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Bone Marrow Derived Macrophage (BMDM) differentiation and maintenance

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Protocol status: Working
We use this protocol and it's working

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Abstract

This protocol describes the isolation of bone marrow derived macrophages.

Attachments

724-1815.docx
17KB

Materials

Reagents required

- L929 conditioned media
- Penicillin/Streptomycin **Thermo Fisher Scientific Catalog #**Invitrogen 15140-122
- Gibco™ Fetal Bovine Serum **Fisher Scientific Catalog #**16-140-071
- Gibco™ DMEM/F-12 HEPES **Thermo Fisher Scientific Catalog #**11330032
- GlutaMAX™ Supplement **Gibco - Thermo Fisher Scientific Catalog #**35050061

1

Note

Note: For mouse bone marrow-derived macrophage (BMDMs) primary cultures, each experiment involved age and sex matched C57BL/6 mice between 3-9 months of age. *Lrrk2*KO (Lrrk2$^{tm1.1Mjff}$) and G2019S (Lrrk2$^{tm1.1Hlme}$) homozygous knockin mice were obtained from The Jackson Laboratory[76].

Euthanize mice via CO_2 or isoflurane inhalation and cervical dislocation.

2 Collect femurs and flush cavities with 5 mL ice-cold PBS.

3 Then, collect bone marrow cells by centrifugation 0.3 rcf, 00:05:00 . 5m

4 Resuspend the resulting pellet and differentiate for 6 days in culture media containing: DMEMF12 supplemented with 20% FBS, 20% L929 conditioned media, 1% penicillin-streptomycin and 1% GlutaMAX^{TM}.