KEROS THERAPEUTICS

KER-050, a novel inhibitor of TGFβ superfamily signaling, induces red blood cell production by promoting multiple stages of erythroid differentiation

Marina Feigenson, Remya Nathan, Chris M Materna, Alana Gudelsky, Evan Lema, Claire C Tseng, ffolliott M Fisher, Jasbir Seehra, Jennifer L Lachey

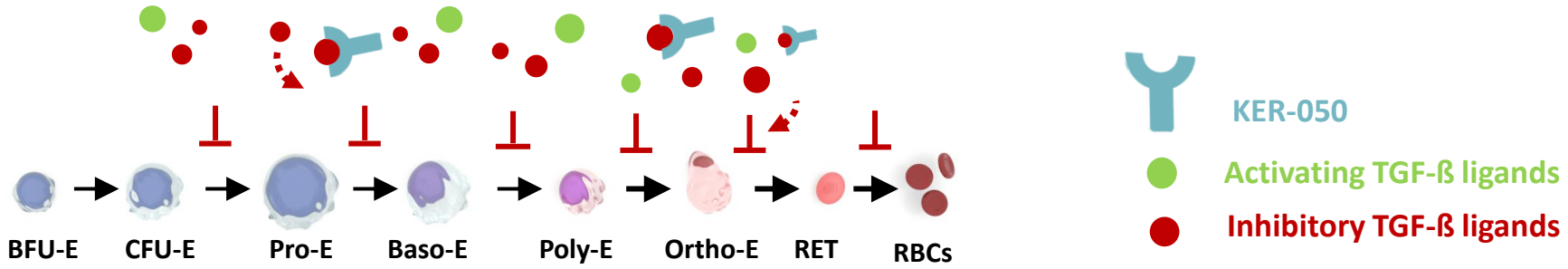
DISCLOSURES OF COMMERCIAL SUPPORT

Name of Company	Research support	Employee	Consultant	Stockholder	Speaker's Bureau	Scientific Advisory Board	Other
Keros Therapeutics		Marina Feigenson					
Keros Therapeutics		Remya Nathan					
Keros Therapeutics		Chris M Materna					
Keros Therapeutics		Alana Gudelsky					
Keros Therapeutics		Claire C Tseng					
Keros Therapeutics		ffolliott M Fisher					
Keros Therapeutics		Jasbir Seehra		Yes			
Keros Therapeutics		Jennifer L Lachey		Yes			

Introduction

- Diseases such as myelodysplastic syndrome (MDS) and myelofibrosis (MF) are characterized by ineffective hematopoiesis, which can result in one or multiple cytopenias.
- Current treatment options to address anemia in these diseases target discreet stages in erythropoiesis, whereas defects leading to ineffective hematopoiesis can occur throughout the pathway. Therefore, a treatment that more globally modulates hematopoiesis has the potential to treat broad patient groups.
- The transforming growth factor beta (TGF-β) superfamily plays a key role in both activating and inhibiting differentiation of erythroid precursors to regulate erythropoiesis.
- KER-050, a modified ActRIIA ligand trap, is designed to promote erythropoiesis by blocking signaling of the inhibitory signals.

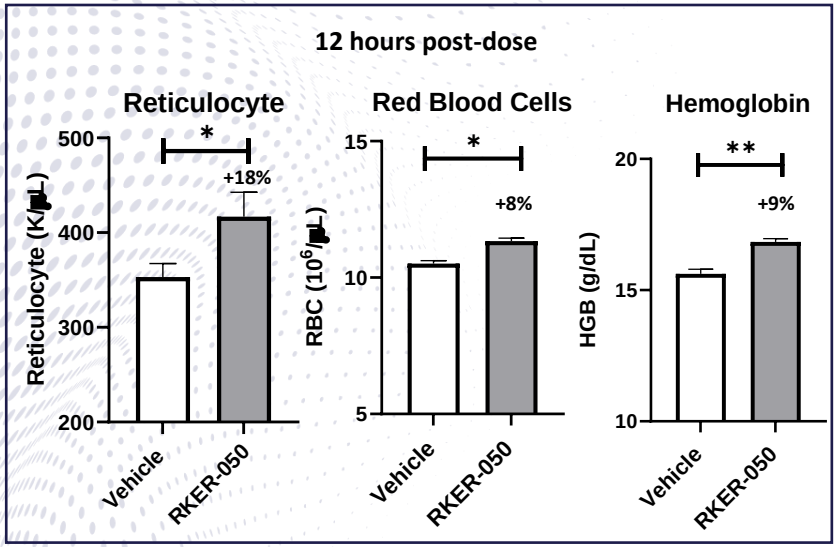
The objective of this study is to characterize the mechanism and time course of KER-050-mediated effects on RBC production and changes in erythroid precursor cell populations in mice

