Published in final edited form as:

Crit Rev Oncol Hematol. 2012 December ; 84(Suppl 1): e49–e57. doi:10.1016/j.critrevonc.2010.09.002.

Therapeutic strategies and mechanisms of tumorigenesis of HER2-overexpressing breast cancer

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1. Abstract

The receptor tyrosine kinase HER2 is overexpressed in approximately 25% of breast cancers. HER2 acts as a signal amplifier for its siblings, namely three different transmembrane receptors that collectively bind with 11 distinct growth factors of the EGF family. Thus, overexpression of HER2 confers aggressive invasive growth in preclinical models and in patients. Specific therapies targeting HER2 include monoclonal antibodies, antibody-drug conjugates, small molecule tyrosine kinase inhibitors, as well as heat shock protein and sheddase inhibitors. Two of these drugs have shown impressive – yet mostly transient – efficacy in patients with HER2 overexpressing breast cancer. We highlight the biological roles of HER2 in breast cancer progression, and overview the available therapeutic armamentarium directed against this receptor-kinase molecule. Focusing on the mechanisms that confer resistance to individual HER2 targeting agents, we envisage therapeutic approaches to delay or overcome the evolvement of resistance in patients.

Keywords

breast cancer; growth factor; kinase inhibitor; oncogene; monoclonal antibody; receptor tyrosine kinase

2. Introduction

Polypeptide growth factors, such as the epidermal growth factor (EGF), play essential roles in embryogenesis, and they participate in tissue renewal and remodelling throughout adulthood. Examples of the late functions include wound healing and mammary gland morphogenesis. Growth factor actions are mediated by a group of 58 transmembrane glycoproteins, collectively named receptor tyrosine kinases (RTKs), whose cytoplasmic domains harbor an enzymatic activity, namely tyrosine-specific phosphorylation [1]. The family of EGF-like molecules, which comprises 11 different ligands sharing a 50–60 amino acid receptor-binding domain, bind with subtype I RTKs, also called the ErbB or HER family. Each receptor consists of an extracellular ligand-binding domain, a transmembrane domain, and a tyrosine kinase portion [2]. Upon ligand binding, the otherwise inactive monomeric receptors form active homodimers or heterodimers, thereby leading to receptor phosphorylation and signaling via various biochemical pathways (Fig. 1). Amongst the most prominent pathways are the phosphoinositide-3-kinase (PI3K)/AKT and the mitogen activated protein kinase (MAPK) pathway [3].

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Two ErbB receptors deviate from the prototypic mode of RTK signaling. The tyrosine kinase portion of ErbB-3/HER3 is defective [4], but ErbB-3/HER3 is able to bind ligands of the neuregulin family, as well as to trans-activate other members of the family [5]. On the other hand, ErbB-2/HER2 is incapable of ligand binding, although it is able to activate other receptors [6]. Because the four ErbB receptors can form multiple homo- and heterodimeric complexes, and each receptor dimer is able to recruit a specific repertoire of signaling cascades, the ErbB family acts as a layered network that transforms input signals in the form of growth factors, into biological outcomes, such as cell division and migration [7].

Amplification of a genomic region containing the HER2 gene on chromosome 17q12 occurs in approximately 25% of invasive breast tumors [8]. Multiple flanking genes promoting the growth of breast cancer, such as GRB7 and topoisomerase II, are frequently co-amplified along with HER2 [9]. HER2 overexpression is frequently accompanied by the occurrence of truncated forms of the receptor resulting from proteolytic cleavage, alternative translation initiation or alternative pre-mRNA splicing. These short forms are characterized by enhanced oncogenic potential [10]. Our review addresses the biochemistry underlying HER2-mediated tumorigenesis, as well as therapeutic strategies that target HER2. The reader is referred to several recent reviews that cover other aspects of HER2 [11–14].

3. The biochemistry underlying HER2's action in embryogenesis and oncogenesis

HER2 plays essential roles, primarily as a co-receptor, during embryonic development, as well as in adulthood. In embryonic development, neuregulin-promoted HER2 heterodimers with ErbB-4/HER4 are crucially involved in development of the heart, as revealed by selective inactivation of the three components [15–17]. Similarly, heterodimers containing HER2 and ErbB-3/HER3 are critical for proper development of Schwann cells and specific ganglia of the peripheral nervous system [18–20]. Interestingly, these functions of HER2 seem to derive from an ability to support migration of progenitor cells of the neural crest. Indeed, HER2 occupies central roles in additional cell functions, such as metabolism, differentiation and proliferation. These cellular functions of HER2 are unique among other RTKs: Not only this kinase is unable to bind a growth factor of its own, it serves as the preferred heterodimeric partner of ligand-activated receptors [21]. Once it joins into heterodimers, HER2 increases ligand binding affinities [6; 22] and prolongs signaling by delaying the onset of ligand-induced receptor endocytosis [23–25].

Similar to the impaired endocytosis of HER2, internalization and degradation of ErbB-3/HER3 are defective [23; 26; 27]. Nevertheless, when recruited into heterodimers, ErbB-3/HER3 potently couples to downstream signaling pathways. Especially important is the heterodimer comprising HER2 and ErbB-3/HER3: this complex is endowed with potent oncogenic activities [28]. Accumulating evidence suggest that ErbB receptors may also be recruited into heteromeric complexes with other cell surface receptors, such as the receptors for the hepatocyte growth factor and the insulin-like growth factor 1 [29–31]. In summary, these unique biochemical attributes of HER2 enable formation of especially active heterodimeric complexes, which explains why this ligand-less RTK is transforming [32; 33].

4. Clinically approved drugs that target HER2

Paul Ehrlich' vision of targeted therapy was originally presented by some 100 years ago, and materialized into immunotherapy with the development of the first monoclonal antibodies [34]. It took a quarter of a century to transform murine monoclonal antibodies into human therapeutics: Rituximab (Rituxan), a monoclonal antibody against CD20, and Trastuzumab (Herceptin), a monoclonal antibody to HER2, were approved for clinical use in lymphoma

and in breast cancer, respectively, in the last three years of the 20th century [reviewed in [35]]. Despite their wide distribution, both antigens emerged as effective targets for immunotherapy, especially when the antibodies are administered together with chemotherapy [36–38]. So far, only two drugs that target HER2, Trastuzumab and a kinase inhibitor called Lapatinib/Tykerb, are approved for clinical application in breast cancer, but several novel drugs are in development (see Fig. 2).

