A HIGH-THROUGHPUT CRISPRi SCREENING PLATFORM TO UNRAVEL FUNCTIONAL LONG NON-CODING RNAS

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INTRODUCTION

- Long non-coding RNAs (lncRNAs) have been demonstrated to regulate various cellular processes. Only a minority of the vast repertoire of annotated lncRNAs has been functionally characterized.
- A significant gap still persists in developing models to predict their functions based on inherent features.
- This highlights the need for systematic identification of functional lncRNAs through high-throughput screening (HTS) and the exploration of potential links between their features and functions.
- Our work primarily focuses on identifying functional lncRNAs and integrating their features to establish a foundation for developing lncRNA functional prediction models.

OBJECTIVE

Establish a high-throughput CRISPRi arrayed screening platform that is applicable across diverse cell types and cellular conditions to facilitate the systematic identification of functional lncRNAs.
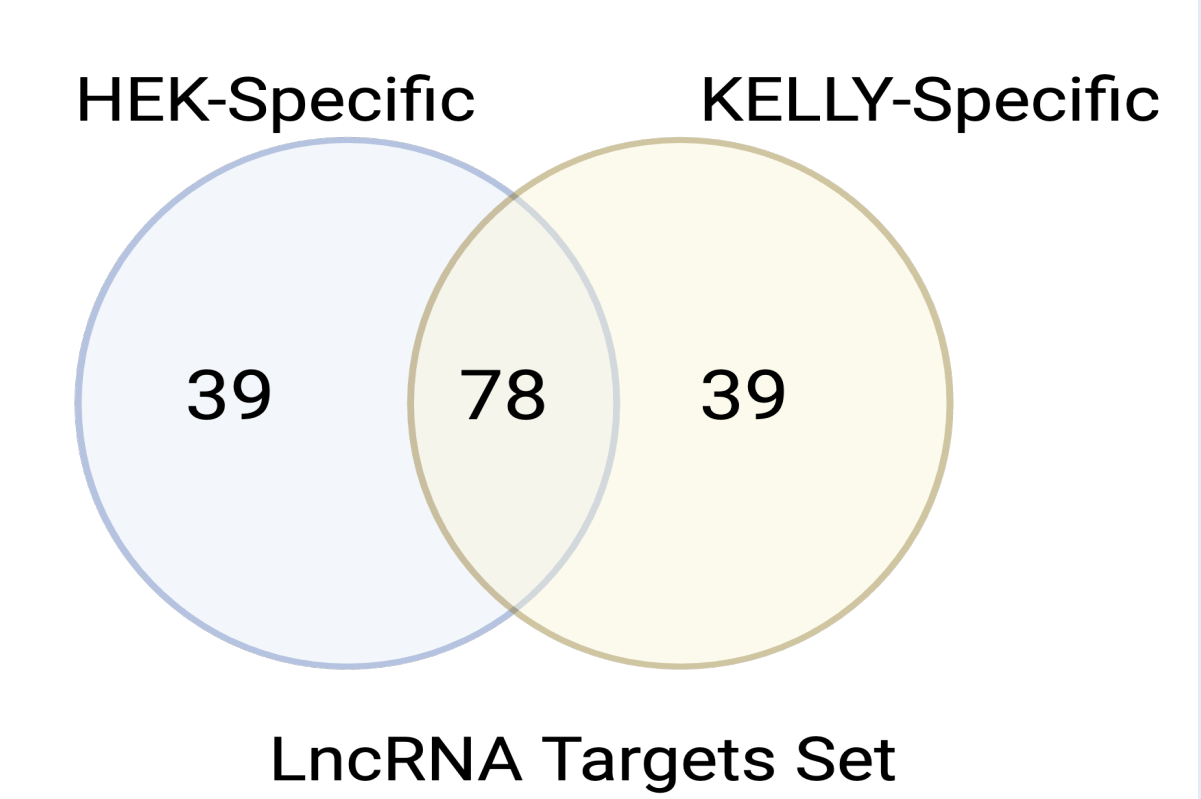
METHODS
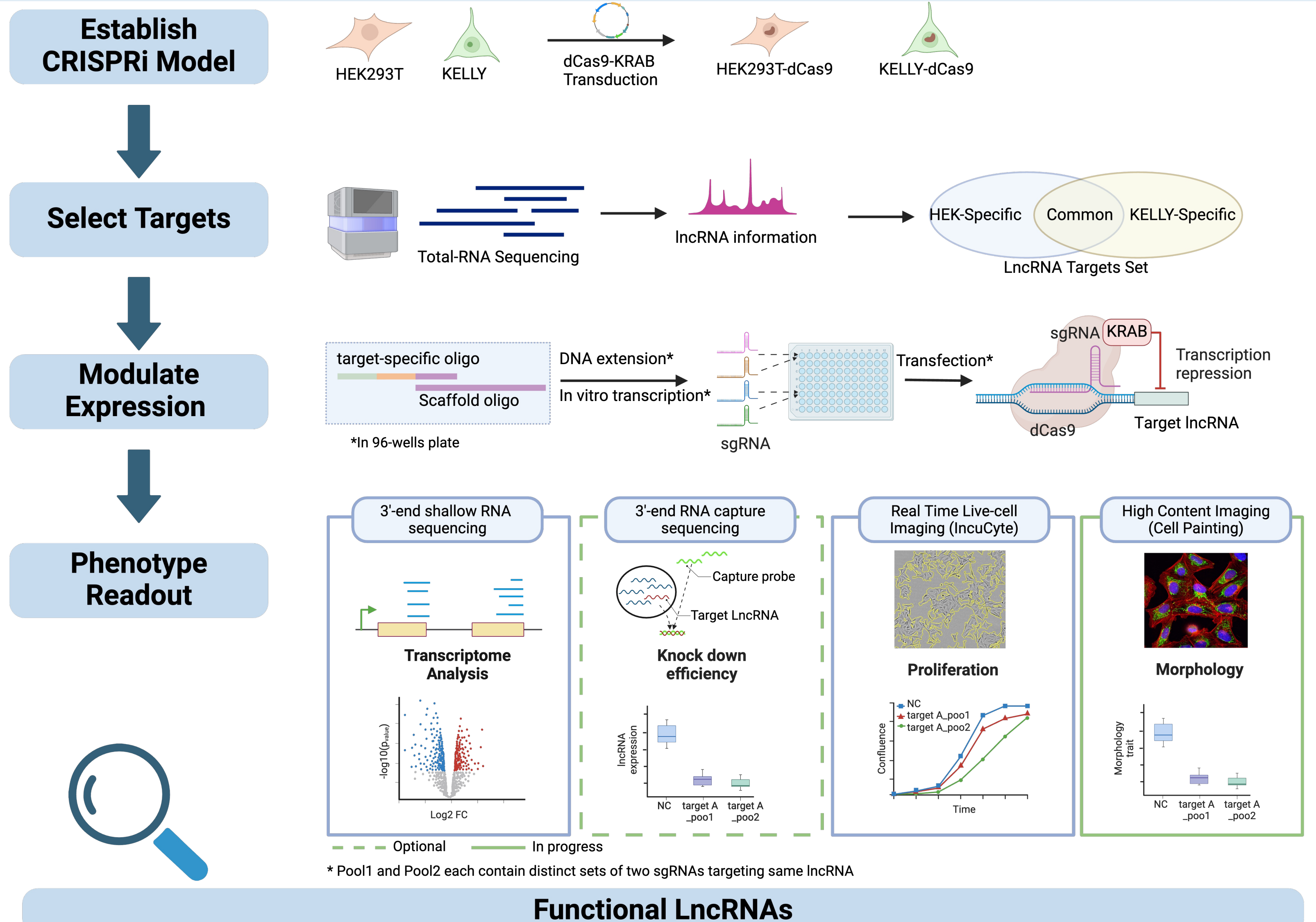
RESULTS

