A half-life extended, tumor selective IL-12 activated tumor infiltrating immune cells and demonstrated anti-tumor activity in the MC38 syngeneic mouse model


Waltham, MA, USA

Intestinal-12 (IL-12) is a proinflammatory cytokine that bridges innate and adaptive immunity via induction of T helper 1 differentiation and promoting cytolytic activity of natural killer, as well as T cells. IL-12 has demonstrated potent anti-tumor activity in syngeneic mouse models and promising anti-tumor activity in humans. However, clinical development of IL-12 has been limited by severe systemic toxicities. To overcome toxicity and improve the therapeutic index of IL-12 in a clinical setting, XTX301 was engineered as a potent, half-life extended and masked IL-12. The masking domain of XTX301 is designed to pharmacologically inactivate IL-12 in non-tumor-infiltrating cells while in plasma from healthy donors. XTX301, a half-life extended tumor-selective IL-12, demonstrated cleavage in plasma from healthy donors, RCC, melanoma, or H&N cancer patients. XTX301 was incubated with plasma samples at 37°C for 11 days. The cleavage was evaluated by Western Blot. XTX301 was activated by cells from human primary tumors across a broad range of solid tumor types (with cleavage occurring in 50-85% of samples tested).

**XTX301 cleavage by cells from human primary tumors**

**Ex vivo proteolytic activation of XTX301 by human plasma and tumors**

**Figure 7:** Ex vivo cleavage analysis of XTX301 by six solid tumor types. (A) Study Design: Single dose XTX301 was incubated with the cells at 37°C for 24 hrs. The cleavage was evaluated by Western Blot. (B) XTX301 was activated by cells from human primary tumors across a broad range of solid tumor types (with cleavage occurring in 50-85% of samples tested).

**XTX301 was minimally cleaved by human plasma**

**Figure 8:** Ex vivo cleavage analysis of XTX301 by human plasma from healthy donors, RCC, melanoma, prostate or H&N cancer patients. XTX301 was incubated with plasma samples at 37°C for 11 days. The cleavage was evaluated Western Blot. No significant cleavage of XTX301 was observed in plasma from healthy donors.

**Conclusions**

XTX301, a half-life extended tumor-selective IL-12 molecule, demonstrated cleavage in human tumor samples but was minimally cleaved in human plasma derived from patients with tumor burden, indicating tumor-selective activation. A mouse surrogate of XTX301, mXTX301, induced immune activation in the tumor microenvironment of a syngeneic mouse model. Furthermore, in vivo data demonstrated that XTX301 potently inhibited tumor growth in a mouse model with improved tolerability compared to a non-tumor-selective IL-12 molecule. In conclusion, XTX301 has the potential to achieve potent patient anti-tumor activity while widening the potential therapeutic index of IL-12 treatment.