4.1. Trastuzumab

4.1.1. Clinical applications of Trastuzumab—Trastuzumab, a humanized mouse monoclonal antibody specific to human HER2, was initially approved for treatment of patients with HER2 overexpressing metastatic breast cancer. This approval was based on several studies demonstrating single agent activity [39; 40] and two randomized studies showing that addition of Trastuzumab to chemotherapy significantly prolonged patient survival [41; 42]. Because Trastuzumab also enhances the efficacy of neoadjuvant chemotherapy in operable or locally advanced HER2-positive tumors [43], the antibody currently represents the standard of care for patients with early or advanced stages of HER2-overexpressing breast cancer.

4.1.2. Mechanisms of action of Trastuzumab—The clinical efficacy of Trastuzumab likely entails a combination of immunological and non-immunological mechanisms. The ability of Trastuzumab to elicit antibody-dependent cellular cytotoxicity (ADCC) critically influences the efficacy of Trastuzumab-based therapies [44–46]. Non-immunological mechanisms of Trastuzumab action include the inhibition of HER2 activation and downstream signaling. Alternatively, Trastuzumab may act by removing HER2 from the cell surface [47; 48]. Because it binds to an epitope near the cleavage site of HER2's extracellular domain [49], Trastuzumab inhibits HER2 activation by metalloproteinase-mediated shedding of the extracellular domain [50]. Moreover, Trastuzumab binding may lead to disassembly of basally active, ligand-independent signaling complexes formed by HER2 [51; 52]. The resulting interference with HER2-mediated downstream signaling processes shuts down cell proliferation, angiogenesis, invasive growth, resistance to apoptosis, and DNA repair, thus sensitizing tumor cells to conventional therapeutic modalities such as chemotherapy, endocrine treatment and radiotherapy [53–58].

4.1.3. Mechanisms of resistance to Trastuzumab—Despite potent activity, the majority of HER2-overexpressing breast cancers do not respond to treatment with single-agent Trastuzumab, and in the remainder the antibody loses efficacy within one year of treatment initiation. Trastuzumab resistance can be achieved by mechanisms leading to epitope loss, as well as by mechanisms that pre-empt HER2 action. Epitope loss can result from expression of receptor variants lacking the Trastuzumab-binding site [59; 60]. Similarly, epitope loss may be induced by antigen masking via association of HER2 with the MUC4 glycoprotein [61; 62], and by aberrant HER2 glycosylation [63; 64]. Resistance to Trastuzumab by uncoupling downstream signaling pathways from HER2 may occur also by kinase switching. Examples include activation of the PI3K pathway through enhanced activity of the insulin-like growth factor receptor 1 or the hepatocyte growth factor receptor, c-MET [65–67] and/or by mutations conferring constitutive activity of downstream components. For instance, loss of the lipid phosphatase PTEN [68] and gain-of-function mutations of PI3K [69; 70] have been shown to enhance resistance to Trastuzumab.

4.1.4. Combinatorial antibody therapies employing Trastuzumab—The enhanced therapeutic efficacy of Trastuzumab in combination with conventional therapeutic modalities has fostered combinations of the antibody with other targeted therapies. Aside from simultaneous targeting of multiple pathways essential for cancer growth (e.g., by

combining HER2-targeted therapies with anti-angiogenic agents [71]), an emerging therapeutic strategy entails the combination of two antibodies targeting distinct epitopes of HER2. The proposed mechanisms include enhanced receptor degradation [47; 72] and improved recruitment of immune effector cells [73]. A recent phase II trial, combining Trastuzumab with a second HER2-directed antibody, Pertuzumab, in the treatment of Trastuzumab-refractory metastatic breast cancer found the combination to be effective in 50% of treated patients [74]. Pertuzumab, like Trastuzumab, is a monoclonal antibody targeting HER2, but it binds to a receptor domain that mediates heterodimerization of HER2 with ligand-bound ErbB receptors including EGFR and ErbB-3/HER3. As single agent, Pertuzumab showed only moderate efficacy in patients with Trastuzumab-refractory HER2 overexpressing breast cancers. In combination with Trastuzumab, however, Pertuzumab may induce significant clinical responses in many patients, including those, in which either antibody alone had failed [75–78].

4.2. Lapatinib

Small molecule kinase inhibitors bind either reversibly or irreversibly to the nucleotide-binding cleft of their target kinases. Lapatinib is a highly specific, reversible inhibitor that blocks the catalytic action of both HER2 and EGFR [79]. Experiments in vitro and in animals established the ability of Lapatinib to inhibit both the intact form of HER2 and the truncated intracellular form (p95-HER2), which is not recognized by Trastuzumab [80].

4.2.1. Clinical applications of Lapatinib—The combination of Lapatinib and chemotherapy (capecitabine) was found to be superior to capecitabine alone in women with HER2-positive advanced breast cancer that progressed after treatment with regimens that included Trastuzumab, an anthracycline and a taxane [81; 82]. In addition, Lapatinib demonstrated clinical activity and was well tolerated as first-line monotherapy in HER2-amplified, locally advanced or metastatic breast cancer [83]. Potential future applications of Lapatinib may include inflammatory breast cancer, an aggressive disease with a relatively high frequency of HER2 overexpression [84].

4.2.2. Mechanisms of action of Lapatinib—Lapatinib response correlated with EGFR and HER2 expression levels in patients' tumors, and associated with increased pre-treatment expression of phosphorylated-HER2 (p-HER2) [85]. Lapatinib is able to induce apoptosis of Trastuzumab-resistant breast cancer cells via alteration of IGF-1 signaling [86], and also block NRG-induced p95-HER2/HER3 heterodimer formation [87].

4.2.3. Mechanisms of resistance to Lapatinib—Mechanisms of resistance to Lapatinib include enhanced engagement of compensatory signaling pathways, such as increased estrogen receptor activity [88], calcium-dependent activation of RelA, a pro-survival subunit of NFκB [89], Ras-induced MEK activation [90] and hyperactivation of PI3K [91].

