Breast & Colon Cancer: Molecular Alterations & Therapeutic Targets
Part 1

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FMI

June 1, 2007
Molecular targets for cancer therapy

- In the past 25 yrs cancer research has generated a large body of knowledge on molecular mechanisms controlling all aspects of normal cellular physiology/signaling pathways.

- These studies have also provided knowledge on molecules & mechanisms underlying cancer development.

- **Goal** - to design rational/molecularly targeted therapeutics for cancer treatment.

Major goal of lectures

- Present studies from the area of breast & colon cancer to show how molecular analyses can be used to achieve the ultimate goal of providing better therapies for patients.
Molecular targets for cancer therapy

- **Breast cancer**
  1 in 8 women develop the cancer
  30% will die of metastatic disease
  - incidence has been decreasing
  - therapeutics targeting proteins involved in breast cancer are in clinical use

- **Colon cancer**
  3rd most common cancer
  - therapeutics targeting proteins involved in colon cancer are in preclinical testing; drugs targeting angiogenesis have been approved

P. Ravdin et al 2007 NEJM 356:1670
The original description of pathological stages of colorectal cancer as defined by Duke in 1932

Box 1 | Relative survival for colorectal cancer by modified Dukes’ stage

The survival of patients with colorectal cancer decreases markedly with disease progression (see figure; data from REF 20).

The original description of staging for rectal cancer was described by Cuthbert Dukes in 1932 (REF 89), and included stages A–C, as outlined below. This staging system was modified in 1973 to include a description for metastatic disease (stage D) 90.

- Dukes’ A: tumour confined to the bowel wall with no lymph-node metastases.
- Dukes’ B: tumour spread confined to the extrarectal or extracolonic tissues by direct continuity, but with no lymph-node metastases.
- Dukes’ C: lymph-node metastases.
- Dukes’ D: distant metastases.

The colorectal adenoma-carcinoma sequence

- A well defined molecular/pathological sequence of colorectal cancer development has emerged over the past 15 yrs based on the pioneering work of Vogelstein & colleagues.

Gene Expression Patterns & Breast Cancer

- A well defined molecular/pathological sequence of breast cancer is not available.

- Gene expression profiling is a broad method for molecularly defining tumor types.

- Tumor sub-types with distinct gene expression patterns have been described.
  - prognosis
  - diagnosis
  - new therapeutic targets
Gene expression profiling & breast cancer

Breast cancers fall into 5 clinically-relevant subtypes*

* cDNA microarrays on a core set of 8100 genes were carried out, then **427 unique genes** formed a basis for classification based upon significantly greater variation in expression between different tumors than between paired samples from same tumor.

T. Sorlie et al PNAS 2001 98:10869; ibid 2003 100:8413
Gene expression profiling & breast cancer

- Breast cancers fall into 5 clinically-relevant subtypes:
  - Basal
  - ErbB2+
  - Normal
  - Luminal B
  - Luminal A

Luminal have best prognosis
Basal have worst prognosis

Overall survival
Disease-free survival
What influences Breast Cancer development?

- Hormones
- Genetic & Epigenetic Alterations
Many of these factors are related to estrogen exposure.
Estrogen & Breast Cancer

- Estrogens play an essential role in development of the normal breast & in cancer.
- The life time exposure to estrogens has a strong influence on cancer development.
  
  Loss of ovarian function prevents breast cancer

  Early onset of menarche & late menopause

  Low incidence of male breast cancer

  Early full-term pregnancy reduces risk of breast cancer (post-menopausal ER+ type).
Estrogen & Breast Cancer - Timeline

1896
George Beatson described that removal of the ovaries from women with metastatic breast cancer sometimes led to tumor regression.

