

Modulating Ceramides and Sphingosine-1-Phosphate

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Summary: BXQ-350 is a nanovesicle formulation of Saposin C, an allosteric activator of sphingolipid metabolism, which was investigated in a Phase 1 dose-escalation safety study in all-comers cancer patients with advanced solid malignancies (NCT02859857). Results showed that BXQ-350 is well tolerated and exhibits signs of potential single agent clinical activity across multiple tumor types. Analysis of preclinical and clinical samples demonstrated that BXQ-350 increases ceramides and decreases sphingosine-1-phosphate (S1P) while it may positively impact the innate and adaptive immune systems.

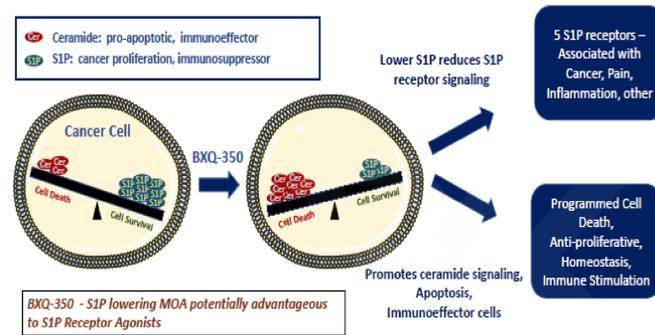
Acknowledgement: Patients who participated in the trial and their families, clinicians and staff at investigational sites, Bexion's personnel

1. Background: Ceramides and S1P are sphingolipids, a class of bioactive signaling lipids implicated in many cellular pathways and cellular processes:

- ceramides are pro-apoptotic, down regulate oncogenic pathways and stimulate immuno-effector cells (1)
- S1P promotes survival and proliferation of cancer cells, activates oncogenic pathways, and stimulates immuno-suppressor cells (2).

Investigation of BXQ-350's *ex vivo* properties indicated, thus far, that

2. BXQ-350: BXQ-350 is a nanovesicle formulation of Saposin C, an allosteric activator of sphingolipid metabolism, and dioleoyl-phosphatidylserine (DOPS).



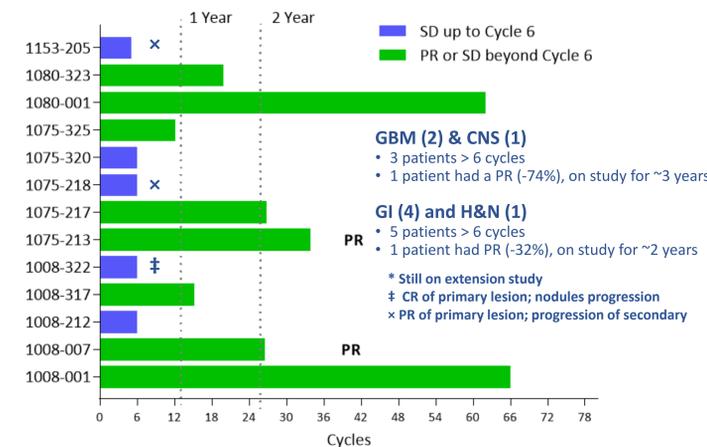
3. Phase 1 Study: A Phase 1 dose-escalation safety study in all-comers cancer patients with advanced solid malignancies (NCT02859857) investigated the safety profile and potential clinical activity of BXQ-350 as a single agent:

- Accelerated dose escalation from 0.7 mg/kg up to 2.4 mg/kg
- Disease assessment at Cycle 2, 4, 6...

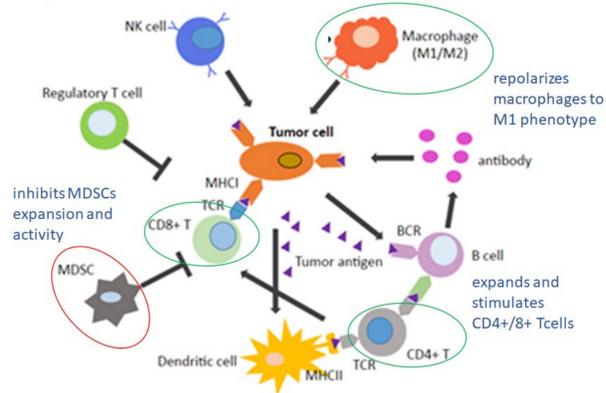
Once Daily IV Infusion over 45 min ± 15 min			
Cycle 1 Week 1	Cycle 1 Week 2	Cycle 1 Week 3 & Week 4	Cycle 2 & thereafter
Days 1-5 (5 consecutive days)	3x/week (every other day)	Once every 7 days (± 3 days)	Once every 28 days (± 3 days)

4. Safety Profile: BXQ-350 is safe and well-tolerated; no DLT observed and a MTD was not reached. The three most common adverse events reported are fatigue, nausea and flushing/infusion reactions.