5. Experimental therapeutic approaches targeting HER2

Currently, several novel strategies to target HER2 are in various phases of pre-clinical and clinical development (see Fig. 2). HSP90 inhibitors are able to block signaling downstream to HER2 and p95-HER2 [92], and they can inhibit growth of human HER2-overexpressing mammary xenografts [93]. Mechanistically, HSP90 inhibitors bind to the ATP/ADP binding pocket of HSP90, thereby impairing the chaperoning function and leading to proteasomal degradation of HER2 [94; 95]. Indeed, two compounds have already shown activity in combination with Trastuzumab in antibody-refractory breast cancer patients [96; 97]. Yet another novel strategy combines inhibitors of matrix metalloproteinases with anti-HER2

targeted therapies [98]. An interesting approach makes use of irreversible pan-HER/Erbb kinase inhibitors like Neratinib [99; 100], which has already demonstrated activity as single agent and in combination with Trastuzumab [101; 102]. Last, it is notable that tumors that become independent on HER2 signaling still maintain expression of the oncoprotein. This attribute is exploited by the antibody-drug conjugate Trastuzumab-DM1 [103], which delivers the anti-microtubule agent DM1 to HER2-positive cells, and has shown activity in early trials [104; 105].

6. Perspectives

Detailed characterization of the aberrant expression of HER2 in breast lesions, along with deep understanding of the biochemistry underlying the tumorigenic effects of an overexpressed HER2, culminated in the approval of the first drug targeting the oncoprotein in mammary carcinoma in 1998. The next drug, Lapatinib, entered clinical use less than a decade later, but primary and secondary (evolving) patient resistance emerged as a major obstacle in the mission to eradicate HER2 overexpressing tumors. We predict that similarly deep understanding of other oncogenic mechanisms will guide future attempts to target additional oncoproteins and identify novel drug combinations.

Acknowledgments

AE is supported by a postdoctoral fellowship of the Minerva Foundation. Our research is supported by grants from the National Cancer Institute (4R37CA072981), the Seventh Framework Program (FP7) of the European Commission, the German-Israeli Project Cooperation (DIP/DFG), the Israel Cancer Research Fund, the Dr. Miriam and Sheldon G. Adelson Medical Research Foundation, the Kekst Family Institute for Medical Genetics, the Kirk Center for Childhood Cancer and Immunological Disorders, the Women's Health Research Center funded by Bennett-Pritzker Endowment Fund, the Marvelle Koffler Program for Breast Cancer Research, the Leir Charitable Foundation and the M.D. Moross Institute for Cancer Research. Y.Y. is the incumbent of the Harold and Zelda Goldenberg Professorial Chair.

References

1. Witsch E, Sela M, Yarden Y. Roles for growth factors in cancer progression. *Physiology* (Bethesda). 2010; 25:85–101. [PubMed: 20430953]
2. Olayioye MA, Neve RM, Lane HA, Hynes NE. The ErbB signaling network: receptor heterodimerization in development and cancer. *Embo J*. 2000; 19:3159–67. [PubMed: 10880430]
3. Yarden Y, Sliwkowski MX. Untangling the ErbB signalling network. *Nat Rev Mol Cell Biol*. 2001; 2:127–37. [PubMed: 11252954]
4. Berger MB, Mendrola JM, Lemmon MA. ErbB3/HER3 does not homodimerize upon neuregulin binding at the cell surface. *FEBS Lett*. 2004; 569:332–6. [PubMed: 15225657]
5. Jura N, Shan Y, Cao X, Shaw DE, Kuriyan J. Structural analysis of the catalytically inactive kinase domain of the human EGF receptor 3. *Proc Natl Acad Sci U S A*. 2009; 106:21608–13. [PubMed: 20007378]
6. Klapper LN, Glathe S, Vaisman N, Hynes NE, Andrews GC, Sela M, et al. The ErbB-2/HER2 oncoprotein of human carcinomas may function solely as a shared coreceptor for multiple stroma-derived growth factors. *Proc Natl Acad Sci U S A*. 1999; 96:4995–5000. [PubMed: 10220407]
7. Citri A, Yarden Y. EGF-ERBB signalling: towards the systems level. *Nat Rev Mol Cell Biol*. 2006; 7:505–16. [PubMed: 16829981]
8. Slamon DJ, Clark GM, Wong SG, Levin WJ, Ullrich A, McGuire WL. Human breast cancer: correlation of relapse and survival with amplification of the HER-2/neu oncogene. *Science*. 1987; 235:177–82. [PubMed: 3798106]
9. Neve RM, Chin K, Fridlyand J, Yeh J, Baehner FL, Fevr T, et al. A collection of breast cancer cell lines for the study of functionally distinct cancer subtypes. *Cancer Cell*. 2006; 10:515–27. [PubMed: 17157791]

