CBX-12: A low pH targeting alphalex™-exatecan conjugate for the treatment of solid tumors

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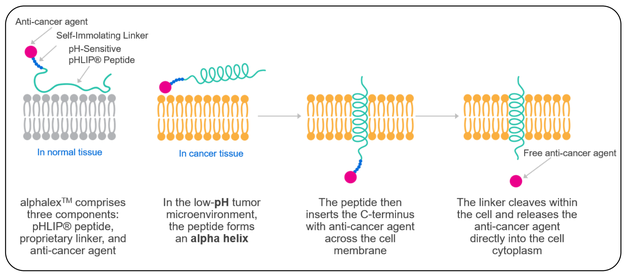
Abstract

Topoisomerase inhibitors are potent DNA damaging agents with great potential as anti-cancer drugs for a wide range of solid tumors. However, dose-limiting toxicities such as myelosuppression and gastric toxicity have prevented them from reaching their full clinical potential. Targeting topoisomerase inhibitors with antibodies (i.e., antibody-drug conjugates; ADCs) is one solution to enhance the therapeutic window of these agents, but this approach typically limits applicability to a small subset of patients with tumors expressing the target antigen.

We have recently developed the alphalex™ tumor-targeting platform to overcome the limitations of ADC-based therapeutic strategies. Rather than targeting a specific antigen, alphalex™ consists of a unique variant of pH-Low Insertion Peptide (pHLIP®; References 1-3) which targets the low pH environment of the tumor, a universal feature characteristic of all tumors due to the Warburg effect. These alphalex™ conjugates form an alpha helix only in low pH conditions, allowing for insertion of the peptide within the cancer cell membrane, delivery of C-terminal warheads across the membrane, and subsequent intracellular release of the agent via glutathione reduction of the linker, thereby allowing for tumor-specific intracellular delivery in an antigen-independent manner.

Cybrexa has synthesized and developed CBX-12, an alphalex™ conjugate of the potent topoisomerase inhibitor, exatecan. CBX-12 provides additional proof of mechanism to the alphalex™ platform by displaying remarkable tumor-targeting and efficacy in preclinical models. These superior properties of CBX-12 allow us to greatly enhance efficacy while avoiding dose-limiting bone marrow toxicity when administering equimolar doses of unconjugated exatecan. We have demonstrated that our lead alphalex™ candidate, CBX-12, is both safe and has potent anti-tumor activity in preclinical models, and we plan to rapidly move forward with the clinical development of CBX-12.

alphalex™ Enables Antigen-Independent Tumor Targeting



Specific Release of Exatecan Payload in Tumor by CBX-12

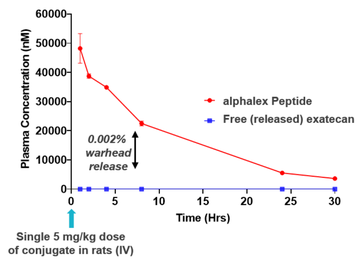


Figure 1. Plasma PK of alphalex peptide and released exatecan after a single i.v. dose of 5mg/kg CBX-12 in the rat. Less than 0.002% of the exatecan warhead is released after 30h in circulation. Data are expressed as means ± SEM.

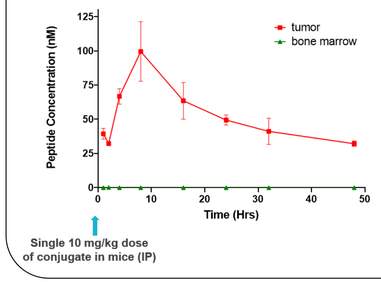


Figure 2. Tumor and bone marrow PK of alphalex peptide after a single i.p. dose of 10mg/kg CBX-12 in mouse. Data are expressed as means ± SEM.