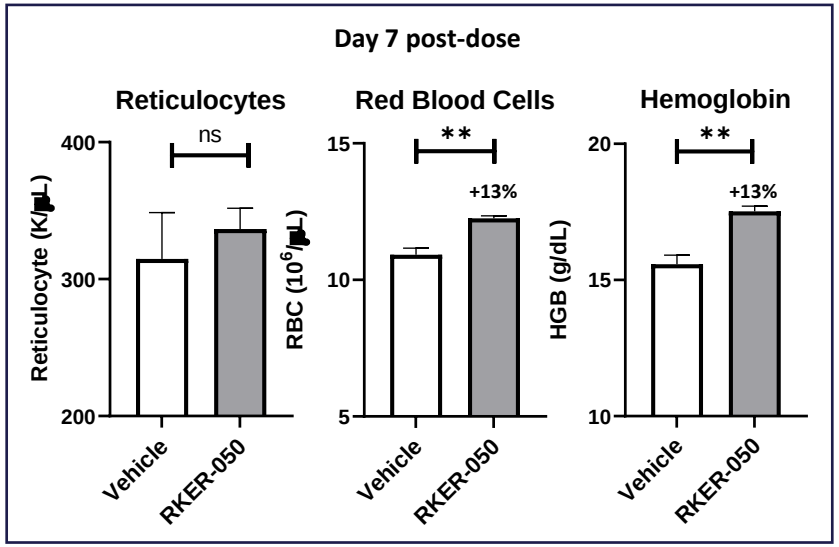


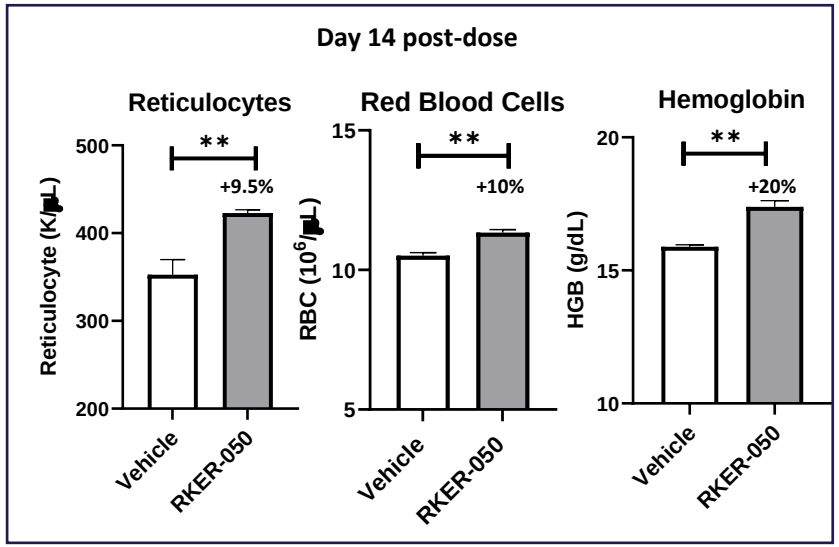
Methods

- Eleven-week-old mice were treated with a single intraperitoneal injection of 10 mg/kg dose of the research form of KER-050 (RKER-050).
- Changes in hematological parameters in peripheral blood and in erythroid progenitors in bone marrow were measured at multiple time points from 12 hours through 51.
- Hematological parameters were measured in peripheral blood by IDEXX BioAnalytical.
- Erythroid progenitors were measured by harvesting bone marrow cells and staining with antibodies against Ter119 (PE-conjugated) and CD71 (FITC-conjugated) and analyzed by flow cytometry.
- BFU-E and CFU-E numbers were analyzed by counting the number of colonies formed by culturing bone marrow cells in a semi-solid media.
- Enucleated erythroid progenitors were measured by staining the bone marrow cells with DRAQ5 nuclear fluorescent dye and evaluating by flow cytometry the number of DRAQ5+ cells in Ter119+ (erythroid cells) population.
- Erythropoietin serum levels were measured by mouse EPO Immunoassay (ELISA).

