

September 04, 2016

Teena Mehta, Science Editor
Journal of Visualized Experiments
1 Alewife Center, Suite 200
Cambridge, MA 02140

Dear Teena Mehta,

We are writing to submit our manuscript entitled, "Isolation, Characterization, and Purification of Tissue Resident Macrophages from Tissues Affected by Obesity-Related Inflammation," for consideration for publication in the Journal of Visualized Experiments (JoVE). This manuscript emerges from our experience in isolating tissue resident macrophages from diet- induced inflamed tissue sources. Primarily this article endeavors to help researchers achieve adequate isolations of macrophages for further characterization in similar models. Through years of consulting with resident experts and following suite with their recommendations and subsequent optimization trials, our research team was able to achieve superb enzymatic isolations and flow cytometry based characterization of tissue resident macrophage populations from the liver, white adipose tissue and aorta. We think researchers would benefit greatly from this format as fatty and inflamed tissues are oftentimes difficult to isolate, digest and process. This manuscript emphasizes troubleshooting options, in which case we have incorporated a comprehensive guide of alternative approaches to help researchers with various options for enriched and adequate isolations.

All authors have approved the written component of this manuscript and it's submission to the Journal of Visualized Experiments and those listed in the author byline have contributed sufficiently to the project. The author contributions are as follows, Joselyn Allen: Substantial contribution to conception and design, data acquisition and interpretation, primary writer and editor of the manuscript; Adwitia Dey: Substantial contribution to drafting and revising the manuscript; Ruth Nissly: Substantial contribution to data acquisition and provided technical expertise; Gayathri Balandaram: Substantial contribution to data acquisition and provided technical expertise; Jeffrey M. Peters: Principal Investigator and provided final approval of the version to be published; Pamela A Hankey-Giblin: Substantial contribution to the revision of the manuscript, provided final approval of the version to be published, primary Principal Investigator and corresponding author.

This manuscript has not been published and is not under consideration for publication elsewhere. If relevant, we briefly discussed these methods in a recently published Journal of Immunology publication (2016). We have no conflicts of interest to disclose, but we do respectfully request that Dr. Peter Murray not be considered as to review our manuscript. If you feel that the manuscript is appropriate for JoVE, we suggest the following reviewers, Ajay Chawla (University of California San Francisco), Philip R. Taylor (Cardiff University School of Medicine), Alan R. Saltiel (University of California San Diego School of Medicine), Tim P. Bushnell (University of Rochester Medical Center), Ernest M. Meyer (University of Pittsburgh Cancer Institute and David Adams (University of Michigan Medical School).

We are grateful to JoVE editors Teena Mehta and Ronald Myers for their guidance and ultimate patience with our submission.

We humbly thank you for taking our manuscript into consideration and review. We sincerely appreciate your time and enthusiastically look forward to your response.

Kind Regards,
Joselyn N. Allen (First Author)
Pamela A Hankey-Giblin (Correspondence Author)