# Next-Gen Biosensors: Aptamers and SPRi for Unmatched Binding Evaluation

## Excellence in Research: Stimuli-Responsive Exosomes for Quantitative Biosensing

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## PROJECT BACKGROUND

Aptamers are short, synthetically derived ligands that take advantage of oligonucleotides' diverse combinatorial capabilities to achieve strong binding affinity with a high degree of specificity toward a target of interest [1, 2, 3]. Conventional research-grade antibodies often show significant batch-to-batch and/or brand-to-brand variability with some studies stating that as much as 51% of available product cannot be successfully validated [4]. Unlike conventional antibodies, aptamers are sequence-defined and chemically synthesized [4]. This makes them highly consistent and easy to modify when compared to antibodies. Aptamers also have fewer target limitations than conventional antibodies and can be generated to target toxic small molecules, non-immunogenic targets, and even single molecules.

The ability to design and readily produce highly specific and selective aptamers for a broad range of targets makes them a promising tool in the areas of diagnostic and therapeutic research, especially in areas related to the identification of biomarkers. However, for aptamer-based biosensors to become utilized on a broader scale, the binding affinity must be evaluated to confirm that a high degree of both specificity and selectivity are being achieved. While there are many methods of evaluating the binding, Surface plasmon resonance imaging (SPRi) is the method that we chose to quantitatively evaluate the binding events occurring between our aptamers and chosen targets. This method will provide a snapshot of each individual binding event, allowing us to strictly vet the accuracy of our aptamers before utilizing them as targets for in future biosensor applications.
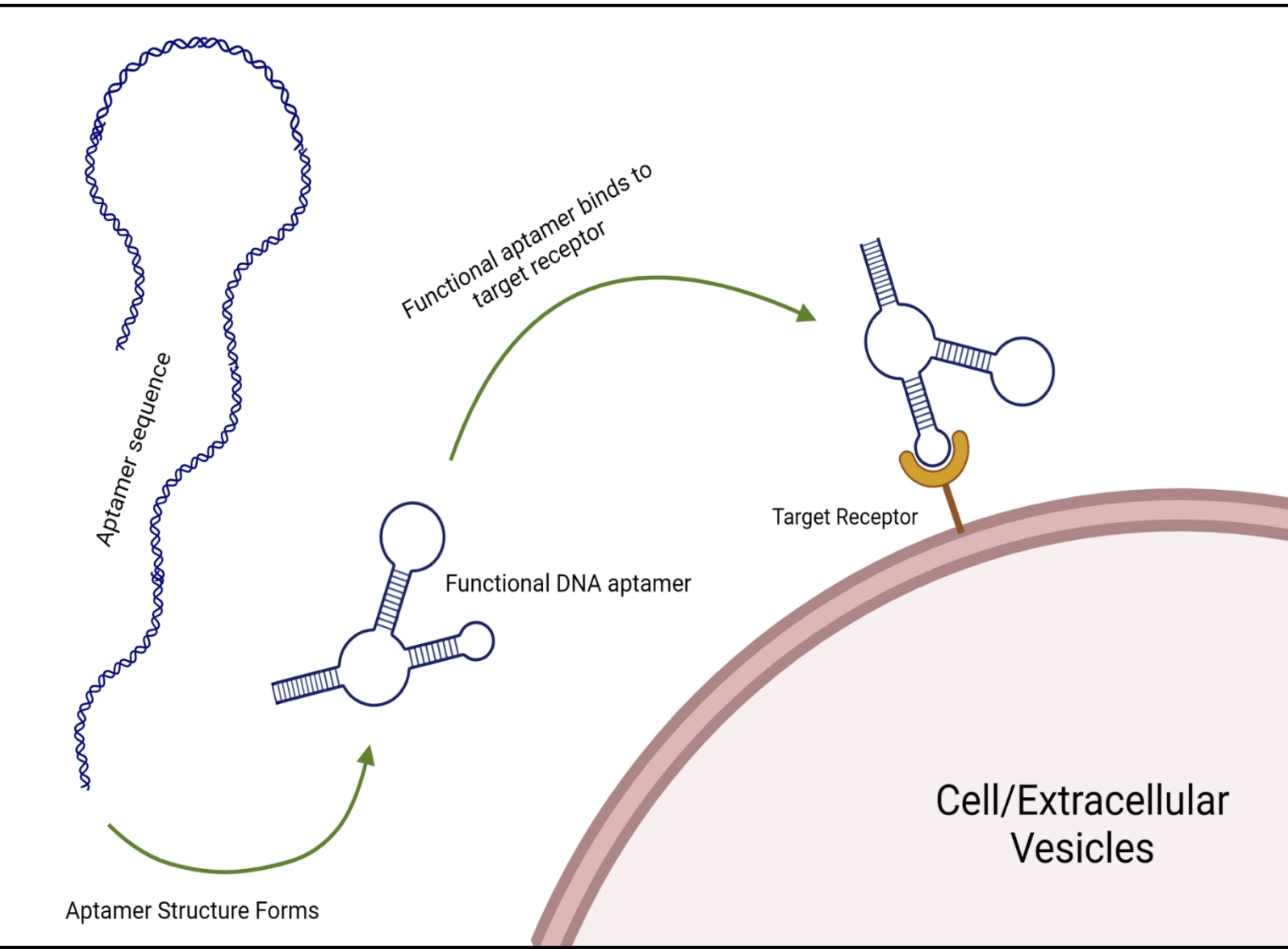
**Figure 1.** Diagram illustrating the process of aptamer attachment to a target receptor. Aptamers are initially linear ssDNA that fold into specific orientations due to the positioning of certain groups along the strand. The structure of an aptamer allows it to achieve strong and specific binding to a desired target. Created with BioRender.com
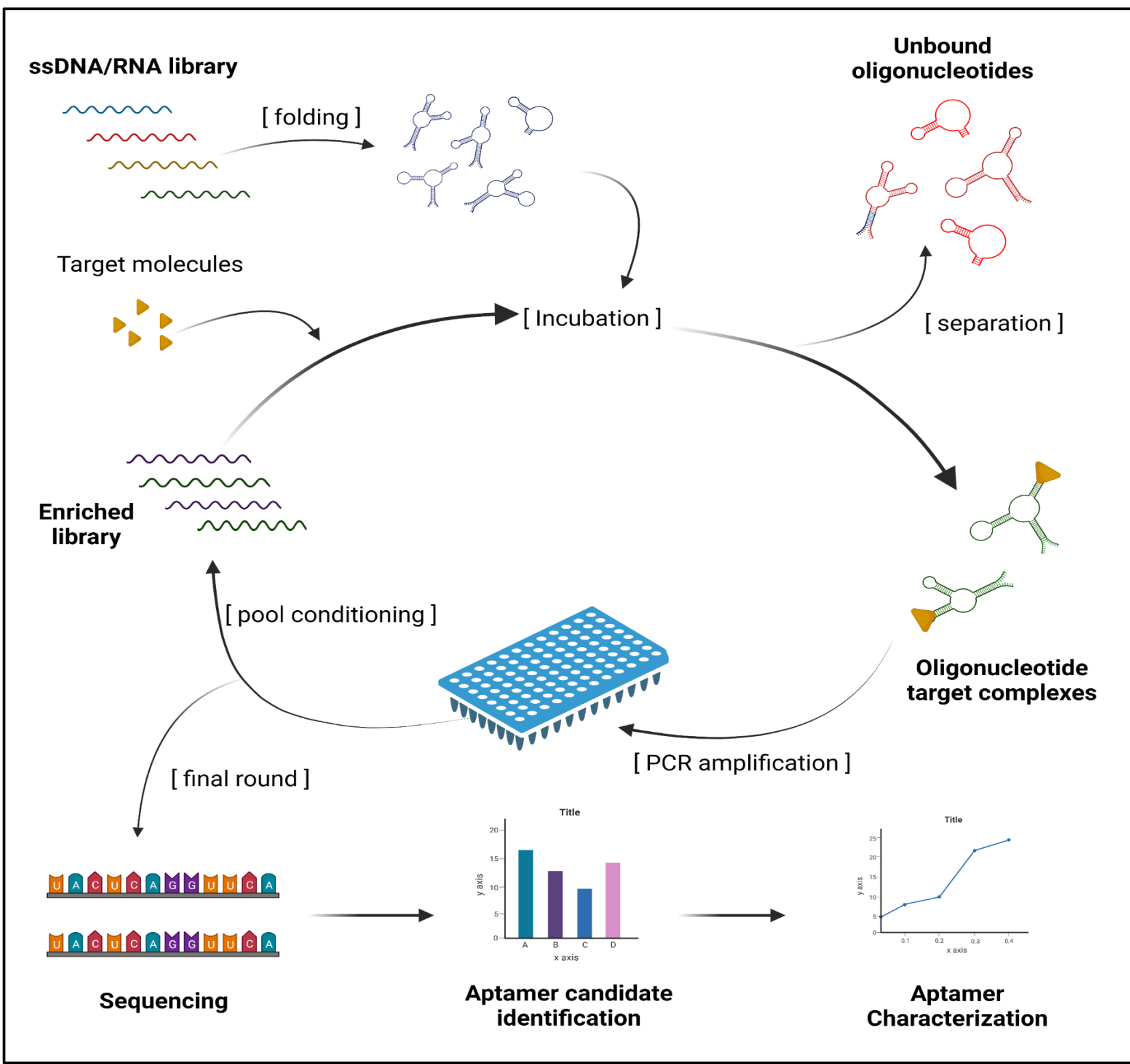
## MATERIALS & METHODS