A single dose of RKER-050 resulted in rapid and sustained increases in hematological parameters

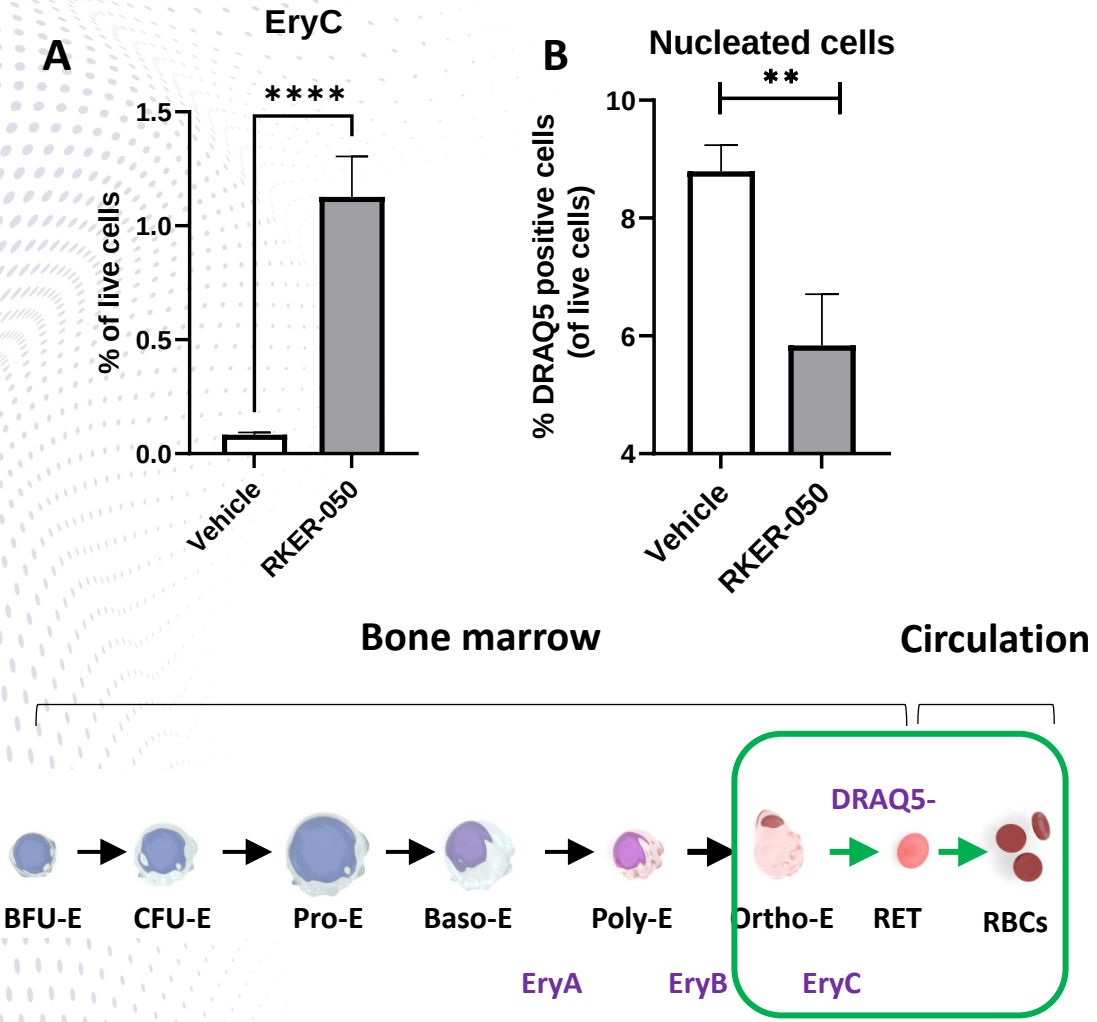






- Mice treated with RKER-050 exhibited 18% increase in RET, 8% in RBCs and 9% in HGB 12 hours after treatment. This timing is consistent with maturation of late-stage progenitors.
- These effects were maintained until at least 14 days post a single dose of RKER-050.

RKER-050 accelerated maturation of late-stage erythrocyte precursors and outflux of reticulocytes into circulation



Treatment with RKER-050:

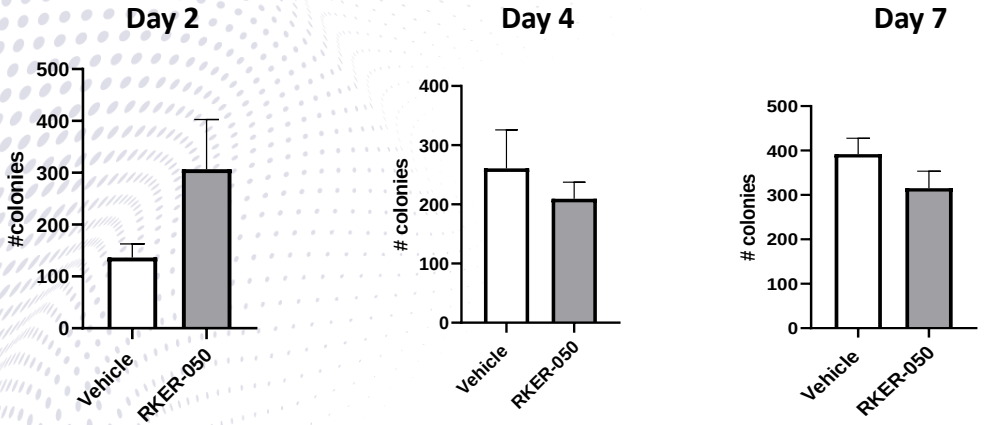
A) **Led to increases** in late orthochromatic erythroblasts (EryC) at 24HR

B) **Reduced** the number of nucleated erythroid cells in the bone marrow at 48HR

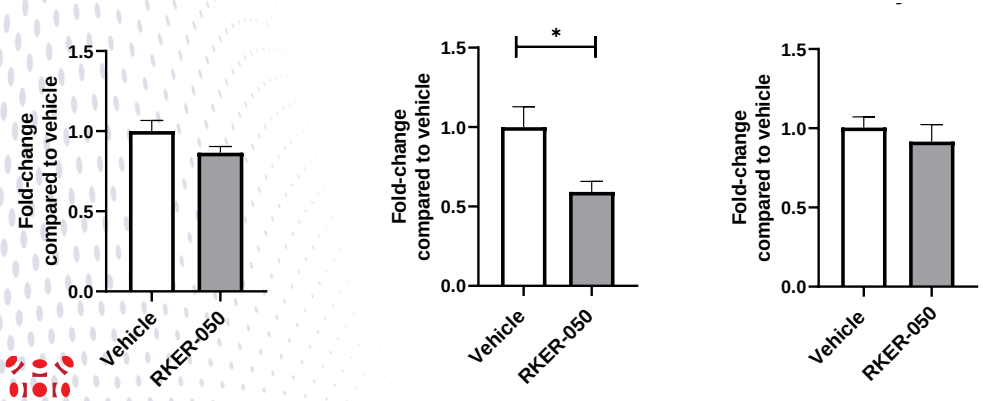
These data demonstrate increased differentiation and outflux of late-stage erythroid progenitors from the bone marrow to the circulation

RKER-050 increased early-stage precursors and numbers of cells progressing through erythropoiesis