Figure 1. Cell-type specificity of selected lncRNAs

Figure 2. Molecular and cellular phenotype of lncRNA RP11-14O19.2
(A) Volcano plots representing differentially expressed genes upon lncRNA RP11-14O19.2 knockdown with two different sgRNA pools. (B) Bubble plot for Hallmark gene set enrichment analysis results. (C) Cell confluence measured by IncuCyte, comparing RP11-14O19.2 sgRNAs with the non-targeting sgRNA group. (D) Images obtained from Cell Painting.
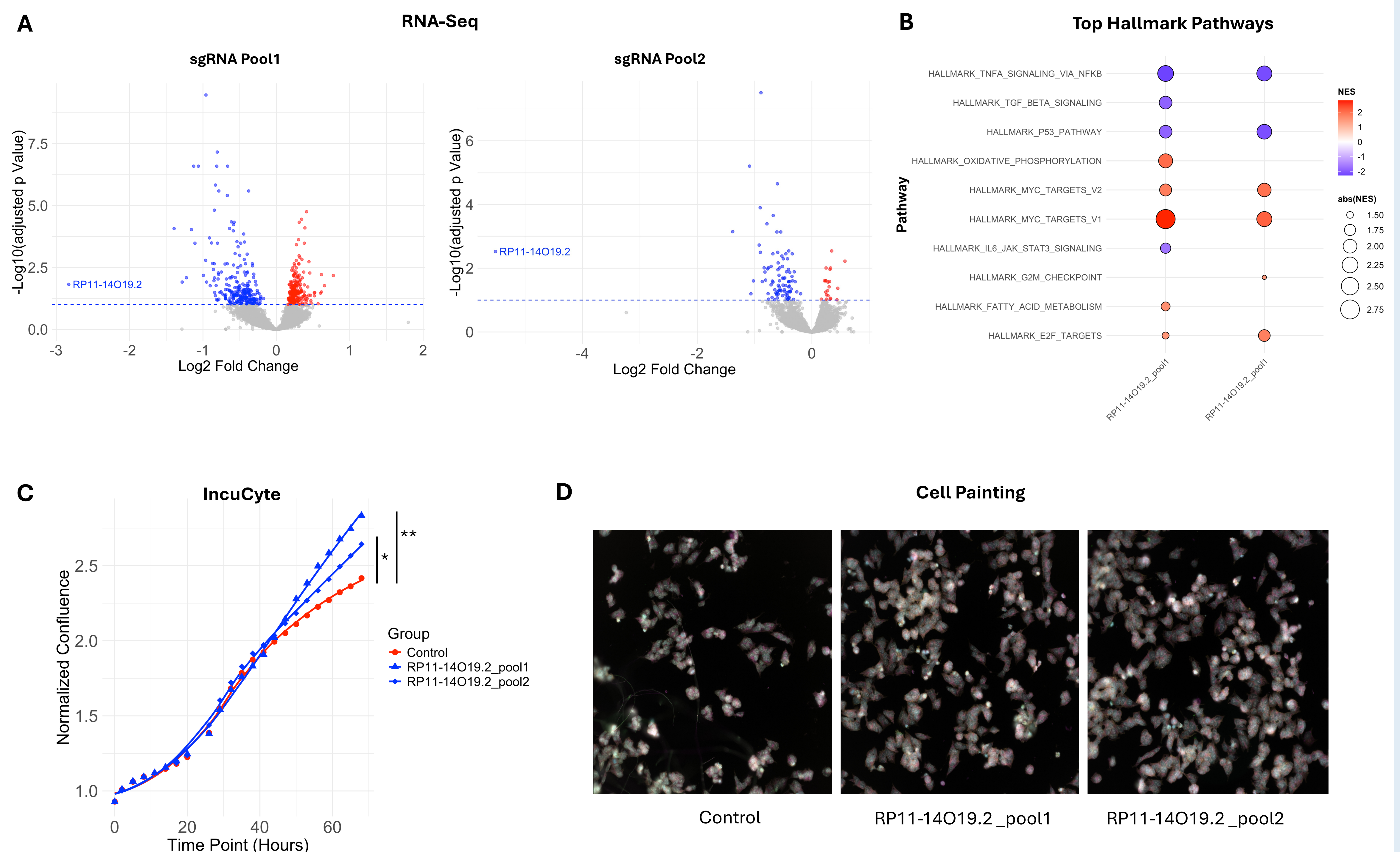
PERSPECTIVES
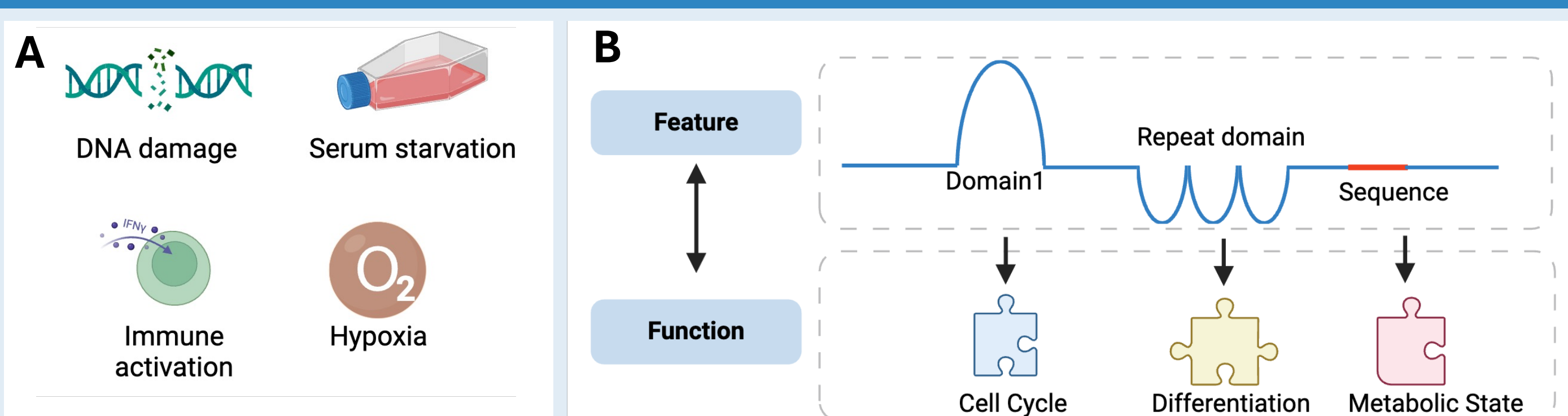
A. We aim to intergrade multiple perturbations into the platform to explore functional lncRNAs across various cellular states.

B. Further analysis of the sequence and structural features of the functional lncRNAs identified in this screen may uncover structure-function relationships and contribute to the development of a functional prediction model.

CONCLUSION

- This platform enables potent knockdown of lncRNAs expression.
- By integrating multiple high-throughput phenotypic readouts, this platform enables the systematic identification of functional lncRNAs across diverse biological processes.
- This platform is applicable across various cell types and under different cellular conditions, facilitating the exploration of cell-state- and cell-type-specific lncRNA functionality.

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