STX-003: Cancer Immunotherapy with systemic delivery of mRNA utilizing programmable genetic circuits for precise regulation of IL-12 expression and reduced toxicity

Gautam N. Shenoy, Wen Allen Tseng, Emily A. Clinton, Ryan A. Patenaude, Wuhbet Abraham, Zhijun Su, Madhuri Dey, Justin H. Letendre, Maeve M. Pattie, John T. Sexton, Emily Tjon, Nandita Satish Menon, Isma Zarin, Breanna Burgett, Aidan Coia, Akshay Mavani, Weiyu Zhao, Kevin Shi, Alexander C. Lemaire, Julian Quiñones, Ryan T. Sowell, Jaspreet S Khurana, Joseph Barberio, Prashant R. Nambiar, Jacob R. Becraft, Tasuku Kitada

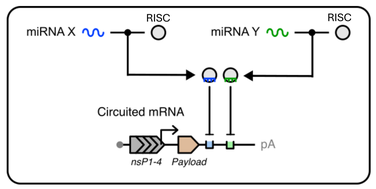
Strand Therapeutics Inc. 20 Overland St, Boston, MA 02215, USA

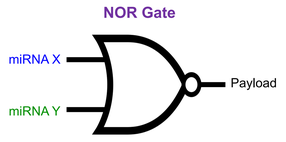
ABSTRACT # 3472

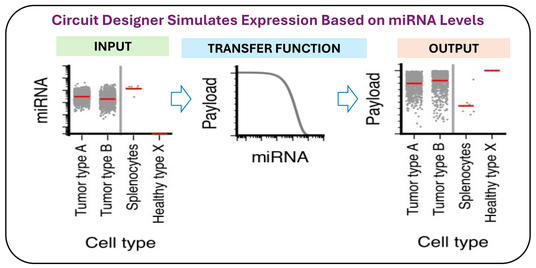
INTRODUCTION

Immunotherapies have revolutionized the treatment of solid tumors, which represent 90% of adult human cancers. Therapeutic delivery of IL-12, a potent immunostimulatory cytokine, is robustly effective in preclinical models. However, systemically delivered IL-12 is poorly tolerated potentially due to its off-target activity. To overcome this challenge, we developed an mRNA platform that utilizes programmable genetic circuits to regulate the expression of an encoded protein in response to microRNAs (miRNAs), enabling precise control of payload expression in the target tissues. Using this platform we developed STX-003, a systemically delivered self-replicating mRNA encoding IL-12 which bears programmable genetic circuitry to limit the expression of payload in off-target tissues while preserving expression in the tumor.

IN SILICO MODELING INFORMS CIRCUIT SELECTION FOR STX-003



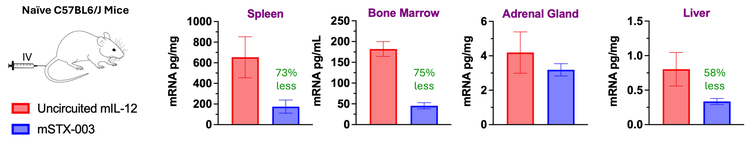




INPUT		OUTPUT
miRNA X	miRNA Y	Payload
0	0	1
0	1	0
1	0	0
1	1	0

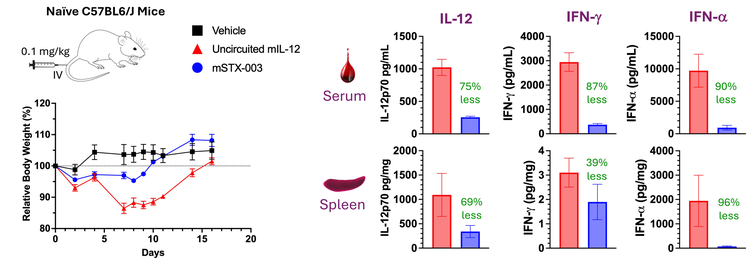
- Unique miRNA signatures were identified using *in silico,* and high throughput *in vitro* screening.
- Candidate circuit was selected based on the cell and tissue types to be detargeted.
- Circuited mRNA were formulated into LNPs and administered IV in mouse models to conduct proof-of-concept studies.

CIRCUIT REDUCES mRNA LEVELS IN OFF-TARGET TISSUES



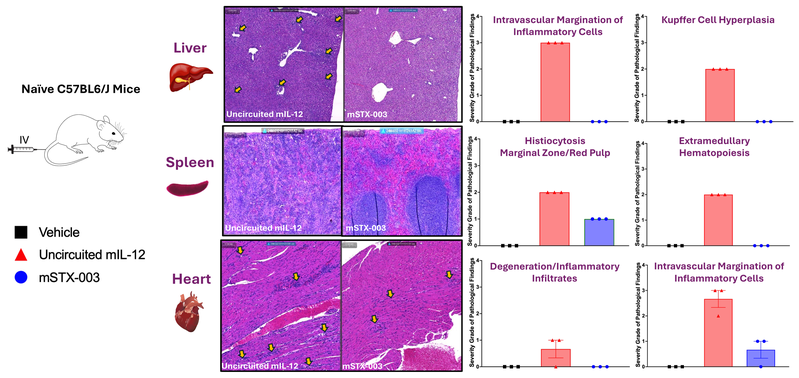
- The presence of the circuit robustly knocked down mRNA levels in multiple off-target tissues in naïve mice. Data shown is at Tmax.

