Review Article

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Breast cancer and therapeutic deployment of growth factor receptors

Gajanan V. Sherbet

Abstract

Growth factors and their receptor play a major part in normal growth and differentiation and also in tumour development and progression. Mutations or over-expression of growth factor receptors is associated with aggressive cancers and poor prognosis for patients. Growth factor receptors are transmembrane tyrosine kinase proteins that transduce growth factor signals imparted by their binding to specific receptors leading ultimately to the induction of cell proliferation. HER2 is a human epidermal growth factor receptor. Approximately 25% of breast cancers show HER2 gene amplification and this correlates with aggressive behaviour and poor prognosis. The deployment of Herceptin (Trastuzumab), a humanised chimeric antibody against HER2, to treat HER2+ patients, has emerged as a successful approach to the treatment of breast cancers that over-express HER2 and are resistant to tamoxifen. These patients could benefit from anti-oestrogen therapy combined with blockade of HER2 signalling. Post-menopausal patients with advanced breast cancer appear to benefit significantly from this combination therapy. Combination of Herceptin with chemotherapy might yield considerable benefits in terms of reduction of recurrence and mortality. The efficacy of conjugates of anti-HER2 antibodies with cytotoxic drugs to achieve targeted delivery of the cytotoxic agents is being evaluated. The toxicity associated with the administration of monoclonal antibodies has been recognised. Cardiotoxicity, pulmonary toxicity and infusion-related problems such as anaphylaxis occur, albeit infrequently, with monoclonal antibody therapies. The EGFr (epidermal growth factor receptor) inhibitor Lapatinib (Tykerb) is a protein kinase inhibitor (a 4-anilinoquinazoline derivative), which inhibits growth factor signalling by binding to the ATP-binding pocket of both EGFr and HER2 receptor proteins. Lapatinib has shown much promise in clinical trials in patients with advanced metastatic breast cancer and is believed to have little cardiac toxicity. A strategy similar to that adopted with EGF family growth factor receptors has been used to target the vascular endothelial growth factor receptor (VEGFr) and inhibit signalling by VEGF. Avastin (Bevacizumab) is a humanised monoclonal anti-VEGFr antibody. Avastin combined with Paclitaxel improves progression-free survival and response rate in patients with advanced breast cancer. However, on account of possible side effects, Avastin has not received general approval.

Key words: Avastin (Bevacizumab), VEGFr inhibitor, EGFr Epidermal growth factor receptor, ER Oestrogen receptor, Growth factor signalling, HER2 Human epidermal growth factor receptor 2, Herceptin (Trastuzumab), Lapatinib (Tykerb) inhibitor of EGFr, Receptor tyrosine kinases, Tamoxifen resistance, VEGFr Vascular endothelial growth factor receptor

Introduction

Adjuvant modes of breast cancer therapy following surgical intervention mainly revolve round radiation therapy, chemotherapy, or hormone therapy designed to eliminate residual cancer cells. The increase in the incidence of breast cancer with age has sharply focused attention on the link between incidence and progression. It follows from this that approaches to successful treatment and patient management would converge on hormonal status as a beneficial mode of targeted therapy. A number of growth factors, besides the steroid hormones oestrogen and progesterone, are closely involved in the growth and metastatic spread of breast cancer. Recent years have seen intensive studies of the mechanisms of function of growth factors and the pathways by which they stimulate the growth of cancer cells. These studies have led the way to the targeting growth factor function as a means of controlling breast cancer development and secondary spread.

The growth factor receptors are transmembrane proteins. The binding of growth factors to the external domain activates these receptors which have tyrosine kinase activity. This activation therefore leads to the phosphorylation of signalling proteins down stream in the signalling cascade. This in turn leads to the expression of genes associated with cell proliferation and often also to the inhibition of apoptotic loss of cells.

