

What targeted therapy for cancer is and how it is achieved: a review

In this article, Max Summerhayes describes ways in which tumours are targeted by cytotoxic drugs

Targeted therapy is sometimes discussed as if it were a new concept but this is clearly not the case because all cancer treatments must have a target (known or unknown) and even “non-selective” cytotoxic drugs must have a greater impact on malignant cells than on normal ones. Without some degree of selectivity, it would be impossible to destroy tumour cells without killing, or at least causing unacceptable harm to, the patient.

In the case of conventional cytotoxic drugs, targeting is often achieved by means of scheduling. Healthy tissues damaged by cytotoxic agents generally recover more quickly than cancerous ones and this property is exploited by giving pulses of chemotherapy every three to four weeks, which allows sufficient recovery of normal tissues between doses with the minimum possible regrowth of the tumour. Treatment scheduling is not usually considered drug targeting, but it is vital to focus the destructive power of chemotherapy where it is required. However, it is not the only way in which “non-selective” cytotoxic agents are targeted. Some have inherently greater activity than others against specific tumour types. For example, among the platinum analogues oxaliplatin, but not carboplatin or cisplatin, is a useful drug for treating colorectal cancer. In addition, nephrotoxicity, although dose-limiting for cisplatin, is not a prominent side effect of the other two drugs. Differences of this type imply that conventional cytotoxic drugs can be, to a significant degree, targeted to specific types of cancerous and healthy tissue.

Despite these observations, and the fact that the concept of targeted therapy is as old as Paul Ehrlich’s quest for a “magic bullet” in the early years of the 20th century, conventional cytotoxic drugs are not generally regarded as targeted therapies. This may reflect the process that led to their development as clinical agents. Generally, conventional cytotoxics emerged from programmes involving the synthesis and screening of large numbers of compounds for cytotoxic activity or the serendipitous discovery that a compound has such activity. In other words, development was driven by the properties of the available agents rather than a knowledge of the cancerous and healthy tissues with which they would interact.

In recent years, however, considerable effort has been expended in exploiting our growing knowledge of physiological and pathological processes to design agents with

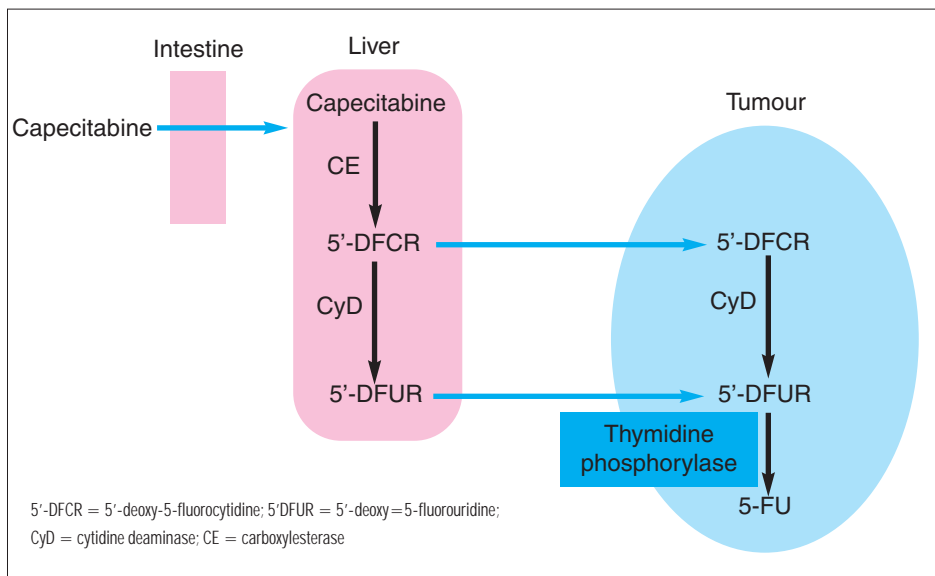


Figure 1: Three step enzymatic conversion of the non-cytotoxic pro-drug capecitabine to 5-fluorouracil resulting in preferential delivery of capecitabine to tumour tissue

properties that render them selectively active against tumour cells. These approaches can be divided into two main types: pharmacokinetic and pharmacodynamic targeting.

