REALIZING THE POWER OF RED™
A NEW ERA IN CELLULAR MEDICINE

SEPTEMBER 2020
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LEVERAGE HISTORY OF ADMINISTERING RED BLOOD CELLS

MODULAR PLATFORM THAT MIMICS IMMUNE BIOLOGY

BROAD PIPELINE

ADVANTAGEOUS TOLERABILITY

SCALABLE OFF-THE-SHELF

POTENTIALLY TRANSFORMATIVE CELLULAR THERAPIES

Cancer
Autoimmune

Red Cell Therapeutics™: The Future of Cellular Therapy
### Building a Broad and Diverse Pipeline

<table>
<thead>
<tr>
<th>PRODUCT CATEGORY</th>
<th>PROGRAM</th>
<th>PRECLINICAL</th>
<th>IND ENABLING</th>
<th>PHASE 1</th>
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<tbody>
<tr>
<td><strong>CANCER</strong></td>
<td>RTX-240</td>
<td>R/R Solid Tumors</td>
<td></td>
<td>Enrolling*</td>
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<td></td>
<td>RTX-240</td>
<td>R/R Acute Myeloid Leukemia</td>
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<td>IND Cleared; Recruiting</td>
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<td>RTX-321 aAPC (HPV 16+)</td>
<td>R/R HPV-16+ Solid Tumors</td>
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<td>IND Filing Expected by Year-End</td>
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<td>RTX-224</td>
<td>R/R Solid Tumors</td>
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<td></td>
<td>RTX-aAPC</td>
<td>Cancer</td>
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<td><strong>AUTOIMMUNE DISEASES</strong></td>
<td>RTX-T1D</td>
<td>Type 1 Diabetes</td>
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<td>RTX-TBD</td>
<td>Other Programs</td>
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*First patient dosed” announced via press release on May 7, 2020, updates provided on June 30 and August 10, 2020.*
The Promise of Red Cell Therapeutics™: Highly Potent, Allogeneic and Off-the-Shelf

RED PLATFORM®

- EARLY PROGENITOR CELLS
- EXPANSION & DIFFERENTIATION
- ENUCLEATION & MATURATION
- RED CELL THERAPEUTIC
- 100-1000’s OF DOSES

SINGLE HEALTHY O- DONOR

GENETIC ENGINEERING

100-1000's OF DOSES
Rubius Therapeutics’ Differentiated Oncology Approaches

BROAD IMMUNE SYSTEM STIMULATION

Stimulate adaptive and innate immunity through immune cell agonists

- Presentation of synergistic co-stimulatory ligands and cytokines
- Biodistribution may reduce toxicities
- Potential broad therapeutic window

ANTIGEN-SPECIFIC IMMUNE STIMULATION

Drive unique, in vivo antigen-specific immune responses

- Direct MHC I antigen presentation
- Co-stimulatory ligand induces significant quantity of CD8+ T cells
- Cytokine potently stimulates desired quality of killer T cells

Natural ligand presentation drives potent cell-cell interactions in preclinical models
RTX-240: BROAD IMMUNE SYSTEM STIMULATION
The Untapped Potential of Immune Agonists

**BLOCKING IMMUNE CHECKPOINTS WORKS**

- Checkpoint inhibitors (CPIs) release the “brakes” in cancer immunity
- Costimulatory and proinflammatory cytokine pathways push the “accelerator”
- CPIs are limited to certain cancers, most patients do not benefit and rapid disease progression is common
- Harnessing the immune system to treat cancer works
- New combinatorial approaches with CPIs are needed

**STRONG BIOLOGIC RATIONALE FOR AGONISTS**

- 80 agents in development targeting stimulatory pathways, such as 4-1BB, OX40, GITR, CD27, IL-15, IL-12, and IL-2
- Anti-4-1BB mAbs showed promise in solid tumors (utmilumab), but potent approaches (urelumab) have had toxicity
- IL-15 agonist (ALT-803) showed promise in AML Phase 1 study
  - Multiple other approaches being explored in combo
- These potent approaches limited by severe toxicities

- 4-1BB and IL-15 are promising pathways
- Clinical utility limited by toxicity, particularly in combination therapy
RTX-240: Enrolling Phase 1/2 Clinical Trial in Advanced Solid Tumors; Recruiting for Relapsed/Refractory Acute Myeloid Leukemia

