Wu Tsai Human Performance Alliance
UC San Diego

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Notes for cultured human satellite cells (SCs) and FAPs stored in the biobank.

- For culture media, we recommend using DMEM low glucose with 20% FBS, 5ng/ml bFGF, 1% Penicillin-Streptomycin
- For thawing, we recommend seeding all cells (~1million) from a single cryovial into a T-175 with 20-25 mL of culture media.
- For passaging, we use 0.25% Trypsin-EDTA for 5 mins at 37C.
- For expansion, we seed the cells at 3500 cells/cm^2, change media every two days, and passage when the cells reach ~75% confluence (we do not let the cells grow to higher confluency as they will start fusing).
- For freezing, we use 1 million live cells per 1ml of CryoStor CS10 per cryovial.

List of materials we use:

DMEM low glucose	11054-020	Gibco™
Fetal Bovine Serum (FBS)	FB-01	Omega Scientific
Recombinant Human FGF basic/FGF2/bFGF	233-FB	R&D Systems
Penicillin-Streptomycin (10,000 U/mL)	15140122	Thermo Fisher Scientific
0.25% Trypsin-EDTA	25200-056	Gibco™
CryoStor CS10	07930	Stem Cell Technologies

Quality control:
No contamination has been detected during expansion. Some but not all samples were tested for mycoplasma.

A random set of samples at p3 has been used for quality control after a freeze-thaw cycle:

- All tested samples expanded.
- All the tested samples maintained lineage-specific markers (CD56 for SCs, CD140a for FAPs).
- SCs were able to fuse and make myofibers (MyHC+).
- FAPs were able to differentiate into fibroblasts (fibronectin+) and adipocytes (FABP4+).

For your publication:

- If these instructions were helpful, please cite https://doi.org/10.1152/ajpcell.00023.2024
- Please acknowledge the use of biobank samples in the methods and acknowledgment sections of your publication.
- Please send your publication to HPBiobank-G@ucsd.edu and we will be happy to mention it on our webpage.

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