

ErbB3 Upregulation as a Mechanism of Resistance to BRAF/MEK Inhibitors in the BRAF Mutant Setting

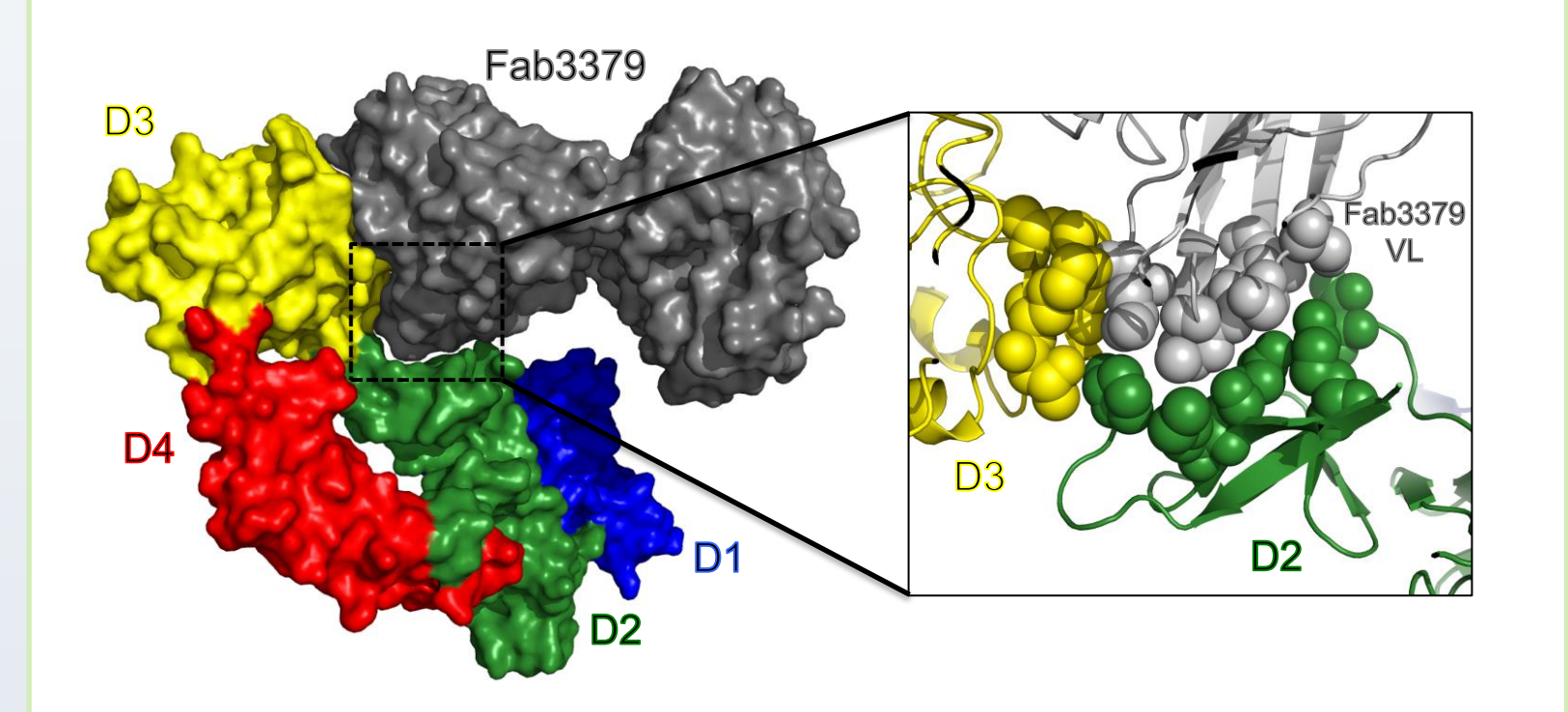
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Introduction