"On the treatment of inoperable cases of carcinoma of the mamma: suggestions for a new method of treatment with illustrative cases". Lancet (1896)

1962
Jensen & Jacobsen identified the target for estrogen action, the estrogen receptor (ER).

"Basic guides to the mechanism of estrogen action"
Recent Prog Horm Res (1962)

1971
Jensen correlated the presence of ER with hormone responsiveness of the breast tumor.

"Estrogen receptors & breast cancer response to adrenalectomy"
Natl Cancer Inst Monogr, 1971
The estrogen receptor as target for therapy

- ER$^+$ tumors depend upon estrogen for proliferation.

- In 1971 Jensen correlated the presence of ER with hormone responsiveness of the breast tumor.

- ER antagonists were some of the first targeted, rational therapeutics. (proposed in 1936 by Lacassagne)

Many primary breast tumors are ER$^+$ and depend upon ER action for growth.
New perspectives into the biological and clinical relevance of oestrogen receptors in the human breast

V Speirs\(^1\) and RA Walker\(^2\*)

\(^1\)Leeds Institute of Molecular Medicine, Welcome Trust Bremner Building, St James’s University Hospital, Leeds LS9 7TF, UK

\(^2\)Department of Cancer Studies and Molecular Medicine, Robert Kilpatrick Building, Leicester Royal Infirmary, PO Box 65, Leicester LE2 7LX, UK
The estrogen receptor as a therapeutic target in breast cancer

The non-steroidal, anti-estrogen Tamoxifen has been used since 1973 for breast cancer therapy.

- Unfortunately after a few years most breast cancer patients relapse & show metastases during treatment with tamoxifen.

- Why?
  Has proliferation become independent of ER action?
In postmenopausal women, adrenal & ovarian androgens are converted into estrogens, in different peripheral tissues: muscle, skin, normal breast & in breast cancer cells, by the enzyme **aromatase**.

**Aromatase inhibitors & breast cancer**
The non-steroidal, anti-estrogen Tamoxifen has been used since 1973 for breast cancer therapy.

Aromatase inhibitors were recently approved for clinical use.

Tamoxifen blocks ER transcriptional activity.

Aromatase inhibitors lower the level of estrogens in the skin, adipose tissue & the tumor; this lowers ER activity & blocks cancer cell proliferation.

The estrogen receptor as a therapeutic target in breast cancer
Aromatase inhibitors & breast cancer

- Some have shown efficacy in patients who relapsed on tamoxifen therapy.

- These results suggest that tumors still require estrogen/ER for proliferation; lowering the level of estrogen in the tumor & adipose tissue has an impact on disease.

- These results are very important for breast cancer patients:
  1. after relapse on tamoxifen there is another therapeutic option with proven efficacy
  2. anti-estrogen therapy is relatively non-toxic
Ultimately most ER+ breast cancer patients that respond to anti-estrogen based therapies (tamoxifen or AI) relapse on treatment.

Why does resistance emerge?
More later...
What influences breast cancer development?

- **Hormones**
  - loss of tumor suppressor genes
  - DNA or transcriptional level
  - activation of dominant oncogenes: mutation, amplification, transcriptional

- **Genetic & Epigenetic**

Specific Molecular Alterations & Therapies

- Development of targeted therapeutics based upon characterizing specific molecular alterations in tumors. e.g., ErbB2 gene amplification
**Tumor suppressor genes & breast cancer**

- **BRCA 1 & BRCA2**

Existence of genes responsible for inherited predisposition to breast & ovarian cancer was suggested more than 100 yrs ago.

Due to foresight of certain human geneticists, G Lenior & MC King, who collected material from “cancer families”, linkage analyses were used to determine chromosomal location of the genes, this was followed by their positional cloning.
What characterizes a family with hereditary breast cancer?

Features That Indicate Increased Likelihood of Having BRCA Mutations

- Multiple cases of early onset breast cancer
- Ovarian cancer (with family history of breast or ovarian cancer)
- Breast and ovarian cancer in the same woman
- Bilateral breast cancer
- Ashkenazi Jewish heritage
- Male breast cancer

10-15% of all breast & ovarian cancers are inherited. BRCA1 & BRCA2 are responsible for <50% of these.
What are the functions of BRCA1 & BRCA2?

