Heterokaryons (cell fusions)

Fusing cells is one method for quickly introducing a particular protein into a cell at reasonable levels and following its localisation. This can help avoid overexpression artefacts seen with transient transfection. Cells expressing two different fusion proteins can also be fused and the interaction of those proteins monitored over time. We have used this approach to study such things as the snRNP assembly pathway and the targeting of PP1 by regulatory subunits.

Reagents

Hybri-Max PEG solution (50% w/v polyethelene glycol in DPBS without calcium), SIGMA P7181 is used as a fusogen to obtain hybridomas for monoclonal antibody production. It also induces cell hybridisations. Hybri-Max comes ready to use and can be diluted to desired concentration using sterile DPBS. Can be stored at -20°C in aliquots.

Procedure

1. Co-culture the cells of interest (e.g. trypsinize the two separate dishes, mix the cells and let settle into the same dish) and leave overnight.
2. Pour media of cells and add 50% Hybrimax PEG solution (Sigma) to cover.
3. Leave on for 90 seconds and then rinse off with several media washes. Fusion occurs within 20-30 minutes, and can be observed down the light microscope as large continuous cytoplasms with multiple nuclei.