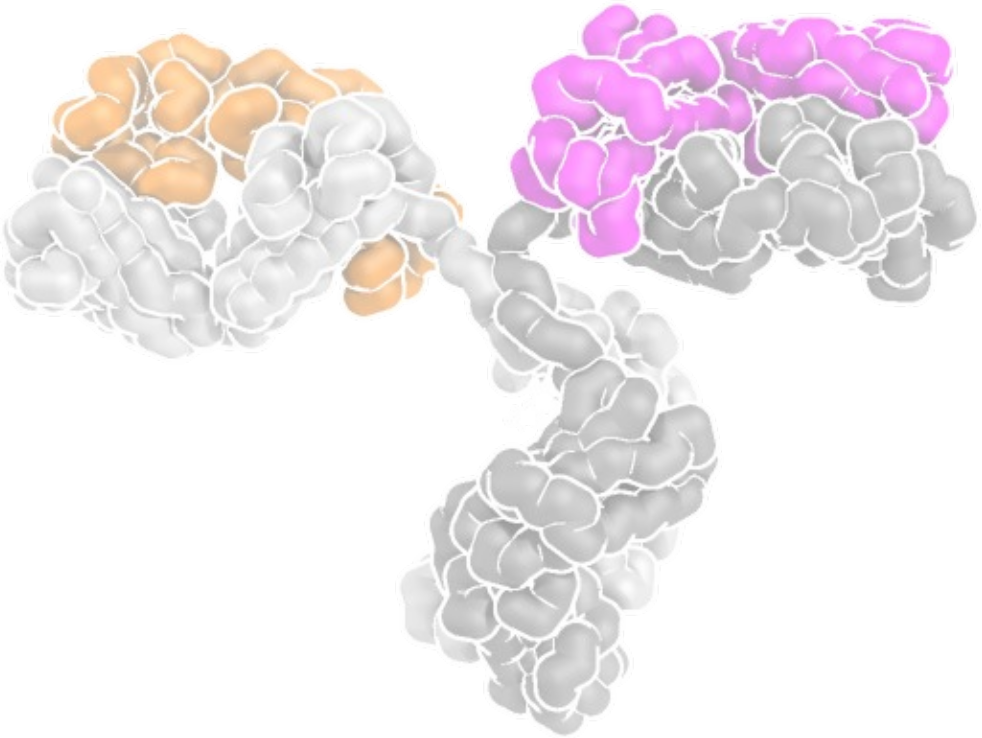
Class core values

1. Be **respect**ful to yourself and others
2. Be **confident** and believe in yourself
3. Always do your **best**
4. Be **cooperative**
5. Be **creative**
6. Have **fun**
7. Be **patient** with yourself while you learn
8. Don’t be shy to **ask “stupid” questions**



Week 9, Lecture 1

Antibodies: a case study

Learning Objectives

1. Identify the potentials and challenges of antibodies
2. Apply knowledge from the course to find methods for antibody engineering
3. Critically evaluate the advantages and disadvantages of different protein engineering methods for improving antibodies

Our immune system is very complex and highly functional

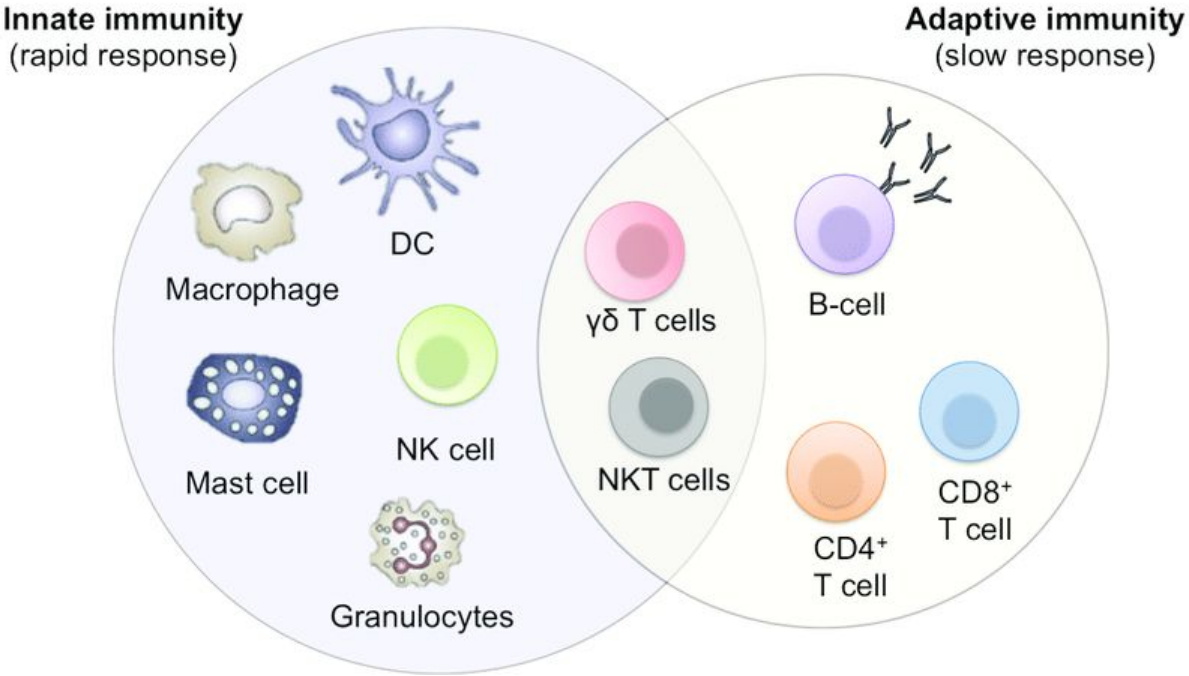
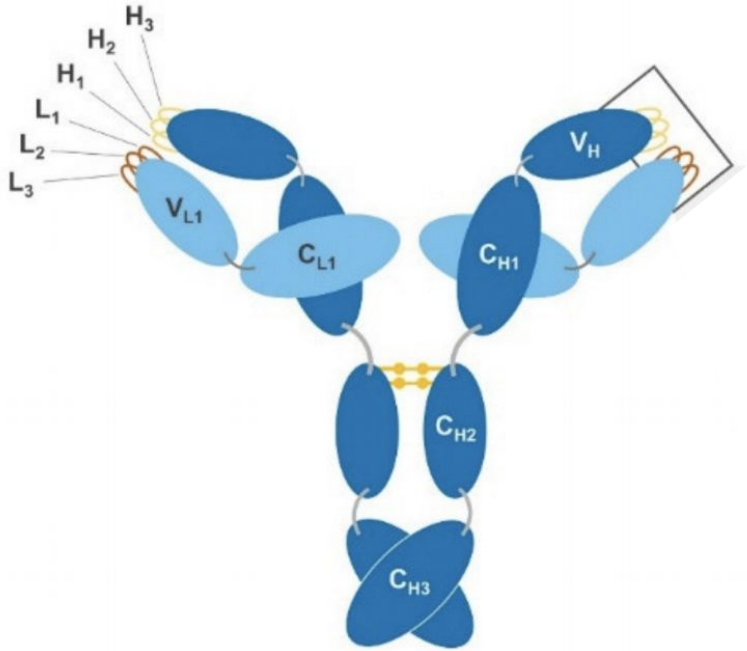


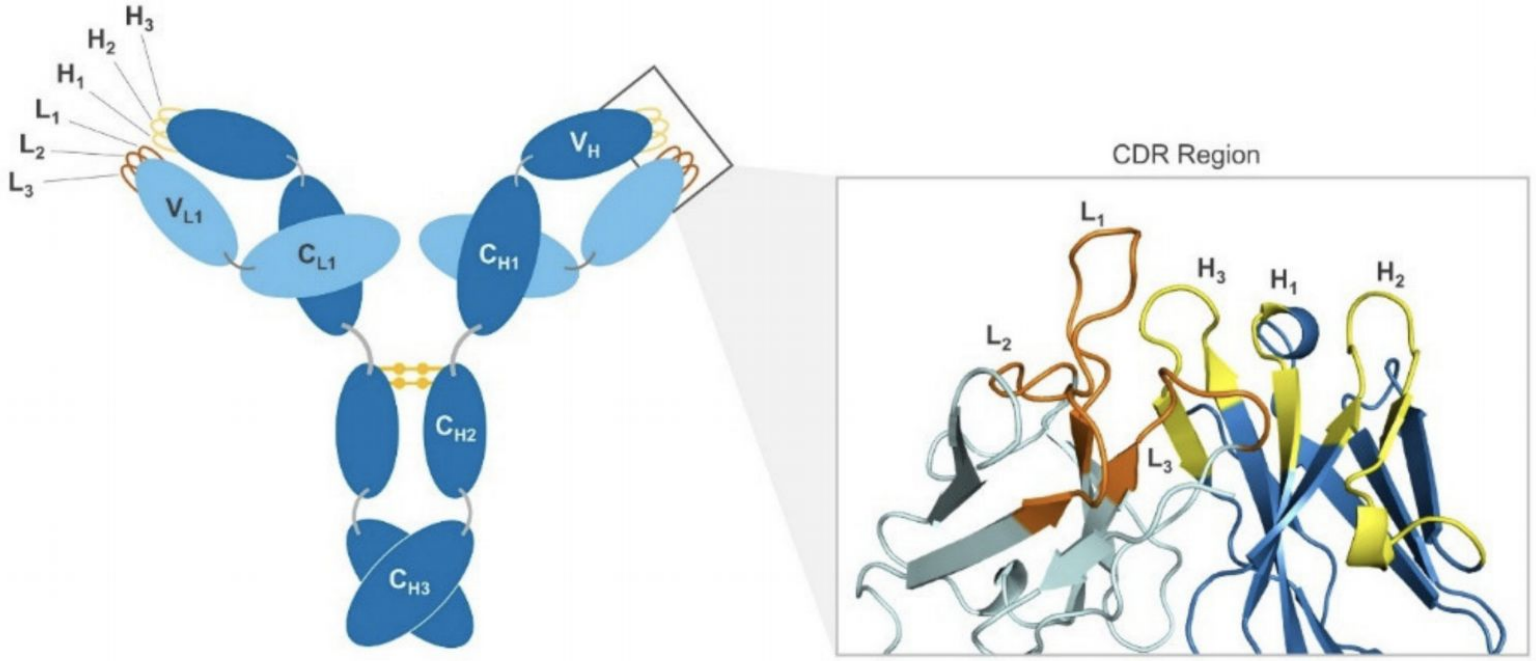
Figure from Cathrin Ritter's thesis, adopted from *Nature Review Cancer* **2004**, 4,11-11

