The nonclinical safety profile of human MAPC cells has been adequately characterized and support its safe progression in clinical development for various indications.

6. EFFECTS IN HUMANS

6.1. Pharmacokinetics and Product Metabolism in Humans

The evaluation of absorption, metabolism, and excretion is not relevant to a cell-based therapy. MultiStem is neither metabolized nor excreted in a conventional sense. Clinical pharmacology data do not currently exist for MultiStem due to the lack of methods to track stem cells compared to traditional pharmacokinetic methods used in the study of chemical and biological products.

6.2. Safety and Efficacy

The clinical experience with the MultiStem product to date comprises five completed, two terminated, and five ongoing clinical trials. These trials are evaluating the safety, tolerability, and preliminary efficacy of MultiStem for different indications, ie, treatment following AMI (separate AMI-07-001 and B02-02 trials), prophylaxis of GvHD during and after HSCT (GVHD-2007-001), treatment for acute ischemic stroke (B01-02, B01-03, and B01-04) and UC (B3041001), support during and after liver transplant (MiSOT-I), and treatment of ARDS (B04-01, B04-02, and B04-03) and trauma (B06-01). These trials use different MultiStem formulations and concentrations, but identical cellular constituents. The 5 completed trials are in AMI (AMI-07-001), UC (B3041001), ischemic stroke (B01-02), ARDS (B04-01), and GvHD (GVHD-2007-001). The 2 terminated trials were in liver transplant (MiSOT-I) and AMI (B02-02). Ischemic stroke (B01-03 and B01-04), ARDS (B04-02 and B04-03) and trauma (B06-01) trials are ongoing with 189, 56, 33,10, and 0 subjects enrolled and dosed, respectively, as of this document's cutoff date.

MultiStem was delivered locally to the heart in completed and ongoing AMI trials and was administered IV in the completed GvHD, ischemic stroke, ARDS and UC trials and the ongoing ARDS, ischemic stroke, and trauma trials. The terminated MiSOT-I liver transplant study delivered MultiStem via portal and systemic IV circulation.

As outlined in Table 6-1, the product has been shown to be well tolerated in a total of approximately 413 participants in MultiStem clinical trials to date (25 Nov 2020 cutoff) using doses ranging from 20 million cells administered through local cardiac injection to ≥ 1.2 billion cells administered through systemic IV injection. In addition, MultiStem has been shown to be well tolerated using doses of 50 and 100 million cells administered via trans-arterial catheter to patients with AMI receiving percutaneous coronary intervention.

Table 6-1. Overview of Designs and Status of Completed/Terminated and Ongoing MultiStem Studies

Indication (Study Title/ID No.) Study Status	Reg Ref No. (Country) No. of Sites Study Duration	Study Purpose and Design	Primary End Point(s)	MultiStem Dose and Schedule	Planned Enrollment: Main Inclusion Criteria	Patient Disposition	FPI/LPO
Acute Myocardial Infarction-STEMI (AMI) (A phase 1, multicenter, dose-escalation trial evaluating the safety of allogeneic AMI MultiStem in patients with acute myocardial infarction/AMI-07-001) Completed	IND: 13554 (US) 7 sites 2-year follow-up	Open-label, multi-center, dose-escalation trial to evaluate the safety of specified doses of AMI MultiStem administered via trans-arterial catheter in patients with acute myocardial infarction receiving percutaneous coronary intervention	1. Assessment of acute adverse events during the first 24 hours 2. Assessment of post-acute adverse events up to 30 days	Cohorts at single 20, 50, or 100 million cell doses and registry cohort	28-37 subjects: Males or females, 18-85 years of age who had a diagnosis of first time ST elevation myocardial infarction (STEMI) with reduced ejection fraction (LVEF between 30% and 45%)	<u>Overall:</u> 25 enrolled 23 completed 2 premature termination <u>20 Million cohort</u> 6 enrolled 5 completed 1 withdrawn <u>50 Million cohort</u> 7 enrolled 6 completed 1 withdrawn <u>100 Million cohort</u> 6 enrolled 6 completed <u>Registry cohort</u> 6 enrolled 6 completed	Sep 2008/ Feb 2010

Table 6-1. Overview of Designs and Status of Completed/Terminated and Ongoing MultiStem Studies

Indication (Study Title/ID No.) Study Status	Reg Ref No. (Country) No. of Sites Study Duration	Study Purpose and Design	Primary End Point(s)	MultiStem Dose and Schedule	Planned Enrollment: Main Inclusion Criteria	Patient Disposition	FPI/LPO
Graft versus Host Disease (GvHD) (A phase 1, multicenter, dose-escalation trial evaluating maximum-tolerated dose of single and repeated administration of allogeneic MultiStem in patients with acute leukemia, chronic myeloid leukemia, or myelodysplasia/ GVHD-2007-001) Completed	IND: 13507 EudraCT: 2010-018760-16 (US, Belgium) 7 sites 100-day follow-up	Open-label, multicenter, 2-armed, dose escalation trial to assess the safety of MultiStem in subjects with hematological malignancy after hematopoietic stem cell transplantation (HSCT) in patients receiving MultiStem via systemic IV infusion	Maximum-tolerated dose, as determined by the CRM, evaluating dose limiting toxicities through 30 days after administration of the last MultiStem dose	First arm: single doses at 1, 5, or 10 million cells/kg Second arm: 1 or 5 million cells/kg administered weekly for three weeks or 5 million cells/kg administered weekly for 5 weeks	36 subjects: Males or females, 18-65 years of age inclusive, scheduled for a HSCT due to a diagnosis of acute myeloid leukemia, acute lymphoblastic leukemia, chronic myelogenous leukemia, or myelodysplastic syndrome	Overall 36 enrolled 30 completed 6 premature termination 1 million ×1 6 enrolled 5 completed 1 withdrawn 5 million ×1 3 enrolled 2 completed 1 withdrawn 10 million ×1 9 enrolled 7 completed 2 withdrawn 1 million ×3 3 enrolled 3 completed 5 million ×3 3 enrolled 2 completed 1 withdrawn 5 million ×5 12 enrolled 11 completed 1 withdrawn	Oct 2008/ Nov 2011

Table 6-1. Overview of Designs and Status of Completed/Terminated and Ongoing MultiStem Studies