The BRCA proteins have been implicated in many different processes including DNA repair & recombination, cell cycle control & transcription.

The BRCA proteins are important for the maintenance of genomic stability.
**BRCA proteins & DNA Repair**

Mouse cells deficient in BRCA2 show spontaneous aberrations in chromosome structure.

Aberrations in the usual U-shaped mouse chromosomes are visible. (enlarged on the right)

Venkitaraman 2002 Cell 108: 171
BRCA proteins & DNA Repair

- A cell undergoes 10-20 double strand breaks/day - in addition to other types of DNA damage.

Venkitaraman 2002 Cell 108: 171

- In S phase - HR is the preferred repair pathway
A cell undergoes 10-20 double strand breaks/day - in addition to other types of DNA damage.

The preferred error-free HR pathway does not function in cells lacking BRCA proteins.
**BRCA1 has multiple functions**

- BRCA1 has a broad role linked to a range of processes implicated in different cell cycle checkpoints.

**G2/M:**
DNA decatenation to separate entangled chromosomes

**S-phase:**
Repair of DS DNA breaks by homologous recombination

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A. Ashworth August 2005 Cancer Cell
BRCA1 has multiple functions

- BRCA1 has a broad role linked to a range of processes implicated in different cell cycle checkpoints.

BRCA2 has a more specific role in repair of DNA DS breaks.

G2/M:
DNA decatenation to separate entangled chromosomes

S-phase:
Repair of DS DNA breaks by homologous recombination

A. Ashworth August 2005 Cancer Cell
ATM & ATR kinases are activated in response to DNA DS breaks. ATM & ATR phosphorylate multiple proteins to transduce the DS break response signal. DNA repair is stimulated via NHEJ or HR. Cell cycle is stalled via CH2 & CHK1.
BRCA2, Rad51 & DNA repair

BRCA2 is required for the regulation of RAD51-mediated homologous recombinational repair.
**BRCA2, Rad51 & DNA repair**

Stabilization of RAD51 nucleoprotein filaments by the C-terminal region of BRCA2

Fumiko Esashi\(^1\), Vitold E Galkin\(^2\), Xiong Yu\(^2\), Edward H Egelman\(^2\) & Stephen C West\(^1\)

Rad51 monomers bind the BRC motifs

Rad51 filaments bind the C-T erminal Domain (CTD)

Ser3291-P blocks Rad51 association

**BRCA2, Rad51 & DNA repair**

Stabilization of RAD51 nucleoprotein filaments by the C-terminal region of BRCA2

Fumiko Esashi¹, Vitold E Galkin², Xiong Yu², Edward H Egelman² & Stephen C West¹

Rad51 monomers bind the BRC motifs

Rad51 filaments bind the C-terminal domain (CTD)

CTD association of Rad51 filaments facilitates efficient nucleaction of RAD51 multimers on DNA - HR is stimulated

Based on the knowledge of the roles of BRCA proteins in DNA repair the next goal was to design inhibitors that would specifically kill tumor cells with BRCA mutations - and spare normal cells.

**Targeting BRCA tumors**

- Synthetic lethality: A gene (pathway) that has no phenotype when deleted (inhibited) alone is made essential by loss of a redundant partner (pathway).

R. Kennedy & A. D'Andrea 2006 JCO 24:3799
Specific DNA lesions or lesions that block or cause the collapse of replication forks are very dependent on HR-based DNA repair.

SSBs are repaired by base excision repair pathway; inhibition of this pathway increases the number of unrepaired SSBs leading to collapse of replication forks & increase in DSBs at replication forks.
	poly(ADP-ribose) polymerase (PARP1) is critical for base excision repair.
**Targeting BRCA tumors**

- Based on the knowledge of the roles of BRCA proteins in DNA repair, the next goal was to design inhibitors that would specifically kill tumor cells with BRCA mutations - and spare normal cells.