Antibodies can detect a plethora of antigens using a conserved structure



Antibodies **2020**, *9*, 12; doi:10.3390/antib9020012

Antibodies can detect a plethora of antigens using a conserved structure



Antibodies **2020**, *9*, 12; doi:10.3390/antib9020012

Antibodies offer a great therapeutic potential

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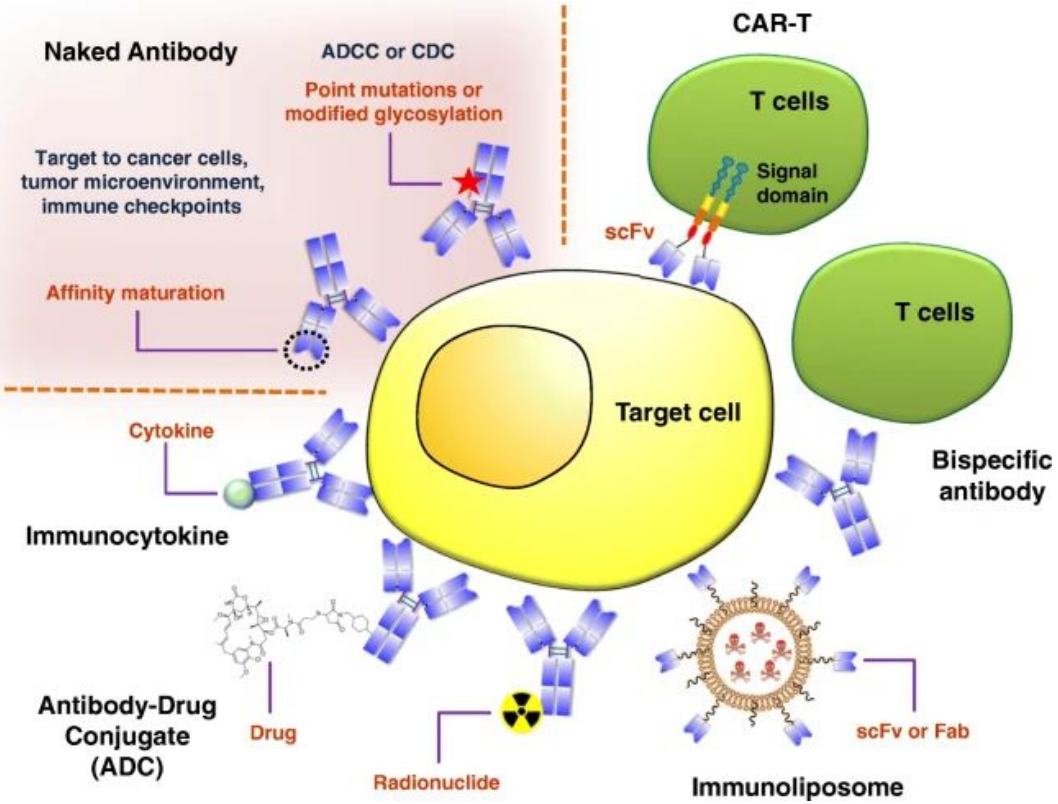
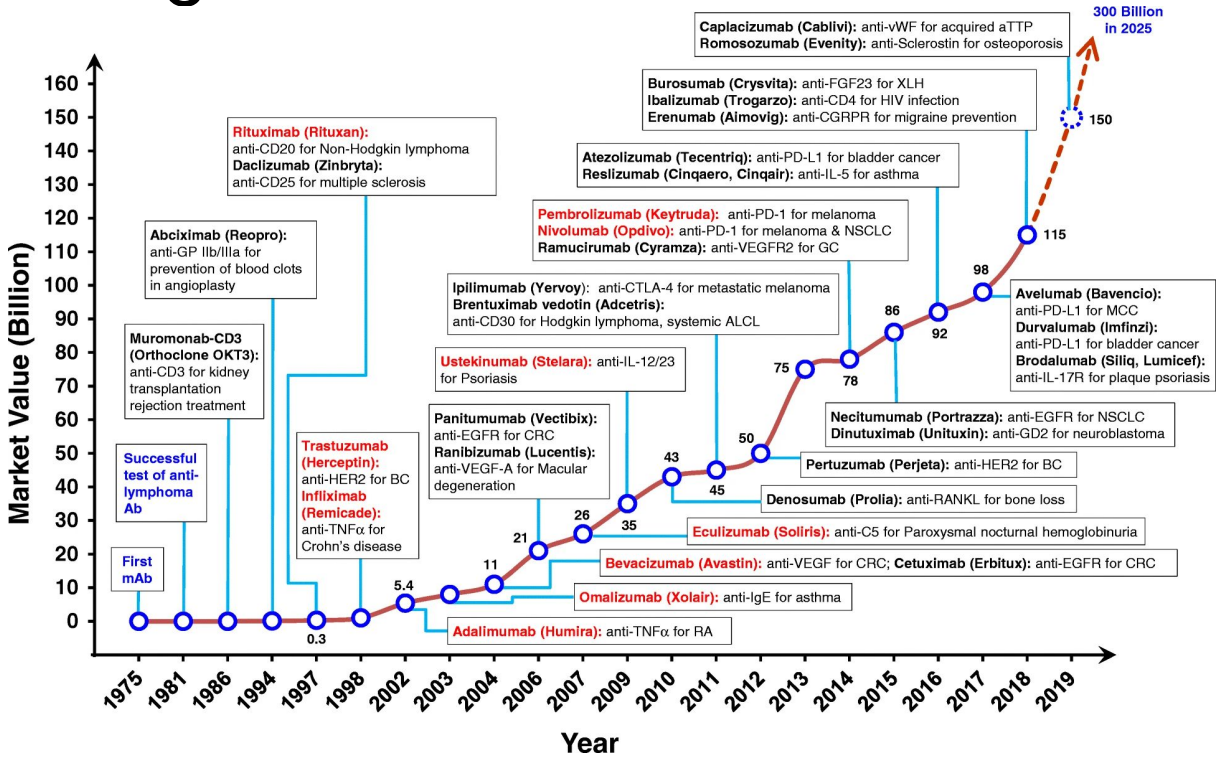


Journal of Biomedical Science 27, Article number: 1 (2020)

Antibodies offer a great therapeutic potential

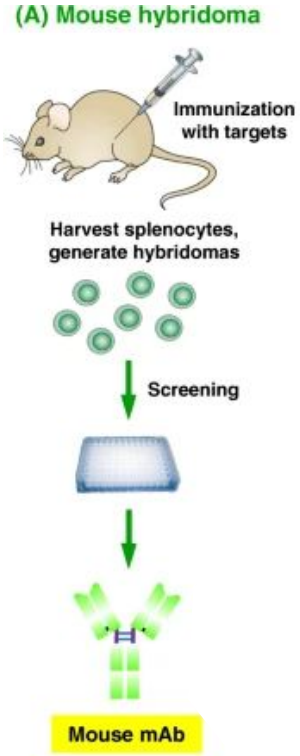
… and a great market!

Antibodies offer a great therapeutic potential … and a great market!



Journal of Biomedical Science 27, Article number: 1 (2020)

How can we obtain antibodies?



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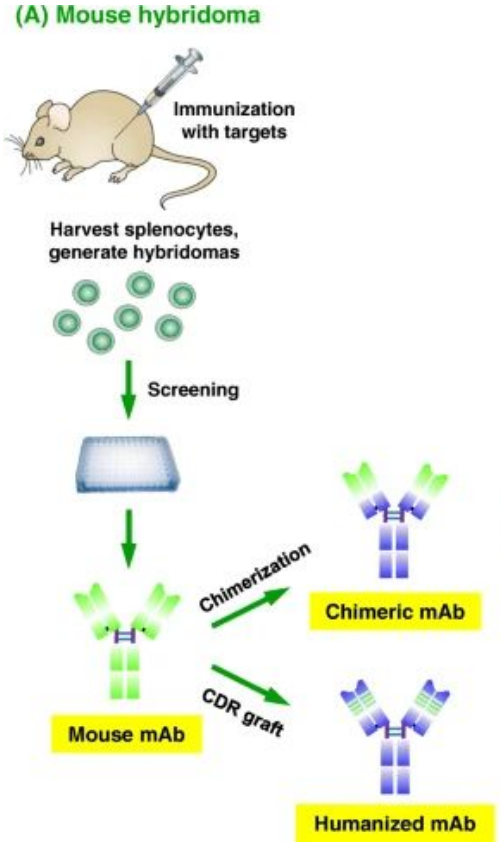
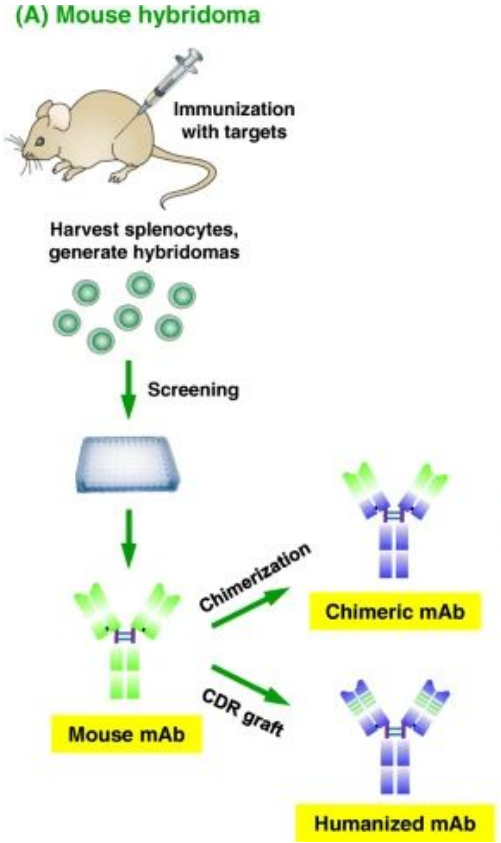
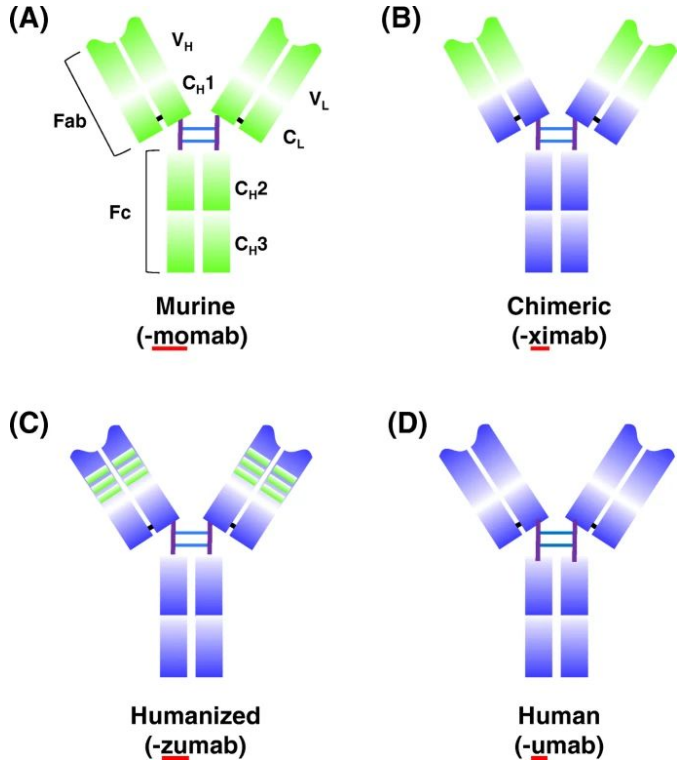


