Quantitative Analysis of HER2 Overexpression Effect on EGF Receptor Signaling

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***Short Abstract* —Overexpression of HER2, the preferred dimerization partner of other EGF receptors, not only shifts EGF receptor distribution between different cellular compartments, but also alters their signaling properties. We have investigated the effect of HER2 overexpression on EGF receptor signaling using both experimental approaches and computational methods. Epithelial cell lines expressing varying levels of HER2 receptors were constructed and receptor phosphorylation level upon ligand stimulation was monitored. Data collected in experimental measurements were then analyzed in the context of a multi-receptor cellular network model, which can be further applied to predict EGF receptor mediated biological response.**

I. Purpose

OVEREXPRESSION of epidermal growth factor (EGF) receptor 2 (HER2) has been associated with high rate of tumor formation [1]. Upon forming heterodimers with ligand activated EGF receptors, it is rapidly transphosphorylated. HER2 mediated signaling leads to prolonged and enhanced downstream signal transduction processes that regulate cell growth, differentiation and migration [2]. HER2 is also involved in regulating EGF receptor signal transduction through mediating EGF receptor trafficking [3]. While EGF receptor down-regulation through lysosomal degradation is the main process to negatively regulate its signal strength and duration, the presence of HER2 can either reduce EGFR-HER2 heterodimer internalization or enhance receptor recycling, therefore efficiently sustains EGF signaling intensity. Numerous experimental approaches and computational methods have been applied to discover the underlying mechanisms which remain unclear in EGF receptor system. We intend to quantitatively examine the effect of HER2 overexpression on EGR receptor signaling using both methods.

II. Methods and Results

A. Experimental Method

Human mammary epithelial cell line (HMEC) 184A1-1 was transfected with HER2 gene and then subcloned to establish a set of genetically related cell lines expressing different levels of HER2. Among them three clones were selected, cultured as described [4], and activated with 100ng/ml EGF for various times. EGF receptor mass and surface/internal phosphorylated receptor levels were simultaneously monitored using the ELISA technique [5].

B. Kinetic Modeling

A simple model of EGFR and HER2 interaction that includes their trafficking and signaling properties was constructed. ODEs quantifying both biochemical reactions and trafficking dynamics in the system were solved analytically. Model parameters were optimized by minimizing the root-mean-squared deviation between the model generated result and the experimental data.

C. Results

Our experimental results confirm that: 1) Internalization and degradation rate of EGFR slow down as the HER2 expression level increase. 2) While HER2 overexpression shifts the bias in EGFR signaling towards cell surface, it leads to an enhanced EGFR activation duration as well. These observations were used to deduce critical parameters in our kinetics model, which can further provide more receptor interaction information not available from the experiment.

III. Conclusion

Combining our cellular model with appropriate experiments that utilize a series of engineering HME cells expressing different levels of HER2, we could systematically investigate how overexpression of HER2 regulates the spatial distribution of EGFR and how it affects EGFR signaling patterns.

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