**Targeting the DNA repair defect in BRCA mutant cells as a therapeutic strategy**

Hannah Farmer, Nuola McCabe, Christopher J. Lord, Andrew N. J. Tuttle, Damien A. Johnson, Tobias B. Richardson, Manuela Santarosa, Krystyna J. Dillon, Ian Hickson, Charlotte Knights, Niall M. B. Martin, Stephen P. Jackson, Graeme M. Smith & Alan Ashworth

1 Cancer Research UK Gene Function and Regulation Group and 2 The Breakthrough Breast Cancer Research Centre Institute of Cancer Research, Fulham Road, London SW3 6JB, UK
3 Guy’s Hospital, St Thomas’ Street, London SE1 9RT, UK
4 Kudos Pharmaceuticals Ltd, Cambridge Science Park, Cambridge CB4 0WG, UK
5 Wellcome Trust and Cancer Research UK, Garden Institute of Cancer and Developmental Biology and Department of Zoology, University of Cambridge, Tennis Court Road, Cambridge CB2 1QH, UK
6 Present address Division of Experimental Oncology, CBO-SGC, Antonio Nardini 2, Italy
7 The Institute for Cancer Studies, University of Sheffield, Medical School, Broom Hill Road, Sheffield S10 2RX, UK
8 Department of Genetics, Microbiology and Toxicology, Arthritis Laboratory, Stockholm University S-106 91 Stockholm, Sweden
9 Northern Institute for Cancer Research, University of Newcastle upon Tyne, Medical School, Newcastle upon Tyne, NE2 4HN, UK

- KU005864 PARP1 inhibitor - IC50 = 3.2 nM

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Nature 2005 vol 434, p913 & p917
Targeting BRCA tumors with a PARP inhibitor

PARP inhibition selectively blocks the in vivo growth of BRCA2-deficient tumors & does not affect growth of WT tumors.


In the absence of BRCA1 or BRCA2, PARP inhibition leads to persistent SS gaps in DNA that collapse into DS breaks at a replication fork. Repair of these breaks by error-prone DSB repair mechanisms causes many aberrations that eventually lead to cell death.
Various PARP inhibitors are in clinical development.
Gene expression profiling & breast cancer

- BRCA1 tumors have a basal cancer “expression signature”
  (Turner et al 2004 Nature Reviews Cancer vol 4)

- Would PARP inhibition also inhibit growth of tumors with a Basal phenotype? Are there sporadic cancers with BRCA1 mutations - in the basal-like group?

![Gene expression profiling diagram]
BRCA1 & BRCA2 have not been implicated in sporadic breast cancer

In contrast to other inherited tumor suppressor genes (APC in colon cancer), sporadic breast cancers do not carry mutations in BRCA genes.

Other mechanisms contribute to loss of BRCA expression or function in breast cancer.
Epigenetic mechanisms of BRCA1 inactivation

- BRCA1 transcription is silenced by promoter methylation in sporadic breast & ovarian cancers.

<table>
<thead>
<tr>
<th>Cancer type</th>
<th>Somatic BRCA mutations</th>
<th>BRCA1 methylation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Breast</td>
<td>Very rare(^7,8)</td>
<td>11–14(^{26,28})</td>
</tr>
<tr>
<td>Ovarian</td>
<td>&lt; 5(^{29,48,71})</td>
<td>5–31(^{26,28,30})</td>
</tr>
<tr>
<td>HNSCC</td>
<td>ND</td>
<td>0(^34)</td>
</tr>
<tr>
<td>NSCLC</td>
<td>ND</td>
<td>4(^34)</td>
</tr>
<tr>
<td>Cervical</td>
<td>ND</td>
<td>6.1(^35)</td>
</tr>
</tbody>
</table>

Turner et al 2004 Nature Reviews Cancer vol 4
Mutations in BRCA2 interacting proteins

PALB2 binds N-terminus of BRCA2 & its binding is essential for BRCA2 DSB DNA repair activity.