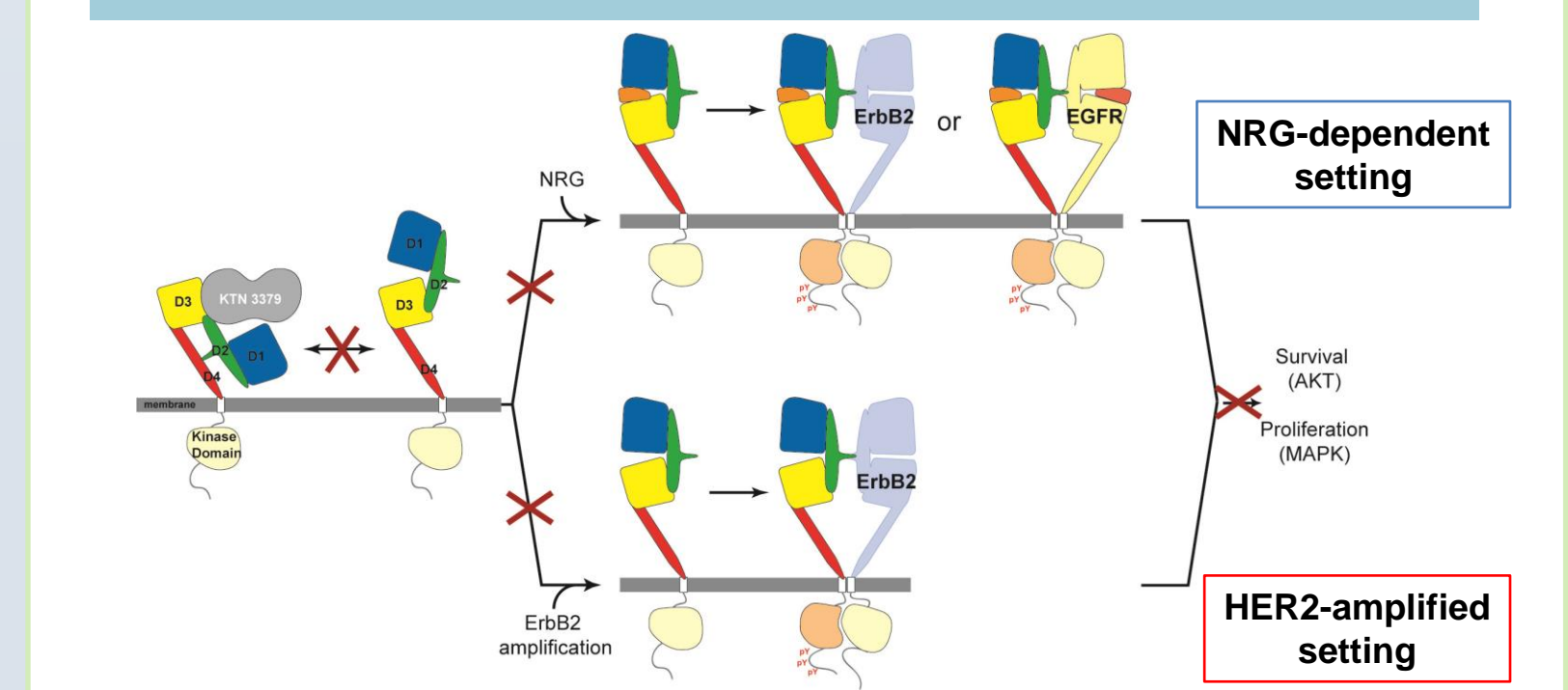
- KTN3379 is an IgG1 monoclonal antibody against ErbB3 that inhibits both ligand-dependent and –independent activation
- Three point mutations (YTE) in the Fc region of KTN3379 should increase antibody serum half-life through enhanced affinity to FcRN
- ErbB3 is a kinase-dead member of the ErbB family that functions as an obligate heterodimer with other ErbB receptors
- ErbB3 activation is mediated by its cognate ligand neuregulin/heregulin (NRG/HRG), or in the absence of ligand where ErbB2 is overexpressed
- ErbB3 provides strong pro-survival signals in solids tumors by engaging the PI3K/AKT signaling pathway
- ErbB3 upregulation may drive resistance to targeted therapies

KTN3379 Locks ErbB3 in its Autoinhibited Conformation



- KTN3379 binds to the hinge region between domains 2 and 3 and stabilizes ErbB3 in its inactive conformation *Lee S. et al. 2015. PNAS*
- Although the antibody does not bind to the ligand binding site, it allosterically precludes NRG binding to ErbB3