Growth factors, growth factor receptors and tumour growth and progression

Growth factors and their receptor play a major part in normal growth and differentiation and also in tumour development and progression. Growth factors promote proliferation and induce cancer invasion. Certain growth factors, e.g. the insulin-like growth factor (IGF) might promote tumour growth by inhibiting apoptotic loss of cells[1]. Mutations or over-expression of growth factor receptors is associated with aggressive cancers and poor prognosis for patients. Growth factor receptor genes are amplified in a number of human cancers and this is reflected in the expression of the respective receptor proteins in the cancers. Growth factor receptors are transmembrane tyrosine kinase proteins and transduce the signals imparted by the binding of the growth factors to their specific receptors; this signal transmission ultimately results in the induction of cell proliferation.

Among growth factors of note in the context of this editorial are the epidermal growth factor (EGF) and the Heregulins constituting a family of EGF related growth factors. There are several isoforms of Heregulin generated by alternative RNA splicing of the heregulin gene; these isoforms bind to their receptors with different degree of affinity. The epidermal growth factor receptor (EGFr) family (also often referred to as the erb

family) includes HER1 (human epidermal growth factor receptor 1 also called EGFr), HER2, HER3 and HER4. These receptor proteins significantly resemble one another in aminoacid sequence[2]. Heregulin can bind HER3 and HER4 receptors but the ligand for HER2 has not been identified[3] and so HER2 is often described as an orphan receptor. The receptor protein consists of an external domain that binds growth factors, a transmembrane domain and an intracellular domain which possesses tyrosine phosphorylation sites[4]. Growth factors and their paralogues bind to these receptors and induce receptor oligomerisation. This activates the cytoplasmic kinase domains, which phosphorylate and activate target proteins that induce the expression of genes responsive to the growth factors. The binding and activation of the receptors is a highly specific process, but often more than one receptor might be involved in the signalling process. In this event heterodimerisation would occur between different receptors; this seems to enhance the affinity of ligand binding[5]. This process of engagement of co-receptors to enhance growth factor signalling has been described as cross-talk between the receptors. HER2 is known to be involved in cross-talk with EGFr, HER3 as well as HER4. So HER2 seems to occupy a pre-eminent position in the signalling cascade, but recently it has been suggested that HER3 might also be a prominent participant[6].

The EGFr family of receptors have been intensively investigated for their potential relationship to cancer progression and prognosis and as a potential route for treatment and patient management. Approximately 25% of breast cancers show HER2 gene amplification and this correlates aggressive behaviour and poor prognosis[7-10]. However, on the positive side the presence of HER2 receptors has provided a new treatment modality for many patients.

Monoclonal antibodies (Herceptin) have been raised against the external domain of HER2. Herceptin has provided a highly successful mode of treatment for metastatic breast cancer showing high HER2 expression and HER2 gene amplification[11-13]. Blocking receptor function with Herceptin inhibits tumour growth and possibly also microvascular density associated with tumours and vascular permeability. Furthermore, Herceptin treatment appears to reduce VEGF (vascular endothelial growth factor) expression, tumour associated microvascular density and cell proliferation in breast cancers xenografted into mice[14]. Also in murine tumour models, Herceptin reduces the number of circulating cancer cells even under circumstances where the tumour is resistant to Herceptin treatment[15], which could be a manifestation of its effects on the vasculature independently of its inhibition of HER2 signalling.

HER2 expression and tamoxifen resistance

Breast cancer growth is influenced by the sex steroid hormones oestrogen and progesterone and growth factors such as EGF and HER2 ligands. Patients with tumours that are oestrogen receptor (ER) positive are treated with tamoxifen. The latter binds ER and competitively blocks oestrogen signals. In the context of the deployment of Herceptin to treat HER2+ patients, it has emerged that tumours over-expressing HER2 are resistant to tamoxifen. These patients could benefit from anti-oestrogen therapy combined with blockade of HER2 signalling[16]. A randomized trial has indicated that post-menopausal patients with advanced breast cancer can benefit significantly from this combination therapy[17].