10. Anido J, Scaltriti M, Bech Serra JJ, Santiago Josef B, Todo FR, Baselga J, et al. Biosynthesis of tumorigenic HER2 C-terminal fragments by alternative initiation of translation. *Embo J*. 2006; 25:3234–44. [PubMed: 16794579]
11. Baselga J, Swain SM. Novel anticancer targets: revisiting ERBB2 and discovering ERBB3. *Nat Rev Cancer*. 2009; 9:463–75. [PubMed: 19536107]
12. Jones KL, Buzdar AU. Evolving novel anti-HER2 strategies. *Lancet Oncol*. 2009; 10:1179–87. [PubMed: 19959074]
13. Nahta R, Shabaya S, Ozbay T, Rowe DL. Personalizing HER2-targeted therapy in metastatic breast cancer beyond HER2 status: what we have learned from clinical specimens. *Curr Pharmacogenomics Person Med*. 2009; 7:263–74. [PubMed: 20300449]
14. Sauter G, Lee J, Bartlett JM, Slamon DJ, Press MF. Guidelines for human epidermal growth factor receptor 2 testing: biologic and methodologic considerations. *J Clin Oncol*. 2009; 27:1323–33. [PubMed: 19204209]
15. Gassmann M, Casagrande F, Orioli D, Simon H, Lai C, Klein R, et al. Aberrant neural and cardiac development in mice lacking the ErbB4 neuregulin receptor. *Nature*. 1995; 378:390–4. [PubMed: 7477376]
16. Lee KF, Simon H, Chen H, Bates B, Hung MC, Hauser C. Requirement for neuregulin receptor erbB2 in neural and cardiac development. *Nature*. 1995; 378:394–8. [PubMed: 7477377]
17. Meyer D, Birchmeier C. Multiple essential functions of neuregulin in development. *Nature*. 1995; 378:386–90. [PubMed: 7477375]
18. Morris JK, Lin W, Hauser C, Marchuk Y, Getman D, Lee KF. Rescue of the cardiac defect in ErbB2 mutant mice reveals essential roles of ErbB2 in peripheral nervous system development. *Neuron*. 1999; 23:273–83. [PubMed: 10399934]
19. Woldeyesus MT, Britsch S, Riethmacher D, Xu L, Sonnenberg-Riethmacher E, Abou-Rebyeh F, et al. Peripheral nervous system defects in erbB2 mutants following genetic rescue of heart development. *Genes Dev*. 1999; 13:2538–48. [PubMed: 10521398]
20. Riethmacher D, Sonnenberg-Riethmacher E, Brinkmann V, Yamaai T, Lewin GR, Birchmeier C. Severe neuropathies in mice with targeted mutations in the ErbB3 receptor. *Nature*. 1997; 389:725–30. [PubMed: 9338783]
21. Tzahar E, Waterman H, Chen X, Levkowitz G, Karunakaran D, Lavi S, et al. A hierarchical network of interreceptor interactions determines signal transduction by Neu differentiation factor/neuregulin and epidermal growth factor. *Mol Cell Biol*. 1996; 16:5276–87. [PubMed: 8816440]
22. Pinkas-Kramarski R, Lenferink AE, Bacus SS, Lyass L, van de Poll ML, Klapper LN, et al. The oncogenic ErbB-2/ErbB-3 heterodimer is a surrogate receptor of the epidermal growth factor and betacellulin. *Oncogene*. 1998; 16:1249–58. [PubMed: 9546426]
23. Baulida J, Kraus MH, Alimandi M, Di Fiore PP, Carpenter G. All ErbB receptors other than the epidermal growth factor receptor are endocytosis impaired. *J Biol Chem*. 1996; 271:5251–7. [PubMed: 8617810]
24. Pinkas-Kramarski R, Soussan L, Waterman H, Levkowitz G, Alroy I, Klapper L, et al. Diversification of Neu differentiation factor and epidermal growth factor signaling by combinatorial receptor interactions. *Embo J*. 1996; 15:2452–67. [PubMed: 8665853]
25. Worthylake R, Wiley HS. Structural aspects of the epidermal growth factor receptor required for transmodulation of erbB-2/neu. *J Biol Chem*. 1997; 272:8594–601. [PubMed: 9079690]
26. Waterman H, Alroy I, Strano S, Seger R, Yarden Y. The C-terminus of the kinase-defective neuregulin receptor ErbB-3 confers mitogenic superiority and dictates endocytic routing. *Embo J*. 1999; 18:3348–58. [PubMed: 10369675]
27. Waterman H, Sabanai I, Geiger B, Yarden Y. Alternative intracellular routing of ErbB receptors may determine signaling potency. *J Biol Chem*. 1998; 273:13819–27. [PubMed: 9593726]
28. Wallasch C, Weiss FU, Niederfellner G, Jallat B, Issing W, Ullrich A. Heregulin-dependent regulation of HER2/neu oncogenic signaling by heterodimerization with HER3. *Embo J*. 1995; 14:4267–75. [PubMed: 7556068]
29. Balana ME, Labriola L, Salatino M, Movsichoff F, Peters G, Charreau EH, et al. Activation of ErbB-2 via a hierarchical interaction between ErbB-2 and type I insulin-like growth factor receptor in mammary tumor cells. *Oncogene*. 2001; 20:34–47. [PubMed: 11244498]

30. Brantley-Sieders DM, Zhuang G, Hicks D, Fang WB, Hwang Y, Cates JM, et al. The receptor tyrosine kinase EphA2 promotes mammary adenocarcinoma tumorigenesis and metastatic progression in mice by amplifying ErbB2 signaling. *J Clin Invest.* 2008; 118:64–78. [PubMed: 18079969]
31. Liu D, Aguirre Ghiso J, Estrada Y, Ossowski L. EGFR is a transducer of the urokinase receptor initiated signal that is required for in vivo growth of a human carcinoma. *Cancer Cell.* 2002; 1:445–57. [PubMed: 12124174]
32. Di Fiore PP, Pierce JH, Fleming TP, Hazan R, Ullrich A, King CR, et al. Overexpression of the human EGF receptor confers an EGF-dependent transformed phenotype to NIH 3T3 cells. *Cell.* 1987; 51:1063–70. [PubMed: 3500791]
33. Hudziak RM, Schlessinger J, Ullrich A. Increased expression of the putative growth factor receptor p185HER2 causes transformation and tumorigenesis of NIH 3T3 cells. *Proc Natl Acad Sci U S A.* 1987; 84:7159–63. [PubMed: 2890160]
34. Kohler G, Milstein C. Continuous cultures of fused cells secreting antibody of predefined specificity. *Nature.* 1975; 256:495–7. [PubMed: 1172191]
35. Ben-Kasus T, Schechter B, Sela M, Yarden Y. Cancer therapeutic antibodies come of age: targeting minimal residual disease. *Mol Oncol.* 2007; 1:42–54. [PubMed: 19383286]
36. McLaughlin P, Grillo-Lopez AJ, Link BK, Levy R, Czuczman MS, Williams ME, et al. Rituximab chimeric anti-CD20 monoclonal antibody therapy for relapsed indolent lymphoma: half of patients respond to a four-dose treatment program. *J Clin Oncol.* 1998; 16:2825–33. [PubMed: 9704735]
37. Baselga J, Tripathy D, Mendelsohn J, Baughman S, Benz CC, Dantis L, et al. Phase II study of weekly intravenous recombinant humanized anti-p185HER2 monoclonal antibody in patients with HER2/neu-overexpressing metastatic breast cancer. *J Clin Oncol.* 1996; 14:737–44. [PubMed: 8622019]
38. Pegram MD, Lipton A, Hayes DF, Weber BL, Baselga JM, Tripathy D, et al. Phase II study of receptor-enhanced chemosensitivity using recombinant humanized anti-p185HER2/neu monoclonal antibody plus cisplatin in patients with HER2/neu-overexpressing metastatic breast cancer refractory to chemotherapy treatment. *J Clin Oncol.* 1998; 16:2659–71. [PubMed: 9704716]
39. Cobleigh MA, Vogel CL, Tripathy D, Robert NJ, Scholl S, Fehrenbacher L, et al. Multinational study of the efficacy and safety of humanized anti-HER2 monoclonal antibody in women who have HER2-overexpressing metastatic breast cancer that has progressed after chemotherapy for metastatic disease. *J Clin Oncol.* 1999; 17:2639–48. [PubMed: 10561337]
40. Vogel CL, Cobleigh MA, Tripathy D, Gutheil JC, Harris LN, Fehrenbacher L, et al. Efficacy and safety of trastuzumab as a single agent in first-line treatment of HER2-overexpressing metastatic breast cancer. *J Clin Oncol.* 2002; 20:719–26. [PubMed: 11821453]
41. Slamon DJ, Leyland-Jones B, Shak S, Fuchs H, Paton V, Bajamonde A, et al. Use of chemotherapy plus a monoclonal antibody against HER2 for metastatic breast cancer that overexpresses HER2. *N Engl J Med.* 2001; 344:783–92. [PubMed: 11248153]
42. Marty M, Cogne F, Maraninchi D, Snyder R, Mauriac L, Tubiana-Hulin M, et al. Randomized phase II trial of the efficacy and safety of trastuzumab combined with docetaxel in patients with human epidermal growth factor receptor 2-positive metastatic breast cancer administered as first-line treatment: the M77001 study group. *J Clin Oncol.* 2005; 23:4265–74. [PubMed: 15911866]
43. Untch M, Rezaei M, Loibl S, Fasching PA, Huober J, Tesch H, et al. Neoadjuvant treatment with trastuzumab in HER2-positive breast cancer: results from the GeparQuattro study. *J Clin Oncol.* 2010; 28:2024–31. [PubMed: 20308670]
44. Clynes RA, Towers TL, Presta LG, Ravetch JV. Inhibitory Fc receptors modulate in vivo cytotoxicity against tumor targets. *Nat Med.* 2000; 6:443–6. [PubMed: 10742152]
45. Varchetta S, Gibelli N, Oliviero B, Nardini E, Gennari R, Gatti G, et al. Elements related to heterogeneity of antibody-dependent cell cytotoxicity in patients under trastuzumab therapy for primary operable breast cancer overexpressing Her2. *Cancer Res.* 2007; 67:11991–9. [PubMed: 18089830]
46. Musolino A, Naldi N, Bortesi B, Pezzuolo D, Capelletti M, Missale G, et al. Immunoglobulin G fragment C receptor polymorphisms and clinical efficacy of trastuzumab-based therapy in patients

