

May 05, 2020

Version 1

## Amplicon clean-up using SPRI beads V.1



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

DOI

[dx.doi.org/10.17504/protocols.io.bfwpjpdn](https://dx.doi.org/10.17504/protocols.io.bfwpjpdn)

Muhammad Faisal<sup>1</sup>, Olin Silander<sup>1</sup>, Nikki Freed<sup>1</sup>

<sup>1</sup>Massey University

Coronavirus Method De...



Nikki Freed

### Create & collaborate more with a free account

Edit and publish protocols, collaborate in communities, share insights through comments, and track progress with run records.

Create free account

OPEN  ACCESS



DOI: <https://dx.doi.org/10.17504/protocols.io.bfwpjpdn>

**Protocol Citation:** Muhammad Faisal, Olin Silander, Nikki Freed 2020. Amplicon clean-up using SPRI beads. **protocols.io** <https://dx.doi.org/10.17504/protocols.io.bfwpjpdn>

**License:** This is an open access protocol distributed under the terms of the **[Creative Commons Attribution License](#)**, which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited



**Protocol status:** In development

**We are still developing and optimizing this protocol**

**Created:** May 02, 2020

**Last Modified:** May 05, 2020

**Protocol Integer ID:** 36527

**Keywords:** using spri bead, amplicon clean, spri bead

## Materials

### MATERIALS

⊗ Agencourt AMPure XP beads

### 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

## Protocol materials

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

⊗ Agencourt AMPure XP beads

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

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









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

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







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

## Troubleshooting

## 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  [go to step #7](#) 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 it's 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  $\mu\text{L}$  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  $\mu\text{L}$  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