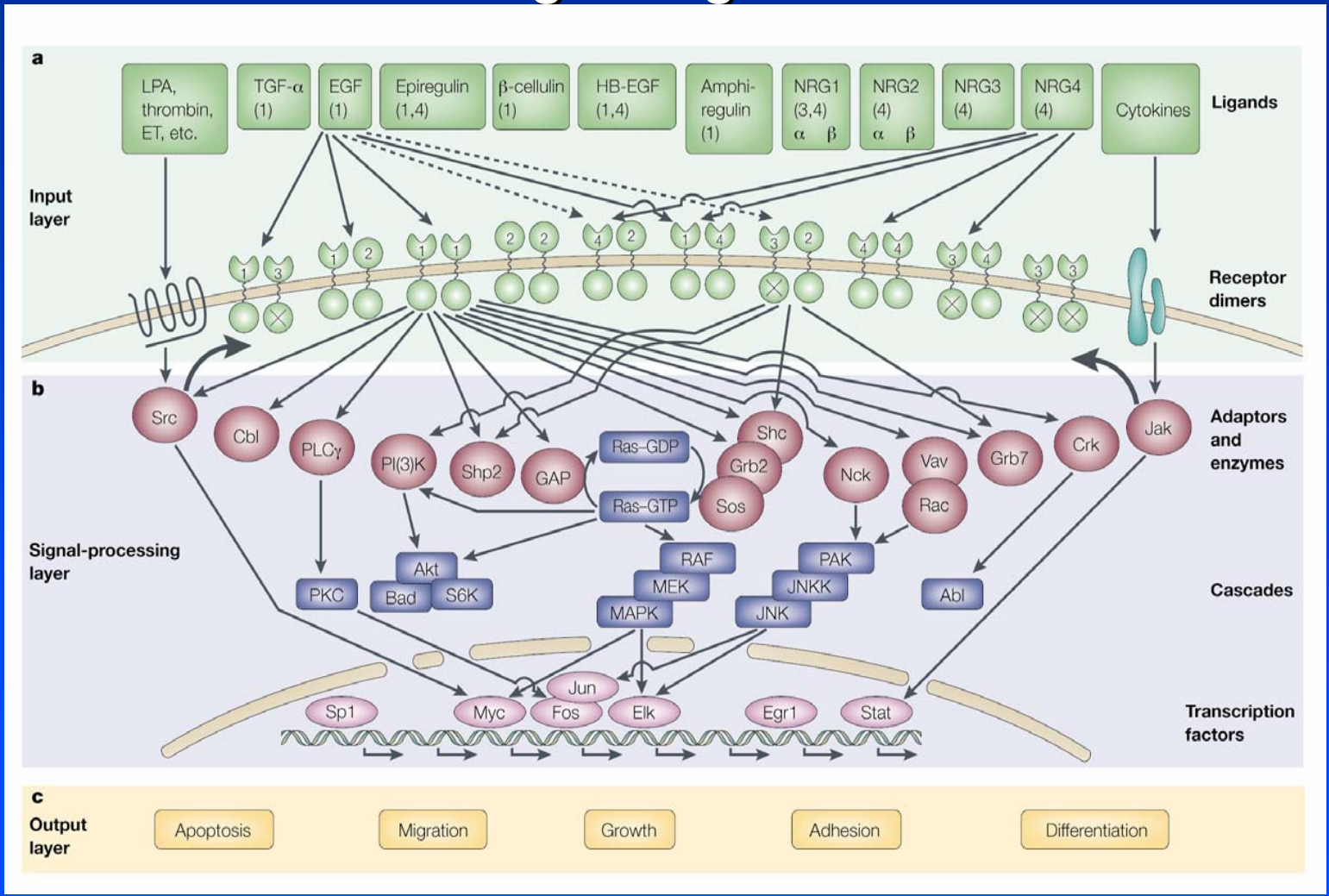
Monoclonal Antibodies in Cancer

Ralph Schwall, PhD
Associate Director, Translational Oncology
Genentech, Inc.

Disclaimer

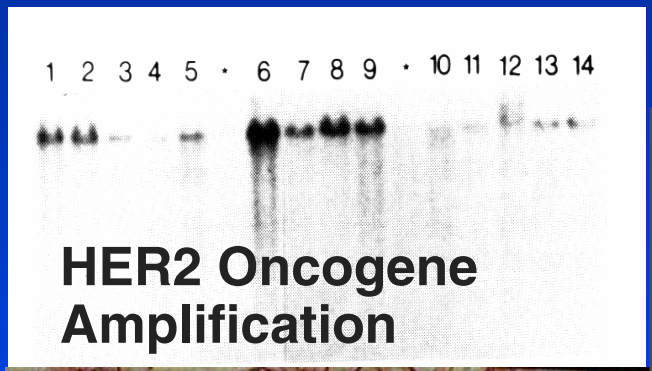
- I had nothing to do with Herceptin
- Using lessons learned in new antibody projects

The HER2/ErbB Signaling Network



Yarden Y, Sliwkowski MX. *Nat Rev Mol Cell Biol.* 2001;2:127-137.

Why Target Human EGF Receptor 2

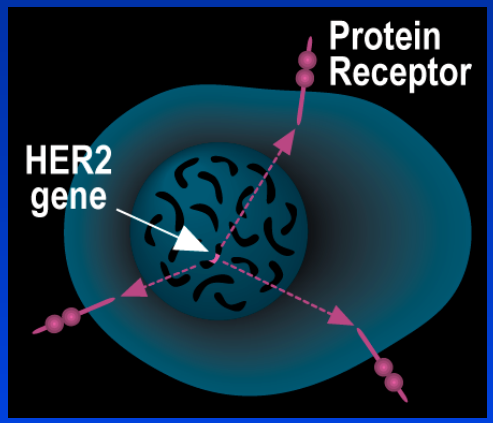


Shortened Median Survival

HER2 overexpressing	3 yrs
HER2 normal	6 - 7 yrs

Slamon et al, 1987

Herceptin

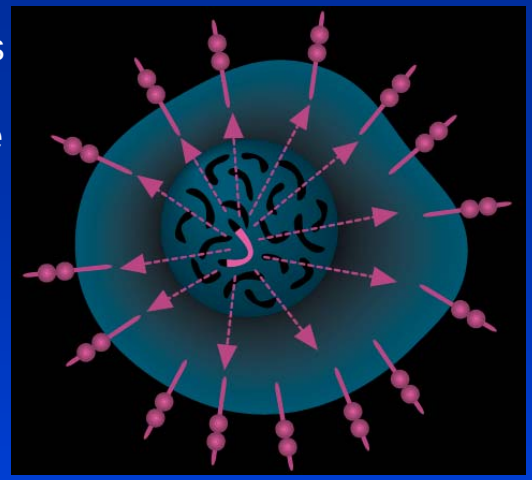


Normal Cell

In normal breast tissue cells, the HER2 gene produces a protein receptor on the cell surface. These growth factor-like receptors are thought to play a role in normal cell growth by signaling the cell to divide and multiply.

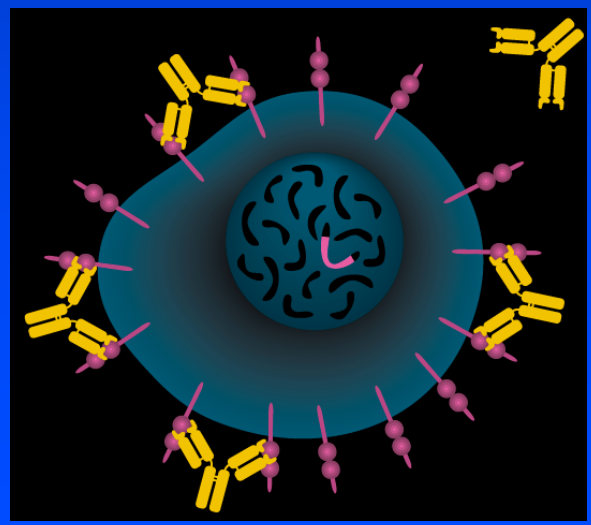
HER2 Overexpressing Cancer Cell

Cancerous breast tissue cells that overexpress (or overproduce) the HER2 gene produce extra protein receptors on the cell surface. which triggers the cell to divide and multiply at an accelerated rate, thus contributing to tumor growth.

