

Amplicon clean-up using SPRI beads

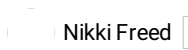
Forked from [Amplicon clean-up using SPRI beads](#)

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In Development dx.doi.org/10.17504/protocols.io.bfwpjpdn

Coronavirus Method Development Community



Nikki Freed

MATERIALS

NAME	CATALOG #	VENDOR
Agencourt AMPure XP beads		

STEPS MATERIALS

NAME	CATALOG #	VENDOR
Agencourt AMPure XP	A63880	Beckman Coulter
QuantiFluor(R) ONE dsDNA System, 100rxn	E4871	Promega
Elution Buffer (EB)	19086	Qiagen

MATERIALS TEXT

Freshly prepared 80% ethanol

EQUIPMENT










NAME	CATALOG #	VENDOR
Quantus	E6150	

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
by [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)**
by 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
by Promega
Catalog #: E4871



Quantus
Fluorometer
Promega E6150 [↗](#)