CBX-12 Avoids the Dose Limiting Bone Marrow and GI Toxicities of Exatecan

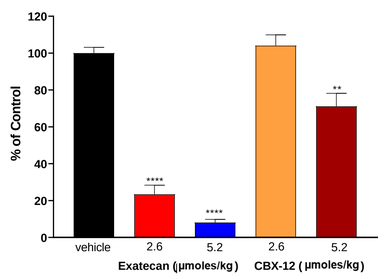


Figure 3. Total bone marrow counts from the femurs of tumor bearing nude mice after dosing of 2.6 and 5.2µmoles/kg of either CBX-12 (equivalent to 10, 20mg/kg CBX-12) or unconjugated exatecan (equivalent to 1.15, 2.3mg/kg exatecan) dosed QDx4 i.p. Data are expressed as means ± SEM.

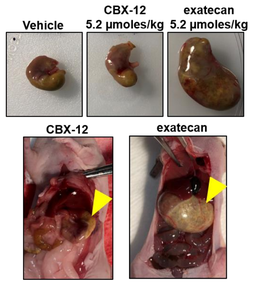


Figure 4. Stomachs of tumor bearing nude mice either excised (top row) or *in situ* (bottom row; highlighted by arrow) after dosing of 5.2µmoles/kg of either CBX-12 (equivalent to 20mg/kg CBX-12) or unconjugated exatecan (equivalent to 2.3mg/kg exatecan) dosed QDx4 i.p.

CBX-12 Tumor Targeting Enables Durable and Potent Anti-Tumor Activity

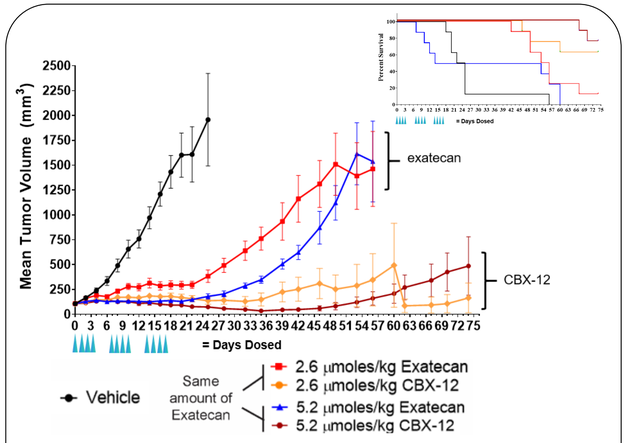


Figure 5. Comparison of efficacy of dosing equimolar amounts of either CBX-12 or unconjugated exatecan in nude mice bearing HCT116 colorectal cancer flank tumors. Animals were dosed QDx4/week i.p. for three weeks. Inset displays Kaplan Meier survival curve. Data are expressed as means ± SEM.

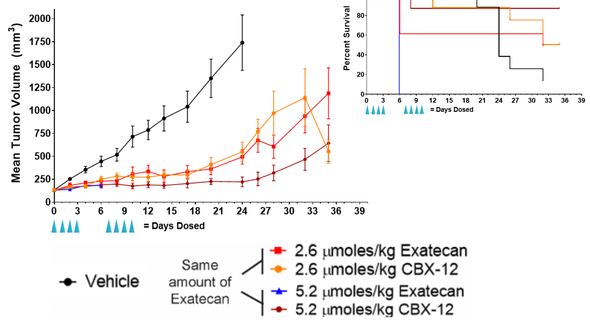


Figure 6. Single agent efficacy of CBX-12 in nude mice bearing MKN45 HER2 negative gastric cancer flank tumors. Animals were dosed QDx4/week i.p. for two weeks. Inset displays Kaplan Meier survival curve. Data are expressed as means ± SEM.