Pharmacokinetic targeting

Pharmacokinetic targeting depends on the preferential delivery of a non-specific cytotoxic agent to tumours. This can be achieved by harnessing biochemical pathways within tumour cells to activate a pro-drug, using physical aspects of drug formulation, employing tumour specific antibodies as drug delivery systems or applying an exogenous stimulus selectively to the tumour in order to activate an inactive pro-drug *in situ*. Some examples of these approaches follow.

Use of tumour biochemistry The fluoropyrimidine pro-drug capecitabine (Xeloda) was designed to overcome the poor and erratic oral bioavailability of the parent drug 5-fluorouracil (5-FU) and its limited tumour specificity. Capecitabine is well absorbed after oral administration and is converted into 5-FU in a three-step enzymatic process. As illustrated in Figure 1, the first step takes place in the liver while the last step requires thymidine phosphorylase, an enzyme whose levels are elevated in many common solid tumours. Following systemic capecitabine administration this can result in more 5-FU in tumours than in healthy surrounding tissues. Preferential delivery of 5-FU to tumour tissues probably explains why, in randomised

trials comparing it with conventional intravenous regimens of 5-FU, capecitabine has produced significantly higher response rates in advanced colorectal cancer,¹ and tends towards improved survival in inoperable gastric cancer^{2,3} and in the adjuvant treatment of colorectal cancer,⁴ while reducing the incidence of key fluoropyrimidine toxicities, notably leukopenia. However, it should be noted that capecitabine is associated with a higher incidence of “hand-foot syndrome” (reddening and soreness of the palms of the hands and soles of the feet) than IV 5-FU, demonstrating that actions designed to target a drug to tumour tissue can also exaggerate its actions on normal tissues with appropriate characteristics.⁴

Physical formulation The anthracycline doxorubicin is a potent cytotoxic agent with a broad spectrum of antitumour activity and it is widely used. However, its use is complicated by cardiotoxicity, which means that a cumulative dose of 450mg/m² cannot be safely exceeded. In recent years, liposomal formulations of doxorubicin (and the related drug daunorubicin) have been developed in which the free drug is entrapped in lipid vesicles.⁵ This entrapment dramatically modifies the pharmacokinetic behaviour of the anthracycline, especially when the lipid envelope incorporates polyethylene glycol as in the case of Caelyx.⁵ Following IV administration, the liposomes are largely restricted to the intravascular space. However, the incomplete

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and leaky tumour vasculature results in their preferential accumulation in cancers where they slowly release free drug. This has been reported to result in increased concentrations of doxorubicin in some tumour tissues^{5,6} and reduced levels in normal tissues. Thus the cytotoxic agent is targeted towards the tumour and away from normal tissues, including the heart. This probably explains the reduced cardiotoxicity of liposomal anthracyclines.

Again hand-foot syndrome — rarely seen with conventional anthracycline formulations — emerges as a side effect after liposomal encapsulation, possibly as a consequence of prolonged tissue exposure.⁵

Exogenous activation Exogenous activation is the principle behind photodynamic therapy, whereby a light sensitive non-cytotoxic pro-drug is administered systemically and given time to distribute around the body. A laser light source is then directed at superficial cancers, such as those of the head, neck, oesophagus and lung. This activates the pro-drug and results in a local cytotoxic action.

Pharmacodynamic targeting

Although all the above approaches are valuable, they rely ultimately on the selective delivery or production *in situ* of non-specific cytotoxic agents and, as such, they have an inherent weakness — completely selective delivery is impossible, making some degree of systemic toxicity inevitable. Even when, as in photodynamic therapy, the activation of the drug can be well localised anatomically, there is little or no specificity at cellular level and treatment will result in the death of healthy “bystander” cells as well as malignant ones. For this reason, among others, there has been great excitement in recent years about the increasing understanding of cancer biology, which has opened up the possibility of pharmacodynamically targeting malignancies.