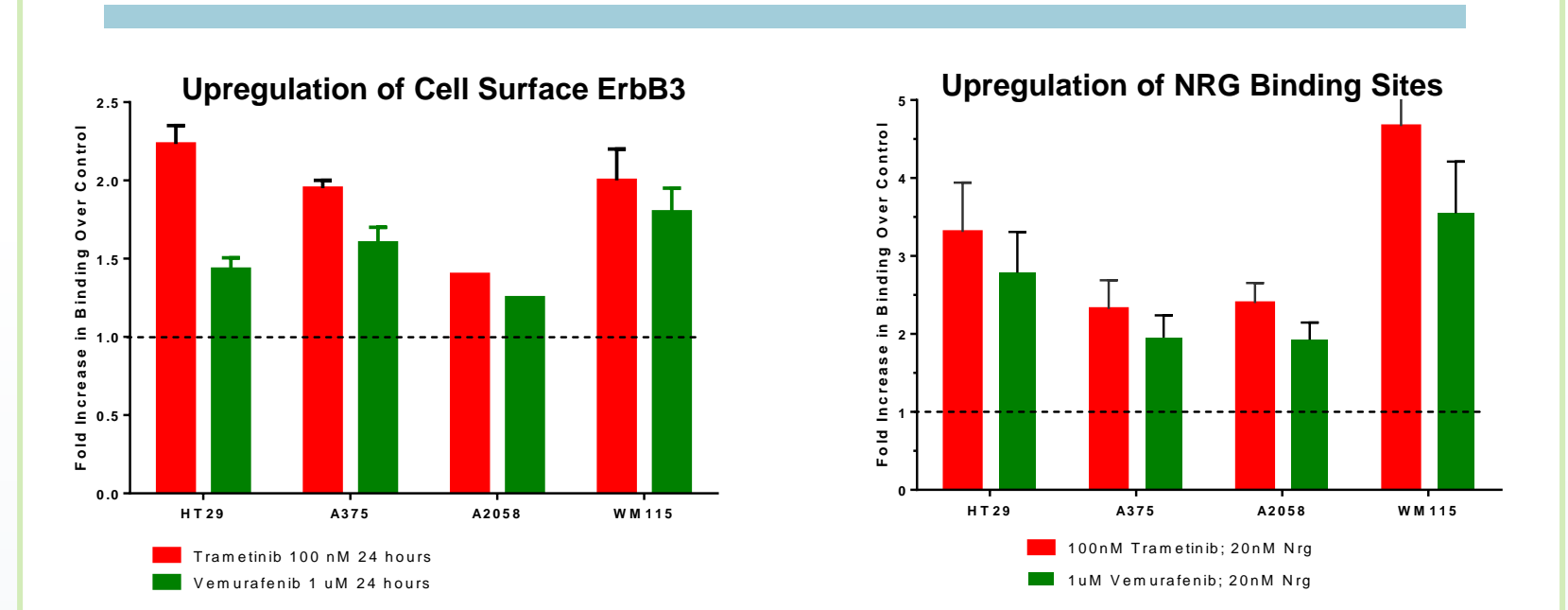
KTN3379 Inhibits ErbB3 Irrespective of its Mechanism of Activation



- ErbB3 exists in an equilibrium between a tethered and an extended state (left). NRG stabilizes the extended conformation of ErbB3 promoting heterodimerization and subsequent activation (top panel). In ErbB2-overexpressing tumors, ErbB3 can be activated independent of ligand (bottom panel)
- By blocking ErbB3 at the first step of activation, KTN3379 can inhibit either mode of ErbB3 activation