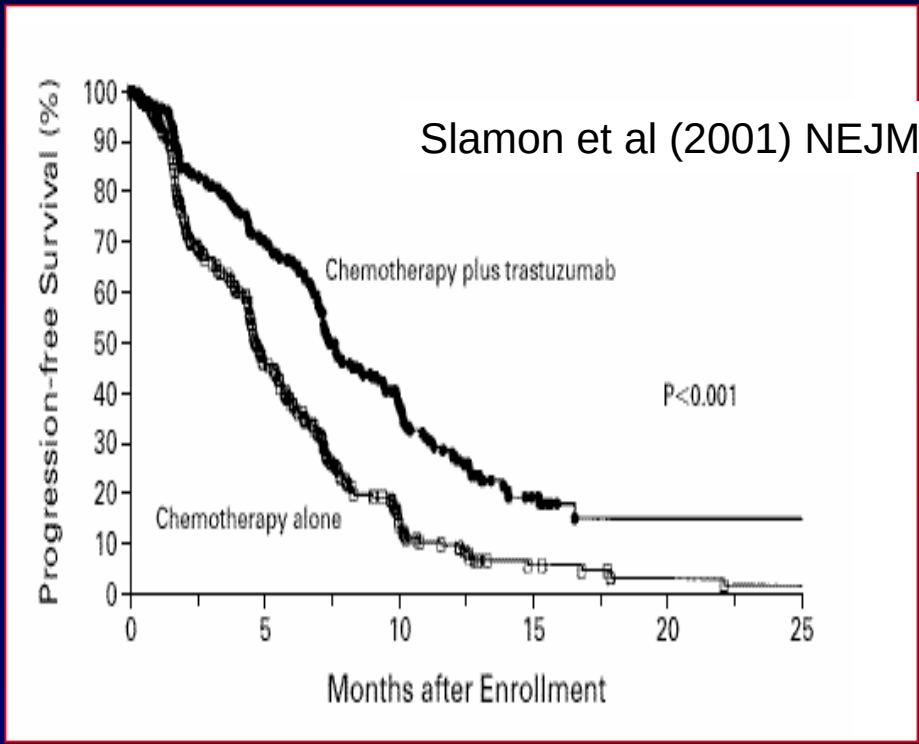


Herceptin® (Trastuzumab)

Herceptin (a HER2 antibody) binds to numerous HER2 receptor sites found on the cell surface, blocking the receptor sites and possibly preventing further growth by interrupting the growth signal. As a result, the HER2 antibody may slow progression of the disease.

