

ChargeSwitch® PCR Clean-Up Kit

Catalog No. CS12000-10

Quantity: 960 reactions

Store at Room Temperature

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Contents

The ChargeSwitch® PCR Clean-Up Kit is shipped at room temperature. Upon receipt, store all components at room temperature. All components are guaranteed stable for 6 months when stored properly. The components supplied in the ChargeSwitch® PCR Clean-Up Kit are listed in the following table. The reagents supplied are sufficient to perform 960 purifications.

Component	Amount*
ChargeSwitch® Magnetic Beads (25 mg/mL in 10 mM MES, pH 5.0, 10 mM NaCl, 0.1% Tween 20)	11 mL
ChargeSwitch® Purification Buffer (N5)	65 mL
ChargeSwitch® Wash Buffer (W12)	2 × 200 mL
ChargeSwitch® Elution Buffer (E5; 10 mM Tris-HCl, pH 8.5)	48 mL

*Note: Some reagents may be supplied in excess of the amount needed.

Description

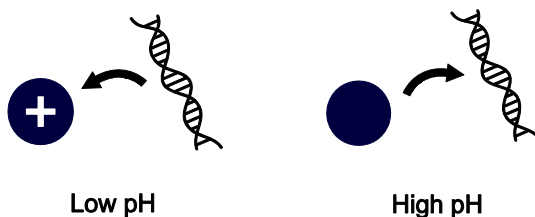
The ChargeSwitch® PCR Clean-Up Kit allows rapid and efficient purification of PCR products from salts, primers, dNTPs, and other non-nucleic acid reagents. The kit is designed for automated processing of large numbers of samples in 96-well plates using a liquid handling robot. The kit is designed for the purification of DNA fragments ranging from 90 bp–40 kb and the purified PCR product is suitable for any downstream applications of choice.

The PCR product is purified in less than 10 minutes with the ChargeSwitch® Technology using an automated liquid handling robot. For more information on the Charge Switch® Technology, see the following section.

Use of the ChargeSwitch® PCR Clean-Up Kit has been demonstrated on the Tecan Genesis® robotic workstation to purify PCR products in a fully automated system from large numbers of samples in a 96-well format. Other liquid handling robots are suitable provided that each is equipped with a gripper arm, a 96-well magnetic separator, and other additional hardware as described on page 2. This manual provides general guidelines and a protocol that can be used to develop a script for your robot.

The ChargeSwitch® Technology

The ChargeSwitch® Technology is a novel magnetic bead-based technology providing a switchable surface that is charge dependent on the surrounding buffer pH to facilitate nucleic acid purification. The ChargeSwitch® chemistry is ideal for purification of DNA using liquid handling robots, avoiding the need for centrifugation steps or the use of ethanol or chaotropic salts. In low pH conditions, the ChargeSwitch® Magnetic Beads have a positive charge and binds the negatively charged nucleic acid backbone (see figure). Proteins and other contaminants are not bound and are washed away using the wash buffer. To elute nucleic acids, the charge on the surface is neutralized by raising the pH to 8.5 using a low-salt elution buffer (see figure). Purified DNA elutes instantly into this elution buffer.



Safety Information

Follow the safety guidelines below when using the ChargeSwitch® Kit.

- Treat all reagents supplied in the kit as potential irritants.
- Always wear a suitable lab coat, disposable gloves, and protective goggles.
- If a spill of the buffers occurs, clean with a suitable laboratory detergent and water. If the liquid spill contains potentially infectious agents, clean the affected area first with laboratory detergent and water, then with 1% (v/v) sodium hypochlorite or a suitable laboratory disinfectant.

Automated Procedure for Purifying PCR Products

Materials Needed

Components not supplied with the kit

- PCR samples
- 96 × 200 µL U-bottomed microtiter plate and 96 Deep Well Block
- Any liquid handling robotic workstation with a gripper arm to process samples in 96-well plates
- Appropriate tips for liquid dispensing and aspiration (see below)
- 96-Well Magnetic Separator (see below)
- Shaker

Components supplied with the kit

- ChargeSwitch® Purification Buffer (N5)
- ChargeSwitch® Magnetic Beads
- ChargeSwitch® Wash Buffer (W12)
- ChargeSwitch® Elution Buffer (E5)

Handling Magnetic Beads

To maximize DNA yield, follow these recommendations when processing your samples:

- Resuspend the ChargeSwitch® Magnetic Beads thoroughly before use.
- Ensure that the robotic tips enter the wells of the plate without interfering with the bead pellet.
- When removing supernatant, aspirate slowly to ensure that the pellet of beads is not disturbed.
- To maximize DNA yield, make sure that all Wash Buffer is removed before elution and the beads are fully resuspended during the elution step.