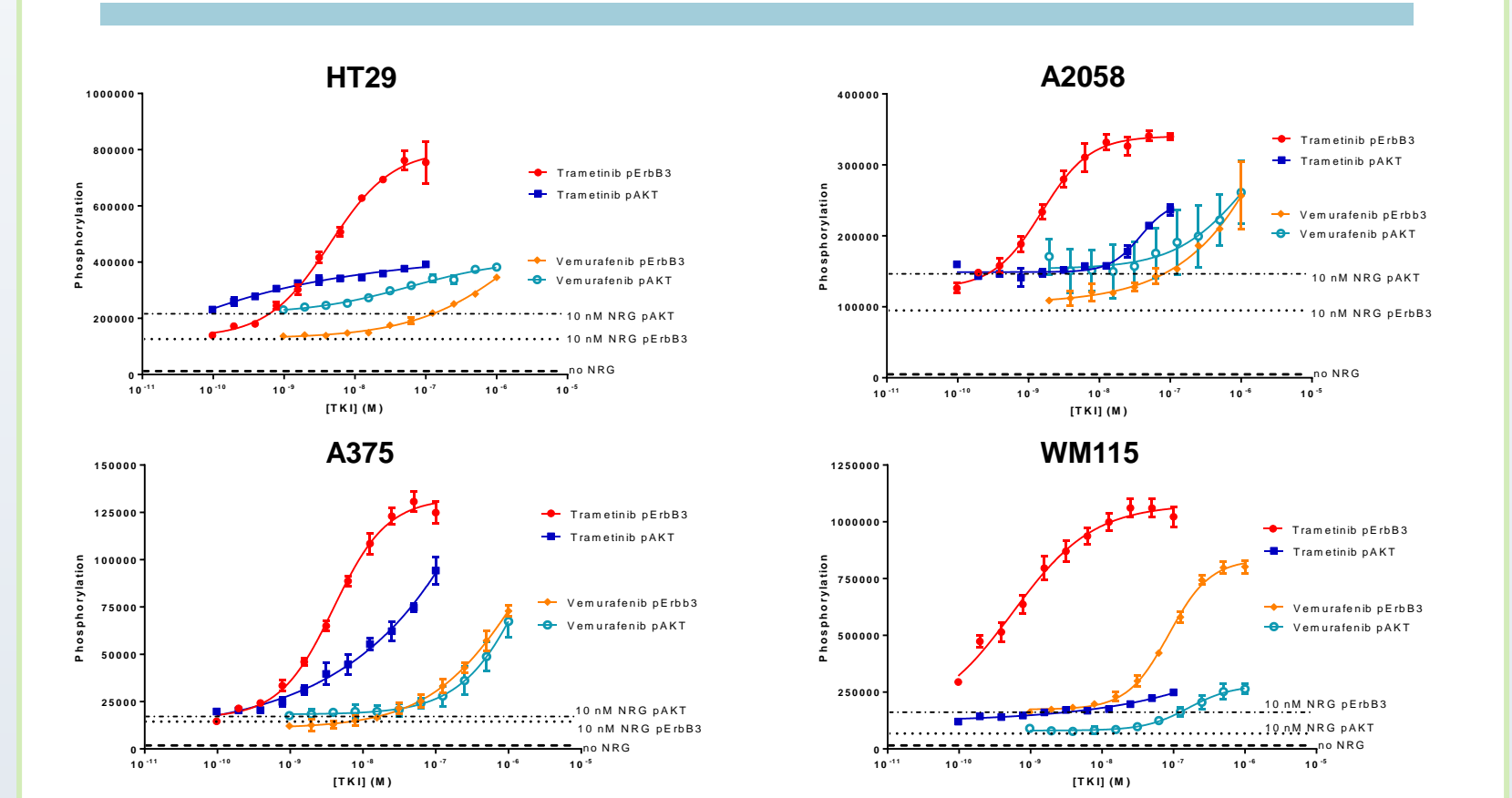
Results

ErbB3 Upregulation upon BRAF/MEK Inhibition in BRAF Mutant Cell Lines



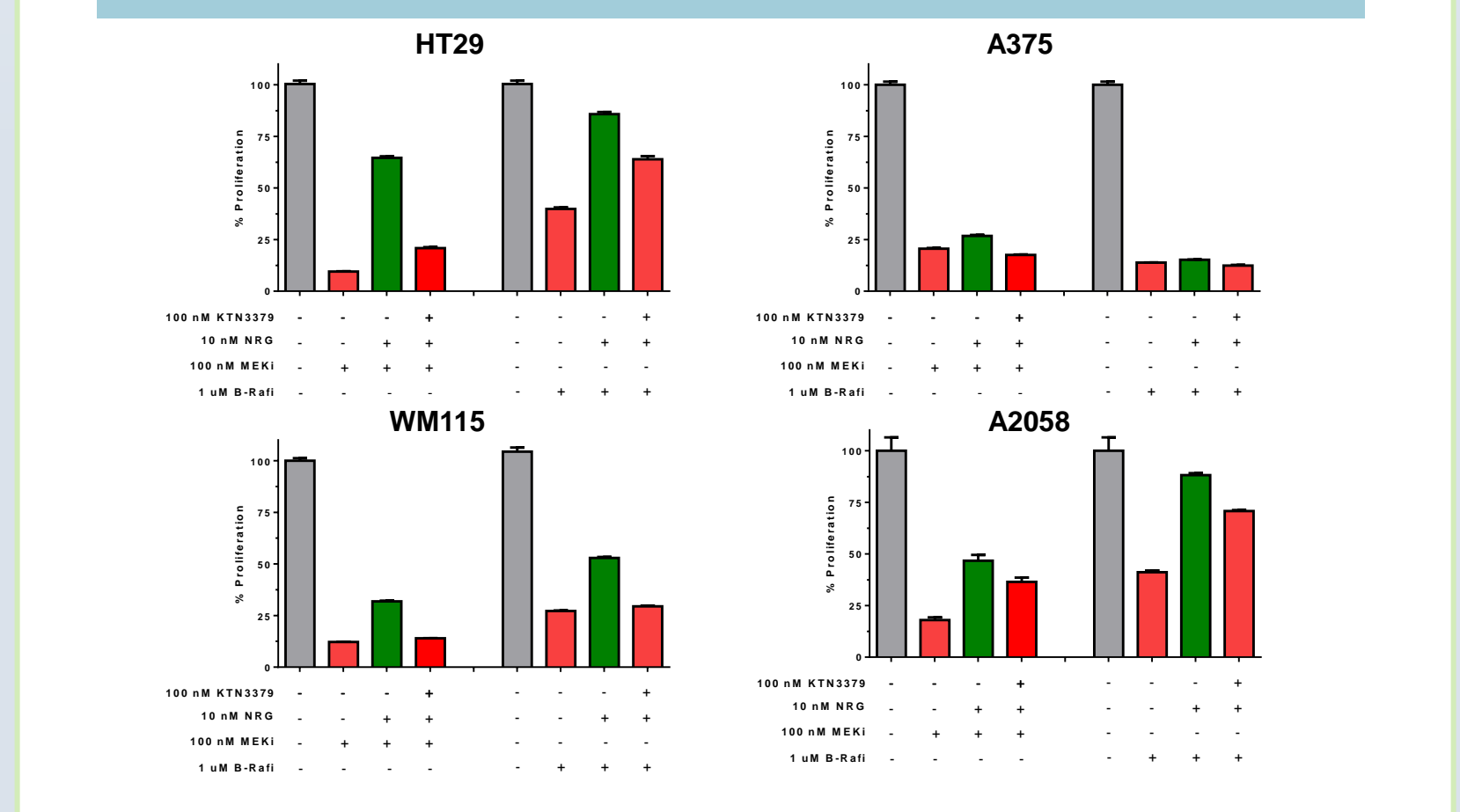
- Treatment of BRAF mutated cell lines with trametinib or vemurafenib for 24 hours results in cell-surface upregulation of ErbB3, as determined with an anti-ErbB3 antibody (left), or fluorescently-labeled NRG1 (right)