BROAD IMMUNE SYSTEM STIMULATION

POTENTIAL BENEFITS:
- Activates existing agonist pathways leading to enhanced potency
- Improved anti-tumor activity
- Overcomes resistance to immunotherapy
- Reduced toxicity given biodistribution

RTX*-240 | (4-1BBL + IL-15TP)

*Rubius Therapeutics Terminology: RTX – Red Cell Therapeutic product candidate; mRBC – mouse surrogate model; RCT – experimental construct
### The Role of T Cells and NK Cells in Oncology

#### T CELLS

- T cells are critical mediators of anti-tumor responses through **cell killing, cytokine production**, and **immune cell activation**
  - CD8 T cells **directly kill tumor** cells that express tumor antigens
  - CD4 T cells **activate dendritic cells** by molecules, such as CD40L
  - CD4 and CD8 T cells **produce inflammatory cytokines** including IFN-γ and TNF
  - CD4 T cells promote **B cell expansion and maturation**

#### NK CELLS

- Natural killer (NK) cells mediate anti-tumor responses both via **cytolytic activity** and **immune activation**
  - NK cells **directly kill tumor cells** that express NK-activating molecules
  - NK cells augment immunotherapy escape mediated by MHC class I loss, by killing “missing self” cells
  - NK cells enhance **dendritic cell and T cell activation**
  - NK cells are targeted to kill by **antibody-dependent cellular cytotoxicity**
  - NK cells have a particularly strong role in **acute myeloid leukemia**

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The Proposed Mechanism of RTX-240 Encompasses Three Distinct Compartments

**PERIPHERAL BLOOD**
- T cell
- NK cell
- RTX
- RBC

**SPLEEN**
- White pulp
- Red pulp
- NK cell
- RTX
- RBC

**TUMOR**
- T cell
- NK cell
- Tumor cell

*Images depict interactions between cells, RTX, and RBC in different compartments.*
The Biodistribution of RBCs can be Exploited to Limit the Toxicity of Effective Targets

Liver

Spleen

mRBC-240 (4-1BBL + IL-15TP)

mRBCs/mm²

Spleen  Liver  Lung  Kidney  Heart

- mRBC-240
- CD31
- Hoechst
mRBC-240 Results in No Liver Toxicity Compared to Anti-4-1BB mAb

SERUM ALT

Normal mice; 4 Doses, 1x10⁹ cells day 0, 3, 7, 10; Sacrifice day 18

LIVER HISTOLOGY

Dugast, et. al., American Association for Cancer Research; Poster #3272, 2019
Treatment with mRBC-240 Expands CD8 and NK Cell Numbers in Spleen and Blood

Normal mice; 4 Doses, 1x10⁹ cells at days 0, 3, 7, 10; Sacrifice day 14
mRBC-240 Significantly Inhibits Tumor Growth as Monotherapy or in Combination with Anti-PD-1 mAb

mRBC-240 reduced tumor burden and was equivalent to anti-4-1BB mAb

mRBC-240 + anti-PD-1 mAb reduced tumor burden vs. anti-PD-1 mAb alone
RTX-240 Stimulated Potent Activation of Immune System

RTX-240 is ~10-Fold Superior to 4-1BB Agonist Antibody in Preclinical Models

Dugast, et. al., American Association for Cancer Research; Poster #3272, 2019
RTX-240 Promotes Expansion and Activation of Cells Driving Innate and Adaptive Immunity

8-DAY PBMC ASSAY

PBMCs + RTXs

Dendritic Cell
T Cell
B Cell
NK Cell

NK CELL NUMBER
CD8 CELL NUMBER

Number of CD8 T Cells

Number of NK Cells

RCT-CTRL
RCT-4-1BBL
RCT-IL-15TP
RTX
4-1BB Ab
rec-IL-15
4-1BB Ag + IL-15

Dugast, et. al., American Association for Cancer Research; Poster #3272, 2019
### RTX-240 Clinical Trial Development Plan