CFU-E

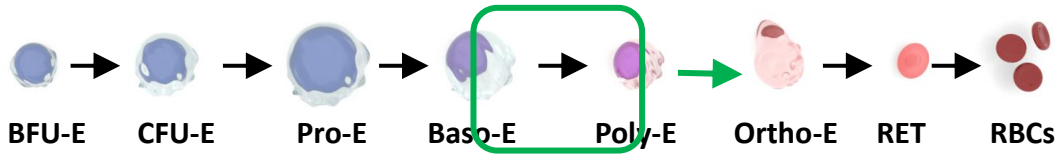


EryB

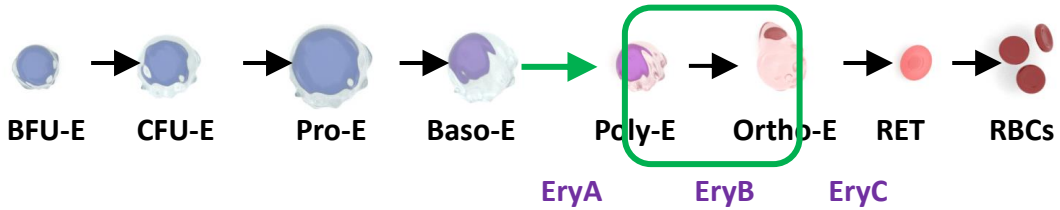


Day 2 – RKER-050 increased early-stage progenitor cells

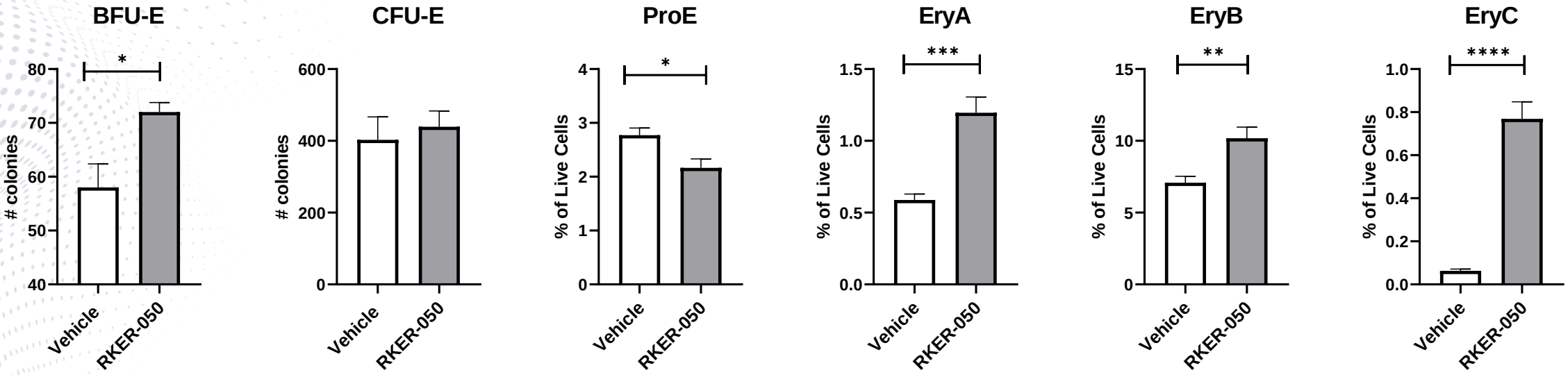
Day 4 – EryB population matured to EryC, depleting its numbers

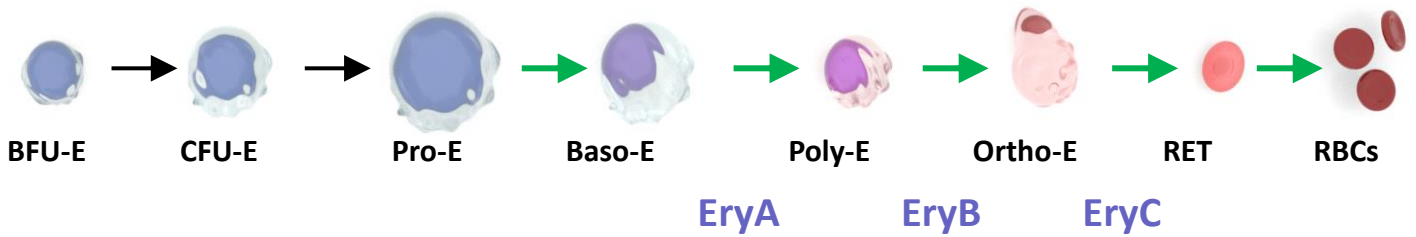


Day 7 – EryB cell pool repleted, demonstrating that the early precursors mobilized on Day 2 differentiate to replenish the progenitor pools