Indication (Study Title/ID No.) Study Status	Reg Ref No. (Country) No. of Sites Study Duration	Study Purpose and Design	Primary End Point(s)	MultiStem Dose and Schedule	Planned Enrollment: Main Inclusion Criteria	Patient Disposition	FPI/LPO
Ulcerative colitis (UC) (A phase 2 randomized, placebo-controlled, parallel group, multi-center study to investigate the safety and efficacy of MultiStem (pf-05285401) in subjects with moderate to severe ulcerative colitis/ B3041001) Completed	IND:14512 EudraCT: 2010-022766-27 Canada CTA NO. 9427-P0006\7-237C (US, Belgium, Canada, Germany, Hungary, Italy, Slovakia, Sweden) 52 sites 1-year follow-up	Multi-center, double-blind, dose-escalation trial to evaluate the safety, tolerability and efficacy of intravenous doses of MultiStem in moderate-to severe ulcerative colitis in patients receiving MultiStem via systemic IV infusion	1. Incidence and severity of adverse events (at Weeks 4, 8, 12, and 16) 2. Change from baseline of endoscopic score at Week 8 as measured by modified Baron score. 3. Change from baseline of Mayo rectal bleeding sub-score at Week 4. 4. Change from baseline of Mayo rectal bleeding sub-score at Week 8	Cohort 1: 300 million total cells or placebo, with crossover at 8 weeks Cohort 2: 750 million total cells or placebo, with crossover at 8 weeks Cohort 3: patients randomized to 1 of the following groups: Group 1 D1: 750 million W8: placebo Group 2 D1: 750 million W8: 750 million Group 3 D1: placebo W8: 750 million Group 4 D1: placebo W8: placebo	128 subjects: Males or females, ≥ 18 years of age with diagnosis of active moderate-to-severe ulcerative colitis	Overall: 106 enrolled 65 completed 40 premature termination Cohort 1 9 enrolled- 8 Day 1 dosed 6 Week 8 dosed 3 complete 5 withdrawn Cohort 2 9 enrolled 9 Day 1 dosed 9 Week 8 dosed 4 complete 5 withdrawn Cohort 3 88 enrolled 88 Day 1 dosed 84 Week 8 dosed 58 completed 30 withdrawn 750 million x2 Group 1 & 3 (750 × 1) 42 enrolled Group 2 (750 million × 2) 21 enrolled- Group 4 (Placebo) 21 enrolled	Feb 2011/ Nov 2014

Table 6-1. Overview of Designs and Status of Completed/Terminated and Ongoing MultiStem Studies

Indication (Study Title/ID No.) Study Status	Reg Ref No. (Country) No. of Sites Study Duration	Study Purpose and Design	Primary End Point(s)	MultiStem Dose and Schedule	Planned Enrollment: Main Inclusion Criteria	Patient Disposition	FPI/LPO
Stroke (double-blind, randomized, placebo-controlled, phase 2 safety and efficacy trial of MultiStem in adults with ischemic stroke (MASTERS-1)/B01-02) Completed	IND: 13852 EudraCT: 2012-005749-18 (US, UK) 36 sites 1-year follow-up	Randomized, double-blind, placebo-controlled, multicenter, dose-escalation trial to assess safety and efficacy of MultiStem in adults with ischemic stroke in patients receiving MultiStem via systemic IV infusion	The global stroke recovery test statistic at Day 90, assessed by global disability neurological deficit and activities of daily living in MultiStem vs Placebo. DLTs between MultiStem and placebo in infusion related allergic reactions, related AEs and related neurologic worsening.	Cohorts 1 and 2: dose escalation from 400 million to 1.2 billion total cells or placebo (3:1) given once at baseline; Cohort 3 at 1.2 billion total cells or placebo (1:1) given once at baseline	136 subjects: Males or females, 18-83 years of age inclusive who have suffered a cortical ischemic stroke within the past 1-2 days	Overall: 134 enrolled 114 completed 20 premature termination cohort 1-400 million/placebo (3:1) 8 enrolled 8 completed 6 treatment 2 placebo cohort 2-1.2 billion/placebo (3:1) 8 enrolled 8 completed 6 treatment 2 placebo cohort 3-1.2 billion/placebo (1:1) - blinded 118 enrolled 98 completed 20 withdrawn	Dec 2011/ Dec 2015

Table 6-1. Overview of Designs and Status of Completed/Terminated and Ongoing MultiStem Studies

Indication (Study Title/ID No.) Study Status	Reg Ref No. (Country) No. of Sites Study Duration	Study Purpose and Design	Primary End Point(s)	MultiStem Dose and Schedule	Planned Enrollment: Main Inclusion Criteria	Patient Disposition	FPI/LPO
Liver transplant (phase 1: safety and feasibility of MultiStem for immunomodulation therapy after liver transplantation: a phase 1 study of the MiSOT study consortium/ MiSOT-I) Terminated	EudraCT: 2009-017795-25 (Germany) 1 site Up to 6 year follow-up	Investigator initiated, single center, open-label, dose-escalation trial to evaluate the safety and feasibility of MultiStem after allogeneic liver transplantation in patients receiving MultiStem via trans-portal and systemic IV infusion	Occurrence of dose limiting toxicity, defined as at least one grade 3 toxicity until day 30.	Two infusions with first at time of transplant via portal circulation and second systemically two days later with up to 150 to 600 million cells per infusion	12 to 24 subjects: Males or females, 18-65 years of age inclusive, undergoing liver transplant	Overall 3 enrolled 3 completed 0 premature termination Cohort 1 (150 million) 3 enrolled 3 completed 0 withdrawn	Aug 2014/ April 2016 (terminated due to poor enrollment)
Acute respiratory distress syndrome (MUST-ARDS: A phase 1/2 study to assess the safety and efficacy of MultiStem cell therapy in subjects with acute respiratory distress syndrome/ B04-01) Completed	IND: 16460 EudraCT: 2015-001586-96 (US, UK) 12 sites Up to 1 year follow-up	Multicenter, safety and efficacy trial conducted in 3 sequential cohorts including dose-escalation with open label and randomized, double-blind, placebo-controlled cohorts	The safety and tolerability within 4 hours of dosing and SUSARs within the first 24 hours after dosing.	Single infusions up to 96 hours post-ARDS diagnosis via systemic circulation with 300 or 900 million cells per infusion	40 subjects: Males or females, 18-90 years of age inclusive, with confirmed ARDS	Overall: 36 enrolled 21 completed 15 premature termination Cohort 1 (300 million) 3 enrolled 3 completed 0 withdrawn Cohort 2 (900 million) 3 enrolled 2 completed 1 withdrawn Cohort 3 (900 million/Placebo; 2:1) 30 enrolled 16 completed 14 withdrawn	Mar 2016/ Jul 2019

Table 6-1. Overview of Designs and Status of Completed/Terminated and Ongoing MultiStem Studies

Indication (Study Title/ID No.) Study Status	Reg Ref No. (Country) No. of Sites Study Duration	Study Purpose and Design	Primary End Point(s)	MultiStem Dose and Schedule	Planned Enrollment: Main Inclusion Criteria	Patient Disposition	FPI/LPO
Acute myocardial infarction-NSTEMI (A phase 2 prospective, randomized, double-blind, sham-controlled, parallel-group, multi-center trial of AMI MultiStem in subjects with non-ST elevation acute myocardial infarction/ B02-02) Terminated	IND: 13554 (US) 12 sites Up to 1-year follow-up	Multi-center, randomized, double-blind, sham-controlled, parallel-group trial to evaluate the safety and efficacy of AMI-MultiStem administered via a micro-infusion catheter in subjects with non-ST elevation acute myocardial infarction (NSTEMI) receiving percutaneous coronary intervention (PCI)	Difference between MultiStem and sham at Day 30 for AEs, severity of AEs, incidence of ventricular arrythmias. Difference between MultiStem and sham at Day 120 in myocardial perfusion measured by MRI	One infusion to the heart via a micro-infusion catheter following successful PCI, 50 million cells or Sham	90 subjects: Males or females, 18-85 years of age inclusive, undergoing PCI following NSTEMI	<u>Overall:</u> 34 enrolled 30 completed 4 premature termination <u>Therapy</u> 17 enrolled 16 completed 1 withdrawn <u>Sham Control</u> 17 enrolled 14 completed 3 withdrawn	Jan 2016/ Jan 2020 (terminated due to poor enrollment)

