

## Expanding phage display technology with luminescence

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The -omics Era has contributed to the disclosure of genes coding for known and novel biologically active peptides. The interaction of those peptides with desired targets can be studied using phage display, mostly using phagemids as vectors. However, complex and time-consuming procedures are usually needed for purification and validation of desired phages. So, a vector that would present the capacity to easily confirm and detect a certain function would be a step forward in display technology.

The present work demonstrates the manipulation of a lytic bacteriophage to simultaneously display a luminescent protein and a selected peptide on its capsid in order to function as a confirmation vector. For this purpose, the compact luminescent protein luciferase NanoLuc® from Promega was genetically inserted in the T7 genome using the Bacteriophage Recombineering of Electroporated DNA (BRED) technique. Additionally, a peptide-encoding DNA sequence, identified using phage display against cancer cells MDA-MB-231, was fused to the T7 10B gene capsid protein. For 10B protein synthesis, translationally biased hosts were selected. *Escherichia coli* BL21 – an F- episome host – was used since F+ (*E.coli* W1485) and F' (*E.coli* JM109) hosts prevent 90% of capsid protein synthesis. This codon selective pressure imposed allowed the variation of the 10A and 10B capsid proteins ratio (T7 capsid complex), changing the luminescent protein and peptide copies. Capsid stoichiometry analysis, ELISA binding tests and highly sensible photonic microscopy were performed to confirm the binding and sensibility of the virion signal reporting system.

This study shows that a manipulated lytic luminescent bacteriophage can be a simpler and faster alternative vector for any peptide display compared to the widely used phagemids. We expect that these kind of vectors will play an important role in fields as molecular recognition and imaging.