The ideal target is one that is found only in tumour cells and that has a vital role in sustaining their viability or is essential for cell replication. However, in cancer therapeutics, as in other areas, the ideal target is a rare commodity and the only one of this type to be successfully identified and exploited, so far, is the Bcr-Abl tyrosine kinase which is found in 95 per cent of patients with chronic myeloid leukaemia (CML). CML is characterised by a specific chromosomal abnormality (the Philadelphia chromosome) caused by part of chromosome 22 breaking off and exchanging with material from chromosome 9. An entirely novel sequence of DNA is created where the genetic material joins after exchange. This encodes for the Bcr-Abl tyrosine kinase. This enzyme is not found in normal cells and its action drives the malignant behaviour of CML cells. Once this was recognised, a search began for selective inhibitors of the enzyme. These included imatinib (originally known as CGP57148 and STI571 and now marketed as Glivec), first described in 1996 by Brian Druker and his colleagues.

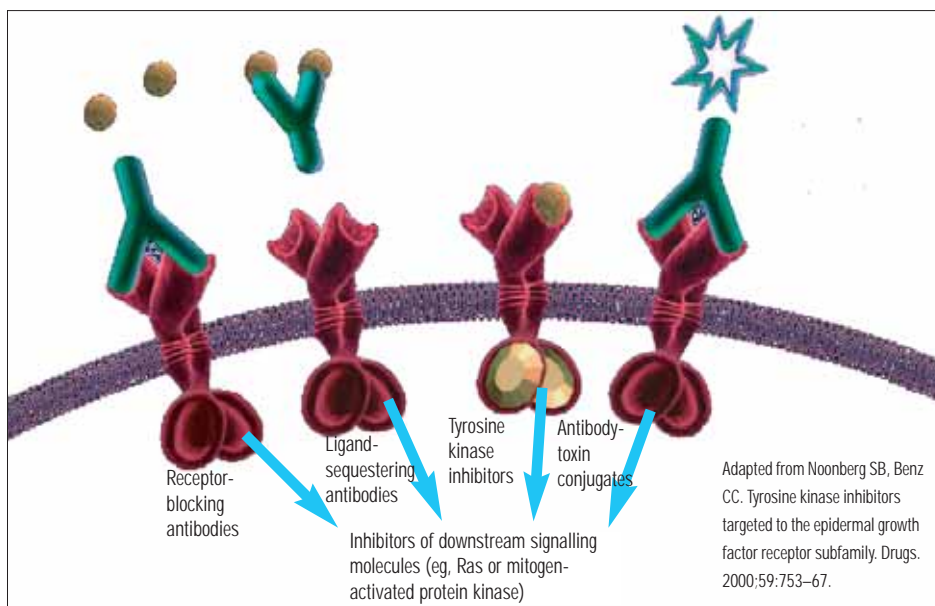


Figure 2: Possible approaches to disruption of HER receptor mediated signalling

At this year's annual meeting of the American Society for Clinical Oncology it was reported that after a median follow-up of 54 months only 9.5 per cent of 553 patients randomised to imatinib in the pivotal trial comparing it with conventional therapy (interferon alfa plus cytosine arabinoside) had stopped treatment for lack of effect and just five because of adverse events while 84 per cent remained free of disease progression on treatment.⁷ Data such as these highlight the way in which imatinib has transformed the treatment and outlook for patients with CML.

Although the efficacy and tolerability of imatinib are far better than other treatment options in CML, it still has side effects, including gastrointestinal disturbances, oedema, myalgia and rash, illustrating the point that identifying the perfect target is only half the battle. The other half lies in producing a drug that can interact with the target specifically, in order to avoid unexpected effects. In the case of imatinib its lack of complete specificity — it also inhibits the platelet-derived growth factor receptor and c-kit (stem-cell factor receptor) linked tyrosine kinases — has been exploited therapeutically. Activation of c-kit drives the growth of malignant gastrointestinal stromal cell tumours. These are rare tumours that resist conventional cytotoxic drugs, and imatinib was the first effective treatment for inoperable tumours of this type.

