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

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🌐 Bone Marrow Derived Macrophage (BMDM) differentiation and maintenance

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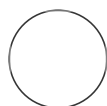
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ABSTRACT

This protocol describes the isolation of bone marrow derived macrophages.

ATTACHMENTS

[724-1815.docx](#)

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 Catalog #35050061

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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 Laboratory⁷⁶.

Euthanize mice via CO₂ 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% GlutaMAXTM.