T. Walsh & M.C. King 2007 Cancer Cell 11:103
PALB2 mutations in familial & sporadic breast cancers

A recurrent mutation in PALB2 in Finnish cancer families

- Frameshift mutation found in 113 families with history of breast & ovarian cancer.
- Same mutation found in 18/1918 sporadic breast cancers.
**PALB2 mutations in familial & sporadic breast cancers**

*PALB2*, which encodes a BRCA2-interacting protein, is a breast cancer susceptibility gene

Identified PALB truncating mutations in 10/923 individuals with familial breast cancer (0/1084 controls).

Important for two reasons:

~50% of inherited breast cancer is due to unknown mutations.

Evidence that the BRCA pathway/complex is involved in sporadic breast/ovarian cancer.
Mutations in BRCA2 interacting proteins

Proteins in red carry germline mutations that predispose to breast cancer.

Figure 1. Interactions of Proteins Associated with Inherited Breast Cancer and with Fanconi Anemia

T. Walsh & M. King 2007 Cancer Cell 11:103
Proteins aberrantly expressed or activated in breast cancer

- ErbB2 & EGF receptor tyrosine kinases
- FGF receptor tyrosine kinases
- Cyclin D1
- c-myc
- ......
ErbB2 overexpression in primary breast cancer

- *ERBB2* gene amplification was the first consistent genetic alteration found in breast cancer (20-25%).

- Patients with high ErbB2 tend to have a poor clinical outcome.

Hynes et al 1989
Gene expression profiling & breast cancer

- ErbB2+ group has a bad prognosis

Overall survival  Disease-free survival

ErbB2+ Basal-like Luminal A
Normal Luminal B  Luminal A  ErbB2+  Basal
ERBB targeted inhibitors

ERBB targeted antibodies: Herceptin Cetuximab

ERBB targeted tyrosine kinase inhibitors (TKIs): Gefitinib Erlotinib

Extracellular Domain

Plasma Membrane

Cytoplasmic Domain

Herceptin was approved for treatment of metastatic ErbB2 overexpressing breast cancer patients in 1998.
**Herceptin®/Trastuzumab & Breast Cancer**

- Herceptin® does not cure metastatic breast cancer - at this stage tumor cells have spread to distant organs.

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**Median time to disease progression**

- 7.4 mos.
- 4.6 mos.

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**Progression-free Survival (%)**

- **Chemotherapy plus trastuzumab**
  - 15 patients-CR
- **Chemotherapy alone**
  - 6 patients-CR

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Slamon et al NEJM 2001 vol 344
Herceptin® & Breast Cancer

Once Herceptin® was approved for treatment of advanced metastatic breast cancer patients, it was tested on “early-stage” ErbB2+ breast cancer patients, in combination with chemotherapy.

Patients all had ErbB2 overexpression & most were lymph node+, i.e., the tumor had spread to LNs and possibly to distant organs.

Results from trials testing Herceptin® in the adjuvant setting* showed that its addition to chemotherapy reduced the risk of recurrence by 50% in women with early stage ErbB2+ cancer.


*adjuvant setting - directly after surgical removal of the tumor
Addition of trastuzumab reduced mortality rate by one third.

CONCLUSIONS
Trastuzumab combined with paclitaxel after doxorubicin and cyclophosphamide improves outcomes among women with surgically removed HER2-positive breast cancer. (clinicaltrials.gov numbers, NCT00004067 and NCT00005970.)

