GBT1118, A POTENT ALLOSTERIC MODIFIER OF HEMOGLOBIN OXYGEN AFFINITY INCREASES TOLERANCE TO HYPOXIA IN MICE

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Introduction

Tissue hypoxia may occur under conditions of reduced ambient oxygen (O2) tensions (eg. at high altitude), increased O2 demand during intense exercise or in lung disease where O2 loading of hemoglobin (Hb) is compromised 1,2. Under these conditions, arterial O2 saturation decreases leading to reduced O2 delivery to tissues and tissue hypoxia when O2 consumption is greater than O2 delivery 1. Short term adaptation to moderate hypoxia mainly involves a reduction in Hb O2 affinity and corresponding increase in O2 unloading to tissues. However, long-term genetic adaptation to extreme hypoxia involves Hb with an increase in O2 affinity, maintaining O2 loading in the lungs and thereby preserving tissue oxygenation16. GBT1118 is a novel orally-bioavailable small molecule that binds covalently and reversibly via Schiff base to the N-terminal valine of the Hb alpha chain and allosterically modulates Hb O2 affinity.

Hypothesis:

We tested the hypothesis that pharmacologically increasing Hb O2 affinity with GBT1118 can augment O2 transport during severe hypoxia.

Methods

Study design: Mice were fitted with a dorsal skinfold window chamber for direct visualization of an intact microvascular bed 3,4.

Mice were then dosed orally with GBT1118 (70 or 140mg/kg) or vehicle only and exposed to stepwise hypoxia by decreasing the fraction of inspired O2 (FiO2) from normoxia (baseline=21% O2) to 15%, 10% and 5% O2 hypoxia. Mice were kept at each hypoxic level for 0.5 hours and an additional 1.5 hours at extreme hypoxia (5% O2) to assess tolerance to hypoxia (survival). To prevent animal stress or discomfort, hypoxia was discontinued if blood pressure dropped below 60 mmHg, and the animal was considered non-survived.

Measurements:

• Hemoximetry was used to measure Hb O2 affinity.
• Blood pressure and heart rate were recorded continuously via a carotid catheter.
• Arterial blood gases measured with rapidlab 248 gas analyzer.
• Lactate was measured using lactate dehydrogenase assay.
• Arterial O2 saturation was measured with a co-oximeter (IL482).
• For microvascular analysis, mice with the window chamber were fixed to the microscopic stage for transillumination with an intravital microscope 4.
• Tissue hypoxia was evaluated via pimonidazole staining.

Results

GBT1118 increased Hb O2 affinity and arterial O2 saturation during hypoxia

Panel A) GBT1118 decreased blood p50 (P50) at which blood is 50% saturated with O2 relative to control indicating increased Hb O2 affinity. Panel B) Arterial blood PO2 decreased in all treatment groups during hypoxia. Panel C) GBT1118 increased arterial blood O2 saturation (SaO2) relative to control indicating an increase in O2 uptake during hypoxia. (Mean ± SD shown; N=6) Baseline (BL)=21%O2 or normoxia

GBT1118 preserved microvascular blood flow and tissue oxygenation during hypoxia

Panel A) GBT1118 maintained arterial blood vessel diameter relative to BL during extreme hypoxia. Blood vessel diameter of control group decreased relative to baseline (BL) during extreme hypoxia. Panel B) GBT1118 increased blood flow relative to control during extreme hypoxia. (Mean ± SD shown; N=6) BL=21%O2 or normoxia

GBT1118 reduced tissue hypoxia and improves survival

Panel A) GBT1118 reduced hypoxic tissues positively stained by pimonidazole relative to control during extreme hypoxia. Panel B) GBT1118 improved survival during extreme hypoxia. (Mean ± SD shown; N=6)

Conclusions

GBT1118 increased Hb O2 affinity, improved O2 uptake in the lungs and thereby improved O2 delivery to tissues and survival during hypoxia.

Hb O2 affinity modulators represent a promising and novel therapeutic strategy for the treatment of hypoxemia associated with lung disorders.

References