Among other factors that might confer tamoxifen resistance is AIB1, the steroid receptor co-activator, which is often amplified in breast cancers. In vitro studies with breast cancer cells and in vivo investigations of murine tumours have suggested the involvement of AIB1 in tamoxifen resistance[18] and in HER2 signalling[19]. In primary breast cancer also AIB1 has been linked with tamoxifen resistance[20, 21]. A second factor deserving discussion in this context is the possibility that EGFr and HER2 signalling systems might interact and contribute in this way to resistance to hormonal therapy. As mentioned elsewhere in this review, EGFr does recruit HER2 as a co-receptor in signal transduction. This is of some significance for patients with ER-negative tumours. For, we showed some years ago that a proportion of ER-ve tumours tended to be EGFr+ve[22-24]. So this would suggest the possibility that patients with ER-ve/EGFr+ tumours could conceivably benefit from Herceptin treatment (see below).

Another possible means by which tamoxifen resistance might arise has been suggested by the finding that tamoxifen and Fulvestrant, also an anti-oestrogen, appear to be able to induce breast cancer cell invasion in the absence of E-cadherin[25]. Cadherins are transmembrane proteins, which have considerable influence on cancer invasion because they alter intercellular and cell-substratum adhesion. E-cadherin is regarded as a suppressor of invasion and growth of carcinomas as the loss or mutation of E-cadherin leads to the acquisition of invasiveness. Borley et al.[25] showed that both tamoxifen and Fulvestrant induced invasion in E-cadherin deficient MCF7 breast cancer cells, but this did not occur after oestrogen depletion. These findings add a new dimension to tamoxifen resistance as potentially being mediated by recurrence resulting from induced invasive ability.

HER2 expression and adjuvant chemotherapy

It has been recognised of late that combination of Herceptin with chemotherapy might yield considerable benefits in terms of reduction of recurrence and mortality. So HER2 expression has come into the reckoning when considering the use of adjuvant chemotherapy. Combining Herceptin with either an anthracycline plus cyclophosphamide or with Paclitaxel, as first-line therapy for metastatic breast cancer over expressing the HER2 receptor, has provided significant benefits in terms of objective response, duration of response and survival as compared with chemotherapy alone. Furthermore, the benefits were related to the degree of HER2 over-expression[26]. A review

of 35 clinical trials has indicated that patients with HER2+ cancers might benefit more from anthracycline-based and taxane-based adjuvant chemotherapy than those with HER2-negative cancers[27]. Indeed, anti-HER2 antibody combined with chemotherapy is superior to HER2 anitibody and anti-oestrogen combination[17]. The benefits of adjuvant chemotherapy with anthracyclines to patients with HER2 over-expressing tumours seem to be beyond reasonable doubt. Gennari et al.[28] have provided a combined analysis of eight studies. HER2+ patients on anthracyclines had superior disease-free as well as overall survival in comparison with patients on non-anthracycline regimen. No such benefits emerged for HER2-ve patients, suggesting that one can exclude these patients from anthracycline adjuvant therapy. Also being investigated is potential synergy between antibodies against other growth factor receptors and anti-HER2 antibodies.

Attempts are also currently in progress to test the efficacy of conjugates of anti-HER2 antibodies with cytotoxic drugs to achieve targeted delivery of the cytotoxic agents. Laboratory studies are underway with Herceptin-platinum(II) complexes[29] and Herceptin-microtubule-depolymerising agents[30].

Contraindications of Herceptin regimen

The toxic side-effects of the administration of monoclonal antibodies were recognised some years ago. Cardiotoxicity, pulmonary toxicity and infusion-related problems such as anaphylaxis occur, albeit infrequently with monoclonal antibody therapies [31, 32]. These toxicities have been described with Herceptin treatment, more so in patients on anthracycline and cyclophosphamide combined with Herceptin[26]. Cardiotoxicity could occur in some patients when Herceptin is administered with anthracyclines[33, 34]. Herceptin itself can be cardiotoxic in patients receiving concurrent or prior anthracyclines[35, 36]. Cardiotoxicity is not due to structural abnormalities but Herceptin might cause myocardial dysfunction. The toxicity of anthracyclines and Herceptin could be brought about by different routes[37]. As Dinh et al.[38] have emphasised, many questions relating to Herceptin treatment still remain unanswered, e.g. optimising treatment, and combination with conventional chemotherapeutic agents, among others. Even with these caveats Herceptin may be regarded as a most efficacious agent in the treatment of HER2+ breast cancers.