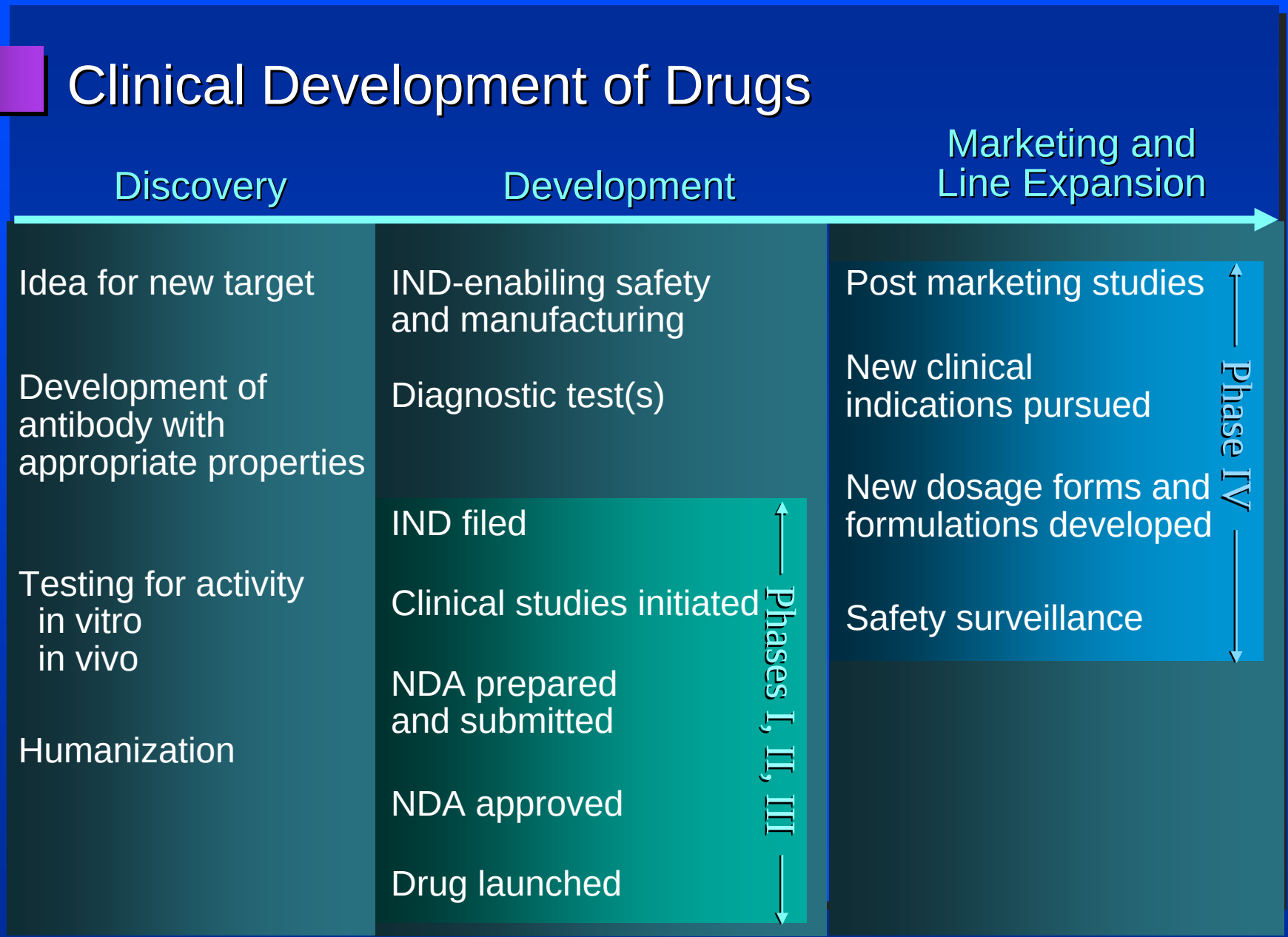


Herceptin™ confers survival benefit in metastatic breast cancer

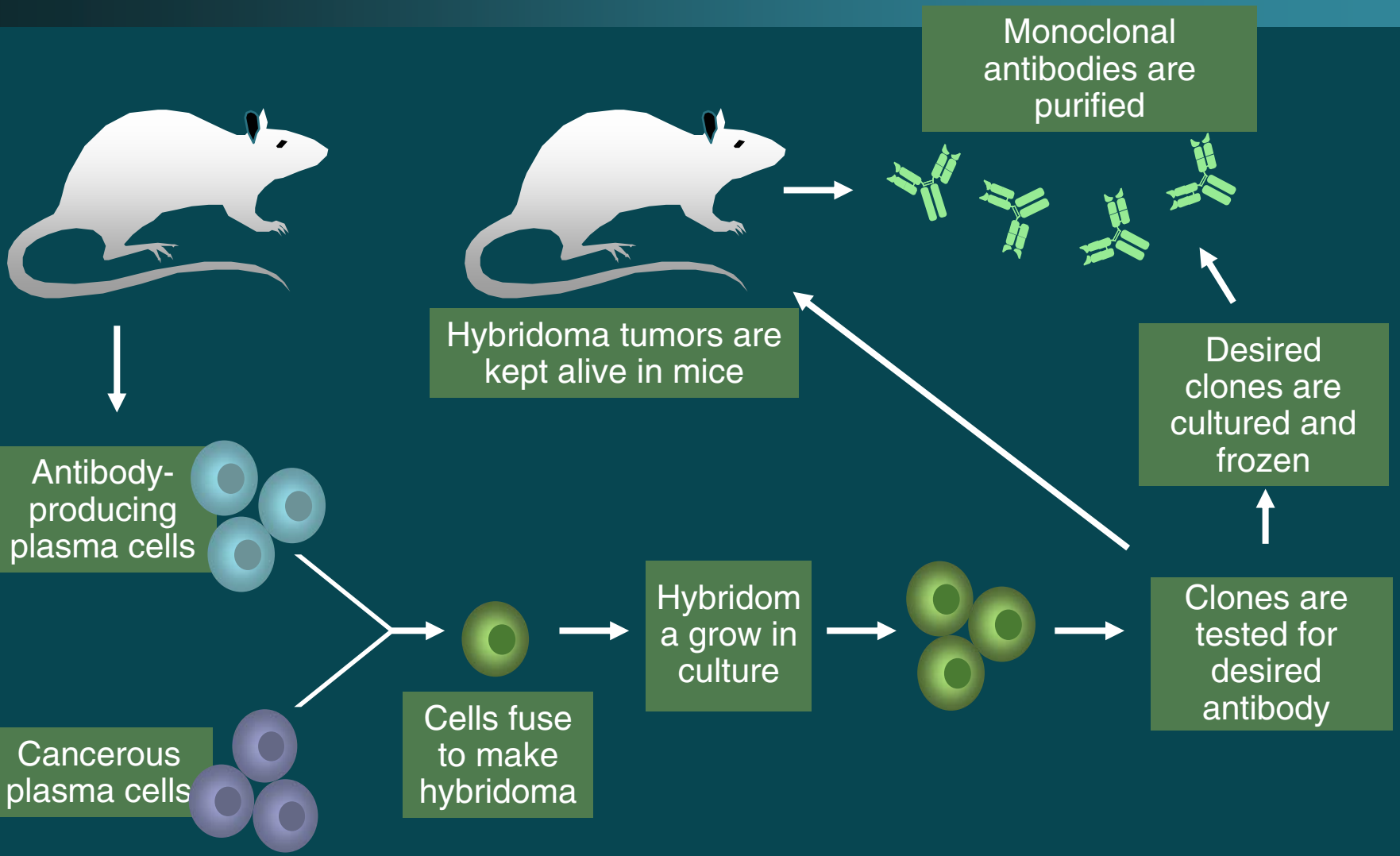


Slamon et al (2001) NEJM 344, 783-792

Clinical Development of Drugs



Production of Monoclonal Antibodies Hybridoma Technology



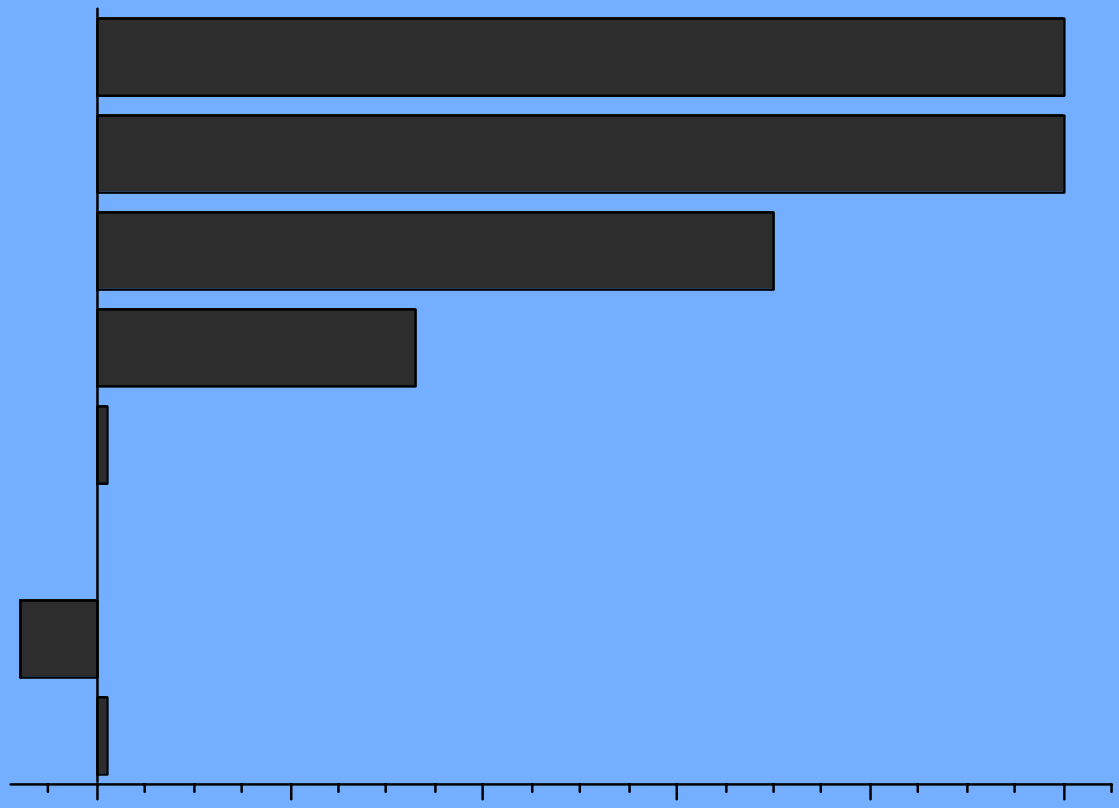
4D5 becomes Herceptin

- In vitro proliferation assays
- Screening of many antibodies for activity
- Murine hybridoma “4D5” was selected out of a panel of anti-HER2 antibodies because it selectively inhibits growth of breast cancer cells overexpress HER2 but not cells with normal levels of HER2.

4D5 only inhibits cells with high HER2 *(Gail Phillips)*

Cell line	Relative HER2 expression	Proliferation (% of Control)					
		4D5	3H4	2C4	7F3	7C2	6E9
HMEC	**1.0**	116	114	109	116	117	103
HBL-100	**1.0**	104	102	103	96	104	105
MCF7	**1.2**	101	113	100	111	112	105
MDA-MD-231	**1.2**	91	100	93	98	104	103
ZR-75-1	**3.3**	102	105	99	97	108	97
MDA-MB-436	**3.3**	97	91	98	93	92	101
MDA-MB-175	**4.5**	62	77	29	48	87	96
MDA-MB-453	**16.7**	61	65	88	80	70	101
MDA-MB-361	**16.7**	63	67	64	76	105	99
BT-474	**25.0**	27	29	60	21	78	91
SK-Br-3	**33.0**	33	40	73	51	82	89

Herceptin Inhibiton of Anchorage-Independent Growth Is Related to HER2 Expression Level



