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Cell migration assay or Transwell assay

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Protocol status: Working

We use this protocol and it's working

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- 1** Cells are washed in cold PBS, detached with 5mM EDTA, centrifuged at 1200 g for 7 min and resuspended in serum-free medium.
- 2** Transwell chambers (Corning Costar) are prepared filling with 600 µl of complete medium the lower chamber.
- 3** The desired amount of cells is plated in 100 µl of serum-free medium in the upper insert (8.0 µm pore size) of each Transwell chamber and incubated at 37°C.
- 4** After 24 hours at 37°C, fill with 600 µl of 5mM EDTA an empty well beside each used Transwell chamber.
- 5** Eliminate DMEM from the upper insert and move it inside an EDTA well.
- 6** Collect DMEM from the lower chamber and put some EDTA inside, to detach cells possibly fallen from the upper insert.
- 7** Detach cells migrated on the underside of the upper insert using a plastic scraper and collecting them in the EDTA well.
- 8** Mix in one tube cells collected from the lower chamber and scraped from the insert and

centrifuge at 1200 g for 7 min, to count the migrated cells.