Supplemental Figures

Malignant Melanoma Cells Differentially Alter Extracellular Matrix Biosynthesis that Promotes Survival in Response to Targets of Mitogen-Activated Protein Kinases Signaling

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Supplemental Figure 1. Examination of integrins that mediate cell ECM interactions
A) Top panel- Schematic of 3D sample preparation. Cells were embedded in laminin rich ECM (IrECM) and cultured for 10 days to recapitulate growth of melanoma in the presence of basement membrane.
(B) Quantification of tumor aggregates shows differences in size and shape (circularity) between aggregates derived from distinct clones, where A375 aggregates are bigger and irregular while A375.S2 clones are smaller and circular. *** indicates a p value<0.001
(C) Real-time PCR confirm that both clones produce FN1 by probing mRNA levels. Actin is a loading control.
Supplemental Figure 2. Examination of integrins that mediate cell ECM interactions
Immunoblots show that αv integrin levels are similar for Scr control cells and shFN cells, whereas β1 levels show a modest increase in shFN cells. GAPDH is a loading control.

Supplemental Figure 3. Cell cycle is unaltered for shFN cells compared to Scr control Tumor cells
Cell cycle analysis revealed negligible differences between shFN cells and scramble control for both isogenic melanoma clones.

Supplemental Figure 4. Effect of architecture on drug response
A) Immunoblots of protein cultured in 2D show differential ERK phosphorylation for each clone in response to inhibitors, despite previously established similarity of viability curves.
(B) Representative images show that colonies are viable and metabolically active as determined by uptake of nitrotetrazolium blue chloride.
(C) Histograms showing the size distribution and numbers of colonies after one month, where drug treatment commenced at the single cell stage in 3D soft agar.

Supplemental Figure 5. 2D drug uptake is unaltered for shFN cells compared to Scr control Tumor cells
Control studies show that rhodamine administered with an inhibitor of a drug transporter illustrates no differences in uptake observed between Scr and shFN cells in 2D (Left panel). Similarly, no differences were observed for fluorescent conjugated taxol (right panel).
Supplemental Figure 1
Supplemental Figure 2
Supplemental Figure 3

A375 scr
G1: 48.07%
S: 7.42%
G2: 43.61%

A375 shFN2
G1: 52.96%
S: 11.63%
G2: 35.1%

A375S2 scr
G1: 41.85%
S: 4.93%
G2: 41.13%

A375S2 shFN2
G1: 40.9%
S: 3.19%
G2: 50.82%
A

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<th>A375-Scr</th>
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B

Measuring Uptake of nitrotetrazolium blue chloride

C

Control | CISPLATIN-i | BRAF-i | ERK-i | BRAF-i AND ERK-i

Supplemental Figure 4