Erythropoietic tone is upregulated 14 days after a single RKER-050 injection

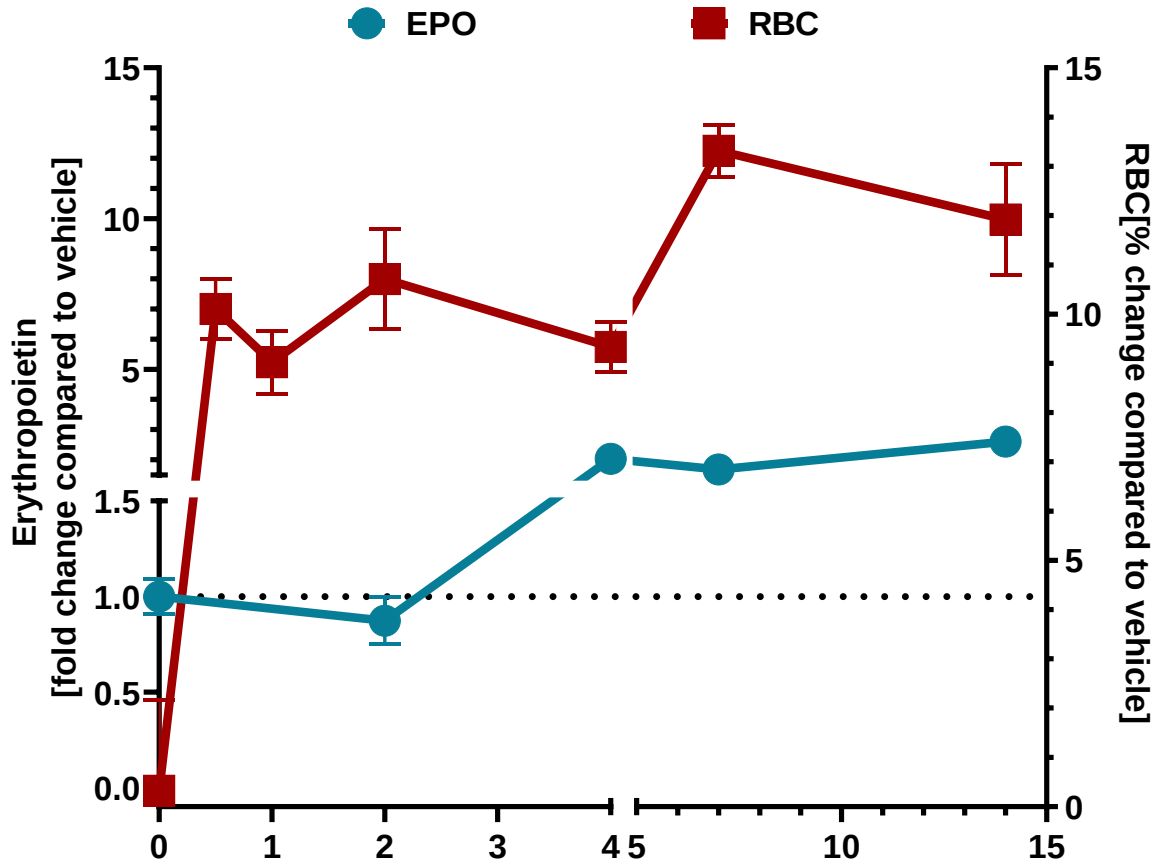




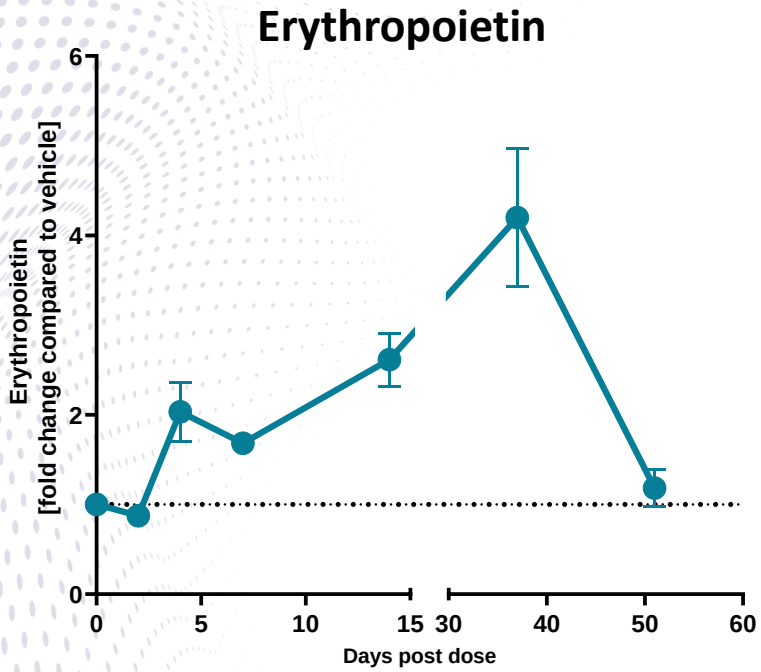
Observed increases in early and late-stage progenitors at day 14 post-single dose support the hypothesis that treatment with RKER-050 has the potential to expand the early progenitor pool that continue to mature and contribute to the overall upregulation of erythropoiesis

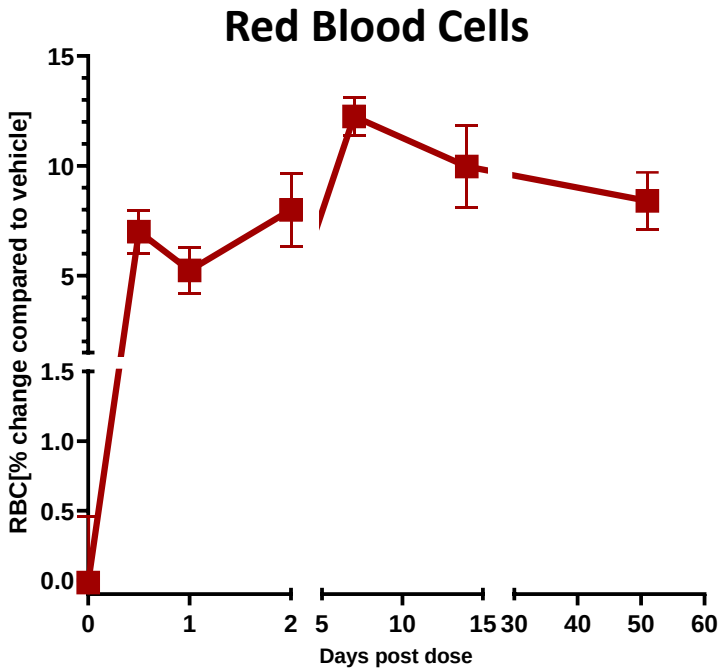
RKER-050 treatment resulted in increase in circulating erythropoietin levels even in the context of increased RBCs