<table>
<thead>
<tr>
<th>RTX-240</th>
<th>DEVELOPMENT PLAN</th>
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<tr>
<td>Relapsed/Refractory or Locally Advanced Solid Tumors</td>
<td><strong>NOW ENROLLING:</strong> Phase 1: Monotherapy Dose-Escalation 4-6 cohorts</td>
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<tr>
<td>Relapsed/Refractory Acute Myeloid Leukemia</td>
<td><strong>Phase 2: Tumor-Specific Expansion Cohort #1</strong></td>
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<td><strong>Phase 2: Tumor-Specific Expansion Cohort #2</strong></td>
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<tr>
<td></td>
<td><strong>IND CLEARED:</strong> Phase 1: Monotherapy Dose-Escalation 4 cohorts, initially transplant ineligible with potential for post-transplant</td>
</tr>
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- cGMP clinical supply being produced at fully owned manufacturing facility
- **Phase 1 Objectives:**
  - Determine safety and tolerability, maximum tolerated dose, recommended Phase 2 dose and dosing interval of RTX-240
  - Assess pharmacodynamics of RTX-240 as measured by changes in T and NK cell number and function relative to baseline
  - Measure anti-tumor activity by Overall Response Rate (ORR)
  - Assess pharmacokinetics of RTX-240
Limited Therapeutics Options for Relapsed/Refractory Acute Myeloid Leukemia (AML)

AML IS A RARE AND AGGRESSIVE CANCER OF THE BLOOD AND BONE MARROW

• Some patients experience complete remission, and then experience a return of the leukemia, or relapsed AML

• Refractory AML occurs when patients have received approximately 2 rounds of chemotherapy, but the disease does not go into remission

Overall outcome for patients with relapsed/refractory AML remains poor
Five-year overall survival rate is approximately 10 percent
RTX-240 Leverages the Untapped Potential of NK Cells in AML

NK CELLS IN AML

- NK cells generate potent **cytolytic activity** and regulate **immune responses** against cancer cells
- NK cells play a key role in the development and eradication of **myeloid malignancies**
  - AML tumor-associated mechanisms often suppress proper NK cell function to avoid immune system recognition
  - Loss of NK activation receptor ligands is a hallmark of leukemia blasts and stem cells
    - Results in AML cells evading the immune system
  - Reconstitution of NK cells post high-dose chemo or transplant are **strong prognostic indicators** of overall survival

NK CELL INTERACTIONS

RTX-240 is designed to induce NK cell proliferation and activation, leading to the killing of AML cells
RTX-240 Induces Activation and Proliferation of NK Cells that are Effective In Vitro Against a Myeloid Leukemia Cell Line

- RTX-240 induces both proliferation and activation of NK cells in vitro
- These activated NK cells are effective in killing the myeloid leukemia cell line K562
RTX-321: ANTIGEN-SPECIFIC IMMUNE STIMULATION
Rubius’ First Engineered aAPC will Target HPV+ Tumors – IND Expected by End of 2020

Replicating immune system function to activate and expand antigen-specific T cells for a potent anti-tumor effect

RTX-321 (aAPC) | HPV 16+ Tumors

Signal 1
HPV-16 Antigen

Signal 2
4-1BBL

Signal 3
IL-12

T Cell

SELECTIVE TUMOR KILLING BY EXPANDING TUMOR-SPECIFIC T CELLS
Need for New Treatment Options in HPV 16-Positive Cancers

The risk of cancer is strongly associated with HPV infection.

High-risk strain HPV 16 accounts for approximately 70% of all cervical cancers and 80% of head and neck cancers associated with HPV infection.

Critical need for new and innovative treatment options for advanced HPV 16-associated cancers.
Rubius’ aAPCs Demonstrate Rapid and Direct Cell-to-Cell Interaction with Target T cells

Significant OT1-mRBC doublets observed by flow with aAPCs

Increase in labeled mRBC-OVA-4-1BBL-IL-12 cells in OT-1 gate observed with aAPCs indicates direct cell-to-cell interaction.

mRBCs labeled with CTFR injected at time 0
At 1h and 17h splenocytes harvested
Evaluated flow for double positive signals
- OT-1 gate X CTFR
Rubius’ aAPC Approach Drives Tumor Regressions in Vivo

