# Exosome isolation from plasma using ExoQuick reagent

**Xandra O. Breakefield** ( breakefield@hms.harvard.edu )
Departments of Neurology and Radiology and Program in Neuroscience, Harvard Medical School, Massachusetts General Hospital, Boston, MA, USA

**Louise C. Laurent** ( llaurent@ucsd.edu )
Department of Reproductive Medicine, University of California, San Diego, La Jolla, CA, USA

**Anil K. Sood** ( asood@mdanderson.org )
Departments of Gynecologic Oncology and Reproductive Medicine and of Cancer Biology and Center for RNA Interference and Non-Coding RNAs, The University of Texas MD Anderson Cancer Center (MDACC), Houston, TX, USA

**Leonora Balaj**
Department of Neurology and Program in Neuroscience, Harvard Medical School, Massachusetts General Hospital, Boston, MA, USA

**Justyna Filant**
Department of Gynecologic Oncology and Reproductive Medicine, The University of Texas MD Anderson Cancer Center (MDACC), Houston, TX, USA

**Srimeenakshi Srinivasan**
Department of Reproductive Medicine, University of California, San Diego, La Jolla, CA, USA

**Xuan Zhang**
Department of Neurology, Harvard Medical School, Massachusetts General Hospital, Boston, MA, USA

**Method Article**

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# Abstract

This protocol describes how to isolate exosomes from plasma using the ExoQuick Plasma Prep and Exosome Precipitation kit. It supercedes an earlier protocol \(http://dx.doi.org/10.1038/protex.2015.108). The new protocol is based on our experience vetting the original protocol in multiple labs. The major change is an increase in input volume of biofluid from 200 µL to 500 µL. We found that the larger input volume led to more reproducible amounts of RNA isolation across multiple experiments. We have also included footnotes to explain changes we made to manufacturers' protocols, and observations that were made while carrying out the protocols.


# Introduction

Extracellular RNAs \(exRNAs) have been found to play an important role in intercellular communication in the body. exRNAs are present in biofluids, in complexes with lipoproteins and ribonucleoproteins, and in extracellular vesicles such as exosomes and microvesicles. Further study and characterization of exRNAs and their carriers could lead to identification of new biomarkers and have potential for development of novel therapeutics.

# Reagents

ExoQuick Plasma Prep and Exosome Precipitation kit \(System Biosciences – EXOQ5TM-1) Phosphate-buffered saline \(PBS)

# Equipment

Microfuge 1.5 mL Microfuge tubes

# Procedure

1. Transfer 500 µL of plasma into a 1.5 mL microfuge tube. 2. Add 5 µL thrombin \(500U/mL) to a final concentration of 5U/mL. 3. Incubate at room temperature for 5 minutes while mixing \(gently flicking tube). 4. Centrifuge at 10,000 x g for 5 minutes. There should be a visible fibrin pellet at the bottom of the tube. 5. Transfer supernatant to new microfuge tube. 6. Add 125 µL of ExoQuick Exosome Precipitation Solution to the plasma and incubate for 30 minutes at 4°C. 7. Centrifuge ExoQuick/plasma mixture at 1,500 × g for 30 minutes at room temperature. 8. Aspirate supernatant. 9. Spin down residual ExoQuick solution by centrifugation at 1,500 × g for 5 minutes at room temperature. 10. Remove all traces of fluid by aspiration, taking great care not to disturb the pellet. 11. Resuspend the pellet in 50 µL sterile PBS and proceed with RNA isolation using SeraMir Exosome RNA Purification Column Kit.

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