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Protocol status: In development
 We are still developing and optimizing this protocol

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Human platelet isolation

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ABSTRACT

Platelets are the primary cellular mediator of thrombosis. This protocol allows for the preparation of washed human platelets. These platelets can then be stimulated or cultured for various purposes.

GUIDELINES

Handle platelets gently at all steps to avoid activating them

MATERIALS

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⊗ Tyrodes Buffer **Boston Bioproducts Catalog #PY-921**

⊗ PGI2 (10mM) **Santa Cruz Biotechnology Catalog #61849-14-7**

SAFETY WARNINGS

Human blood is a biosafety hazard. Be familiar and have approval to work under BSL-2 conditions.

BEFORE START INSTRUCTIONS

Prepare Tyrode's Buffer, or use Tyrode's buffer from a commercial source

1 Use IV butterfly set and draw blood into sodium citrate vacutainers

- 2 Centrifuge at 1200 rpm for 15 minutes at room temperature
- 3 Transfer plasma top fraction to a new tube
- 4 Dilute plasma with an equal volume of Tyrodes buffer
- 5 Add PGI2 to a final concentration of 10uM
- 6 Centifuge at 2800 rpm for 5 minutes, decant the supernatant and gently resuspend the platelet pellet into Tyrode's buffer
- 7 Dilute the platelets 1:20 in fresh Tyrode's buffer for downstream applications such as flow cytometry, or lyse pellet directly for ELISA or western blotting.