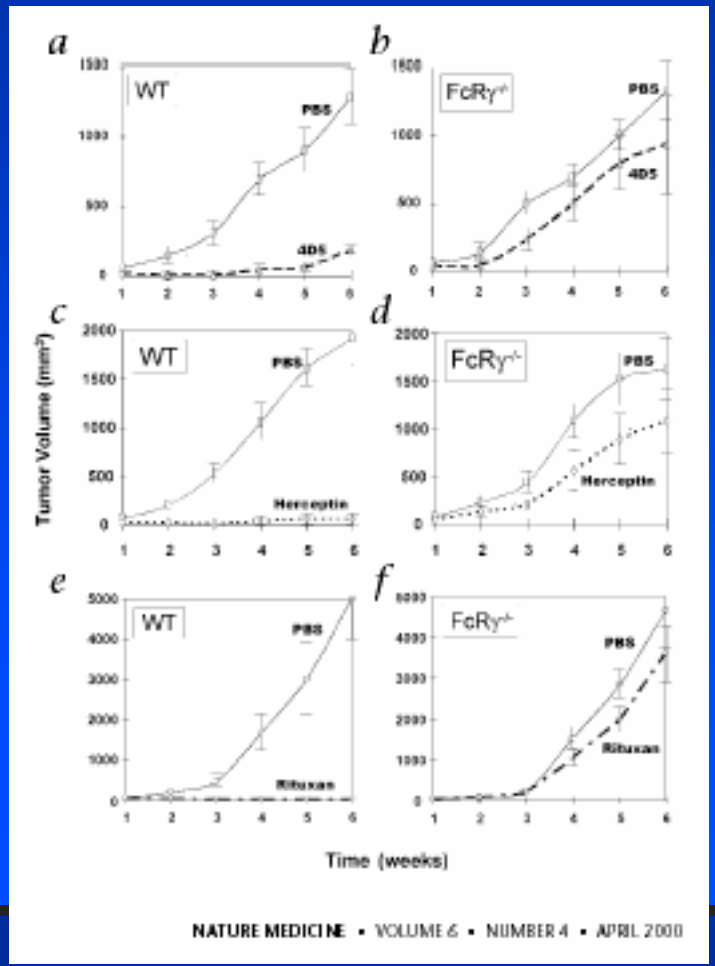
Gail Phillips

Testing Preclinical Efficacy In Vivo

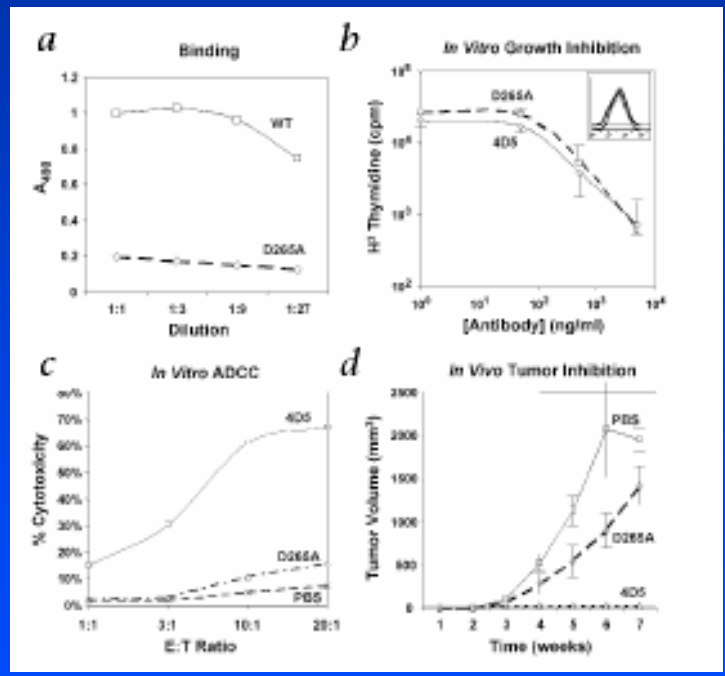
- Antibodies have potential to incorporate immune system effector functions for activity.
- Preclinical testing typically done in nude mouse xenografts
  - Need human cells as target
  - Need immunodeficient host to grow tumor
  - Minimal immune effector function

Antibody-Dependent Cellular Cytotoxicity in Herceptin Mechanism

FcRγ$^{-/-}$ nude mice



D265A-Herceptin Mutant

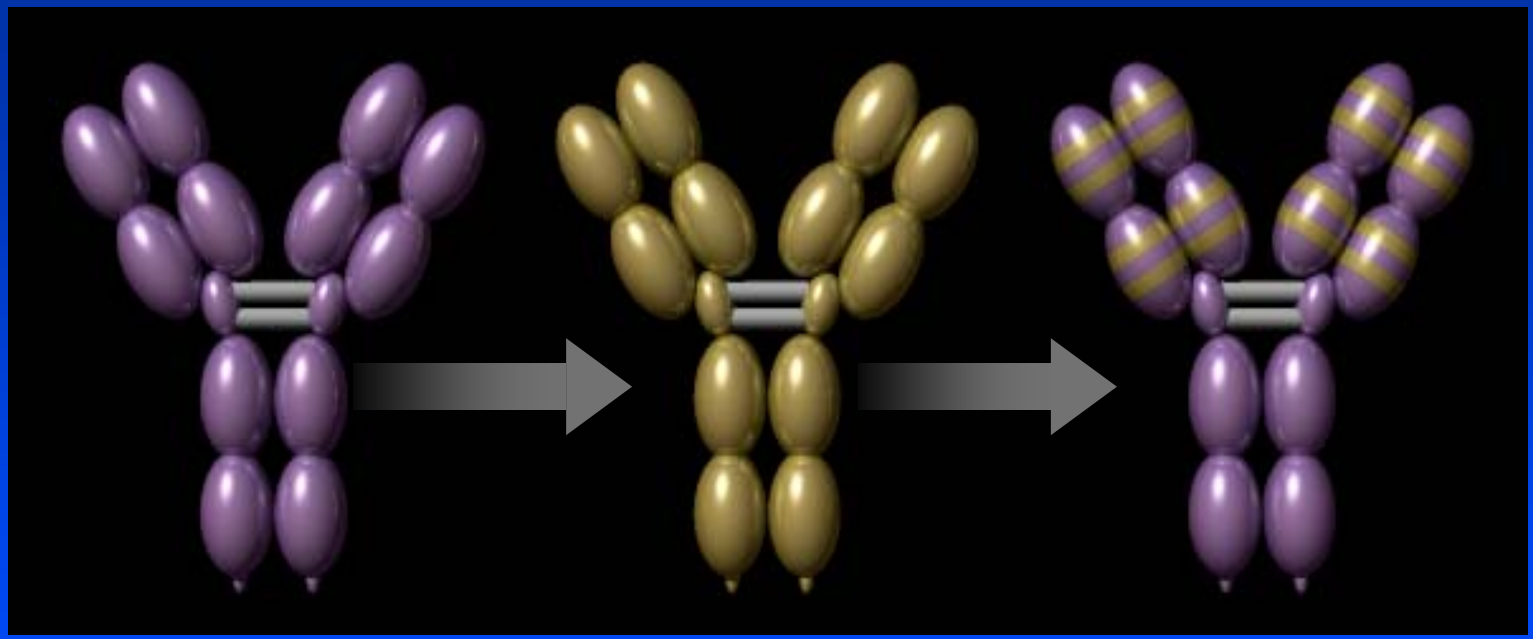


RA Clynes et al (2000) Nature Med 6, 443

Producing clinical grade antibody

- Humanization
- Antigenicity
  - HAMA vs HAHA
- Glycosylation
  - QC
  - quantity

Humanization



Monoclonal Antibody
Human

Monoclonal Antibody
Mouse

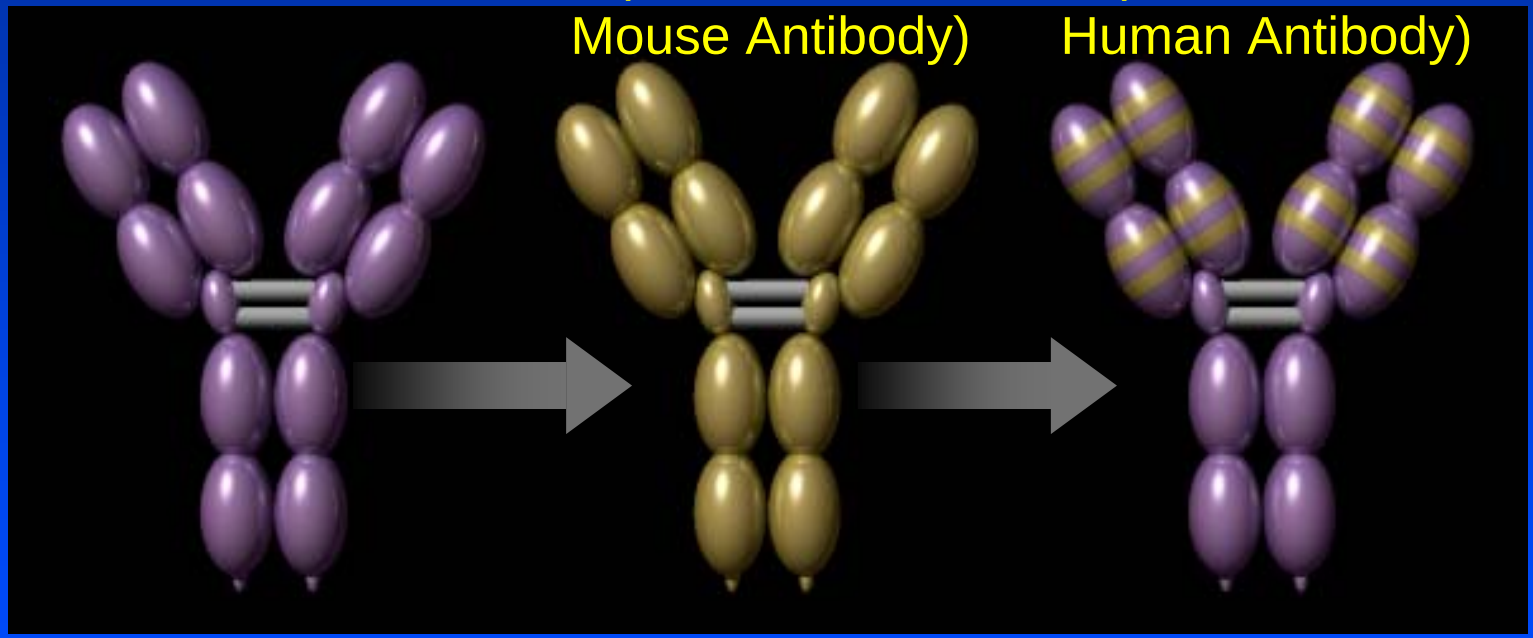
Humanized Antibody

Potential Antigenicity

Minimal

HAMA (Human Anti-Mouse Antibody)

HAHA (Human Anti-Human Antibody)