**Figure 2.** Schematic of SELEX process for the synthesis of aptamers. Created with BioRender.com.
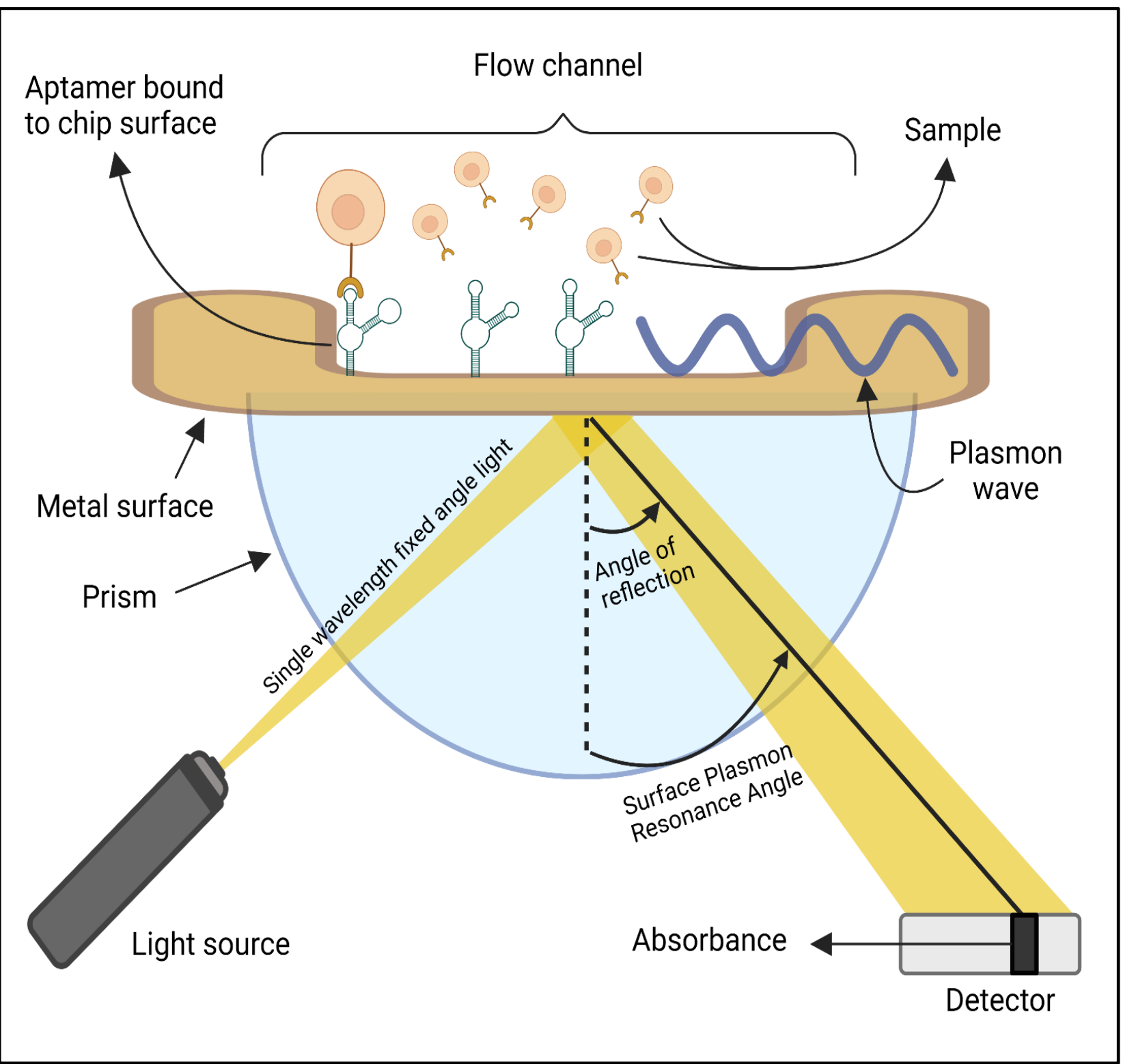
**Figure 3.** Schematic of SPRi mechanism. Created with BioRender.com