TUMOR CONTROL
**EG7.OVA MODEL**

<table>
<thead>
<tr>
<th>Day after tumor randomization</th>
<th>Tumor volume (mm$^3$)</th>
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<tbody>
<tr>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>5</td>
<td>0</td>
</tr>
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<td>10</td>
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<td>20</td>
<td>0</td>
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<tr>
<td>25</td>
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CTRL
⇒ 0/8 regressions

mRBC – OVA + 4-1BBL at 1 x 10$^9$
⇒ 2/8 regressions

mRBC – OVA + 4-1BBL + IL-12 at 6.3 x 10$^7$
⇒ 5/8 regressions; 16-fold lower dose

mRBC – OVA + 4-1BBL + IL-12 at 2.5 x 10$^8$
⇒ 7/8 regressions; 4-fold lower dose

**MODEL DETAILS:**
EG7.OVA is a lymphoma cell line that expresses OVA (antigen from chicken ovalbumin)
OT-1 cells are T cells that recognize OVA

Zhang, et. al., Society for Immunotherapy of Cancer; Poster #P233, 2019
Re-challenge of Cured Mice Demonstrates Memory and Epitope Spreading

Re-challenge with original tumor

Re-challenge with EL4, the “parental” line of EG7.OVA. EL4 expresses other tumor antigens, but not OVA, the antigen that was presented by the aAPC.

Nixon, et. al., American Society of Gene & Cell Therapy; Poster #32, 2020
Rubius’ aAPC Approach Harnesses Endogenous T cells to Control Tumors

mRBC OVA-4-1BBL-IL-12 EXPANDS BOTH ADOPTIVELY TRANSFERRED AND ENDOGENOUS ANTIGEN-SPECIFIC T CELLS

OVA specific T cell expansion
Day 10 (blood)

Endogenous OVA specific T cells

CD45.2; OT1

mRBC or aAPC

EG7.OVA

Day -7

OT-1

D1

D4

D7

mRBC-CTRL or mRBC-aAPC

EG7.OVA

NOTE: No transfer of OT-1 cells
mRBC-aAPC (gp100) Nearly Eliminates Lung Metastases in Melanoma Model

MODEL DETAILS:
B16 is melanoma cell line carrying the tumor associated antigen gp100. Pmel T cells recognize gp100.

Zhang, et. al., Society for Immunotherapy of Cancer; Poster #P233, 2019

LUNG METASTASES COUNT

mRBC-aAPC (gp100)-CTRL

Lung metastases count

mRBC-aAPC (gp100)-CTRL

mRBC-aAPC (gp100)

mRBC-CTRL

aAPC dosing

D0 D1 D4 D8 D14

B16-F10 Pmel T cells Sacrifice

mRBC-gp100-4-1BBL-IL-12

1×10⁸ 2.5×10⁸ 6×10⁷
MODEL:
CD8 T cells are transduced with a TCR specific for HPV 16 E7 peptide cultured for 1 day with aAPCs. ~20% of the cells are HPV tet+ at the start of the culture, i.e., they have the TCR recognizing HPV.
**Expanded T Cells Produce Anti-Tumor Effector Molecules**

**GRANZYME B %**

**LEGEND**

- **RCT-CTRL**
- **RCT-HPV**
- **RCT-HPV-4-1BBL**
- **RTX-321**
- **RCT-CMV-4-1BBL-IL-12**

**IFNγ SECRETION**

**MODEL:**
CD8 T cells are transduced with a TCR specific for HPV 16 E7 peptide cultured for 1 day with aAPCs. ~20% of the cells are HPV tet+ at the start of the culture, i.e., they have the TCR recognizing HPV.
MANUFACTURING
• Highly experienced cell therapy technical operations team with scalable process

• Providing cGMP clinical supply for RTX-240 trial

• Expected to provide supply for RTX-321 trial

• Potential to expand manufacturing capabilities based on future needs
SUMMARY
Financial Position Supports Anticipated 2020 Milestones

- $236.5 million in cash, cash equivalents and investments as of June 30, 2020
- Cash runway into 2022

AN INTEGRATED DEVELOPMENT COMPANY

- Continue to enroll patients in RTX-240 solid tumor; commence dosing in AML clinical trial
- Produce cGMP material for RTX-240 and RTX-321 trials from Rubius site
- File RTX-321 IND by year-end
- Advance the autoimmune pipeline

STRONG FINANCIAL POSITION
REALIZING THE POWER OF RED™
A NEW ERA IN CELLULAR MEDICINE