

FigureS1. ICAM-2 expression inhibited invasion of SK-N-AS cells through Matrigel to the distal side of porous (8 micron) membranes. NB cells were overlaid on solidified Matrigel (upper chamber), with 10% fetal bovine serum in DMEM supplemented with fibronectin (20 μ g/ml) in the lower chamber. Cells that migrated through the Matrigel to the distal sides of membranes were quantitated using an AlphaEase FC software (Alpha Innotech, San Leandro, CA). Results are expressed as area of the membrane in pixels occupied by crystal violet-stained cells. Values shown were calculated from a minimum of three replicates. A t-test was performed using GraphPad Prism 5 software to compare groups. P=0.0353* and P=0.0037**. The value for Control cells in pixels = 10,322 ± 3,044.

Supplementary FigureS2 (Feduska et al.)



FigureS2. Elevated ICAM-2 expression in NB-1691 NB cells inhibited colony growth in soft agar and inhibited disseminated tumor growth in an *in vivo* model of metastatic neuroblastoma. (A) Analysis by t-test demonstrated that ICAM-2 WT suppressed anchorage-independent growth of NB-1691 cells in vitro (P<0.0001). (B) Immunoblots confirmed that transfected NB-1691 cells expressed readily detectable levels of ICAM-2 WT.
(C) Mice injected i.v. with NB-1691 cells transfected to upregulate expression of ICAM-2 WT survived longer than mice receiving cells expressing low detectable ICAM-2. Kaplan-Meier survival plots were analyzed by log-rank test using GraphPad Prism 5 software (P=0.0155*).



Days after s.c. injection of SK-N-AS transfectants

FigureS3. Control and ICAM-2 WT transfectants (5 x 10⁵ cells) produced subcutaneous tumors of equivalent size within the same time frame. When fewer cells were implanted (5 x 10⁴ and 5 x 10³) neither transfectant produced tumors. Mice were injected s.c. with 5 x 10⁵ SK-N-AS Control (red line) or ICAM-2 WT transfectants (blue line), and observed for tumor development. Both transfectants produced tumors of 1,000-1,500 mm³ 45 ± 7 days following implantation, necessitating euthanasia. Neither transfectant produced subcutaneous tumors when 5,000 or 50,000 cells were implanted, within the >2-month duration of the experiment (data not shown). We detected no impact of ICAM-2 on the tumorigenic potential of SK-N-AS NB cells.

Supplementary FigureS4 (Feduska et al.)



FigureS4. Expression of RNA encoding stemness and EMT markers was similar in control SK-N-AS cells and ICAM-2 WT transfectants. No differences were seen in levels of RNA encoding c-Myc (stemness marker) ^{S1}, E-cadherin, Occulidin, Vimentin, or Snail 1 (EMT markers) ^{S2} among the four SK-N-AS NB cell transfectants. Primers used for RT-PCR analysis were: E-cadherin (forward) 5'- CAGCACGTACACAGCCCTAA-3' and (reverse) 5'- ACCTGAGGCTTTGGATTCCT-3'; Occludin (forward) 5'GATGACTTCAGGCAGCCTCG-3' and (reverse) 5'- CTATGTTTTCTGTCTATCATAGTC-3'; Snail 1 (forward) 5'- GAAAGGCCTTCAACTGCAAA-3' and (reverse) 5'-TGACATCTGAGTGGGTCTGG-3'; c-Myc (forward) 5'- CGACTCTGAGGAGGAACA AG-3' and (reverse) 5'- ACCATCAGACTCTGACTTTAGACTTT-3'; GAPDH (forward) 5'- AACATCATCCTGCT-TCCAC-3' and (reverse) 5'- GACCACCTGGTCCTCAGTGT-3'.

^{S1}Bandopadhayay P, Bergthold G, Nguyen B, Schubert S, Gholamin S, *et al.* BETbromodomain inhibition of MYC-amplified medulloblastoma. *Clin Cancer Res.* 2013.

^{S2}Moon H-J, Finney J, Xu L, Moore D, Welch DR, Mure M. MCF-7 cells expressing nuclear associated lysyl oxidase-like 2 (LOXL2) exhibit an epithelial-to-mesenchymal transition (EMT) phenotype and are highly invasive in vitro. *J Biol Chem* 2013; 288: 30000-30080.