How can we obtain antibodies?





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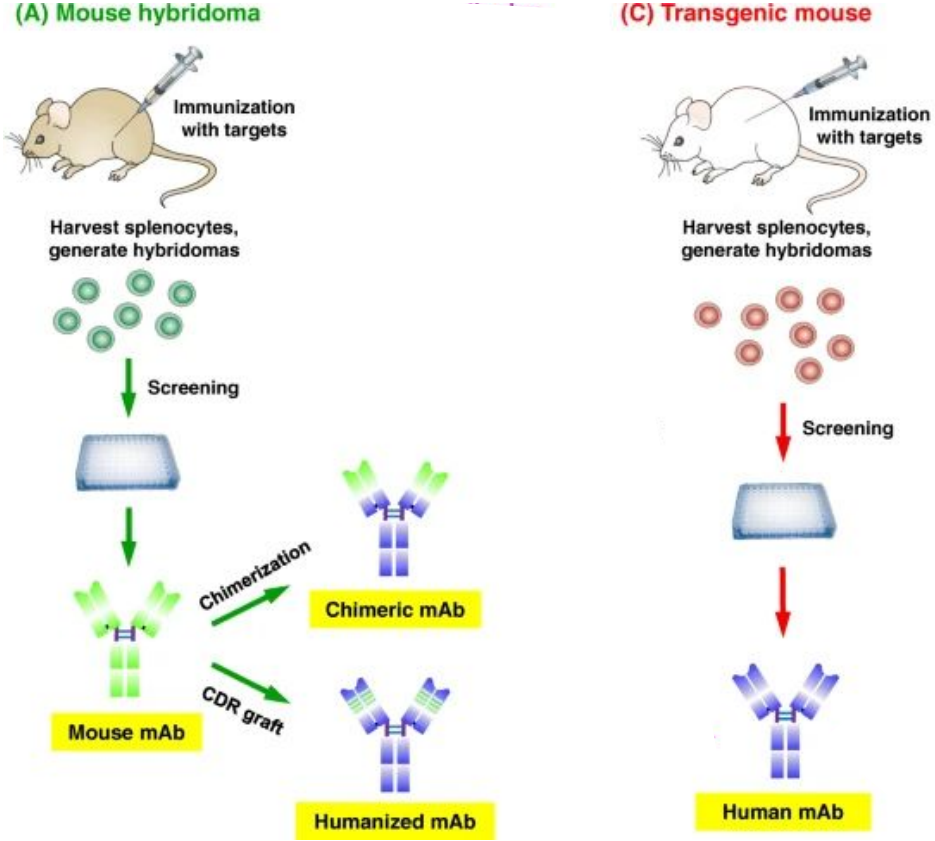
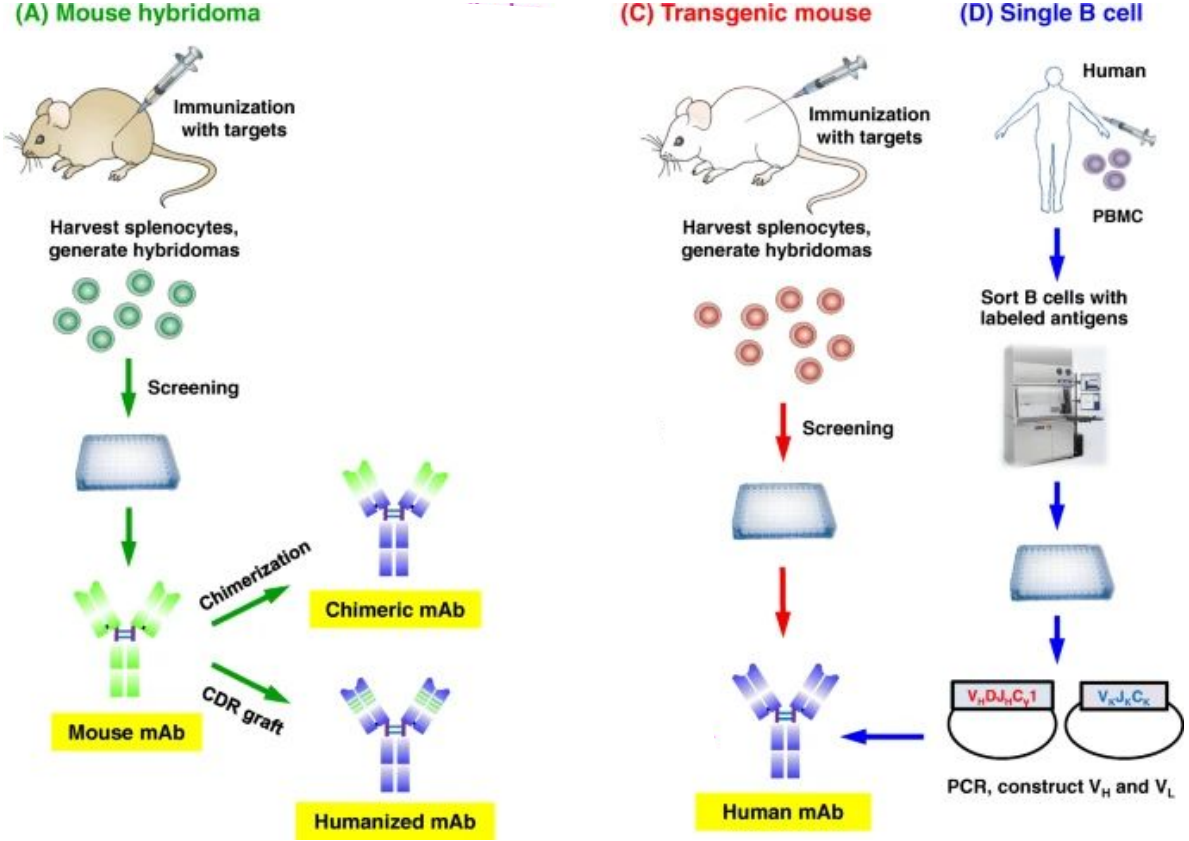


How can we obtain antibodies?



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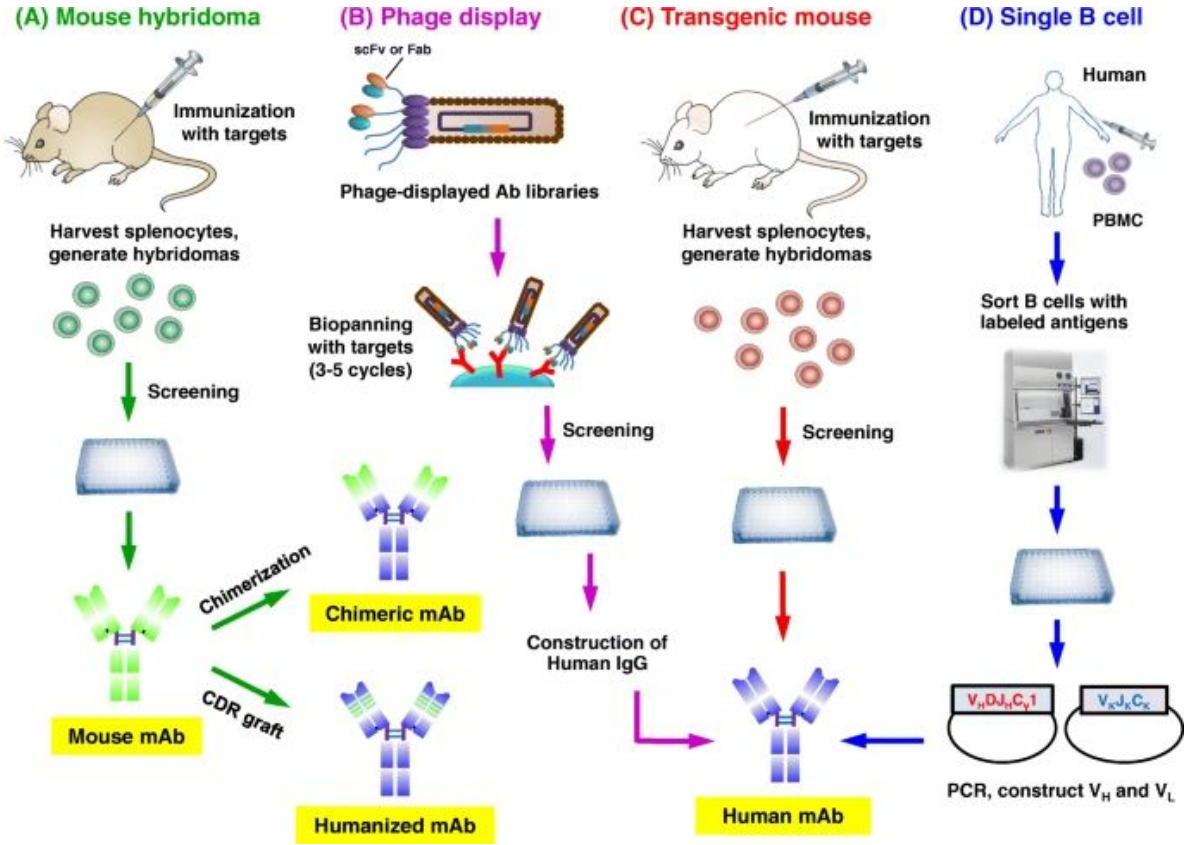
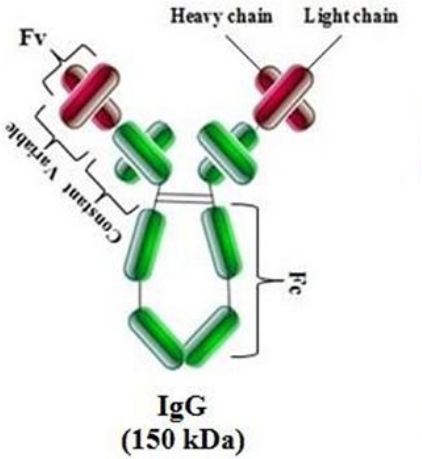