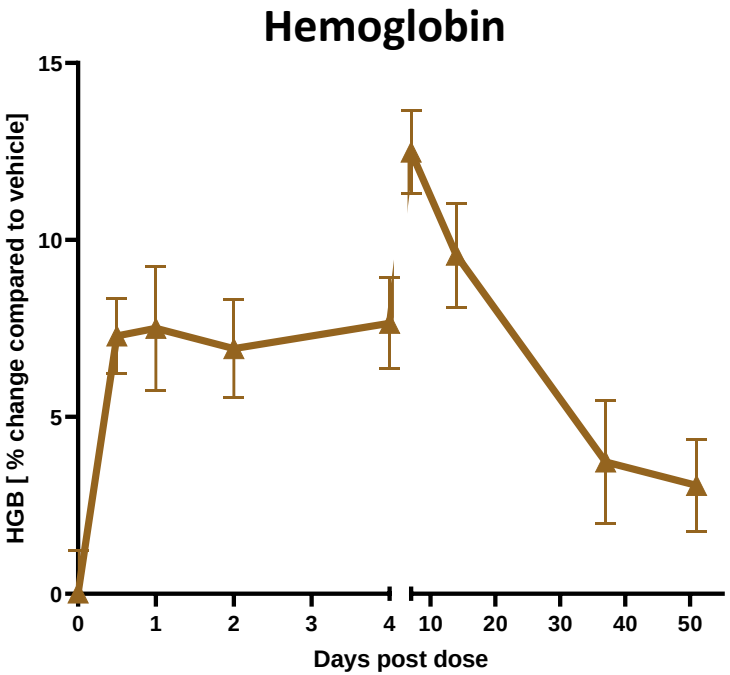
- RKER-050-treated mice exhibited increases in RBCs and EPO levels increased by 2-fold starting at day 4 compared to the vehicle-treated mice.
- Early effects of RKER-050 on RBCs were EPO independent.
- Elevated serum EPO levels starting at day 4 potentially enhances the RKER-050 effect of increasing early progenitor cells, promoting their differentiation and contributing to a sustained effect on erythropoiesis.



A single dose of RKER-050 resulted in a long-lasting effect on erythropoiesis





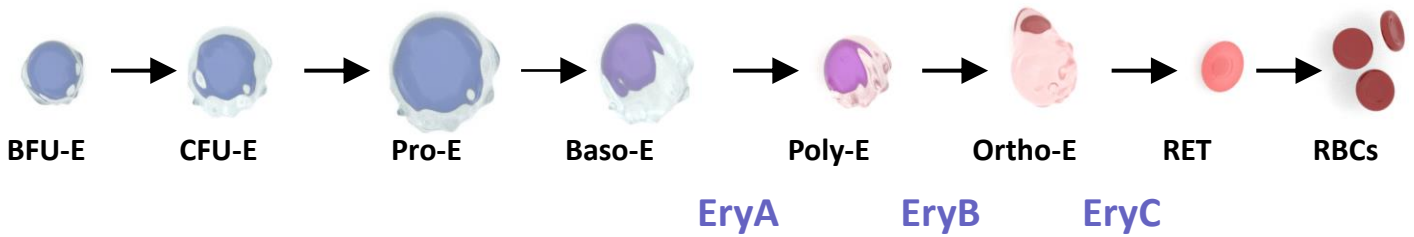


- A single dose of RKER-050 increased serum erythropoietin levels by day 4 through day 37
- A single dose of RKER-050 increased RBC and HGB by 12 hours through at least day 51