Although ideal targets are rare, scientific endeavour has revealed a number of molecules that have appear to have a much greater role in cancers than in mature, healthy tissues stimulating the development of drugs that interact with them. Of these, three groups represent the overwhelming majority of targeted cancer therapies to have entered routine clinical practice so far: hormonal agents, drugs interacting with the HER family of cell surface growth factor receptors and those disrupting angiogenesis (the process of tumour vascularisation).

Hormonal agents Hormonal agents, such as anti-oestrogens and anti-androgens, are much the oldest group of targeted cancer therapies and are often excluded from discussions. However, this should not be the case because they exploit the fact that many breast and prostate cancers have receptors for oestrogen and androgen, respectively, and that stimulation of these receptors drives tumour growth. Consequently, direct and indirect antagonism of sex steroids can inhibit tumour growth. Moreover, the use of anti-androgens and anti-oestrogens is, generally, well tolerated, reflecting the more minor role of sex hormones in many normal tissues.

Although well established, the hormonal treatment of breast cancer has progressed significantly in recent years with the introduction of specific aromatase inhibitors capable of blocking the synthesis of oestrogen in non-ovarian tissues. There are now multiple studies demonstrating that this class of drugs can improve long-term outcomes when used as an adjuvant treatment after surgery for early breast cancer as a substitute for, or in addition to, the oestrogen receptor antagonist tamoxifen.⁸ The National Institute for Health and Clinical Excellence has recently reviewed the evidence for the clinical and cost-effectiveness of the adjuvant use of aromatase inhibitors and recommended that they be made available to NHS patients for this purpose, according to their current marketing authorisations.⁹

HER-family antagonists HER 1–4 (also known as erbB1–4) are a family of structurally similar transmembrane proteins that act as receptors for a variety of growth regulating ligands (eg, heregulins, amphiregulin, transforming growth factor alpha, epidermal growth factor, etc) that are present in extracellular fluid. Binding of a growth factor to the extracellular portion of a receptor facilitates dimerisation with another receptor of the HER family. Ligand binding and dimerisation

trigger a chain of biochemical changes within the cell. These convey the signal encoded by the ligand from the inner surface of the cell to the nucleus, where it modulates gene activity resulting in a variety of biological effects including increased cell proliferation and migration, decreased apoptosis (programmed cell death), angiogenesis and resistance to cytotoxic drugs and ionising radiation.

As well as controlling processes of fundamental importance to the formation and sustainability of cancers, several abnormalities in the HER receptor-signalling pathways have been reported in solid tumours. For example, many solid tumours carry excessive quantities of HER1 (more commonly known as epidermal growth factor receptor; EGFR) and this overexpression has been associated with aggressive cancers, poorer prognosis and worse treatment outcomes. Additionally, a mutant version of HER1 has been identified in some tumours. This is permanently activated, despite lacking a ligand binding domain. Similarly, 20 to 30 per cent of breast cancers have overexpression of HER2 as a consequence of their DNA containing multiple copies of the relevant gene.¹⁰

Survival in women with HER2-positive breast cancer is reduced by up to 50 per cent compared with those whose tumours do not overexpress the protein.¹⁰ Again, overexpression has been associated with early spread of tumour to the lymph nodes and resistance to some drug treatments.

In short, the functions of the HER family suggested that modulation of their activity might be advantageous to cancer patients. Additionally, the aberrant HER signalling seen in many tumours indicated that these might be particularly sensitive to drugs targeted at this process, conferring a degree of selectivity on such interventions.

A number of different approaches can be taken to disrupt HER receptor signalling (see Figure 2, p519). These include:

- Using antibodies raised against the receptors themselves — these might work in several ways — by physically hindering ligand binding or receptor dimerisation or by marking out the cell for attack by the patient's immune system
- Using specific tyrosine kinase inhibitors to block the first step in the intracellular signalling pathway
- Starving the receptor of ligand by using specific antibodies to sequester it.
- Using antibodies against the receptors to deliver drugs or toxins conjugated with them selectively
- Using agents such as inhibitors of Ras and mitogen-activated protein kinase (second messengers) to interrupt the biochemical reactions that carry the signal from the cell surface to the nucleus