Table 6-1. Overview of Designs and Status of Completed/Terminated and Ongoing MultiStem Studies

Indication (Study Title/ID No.) Study Status	Reg Ref No. (Country) No. of Sites Study Duration	Study Purpose and Design	Primary End Point(s)	MultiStem Dose and Schedule	Planned Enrollment: Main Inclusion Criteria	Patient Disposition	FPI/LPO
Stroke (placebo-controlled, double-blind, phase 2/3 efficacy and safety trial of HLCM051 (MultiStem) in patients with ischemic stroke (TREASURE)/ B01-03) Ongoing	IND: 13852 (Japan) 48 sites 1-year follow-up and additional safety follow-up to 2 years	Randomized, placebo-controlled, double-blind, multicenter, phase 2/3 trial to evaluate the efficacy and safety of intravenous administration of HLCM051 compared with placebo in subjects with acute ischemic stroke (within 36 hours of onset)	1) Proportion of subjects with an excellent outcome defined by the functional assessments (Day 90) Excellent outcome is defined as modified Rankin Scale score of ≤ 1 (scale, 0 to 6), NIHSS score of ≤ 1 (scale, 0 to 42), and Barthel Index score of ≥ 95 (scale, 0 to 100). 2) Comparison between the HLCM051 and the placebo groups in key adverse events [Day90]	Infusion of 1.2 billion total cells or placebo (1:1) given once at baseline via systemic circulation	220 subjects: Males or females, 20 years and older of age inclusive who have suffered a cortical ischemic stroke within 18 to 36 hours	189 enrolled 79 completed (1 year) 23 premature termination (1 year)	Nov 2017/ March 2023 (planned)

Table 6-1. Overview of Designs and Status of Completed/Terminated and Ongoing MultiStem Studies

Indication (Study Title/ID No.) Study Status	Reg Ref No. (Country) No. of Sites Study Duration	Study Purpose and Design	Primary End Point(s)	MultiStem Dose and Schedule	Planned Enrollment: Main Inclusion Criteria	Patient Disposition	FPI/LPO
Stroke (MultiStem administration for stroke treatment and enhanced recovered study (MASTERS-2)/ B01-04) Ongoing	IND: 13852 EudraCT: 2019-001680-69 (US, EU, Asia Pacific) 50-80 sites (planned) 1-year follow-up	Randomized, placebo-controlled, double-blind, multicenter international, Phase 3 trial to evaluate the efficacy and safety of intravenous administration of MultiStem compared with placebo in subjects with acute ischemic stroke (within 36 hours of onset)	1) Differences between the MultiStem and placebo treatment groups in the distribution of Day 90 mRS scores will be evaluated by shift analysis. 2) Differences between the MultiStem and placebo treatment groups for the proportion of subjects with excellent functional outcome at Day 90, at Day 365, a mRS score of less than or equal to 2 at Day 90.	Infusion of 1.2 billion total cells or placebo (1:1) given once at baseline via systemic circulation	300 subjects: Males or females, 18 years and older of age inclusive who have suffered a cortical ischemic stroke within 18 to 36 hours of dosing	<u>Overall (blinded)</u> 56 enrolled 24 completed 11 withdrawn	Jul 2018/ Dec 2022 (planned)

Table 6-1. Overview of Designs and Status of Completed/Terminated and Ongoing MultiStem Studies

Indication (Study Title/ID No.) Study Status	Reg Ref No. (Country) No. of Sites Study Duration	Study Purpose and Design	Primary End Point(s)	MultiStem Dose and Schedule	Planned Enrollment: Main Inclusion Criteria	Patient Disposition	FPI/LPO
Acute respiratory distress syndrome (An open-label, standard treatment as a control, multicenter phase 2 study to evaluate the efficacy and safety of HLCM051 (MultiStem) in patients with acute respiratory distress syndrome (ARDS) caused by pneumonia (ONE-BRIDGE)/ B04-02) Ongoing	N/A (Japan) 29 sites 6-month follow-up	A Phase 2 randomized, standard treatment as a control, open-label, multicenter study to evaluate the efficacy and safety of intravenous administration of MultiStem compared to standard care only in subjects with ARDS caused by pneumonia	Pneumonia cohort: Number of days of survival free from mechanical ventilation (ventilator-free days) during 28 days after administration of IP. COVID-19 cohort: The number and rate of adverse events, changes in vital signs and laboratory test values during 180 days after administration of the IP.	Pneumonia cohort: Single, 900 million total cells infused up to 72 hours post-ARDS diagnosis given via systemic circulation along with ARDS standard care or standard care only (2:1) COVID-19 cohort: Single, 900 million total cells infused up to 72 hours post-ARDS diagnosis given via systemic circulation along with ARDS standard care	Pneumonia cohort: 30 subjects: Males or females, 20 to 90 years of age diagnosed with ARDS caused by pneumonia COVID-19 cohort: 5 subjects: Male or female, 20 to 70 years of age diagnosed with ARDS caused by COVID-19	Pneumonia cohort: 28 enrolled 14 completed 10 premature termination COVID-19 cohort: 5 enrolled 0 completed 0 premature termination	Apr 2019/ Jul 2021 (planned)
Trauma (MultiStem for treatment of trauma induced multiple organ failure/systemic inflammatory response syndrome/ B06-01) Ongoing	IND: 19272 (US) 1 site 12-month follow-up	A Phase 2 single center, prospective, placebo-controlled, double-blind, trial to evaluate the efficacy and safety of intravenous administration of MultiStem compared with placebo in severely injured trauma subjects within hours of hospitalization who have survived initial resuscitation.	Compare the incidence, severity, and duration of acute kidney injury within 30 days or hospital discharge	Infusion of 1.2 billion total cells or placebo (3:1) given via the systemic circulation within 8-24 hours post-known injury	156 subjects: Males or females, 18+ years of age diagnosed with a severe trauma injury	0 enrolled 0 completed 0 withdrawn 0 enrolled 0 completed 0 premature termination	Dec 2020/ Aug 2023 (planned)

Table 6-1. Overview of Designs and Status of Completed/Terminated and Ongoing MultiStem Studies

Indication (Study Title/ID No.) Study Status	Reg Ref No. (Country) No. of Sites Study Duration	Study Purpose and Design	Primary End Point(s)	MultiStem Dose and Schedule	Planned Enrollment: Main Inclusion Criteria	Patient Disposition	FPI/LPO
COVID-19 induced acute respiratory distress syndrome (A phase 2/3 study to assess the safety and efficacy of MultiStem Therapy in subjects with acute respiratory distress syndrome (ARDS) Due to Coronavirus disease (COVID-19)/ B04-03) Ongoing	IND: 16460 (US) 10-15 sites 12-month follow-up	A Phase 2/3 study with an open-label lead-in followed by a double-blinded, randomized, placebo-controlled part to evaluate the safety and efficacy of MultiStem therapy in subjects with moderate to severe ARDS due to COVID-19.	Ventilator-free days during 28 days after administration of the investigational product	Cohort 1a and 1 b open label, single infusion, dose escalation 900 million total cells to 1.2 billion total cells (2D) given within 48 hours of ARDS diagnosis Cohort 1 c and 1d -1 or 2 doses, open label, dose escalation 900 million total cells to 1.2 billion total cells (3D) given within 48 hours of ARDS diagnosis Cohort 2 1.2 billion total cells (2D and 3D) or placebo (1:1) given within 48 hours of ARDS diagnosis Cohort 3-TBD 12-month follow-up	300-400 subjects, (50 in cohort 2): Males or females, 18-89 years of age diagnosed with COVID-19 induced ARDS	<u>Overall:</u> 10 enrolled 0 completed 2 premature termination <u>Cohort 1a (900 million 2D)</u> 3 enrolled 0 completed 1 withdrawn <u>Cohort 1b (1.2 billion 2D)</u> 3 enrolled 0 completed 1 withdrawn <u>Cohort 2 (1.2 billion/ placebo; 1:1)</u> 4 enrolled 0 completed 0 withdrawn	Apr 2020/ Aug 2022 (planned)

