ERK-mediated HER2 Thr701 phosphorylation negatively regulates HER2/AKT axis activation through reducing EGFR/HER2 dimerization in a clathrin-dependent manner

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Purpose

Targeting MEK/ERK pathway has been viewed as a promising strategy for cancer therapy. However, MEK inhibition leads to the compensatory PI3K/AKT activation by relieving a negative feedback on ERBB receptors, contributing to the insensitization of cancer cells to MEK inhibitors. Understanding the underlying molecular mechanisms of this event is necessary for development of novel strategy to enhance the anti-tumor activity of MEK inhibitors.

Materials and Methods

HER2 positive and negative breast cancer cell lines were treated with MEK inhibitor. Western blot, immunoprecipitation and *in-vitro* kinase assay were used to investigate the molecular mechanisms of action.

Results

Our data showed that the induction of Akt activity by MEK inhibitors was specifically observed in HER2-positive cancer cells. Silence of HER2 or overexpression of HER2 kinase-dead mutant prevents the induction of Akt activation in response to MEK inhibition, suggesting HER2 as the major regulator for this event. Furthermore, the Thr701 residue of HER2 was demonstrated as a direct phosphorylation target for ERK1/2. Inhibition of this specific phosphorylation prolonged the dimerization of HER2 with EGFR via increasing the protein binding of HER2 with clathrin, leading to the enhanced activities of HER2 and EGFR tyrosine kinase and their downstream Akt pathway.

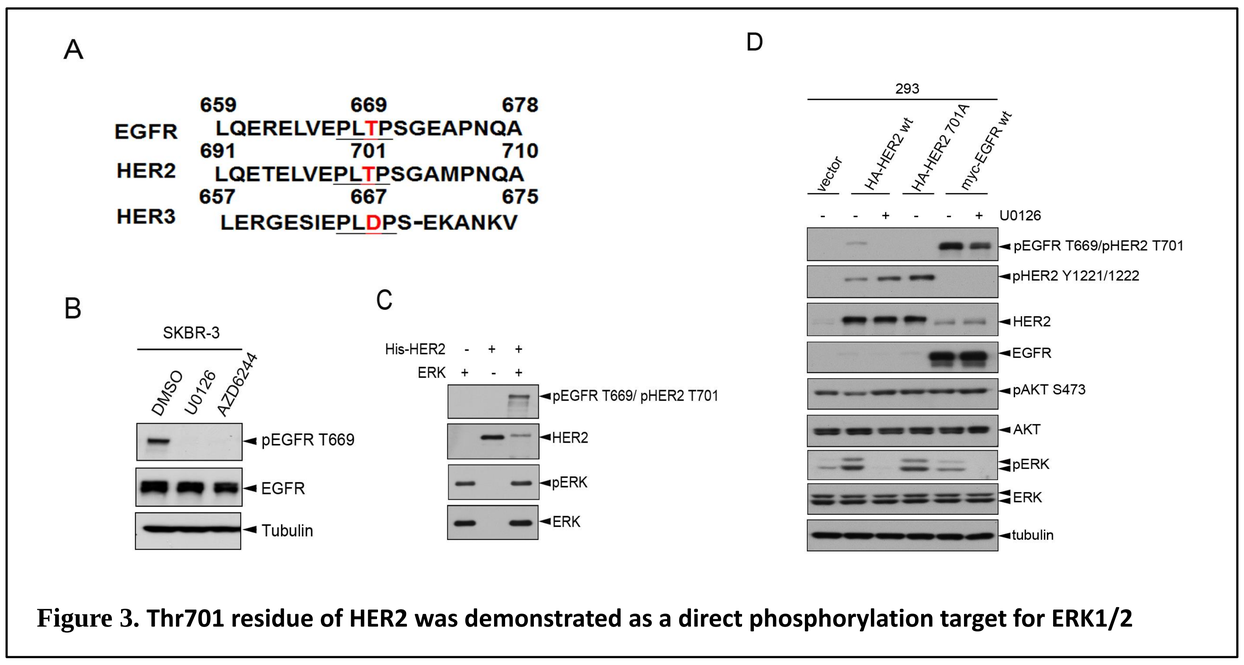


Figure 3. Thr701 residue of HER2 was demonstrated as a direct phosphorylation target for ERK1/2