- with HER-2/neu-positive metastatic breast cancer. *J Clin Oncol.* 2008; 26:1789–96. [PubMed: 18347005]
47. Ben-Kasus T, Schechter B, Lavi S, Yarden Y, Sela M. Persistent elimination of ErbB-2/HER2-overexpressing tumors using combinations of monoclonal antibodies: relevance of receptor endocytosis. *Proc Natl Acad Sci U S A.* 2009; 106:3294–9. [PubMed: 19218427]
 48. Kasprzyk PG, Song SU, Di Fiore PP, King CR. Therapy of an animal model of human gastric cancer using a combination of anti-erbB-2 monoclonal antibodies. *Cancer Res.* 1992; 52:2771–6. [PubMed: 1349849]
 49. Cho HS, Mason K, Ramyar KX, Stanley AM, Gabelli SB, Denney DW Jr, et al. Structure of the extracellular region of HER2 alone and in complex with the Herceptin Fab. *Nature.* 2003; 421:756–60. [PubMed: 12610629]
 50. Molina MA, Codony-Servat J, Albanell J, Rojo F, Arribas J, Baselga J. Trastuzumab (herceptin), a humanized anti-Her2 receptor monoclonal antibody, inhibits basal and activated Her2 ectodomain cleavage in breast cancer cells. *Cancer Res.* 2001; 61:4744–9. [PubMed: 11406546]
 51. Junttila TT, Akita RW, Parsons K, Fields C, Lewis Phillips GD, Friedman LS, et al. Ligand-independent HER2/HER3/PI3K complex is disrupted by trastuzumab and is effectively inhibited by the PI3K inhibitor GDC-0941. *Cancer Cell.* 2009; 15:429–40. [PubMed: 19411071]
 52. Pickl M, Ries CH. Comparison of 3D and 2D tumor models reveals enhanced HER2 activation in 3D associated with an increased response to trastuzumab. *Oncogene.* 2009; 28:461–8. [PubMed: 18978815]
 53. Pietras RJ, Pegram MD, Finn RS, Maneval DA, Slamon DJ. Remission of human breast cancer xenografts on therapy with humanized monoclonal antibody to HER-2 receptor and DNA-reactive drugs. *Oncogene.* 1998; 17:2235–49. [PubMed: 9811454]
 54. Pietras RJ, Poen JC, Gallardo D, Wongvipat PN, Lee HJ, Slamon DJ. Monoclonal antibody to HER-2/neureceptor modulates repair of radiation-induced DNA damage and enhances radiosensitivity of human breast cancer cells overexpressing this oncogene. *Cancer Res.* 1999; 59:1347–55. [PubMed: 10096569]
 55. Dubska L, Andera L, Sheard MA. HER2 signaling downregulation by trastuzumab and suppression of the PI3K/Akt pathway: an unexpected effect on TRAIL-induced apoptosis. *FEBS Lett.* 2005; 579:4149–58. [PubMed: 16023111]
 56. Pegram MD, Konecny GE, O'Callaghan C, Beryt M, Pietras R, Slamon DJ. Rational combinations of trastuzumab with chemotherapeutic drugs used in the treatment of breast cancer. *J Natl Cancer Inst.* 2004; 96:739–49. [PubMed: 15150302]
 57. Cuello M, Ettenberg SA, Clark AS, Keane MM, Posner RH, Nau MM, et al. Down-regulation of the erbB-2 receptor by trastuzumab (herceptin) enhances tumor necrosis factor-related apoptosis-inducing ligand-mediated apoptosis in breast and ovarian cancer cell lines that overexpress erbB-2. *Cancer Res.* 2001; 61:4892–900. [PubMed: 11406568]
 58. Izumi Y, Xu L, di Tomaso E, Fukumura D, Jain RK. Tumour biology: herceptin acts as an anti-angiogenic cocktail. *Nature.* 2002; 416:279–80. [PubMed: 11907566]
 59. Kwong KY, Hung MC. A novel splice variant of HER2 with increased transformation activity. *Mol Carcinog.* 1998; 23:62–8. [PubMed: 9808159]
 60. Scaltriti M, Rojo F, Ocana A, Anido J, Guzman M, Cortes J, et al. Expression of p95HER2, a truncated form of the HER2 receptor, and response to anti-HER2 therapies in breast cancer. *J Natl Cancer Inst.* 2007; 99:628–38. [PubMed: 17440164]
 61. Price-Schiavi SA, Jepson S, Li P, Arango M, Rudland PS, Yee L, et al. Rat Muc4 (sialomucin complex) reduces binding of anti-ErbB2 antibodies to tumor cell surfaces, a potential mechanism for herceptin resistance. *Int J Cancer.* 2002; 99:783–91. [PubMed: 12115478]
 62. Nagy P, Friedlander E, Tanner M, Kapanen AI, Carraway KL, Isola J, et al. Decreased accessibility and lack of activation of ErbB2 in JIMT-1, a herceptin-resistant, MUC4-expressing breast cancer cell line. *Cancer Res.* 2005; 65:473–82. [PubMed: 15695389]
 63. Matsumoto K, Shimizu C, Arai T, Andoh M, Katsumata N, Kohno T, et al. Identification of predictive biomarkers for response to trastuzumab using plasma FUCA activity and N-glycan identified by MALDI-TOF-MS. *J Proteome Res.* 2009; 8:457–62. [PubMed: 19140672]