* = Ongoing enrollment as of 25 Nov 2020.
AE = adverse event; AMI = acute myocardial infarction; ARDS = acute respiratory distress syndrome; CRM = continual reassessment method; DLT = dose limiting toxicity; FPI = first patient in; IP = investigational product; IV = intravenous; LPO = last patient in; NIHSS = National Institutes of Health Stroke Scale; NSTEMI = non-ST elevation acute myocardial infarction.

Table 6-2 presents a cumulative summary of all serious adverse reactions that occurred in any clinical trial with MultiStem until 25 Nov 2020.

Table 6-2. Cumulative Summary of Serious Adverse Reactions

System Organ Class (SOC)/Preferred Term (PT)	MultiStem	Blinded – MultiStem/Placebo
Total Serious Adverse Reactions	8	4
SOC Immune System Disorders		
PT Hypersensitivity	1	0
SOC Blood and lymphatic system disorders		
PT Pancytopenia	1	0
SOC Injury, poisoning and procedural complications		
PT Procedural complication	0	1**
PT Procedural intestinal perforation	0	1**
SOC Nervous system disorders		
PT Convulsions	1*	0
Dyskinesia	0	1
SOC Cardiac disorders		
PT Atrial fibrillation	1	0
Coronary artery dissection	0	1***
SOC Hepatobiliary disorders		
PT Hyperbilirubinemia	1	0
SOC Vascular disorders		
PT Hypotension*	1	0
SOC Neoplasms benign, malignant and unspecified		
PT Acute myeloid leukemia	1	0
SOC General disorders and administration site conditions		
Chills	1	0

* = Case was assessed as possibly related by the investigator but downgraded by the sponsor as a manifestation of the underlying disease.
** = This case was reported related to a study procedure, which occurred after enrollment and informed consent, but before investigational product dosing.
*** = This case was reported by the Investigator as unrelated to investigational product and definitely related to the micro-infusion investigational catheter procedure.

6.2.1. Summary of Trial in Patients with Acute Myocardial Infarction (AMI-07-001)

MultiStem was evaluated in a Phase 1 dose-escalation, open-label, safety trial (AMI-07-001) in patients following an AMI (Penn, 2012). There were no clinically significant changes to vital signs or evidence of allergic reaction associated with MultiStem administration observed immediately following dosing. Over the 30-day post-acute observation period, no infusional toxicities or clinically significant cardiac adverse events or arrhythmias occurred that were deemed to be definitely related to MultiStem.

Below is a listing of serious adverse events considered to be at least possibly related to MultiStem by Investigator and/or Sponsor.

Table 6-3. Listing of Serious Adverse Events Related to MultiStem for the AMI-07-001 Trial

System Organ Class (SOC)/Preferred Term (PT)	MultiStem	Blinded – MultiStem/Placebo
SOC Cardiac disorders		
PT Atrial fibrillation	1	0

6.2.2. Summary of Trial for Prophylaxis in Graft versus Host Disease (GVHD-2007-001)

MultiStem was evaluated in the completed Phase 1 dose-escalation, open-label safety trial (GVHD-2007-001) in patients undergoing HSCT. MultiStem was administered after transplant as a potential prophylactic agent to prevent GvHD. There were no allergic reactions related to MultiStem infusion. No HLA antibody responses were detected and no MultiStem drug product chimerism was observed up to Day 100 post-transplant. The safety profile was consistent with the events that would be expected from the high risk, HSCT population studied.

Below is a listing of 2 serious adverse events considered to be at least possibly related to MultiStem by Investigator and/or Sponsor. The acute myeloid leukemia case was a relapse of the existing disease that the patient was being treated for and was considered possibly related to MultiStem by the Investigator and unrelated by the Sponsor. The Investigator considered the case of "Hyperbilirubinemia" to be possibly related to therapy with MultiStem although the Sponsor assessed the event unrelated to MultiStem.

Table 6-4. Listing of Serious Adverse Events Related to MultiStem for the GVHD-2007-001 Trial

System Organ Class (SOC)/Preferred Term (PT)	MultiStem	Blinded – MultiStem/Placebo
SOC Hepatobiliary disorders		
PT Hyperbilirubinemia	1	0
SOC Neoplasms benign, malignant, and unspecified		
PT Acute myeloid leukemia	1	0

6.2.3. Summary of Trial in Patients with Ischemic Stroke (B01-02)

MultiStem was evaluated in a Phase 2 double-blind, randomized, placebo-controlled safety and efficacy trial in adults following an acute ischemic stroke. The total trial duration for safety and efficacy follow-up was 12 months. The study was completed in 2016 and final data have been reported. The trial is also referred to as MASTERS-1 (MultiStem Administration for Stroke Treatment and Enhanced Recovery Study).

In total, 71 subjects received a single dose of MultiStem, and 63 subjects received a single dose of placebo 24 to 48 hours after stroke onset during the study. Six subjects received 400 million cells and 65 subjects received 1.2 billion cells.

Although an efficacy analysis of Excellent Outcomes (modified Rankin Scale score ≤ 1, National Institutes of Health Stroke Scale score ≤ 1, and Barthel Index score ≥ 95) showed a percentage difference which favored MultiStem treatment, the prospective analysis was not statistically significant for the primary and secondary efficacy endpoints for the trial (Hess, 2017). However, the post-hoc analyses demonstrated that MultiStem treatment had an effect as measured by multiple endpoints (eg, Global Stroke Recovery test statistic, distribution of modified Rankin Scale scores, and Excellent Outcome), especially for subjects who received treatment ≤ 36 hours; this effect became more pronounced through 1 year. Based on these results, a Phase 2/3 trial in Japan referred to as TREASURE and a Phase 3 trial in North American, Europe, and the Asia Pacific region referred to as MASTERS-2 will be conducted to confirm these efficacy results.

Treatment with MultiStem was shown to be safe and generally well tolerated throughout all dose cohorts in MASTERS-1. In particular, the highest dose studied (1.2 billion total cells) was shown to be safe and well tolerated during the study. No subjects had infusion-related allergic reactions, neurological worsening, or reported dose limiting adverse that trigger protocol stopping rules. The majority of adverse events were mild or moderate in severity and consistent with the disease state being studied. There were no clinically significant differences between treatment groups in laboratory findings or vital signs.

Additionally, the results showed MultiStem to be associated with a favorable impact on a range of complications and outcomes following ischemic stroke. Treatment with MultiStem was associated with a reduced incidence of deaths, life-threatening adverse events, urinary tract infection, and secondary infection rates compared to placebo. Additionally, the mean duration of hospitalization and the mean duration of stay in an intensive care unit were shorter in the MultiStem group compared to the placebo group.