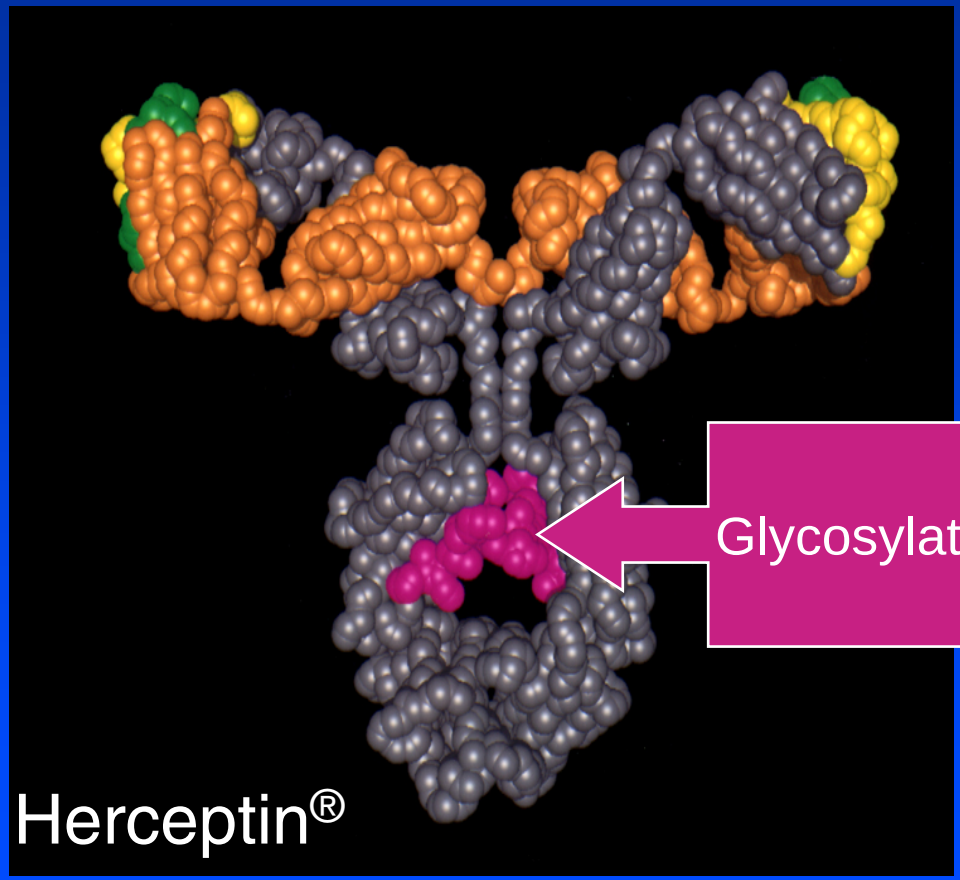


Human Antibody

Mouse Antibody

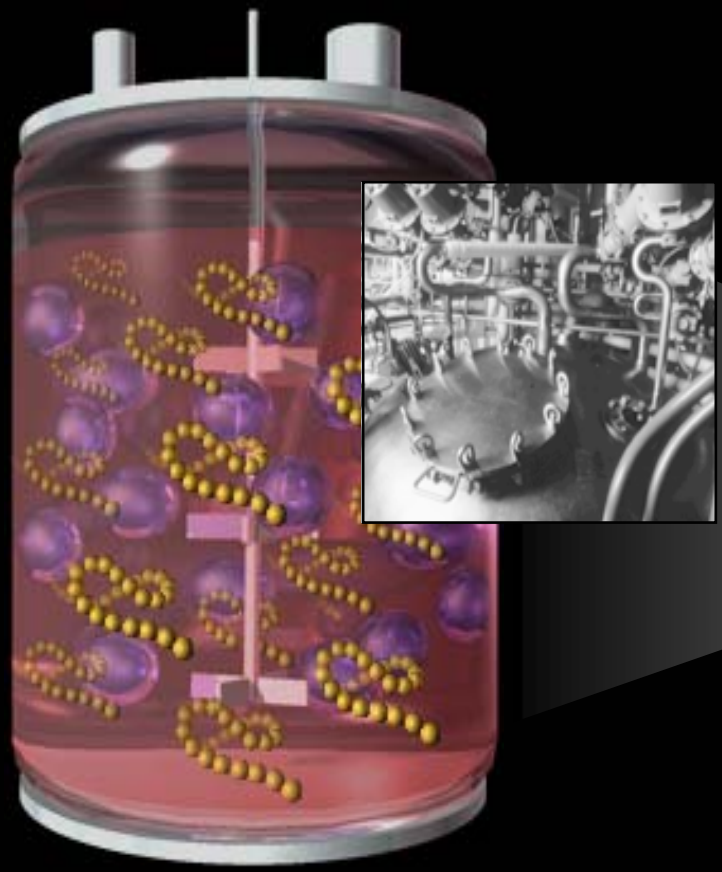
Humanized Antibody

To glycosylate or not to glycosylate?



- Required for effector functions
- CHO cells do it
- e. coli don’t

Manufacturing Process and Product Packaging



Large Scale Fermentation
(1,000 L)

Purification

Packaging

Safey Assessment

- Helps determine starting dose in clinical trials
- Material should be as close as possible to clinical product.
  - Avoid false positive safety signals.
  - Determine manufacturing standards.
- Often need to be done in primates because of limited species cross-reactivity.
  - Monkey anti-human IgG immune response may limit duration.

Clinical Trials

- Identifying patients
- Phase I, II, III paradigm
- Recruiting patients
- Difficulty of adjuvant trials
- Initial trials often in relapsing patients for whom other therapies have failed.

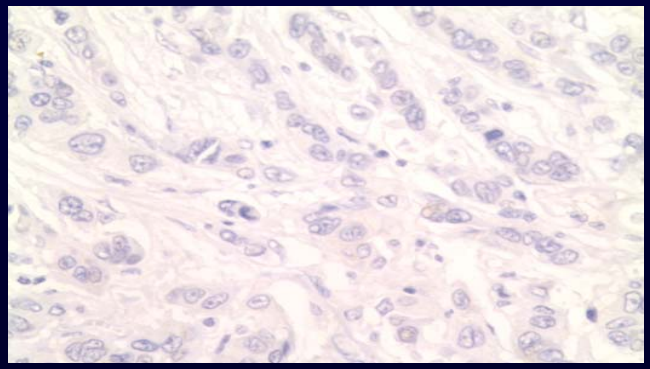
Identifying patients to treat

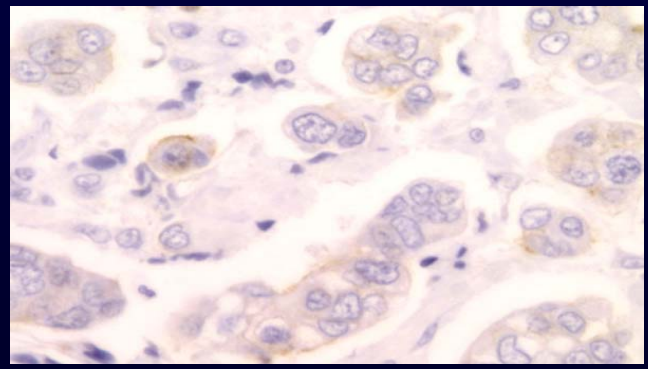
- Herceptin only active against HER2 3+
  - Need biopsy
- Only ~30% of breast cancer patients are HER2 3+
  - need to “see” 3 patients for every one treated
- HercepTest
  - Immunohistochemistry
- FISH
  - Fluorescent In Situ Hybridization

Immunohistochemistry (IHC): Performance Issues (HercepTest®)

- Pre-analytical tissue processing
- Reagent variability
- Antigen retrieval
- Scoring