BRAF/MEK Inhibition Sensitizes Mutant Cell Lines to ErbB3/NRG Signaling



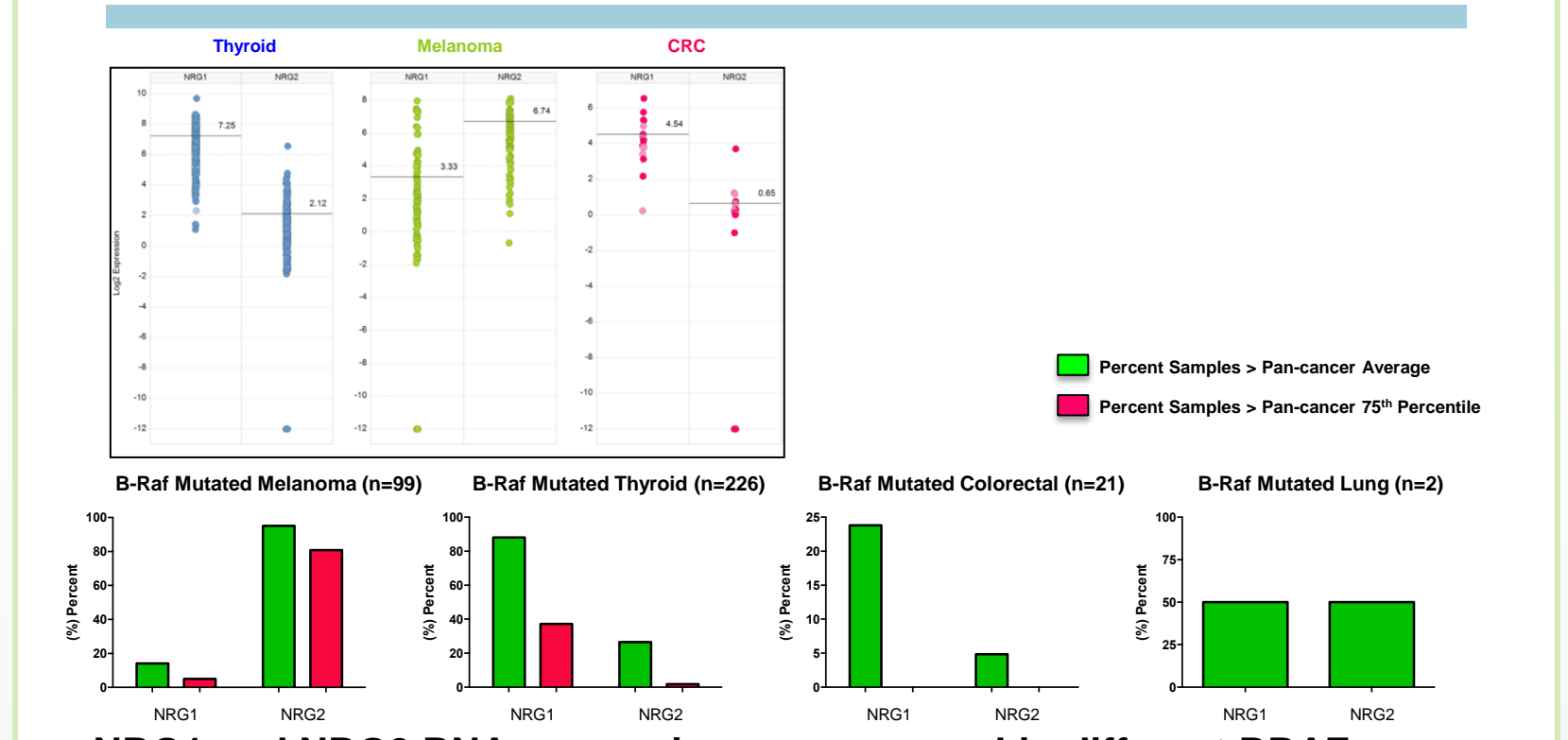
- Titration of trametinib or vemurafenib for 24 hours on BRAF mutated cell lines results in potent upregulation of NRG-induced ErbB3 and AKT phosphorylation relative to untreated samples