64. Palyi-Krek Z, Barok M, Isola J, Tammi M, Szollosi J, Nagy P. Hyaluronan-induced masking of ErbB2 and CD44-enhanced trastuzumab internalisation in trastuzumab resistant breast cancer. *Eur J Cancer*. 2007; 43:2423–33. [PubMed: 17911008]
65. Lu Y, Zi X, Zhao Y, Mascarenhas D, Pollak M. Insulin-like growth factor-I receptor signaling and resistance to trastuzumab (Herceptin). *J Natl Cancer Inst*. 2001; 93:1852–7. [PubMed: 11752009]
66. Nahta R, Hung MC, Esteva FJ. The HER-2-targeting antibodies trastuzumab and pertuzumab synergistically inhibit the survival of breast cancer cells. *Cancer Res*. 2004; 64:2343–6. [PubMed: 15059883]
67. Shattuck DL, Miller JK, Carraway KL 3rd, Sweeney C. Met receptor contributes to trastuzumab resistance of Her2-overexpressing breast cancer cells. *Cancer Res*. 2008; 68:1471–7. [PubMed: 18316611]
68. Nagata Y, Lan KH, Zhou X, Tan M, Esteva FJ, Sahin AA, et al. PTEN activation contributes to tumor inhibition by trastuzumab, and loss of PTEN predicts trastuzumab resistance in patients. *Cancer Cell*. 2004; 6:117–27. [PubMed: 15324695]
69. Berns K, Horlings HM, Hennessy BT, Madiredjo M, Hijmans EM, Beelen K, et al. A functional genetic approach identifies the PI3K pathway as a major determinant of trastuzumab resistance in breast cancer. *Cancer Cell*. 2007; 12:395–402. [PubMed: 17936563]
70. Kataoka Y, Mukohara T, Shimada H, Saijo N, Hirai M, Minami H. Association between gain-of-function mutations in PIK3CA and resistance to HER2-targeted agents in HER2-amplified breast cancer cell lines. *Ann Oncol*. 2010; 21:255–62. [PubMed: 19633047]
71. Pegram M, Chan D, Dichmann R, Tan-Chiu E, Yeon C, Durna L, et al. Phase II combined biological therapy targeting the HER2 proto-oncogene and the vascular endothelial growth factor using trastuzumab (T) and bevacizumab (B) as first line treatment of HER2-amplified breast cancer. *Breast Cancer Res Treat*. 2006; 100 abstract 301.
72. Friedman LM, Rinon A, Schechter B, Lyass L, Lavi S, Bacus SS, et al. Synergistic down-regulation of receptor tyrosine kinases by combinations of mAbs: implications for cancer immunotherapy. *Proc Natl Acad Sci U S A*. 2005; 102:1915–20. [PubMed: 15684082]
73. Spiridon CI, Guinn S, Vitetta ES. A comparison of the in vitro and in vivo activities of IgG and F(ab')₂ fragments of a mixture of three monoclonal anti-Her-2 antibodies. *Clin Cancer Res*. 2004; 10:3542–51. [PubMed: 15161714]
74. Baselga J, Gelmon KA, Verma S, Wardley A, Conte P, Miles D, et al. Phase II trial of pertuzumab and trastuzumab in patients with human epidermal growth factor receptor 2-positive metastatic breast cancer that progressed during prior trastuzumab therapy. *J Clin Oncol*. 2010; 28:1138–44. [PubMed: 20124182]
75. Gelmon K, Fumoleau P, Verma S, Wardley A, Conte P, Miles D, et al. Results of a phase II trial of trastuzumab (H) and pertuzumab (P) in patients (pts) with HER2-positive metastatic breast cancer (MBC) who had progressed during trastuzumab therapy. *J Clin Oncol*. 2008; 26 abstr 1026.
76. Portera CC, Walshe JM, Rosing DR, Denduluri N, Berman AW, Vatas U, et al. Cardiac toxicity and efficacy of trastuzumab combined with pertuzumab in patients with human epidermal growth factor receptor 2-positive metastatic breast cancer. *Clin Cancer Res*. 2008; 14:2710–6. [PubMed: 18451236]
77. Cortés J, Baselga J, Petrella T, Gelmon K, Fumoleau P, Verma S, et al. Pertuzumab monotherapy following trastuzumab-based treatment: Activity and tolerability in patients with advanced HER2-positive breast cancer. *J Clin Oncol*. 2009; 27 abstract 1022.
78. Walshe JM, Denduluri N, Berman AW, Rosing DR, Swain SM. A phase II trial with trastuzumab and pertuzumab in patients with HER2-overexpressed locally advanced and metastatic breast cancer. *Clinical breast cancer*. 2006; 6:535–9. [PubMed: 16595039]
79. Fabian MA, Biggs WH 3rd, Treiber DK, Atteridge CE, Azimioara MD, Benedetti MG, et al. A small molecule-kinase interaction map for clinical kinase inhibitors. *Nat Biotechnol*. 2005; 23:329–36. [PubMed: 15711537]
80. Xia W, Bacus S, Husain I, Liu L, Zhao S, Liu Z, et al. Resistance to ErbB2 tyrosine kinase inhibitors in breast cancer is mediated by calcium-dependent activation of RelA. *Mol Cancer Ther*. 2008; 9:292–9. [PubMed: 20124457]

