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Target Molecules for Immunotherapy of Inflammatory Breast Carcinomas

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INFLAMMATORY BREAST carcinoma (stage IIId) represents a special type of locally advanced breast cancer defined by a triad of rapid-onset clinical signs, including erythema and oedema of the skin, ridging and by the lack of specific histological findings. Usually, a mass is not palpated. Current treatment consists of primary chemotherapy followed by mastectomy (when feasible) and radiation therapy. Such combined modality treatments have led to a large improvement in the long-term survival rate of these previously incurable patients.

Nonetheless, inflammatory breast cancer remains a high-risk disease, and in recent series using combined modality treatments, the 5-year disease-free or relapse-free survival rates were still only in the range of 30–50% [1]. Clearly, more effective approaches and/or newer drugs are warranted.

One promising approach is the use of cytotoxic monoclonal antibodies directed against tumour-associated surface molecules. In fact, the strong inflammatory reaction characteristic of this tumour is expected to favour the uptake of large macromolecules, such as antibodies, whose delivery at the tumour site is often impaired by poor blood flow and poor extravasion in the tumour.

In a systematic search for tumour surface molecules that might act as targets for monoclonal antibodies, inflammatory breast carcinomas of 54 patients treated at the Milan Cancer Institute from 1977 to 1995 as part of approved institutional protocols were investigated by immunohistochemistry for the expression of different cell surface markers.

Mucins and carcino-embryonic antigen, two widely used markers for breast carcinomas, were found to be heterogeneously expressed in 20% of cases, whereas the HER2/neu gene product was highly overexpressed in 34 of 54 cases (63%) (Table 1). This frequency significantly exceeds ($P < 0.01$) that of 22% recently reported on a series of 717 breast carcinomas [2]. Thus, inflammatory carcinomas appear to be especially good targets for passive immunotherapy with monoclonal antibodies directed against the extracellular domain of the HER2/neu oncoprotein [3].

Table 1. Frequency of c-erB-2 overexpression and p53 positivity in 54 inflammatory breast carcinomas

Markers	No. cases/total (%)
$p185^{HER2}$-positive	34/54 (63)
p53-positive	19/54 (35)
$p185^{HER2}$-positive and p53-positive	15/54 (28)
$p185^{HER2}$-positive and p53-negative	19/54 (35)

The anti-HER2/neu antibodies selected for immunotherapy have been shown to inhibit growth in preclinical models [4] and to induce differentiation of tumour cells *in vitro* by up-modulating a functional p53 [5]. Consequently, among patients carrying HER2/neu-overexpressing tumours, those with a wild-type p53 are suitable candidates for therapy with the antibody *per se*. In our series, p53 overexpression was detected by immunohistochemistry with DO7 antibody [6] in 19 of 54 tumours tested (35%) (Table 1). This frequency is significantly higher ($P < 0.01$) than the 18% reported for the series of 717 breast carcinomas [2]. Furthermore, among the 34 HER2/neu-overexpressing carcinomas, no p53 overexpression was observed in 19 cases (55%) and p53 overexpression was detected in the remaining 15 cases (45%).

In conclusion, 63% of patients with inflammatory carcinomas in our series are appropriate candidates for anti-$p185^{HER2}$ monoclonal antibody therapy. Of these, 55% (those with a wild-type protein) might be treated with anti-$p185^{HER2}$ monoclonal antibodies *per se* in order to block cell proliferation and induce differentiation. This kind of treatment is particularly appropriate since these antibodies induce no systemic toxicity, allowing their administration alone or in association with other treatments. However, due to the frequency in breast carcinomas of p53 mutations abolishing the protein transcription (null mutation), which, according to the literature [7, 8] and our own experience, accounts for 30%, the number of patients that may benefit from anti-HER2/neu immunotherapy might be slightly lower. The other 45% of patients with tumours positive for $p185^{HER2}$ and p53 overexpression, as well as those with p53 mutation should benefit from a treatment with the same antibodies rendered cytotoxic by linkage to p53-independent drugs, radioisotopes or toxins to bypass the p53 blockage.

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1. Hortobagyi GN, Buzdar AU. Locally advanced breast cancer: a review including the M.D. Anderson experience. In Ragaz J, Ariel IM, eds. *High-risk Breast Cancer*. Berlin, Springer, 1991, 382–415.
2. Ménard S, Casalini P, Pilotti S, Cascinelli N, Rilke F, Colnaghi MI. No additive impact on patient survival of the double alteration of p53 and c-erbB-2 in breast carcinomas. *J Natl Cancer Inst* 1996, **88**, 1002–1003.
3. Baselga J, Tripathy D, Mendelsohn J, *et al.* Phase II study of weekly intravenous recombinant humanized anti-$p185^{HER2}$ monoclonal antibody in patients with HER2/neu-overexpressing metastatic breast cancer. *J Clin Oncol* 1996, **14**, 737–744.
4. Ohnishi Y, Nakamura H, Yoshimura M, *et al.* Prolonged survival of mice with human gastric cancer treated with an anti-c-ErbB-2 monoclonal antibody. *Br J Cancer* 1995, **71**, 969–973.
5. Bacus SS, Yarden Y, Oren M, *et al.* Neu differentiation factor (Heregulin) activates a p53-dependent pathway in cancer cells. *Oncogene* 1996, **12**, 2535–2547.
6. Righetti SC, Della Torre G, Pilotti S, *et al.* A comparative study of *p53* gene mutations, protein accumulation, and response to

cisplatin-based chemotherapy in advanced ovarian carcinoma. *Cancer Res* 1996, **56**, 689–693.
7. Aas T, Borresen A-L, Geisler S, *et al.* Specific *P53* mutations are associated with de novo resistance to doxorubicin in breast cancer patients. *Nature Med* 1996, **2**, 811–814.
8. Sjögren S, Inganäs M, Norberg T, *et al.* The p53 gene in breast cancer: prognostic value of complementary DNA sequencing versus immunohistochemistry. *J Natl Cancer Inst* 1996, **88**, 173–182.