NRG Rescues and KTN3379 Resensitizes Cells to the Antiproliferative Effects of BRAF/MEK Inhibition



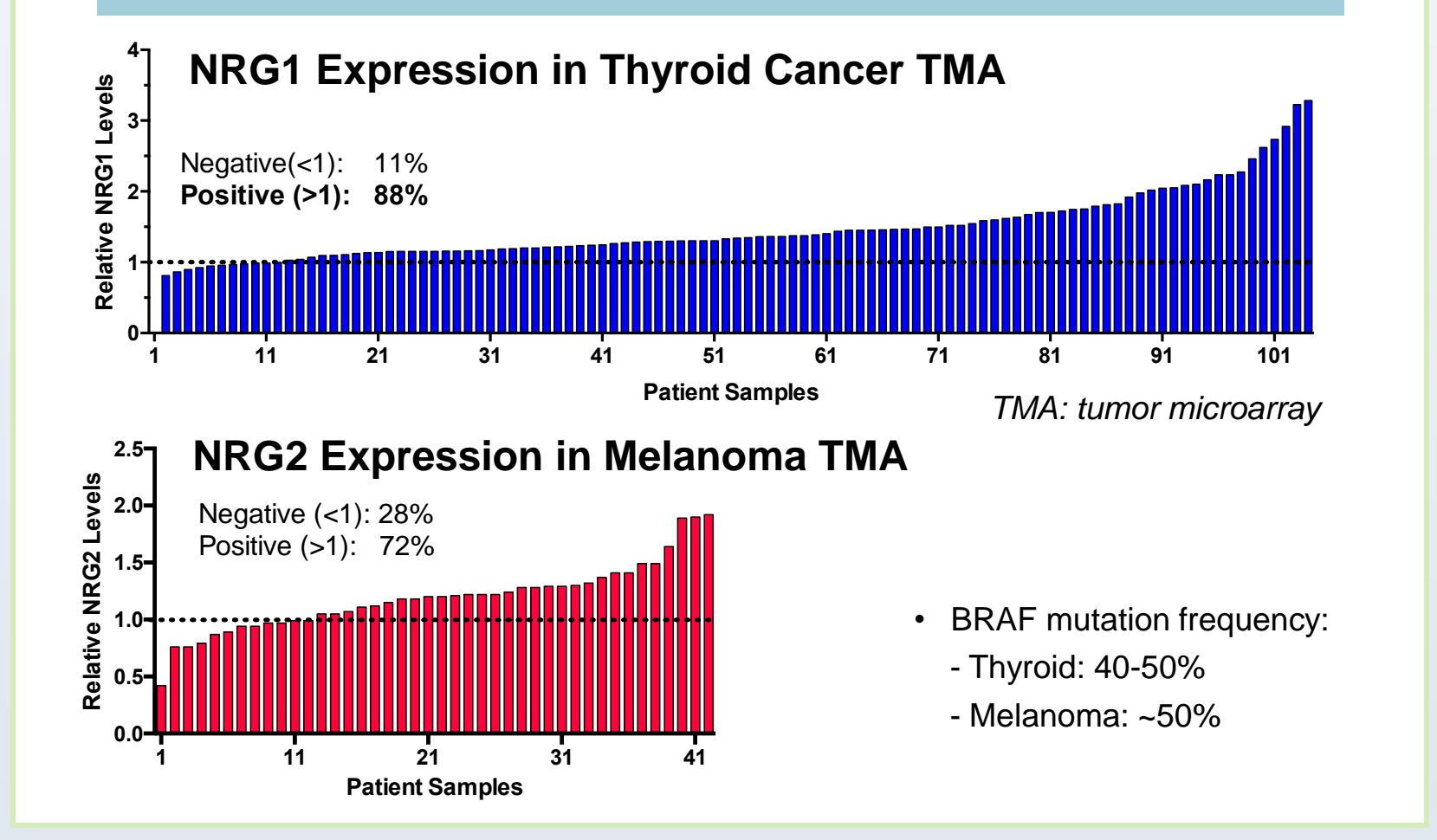
- NRG attenuates the antiproliferative activity of trametinib and vemurafenib

NRG1 and NRG2 are Highly Expressed in BRAF Mutated Thyroid Cancer and Melanoma, Respectively

