Rabbit Stem Cells in Action: Hopping Towards Reliable Screening

Establishment of a rabbit embryonic stem cell line for developmental toxicity testing

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Challenges in Developmental Toxicity Testing

State-of-the-art *in vitro* alternatives for developmental toxicity testing, such as the mouse Embryonic Stem Cell Test, the rat Whole Embryo Culture, and the Zebrafish Embryo Developmental Toxicity Assay, often fail to predict the risk for humans accurately.

A promising approach is the use of human pluripotent stem cells (PSCs), including human induced PSCs (iPSCs) and human embryonic stem cells (ESCs). However, the *in vivo* effects in humans - particularly in pregnant women - remain largely unknown for most drug candidates and chemicals. To validate these PSC assays, they are often compared with animal *in vivo* data, which introduces concerns about species differences.

Key outcome

The aim of this study is to establish a stable rabbit ESC-line that can be further developed into a rabbit ESC-based developmental screening assay with a sensitivity and a specificity equal to or greater than existing *in vivo* models.
This screening assay will be validated by comparing its results with *in vivo* rabbit data and with its rat and human counterpart assays, which utilize commercially available embryonic cell lines. Future research will focus on establishing also a rabbit iPSC-based assay in order to compare the outcome of both the ESC and iPSC-based assays with the in vivo outcome.

Advantages

- Cross-species comparison
- Greater confidence in exposure-informed *in vitro* assays
- Reduction in animal usage
- High-throughput analysis

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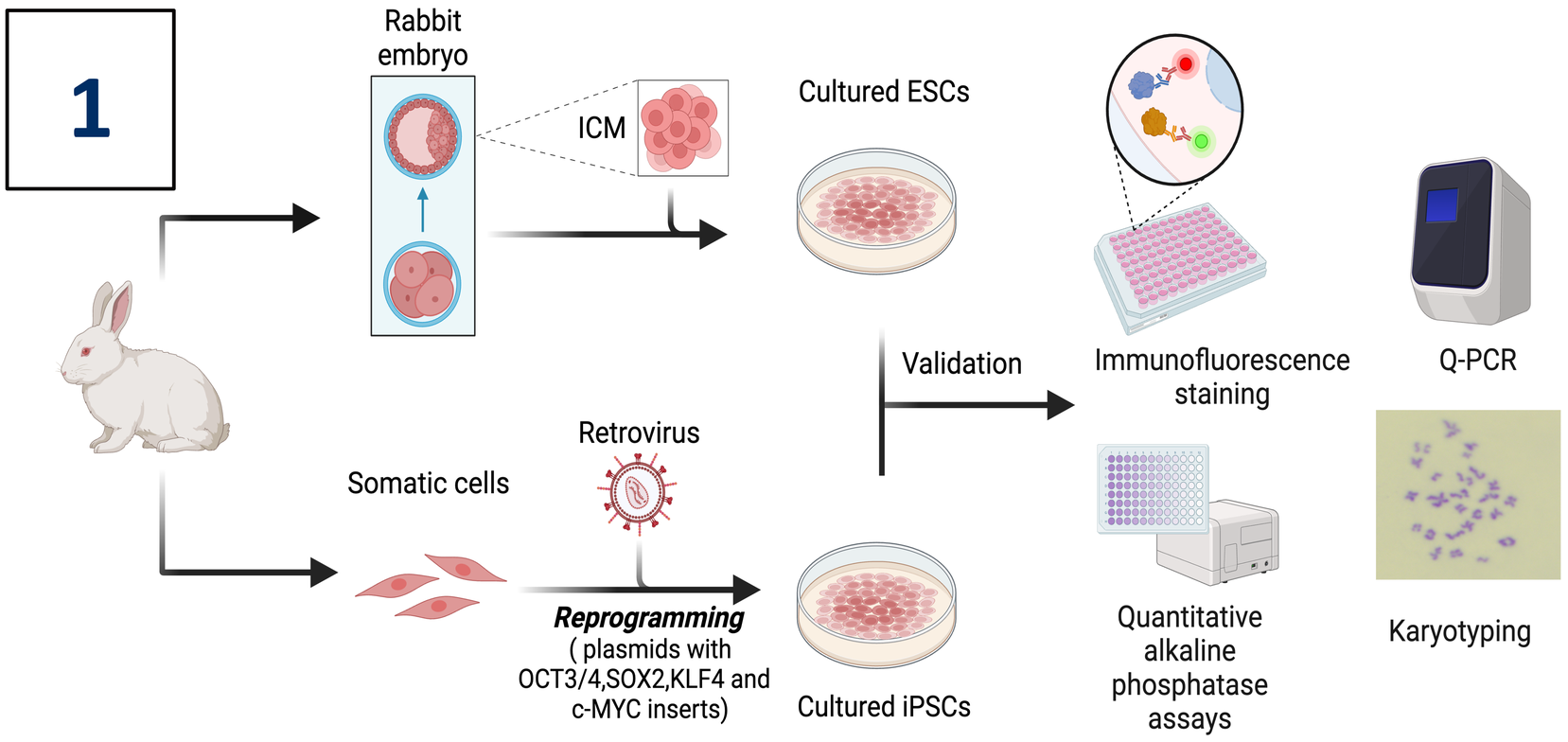
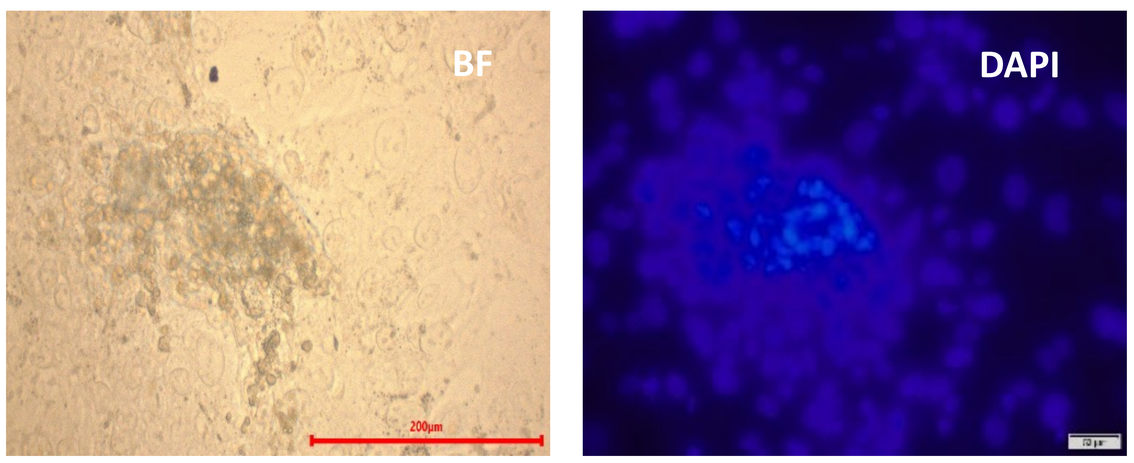