Journal of Biomedical Science 27, Article number: 1 (2020)

Full length antibodies can be challenging to work with



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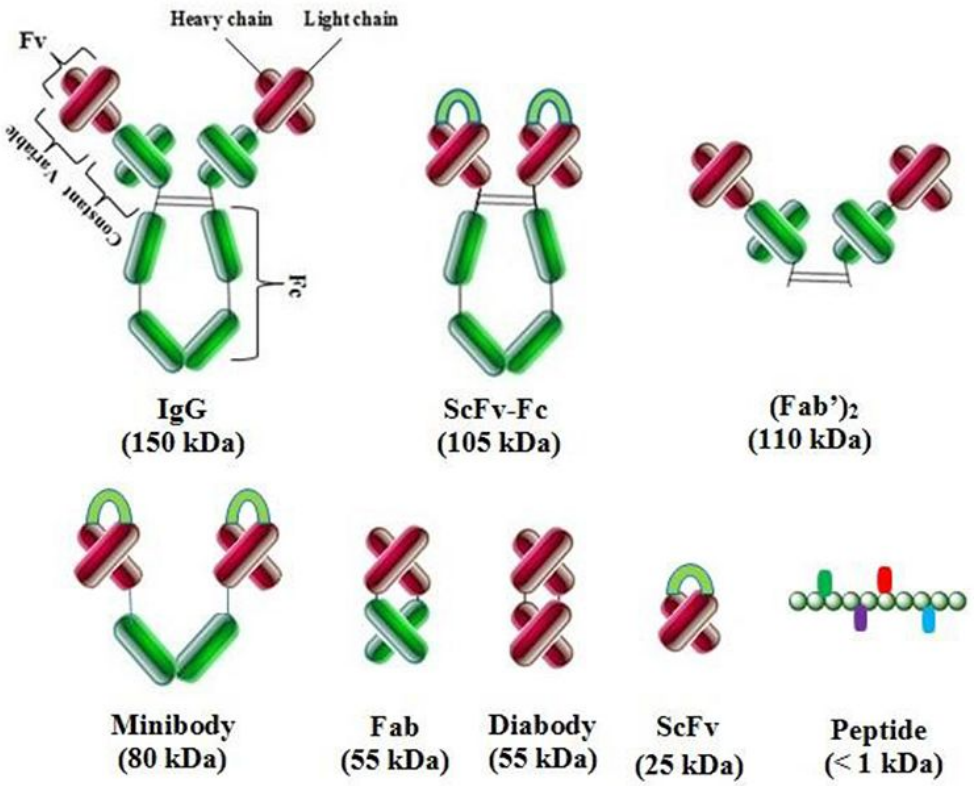

Front. Microbiol., 28 March **2017**

Antibody engineering is one of the most sought after applications of protein engineering

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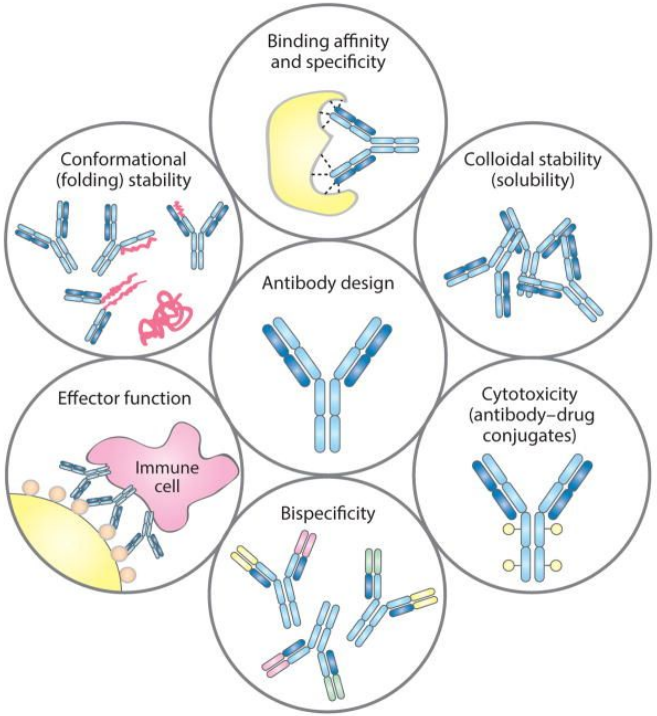
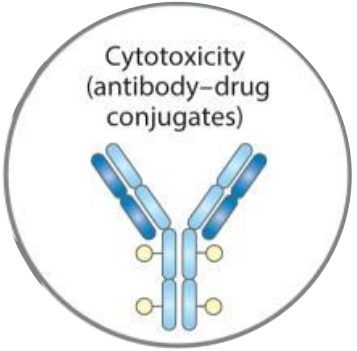
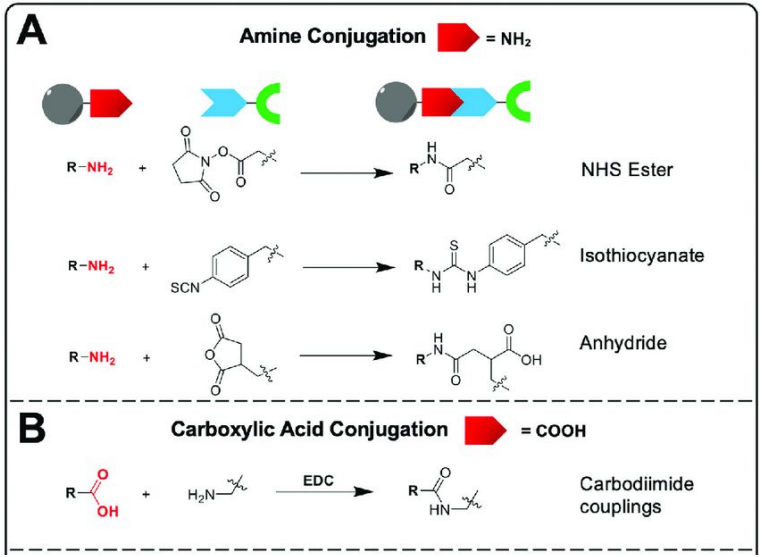
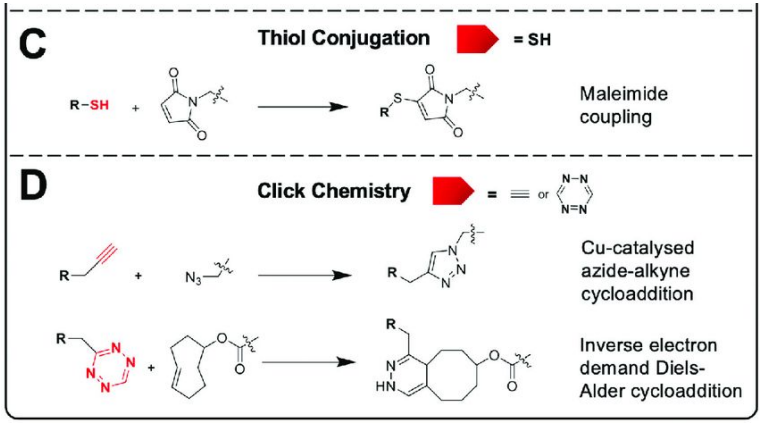


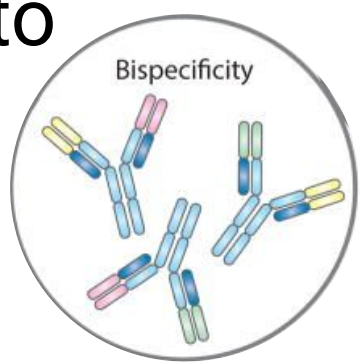
Conjugation of cargo to antibodies can be challenging