No serious adverse events were determined to be related to MultiStem use by the Sponsor.

6.2.4. Summary of Trial in Patients with Ulcerative Colitis (B3041001)

MultiStem was evaluated in a completed Phase 2 dose-escalation safety and efficacy trial (B3041001) in patients with moderate to severe UC. This trial was sponsored by Pfizer Inc and has been completed.

Overall, the completed study showed a favorable safety profile for MultiStem with repeat IV infusions at Day 1 and Week 8 of 300 or 750 million total cells. There were no differences between MultiStem and Placebo for clinical laboratory parameters and vital signs. Adverse events reported were what was expected for this population of patients with active moderate-to-severe UC. No deaths were reported in this study and the number of subjects discontinuing due to adverse events was small and not different between groups.

Weekly repeat infusions of 300 million total cells of MultiStem for the initial patients of Cohort 1 (Day 1, Week 1, and Week 2) were associated with delayed onset infusion reactions (pyrexia and chills) and hypersensitivity, which were transient and treated with standard medications. Weekly re-challenge with MultiStem was avoided during the rest of Cohort 1 and in subsequent Cohort 2 (300 million cells) and Cohort 3 (750 million cells). Repeat dosing was limited to a maximum of two infusions of MultiStem at Day 1 and Week 8, which appeared to be generally well tolerated in this patient population with most adverse events being mild to moderate in severity.

Table 6-5 has a listing of serious adverse events considered to be at least possibly related to MultiStem by Investigator and/or Sponsor from this completed study. A serious adverse event of hospitalization for "hypersensitivity" was reported in Cohort 1. This occurred after the second infusion of investigational product (ie, 1 week between doses). The causality of this event was possibly related to MultiStem or the solution used to dilute the stems cells prior to infusion. The event was reported by the Investigator as an allergic reaction characterized by fever and chills, which subsided after treatment with IV fluids, oxygen, famotidine, paracetamol, diphenhydramine hydrochloride, and IV steroids. No rash or respiratory issues were noted. The patient recovered, received no further infusions, and was followed in the trial to completion. The protocol was modified to allow multiple dosing, but with 8 weeks between MultiStem administration for Cohort 2 and 3.

A serious adverse event of "pancytopenia" was reported by the Investigator in Cohort 3 of the trial. Three days after the second infusion of the product at Week 8, the patient was admitted to the hospital with the initial diagnosis of "aplastic anemia", which was later revised to "pancytopenia". The patient recovered from the event. The Investigator classified the event as related to the investigational product. Pfizer's assessment concluded that although there were other risk factors which could contribute to the reported event, based on the information provided and an unclear temporal association, a causal relationship between the event "pancytopenia" and the blinded study drug could not be excluded. However, Pfizer considered it very likely that a causal relationship between the reported event and the concomitant UC

medication, 6-MP, exists that would explain the patient's recovery, and the improvement in platelet counts in particular.

Table 6-5. Listing of Serious Adverse Events Related to MultiStem for the B3041001 Trial for Ulcerative Colitis

System Organ Class (SOC)/Preferred Term (PT)	MultiStem	Blinded – MultiStem/Placebo
SOC Immune System Disorders		
PT Hypersensitivity	1	0
SOC Blood and lymphatic system disorders		
PT Pancytopenia	1	0
SOC Injury, poisoning and procedural complications		
PT Procedural complication	0	1*
PT Procedural intestinal perforation	0	1*

* = This case was reported related to a study procedure, which occurred after enrollment and informed consent, but before investigational product dosing.

6.2.5. Summary of Trial in Patients undergoing Liver Transplantation (MiSOT-I)

The MiSOT-I protocol was a Phase 1, open label, dose escalation, single arm, single center, safety, and feasibility study of MultiStem in patients undergoing allogeneic liver transplantation. Standard of care pharmacological immunosuppression can achieve reasonable survival of liver grafts and patients. The side effects of this treatment; however, are clinically significant and diminish the overall success of organ transplantation as a curative therapy. It was therefore the objective of this study to implement cellular immunomodulation therapy as an adjunct to standard pharmacological immunosuppression with the ultimate goal of significantly reducing drug-based immunosuppression. The primary objective of this trial was to evaluate short and longer-term safety of MultiStem. the trial was terminated in 2016 due to poor enrollment. Three subjects were enrolled with final study visits completed for the trial. MultiStem was well tolerated with no infusion-related reactions being reported and adverse events were consistent with the disease state being studied. No serious adverse events were reported to be related to MultiStem use.

6.2.6. Summary of Trial in Non-ST-Elevation Acute Myocardial Infarction (B02-02)

The B02-02 protocol was a Phase 2 multi-center, randomized, double-blind, sham-controlled, parallel-group trial to evaluate the safety and efficacy of AMI-MultiStem administered via a micro-infusion catheter in subjects with NSTEMI receiving percutaneous coronary intervention. Approximately 90 subjects were planned to be randomized 1:1 to 50 million cells of AMI-MultiStem or Sham. The primary objectives of this trial were to evaluate safety 30-days post-infusion and assess efficacy (myocardial perfusion as measured by cardiac MRI) at Day 120. The study was terminated in 2020 due to poor enrollment. A total of 34 subjects were enrolled in the trial (17 subjects treated with MultiStem) with no infusion-related reactions reported and adverse events were consistent with the disease state being studied. See listing in Table 6-6 of serious adverse events considered to be at least possibly related to MultiStem by Investigator and/or Sponsor. No serious adverse events have been reported to be related to MultiStem use. One reported serious adverse event of coronary artery dissection was reported related to investigational procedure, but not investigational product.

Table 6-6. Listing of Serious Adverse Events Related to MultiStem for the B02-02 Trial for AMI

System Organ Class (SOC)/Preferred Term (PT)	MultiStem	Blinded – MultiStem/Placebo
SOC Cardiac Disorders		
Coronary artery dissection	0	1*

* = This case was reported by the Investigator as unrelated to investigational product and definitely related to the micro-infusion investigational catheter procedure.

6.2.7. Summary of Trial in Acute Respiratory Distress Syndrome (B04-01)

The ARDS B04-01 protocol is a completed Phase 1/2 double-blind, randomized, placebo-controlled safety and efficacy trial that was conducted in the United States and the United Kingdom. The total trial duration for safety and efficacy follow-up was 12 months. The study was completed and final data reported in 2019.

The study was conducted in 3 sequential cohorts, starting with 2 open-label cohorts (at different dose levels) followed by a randomized, double-blind, placebo-controlled cohort:

- Cohort 1: 3 subjects received 300 million cells via intravenous infusion.
- Cohort 2: 3 subjects received 900 million cells via intravenous infusion.
- Cohort 3: 30 subjects were randomized 2:1 to receive MultiStem therapy or placebo via intravenous infusion. In Cohort 3, MultiStem therapy was administered at the highest tolerated dose from Cohorts 1 and 2, which was 900 million cells.

Inclusion criteria included male or female subjects that were 18 to 90 years of age (inclusive) with new acute onset of moderate to severe ARDS (as per the Berlin definition). Once all diagnostic criteria for ARDS and other eligibility criteria had been met, subjects then had to achieve a 2-hour stable baseline period (pre infusion stability) prior to receiving treatment. The study infusion had to be administered within 96 hours of fulfilling the diagnostic criteria of moderate to severe ARDS.

