

## **Supplementary Methods**

### **PBMC Isolation and storage**

Peripheral blood from consented HNSCC patients or healthy controls was obtained from the University of Cincinnati College of Medicine. Peripheral blood was received in 10mL EDTA tubes and 5mL SST tubes. EDTA samples were processed to isolate PBMCs using Ficoll-Paque PLUS (GE-Healthcare Life Sciences) density gradient centrifugation in SepMate tubes (StemCell Technologies). PBMCs were cryopreserved in Cryostor CS10 (Stem Cell Technologies) and stored in liquid nitrogen. SST samples were spun at 1200xg for 15min. Serum was collected into microcentrifuge tubes and stored at -80C.

### **Drug Treatments**

Metformin (13118) and BP-1-102 (28368) were obtained from Caymen Chemicals. Dexamethasone (S7306), Everolimus (S1120), MHY1485 (S7811), and Flubarabine (S1491) were obtained from Selleck Chemicals. Navarixin (HY-10198) was obtained from Medchem Express.

### **Supplementary Figure 1. Serum Cytokines in healthy patients compared to HNSCC**

**patients treated with metformin.** Healthy control subject serum and HNSCC patient serum from the clinical trial (NCT02325401) pre- and post-metformin were analyzed by ELISA for production of IL-2, IL-4 and TNF $\alpha$ .

**Supplementary Figure 2. T-cell and WBC ratios from clinical trial.** **(A)** white blood cell count before and after metformin in patients from clinical trial (NCT02325401). **(B)**

Absolute leukocyte count before and after metformin in patients from clinical trial (NCT02325401). **(C)** %CD3+ cells as determined by flow cytometry pre and post metformin. **(D)** Ratio of CD8+ cells to CD4+ cells determined by flow cytometry pre and post metformin. **(E)** Flow cytometry of t-cell types pre and post metformin using markers CCR7 and CD45RO. **(F)** Quantification of (E). (A), (B), (C), (D), and (F) analyzed by paired students t-test.

**Supplementary Table 1.** NCT02325401 Clinical trial patient demographics

**Supplementary Table 2.** NCT02083692 Clinical trial patient demographics

**Supplementary Figure 3.** Metformin does not change TGF $\beta$  or IFN $\gamma$  release. ELISA of supernatant collected from peripheral HNC NK cells treated with vehicle, 12mM metformin, 50ng CXCL1, or 10nM Navarixin for 24hrs.