96-Well Magnetic Separator

The 96-Well Magnetic Separator available from Life Technologies (Cat. no. CS15096) is a magnetic separation rack suitable for use in protocols with magnetic beads. The rack can hold up to 96 samples in a deep well plate. The deep well plate fits onto the magnetic separator, associating the array of 24 neodymium magnets with 96 samples for magnetic sample processing (see the following photographs).



Do not freeze the magnetic beads, as frozen beads cannot be used for nucleic acid purifications.

Tip Selection

You may use any tips of choice to dispense and aspirate liquid during the purification procedure. Consider the following factors when choosing an appropriate tip to use:

- Fixed vs. disposable tips
- Tip size vs. head size
- Conductive or non-conductive
- Sterile or non-sterile
- Filtered or non-filtered

Deck Set Up

Once you have the required hardware, you need to configure the deck of your liquid handling robot appropriately to process samples. You may use any suitable configuration of your choice. For more information and details, see www.lifetechnologies.com/support or contact Technical Support.

Automated Procedure for Purifying PCR Products, continued

Automated Protocol

This section provides a general protocol for automated purification of PCR products in a 96-well format. Use the parameters and guidelines provided on page 2, as well as this protocol to develop the script for your liquid handling robot. For more information, see www.lifetechnologies.com/support or call Technical Support.

The following protocol should be used to purify PCR products. The volumes given are on a per sample basis.

1. To ~50 μ L PCR samples in 96-well plates, add 10 μ L ChargeSwitch[®] Magnetic Beads.
2. Add 60 μ L Purification Buffer (N5).
3. Shake at medium speed for 30 seconds to evenly distribute the magnetic beads in the solution.
4. Move samples to the 96-Well Magnetic Separator. Wait for 30 seconds.
5. Aspirate all of the supernatant and discard, leaving behind the pellet of beads.
6. Add 150 μ L Wash Buffer (W12). Move samples to the shaker.
7. Shake at medium speed for 30 seconds to evenly distribute the magnetic beads in the solution.
8. Move samples to the 96-Well Magnetic Separator. Wait for 1 minute.
9. Aspirate all of the supernatant and discard, leaving behind the pellet of beads.
10. Add 150 μ L Wash Buffer (W12). Move samples to the shaker.
11. Shake at medium speed for 30 seconds to evenly distribute the magnetic beads in the solution.
12. Move samples to the 96-Well Magnetic Separator. Wait for 1 minute.
13. Aspirate all of the supernatant and discard, leaving behind the pellet of beads.
14. Add 50 μ L Elution Buffer (E5; 10 mM Tris-HCl, pH 8.5). Move samples to the shaker.
15. Shake at fast speed for 1 minute to evenly distribute the magnetic beads within the solution. Wait for 30 seconds.
16. Move samples to the 96-Well Magnetic Separator. Wait for 1 minute.
17. Slowly aspirate **supernatant containing the purified PCR product** to a 96 \times 200 μ L U-bottomed microtiter plate.

Storage

Store the purified PCR product at -20°C or use the PCR product in the downstream applications of choice.

Accessory Products

The table below lists additional products available from Life Technologies that may be used with the ChargeSwitch® PCR Clean-Up Kit.

Product	Quantity	Catalog No.
96-Well Magnetic Separator	1 rack	CS15096
96 Deep Well Block	1 case of 50	CS15196
ChargeSwitch® PCR Clean-Up Kit	100 purifications	CS12000
Quant-iT™ DNA Assay Kit, High Sensitivity	1000 assays	Q33120
Quant-iT™ DNA Assay Kit, Broad Range	1000 assays	Q33130
Quant-iT™ PicoGreen® dsDNA Assay	1 kit	P7589

A large selection of ChargeSwitch® products is available from Life Technologies for plasmid and genomic DNA purification from various sources.

E-Gel® Agarose Gels are bufferless pre-cast agarose gels designed for fast, convenient electrophoresis of DNA samples. E-Gel® agarose gels are available in different agarose percentage and well format for your convenience. A large variety of DNA ladders is available from Life Technologies for sizing DNA.

For more information on these products, visit www.lifetechnologies.com/support or contact Technical Support.

Product Qualification and Purchaser Notification

Product Qualification and SDS

The Certificate of Analysis provides detailed quality control and product qualification information for each product. Certificates of Analysis are available on our website. Go to www.lifetechnologies.com/support and search for the Certificate of Analysis by product lot number, which is printed on the box.

Safety Data Sheets (SDSs) are available at www.lifetechnologies.com/support.

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