A total of 36 subjects were randomized and received treatment. That was 3 subjects in Cohort 1, 3 subjects in Cohort 2, and 30 subjects in Cohort 3 (20 receiving MultiStem and 10 receiving placebo). The median age across all cohorts was approximately 60 years. The majority of subjects were White and body mass index was similar in all cohorts.

No infusion related adverse events of special interest were seen during the first 4 hours post-infusion or within the first 3 days post-infusion and no serious adverse events possibly related to MultiStem were reported. The majority of adverse events were moderate or severe in severity and consistent with the disease state being studied. No changes in laboratory values, vital signs or physical examination findings were seen that indicated an adverse effect of MultiStem therapy.

Other data collected during this exploratory study showed lower mortality of 25% in the MultiStem treatment group compared to 40% in the placebo group from Day 0 to Day 28. In addition, the MultiStem group had a higher median number of ventilator-free days from Day 0 to Day 28 (18.5 days) compared to the placebo group (6.5 days). The number of Intensive Care Unit-free days from Day 0 to Day 28 was higher in the MultiStem group (median of 12.5 days) compared to the placebo group (4.5 days).

6.2.8. Summary of Japanese Trial in Ischemic Stroke (B01-03)

The TREASURE trial (protocol B01-03) is an ongoing Phase 2/3 double-blind, randomized, placebo-controlled efficacy and safety trial in Japanese patients who have suffered an ischemic stroke. This trial is being conducted in Japan and the Sponsor is HEALIOS K.K.

Approximately 220 subjects will be randomized 1:1 to 1.2 billion cells of HLCM051 (MultiStem) or placebo. A single systemic infusion will be given within 36 hours from onset of an ischemic stroke and subjects will be followed for 1 year as the primary follow-up period with additional safety information collected at 2 years.

The primary objectives of this trial are to evaluate efficacy and safety. The primary efficacy objective will evaluate the proportion of subjects with an excellent outcome at Day 90 (modified Rankin Scale score of ≤ 1, National Institutes of Health Stroke Scale score of ≤ 1, and Barthel Index score of ≥ 95). The primary safety objective will examine comparisons between the HLCM051 and the placebo groups in key adverse events through Day 90.

As of 25-November-2020, the study has enrolled 189 subjects. Table 6-7 has a listing of serious adverse events considered to be at least possibly related to MultiStem by Investigator and/or Sponsor.

A serious adverse event of dyskinesia was reported. The Investigator reported the verbatim term of involuntary movement with onset of hemiballism/hemichorea observed on the left upper extremity 7 days after the stroke and 6 days following administration of blinded investigational product. Symptoms were reported as moderate in severity and gradually improved over the following 10 days. The dyskinesia event was reported as a serious adverse event because discharge from the acute care hospital and transfer to rehabilitation was delayed due to the participant's dyskinesia symptoms, which resolved 26 days after being initially reported. Given the temporal sequence of the event 6 days following blinded investigational product administration, a potential causal relationship between administration of blinded investigational product and dyskinesia could not be ruled out by the Investigator.

Table 6-7. Listing of Serious Adverse Events Related to MultiStem for the B01-03 Trial for Ischemic Stroke

System Organ Class (SOC)/Preferred Term (PT)	MultiStem	Blinded – MultiStem/Placebo
SOC Nervous system disorders		
Dyskinesia	0	1

6.2.9. Summary of International Trial in Ischemic Stroke (B01-04)

The MASTERS-2 trial (protocol B01-04) is an ongoing Phase 2/3 double-blind, randomized, placebo-controlled efficacy and safety trial in adult subjects who have suffered a moderate to moderately-severe, acute cortical ischemic stroke. This trial will be conducted in the US, Europe, Australia, and Taiwan. The Sponsor is Athersys, Inc.

Approximately 300 subjects will be randomized 1:1 to 1.2 billion cells of MultiStem or placebo. A single systemic infusion will be given within 18-36 hours from onset of an ischemic stroke and subjects will be followed for 1 year.

The primary objective of this trial is to evaluate efficacy. The primary efficacy objective will evaluate differences between the MultiStem and placebo treatment groups in the distribution of Day 90 mRS scores by shift analysis. The key secondary efficacy variables will examine differences between the MultiStem and placebo treatment groups for the:

- Proportion of subjects achieving an excellent outcome at Day 365 defined by all of the following criteria: mRS score of ≤ 1, NIHSS total score of ≤ 1, and Barthel Index score of ≥ 95;
- Proportion of subjects achieving an excellent outcome at Day 90 defined by all of the following criteria: mRS score of ≤ 1, NIHSS total score of ≤ 1, and Barthel Index score of ≥ 95; and
- Proportion of subjects with a mRS score of ≤ 2 at Day 90.

As of 25 Nov 2020, the study has enrolled 56 subjects. No serious adverse events have been reported to be related to MultiStem use.

6.2.10. Summary of Japanese Trial in Acute Respiratory Distress Syndrome (B04-02)

The ONE-BRIDGE study (protocol B04-02) is an open-label, standard treatment as a control, multicenter Phase 2 trial to evaluate the efficacy and safety of MultiStem in Japanese patients with acute respiratory distress syndrome (ARDS) caused by pneumonia or Covid-19. This trial is being conducted in Japan and the Sponsor is HEALIOS K.K.

This study consists of 2 cohorts, the ARDS caused by pneumonia cohort (Pneumonia cohort) and the ARDS caused by COVID-19 cohort (COVID-19 cohort) which was added by the amendment protocol in April 2020. In the Pneumonia cohort, 30 subjects will be randomly assigned to the HLCM051 group or the standard therapy group in a 2:1 ratio. In the COVID-19 cohort, a total of 5 subjects are assigned to the HLCM051 group. Patients in the HLCM051 group, in addition to standard ARDS therapy, will receive one unit of HLCM051 that contains 900 million cells, administered as a single IV infusion within 72 hours of ARDS diagnosis.

The primary objective of the Pneumonia cohort in this trial is to evaluate efficacy. The primary efficacy objective will evaluate the number of days of survival free from mechanical ventilation (ventilator-free days) during 28 days after administration of the investigational product. The primary safety objectives will examine comparisons between the HLCM051 and the standard care only groups in adverse events, vital signs, and laboratory test values through Day 180. The primary objective of the COVID-19 cohort is to evaluate safety. The primary safety objectives are adverse events, vital signs, and laboratory test values through Day 180 after administration of the investigational product.

As of 25 Nov 2020, the study has enrolled 28 subjects in the Pneumonia cohort and 5 subjects in the COVID-19 cohort. Table 6-8 has a listing of serious adverse events considered to be at least possibly related to MultiStem by Investigator and/or Sponsor.

A serious adverse event of chills was reported. Per protocol, the study participant received, open-label, investigational product (MultiStem - 900 million cells), by single intravenous infusion following diagnosis of ARDS. The Investigator reported onset of chills ("shivering") about 20 minutes following the end of MultiStem administration. Moderate generalized tremor was accompanied by coughing, increased ventilated tidal volume, and decreased pulse oximetry measurements. The chills event was reported as severe in severity and resolved about 20 minutes after onset. Treatment consisted of up-titration of sedative medications and ventilator and airway management. Resolution of the event was followed by observation of increased body temperature lasting several hours. This event met seriousness criteria based on being judged an Important Medical Event by the Investigator. Given the temporal sequence of the event following MultiStem administration, a potential causal relationship between administration of MultiStem and chills could not be ruled out by the Investigator.

