## Supplemental Figure 1

## A. - C. UMAP plots for all other data sets that were not treated with Navitoclax including OIS cells that were taken off of 4-OHT before collection and were sequenced from the 5’ end, OIS cells that were in 4-OHT when they were collected, and RS cells that were generated from a different clonal cell line than the rest of the data sets. D. - F. Cluster trees for the data sets. G. - I. Senescence markers for cluster-1 and cluster-2.

## Supplemental Figure 2

## A. - C. Senescence markers for cluster -1 and cluster -2 for all Navitoclax treated samples.

## Supplemental Figure 3

## A. UMAP plots for OIS cells treated with Navitoclax that were sequenced from the 5’ end. B. Cluster tree for that data set. C. Violin plots showing marker genes for primary and secondary senescence. D. Bar plots showing Navitoclax preferentially induces apoptosis in Cluster-1 cells.

## Supplemental Figure 4

## A. UMAP plots showing primary and secondary clusters for IMR90 cells from Teo et al. B. Primary and secondary marker genes for Teo et al [11].

## Supplemental Figure 5

## Plots for HUVEC RS cells from Zirkel et al. [18]. A. Feature plots showing the expression of senescence markers in the data set. B. Predicted cell cycle phase. C. (Top) Scatter plot with cells plotted according to their ribosomal protein gene expression on the x-axis and collagen expression on the y-axis. (Bottom) Scatter plot with cells plotted according to their HMGA1 expression on the x-axis and collagen expression on the y-axis.

## Supplemental Figure 6

## Violin plots showing expression of the neomycin resistant marker gene (labeled neo-ras) which we used to measure the expression of the 4-OHT inducible HRAS:G12V transgene. Violin plots show expression between cluster-1 and cluster-2 for RS, OIS, and DDIS cells.

## Supplemental Figure 7

## A. UMAP plots for RS cells generated from the second clonal line (RS Rep2). Top left shows samples. Top right shows identified clusters. Bottom left shows cell cycle score for the G2 phase of the cell cycle. Bottom right shows the cell cycle score for the S phase of the cell cycle. B. Violin plots showing QC parameters for the identified clusters. C. Schematic showing how filtering was performed.

## Supplemental Figure 8

Volcano plots showing predicted upstream regulators (IPA) for cluster-1 (red) and cluster-2 (blue) for each type of senescence. Regulators are plotted according to their z-score on the x-axis which shows if they regulate cluster-1 or cluster-2. Positive values indicate an upstream regulator for cluster-1 and negative for cluster-2. The ‘-log q-value of overlap’ is plotted on the y-axis. This is the negative logarithmic transformation of the false discovery rate, which determines the upstream regulators whose gene sets show a statistically significant overlap with the list of differentially expressed genes in the data. Grey lines indicate significance values. Upstream regulators located above the horizontal line and to the left of the vertical left grey line or right of the vertical right grey line have significant z-scores and false discovery rates. Plots for IMR90 cells from Teo et al. are also shown.

## Supplemental Figure 9

## Hierarchical clustering of cells in cluster-1 and cluster-2 across RS, OIS, and DDIS based on genes with the most significant differences between cluster-1 and cluster-2 (differential expression p < 10-20).