81. Geyer CE, Forster J, Lindquist D, Chan S, Romieu CG, Pienkowski T, et al. Lapatinib plus capecitabine for HER2-positive advanced breast cancer. *N Engl J Med*. 2006; 355:2733–43. [PubMed: 17192538]
82. Ryan Q, Ibrahim A, Cohen MH, Johnson J, Ko CW, Sridhara R, et al. FDA drug approval summary: lapatinib in combination with capecitabine for previously treated metastatic breast cancer that overexpresses HER-2. *Oncologist*. 2008; 13:1114–9. [PubMed: 18849320]
83. Gomez HL, Doval DC, Chavez MA, Ang PC, Aziz Z, Nag S, et al. Efficacy and safety of lapatinib as first-line therapy for ErbB2-amplified locally advanced or metastatic breast cancer. *J Clin Oncol*. 2008; 26:2999–3005. [PubMed: 18458039]
84. Kaufman B, Trudeau M, Awada A, Blackwell K, Bachelot T, Salazar V, et al. Lapatinib monotherapy in patients with HER2-overexpressing relapsed or refractory inflammatory breast cancer: final results and survival of the expanded HER2+ cohort in EGF103009, a phase II study. *Lancet Oncol*. 2009; 10:581–8. [PubMed: 19394894]
85. Spector NL, Xia W, Burris H 3rd, Hurwitz H, Dees EC, Dowlati A, et al. Study of the biologic effects of lapatinib, a reversible inhibitor of ErbB1 and ErbB2 tyrosine kinases, on tumor growth and survival pathways in patients with advanced malignancies. *J Clin Oncol*. 2005; 23:2502–12. [PubMed: 15684311]
86. Nahta R, Yuan LX, Du Y, Esteva FJ. Lapatinib induces apoptosis in trastuzumab-resistant breast cancer cells: effects on insulin-like growth factor I signaling. *Mol Cancer Ther*. 2007; 6:667–74. [PubMed: 17308062]
87. Xia W, Liu LH, Ho P, Spector NL. Truncated ErbB2 receptor (p95ErbB2) is regulated by heregulin through heterodimer formation with ErbB3 yet remains sensitive to the dual EGFR/ ErbB2 kinase inhibitor GW572016. *Oncogene*. 2004; 23:646–53. [PubMed: 14737100]
88. Xia W, Bacus S, Hegde P, Husain I, Strum J, Liu L, et al. A model of acquired autoresistance to a potent ErbB2 tyrosine kinase inhibitor and a therapeutic strategy to prevent its onset in breast cancer. *Proc Natl Acad Sci U S A*. 2006; 103:7795–800. [PubMed: 16682622]
89. Xia W, Bacus S, Husain I, Liu L, Zhao S, Liu Z, et al. Resistance to ErbB2 tyrosine kinase inhibitors in breast cancer is mediated by calcium-dependent activation of RelA. *Mol Cancer Ther*. 9:292–9. [PubMed: 20124457]
90. Zoppoli G, Moran E, Soncini D, Cea M, Garuti A, Rocco I, et al. Ras-induced resistance to lapatinib is overcome by MEK inhibition. *Curr Cancer Drug Targets*. 10:168–75. [PubMed: 20088787]
91. Eichhorn PJ, Gili M, Scaltriti M, Serra V, Guzman M, Nijkamp W, et al. Phosphatidylinositol 3-kinase hyperactivation results in lapatinib resistance that is reversed by the mTOR/ phosphatidylinositol 3-kinase inhibitor NVP-BEZ235. *Cancer Res*. 2008; 68:9221–30. [PubMed: 19010894]
92. Chandarlapaty S, Scaltriti M, Angelini P, Ye Q, Guzman M, Hudis CA, et al. Inhibitors of HSP90 block p95-HER2 signaling in Trastuzumab-resistant tumors and suppress their growth. *Oncogene*. 29:325–34. [PubMed: 19855434]
93. Leow CC, Chesebrough J, Coffman KT, Fazenbaker CA, Gooya J, Weng D, et al. Antitumor efficacy of IPI-504, a selective heat shock protein 90 inhibitor against human epidermal growth factor receptor 2-positive human xenograft models as a single agent and in combination with trastuzumab or lapatinib. *Mol Cancer Ther*. 2009; 8:2131–41. [PubMed: 19671750]
94. Mimnaugh EG, Chavany C, Neckers L. Polyubiquitination and proteasomal degradation of the p185c-erbB-2 receptor protein-tyrosine kinase induced by geldanamycin. *J Biol Chem*. 1996; 271:22796–801. [PubMed: 8798456]
95. Workman P, Burrows F, Neckers L, Rosen N. Drugging the cancer chaperone HSP90: combinatorial therapeutic exploitation of oncogene addiction and tumor stress. *Ann N Y Acad Sci*. 2007; 1113:202–16. [PubMed: 17513464]
96. Modi S, Miller K, Rosen LS, Schneider B, Chap L, Zhong Z, et al. Alveposimycin (KOS-1022) and trastuzumab (T): Activity in HER2+ metastatic breast cancer (MBC). *Breast Cancer Symposium*; 2008; ASCO; 2007. abstract 165

