



Figure S2: KRAS mutation promotes resistance to combinations of RAF, EGFR, and MEK inhibitors, but retains sensitivity to ERK inhibitor combinations. VACO42 cells expressing exogenous KRAS G12D, G13D, or empty vector control were treated with the indicated concentrations of drugs for 3d and relative cell titer was determined. Combinations of RAF, EGFR, and MEK inhibitors, including combinations in clinical use, are shown in (A), and ERK inhibitor combinations are shown in (B). Encorafenib (ENC), cetuximab (CET), dabrafenib (DAB), panitumumab (PAN), trametinib (TRA), vemurafenib (VEM), selumetinib (SEL), VX-11e (VX). (C)

VACO432 cells (VACO) expressing empty vector, KRAS G12D or G13D and the *BRAF* and *KRAS* wild-type CRC cell line COLO-320DM, which is not MAPK dependent, were treated with the indicated concentrations of VX-11e for 3d. VX-11e did not effect the viability of COLO-320DM cells at the concentration range in which it suppresses viability of VACO432 cells, supporting that the effects of VX-11e on VACO432 cell models are due to the specific, on-target effects of this compound. (D) Parental VACO432 cells (VACO) and derivatives made resistant to combined RAF/EGFR inhibition (VACO-RE) or RAF/MEK inhibition (VACO-RM) were treated with the indicated concentrations of VX-11e for 3d, and relative cell titer was determined.