Phage Display Peptide Library Guide

Phage Display Technology Platfor
ALPHA LIFETECH INC.

1. Current peptide screening approaches

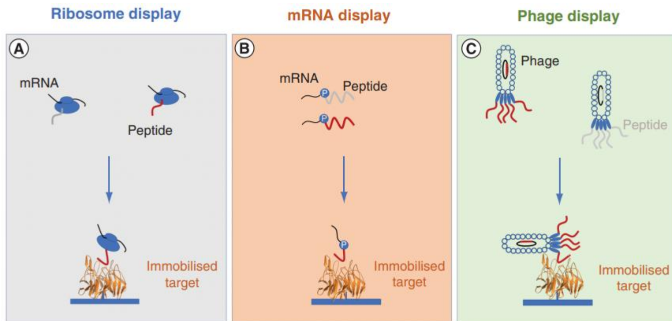

Figure 1. Examples of common peptide library screening approaches. (A) Ribosome display, (B) mRNA display, (C) phage display

(1) **Genetically encoded peptide screening approaches**

The method often screen the library against an immobilized target and are able to provide a means to link a selected peptide that works through physically connecting the peptides being screened in the assay to their coding mRNA to the DNA that encoded it, to enable detection.

A. ribosome display, where the coding mRNA lacks a stop codon, ensuring that the mRNA remains linked to the translated peptide in complex with the ribosome during screening.

B. mRNA display , whereby the peptide is covalently linked to its coding mRNA via a puromycin molecule.

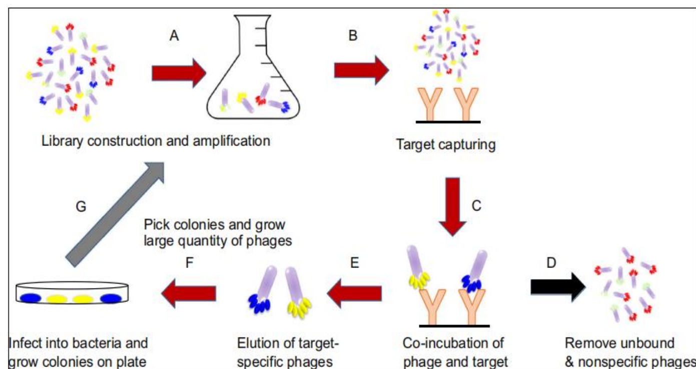

Figure 2. The general scheme of phage display technique and biopanning selection of high affinity peptide.

(2) **Phage display**

Phage display technology is a powerful tool that can be used to select binding partners from a complex library, which takes advantage of the direct connection between phage-displayed peptides and the nucleic acid encoding them. The goal of it is to separate the peptides with high affinity and specificity for the target by conducting repeated affinity selection. This method has been widely applied to rapidly screen of new drugs or lead compounds such as biologically active peptides, proteins and receptors.

Phage-displayed random peptide libraries enable functional access to the peptides and provide a physical link between phenotype (the displayed peptide) and genotype (the encoding DNA); these libraries lend themselves to a screening process in which binding clones are separated from nonbinding clones by affinity purification. Peptides binding to individual targets can be identified by affinity selection (called biopanning). For biopanning, a display library is incubated with an immobilized target, followed by extensive washing to remove nonreacting phages. Binders are usually eluted using acid or high salt and are enriched by amplification in the appropriate host cells. Three to five rounds of biopanning are usually performed in order to obtain targets that bind with high affinity (Fig 2). The primary structure of the peptide can then be determined by sequencing the DNA of individual clones. Using this approach, it is easy to identify peptides that bind specifically to target molecules.

2. Phage display based biopanning consists of five screening steps for selection of peptides.

1. library construction & amplification” where polypeptide-displayed phage libraries were constructed via cloning of combinatorial DNA sequence. This library will be amplified prior to biopanning.
2. The second step is the “target capturing step”, in which the phage library is incubated with target molecule for a specific time to allow binding.
3. The third step is to “remove unbound & nonspecific phages” by using repetitive washing to remove any unbound and non-target specific phages.
4. The fourth step is the “elution step”, in which target-bound phages are separated after a short incubation with low pH buffer or by competitive elution.
5. The fifth step “infection stage”, the eluted phages are infected in bacteria to amplify selected phages, making a new and more selective phage library that should be applied in a next round of biopanning.

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