Bispecificity is a highly desirable feature to engineer in antibodies



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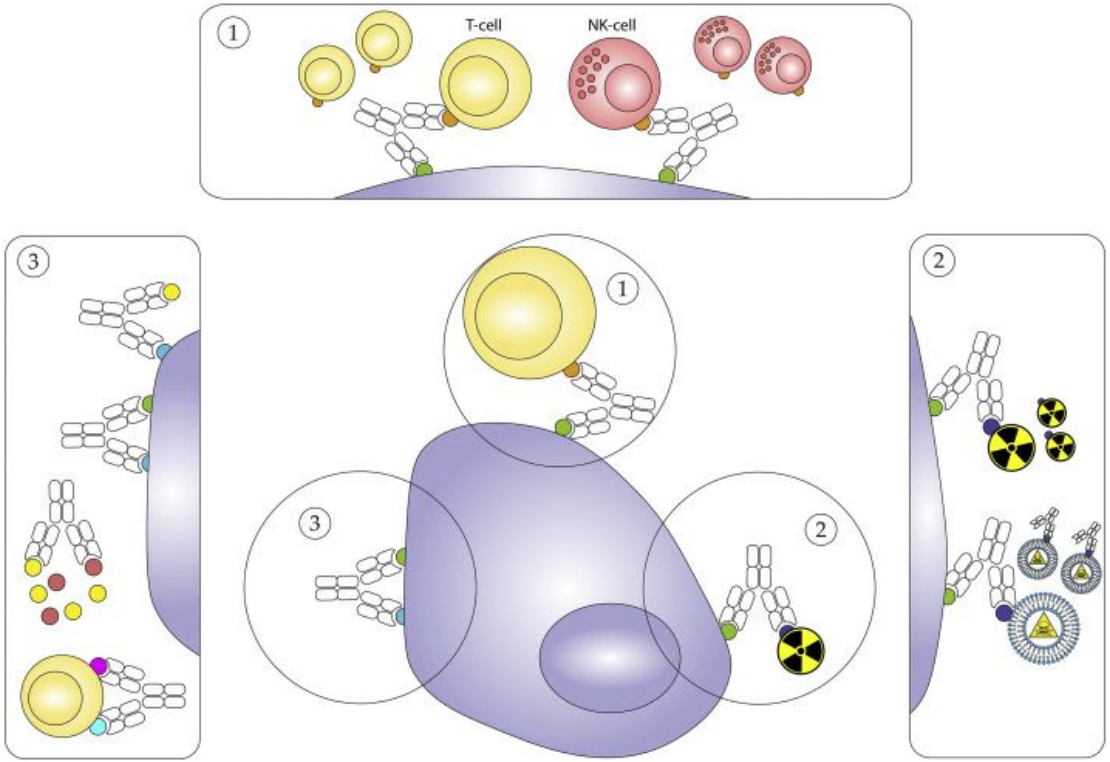
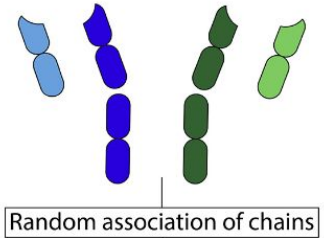


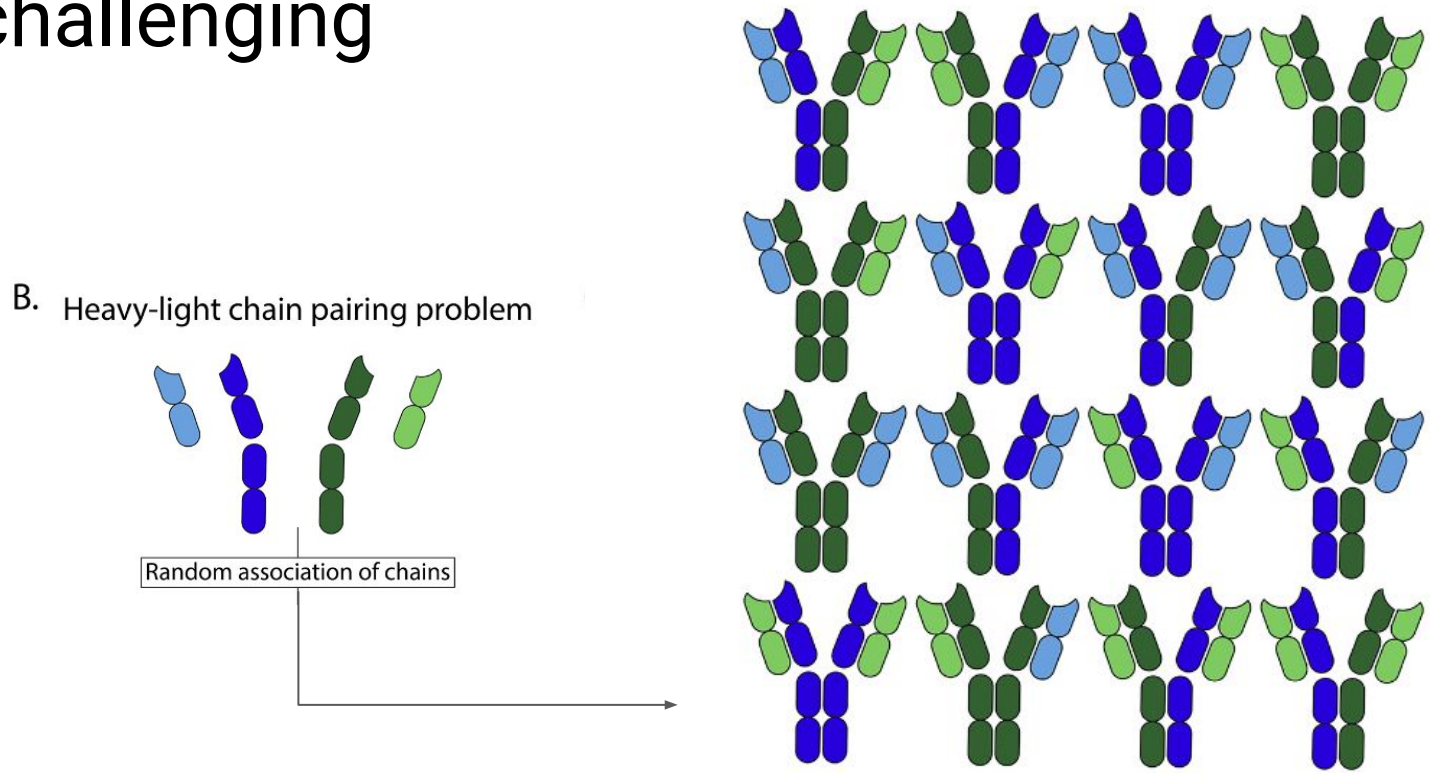
Pharmacology & Therapeutics Volume 201, September **2019,** Pages 103-119