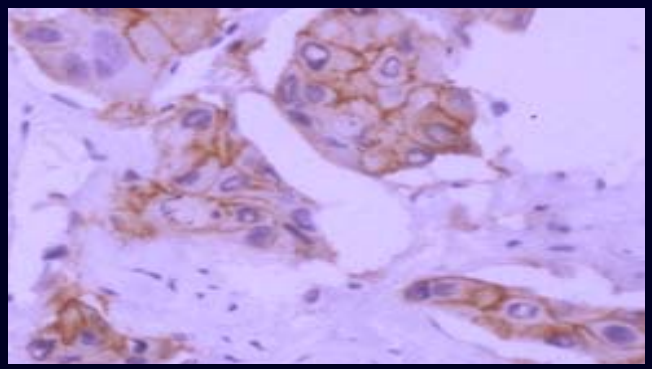
0

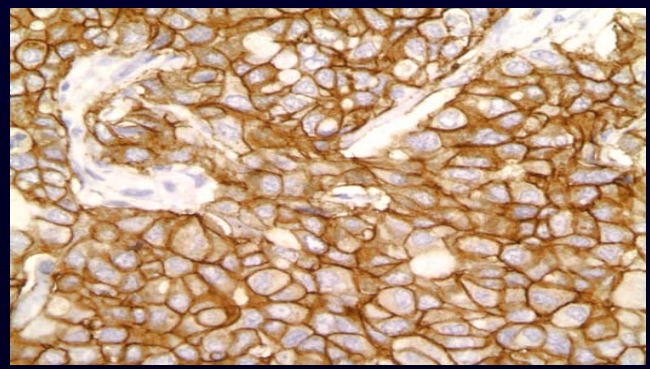




1+

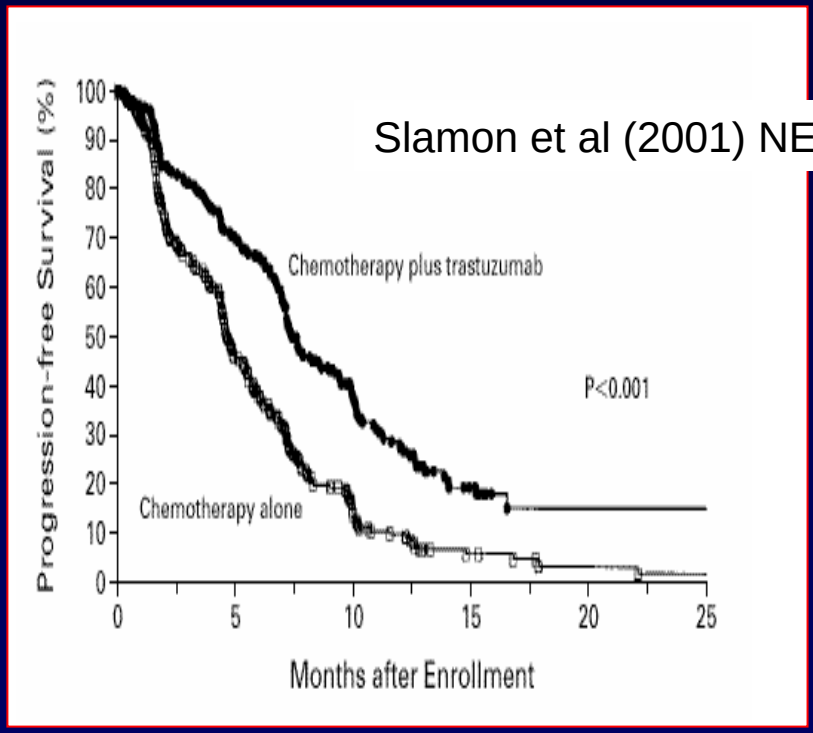
2+





3+

Herceptin™ confers survival benefit in metastatic breast cancer

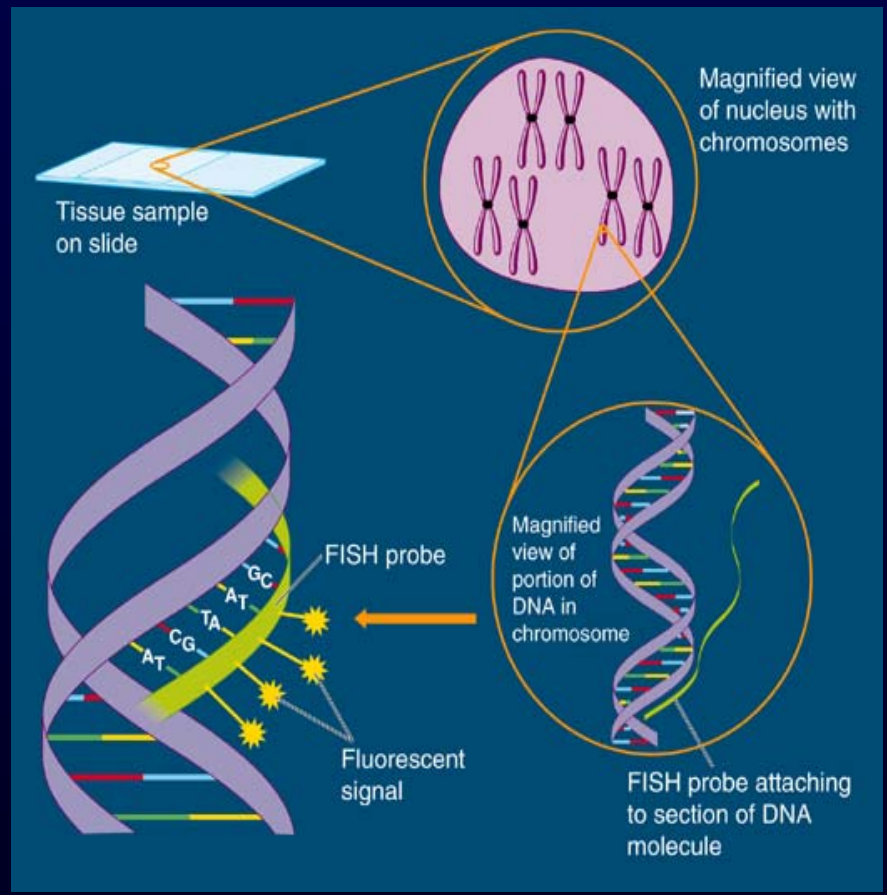


Slamon et al (2001) NEJM 344, 783-792

enrollment based on HercepTest

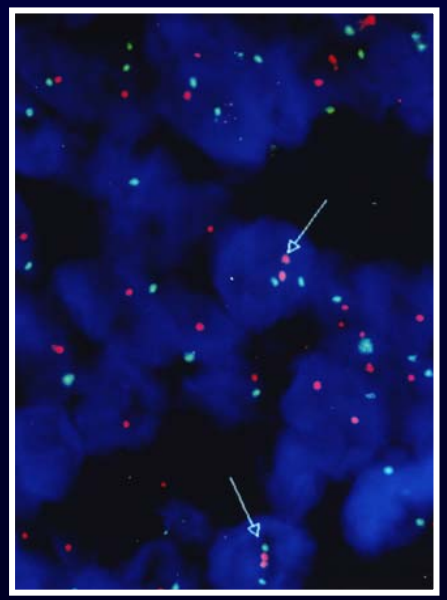
Fluorescence *in situ* Hybridization (FISH): PathVysion™

- Key features:
- Probes
  - Direct labeled
  - HER2 sequence
  - Chromosome 17 centromere
- Interpretation
  - Signal enumeration
  - Ratio of HER2:Chr 17 signals

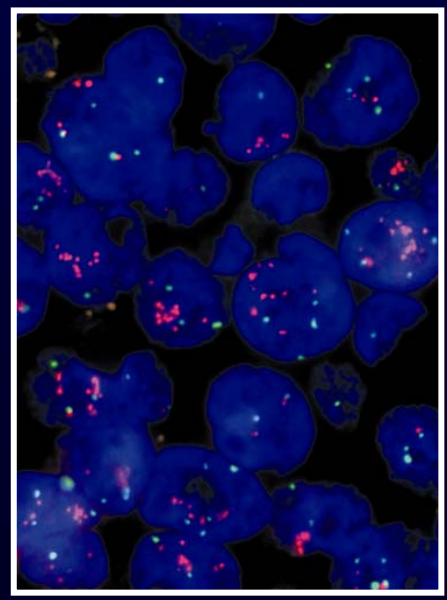


HER2 Diagnostics: Fluorescence *In Situ* Hybridization

- Measures the level of HER2 gene amplification
- PathVysion™ may be preferable due to internal control
- Issue: not performed in-house at all hospitals

