

AntibodyServices:
Antibody Engineering

www.glycotope.com

Contact:

hans.baumeister@glycotope.com

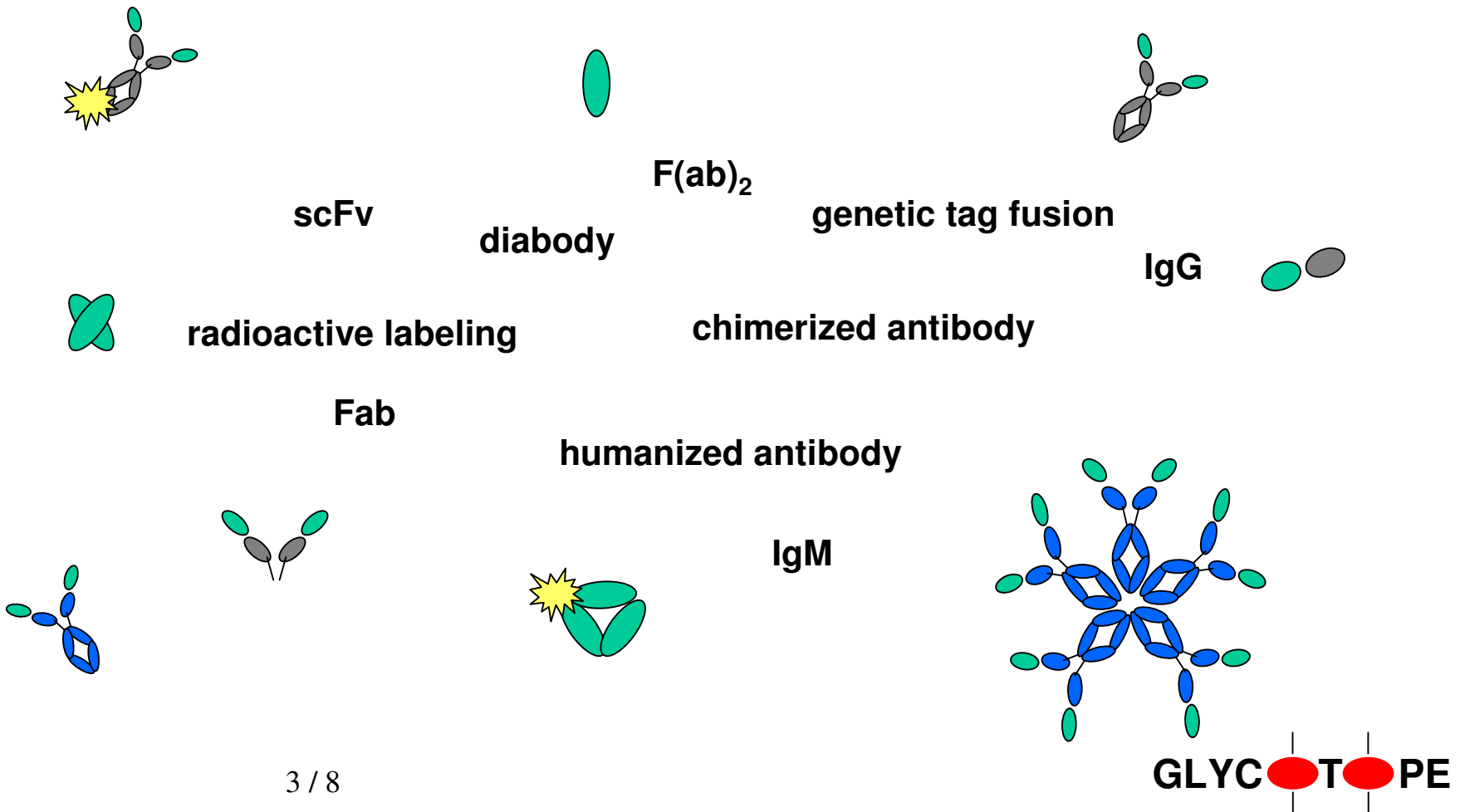
Tel. : +49-30-941084-207

ANTIBODY ENGINEERING

