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

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Amplicon clean-up using SPRI beads V.1

Forked from [Amplicon clean-up using SPRI beads](#)

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Coronavirus Method Development Community

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MATERIALS

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Agencourt AMPure XP beads **Contributed by users**

STEP MATERIALS

Agencourt AMPure XP **Beckman Coulter Catalog #A63880**

QuantiFluor(R) ONE dsDNA System, 100rxn **Promega Catalog #E4871**

Elution Buffer (EB) **Qiagen Catalog #19086**

Freshly prepared 80% ethanol

Ampure XP bead clean up

- 1 Vortex SPRI beads thoroughly to ensure they are well resuspended, the solution should be a homogenous brown colour.

Agencourt AMPure XP **Beckman Coulter Catalog #A63880**

- 2 Add an equal volume (1:1) of SPRI beads to the sample tube and mix gently by either flicking or pipetting. For example add 50 µL room temperature SPRI beads to a 50 µL reaction.
- 3 Pulse centrifuge to collect all liquid at the bottom of the tube.
- 4 Incubate for 00:05:00 at room temperature.
- 5 Place on magnetic rack and incubate for 00:02:00 or until the beads have pelleted and the supernatant is completely clear.
- 6 Carefully remove and discard the supernatant, being careful not to touch the bead pellet.
- 7 Add 200 µL of freshly prepared room-temperature 80 % volume ethanol to the pellet.
- 8 Keeping the magnetic rack on the benchtop, rotate the bead-containing tube by 180°. Wait for the beads to migrate towards the magnet and re-form a pellet. Remove the ethanol using a pipette and discard.

- 9 and repeat ethanol wash.
- 10 Pulse centrifuge to collect all liquid at the bottom of the tube and carefully remove as much residual ethanol as possible using a P10 pipette.
- 11 With the tube lid open incubate for 00:01:00 or until the pellet loses its shine (if the pellet dries completely it will crack and become difficult to resuspend).
- 12 Remove the tube from the magnetic rack. Resuspend pellet in 10 μL molecular grade water or Elution buffer, mix gently by flicking and incubate for 00:02:00 .
Elution Buffer (EB) **Qiagen Catalog #19086**
- 13 Place on magnet and transfer sample to a clean 1.5mL Eppendorf tube ensuring no beads are transferred into this tube.
- 14 Quantify 1 μL product using the Quantus Fluorometer using the ONE dsDNA assay.
QuantiFluor(R) ONE dsDNA System, 100rxn **Promega Catalog #E4871**

Equipment

Quantus

NAME

Fluorometer

TYPE

Promega

BRAND

E6150

SKU

<https://www.promega.co.uk/products/microplate-readers-fluorometers-luminometers/fluorometers/quantus-fluorometer>

LINK