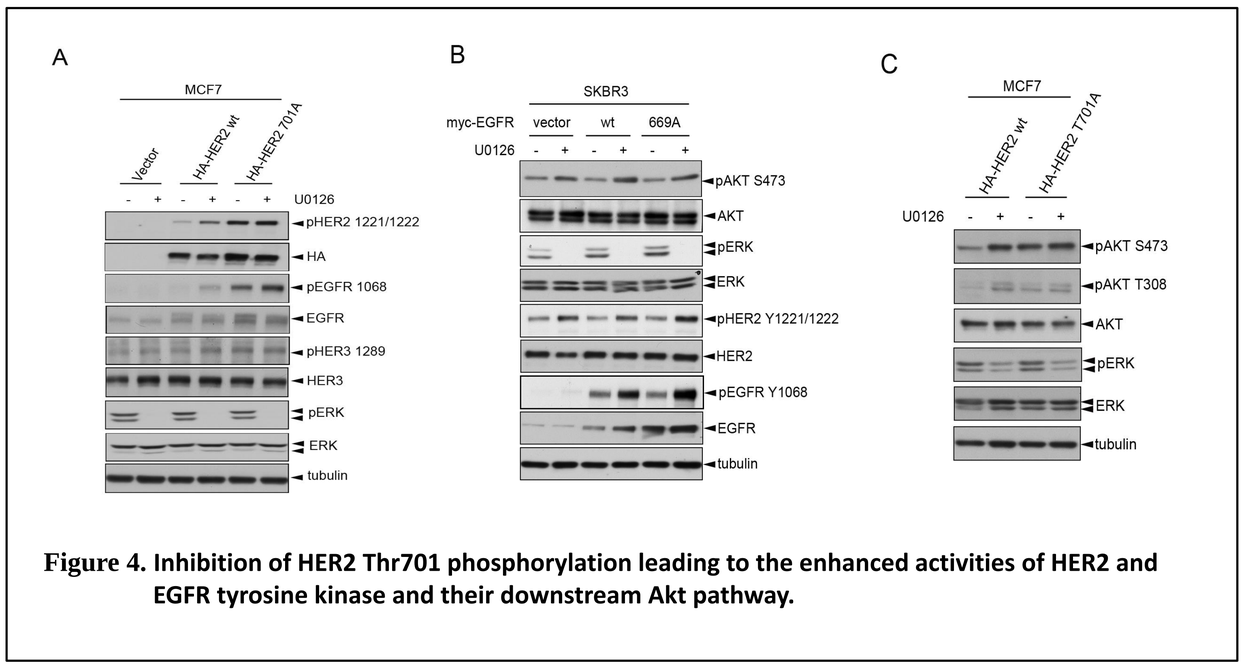


Figure 4. Inhibition of HER2 Thr701 phosphorylation leading to the enhanced activities of HER2 and EGFR tyrosine kinase and their downstream Akt pathway.

