**Introduction**

BNC105 is a compound that exerts an anti-cancer action through selective destruction of tumor blood vessels. A single IV dose of BNC105 causes a very high degree of tumor hypoxia leading to >95% necrosis in rodent models. Despite the dramatic tumor necrosis, tumor recovery becomes evident by day 2 following BNC105 treatment. We conducted immunohistochemical, in vitro and in vivo studies to identify the cellular and molecular basis driving tumor recovery from the significant vascular destruction caused by BNC105. Renal cancer cell lines treated with BNC105 exhibited increased expression of Hif1α, VEGFa and increased phosphorylation of mTOR and 4E-BP1. These observations led us to investigate the potential therapeutic benefit of combining BNC105 with agents inhibiting the signaling pathways corresponding to these proteins.

**Tumor Treatment with BNC105**

**BNC105 Combination with Everolimus**

A panel of renal cancer cell lines, including VHL mutant and VHL wild type, were shown under both normoxic and hypoxic conditions to express high levels of VEGFa. Culturing these cell lines with the mTOR inhibitor Everolimus decreased phosphorylation of p70S6K (Figure 4) and significantly reduced VEGFa expression (Figure 5). These findings demonstrate that Everolimus effectively curtails VEGFa signalling and is appropriate to combine with BNC105 therapy.

**BNC105 Combination with Pazopanib**

The potential benefit of combining BNC105 with the pan-VEGF inhibitor Pazopanib was also investigated. We similarly hypothesised that tumor recovery from the BNC105 induced hypoxia would remain or be sustained in the absence of inhibition of VEGFa receptors. Treatment to RENCA orthotopically implanted tumors with BNC105 alone resulted in 21% TGI and Pazopanib alone 19%. In combination the TGI was 46% (Figure 8A). Furthermore, survival was significantly increased (p<0.0001) than animals treated with monotherapies (Figure 8B).

**Summary & Conclusions**

- **Regions of tumors adapt to the hypoxic shock caused by BNC105 treatment through adaptive survival pathways such as the mTOR pathway and VEGF driven revascularisation.**
- **Specifically targeting and exploiting these hypoxic adaptive survival pathways greatly increases efficacy of renal tumor treatment in combination with BNC105.**
- **These data demonstrate that BNC105 can be combined with Everolimus or Pazopanib, to yield greater anti-tumor efficacy in renal cancer.**
- **A randomised Phase II trial evaluating the potential benefit of combining BNC105 with Everolimus in patients with metastatic renal cancer has finished accrual and is expected to yield results in the first half of 2014.**