5. Efficacy Profile: Results show that 11 patients (~15% of evaluable patients) had a potential clinical benefit and 8 of these patients had a PFS > 6 months across different tumor types. 2 patients are still on study after >5 years.



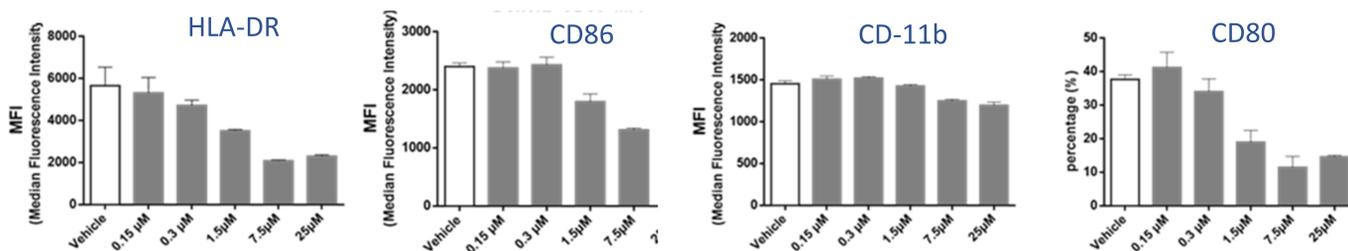
BXQ-350 rebalances the tumor microenvironment towards an anti-tumoral state



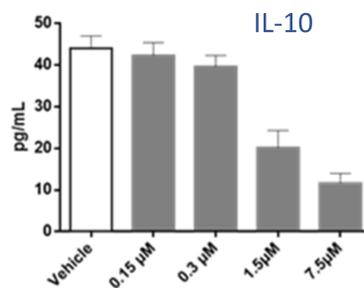
(1) a) E. Assi et al., Modulation of Acid Sphingomyelinase Reprogrammes the Tumour Immune Microenvironment, *Mediators of Inflammation*, 2015, id 370482. doi.org/10.1155/2015/370482; b) N. Beyersdorff et al., Sphingomyelin Breakdown in T cells: Role in Activation, Effector Functions and Immunoregulation, *Biology Chemistry*, 2015, 396 (6-7), 749; c) S. Morad et al., Ceramide-Orchestrated Signalling in Cancer Cells, *Nature Reviews*, January 2019 (13), 51.
(2) a) F. Liu et al., Ceramide Activates Lysosomal Cathepsin B to Suppress Myeloid-Derived Suppressor Cells, *Oncotarget*, 2016, vol 7(51), 83907; b) P. Chakraborty et al., S1P Metabolically Programs T cells to Limit Anti-Tumor Activity, *Cell Reports*, 2019, 28, 1879; c) Y. Liu et al., S1P1 Promotes Tumor-Associated Regulatory T cell Expansion Leading to Poor Survival in Bladder Cancer, *Cell Death and Disease*, 2019, 10:50. doi.org/10.1038/s41419-018-1298-y.

6. BXQ-350 Inhibits MDSCs Differentiation and Suppressive Function: Monocytes are cultured with IL-6 and rhGM-CSF; after 7 days, MDSCs differentiation is ascertained (IL-10 and CD11-b, C80, CD86 and HLA-R expression).

Decrease in expression of specific MDSC markers indicates inhibition of differentiation

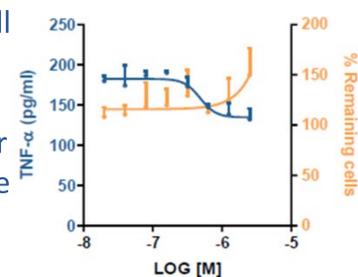


Decreased secretion of IL-10 indicates inhibition of MDSCs' suppressive function



7. BXQ-350 Repolarizes Macrophages to M1 Phenotype:

CD14+ macrophages were seeded at 5,000 cell / well on Day 0, on Day 5 BXQ-350 was added and LPS was added one hour after that. On Day 8, cells were fixed with 4% formaldehyde and TNF-α and viability quantified



And Stimulates T cell Expansion:

Increasing concentrations of BXQ-350 promote proliferation of both CD4+ and CD8+ T cells

