#1064

Treatment Success in Reducing Recurrent *Clostridioides difficile* Infection with Investigational Live Biotherapeutic RBX2660 was Associated with Microbiota Restoration: Consistent Evidence from a Phase 3 Clinical Trial

Ken Blount PhD [1], *Dana Walsh PhD* [1], *Carlos Gonzalez* [2], *Bill Shannon PhD MBA* [2]

[1] Rebiotix Inc., A Ferring Company, Roseville, MN, USA; [2] BioRankings LLC, St. Louis, MO, USA

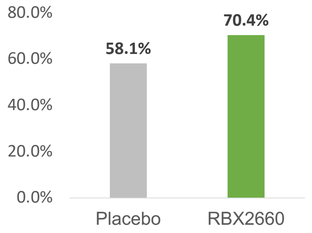
BACKGROUND

- Several investigational microbiota-based live biotherapeutics are in clinical development for reducing recurrence of *Clostridioides difficile* infection (rCDI)
- RBX2660, a liquid suspension of a broad consortium of microbiota, which includes Bacteroidetes and Firmicutes, has been evaluated in >600 participants in 6 clinical trials
- Here we report that RBX2660 induced significant shifts to the intestinal microbiota of treatment responsive participants in PUNCH CD3—a Phase 3 randomized, double-blinded, placebo-controlled trial

METHODS

- PUNCH CD3 (NCT03931941) - a Phase 3 randomized, double-blinded, placebo controlled (2:1) study of RBX2660 versus placebo to reduce recurrent CDI.
- Treatment success was defined as absence of CDI recurrence at 8 weeks after blinded investigational treatment.
- The study utilized an innovative, a priori Bayesian statistical plan that leveraged data from a prior phase 2B trial and included a prespecified threshold of success, defined as a minimum of 0.975 posterior probability of superiority for RBX2660 versus placebo.
- Participants voluntarily submitted stool samples prior to blinded study treatment (baseline), and 1 week (1W), 4 weeks (4W), 8 weeks (8W), 3 months (3M) and 6 months (6M) after receiving study treatment.
- Samples were extracted and sequenced using shallow shotgun methods (Diversigen, Minneapolis, MN)
- Operational taxonomic unit (OTU) data were used to calculate relative taxonomic abundance, alpha diversity, and the Microbiome Health Index (MHI)
- MHI is a biomarker of antibiotic-induced dysbiosis and restoration - and defined as (Bacteroidia + Clostridia) / (Gammaproteobacteria + Bacilli)

RBX2660 CLINICAL EFFICACY



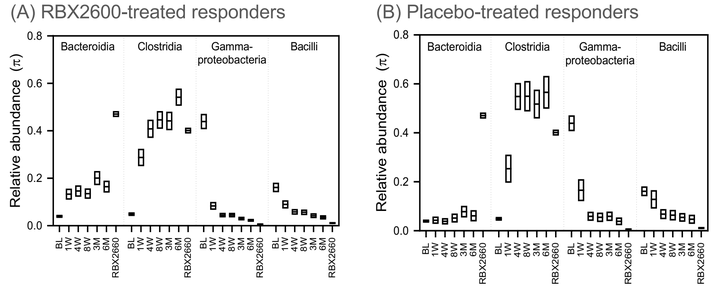
RBX2660 MET THE PRESPECIFIED THRESHOLD OF SUCCESS, with **a 0.986** posterior probability of superiority for RBX2660 vs placebo; treatment success was 70.4% for RBX2660 and 58.1% for placebo.

(reported previously by Lee et al, DDW 2021)

MICROBIOME ANALYSIS

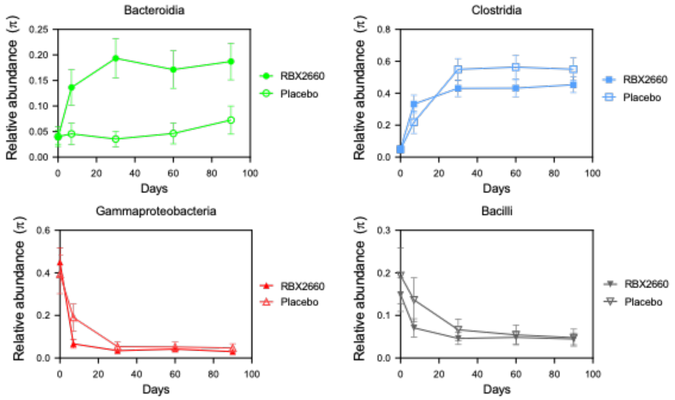
Samples from the blinded phase were available from 179 participants. Samples received after an adjudicated failure event were excluded. All timepoints were not available from each participants.

TREATMENT RESPONDERS DEMONSTRATED RESTORATION OF KEY TAXA



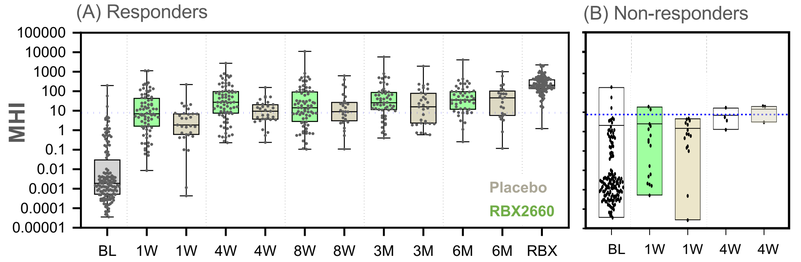
RBX2660-treated responders demonstrated more rapid and more extensive recovery of Bacteroidia and decreased Gammaproteobacteria (A) relative to placebo-treated responders (B).

EVALUATION OF LONGITUDINAL PATIENT-MATCHED SAMPLES DIFFERENTIATES RESTORATION BETWEEN RBX2660-TREATED RESPONDERS AND PLACEBO-TREATED RESPONDERS



Changes among RBX2660-treated responders were significantly different than among placebo-treated responders (p<.001, DM Repeat, 31 participants).

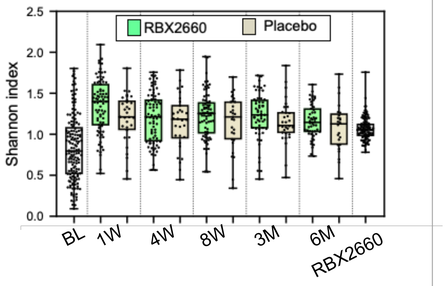
MICROBIOME HEALTH INDEX (MHI) WAS INCREASED AMONG RESPONDERS



Rapid increase of MHI within 1W after treatment (A). MHI among non-responders did not increase as much as responders (B). *Disclaimer: post-treatment samples from non-responders are underrepresented in this analysis (n=43) due to majority of participants recurring before 1W.*

ALPHA DIVERSITY SIGNIFICANTLY INCREASED AFTER TREATMENT IN RESPONDERS

- Alpha diversity is significantly higher than baseline at all post-treatment time points among responders (p<.001, non-parametric test)
- Alpha diversity trended higher in RBX2660 vs placebo responders at 1 week after treatment



CONCLUSIONS

- The PUNCH CD3 clinical trial reported superiority of RBX2660 over placebo in reducing CDI recurrence.
- Among PUNCH CD3 clinical responders, RBX2660 significantly increased taxa associated with health and decreased taxa associated with *C diff* pathology, and these shifts were durable to at least 6 months.
- These clinically-correlated microbiome shifts are consistent with results from multiple trials of RBX2660.