Of these, the first two strategies have already produced drugs that are in everyday clinical use. Examples of receptor directed antibodies include trastuzumab (Herceptin) and

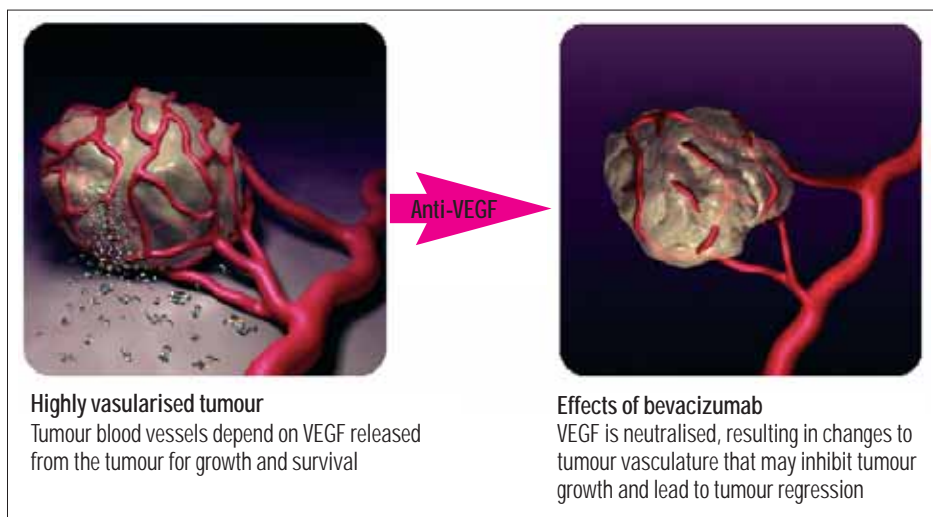


Figure 3: Effect of the the anti-VEGF antibody bevacizumab on tumour vasculature and growth

cetuximab (Erbix), and an example of a specific tyrosine kinase inhibitor is erlotinib.

Trastuzumab Trastuzumab is a chimeric monoclonal antibody created by inserting the antigen-binding site of a mouse monoclonal antibody directed against HER2 into a human immunoglobulin (IgG) skeleton. It was first described by Carter *et al* in 1992.¹¹ Early clinical trials in patients overexpressing HER2 demonstrated that, even in women with heavily pretreated metastatic disease, trastuzumab alone is capable of inducing durable disease remission. Also, when added to standard first-line chemotherapy for metastatic disease, it extends median survival by nine months — an increase of 45 per cent.¹³ These findings encouraged development of the drug for use as an adjuvant to surgery. Data are now available from 10,000 women with HER2-positive breast cancer, who were entered into four randomised controlled trials of standard adjuvant chemotherapy with and without a year of trastuzumab. These consistently showed that the addition of trastuzumab approximately halves the risk of a “treatment failure event” (recurrence, new cancer or death), and reduces the risk of death by 25–33 per cent.^{14–17} This high level of efficacy recently resulted in adjuvant trastuzumab being recommended by NICE for use within the NHS.¹⁸

Trastuzumab is generally well tolerated but, in a few patients, it can produce cardiac impairment so regular monitoring of cardiac function is necessary during treatment. It is unclear whether this toxicity is a consequence of binding to HER2 in non-tumour tissue or an unrelated action of the antibody. Either way, this again shows that it is unrealistic to expect targeted therapies to impact only on specific malignant cells.

Cetuximab Cetuximab is a chimeric antibody raised against the HER1/EGFR receptor. It has been tested in a number of solid tumours (where this receptor is thought to have a role) and it is currently licensed for use, in combi-

nation with irinotecan, by patients failing irinotecan-containing cytotoxic chemotherapy, and in combination with radiotherapy, by patients with locally advanced squamous cell cancer of the head and neck. Its activity in the latter condition is particularly impressive — it improves median overall survival from 29.3 to 49.0 months (hazard ratio for death, 0.74; $P=0.03$).¹⁹

