High-throughput spheroid imaging and formation for using magnetic 3D bioprinting

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High-throughput spheroid imaging and formation for using magnetic 3D bioprinting

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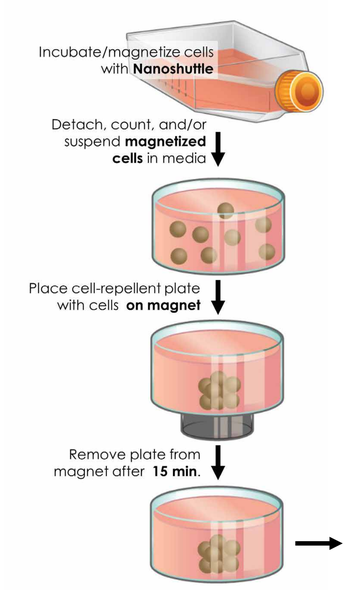
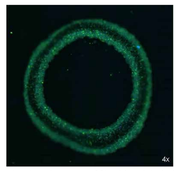
1. Nano3D Biosciences, Inc. Houston, TX USA
2. GE Healthcare, Marlborough, MA, USA

Summary



- 3D cell culture have the potential to recreate native tissue environments
- Current products on the market have technical challenges limiting their use for high-throughput screening.
- Magnetic 3D bioprinting works by magnetizing cells with NanoShuttle (NS), then printing them with magnetic forces in CELLSTAR™ flat-bottom cell-repellent plates
- This platform escapes limitations of others by being reproducible, fast, easy to use, and using flat bottom adaptable for high-content and high-throughput imaging
- 3D cultures can be centered in the middle of the well, while on a flat and high-optical quality surface, for efficient high-throughput and high-resolution imaging

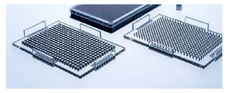


Bioprinted 3D ring of fibroblast labeled with CellTracker™ green and Hoechst nuclei staining. Rings can be easily bioprinted and coupled with real-time imaging for generating would-healing and cell migration models. Rings contract over time corresponding to wound-healing or migration rates of specific cell types.
1. Timm et al. Scientific Reports, October 2013
2. Tseng et al. Scientific Reports, July 2016

Advantages

- Rapid spheroid formation (<24 h)
- Cultures can be easily centered magnetically in the middle of the well
- Unattached spheroids with CELLSTAR™ flat bottom cell-repellent plates
- Magnetized spheroids easy to hold down while removing liquids
- Scalable, up/down, in size from 6- to 1536-well formats
- Print, culture, co-culture, stain, and image spheroids in same plate
- Non-specific to any cell type
- No effect of NS or magnetic field on cell viability and function
- No interference on any endpoint (fluorescence, qRT-PCR, etc)
- No special equipment/media required.

3D in a 2D Workflow

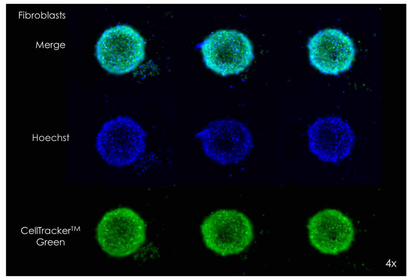


Magnetic 3D bioprinting is the ideal platform for high-throughput 3D culture

384-well bioprinting kit

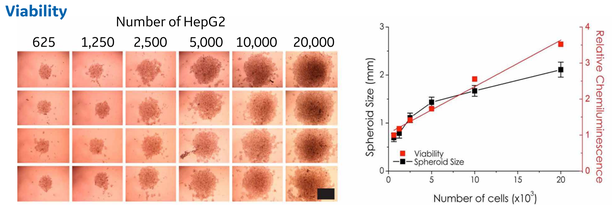
CELLSTAR™ Cell-repellent 384-well plates

Reproducibility & Scalability

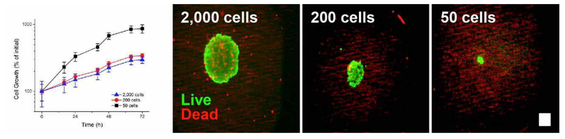


3D fibroblast spheroids after 12 hours of printing (10,000 cells per spheroid).

Viability



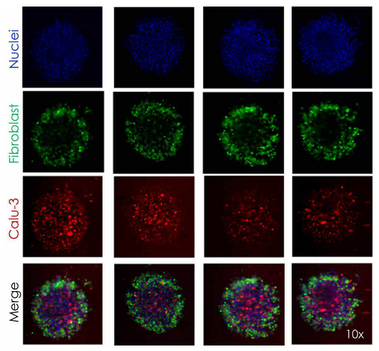
HepG2 spheroids printed at various cell numbers (left), and their size and viability (CellTiter-Glo™, Promega) (right). Scale bar = 500 µm.



Viability of PC3 prostate cancer spheroids of 2,000, 200, and 50 cells measured by RealTime-Glo RealTime-Glo (left) and live/dead staining (right). Scale bar = 250 µm. Tseng et al. Promega Corporation Web site. http://www.promega.com/resources/pubhub/bioprinting-3d-cell-cultures/.

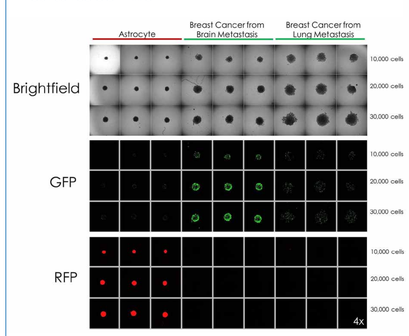
Co-culture

Bioprinted 3D co-cultures of lung adenocarcinoma (Calu-3; CellTracker™ Red) and fibroblasts (CellTracker™ Green) after 16 hours (Hoechst nuclei staining, blue). Cancer cells reproducibly localized inside, while fibroblasts are mostly at the outside of the co-culture.



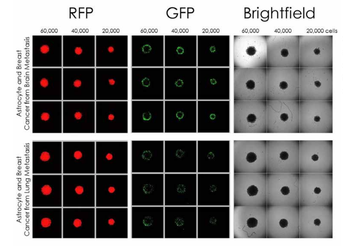
3D Culture and Co-Culture Centered on Flat Surface

Cultures 384-well



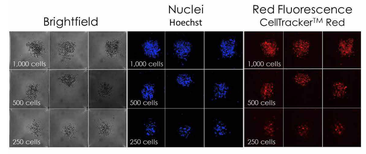
IN Cell Analyzer

Co-cultures 384-well

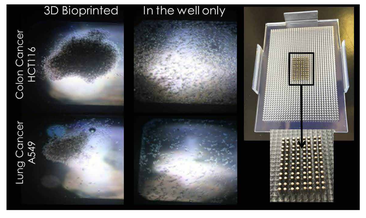


Co-Cultures of bioprinted with varying cell density of astrocytes and breast cancer cells from brain or lung metastatic sites. Cancer cells reproducibly localized outside, while astrocytes are mostly at the center. All cultures positioned at the middle of flat and high-optical quality wells.

1,536-well 3D Bioprinting



1536-Well Spheroids Bioprinted - 15 minutes



Acknowledgements

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References: http://www.n3dbio.com/technical-resources/publications/

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n3D Biosciences, Inc.

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