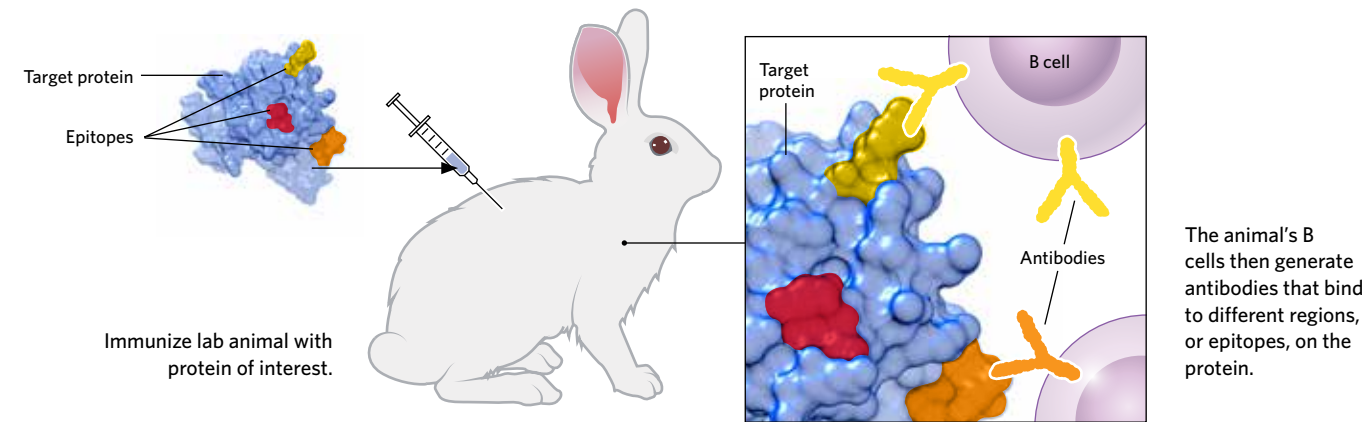


BUILDING BETTER REAGENTS

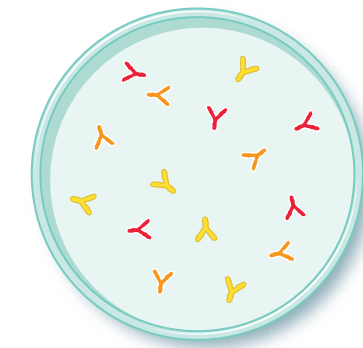
Antibody therapeutics represents the fastest growing sector of pharmaceutical sales, with 47 monoclonal antibodies currently on the market and 300 more in clinical trials. But facing problems of inconsistent, time-consuming, and costly antibody production, some researchers are turning to alternatives—nucleic acid aptamers and protein scaffolds—to target specific proteins of interest, in the lab and in the clinic.

ANTIBODIES



POLYCLONAL ANTIBODIES

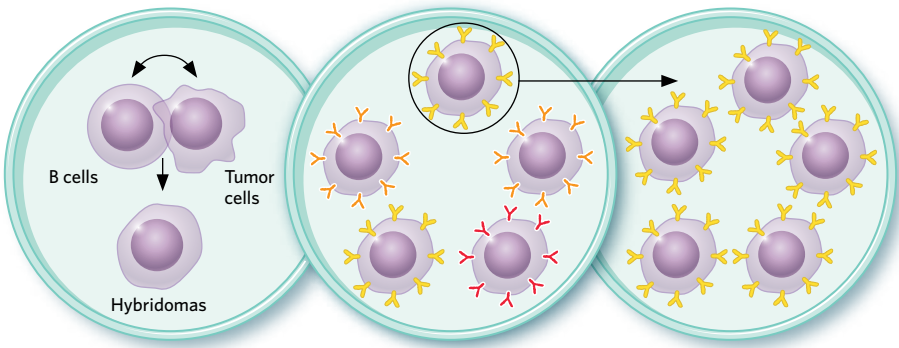
The diverse antibodies that bind to the target protein's numerous epitopes can then be isolated and purified for use.



- **Size:** Large (about 150 kDa)
- **Binds specific epitope?:** Typically no. Diverse antibodies against different epitopes, making them less sensitive to antigen changes than monoclonal antibodies. Antibodies will also vary in affinity and specificity for a given target.
- **Production:** 2–4 months; entirely in animal models
- **Lot-to-lot heterogeneity:** High
- **Shelf life:** Limited

MONOCLONAL ANTIBODIES

Alternatively, the immunized animals' B cells can be isolated from the spleen or lymph nodes and fused with a tumor cell to generate immortal hybridoma lines. Those cell lines that produce the desired antibody against a specific epitope of the target protein can then be grown in large bioreactors to scale up production of the antibody.