Erlotinib Erlotinib (Tarceva) is an orally administered inhibitor of the HER1/EGFR receptor tyrosine kinase that has been most extensively investigated in the treatment of non-small-cell lung cancer where it is licensed for single-agent use in patients who have relapsed after at least one prior chemotherapy regimen. The marketing authorisation in this indication was based on the BR21 study comparing erlotinib with placebo. This demonstrated that, used second- or third-line, erlotinib improves median survival by 45 per cent,²⁰ while improving or slowing deterioration of symptoms and improving quality of life.²¹ It lacks the sometimes life-threatening myelotoxicity associated with conventional cytotoxic chemotherapy, but has its own characteristic side effects: diarrhoea and rash. Of these, mild-moderate rash is the most common. Rash affected 76 per cent of patients in the BR21 study and it is also common after other HER1/EGFR antagonists (both small molecules and antibodies²²), suggesting that it is probably a consequence of the disruption of HER1/EGFR-mediated signalling in normal tissues.

Agents inhibiting angiogenesis In 1971, Judah Folkman hypothesised that tumour growth depended on the establishment of a network of blood vessels. This process represents an attractive therapeutic target because it has little role in most mature normal tissues and involves normal vascular cells that are genetically stable and less likely than tumour cells to develop drug resistance. It was established that tumours secrete

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growth factors that trigger angiogenesis, stimulating nearby blood vessels to bud and infiltrate the tumour, permitting growth and metastatic spread via the circulation. Among these, vascular endothelial growth factor (VEGF) was identified as having a pivotal role. Elevated levels of this protein are found in a wide variety of human tumours. As with therapeutic disruption of signalling through the HER family of receptors, more than one approach can be taken to inhibiting VEGF-mediated angiogenesis, as the following examples show.

Bevacizumab Work by Ferrara *et al* led first to the development of a murine, VEGF-specific, antibody designed to sequester VEGF and neutralise its angiogenic stimulus, as shown in Figure 3 (p520). This antibody successfully inhibited the growth of a variety of human tumours growing as xenografts in mice and it was subsequently humanised (as bevacizumab) to permit its use in humans.²³ Currently, the drug is licensed, under the proprietary name Avastin, for use in conjunction with 5-FU based cytotoxic chemotherapy for the first-line treatment of metastatic colorectal cancer. In this condition it improves median survival by 30 per cent when added to combination chemotherapy with 5-FU, folinic acid and irinotecan.²⁴ Recently, it has also been demonstrated to improve long-term outcomes in breast¹⁷ and lung²⁵ cancers and is being developed for the treatment of a wide variety of tumours.

Sorafenib and sunitinib Two small molecule tyrosine kinase inhibitors, sorafenib (Nexavar) and sunitinib (Sutent), have been recently licensed for the treatment of advanced renal cell cancer relapsing after, or unsuitable for, cytokine therapy with interferon alfa or interleukin-2. Both drugs inhibit a selection of kinase enzymes, including tyrosine kinases linked to the VEGF receptor family. As such, they have antiangiogenic activity which probably contributes to their activity in relapsed renal cell cancer, where sorafenib has been shown to double progression-free survival from 12 to 24 months relative to placebo²⁶ and sunitinib has elicited tumour responses in cytokine resistant tumours.²⁷

It is currently impossible to say how important their ability to inhibit non-VEGF-linked kinase enzymes is to their clinical activity in this condition. However, it is likely that the ability of sunitinib to inhibit c-kit tyrosine kinase is central to its activity in its other licensed indication: imatinib-resistant gastrointestinal stromal cell tumours.²⁸

Conclusion

The concept of targeting therapies at cancerous cells is not new, indeed it is implicit in any attempt to treat malignancy without causing undue harm to the patient. What is novel, is the opportunity that scientists now have to design drugs that interact selectively with those processes which underpin malignant transformation.

In recent years this has led to the development of a large number of new, effective and generally well tolerated anti-cancer agents. However, few targets are truly tumour specific and some agents lack absolute target specificity. This is one of the challenges to those developing and using targeted anti-cancer drugs. A second article, to be published next week, will discuss other challenges.