… but obtaining bispecific antibodies is very challenging

B. Heavy-light chain pairing problem



… but obtaining bispecific antibodies is very challenging



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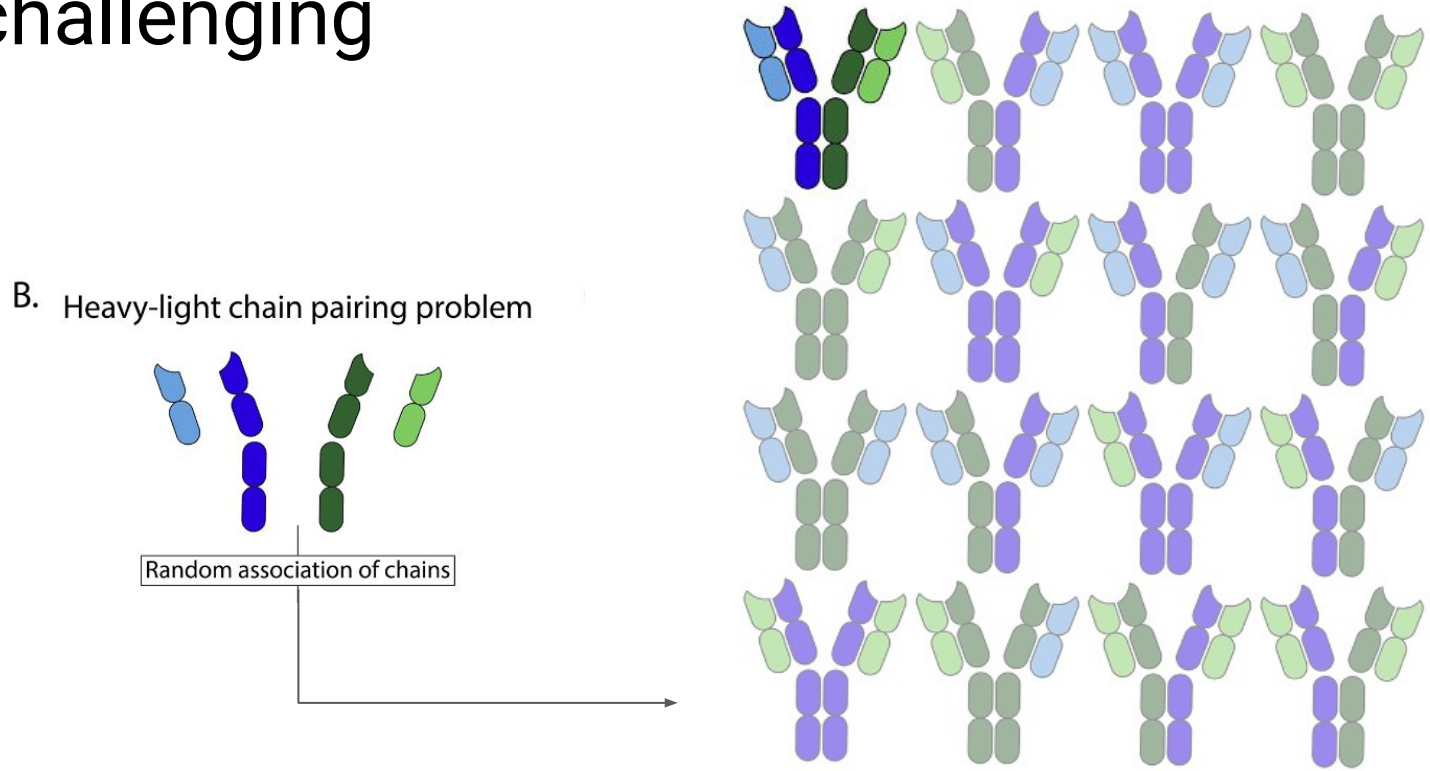
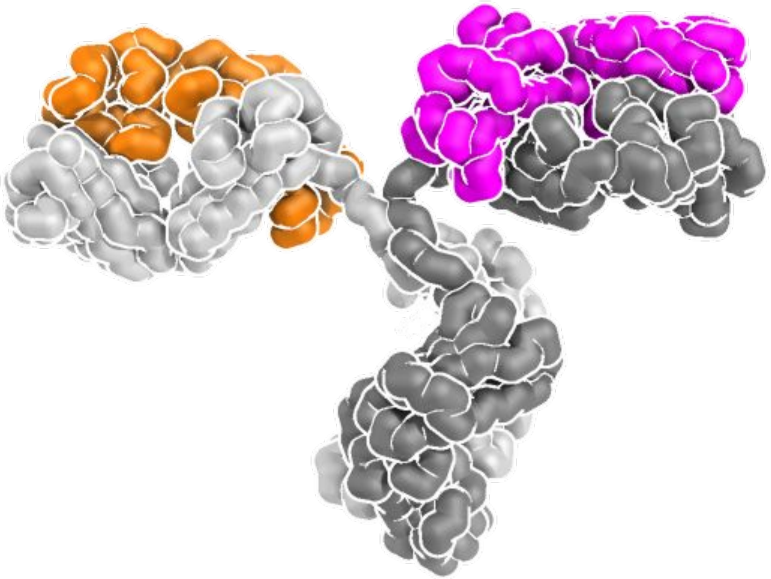
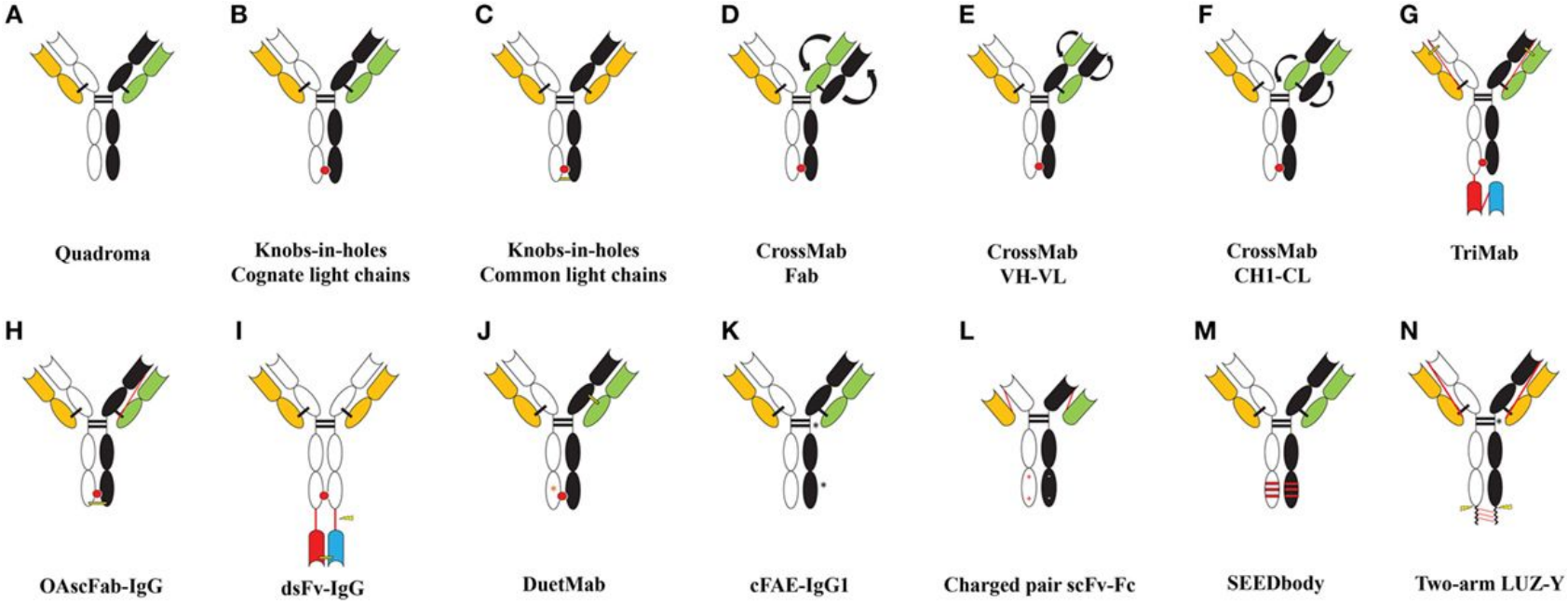


In-class activity

Engineer bispecificity



There are multiple ways to engineer bispecificity



Front. Immunol., 26 January **2017**

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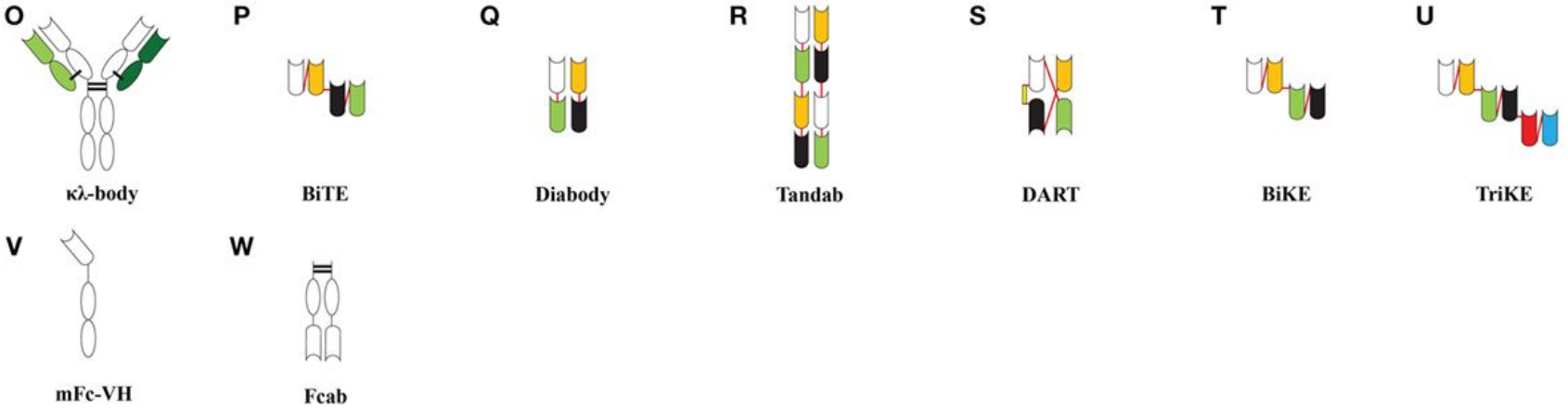
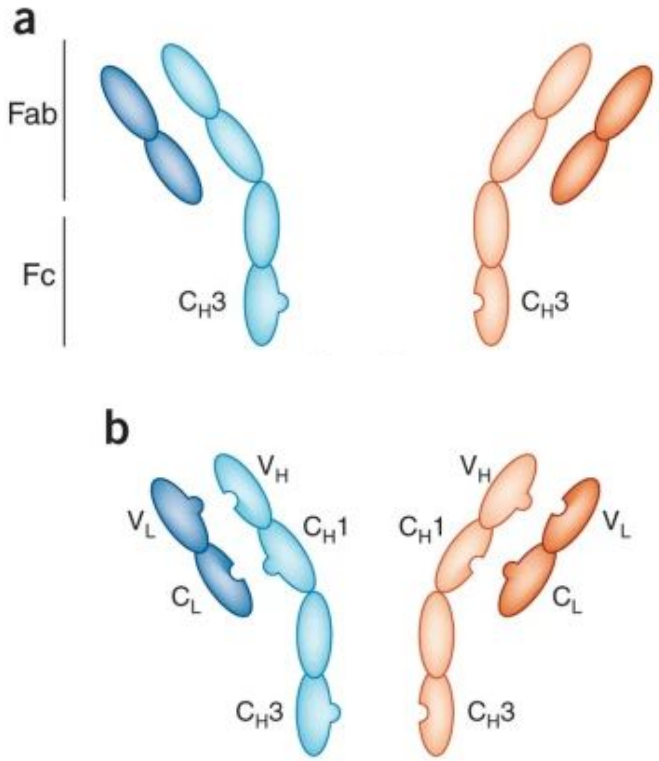


Simple strategies can result in huge impact



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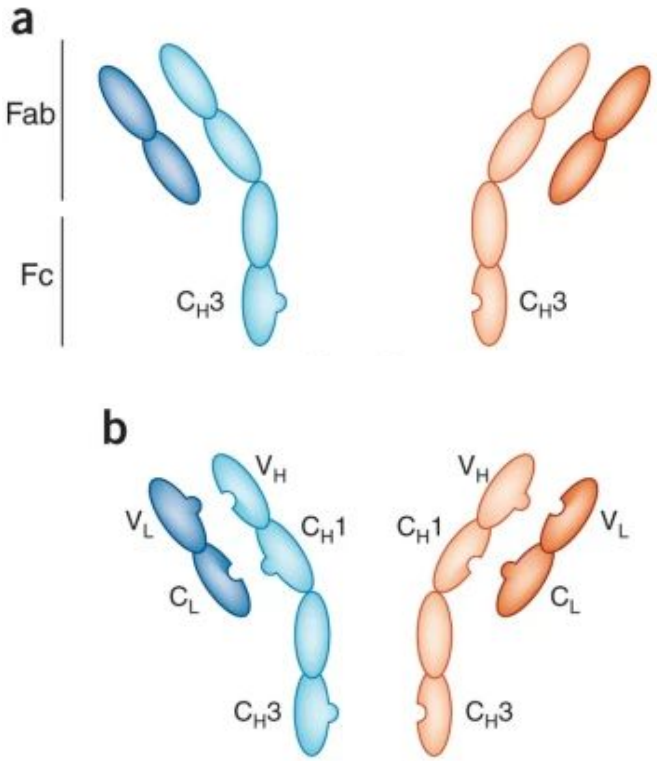
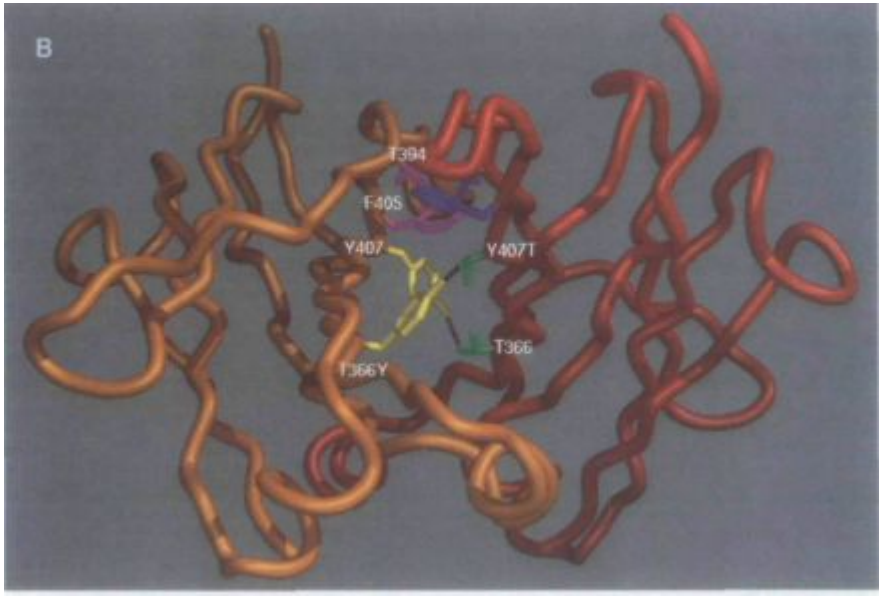
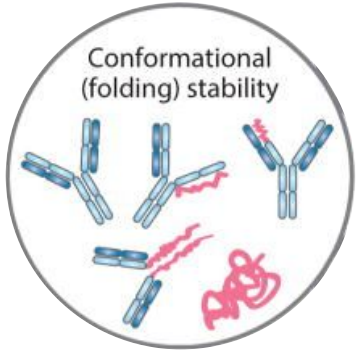




