

BLVRB is the metabolic driver in breast cancer

20% of breast cancers exhibit the overexpression of HER2 (ErbB2) receptor that is associated with more aggressive disease and worse patient prognosis. Although HER2-directed therapies have significantly improved the clinical outcome, many patients do not benefit from them because of intrinsic and acquired resistance. Therefore, the identification of novel therapeutic targets and their pharmacological inhibitors are needed to improve the survival of these high-risk patients.

Our preliminary study has identified BLVRB (Biliverdin IX β reductase) as a novel putative target linked to progression and therapy-resistance, specifically in Her2+ breast cancer. We found that BLVRB is frequently upregulated in HER2+ cancer, predicting poor survival and resistance to therapies. BLVRB is a cellular redox regulator within the heme degradation pathway that catalyzes the NADPH-dependent reduction of multiple substrates and plays an essential role in maintaining cellular redox homeostasis. We demonstrated that BLVRB-deficiency in stem cells leads to a selective loss of glutamine entry to tricarboxylic acid (TCA) cycle, and exaggerated accumulation of reactive oxygen species (ROS), and cell death. While together these data support the unrecognized significance of BLVRB in mammary tumorigenesis, its oncogenic function has not been identified. Our feasibility proof-of-principle study aims to establish the role of BLVRB in breast cancer progression and therapy resistance.

Furthermore, formulated on (1) computer-aided in silico thermodynamic modeling, (2) a structure similarity screen (>35 million compounds) incorporating specificity/affinity restrictions, and (3) confirmatory compound/BLVRB crystal structures, we identified the most potent selective BLVRB inhibitors described to date.

Hypothesis or Objective: We propose to develop ***BLVRB-selective inhibitors as an innovative approach for the intervention of Her2+ breast cancer bioenergetics and metabolism by modulating cellular glutamine utilization and redox homeostasis.*** As a **feasibility study**, we propose to test BLVRB inhibition in proof-of-principle experiments *in vitro* (Aim 1) and MMTV/ErbB2 mouse model of Her2+ breast cancer (Aim 2). We will validate the therapeutic utility of targeting of BLVRB in HER2+ breast cancer in the BLVRB-deficient mouse model of HER2 positive breast cancer and *in vitro* studies testing isogenic cell lines with genetically ablated BLVRB (dox-inducible CRISPR) and specific BLVRB inhibitors. Importantly, unlike current glutamine-modulating strategies designed to limit TCA glutamate uptake, the approach is taken in this application (1) targets a cellular enzyme (BLVRB) that is specifically upregulated in invasive breast cancer and (2) that retains the unique capacity of shunting glutamate from TCA uptake. We postulate that this approach enhances target specificity with putative effects on improved efficacy.

Specific Aims:

Aim 1: Validate and delineate molecular mechanisms of BLVRB inhibitors to selectively block metabolic functions in ErbB2-positive breast cancer cells *in vitro*.

Aim 2. Test whether genetic BLVRB deficiency suppresses ErbB2-driven mammary tumorigenesis.

The implementation of this collaborative research will provide critical data for RO1 application and layout a framework for future mechanistic studies establishing BLVRB as a metabolic driver in breast cancers, as well as clinical applicability. We have already made progress in characterizing lead compounds that can further be optimized by medicinal chemistry to advance toward drug development.