These results suggest that trastuzumab acts as a chemosensitizer when given together with chemotherapy directly after surgery.
Remission of human breast cancer xenografts on therapy with humanized monoclonal antibody to HER-2 receptor and DNA-reactive drugs

Richard J Pietras¹, Mark D Pegram¹, Richard S Finn¹, Daniel A Maneval² and Dennis J Slamon¹

¹Division of Hematology-Oncology, Department of Medicine, UCLA School of Medicine, Los Angeles, California 90095, and ²Genentech, Inc., 460 Point San Bruno Boulevard, South San Francisco, California 90078, USA

In vitro ErbB2⁺ model to test DNA repair in the absence or presence of trastuzumab.

Trastuzumab blocks the ability of ErbB2⁺ tumor cells to repair cis-platin induced DNA damage.

Why is this important?
**ErbB2^+ tumors & DNA repair**

- Blocking ErbB2 with Herceptin® (TKIs ?) may make the tumor cells very sensitive to chemotherapeutics.
ErbB2+ tumors & DNA repair

Drugs tested with Herceptin®

In clinical trials:
Doxorubicin (Topo2 inhibitor—blocks replication by intercalation)
Cyclophosphamide (alkylating agent)
Paclitaxel (MT stabilizer)

In vitro:
Cis-platin (DNA cross-linker, alkylating agent)

ErbB2+ Cancer Cell

Herceptin

Elevated ErbB2+

Signaling Pathway 1
Akt, ERK, mTOR etc

Signaling Pathway 2
??

Survival

Chemotherapeutic drugs

Chemotherapeutic drugs
ERBB Receptors and Breast Cancer: Clinical Problems to Solve

Understand mechanisms underlying activity of ERBB targeted inhibitors.

What mechanisms contribute to resistance? June 21st - Cellular Signaling

ERBB inhibitors in combination with other anti-cancer drugs. Best choice?

ERBB receptors & resistance to anti-ER targeted therapies.
Mechanisms contributing to Tamoxifen resistance

- ERBB2 expression levels increase in tumors at time of relapse on tamoxifen.

(Dowsett & colleagues - Gutierrez et al 2005 JCO 23:2469)
Mechanisms contributing to Tamoxifen resistance

- ERBB2 gene amplification observed in some patients who were ErbB2- at the start of Tamoxifen treatment.
  Dowsett & colleagues - Gutierrez et al 2005 JCO 23:2469

FISH + = ErbB2 gene amplification
Estrogen receptor has multiple P sites.

New perspectives into the biological and clinical relevance of oestrogen receptors in the human breast

V Speirs\(^1\) and RA Walker\(^2\)\(^*\)

\(^1\)Leeds Institute of Molecular Medicine, Welcome Trust Brenner Building, St James’s University Hospital, Leeds LS9 7TF, UK

\(^2\)Department of Cancer Studies and Molecular Medicine, Robert Kilpatrick Building, Leicester Royal Infirmary, PO Box 65, Leicester LE2 7LX, UK
Mechanisms contributing to Tamoxifen resistance

Estrogen receptor has multiple P sites.

When signaling pathway activity is elevated—e.g., in ErbB2 overexpressing tumors cells, these sites are P.

ER transcriptional activity is high—even in the absence of estrogens.

<table>
<thead>
<tr>
<th>ER - P sites</th>
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<tbody>
<tr>
<td>Ser 104 Cdk2</td>
</tr>
<tr>
<td>Ser 118 ERK 1/2, cdk7 &amp; others</td>
</tr>
<tr>
<td>Ser 167 p90 RSK &amp; AKT</td>
</tr>
<tr>
<td>Ser 115 MEKK1 (H Lee et al 2000 Mol Endo 14)</td>
</tr>
<tr>
<td>Thr 311 p38 MAPK (H Lee et al 2002 MCB 22)</td>
</tr>
<tr>
<td>Ser 305 PKA (R Michalides et al 2004 Cancer Cell)</td>
</tr>
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</table>

Elevated activity of other RTKs: IGFR-1 or FGFR also impact on ER activity & are likely to play a role in the development of tamoxifen resistance.
Anti-Tumor Approaches using Antibodies