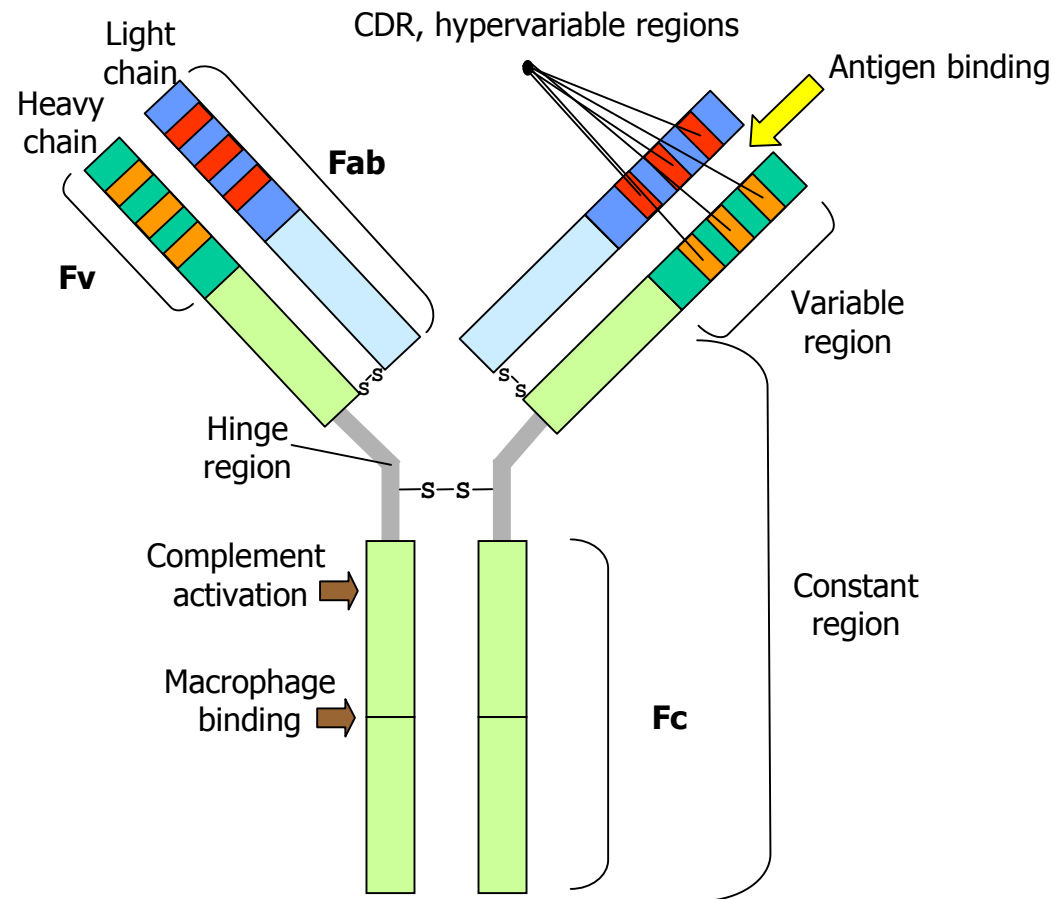
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Choose the Antibody Format You Need

For Each Application, Indication and Therapeutic Strategy the Optimal Antibody Construct