Conclusions

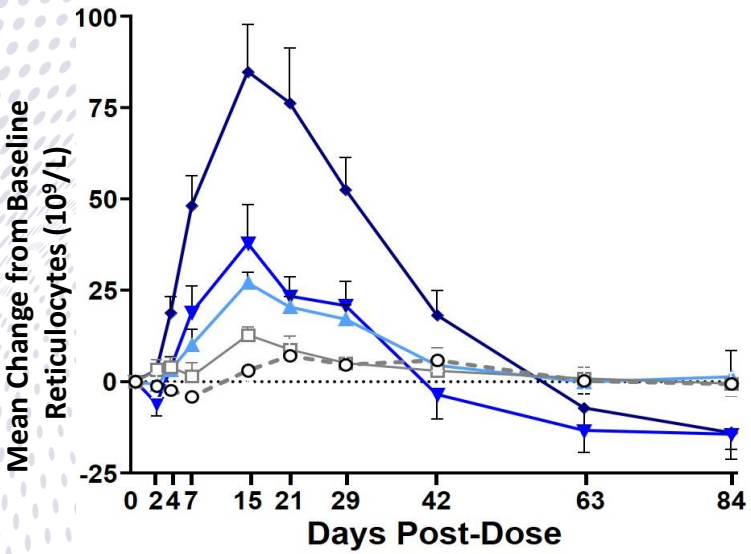
- **Our results suggest that the combination of RKER-050 effects on erythropoiesis potentially contributes to a rapid increase in RBCs and a continuous supply of progenitors that allows for sustained upregulation of erythropoiesis.**

- **The ability of KER-050 to potentially target multiple stages along the erythropoiesis cascade makes it a potential therapeutic candidate for diseases that cause anemia due to ineffective erythropoiesis, including myelodysplastic syndrome and myelofibrosis, where defects can arise throughout the erythropoietic pathway.**

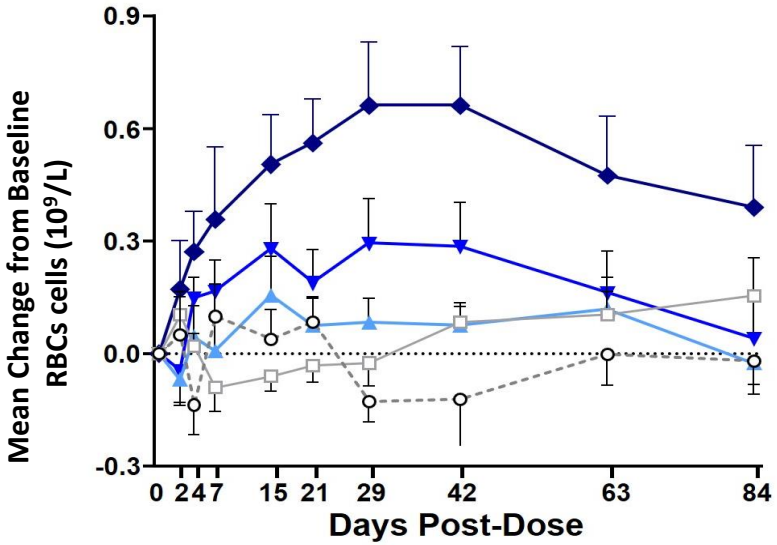


KER-050 exhibited robust effect on erythropoiesis in healthy human volunteers

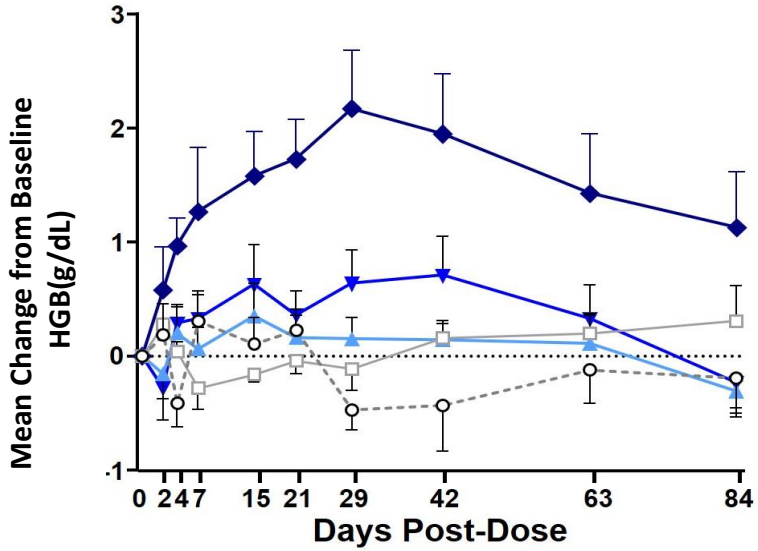
Reticulocytes



Red Blood Cells



Hemoglobin



Placebo
0.05 mg/kg
0.5 mg/kg
1.5 mg/kg
4.5 mg/kg

- Rapid increase in reticulocytes potentially indicative of effect on terminal differentiation
- Observed increases in HGB beyond day 15 potentially indicative of effect on early precursors
- Change in RBCs was durable for up to 84 days after single KER-050 dose

KER-050 is currently in Phase 2 clinical trial in MDS, and Phase 2 trial in MF is scheduled to start in 2021