W. Wels et al 1992
Cancer Res 52:6310-17.

scFv-toxin
Structure of Single Chain Antibody - scFv

Hybridoma cells producing FRP5, a murine mAb that binds the extracellular domain of human ErbB2, were used to isolate cDNAs encoding the specific heavy and light chain variable domains. I.-M. Harwerth et al. 1992 JBC 267:15160-67.
Targeting ErbB2 with specific scFv toxins

(W. Wels et al 1992 Cancer Res)

Bifunctional antibody fragments for directed cancer therapy

Exotoxin A from Pseudomonas aeruginosa

Tumor cell-specific antibody

Recombinant single chain antibody

Effector with novel binding properties
Chimeric scFv-Toxin Fusion Protein

Domain Structure of Pseudomonas Exotoxin A

scFv-ETA

- VH
- Linker
- VL

scFv

ETA 252-613
**scFv-Toxin Binds ErbB2 & Internalizes into Tumor Cells**

Toxin's Receptor

- α2-macroglobulin receptor

Cell Surface

- ErbB2

Cytosol

Internalization

- Internalized ErbB2 enters the endosomes

ErbB2

Endosome

- pH ↓

Processing

- Inhibition of Protein Synthesis

Trans-Golgi

- KDEL receptor

Golgi

Endoplasmic Reticulum

Translocation

W.Wels
**scFv-Toxin Specifically Kills ErbB2⁺ Cells**

Specificity and cytotoxic activity of scFv(FRP5)-ETA

<table>
<thead>
<tr>
<th></th>
<th>MDA-MB453</th>
<th>SKBR3</th>
<th>MDA-MB468</th>
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<td>control</td>
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<td><img src="image2" alt="Control" /></td>
<td><img src="image3" alt="Control" /></td>
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<tr>
<td>scFv(FRP5)-ETA 1 µg/ml</td>
<td><img src="image4" alt="Treatment" /></td>
<td><img src="image5" alt="Treatment" /></td>
<td><img src="image6" alt="Treatment" /></td>
</tr>
</tbody>
</table>

W. Wels

ErbB2⁺  ErbB2⁻
**ErbB2-Overexpressing Tumor Xenografts are Sensitive to FRP5-ETA**

**Treatment of established breast carcinoma with scFv(FRP5)-ETA**

MAXF1162 human breast carcinoma xenografts

Intratumoral injection of 8 μg scFv(FRP5)-ETA on days 25, 27, 29, 33, 35 p.i.

W. Wels

PBS
day 37 p.i.

scFv(FRP5)-ETA
day 37 p.i.
Recombinant scFv(FRP5)-ETA was produced as an experimental drug under GMP conditions & provided by Ciba Geigy AG, Basel.
Compassionate Use Clinical Trial

G2M Cancer Drugs AG, Frankfurt

Intratumoral application of scFv(FRP5)-ETA

Metastatic breast carcinoma
Total dose 1.8 mg

Malignant melanoma
Total dose 2.8 mg

(Azemar et al 2003 Breast Cancer Res Treatment)
Phase 1 dose-finding study

Goals:
- determination of MTD
- dose-limiting toxicity of IV injected scFv(FRP5)-ETA

Protocol:
- 5 daily infusions - 2 consecutive weeks
- 5 doses

Results:
N = 17
2 Stable disease
3 Clinical activity
11 Disease progression

von Minckwitz et al 2005 Breast Cancer Res 7:R617
Phase 1 dose-finding study

Major conclusions:

- scFv-ETA can be safely administered IV at doses up to 12.5 μg/ml per day
- potentially therapeutic serum concentrations were reached
- major dose-limiting side effect was hepatotoxicity

von Minckwitz et al 2005 Breast Cancer Res 7:R617

Novartis licensed scFv(FRP5)-ETA to G2M Cancer Drugs AG, Frankfurt
Licensed to TopoTarget A/S Denmark in 2005