

Scientific Report of STSM-Topic: ReproCell – Somatic cell reprogramming for nuclear transfer in fish

During my stay at the INRA institute I learned about cell nuclear transfer of goldfish somatic cells into unfertilized goldfish eggs and we compared protocols for goldfish cell cultures including their analysis. Culture preparations for goldfish skin, fin and scale-derived cells were compared and for every tissue we picked the best working protocol for further analysis. For one protocol fin and skin pieces were chopped into 1–3 mm² explants and mildly digested in 0.2 mg/mL collagenase for 30 min at 25 °C, to allow a better release of the cells. In another protocol we cut and digested the tissue pieces and plated the cell suspension. Scales were picked from skin tissue and directly plated into the dish. We found that the protocol according to Moritz and Labbe et al. (2008) was the best for fin, while modified protocols according to Kruse et al. (2006) and Rakers et al. (2011) gave the best results for scales and skin. These protocols will be exchanged now between both labs for further replicates. To check for typical gene maps of cultured primary cells, we compared cells and tissue from day 0 and day 6 of scales and fin. We performed qPCR for the control (18 S RNA) as well as for Cytokeratin 49 (CK49) and Cytokeratin 8 (CK8). The amount of CK49 was significantly increased in scale cells at day 6, while CK8 did not change in fin and scale cells and tissue. Further experiments will now be performed to check other genes (collagen 1a1, vimentin). We had also several meetings and discussions about fish cell cultures and exchanged ideas on cryopreservation of aquatic genetic resources. Resulting from the first experiments, we will perform further studies in our laboratories.

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