

Protocol | Usage of SciNX media and the adaptation process

In following, the steps for a general medium adaptation is descripted:

Step	Description
A	Culture your desired cell line to 60 - 80 % confluency in standard medium (StdM)
B (Option 1) Direct adaptation	 Remove used medium; Wash with DPBS w/o Ca²⁺/Mg²⁺; Add 200 µl SciNX per cm²; Incubate at desired temperature, optionally with 5 % CO₂, for at least 48 – 72 h or towards 80 % confluency. No media change is needed; Proceed with passaging (step C).
B (Option 2) Sequential adaptation	 Alternatively, apply 200 µl of a mixture of SciNX/StdM (1:1; ratio 0.5) per cm² without washing with DPBS; Incubate for at least 48 – 72 h or towards 80 % confluency; Proceed with passaging (step C); Steadily reduce the concentration of StdM after each incubation cycle. Proceed the same manner as above, but steadily increase the ratio of SciNX/StdM from 0.5 to 0.75, 0.9 and finally 1.0 after passaging.
C Passaging	 Coat the desired vessel with structure proteins if needed (e.g. Laminin) according manufacture's protocol; Remove used medium Wash with DPBS w/o Ca²+/Mg²+ Dissociate cells; Centrifugation of cell suspension at 300·g, 3 minutes, room temperature; Resuspend cell pellet in medium and cell counting; Transfer cell suspension to desired vessel with 200 µl medium as well as 10³-10⁴ cells per cm², respectively; Repeat passaging every 4-7 days.
D	Adaption is successful after achieving at least 3 continuous passages from SciNX.

Notes | Troubleshooting

Supplements	Beneficial supplements can be added for specific cell types. In general, the SciNX media incorporate ready-to-use solutions for multiple mammalian cell lines and need minor supplementation only. Standard supplementations are EGF, IGF-1 or insulin, bFGF and other cell specific factors. The SciNX media formulation is capable to maintain cell functions for 6 - 9 days even at 100% confluency.
Adherence	Coated vessels are recommended. Depending on cell types and the cell line used there is no pre-coating of vessels needed. These preferences have to be further tested by the customer. Some cell types forms spheroids or low-level clustered cell suspensions, which are capable to grow similar or better than on adherent mode. As an alternative, add additional CaCl ₂ (sterile; up to 4 mM).
Coatings	Laminin- or collagen-based coatings are preferred, which strongly depends on cell type and cell line. Please use manufacturer's protocol and iteratively optimize the conditions.
Poor adaptation performance	In case of no cell growth is observed for several days, contact us first for further support or add known growth factors for your specific cell type in higher or even lower concentrations. In addition, keep the batch-to-batch variations of the growth factors in mind and test different batches/suppliers.