ICM = inner cell mass; ESCs = embryonic stem cells; iPSCs= induced pluripotent stem cells; Q-PCR= quantitative polymerase chain reaction; Created with BioRender.

Current Data



Co-staining of Oct4 (pluripotent marker) and CDX2 (trophoblast marker) in ICM culture. Imaged with an inverted fluorescence microscope (Olympus IX71).

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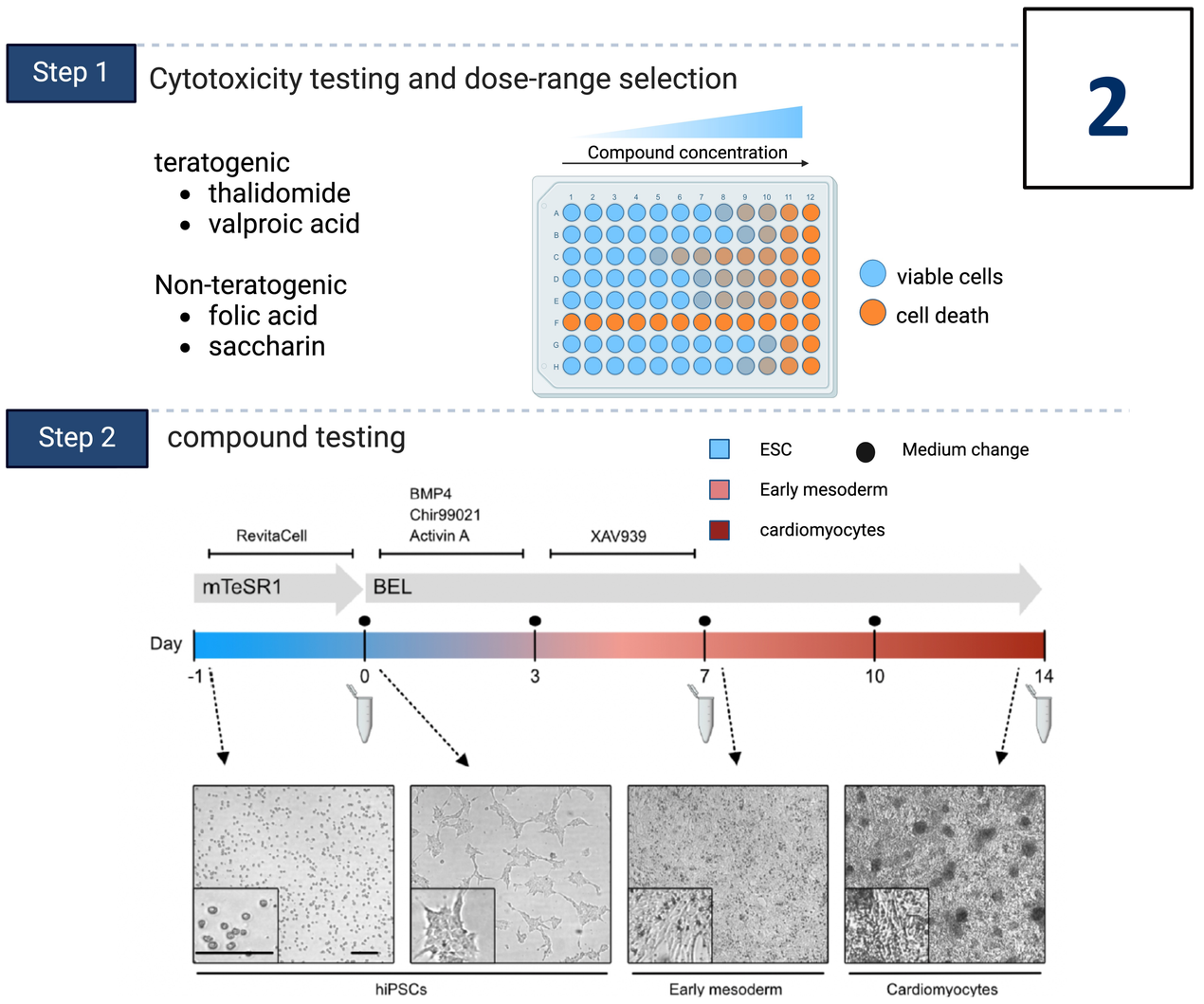

Overview of the assay setup with human iPSCs and differentiation towards cardiomyocytes. Image adapted from Jamalpoor, A. et al., 2022.; Created with BioRender.

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Acknowledgments

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