DECLARATION OF INTEREST Maxwell Summerhayes is a full-time employee of Roche Products Ltd, manufacturer of several of the drugs referred to in this article, specifically bevacizumab, capecitabine, erlotinib, rituximab and trastuzumab.

References

- Twelves C on behalf of the Xeloda Colorectal Cancer Group. Capecitabine as first-line treatment in colorectal cancer: pooled data from two large, phase III trials. *European Journal of Cancer* 2002;38:S15–20.
- Cunningham D, Rao S, Starling N, Iveson T, Nicolson M, Coxon F *et al*. Randomised multicentre phase III study comparing capecitabine with fluorouracil and oxaliplatin with cisplatin in patients with advanced oesophagogastric cancer: The REAL 2 trial. Abstract LBA4017 (and associated on-line presentation). Available at: www.asco.org (accessed 16 October 2006).
- Kang YK, Kang WK, Shin DB, Chen J, Xiong J, Wang J *et al*. Randomised phase III trial of capecitabine/cisplatin (XP) vs continuous infusion of 5-FU/cisplatin (FP) as first-line therapy in patients (pts) with advanced gastric cancer (AGC): efficacy and safety results. Abstract LBA4018 (and associated on-line presentation). Available at: www.asco.org (accessed 16 October 2006).
- Twelves C, Wong A, Nowacki MP, Abt M, Burris III H, Carrato A *et al*. Capecitabine as adjuvant treatment for stage III colon cancer. *New England Journal of Medicine* 2005;352:2696–704.
- Sharpe M, Easthope SE, Keating G, Lamb HM. Polyethylene glycol-liposomal doxorubicin. *Drugs* 2002;62:2089–126.
- Kanter PM, Klaich G, Bullard GA, King GA, Pavelic ZP. Preclinical toxicology of liposome encapsulated doxorubicin (TLC D-99): comparison with doxorubicin and empty liposomes in mice and dogs. *In Vivo* 1993;7:85–95.
- Druker BJ, Guilhot F, O'Brien S, Larson RA. Long-term benefits of imatinib (IM) with chronic myelogenous leukaemia 5-year update from the IRIS study. *Journal of Clinical Oncology* 2006;24 (18S): 338s (Abstract 6506).
- Smith I. Adjuvant treatment for early breast cancer. *Annals of Oncology* 2005;16 (Supplement 2):ii182–ii187.
- National Institute for Health and Clinical Excellence. Breast cancer (early) — hormonal treatments — final appraisal determination. Available at: www.nice.org.uk (accessed 26 September 2006).
- Slamon DJ, Clark GM, Wong SG, Levine 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.
- Carter P, Presta L, Gorman CM, Ridgway JBB, Henner D, Wong WLT *et al*. Humanisation of an anti-p185HER-2 antibody for human cancer therapy. *Proceeding of the National Academy of Sciences USA* 1992;89:4285–9.
- Cobleigh MA, Vogel CL, Tripathy D, Robert NJ, Scholl S, Fehrenbacher L *et al*. Multinational study of the efficacy and safety of humanized anti-HER-2 monoclonal antibody in women who have HER-2-overexpressing breast cancer that has progressed after chemotherapy for metastatic disease. *Journal of Clinical Oncology* 1999;17:2639–48.
- Slamon DJ, Leyland-Jones B, Shak S, Paton V, Bajamonde A, Fleming T *et al*. Use of chemotherapy plus a monoclonal antibody against HER-2 for metastatic breast cancer that overexpresses HER-2. *New England Journal of Medicine* 2001;344:783–92.
- Piccant-Gebhart MJ, Procter M, Leyland-Jones B, Goldhirsch A, Untch M, Smith I *et al*. Trastuzumab after adjuvant chemotherapy in HER-2-positive breast cancer. *New England Journal of Medicine* 2005;353:1659–72.
- Romond EH, Perez EA, Bryant J, Suman VJ, Geyer CE, Davidson NE *et al*. Trastuzumab plus adjuvant chemotherapy for operable HER-2-positive breast cancer. *New England Journal of Medicine* 2005;353:1673–84.