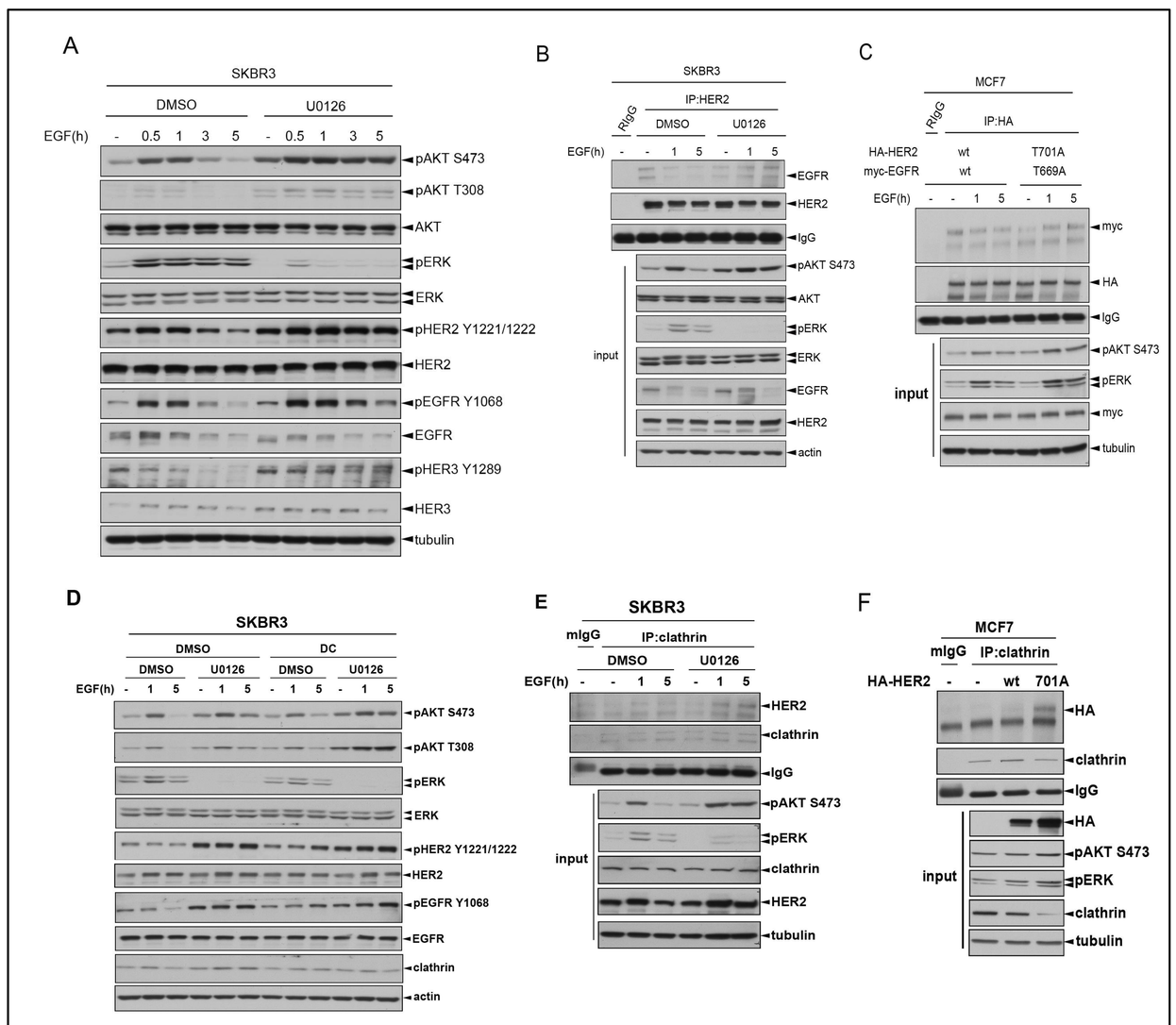


Figure 5. Inhibition of HER2 Thr701 phosphorylation prolonged the dimerization of HER2 with EGFR via increasing the protein binding of HER2 with clathrin