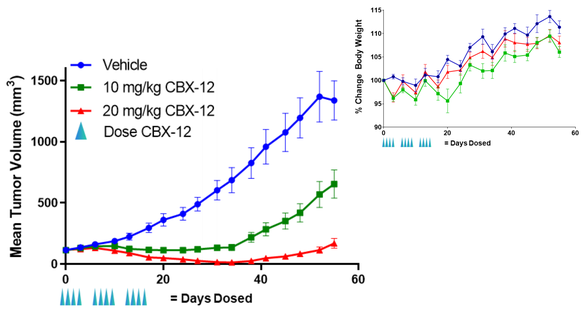


Figure 7. Single agent efficacy of CBX-12 in SCID mice bearing JIMT-1 HER2 positive, trastuzumab-refractory breast cancer flank tumors. Animals were dosed QDx4/week i.p. for three weeks. Inset displays percent change in body weight. Data are expressed as means ± SEM.

CBX-12 Is Effective as Both Single Agent and in Combination in a TNBC Model

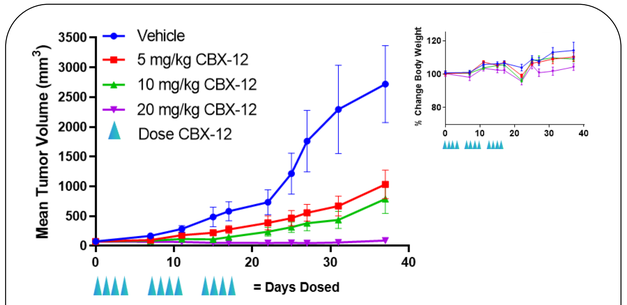


Figure 8. Single agent efficacy of CBX-12 in nude mice bearing MDA-MB-231 triple negative breast cancer flank tumors. Animals were dosed QDx4/week i.p. for three weeks. Inset displays percent change in body weight relative to day 0. Data are expressed as means ± SEM.

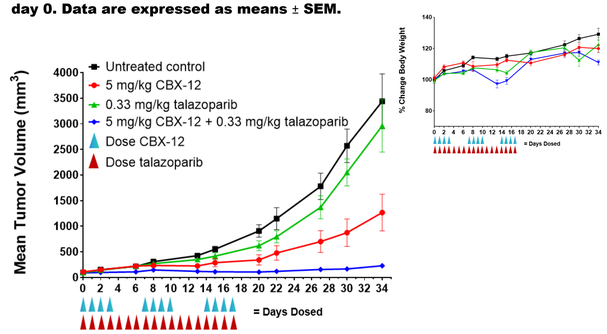


Figure 9. Combination efficacy of CBX-12 and talazoparib in nude mice bearing MDA-MB-231 triple negative breast cancer flank tumors. Animals were dosed QDx4/week i.p. for three weeks with CBX-12 and QDx18 p.o. with talazoparib. Inset displays percent change in body weight relative to day 0. Data are expressed as means ± SEM.

Conclusions

- **CBX-12 displays exquisite plasma stability and tumor targeting ability *in vivo*.**
- **CBX-12 does not display the severe bone marrow and gastric toxicities that limited the clinical utility of exatecan.**
- **CBX-12 is safe and displays potent, antigen independent anti-tumor activity in multiple preclinical models.**
- **Cybrexa plans to rapidly move forward with the clinical development of CBX-12 as our lead candidate.**

References

[1] Rather than targeting a specific antigen, alphalex™ includes a pHLIP® peptide. pHLIP® peptides are a family of pH-Low Insertion Peptides that target acidic cell surfaces. pHLIP® was developed at Yale University and the University of Rhode Island, and is exclusively licensed to pHLIP, Inc

[2]Wyatt LC, Lewis JS, Andreev OA, Reshetnyak YK, Engleman DM. Applications of pHLIP Technology for Cancer Imaging and Therapy. Trends Biotechnol. 2017. Jul; 35(7):653-664.

[3]Wyatt LC, Moshnikova A, Crawford T, Engleman DM, Andreev OA, Reshetnyak YK. Peptides of pHLIP family for targeted intracellular and extracellular delivery of cargo molecules to tumors. Proc Natl Acad Sci USA. 2018 Mar 20;115(12):E2811-2818.