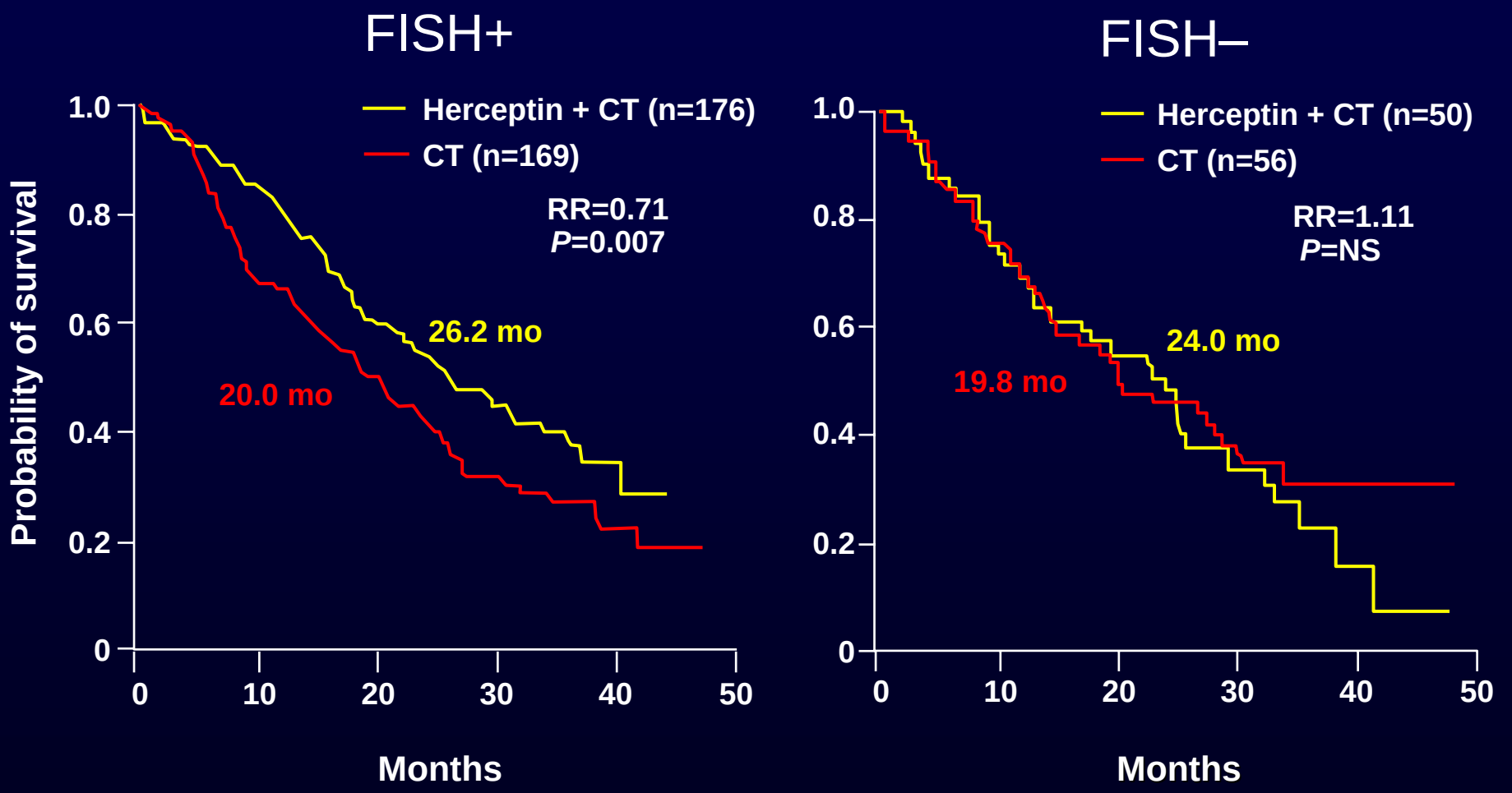


<2.0 not amplified
(FISH–)



≥2.0 amplified
(FISH+)

Herceptin® Combination Pivotal Trial: Overall Survival



Update of Mass R, et al. *Proc Am Soc Clin Oncol.* 2001;20:22a. Abstract 85.

Phase I = “First in Human” studies

- n = 10s
- Patients or healthy volunteers
- Start with single dose, escalate to multi-dose
- Purpose
  - Determine safety and tolerability
  - Determine pharmacokinetics
  - Determine dose for later phases

Phase II = “Small Population” studies

- n = 10s-100s
- Determine safety and efficacy in relevant populations
  - eg, HER2 3+ (FISH+)
- Subgroup analysis
  - eg, prior therapies
- Additional dose-ranging
- Randomized, placebo-controlled
  - nobody wants to be in placebo group
- Drug interactions

Phase III = “Pivotal” studies

- n = 100s - 1000s
- Confirm efficacy with good statistics
- Determine true clinical benefit and its magnitude
  - Increased survival
    - Not always predictable from response rate
  - Quality of life??
- Typically similar to phase II

Phase IV = “Post marketing” studes

- n = 1000s - millions
- Extend safety, dose-schedule
- Test new indications not covered in phase II, III
  - eg Herceptin in early breast cancer

Delivery of Therapeutic Antibodies

- Long half-life
  - can de dosed weekly, or every 2-3 weeks.

- Must be delivered IV, requires clinic visit, iv access.
  - Subcutaneous dosing would be ideal, but difficult
    - 5 mg/kg = 350 mg/patient
      - ~3.5 ml at 100 mg/ml

Recruiting patients

- Competition between new therapies for patients
- <15% of eligible patients participate in clinical trials
  - Stigma that there's nothing else
  - Reimbursement
  - Hassle of coming to clinic every week
- New drugs often tested in combination with current standard of care.

Recruiting patients (cont'd)

- Cross over design (example)

Taxol + Antibody X

Taxol + Placebo $\xrightarrow{\text{progression}}$ Taxol + Antibody X

- Good for recruiting patients
- Bad for interpreting results
  - May make your drug look less effective

Measuring response

- Response rate = tumor shrinkage
  - PR = Partial Response = >50% decrease
  - CR = Complete Remission
- Time to progression vs survival
- Avastin in breast cancer increased response rate to chemotherapy but failed to improve survival.
- Approvable endpoints vs pharmacodynamic markers of response
  - Iressa rash

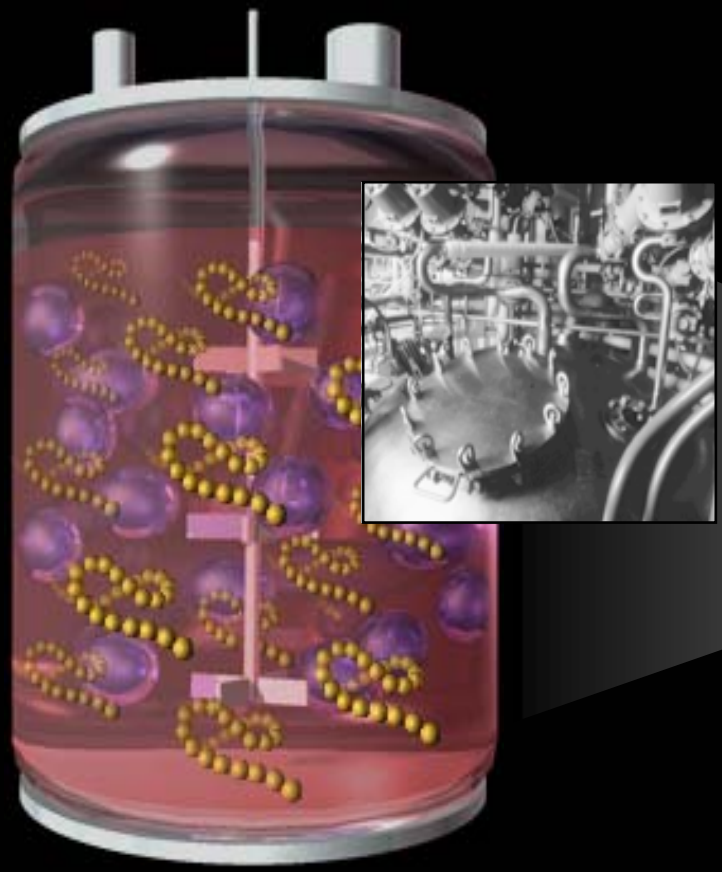
Adjuvant Clinical Trials

- “Adjuvant” therapy is given to patients in whom detectable disease has been surgically removed but there is chance that tumor cells have spread to other sites
  - local lymph nodes positive
- Purpose: to minimize/prevent relapse of disease later in life.
- Most likely scenario for antibody therapies to be efficacious
  - low bulk
  - minimal selection

Difficulties with Adjuvant Clinical Trials

- Only fraction of patients will develop disease later
  - can't predict who
    - need to treat more patients
- Relapse often takes years
  - long clinical trials.
- need to know drug has activity before exposing patients to this.