Antibody Features



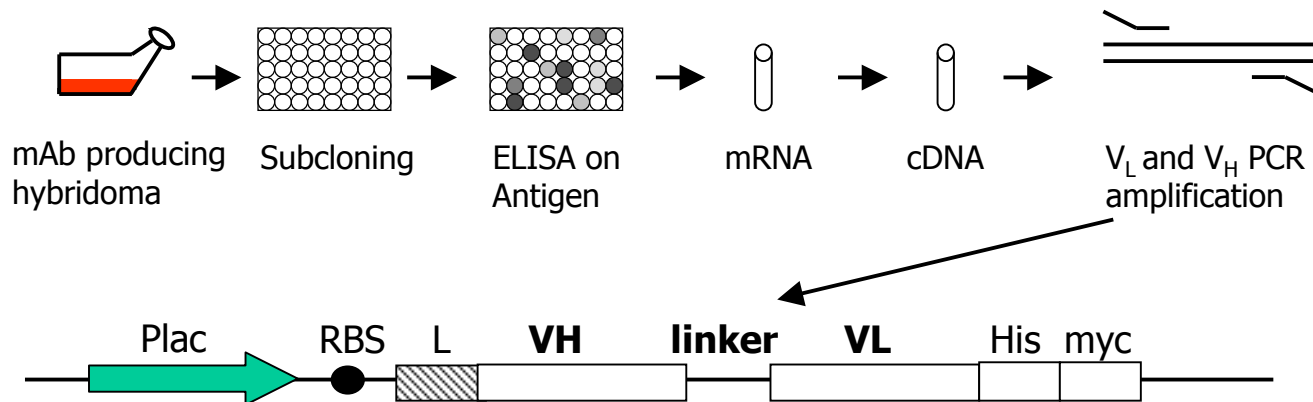
Antibody Sequence Cloning

Functional antibody sequences can be cloned from:

- Monoclonal cell line, e.g. hybridoma
- Spleenocytes from immunized mouse

Identification of functional immunoglobulin genes.

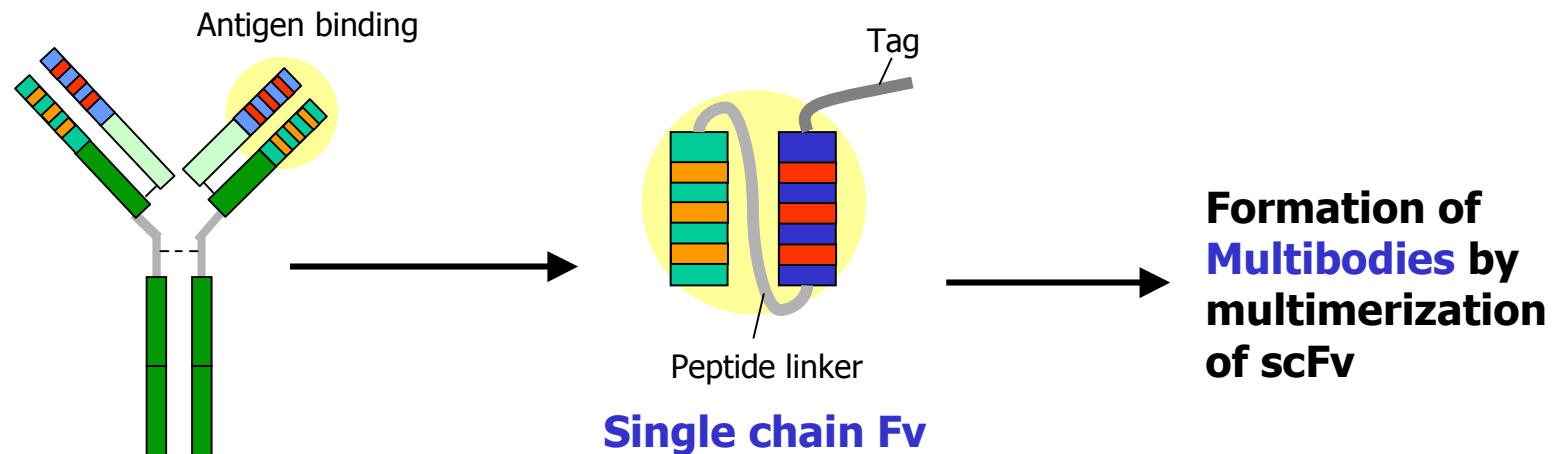
An often occurring problem in cloning of functional V regions is the sequence heterogeneity of hybridoma cell lines due to point mutations, insertions and deletions. Functionality of cloned genes is checked by antigen binding assays (ELISA, FACS and/or Immunofluorescence) following cloning and bacterial expression of the variable region antibody fragments.