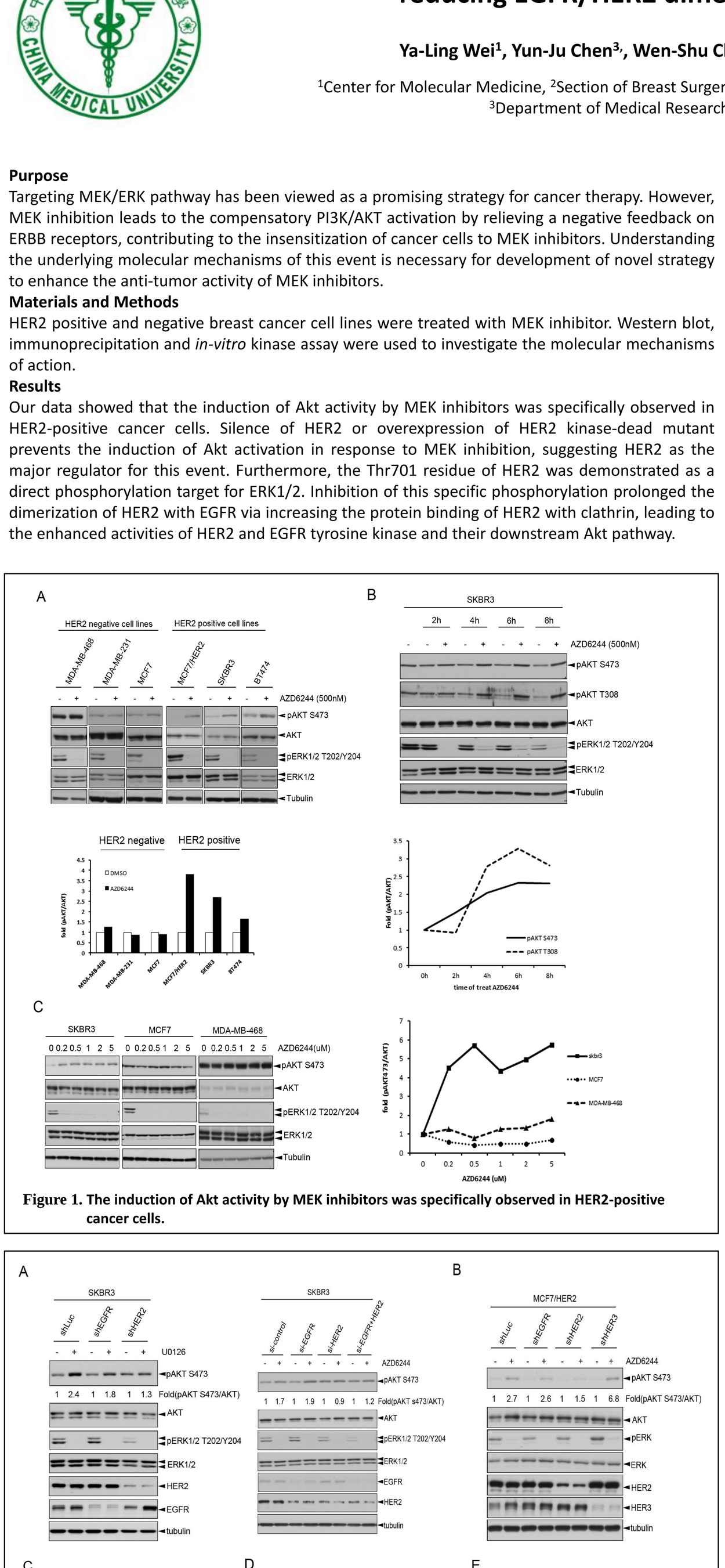


Figure 1. The induction of Akt activity by MEK inhibitors was specifically observed in HER2-positive cancer cells.