## APPLICATIONS

Biosensing, Bioimaging, Diagnostics

Therapeutic Tools, Drug Delivery

Analytical Reagents, Hazard Detection, Food Inspection

## FUTURE DIRECTIONS

The sensitivity of aptamers, combined with the ability to synthesize them consistently and more cheaply, makes them ideal candidates for future research in the diagnostic space. They will provide the ability to produce high volumes of selective tags for wide-ranging molecular and cellular targets. These characteristics also make them very promising, especially in areas related to biomarker research.

With this research, we aim to enable further utilization of aptamers as targets for new isolation, purification, and analysis of disease biomarkers. Such new methods will aid efforts to improve early methods of detection for a plethora of diseases that are difficult to diagnose.

## EXPERIMENTAL FACTORS

### OBJECTIVE 1

**Identify cell lines with surface markers of interest and use coding sequences to create aptamers.**
To design aptamers that target specific cell types with a high degree of selectivity and affinity, our first step is to identify the surface receptors and then identify the appropriate coding sequence that will target these specific surface molecules.

### OBJECTIVE 2

**Test the binding affinity of aptamers using SPRi.**
The first phase of evaluation will focus on single populations of cells against the aptamers developed to target a specific target on their surface. From these experiments we will ascertain the precise quantitative measure of each aptamer's affinity.

### OBJECTIVE 3

**Test the selectivity and affinity of aptamers concurrently using SPRi.**
The second phase of evaluation will test the aptamers against a mixed population of cells, where only some of the population has the specific surface targets. From these experiments we will gather a more precise evaluation of the selectivity in combination with the affinity.

| CELL TYPE | SURFACE RECEPTOR | DISEASE RELEVANCE |
|---|---|---|
| Neuronal cells | L1CAM, APLP1, ATP1A3, | Neurodegenerative |
| Mammary epithelial cells | ER-α, HER-2 | Breast cancer |

**Table 1.** Reference cell lines of interest and the receptors they carry.

## ACKNOWLEDGEMENTS

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