

ASTROGLIAL CULTURES

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Mouse cortical cells are prepared as for mixed cultures but they are seeded on laminin-coated wells (20µg/ml)

When astrocytes are confluent (generally 6-7 DIV) add Ara-C 10µM (Ara-C kills dividing cells). When astrocytes are confluent they stop dividing and microglia starts proliferating. If Ara-C is added at the right time (just when astrocytes reach confluency) it will kill microglia and not astrocytes.

Keep cells in Ara-C for 4-5 days.

Remove Ara-C and use.

Note: The protocol laminin+Ara-C gives astroglial cultures with <2% microglia

Alternatively astroglial cultures can be prepared using Ara-C but no coating, or polylysine coating (laminin is quite expensive). This protocol results in approximately 5% microglial content.