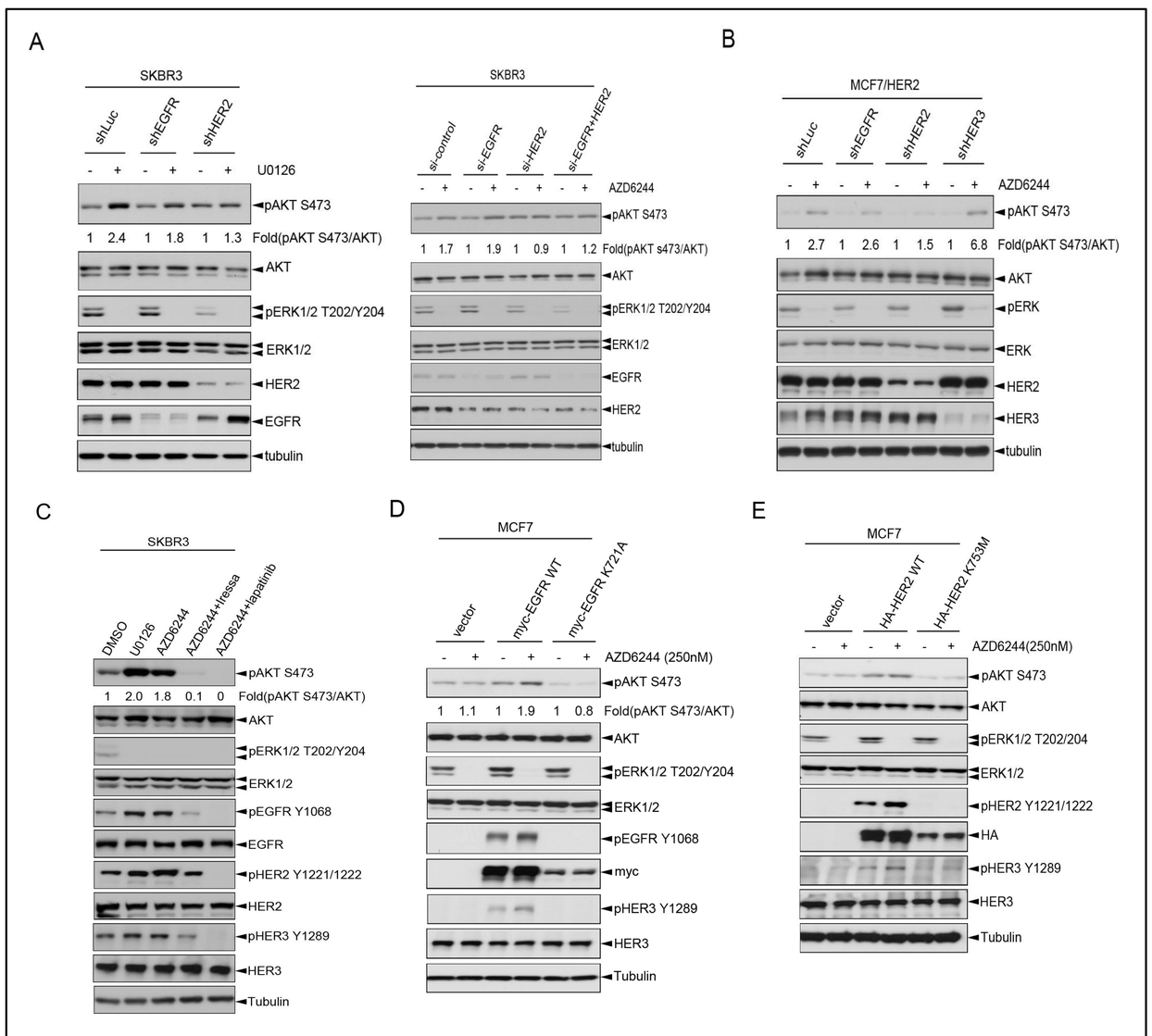
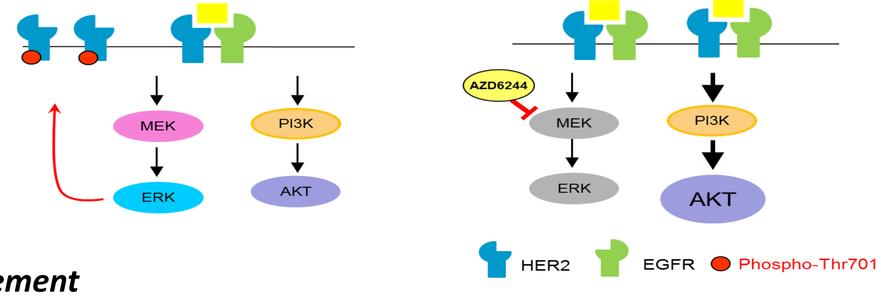


Figure 2. Silence of HER2 or overexpression of HER2 kinase-dead mutant prevents the induction of Akt activation in response to MEK inhibition

Conclusion

These results not only provide the molecular insight into the mechanisms of MEK inhibitor-induced Akt activation but also suggest that targeting protein interaction between HER2 and clathrin may enhance the therapeutic efficacy or MEK inhibitors.



Acknowledgement

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