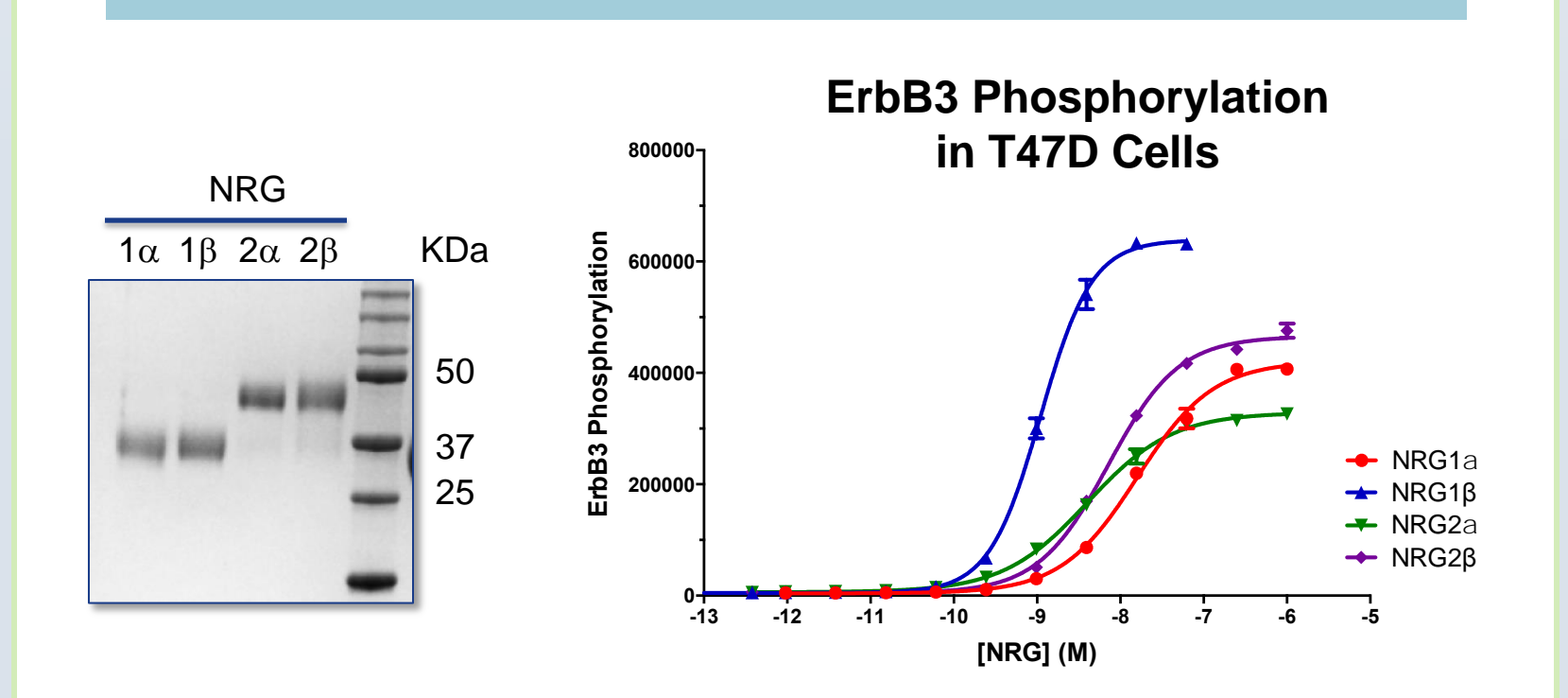


- NRG1 and NRG2 RNA expression was surveyed in different BRAF mutated tumor types
- NRG1 is highly expressed in thyroid cancer, whereas NRG2 is highly expressed in melanoma

NRG1 and NRG2 Expression is Highly Prevalent in Thyroid Cancer and Melanoma, Respectively