Antibody scFvs and Multibodies

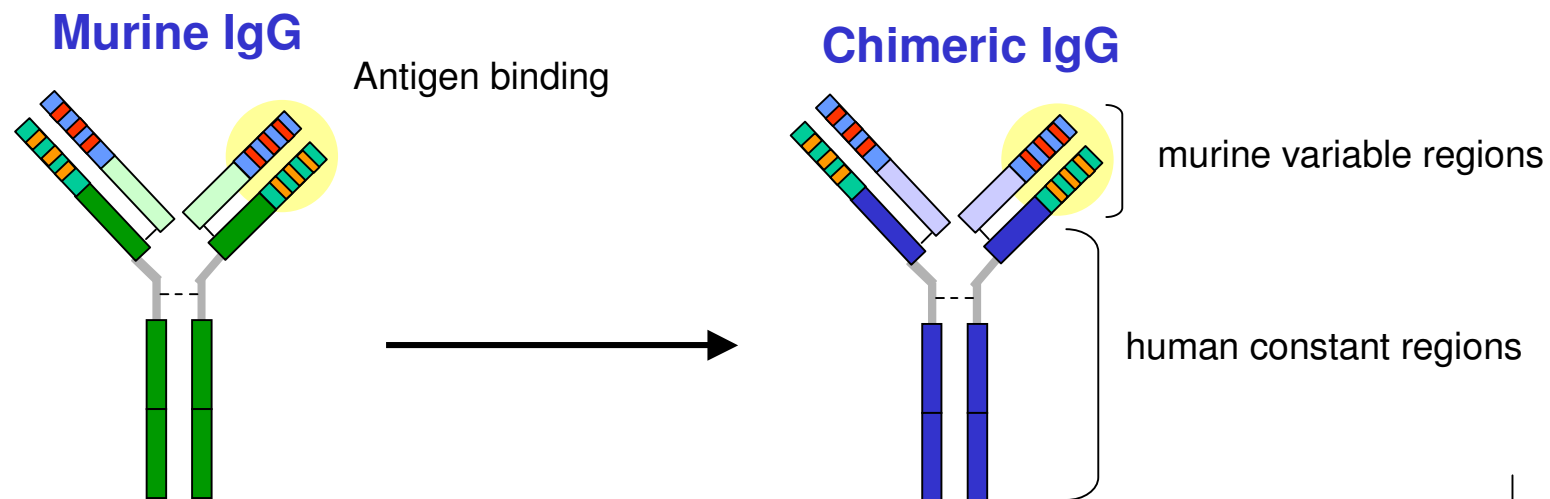
- Single chain antibody fragments (scFv) are constructed by connecting variable heavy and variable light chain through a linker.
- Multimerization of scFv is obtained by e.g. optimization of the linker length.
- Genetic fusion of antibody fragments to a variety of common protein tags for detection and purification is possible.
- Functionality of antibody fragments is checked after bacterial expression.

Murine IgG



Antibody Chimerization

- Own vectors for chimerization based on genomic sequences for the human constant domains and cloning cassettes for the insertion of cDNA from the variable domains of murine antibodies.
- Vectors for cIgG1 and cIgM (with and without human J chain) are available; others can be constructed on request.
- Functionality of chimeric antibodies is checked after transient expression in COS cells.
- Functional antibody is stably transformed into suitable cell lines, e.g. CHO expression cells, adapted to serum free conditions and purified from supernatant by protein A or affinity chromatography.



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GLYCOTYPE

Antibody Humanization

Reducing Immunogenicity of a Murine Antibody

- Choice of frameworks and CDR regions is based on a combination of analyses of the homology of the mouse framework to human germ line sequences, human consensus sequences and canonical structures.
- Construction of a set of different humanized heavy and light chain variable domains.
- A series of functional humanized antibody fragments is selected on the level of antibody fragments on the basis of their functional binding affinity and specificity as well as their producibility in bacteria or mammalian cell lines.

Example: Screening of selected humanized clones of a murine carbohydrate-specific antibody on different antigens

AG1: protein antigen carrying CH structure;
AG2: same protein antigen with irrelevant CH structures;
AG3: synthetic CH conjugate;
Positive control (PC): the murine Ab, negative control: irrelevant Ab.

