Cytospins

Please note – this protocol has been optimised for certain conditions and is meant as a guide only, you may need to alter some parameters to suit your own experiment

NOTE: If doing cytospins on freshly sorted cells some cell types may require "resting and rounding up" in culture for a couple of hours prior to spinning to give accurate morphology.

Different cells will need to be aliquoted at different concentrations (eg, dendritic cells will need to be at a lower concentration than lymphocytes or due to their large size they will end up stacked on top of one another). Similarly, different applications post-cytospin will require cells at different concentrations, it is therefore recommended that you optimise the concentration of cells required for each different application you attempt.

1. Prepare cell suspension at 2 x 10^5 - 1 x 10^6 cells/ml in PBS
2. Label slides in pencil (texta may come off during staining procedures)
3. Sandwich a filter paper between the slide and a sample holder, and line up holes. Ensure that filter paper does not have a circular dent (caused by previous use) on the side which is against the slide
4. Slide the assembled unit into the clip in cyto-centrifuge
5. Aliquot 50 µl heat-inactivated foetal calf serum into sample holder, followed by 100 µl of cell suspension
6. Spin for 8 minutes at no more than 1000 rpm. If cells are unhealthy/old/fragile, reduce speed to 600 rpm.
7. Allow slides to air-dry for approx 15 - 20 minutes (or until spot is opaque)
8. Stain as desired.

9. H&E prior to other applications