Supplemental Figure 2. Cobimetinib and GDC-0994 activity in BRAF mutant cell lines and combination activity in the A549 KRAS mutant cell line.

(a) Colo205 (BRAF^{V600E}, colorectal) or (b) A375 (BRAF^{V600E}, melanoma) cells were treated with DMSO or 0.5x EC50 concentrations of either cobimetinib (0.005 µM) or GDC-0994 (0.1 µM) for the indicated number of hours followed by analysis of pathway activation at the level of MEK, ERK, and p90RSK relative to the loading control GAPDH. (c) A549 (KRAS^{G12S}, NSCLC) cells were treated with DMSO single agent cobimetinib at 0.25 µM, single agent GDC-0994 at 1.25 µM or the combination of half concentrations of both agents (0.125 µM cobimetinib and 0.625 µM of GDC-0994) for the indicated number of hours followed by analysis of pathway activation at the level of p90RSK relative to the loading controls of Actin and GAPDH. The graph below shows quantification of the immunoblot above for normalized p-p90RSK ([p-p90RSK/actin]/[total p90RSK/actin]) following treatment with cobimetinib (green triangles), GDC-0994 (blue squares) or the combination (red diamonds) in A549 cells.