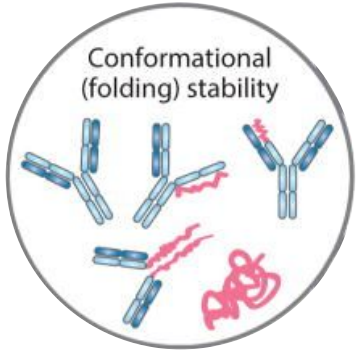
(original idea, Crick 1952)

Enhancing stability of antibodies

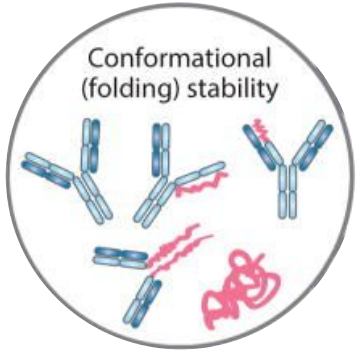


Enhancing stability of antibodies

Grafting => can cause instability in folding



Enhancing stability of antibodies

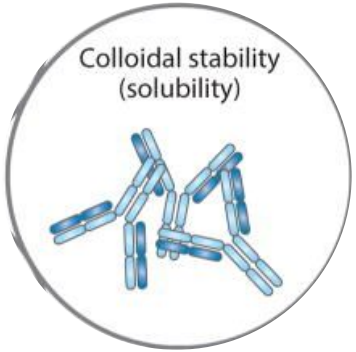


Grafting => can cause instability in folding

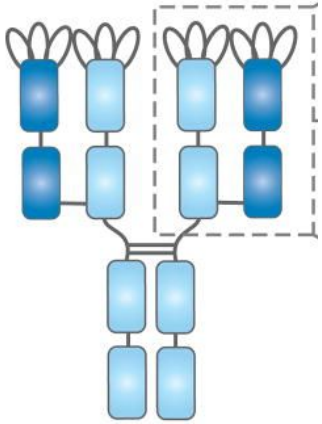
Stabilization:

- Knowledge-based
- Statistical
- Structure-based

Enhancing solubility of antibodies

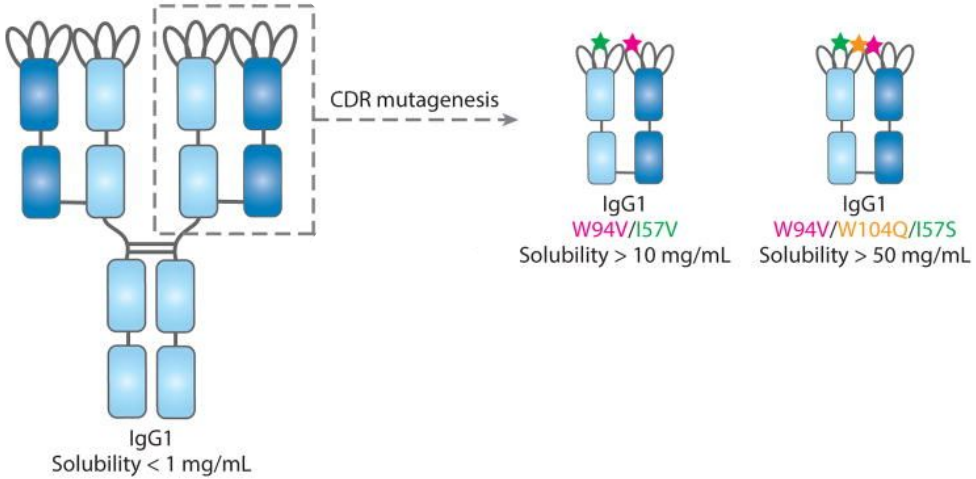


Enhancing solubility of antibodies

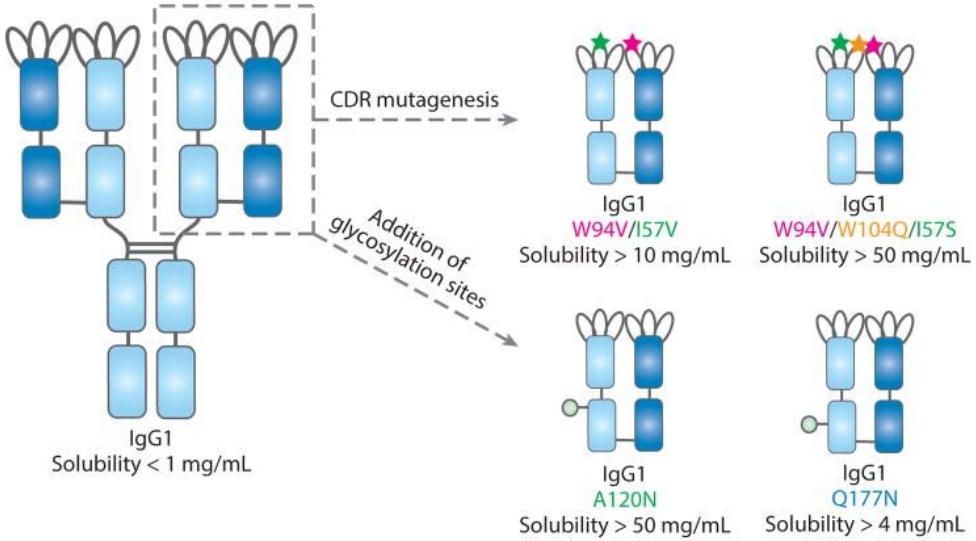


IgG1
Solubility < 1 mg/mL

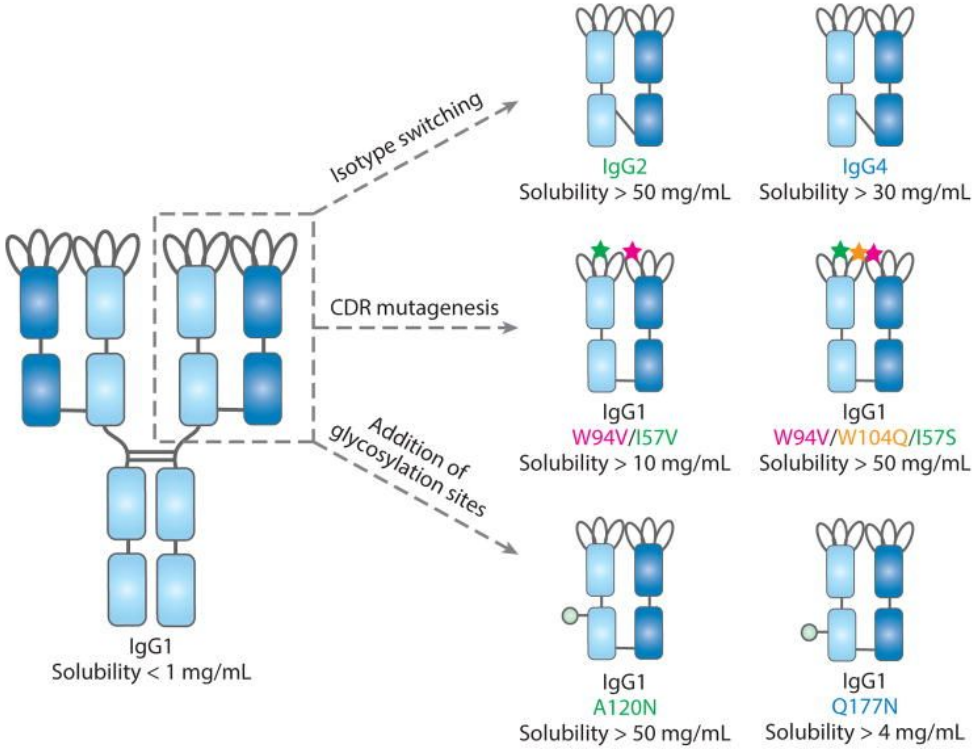
Enhancing solubility of antibodies



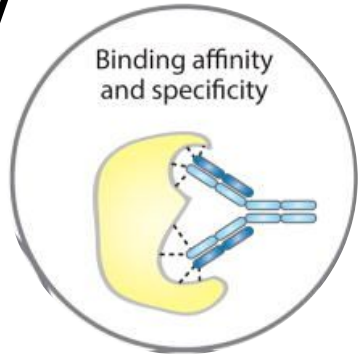
Enhancing solubility of antibodies



Enhancing solubility of antibodies



Engineering binding affinity and specificity



Engineering binding affinity and specificity

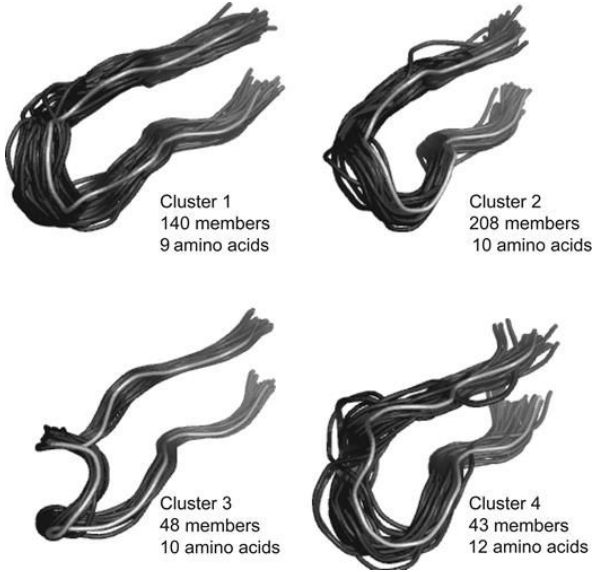
Holy grail:

Predict what sequences bind best

Engineering binding affinity and specificity

Holy grail:

Predict what sequences bind best

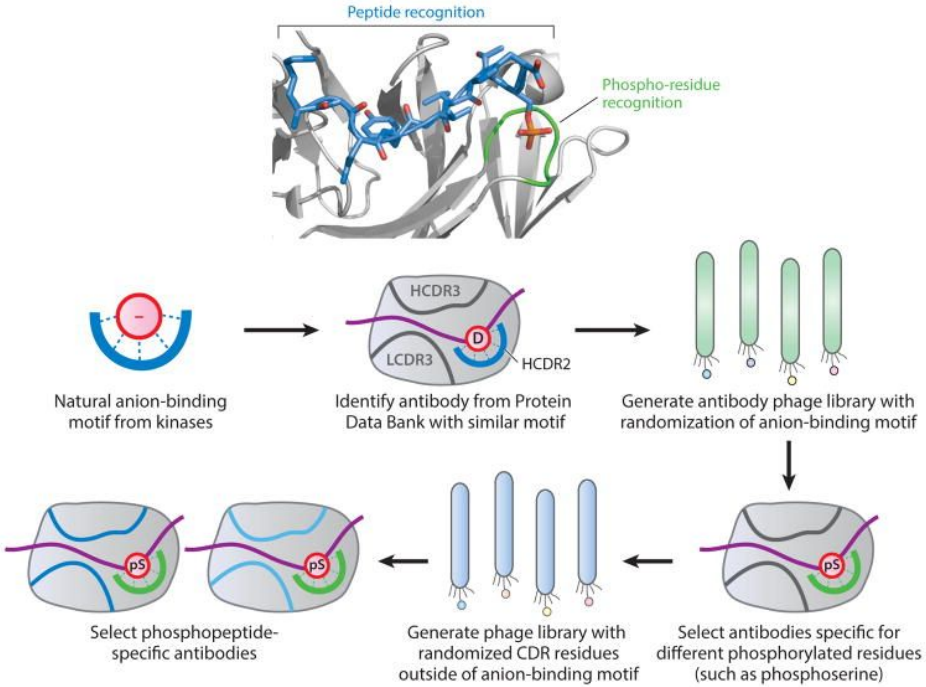


Engineering binding affinity and specificity

Holy grail:

Predict what sequences bind best

Grafting known binding motifs



Nature Biotech, **2018**, 31, 916-921

Engineering binding affinity and specificity

Holy grail:

- Predict what sequences bind best

Grafting known binding motifs

Mostly optimizing current ones

- Optimizing electrostatic interactions
- Insight from different variants

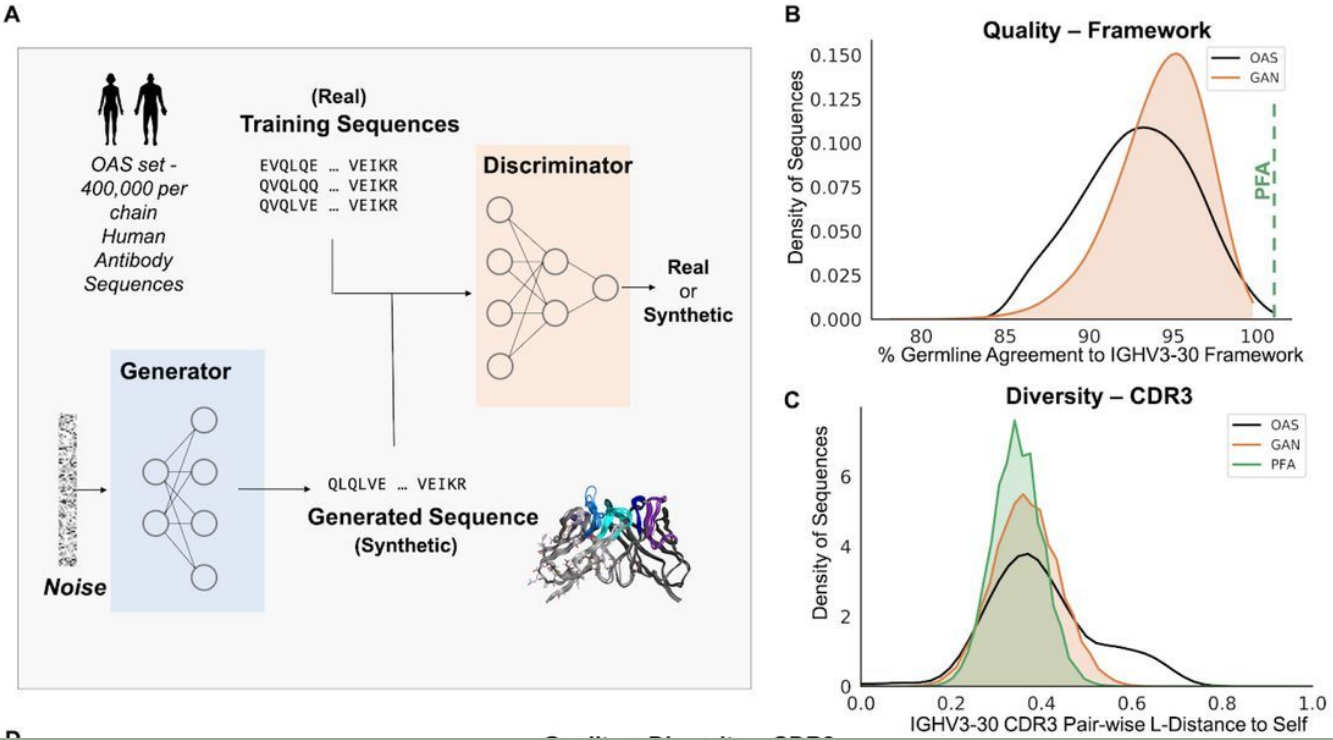
Antibody design is the challenge of finding a needle in a haystack

In-class activity

Finding needle in haystack

Image from medium, finding needle in a haystack

Learning methods can be very helpful to better search the sequence space



Amimeur et al, bioRxive **2020**

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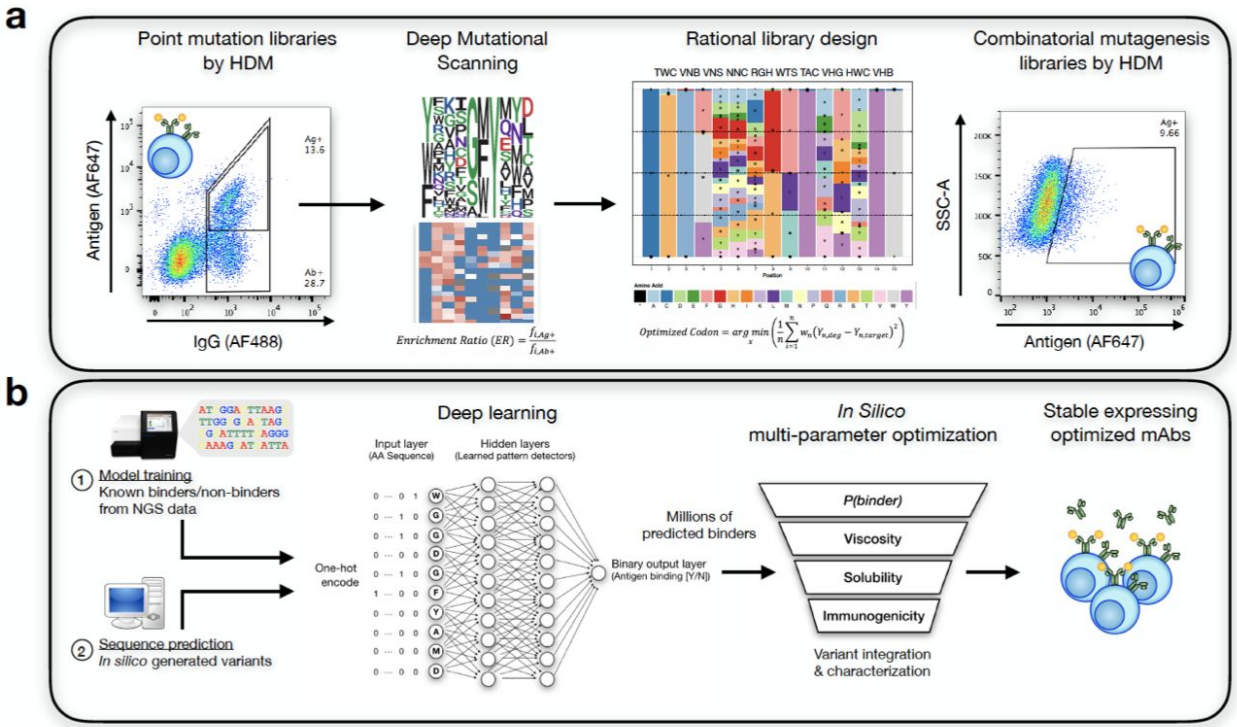

For the next lecture:

1. Make sure you submit your final specific aims page

2. JC ranking and reporting

Next lecture:

Entrepreneurship in protein engineering