CIRCUIT IMPROVES TOLERABILITY BY LOWERING SYSTEMIC LEVELS OF IL-12 AND DRUG-ASSOCIATED BIOMARKERS



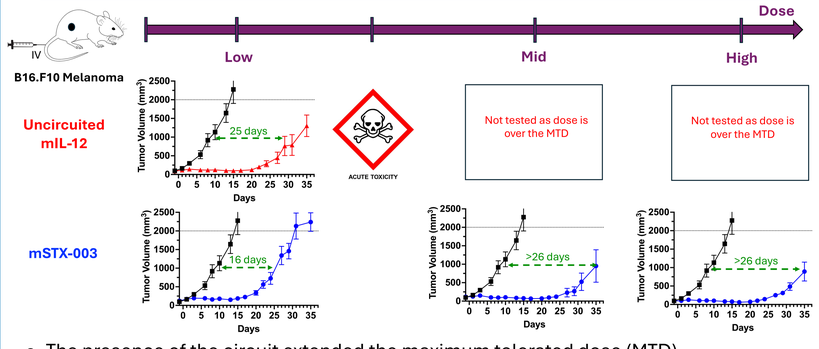
- The presence of the circuit lowered test article-related body weight loss as well as recovery time in naïve C57BL6/J mice.
- This correlated with lower levels of IL-12, IFN-γ & IFN-α in circulation as well as in the spleen and other off-target tissues. Data shown is at the respective Tmax.

CIRCUIT PROTECTS TISSUES FROM TOXICITY ASSOCIATED WITH SYSTEMIC IL-12 EXPOSURE



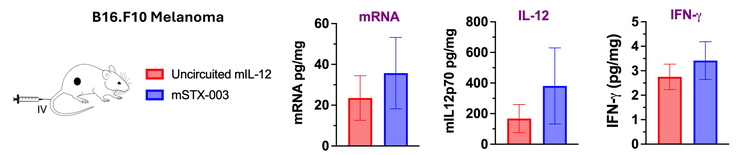
- The presence of the circuit alleviated histopathological findings associated with systemic IL-12 exposure in liver, spleen & heart of naïve mice seen at 17 days post-dose.

CIRCUIT EXPANDS THERAPEUTIC INDEX IN A MOUSE TUMOR MODEL



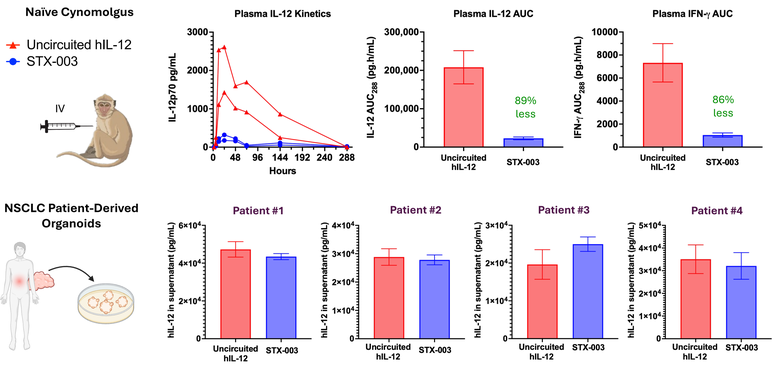
- The presence of the circuit extended the maximum tolerated dose (MTD).
- A dose-dependent increase in tumor control and survival was seen with mSTX-003.

CIRCUIT PRESERVES PAYLOAD EXPRESSION IN TUMORS



- The levels of mSTX-003 mRNA, IL-12 and IFN-γ levels in B16.F10 tumors were comparable with or without the circuit. Data shown is at the respective Tmax.

STX-003 REPRESSES PAYLOAD IN NON-HUMAN PRIMATES WHILE PRESERVING EXPRESSION IN HUMAN TUMORS



- Intravenous administration of STX-003 was well tolerated in Cynomolgus monkeys.
- The presence of the circuit resulted in an 89% reduction in circulating hIL-12 and an 86% reduction in circulating IFN-γ.
- Transfection of circuited and uncircuited mRNA into patient-derived NSCLC organoids yielded comparably robust expression of hIL-12 payload.

CONCLUSIONS

- **STX-003 expresses its IL-12 payload in mouse tumors while selectively repressing its expression in non-target tissues, improving tolerability and protecting these tissues from off-target toxicity.**
- **STX-003 expands the therapeutic index in mouse tumor models by extending the MTD while maintaining effective tumor control upon systemic administration.**
- **STX-003 circuit functions robustly by repressing payload expression in Cynomolgus monkeys, and is well tolerated at the tested doses.**
- **These data demonstrate the potential of STX-003, our programmable mRNA circuit platform, for expanding the therapeutic index of potent drugs that are limited by systemic toxicities associated with off-target expression.**