Manufacturing Process and Product Packaging



Large Scale Fermentation
(12,000 L)

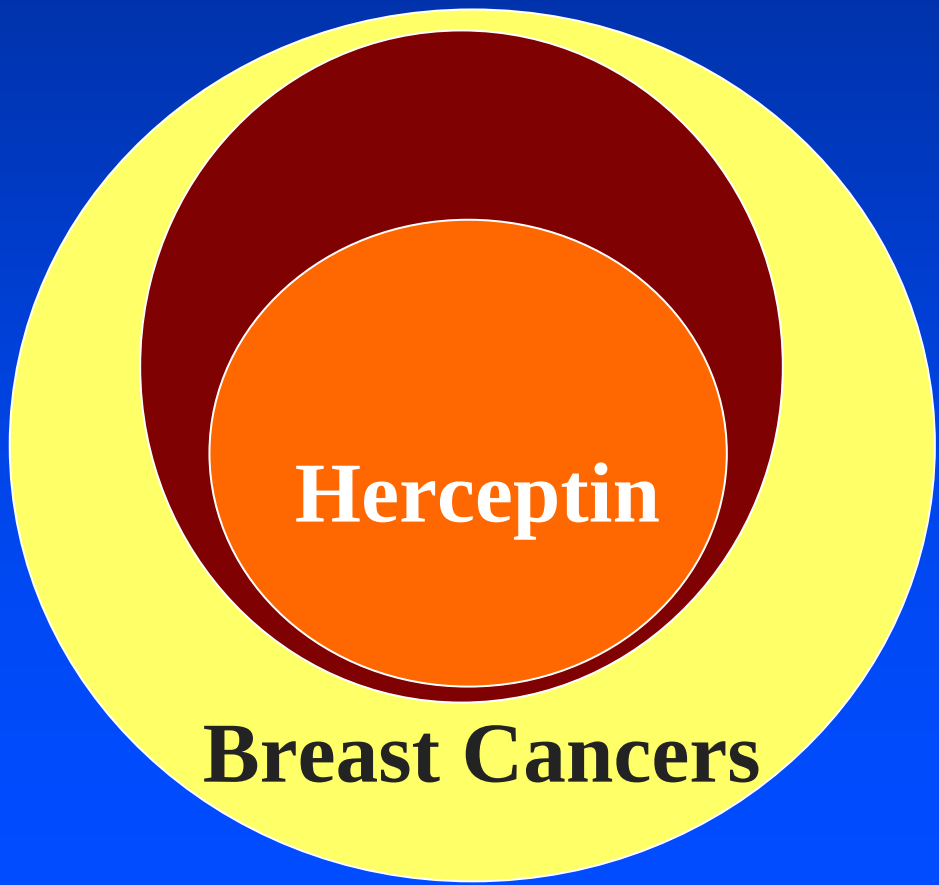
Purification

Packaging

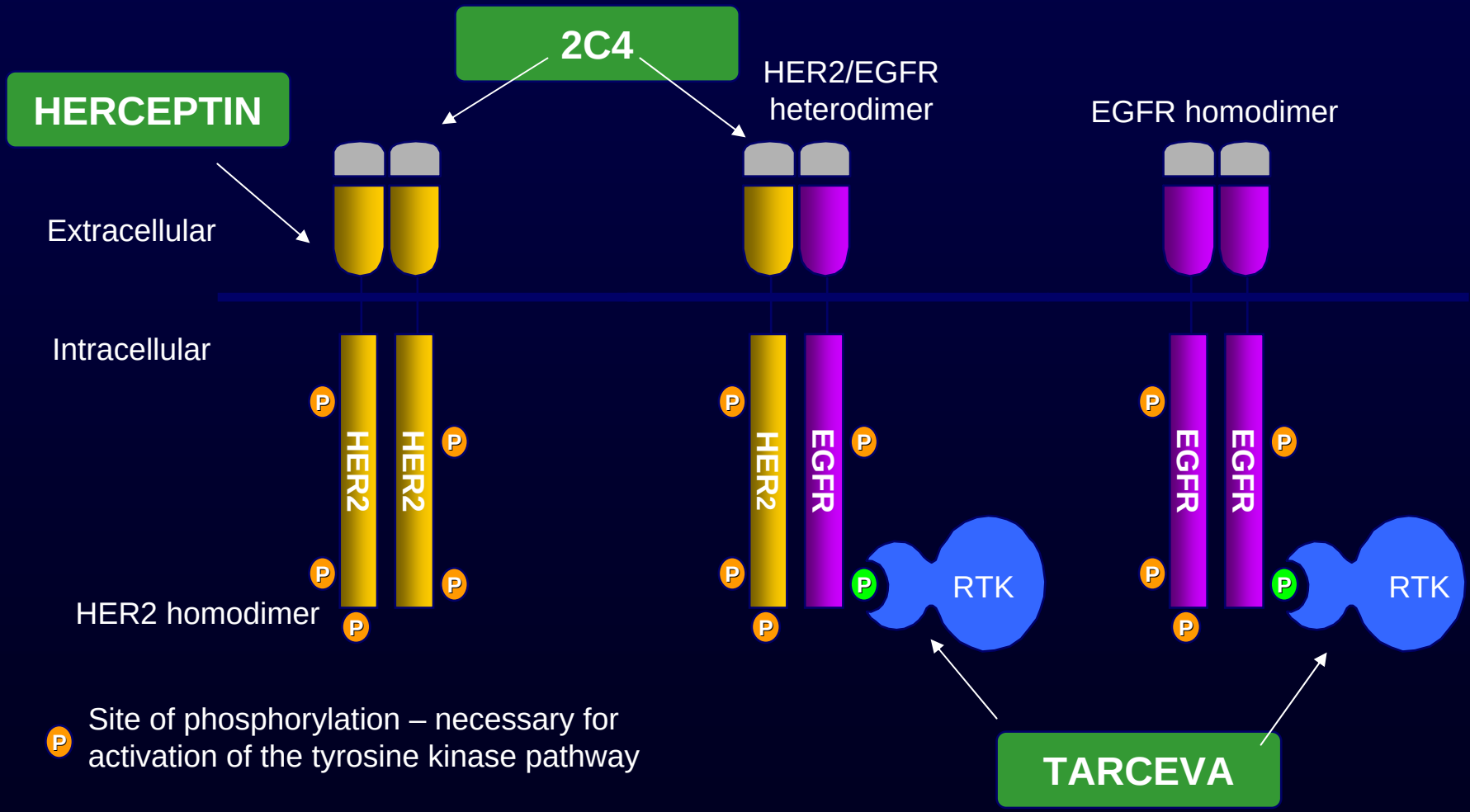
Meeting the market

- Advocates help encourage clinical trial participation
- Time between end of trial and approval difficult

Breast Cancer Subpopulations



The HER Family Presents Several Targets for Biologic Therapies



Issues Surmounted by Herceptin

- Identification
- Production
- Preclinical Efficacy
- Safety Assessment
- Clinical Trials

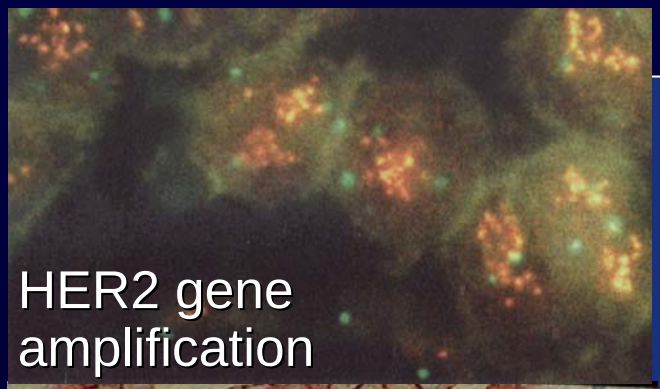
Acknowledgements

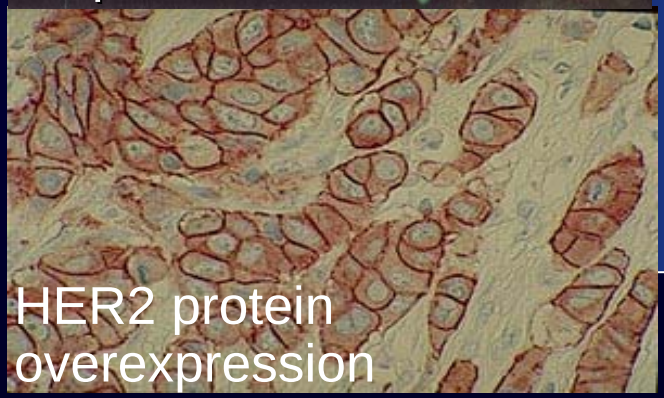
- Virgnia Paton
- Gail Phillips
- Mark Sliwkowski
- Genentech Herceptin team
- Graphics

Issues in developing monoclonal antibodies as cancer therapeutics

- Identification of antibody
- Preclinical Efficacy
- Production on appropriate version of antibody
- Safety Assessment
- Clinical Trials

Targeting HER2: Scientific Rationale





Shortened median survival*	
HER2 positive	3 years
HER2 normal	6–7 years

*Combined metastatic and adjuvant patients.

Slamon D, et al. *Science.* 1987;235:177-182.
Pauletti G, et al. *J Clin Oncol*. 2000;18:3651-3664.