Electrical & Computer Engineering

D E F E N S E

Louisiana State University

From Photolithography to Resin 3D Printing: Evaluation of Microfabrication Technologies and Development of an Integrated Electromechanical Platform for Zebrafish Egg Orientation in Somatic Cell Nuclear Transfer to Support Genetic Resource Preservation

a dissertation to be defended by

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Abstract—The preservation and utilization of genetic resources from aquatic species are constrained by the limited availability of scalable technologies for processing, manipulating, and cryopreserving gametes and embryos. Although microfluidic and lab-on-a-chip systems can improve the precision, reproducibility, and quality assurance of these procedures, their adoption is often limited by the cost, complexity, and fabrication time associated with conventional photolithography and polydimethylsiloxane soft lithography. This dissertation investigates resin-based 3D printing as an accessible fabrication method for biological microdevices and applies this approach to the development of an integrated platform for three-dimensional orientation of zebrafish eggs for somatic cell nuclear transfer (SCNT).

First, we evaluated the dimensional accuracy, precision, fabrication time, and cost of industrial DLP/SLA and consumer-grade resin printers using microchannels and multiscale test structures, and compared them with conventional microfabrication. The industrial printer produced features ranging from 0.1 to 1.0 mm with dimensional errors below approximately 4%. In contrast, the consumer printer maintained errors below approximately 6% for features between 0.4 and 1.0 mm but exhibited larger deviations at smaller dimensions. Although conventional lithography provided the greatest accuracy for the smallest tested features, industrial resin printing reduced fabrication time from approximately two days to less than 30 min and substantially decreased the per-device material and processing cost. These findings establish the dimensional ranges over which resin printing can provide a practical alternative to conventional microfabrication.

Building on this fabrication assessment, a modular egg orientation device was designed, simulated, fabricated, and experimentally evaluated for zebrafish SCNT. The platform integrates electrically induced in-plane rotation about the vertical axis, motor-driven hydrodynamic out-of-plane rotation about an adjustable shaft axis, a manually rotatable positioning stage, and mechanisms for post-orientation egg stabilization and injection-needle access. A resin-printed four-electrode electrorotation module generated a rotating electric field using four 90° phase-shifted signals. A custom phase-splitter circuit produced these signals from a conventional two-channel function generator, reducing instrumentation requirements and system complexity. Electrorotation was demonstrated using dielectric microspheres, unfertilized zebrafish eggs, and embryos. Stable zebrafish egg rotation was achieved at approximately 16.1 Vpp and 700–900 kHz, with angular velocities of approximately 0.3–0.4 rad s^{-1}. The mechanical module transferred shaft motion to the egg through liquid-mediated hydrodynamic and interfacial coupling. Across shaft velocities of 1–20 rpm, the mean shaft-to-egg angular-velocity transfer ratio was 0.907 when the egg intersected the liquid–air interface and 0.752 when the egg was fully immersed. Supplementation of embryonic medium with polyvinylpyrrolidone improved motion transfer, with the highest average transfer observed at 1.0% PVP. The integrated platform enabled three-dimensional micropyle alignment, egg stabilization, and needle access within a workflow of less than 5 min.

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