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5-Fluorouracil-induced Raynaud's Phenomenon

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THERE ARE increasing numbers of reports describing acute vascular toxicity following the administration of cytotoxic chemotherapy. 5-Fluorouracil (5-FU) is a widely used cytotoxic agent. Although relatively unknown, 5-FU-induced coronary artery vasospasm is well described in the literature [1–3]. Nevertheless, perhaps surprisingly, reports of digital ischaemia and associated Raynaud's phenomenon have been described very rarely in relation to 5-FU, and usually in the context of it being used in combination with other drugs [4, 5]. We report a case of a man who developed significant digital ischaemia and Raynaud's phenomenon after receiving 5-FU/leucovorin (LV) based chemotherapy.

A 58-year-old caucasian man underwent antero-posterior resection for a Dukes' B rectal carcinoma in March 1991. He was a non-smoker and the only other history of note at presentation was mild hypertension for which he was prescribed atenolol (50 mg/day) a year earlier by his General Practitioner. In May 1995, he presented with hepatomegaly. Multiple liver metastases were confirmed on computerised tomography (CT) scanning and he was administered 5-FU based chemotherapy. The treatment comprised LV 200 mg/m^2 intravenous infusion (i.v.) over 2 h, then 5-FU 400 mg/m^2 i.v. over 5 min, followed by 400 mg/m^2 i.v. over 22 h on day 1, all repeated on day 2, of a 2 week cycle. Following six cycles, he achieved an objective radiological response, but by cycle 7 he developed severe Raynaud's phenomenon with digital ischaemia. The symptoms worsened with each subsequent cycle and the treatment was discontinued after the 9th cycle. At the same time atenolol was stopped. Examination revealed a digital infarct on the tip of the middle finger of the left hand; nailfold microscopy showed abnormal capillaries. Thermography confirmed severely reduced flow in that finger. An electrocardiogram and echocardiography performed at the time were reported as normal. Antinuclear antibody (ANA) was 1:80 speckled, but other serology was negative. The erythrocyte sedimentation rate (ESR) was raised at 57 mm/1st hour. On subsequent follow-up, the digital infarct had healed completely and he remained asymptomatic. Twelve months later, ANA at 1:80 persisted with a speckled pattern. The patient died from further disease progression in February 1997. The mechanism leading to acute ischaemia in this case is uncertain, although direct vascular toxicity, alteration of platelet activity or the induction of a hypercoagulable state following the administration of 5-FU are possible explanations [6–8]. 5-FU, either alone or in combination with other antineoplastic agents, has been associated with acute vascular events, in particular coronary artery spasm [1–3, 9]. Our patient, who was relatively young, had no signs suggestive of diffuse atherosclerosis and no significant cardiac history; he had been taking a selective β-blocker for 6 years prior to developing Raynaud's phenomenon. Despite the association between β-blockers and the development of Raynaud's, we feel that there was a clear temporal relationship from the time of administration of 5-FU and the onset of symptoms. Stopping the 5-FU provided adequate relief of symptoms. Increased awareness of this 5-FU-related side-effect should help minimise exposure to the drug in susceptible individuals.

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1. Pottage A, Holt S, Ludgate S, *et al.* Fluorouracil cardiotoxicity. *Br Med J* 1978, **6112**, 547.
2. Labianca R, Beretta G, Clerici M, *et al.* Cardiac toxicity of 5-fluorouracil: a study on 1083 patients. *Tumori* 1982, **68**, 505–510.
3. Freeman NJ, Costanza ME. 5-Fluorouracil-associated cardiotoxicity. *Cancer* 1988, **61**, 36–45.
4. Werquin S, Kacet S, Caron J, *et al.* Raynaud's phenomenon and finger necrosis after treatment of ovarian seminoma with bleomycin, vinblastine and 5-fluorouracil. *Ann Cardiol Angeiol* 1987, **36**, 409–412.
5. Bleiberg H, Conroy T, Paillot B, *et al.* Randomised phase II study of cisplatin and 5-fluorouracil (5-FU) versus cisplatin alone in advanced squamous cell oesophageal cancer. *Eur J Cancer* 1997, **33**, 1216–1220.
6. Mosseri M, Fingert HJ, Varticovski L, *et al.* In vitro evidence that myocardial ischaemia resulting from 5FU chemotherapy is due to protein kinase C mediated vasoconstriction of vascular smooth muscle. *Cancer Res* 1993, **53**, 3028–3033.
7. Cwikiel M, Persson SU, Larsson H, *et al.* Changes of blood viscosity in patients treated with 5-fluorouracil—a link to cardiotoxicity? *Acta Oncol* 1995, **34**, 83–85.
8. Kuzel T, Esparaz B, Green D, *et al.* Thrombogenicity of intravenous 5-fluorouracil alone or in combination with cisplatin. *Cancer* 1990, **65**, 885–889.
9. Esklisson J, Albertsson M, Mercke C, *et al.* Adverse cardiac effects during induction chemotherapy treatment with cisplatin and 5-fluorouracil. *Radiother Oncol* 1988, **13**, 41–46.