Table 6-8. Listing of Serious Adverse Events Related to MultiStem for the B04-02 Trial for ARDS

System Organ Class (SOC)/Preferred Term (PT)	MultiStem	Blinded – MultiStem/Placebo
SOC General Disorders and Administration Site Conditions		
Chills	1	0

6.2.11. Summary of Trial in Trauma (B06-01)

The MATRICS trial (protocol B06-01), is a Phase 2 double-blind, randomized, placebo-controlled efficacy and safety trial in severely injured trauma subjects within hours of hospitalization who have

survived initial resuscitation. This trial is being conducted at a single center in the USA and the Sponsor is Athersys, Inc.

Approximately 156 subjects will be randomized to 1.2 billion cells of MultiStem or placebo. A single intravenous infusion will be given within 8-24 hours from onset of the known trauma injury and subjects will be followed for 1 year.

The primary objectives of this trial are to evaluate efficacy and safety. The primary efficacy outcome measure will be to compare the incidence, severity and duration of AKI in the first 30 days after injury.

As of 25 Nov 2020, no subjects have been enrolled into the study.

6.2.12. Summary of Trial in COVID-19 Induced Acute Respiratory Distress Syndrome (B04-03)

The MACoVIA trial (protocol B04-03), is a multicenter trial featuring an open-label lead-in followed by a double-blinded, randomized, placebo-controlled Phase 2/3 part to evaluate the safety and efficacy of MultiStem therapy in subjects with moderate to severe ARDS due to COVID-19. This trial is being conducted in the US. The Sponsor is Athersys, Inc.

The study will enroll approximately 300 to 400 subjects and consist of 3 cohorts. Cohorts 1a, 1b, 1c, and 1d are open-label, with a single active treatment arm. Cohort 2 is a double-blind, randomized, placebo-controlled Run-In phase to evaluate the efficacy of MultiStem in the current clinical climate. The safety and, as applicable, efficacy data from the study will be reviewed by an independent Data and Safety Monitoring Board (DSMB), and the analysis of results from Cohort 2 will guide the design of Cohort 3. The DSMB will determine the recommended dose for Cohorts 2 and 3; and the sample size needed in Cohort 3. Subjects will be followed for 1 year.

The primary objectives of this study are to evaluate the safety and efficacy. The primary efficacy endpoint is Ventilator Free Days from Day 0 through Day 28.

As of 25 Nov 2020, 10 subjects (3 in Cohort 1a, 3 in Cohort 1b and 4 in Cohort 2) have been enrolled into the study. A serious adverse event of hypotension was reported.

Table 6-9. Listing of Serious Adverse Events Related to MultiStem for the B04-02 Trial for ARDS

System Organ Class (SOC)/Preferred Term (PT)	MultiStem	Blinded – MultiStem/Placebo
SOC General disorders and administration site conditions		
Hypotension	1	0

The SAE of hypotension was deemed related by the Investigator but deemed not related by the Sponsor. The final FDA rule was applied.
SAE = serios adverse event.

6.3. Reference Safety Information

A summary of the treatment-related serious adverse events experienced during each clinical trial with MultiStem through 25 Nov 2020 are presented in Table 6-3, Table 6-4, Table 6-5, Table 6-6 , Table 6-7, and Table 6-8 along with a summary in Table 6-2. These results show that there are limited serious adverse events seen with no events occurring in more than one subject. Therefore, all serious adverse events at least possibly related to the study drug will be considered as unexpected and will be reported to the Competent Authorities as per current legislation.

7. SUMMARY OF DATA AND GUIDANCE FOR THE INVESTIGATOR

7.1. Mode of Action and Intended Indications

MultiStem is a cell therapy medicinal product originating from adherent adult stem cells taken from the bone marrow of a non-related donor and expanded *ex vivo*. MultiStem appears capable of delivering a therapeutic benefit through more than one mechanism of action. Factors expressed by MultiStem are believed to reduce inflammation and regulate immune system function, protect damaged or injured cells and tissue, promote formation of new blood vessels, and augment tissue repair and healing (Auletta, 2010).

Based on these multipotent and immunoregulatory properties, MultiStem is being investigated in numerous indications including treatment of AMI, prevention of GvHD, treatment of ischemic stroke and UC, as an adjunct to immunotherapy in SOT, and in treatment of ARDS. The rationale for MultiStem in these indications is discussed in Section 3.1.

7.2. Posology and Method of Administration

MultiStem is a sterile aqueous suspension of viable human cells. Administration of MultiStem may differ by indication and clinical trial (see Section 4). Details of the dosing regimen are included in the clinical trial protocols.

7.2.1. Acute Myocardial Infarction

MultiStem was administered directly into the adventita of the target coronary vessel in patients with ST elevation AMI. To date, single doses of 20, 50, or 100 million total cells have been administered in this indication.

MultiStem is being administered directly into the adventita of the target coronary vessel in patients with non-ST elevation AMI. Single doses of 50 million total cells are being administered in this indication.

7.2.2. Graft versus Host Disease

MultiStem is administered via the IV route in patients who have undergone myeloablation and HSCT resulting in the potential for developing GvHD. To date, single doses of 1, 5, or 10 million cells/kg body weight and multiple doses of 1 or 5 million cells/kg weekly for three weeks or 5 million cells/kg weekly for 5 weeks have been administered in this indication.

7.2.3. Stroke

MultiStem is administered via the IV route in patients with ischemic stroke. To date, single doses of 400 million or 1.2 billion total cells have been administered in this indication.

7.2.4. Ulcerative Colitis

MultiStem is administered via the IV route in patients with UC. To date, single doses of 300 or 750 million total cells or two doses of 750 million total cells per dose have been administered in this indication.

7.2.5. Solid Organ Transplant

For patients undergoing liver transplantation, MultiStem is administered via the portal vein and via the IV route for subsequent doses. To date, multiple doses have been administered of 150 to 600 million total cells per the initial portal vein infusion followed by IV infusion for the second dose.

7.2.6. Acute Respiratory Distress Syndrome

MultiStem is administered via the IV route in patients with ARDS. Single doses of 300 million, 900 million or 1.2 billion total cells have been administered in this indication.

7.2.7. Trauma

MultiStem will administered via the IV route in patients who have suffered a severe trauma injury. Single doses of 1.2 billion total cells will be administered in this indication.

7.3. Risks, Side Effects, Precautions, and Special Monitoring

As of 25 Nov 2020, approximately 413 patients have received MultiStem, including an estimated 123 ischemic stroke patients and 32 ARDS patients from ongoing studies. The patients confirmed to have received MultiStem in completed or terminated studies include 36 AMI patients receiving transcoronary injection; 36 GvHD patients receiving IV infusion through a central line, 84 ulcerative colitis (UC); 71 acute ischemic stroke; 26 ARDS; and 2 expanded access use patients receiving IV infusion through a peripheral line; and 3 liver transplant patients via the portal circulation and through a peripheral line. There were no infusional or allergic reactions reported in the completed AMI, GvHD, ARDS, and stroke trials and adverse events reported were consistent with the disease state under study. There have been no adverse events definitely associated with MultiStem use in any of the completed trials or 2 expanded access use cases. In the completed UC trial, one serious adverse event of hypersensitivity, as further described below, and one serious adverse event of pancytopenia have been reported and considered possibly related to MultiStem or the product used to dilute the stem cells. In the ongoing open-label ARDS trial in Japan (B04-02), a serious adverse event of chills has been reported possibly related to MultiStem. In the ongoing international stroke trial (B01-04), there has been one report of a non-serious, Grade 2 infusion related reaction that resolved after treatment. In the ongoing B04-03 study in the US, there was a serious adverse event of hypotension reported by the Investigator as related to MultiStem; however, the sponsor deemed this SAE related to the underlying disease and not related to MultiStem. The final FDA rule was applied.

