

MEF Feeder production.

Culture MEF and expand it..

(Usually expand 40-50 15cm plates. Cell quality is very important, so do not let them senescence) Maintain them 30-80% confluency

Harvest cells by trypsinization, and put it in 2x 50cm tubes in DME.

Irradiate

(6000rads, 60Gys)

After irradiation, freeze all the cells.

(Usually you can make 30~40 vials)

When you thaw first vial, count live cell number using trypan blue and then plate one million cells per 10 cm plate for mouse ESCs.