- Slamon D, Eiermann W, Pienkowski RN, Martin M, Pawlicki M, Chan M *et al*. Phase III randomized trial comparing doxorubicin and cyclophosphamide followed by docetaxel (AC > T) with doxorubicin and cyclophosphamide followed by docetaxel and trastuzumab (AC > TH) with docetaxel, carboplatin and trastuzumab (TCH) in HER-2 positive early breast cancer patients: BCIRG 006 study. Available at <http://www.abstracts2view.com> (accessed 26th September 2006).
- Hampton P. Monoclonal antibody therapies shine in breast cancer clinical trials. *Journal of the American Medical Association* 2005;293:2895–989.
- National Institute for Health and Clinical Excellence. Technology Appraisal Guidance 107: Trastuzumab for the adjuvant treatment of early stage HER2-positive breast cancer. Available at: www.nice.org.uk (accessed 26 September 2006).
- Bonner JA, Harari PM, Giralt J, Azarnia N, Shin DM, Cohen RB *et al*. Radiotherapy plus cetuximab for squamous-cell carcinoma of the head and neck. *New England Journal of Medicine* 2006;354:567–78.
- Shepherd FA, Pereira JR, Ciuleanu T, Tan EH, Hirsh V, Thongprasert S *et al*. Erlotinib in previously treated non-small-cell lung cancer. *New England Journal of Medicine* 2005;353:123–132.
- Bezjak A, Tu D, Seymour L, Clark G, Trajkovic A, Zukin M *et al*. Symptom improvement in lung cancer patients treated with erlotinib: Quality of Life analysis of the National Cancer Institute of Canada Clinical Trials Group Study BR21. *Journal of Clinical Oncology* 2006;24:3831–7.
- Perez-Soler R, Delord JP, Halpern A, Kelly K, Krueger J, Sureda BM *et al*. HER1/EGFR inhibitor-associated rash: future directions for management and investigation. *Outcomes from the HER1/EGFR inhibitor rash management forum*. *The Oncologist* 2005;10:345–56.
- Presta LG, Chen H, O'Connor SJ, Chisholm V, Meng YG, Krummen L *et al*. Humanisation of an anti-VEGF monoclonal antibody for the therapy of solid tumors and other disorders. *Cancer Research* 1997;57:4593–9.
- Hurwitz H, Fehrenbacher L, Novotny W, Cartwright T, Hainsworth J, Heim W *et al*. Bevacizumab plus irinotecan, fluorouracil and leucovorin for metastatic colorectal cancer. *New England Journal of Medicine* 2004;350:2335–42.
- Sandler AB, Gray R, Brahmer J, Dowlati A, Schiller JH, Perry MC *et al*. Randomized phase II/III trial of paclitaxel (P) plus carboplatin (C) with or without bevacizumab (NSC 704865) in patients with advanced non-squamous non-small cell lung cancer (NSCLC): An Eastern Cooperative Oncology Group (ECOG) Trial – E4599. *Journal of Clinical Oncology* 2005;23(16S; June 1 supplement): Abstract 4.
- Escudier B, Szczylik C, Eisen T, Stadler WM, Schwartz B, Shan M *et al*. Randomized phase III trial of the Raf kinase and VEGFR inhibitor sorafenib (BAY 43-9006) in patients with advanced renal cell carcinoma (RCC). *Journal of Clinical Oncology* 2005; 23 (16S; June 1 supplement): Abstract 4510.
- De Mulder PH, Roigas J, Gillessen S, Srinivas S, Pisa P, Vogelzang M *et al*. A phase II study of sunitinib administered in a continuous daily regimen in patients with cytokine refractory metastatic renal cell carcinoma (mRCC). *Journal of Clinical Oncology* 2006;24 (18S; June 20 suppl.): Abstract 4529.
- Casali PG, Garrett CR, Blackstein ME *et al*. Updated results from a phase III trial of sunitinib in GIST patients (pts.) for whom imatinib (IM) therapy has failed due to resistance or intolerance. *Proceedings of the American Society of Clinical Oncology Annual Meeting*. 2006; 24 (16S; June 20 suppl.): Abstract 9513.