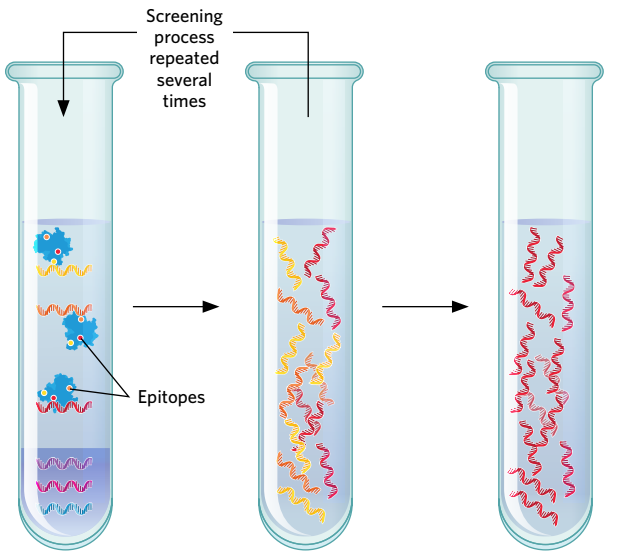


- **Size:** Large (about 150 kDa)
- **Binds specific epitope?:** Yes. As they offer specific recognition of a single epitope on the target protein, monoclonal antibodies are sensitive to molecular changes of that epitope and offer precise molecular recognition of a group of structurally similar molecules.
- **Production:** Six months; requires animal models and the use of expensive cell cultures of higher eukaryotes for growth in bioreactors of up to 2,000 L
- **Lot-to-lot heterogeneity:** Low, though downstream production processes and drift in the cell line's antibody expression can introduce variation
- **Shelf life:** Limited

ANTIBODY ALTERNATIVES

NUCLEIC ACID APTAMERS

Aptamers are short molecules of single-stranded DNA or RNA, typically less than 100 nucleotides in length, that form specific 3-D structures capable of binding target proteins.

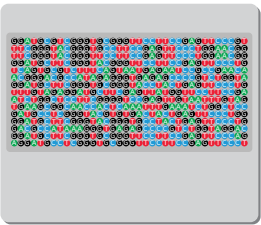


Screen a target protein against a library of nucleic acids. Filter for bound reagents.

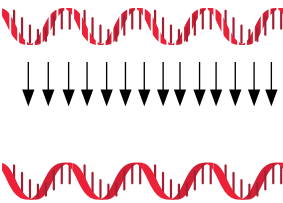
Amplify remaining aptamers with PCR, and screen again.

After many rounds of screening, an aptamer that efficiently binds a single epitope is chosen.

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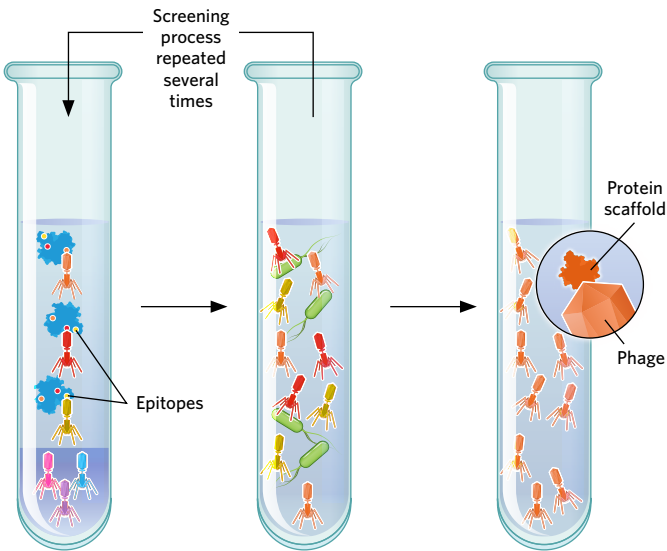
Sequence and chemically synthesize the chosen aptamer.



- **Size:** Small (<25 kDa), opening up new targets that were previously inaccessible to antibodies
- **Binds specific epitope?:** Yes
- **Production:** Weeks; chemically synthesized
- **Lot-to-lot heterogeneity:** Very low
- **Shelf life:** Stable at room temperature for months

PROTEIN SCAFFOLDS

Protein scaffolds, formed from polypeptide fragments or whole proteins, have similarly specific interactions with desired target molecules.

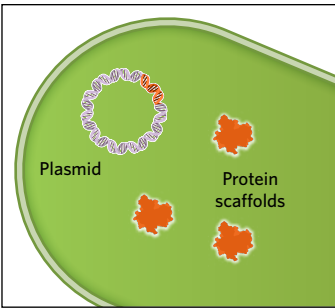


Screen a target against a library of protein scaffolds presented on the surface of bacteriophages. Filter for bound reagents and rinse away unbound ones.

Infect *E. coli* with bacteriophage carrying positive binders to amplify a new, enriched library and screen again.

After many rounds of screening, a scaffold that efficiently binds a single epitope is chosen.

Introduce a plasmid encoding the desired scaffold into *E. coli* to scale up production.



- **Size:** Small (~15 kDa), opening up new targets that were previously inaccessible to antibodies
- **Binds specific epitope?:** Yes
- **Production:** Weeks; entirely in vitro with lower organisms such as bacteria
- **Lot-to-lot heterogeneity:** Very low
- **Shelf life:** Stable at room temperature for months