The cellular component of MultiStem is sourced from bone marrow from a human donor. As such it carries a potential risk of viral and non-viral contamination or infection when infused into patients. The bone marrow donation program has been accredited by the FDA and is compliant with EU regulations on cells and tissues for acquisition of human tissues, providing safeguards against unintentional contamination of the sample following collection and to ensure that quality of the cells is maintained. The manufacturing process used for expansion and harvest of cells has a number of steps where there is a risk for introduction of adventitious viral and non-viral agents. Thus, the manufacturing process involves a number of quality oversight procedures to minimize and identify such potential risks, including manufacturing under current Good Manufacturing Processes, certification or testing of all raw materials that come into contact with cells, and product sterility testing at multiple stages.

The major potential risks of administration of MultiStem, similar to that of cell-based therapies in general, include the immunogenic risk to patients and the potential for infusional toxicity. Although there is very limited persistence of MultiStem cells in animals and no evidence of toxicity or tumorigenicity in the numerous animal studies conducted, these cannot be excluded as potential risks.

To date, there has only been one serious adverse event of allergic reaction following MultiStem drug product administration in the patients exposed. This occurred in a UC trial patient following their second infusion and was characterized by fever and chills, as measured by changes in temperature. The reaction resolved after treatment at the hospital, including IV steroids. The diluent (HTS) used in the UC trial is different from the diluent (Plasma-Lyte A or equivalent) used in the AMI, GvHD, ARDS or ischemic stroke trials. There has only been one serious adverse event of chills possibly related to MultiStem drug product administration in the patients exposed. This occurred in an open-label ARDS trial patient and was characterized as shivering about 20 minutes following the end of MultiStem administration with the event being reported as severe in severity and resolved about 20 minutes after onset. There have been no allergic reactions or serious adverse events definitely related to MultiStem in any of the completed AMI, GvHD, ARDS, and stroke trials. In the GvHD study, no HLA antibody responses were detected and no MultiStem drug product chimerism was observed up to Day 100 post-transplant. The safety profile in the

AMI, GvHD, stroke, ARDS and UC trials was otherwise consistent with the events that would be expected from the high-risk populations being studied. Additionally, a series of *in vivo* safety studies were conducted to evaluate the immune responses occurring upon IV administration of multiple high doses of allogeneic MultiStem (5 to 50 million cells/kg) in rats. Infusion with high doses (50 million cells/kg) of allogeneic MultiStem did not induce alloreactive antibody formation or allo-sensitization. With allogeneic cell-based therapies there is a risk of triggering an immunological response (host versus graft reaction). MultiStem has been shown not to activate allogeneic T-cells in MLRs and has also been shown to suppress an allo-reaction between two mismatched lymphocytes. This allows MultiStem to be administered to patients without the need for tissue matching.

There have been no cases of other infusional toxicities reported in the completed AMI, GvHD, stroke, ARDS, and UC trials, or in the expanded access use cases. Moreover, infusional toxicity was evaluated in multiple animal studies. The results indicated that IV infusion of allogeneic MultiStem was well tolerated, without evidence of pulmonary distress, mortality or biologically significant change in body weight after infusion of single doses and no cumulative side effects after five repeated infusions of MultiStem. Single doses of 40 million MultiStem cells/dose (200 million cells/kg administered IV) or 10 million MultiStem cells/dose (500 million cells/kg administered SC) have been shown to be well tolerated in rats or mice, respectively. Additionally, 200 million MultiStem cells delivered into the adventitial layer of the coronary artery by a transarterial catheter were well tolerated in a pig AMI model. However, in pigs anesthetized with isoflurane, which causes a significant reduction in blood pressure on its own (~35%-40% lower than normal), transient hypotension has been observed shortly (1-2 minutes) following initiation of rapid intravenous infusion of MAPC cells and lasting for several minutes during the infusion, without evidence of impact on pulmonary function. This has not been observed in human subjects in any of the ongoing or completed trials and has not been observed in safety pharmacology studies in rodents evaluating rapid intravenous infusion of high doses of MultiStem cells or in other animal models with intravenous MAPC administration.

Although the risk of allergic reaction or infusional toxicity cannot be ruled out, they can be mitigated by dose escalation designs and by careful safety evaluations conducted before escalating the doses in subsequent cohorts of patients. Furthermore, vital sign and temperature measurements should be taken during and after each MultiStem administration. Any event of infusional toxicity or allergic response, defined as clinically significant deviations in blood pressure, heart rate, respiratory rate, temperature, and oxygen saturation, should also be recorded. In the event of infusional toxicity or allergic response and if flushing, sudden rash, or difficulty breathing occur, the infusion should immediately be slowed or stopped, or in cases of severe reactions (Grade 3 or higher) the infusion should be immediately terminated. Administration of MultiStem cells should be performed in an inpatient setting with access to equipment and staff qualified to provide appropriate emergency care.

Biodistribution and persistence of MultiStem cells was evaluated in NOD/SCID mice and in rodent disease models. Most of the MultiStem cells were cleared from tissues within a few weeks of administration. There was no evidence of tumorigenicity in SC and IV nude mouse tumorigenicity studies or in any other nonclinical studies where tissues were evaluated. Whereas embryonic stem cells in their undifferentiated state can cause the formation of teratomas when administered to animals, this effect has never been observed with MultiStem in nonclinical tumorigenicity studies or with other adult-derived stem cell products.

7.4. Interaction with Other Medicinal Products and Other Forms of Interaction

Clinical pharmacology studies have not been conducted with the MultiStem cell product. Human MultiStem cell viability and activity were assessed *in vitro* in the presence of drugs used as standard of care for each indication under clinical investigation (see Section 5.1.4). The results of these studies indicate that concomitant drug regimens should not significantly impact the viability or functionality of MultiStem cells.

7.5. Undesirable Effects

The clinical experience with the MultiStem product to date comprises 5 completed studies; 2 terminated studies; 2, single patient expanded access cases that have been terminated; and 5 ongoing trials which are being conducted using different MultiStem formulations and concentrations, but identical cellular constituents. While MultiStem was delivered locally to the heart in the AMI trial, it was infused as single and multiple doses IV in patients for prevention of GvHD and in patients with ischemic stroke. MultiStem was also infused as single and multiple IV doses in patients during the completed UC trial. The safety profile has been consistent with the events that would be expected from the high-risk populations being studied.

7.6. Overdose

The highest single dose administered to date (25 Nov 2020) in clinical studies was approximately 1.25 billion cells. No specific antidote exists for the treatment of MultiStem overdose.

7.7. Pregnancy and Lactation

Formal reproductive and developmental toxicity studies have not been conducted with human MultiStem cells.

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