



NYU



# CARPOOL: A High Throughput Drug Screening Platform

**High throughput screening platform without the need for complex instrumentation or expensive consumables and compatible dead cell removal approach.**

**Technology ID**

SUL02-02

**Category**

Life Sciences/Research tools/Drug Development

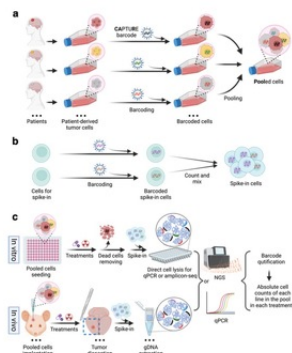
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**Figure. Workflow of CARPOOL for high throughput therapeutic screening in vitro and in vivo with patient-derived glioblastoma spheroid cultures (GSCs).** (a) Preparation of barcoded cells for therapeutic screening. Each patient-derived GSC was tagged by a unique DNA barcode, and the barcoded GSCs were pooled for treatment as a pool. (b) Preparation of spike-in cells to generate standard curve for absolute cell counting. Additional cell lines were labelled with different barcodes, and a series of known counts of different barcodes were mixed as spike-in cells. (c) Therapeutic screening workflow in vitro and in vivo. For in vitro, pooled GSCs were seeded in 96-well plates for treatment. Non-viable cells were removed by PBS induced live cell attachment. On spike-in cells were loaded into each well before cell lysis. The barcodes counts were accessed by amplicon NGS or amplicon qPCR, and spike-in cells counts were used to obtain absolute cell counts for each GSCs. For in vivo, the pooled GSCs were intracranially implanted followed by treatments. The quarter brains with GSCs implantation were harvested, and spike-in cells were added before genome DNA (gDNA) extraction. The barcodes counts were accessed by amplicon NGS or amplicon qPCR, and spike-in cells counts were used to obtain absolute cell counts for each GSCs.



## Technology

CARPOOL revolutionizes high-throughput therapeutic screening by employing individually tagged cell lines using lentiviral barcodes from the CAPTURE library. This innovative method facilitates accurate and parallel interrogation of numerous cell lines for various applications, including drug and radiation sensitivity screening, growth condition surveys, and combinatorial evaluations. Barcode counts pre- and post-treatment are determined via next-generation sequencing (NGS) or quantitative PCR (qPCR), with spike-in controls used to obtain absolute cell counts. (See Figure 1) Compatibility with both NGS and qPCR ensures versatility, with NGS preferred for enhanced efficiency. CARPOOL has been validated *in vitro* and *in vivo* for therapeutic screening with patient-derived glioblastoma spheroid cell lines (GSCs), demonstrating its efficacy in advancing pre-clinical drug screening.

## Background

Current pre-clinical therapy evaluation often relies on a limited number of cell models due to the cost and time constraints of conducting experiments across numerous models and conditions. This approach overlooks the genomic diversity seen in diseases like cancer, contributing to high failure rates in drug development. CARPOOL addresses this gap by enabling high-throughput therapeutic screening across a pool of cell lines and by consolidating evaluations, CARPOOL optimizes resource utilization, allowing researchers to study 20 or more cell lines using the resources typically allocated for a single line study, thereby maximizing

coverage of tumor heterogeneity and enhancing pre-clinical drug screening efficiency. This technology is compatible with NGS, which has an estimated global market size of \$8.4 billion in 2023 and is expected to grow at a compound annual growth rate (CAGR) of 21.7% from 2024 to 2030.

## Development Status

This technology has been validated *in vitro* and *in vivo* using malignant brain tumor models. It has been applied to a large drug library as a screening tool. A comprehensive computational workflow has been established for the general analysis of CARPOOL experimental data. The technology is ready for application a wide variety of cell models in any number of diseases.

## Applications

- Preclinical drug screening for therapeutics development.
- Radiation sensitivity screening, particularly for GBM (Glioblastoma).
- Input tool for precision medicine, assessing cell response rates to different drugs based on molecular markers using predictive machine learning models that could lead to drug identification for repurposing.

## Advantages

- **Compatible** for pre-clinical validation in both *in vitro* and *in vivo* settings.
- **High-throughput:** This approach optimizes resources by pooling multiple lines and provides data output equivalent to approximately 20 similar studies in a single study, effectively eliminating variation between wells or mice.
- **High sensitivity:** Ensures high sensitivity by obtaining single-cell count resolution.
- **Fast and efficient:** Accelerates the generation of comprehensive therapeutic datasets.
- **Compatibility:** CARPOOL can be used with both regular NGS service or qPCR.

## Intellectual Property

NYU has filed for a provisional patent covering the compositions and methods of use of CARPOOL.