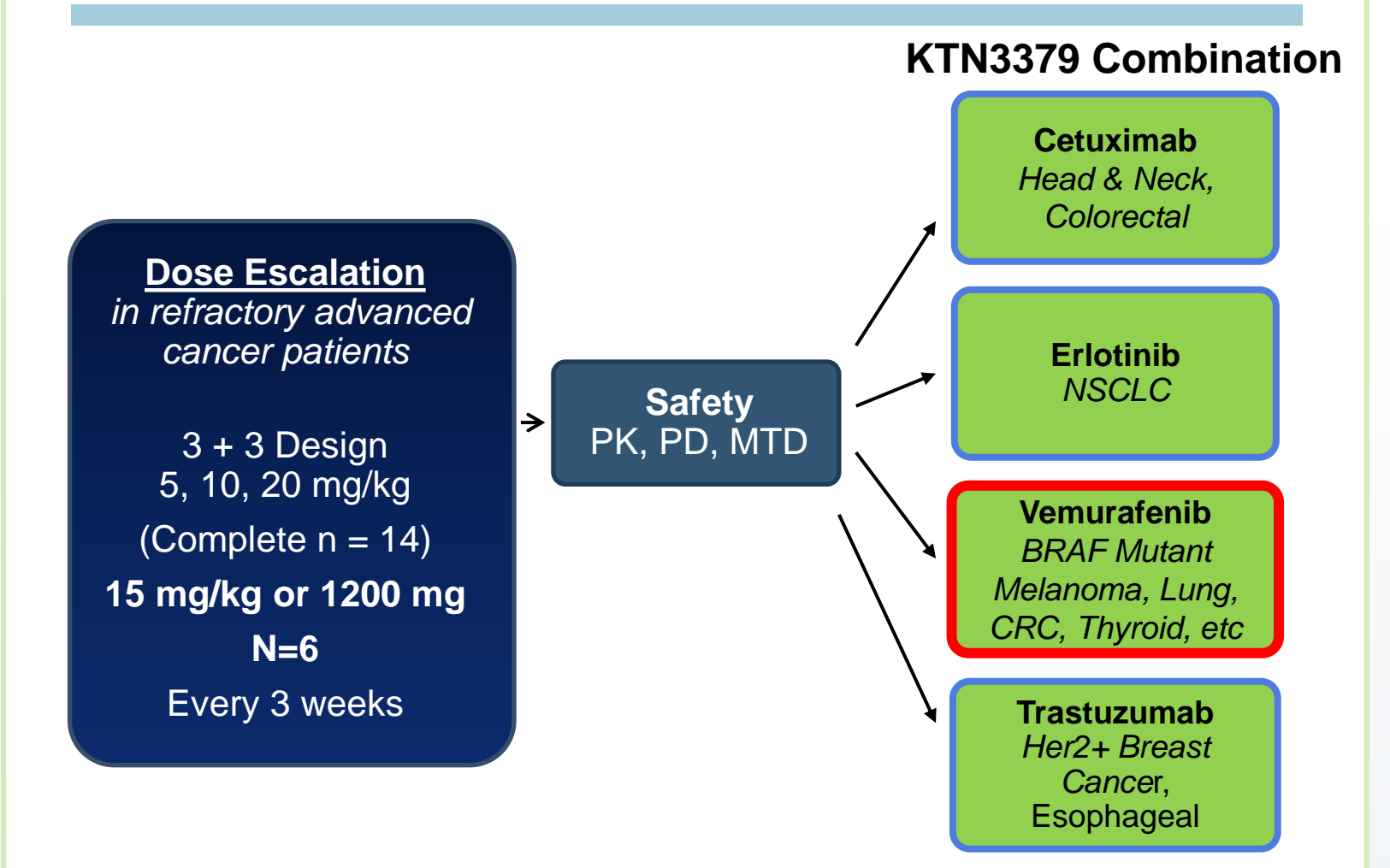


NRG1β is a More Potent ErbB3 Agonist than NRG1α, NRG2α or NRG2β



- The full extracellular domains of the two major NRG1 and NRG2 isoforms were secreted from HEK-293 cells, purified and used to stimulate ErbB3 phosphorylation in T47D cells
- NRG1β activates ErbB3 with an approximately 10-fold greater potency than the other NRG species

Ongoing Phase 1b Evaluation of KTN3379 Combinations in Selected Tumors



Preliminary Results (Bauer et al ASCO 2015 abstr 2598)

- MTD not established with KTN3379 monotherapy up to maximum administered dose of 20 mg/kg
- Most common toxicities for KTN3379 alone or in combination have included diarrhea and rash
- KTN3379 can be combined with vemurafenib, cetuximab, erlotinib, trastuzumab at 15 to 20 mg/kg Q3W
- All patients achieved serum concentrations above those required for maximal antitumor activity in animal tumor models
- Pharmacodynamic biomarker analyses showed soluble circulating ErbB3 levels were increased in all patients at all doses
- Enrollment continues in the four phase 1b cohorts
- Preliminary tumor response (NSCLC, CRC) data support Phase 2 studies in BRAF mutant cancers and other settings (eg., Head & Neck cancer)

Conclusions

- Targeting ErbB3 with KTN3379 hampers adaptive resistance to BRAF/MEK inhibitors in tumors with an oncogenic MAPK signaling drive
- ErbB3 is upregulated in response to BRAF/MEK inhibition
- NRG-stimulated activation of ErbB3 and AKT is potentiated dose dependently with trametinib or vemurafenib treatment of BRAF mutant tumor cell lines
- NRG attenuates the antiproliferative effects of BRAF/MEK inhibitors, however treatment with KTN3379 resensitizes the tumor cells to inhibitors
- Tumors with a high prevalence of BRAF mutation express NRG1 (thyroid) and/or NRG2 (melanoma) which may provide autocrine stimulation of ErbB3 and contribute to adaptive resistance to BRAF/MEK inhibitors
- NRG1β is the most potent ErbB3 ligand tested
- An ongoing Phase 1b study combining KTN3379 with vemurafenib in BRAF mutated tumors support further development in BRAF mutant cancer and other settings (eg., Head & Neck cancer)