EGFr inhibitor Lapatinib (Tykerb) in breast cancer treatment

As stated earlier, EGFr is over expressed in a proportion of breast cancers that are ER-negative. EGFr expression also correlates with the expression of metastasis promoting genes. Further, in the light of the function of EGFr in conjunction with HER2, it would be of considerable clinical benefit to test the effects of EGFr inhibitors in breast cancer treatment. Lapatinib is a powerful dual inhibitor of EGFr and HER2 with marked pharmacological potential.

Lapatinib is a protein kinase inhibitor (a 4-anilinoquinazoline derivative), which inhibits growth factor signalling by binding to the ATP-binding pocket of both EGFr and HER2 receptor proteins and so prevents autophosphorylation of the receptor and inhibits the signalling cascade leading to the suppression of the growth of tumours, including advanced or metastatic breast cancers resistant to Herceptin[39]. Objective responses have been achieved in 28% of patients with untreated HER2-positive tumours[40]. Clinical trials have provided promising results; Lapatinib is clinically very effective especially in advanced or metastatic breast cancer and patients with brain metastases. A phase III trial assessing the efficacy of combination of Lapatinib with Capecitabine, which is converted to 5-Flurouracil and inhibits DNA synthesis, seems to suggest a significant slowing down of disease progression by the combination as compared with capecitabine alone[41]. The efficacy of combining Lapatinib with other conventional chemotherapeutic agents is being evaluated. First-line Paclitaxel-Lapatinib combination gave significant benefits to HER-2-positive patients[42].

Lapatinib could be functioning synergistically with HER2 inhibitors, for its effect on prominently EGFr over-expressing cancers, such as colorectal cancer or squamous cell carcinoma of the head and neck, is said to be unexceptional and moderate[43]. According to Press et al.[44] the benefits of Lapatinib appear to be restricted to patients with HER2 over-expressing cancers. Lapatinib has no cardiac toxicity but does produce other toxic effects[45]. However, its toxicity whilst administered in combination with other anticancer agents has not been appraised.

Avastin in breast cancer treatment

A different route to control of tumour growth and metastatic spread has been afforded by inhibitors that target the microvasculature associated with tumours. Tumours induce the formation of neovascularisation so that tumour cells can access the vascular system and become disseminated to form distant metastases. The neovasculature is induced by VEGF, which transduces its effects by binding specifically to its receptors VEGFr (see[46]). A strategy similar to that adopted with EGF family growth factors has been used to target VEGFr, inhibit its function and inhibit the signalling by VEGF. Avastin (Bevacizumab), a humanised monoclonal anti-VEGFr antibody, is such an inhibitor. Avastin with Paclitaxel chemotherapy has been found to enhance progression-free survival and improve response rate in patients with advanced breast cancer[47]. However, on account of possible side effects, there is some reluctance to the use of Avastin. It has been approved in Europe for first line treatment of women with metastatic breast cancer, but has not been approved for use by the National Institute for Health and Clinical Excellence UK[48].

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AUTHOR DETAILS
GAJANAN V SHERBET, The Institute for Molecular Medicine, Huntington Beach CA, USA and School of Electrical, Electronic and Computer Engineering, University of Newcastle upon Tyne UK
CORRESPONDENCE: GAJANAN V SHERBET, School of Electrical, Electronic and Computer Engineering, University of Newcastle upon Tyne, Merz Court, Newcastle upon Tyne, NE1 7RU, U.K.
Email: gajanan.sherbet@ncl.ac.uk

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