97. Modi S, Sugarman S, Stopeck A, Linden H, Ma W, Kersey K, et al. Phase II trial of the Hsp90 inhibitor tanespmycin (Tan) + trastuzumab (T) in patients (pts) with HER2-positive metastatic breast cancer (MBC). *J Clin Oncol*. 2008; 26 abstract 1027.
98. Witters L, Scherle P, Friedman S, Fridman J, Caulder E, Newton R, et al. Synergistic inhibition with a dual epidermal growth factor receptor/HER-2/neu tyrosine kinase inhibitor and a disintegrin and metalloprotease inhibitor. *Cancer Res*. 2008; 68:7083–9. [PubMed: 18757423]
99. Tsou HR, Overbeek-Klumpers EG, Hallett WA, Reich MF, Floyd MB, Johnson BD, et al. Optimization of 6,7-disubstituted-4-(arylamino)quinoline-3-carbonitriles as orally active, irreversible inhibitors of human epidermal growth factor receptor-2 kinase activity. *J Med Chem*. 2005; 48:1107–31. [PubMed: 15715478]
100. Rabindran SK, Discafani CM, Rosfjord EC, Baxter M, Floyd MB, Golas J, et al. Antitumor activity of HKI-272, an orally active, irreversible inhibitor of the HER-2 tyrosine kinase. *Cancer Res*. 2004; 64:3958–65. [PubMed: 15173008]
101. Burstein HJ, Sun Y, Dirix LY, Jiang Z, Paridaens R, Tan AR, et al. Neratinib, an irreversible ErbB receptor tyrosine kinase inhibitor, in patients with advanced ErbB2-positive breast cancer. *J Clin Oncol*. 2010; 28:1301–7. [PubMed: 20142587]
102. Swaby R, Blackwell K, Jiang Z, Sun Y, Dieras V, Zaman K, et al. Neratinib in combination with trastuzumab for the treatment of advanced breast cancer: A phase I/II study. *J Clin Oncol*. 2009; 27 abstract 1004.
103. Lewis Phillips GD, Li G, Dugger DL, Crocker LM, Parsons KL, Mai E, et al. Targeting HER2-positive breast cancer with trastuzumab-DM1, an antibody-cytotoxic drug conjugate. *Cancer Res*. 2008; 68:9280–90. [PubMed: 19010901]
104. Vogel CL, Burris HA, Limentani S, Borson R, O'Shaughnessy J, Vukelja S, et al. A phase II study of trastuzumab-DM1 (T-DM1), a HER2 antibody-drug conjugate (ADC), in patients (pts) with HER2+ metastatic breast cancer (MBC): Final results. *J Clin Oncol*. 2009; 27 abstract 1017.
105. Krop IE, Beeram M, Modi S, Jones SF, Holden SN, Yu W, et al. Phase I Study of Trastuzumab-DM1, a HER2 Antibody-Drug Conjugate, Given Every 3 Weeks to Patients With HER2-Positive Metastatic Breast Cancer. *J Clin Oncol*. 2010

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Figure 1. Signal transduction pathways instigated by HER2, co-receptors and EGF-like growth factors

Heterodimers of HER2/ErbB-2 and either EGFR/ErbB-1 or the kinase-defective ErbB-3/HER3 (note the letter X that symbolizes a defective cytoplasm-facing kinase domain) are shown, along with the growth factor ligands they bind. All ligands share an epidermal growth factor (EGF) motif of 50–60 amino acids. They include, in addition to EGF, epiregulin (EPG), transforming growth factor alpha (TGF- α), heparin-binding epidermal growth factor-like factor (HB-EGF), amphiregulin (AR), epiregulin (EPR) and betacellulin. Another group includes 4 classes of neuregulins (NRGs). Note that HER2 is unable to bind a ligand. Nevertheless, HER2 takes part in signaling via its own constitutive phosphorylation, as well as by trans-activation of its heterodimerization partners. Tyrosine phosphorylated

receptors are coupled to several biochemical cascades, including the phosphoinositide-3-kinase (PI3K) pathway and the extracellular signal-regulated kinase (ERK), which belongs to the MAPK family. Activation of ERK/MAPK is mediated via the RAS- RAF-MEK pathway and leads to cellular proliferation via the activation of a number of nuclear targets, including the AP1 (FOS and JUN) complex, MYC, which regulates cell cycle progression, and ELK1, a member of the ETS family of transcription factors. SHC and GRB2 are adaptor proteins sharing the ability to bind each other, as well as tyrosine phosphorylated receptors. The EGFR/HER2 heterodimer also couples to phospholipase C (PLC) and the downstream protein kinase C. On the other hand, ErbB-3/HER3-containing heterodimers strongly activate another kinase, AKT, via a lipid kinase, PI3K, leading to activation of mTOR (mechanistic target of rapamycin). Activation of AKT blocks signaling via BAD, a BH3-only protein, which contributes to tissue homeostasis by regulating initiation of apoptosis. Activation of AKT inhibits FKHR and the cyclin-dependent kinase inhibitor p27^{KIP}. The forkhead box O1 (FKHR, FOXO1) transcription factor is a member of the FOXO family of transcription factors, involved in tumor suppression and cell death.

Figure 2. Clinically approved and experimental therapeutic strategies targeting ErbB-2/HER2 in carcinomas

Trastuzumab, a humanized monoclonal antibody directed against the extracellular domain of HER2, is approved for the treatment of HER2-overexpressing breast cancer. The antibody recruits immune effector mechanisms and can induce apoptosis, block angiogenesis and inhibit tumour cell proliferation. Similarly, Pertuzumab is able to prevent heterodimerization of HER2 with other family members. Unlike the ultimate specificity of Trastuzumab and Pertuzumab to HER2, tyrosine kinase inhibitors like the reversible inhibitor Lapatinib (approved for treatment of breast cancer) and the irreversible inhibitor Neratinib variably inhibit a broad range of tyrosine kinases. The drug has completed phase II clinical trials. HSP90 is a molecular chaperone required for proper folding of protein kinases like HER2. Hence, HSP90 inhibitors, such as 17-AAG, which block the ATP/ADP binding pocket of HSP90 and target HER2 for proteasomal degradation are in clinical trials. A naturally occurring truncated form of HER2, p95-HER2, has been implicated as a mechanism conferring resistance to Trastuzumab. Its formation is mediated by processing of the

membrane bound HER2 by matrix metalloproteinases (MMPs) of the ADAM (a disintegrin and metalloproteinase) family. INCB3619 and INCB7839 are potent inhibitors of ADAM10 and ADAM17. ADAM10 is the principle sheddase for different molecules associated with tumor cell proliferation, whereas ADAM17 is the main sheddase for the EGFR ligands TGF- α , AR, NRGs, and HB-EGF. These and similar inhibitors may effectively block truncation of HER2 and onset of patient resistance to Trastuzumab, but clinical testing has not been completed.