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ChargeSwitch® PCR Clean-Up Kit

Catalog No. CS12000

Quantity: 100 reactions

Store at Room Temperature

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Contents

The ChargeSwitch[®] PCR Clean-Up Kit is shipped at room temperature. 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 100 purifications.

Component	Amount
ChargeSwitch® Magnetic Beads (25 mg/mL in 10 mM MES, pH 5.0, 10 mM NaCl, 0.1% Tween® 20)	1 mL
ChargeSwitch [®] Purification Buffer (N5)	8 mL
ChargeSwitch [®] Wash Buffer (W12)	40 mL
ChargeSwitch [®] Elution Buffer (E5; 10 mM Tris-HCl, pH 8.5)	10 mL

Note: Some reagents in the kit maybe provided 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.

After PCR, the PCR product is purified in less than 10 minutes using the ChargeSwitch[®] Technology without the use of a centrifuge or organic solvents. For more information on the Charge Switch[®] Technology, see the following section. The kit is designed for the purification of DNA fragments ranging in size from 90 bp–40 kb and the purified PCR product is suitable for any downstream applications of choice.

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. In low pH conditions, the ChargeSwitch[®] Magnetic Beads have a positive charge and binds the negatively charged nucleic acid backbone (see the following 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 the following figure). Purified DNA elutes instantly into this elution buffer.



System Specifications

Starting Material:	25–50 μL PCR sample
DNA Fragment Size:	90 bp-40 kb
Bead Binding Capacity:	1 mg beads bind ~25 μg DNA
Elution Volume:	50 µL

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.

Purifying PCR Products

Materials Needed

Components not supplied with the kit

- 25–50 µL PCR sample
- MagnaRack[™] Magnetic Rack (Cat. no. CS15000)
- Sterile 1.5-mL microcentrifuge tubes
- Adjustable pipettes and aerosol barrier pipette tips

Components supplied with the kit

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

Handling Magnetic Beads

Follow the recommendations below for best results:

- During the mixing, washing, and resuspending steps of the ChargeSwitch[®] Magnetic Beads, mix beads by pipetting up and down gently as directed in the protocol.
- Always mix the contents by pipetting up and down gently to avoid forming bubbles after bead addition.
- Do not allow the beads to dry as drying reduces the bead binding efficiency.
- To aspirate the supernatant after bead washing, place the pipette tip away from the beads by angling the pipette such that the tip is pointed away from the pellet and carefully remove the supernatant without disturbing or removing any beads (see the following figure).



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

Binding DNA

Note: Instructions are provided to purify the PCR product from 25–50 μ L PCR sample. If you wish to process >50 μ L of PCR sample, be sure to scale-up the volume of all reagents accordingly.

- 1. Vortex the tube containing the ChargeSwitch[®] Magnetic Beads to fully resuspend and evenly distribute the beads in the storage buffer.
- 2. Transfer 25–50 µL PCR sample to a sterile 1.5-mL microcentrifuge tube.
- 3. Add 25 µL (for 25 µL PCR sample) or 50 µL (for 50 µL PCR sample) Purification Buffer (N5) to the tube.
- 4. Add 10 µL ChargeSwitch® Magnetic Beads to the tube, and pipet up and down gently to mix without forming bubbles.
- 5. Incubate at room temperature for 1 minute.
- 6. Place the sample on the MagnaRack[™] base station for 1 minute or until the beads form a tight pellet.
- 7. Without removing the tube from the MagnaRack[™] base station, carefully remove and discard the supernatant without disturbing the pellet of beads by angling the pipette such that the tip is pointed away from the pellet (see **Handling Magnetic Beads**).
- 8. Proceed immediately to **Washing DNA**.

Washing DNA

- 1. Remove the tube containing the pelleted magnetic beads from the MagnaRack[™] base station (step 7, **Binding DNA**).
- 2. Add 150 µL Wash Buffer (W12) to the tube and pipet up and down gently to mix the sample without forming bubbles.
- 3. Place the sample on the MagnaRack^M base station for 1 minute or until the beads form a tight pellet.
- 4. Without removing the tube from the MagnaRack[™] base station, carefully remove and discard the supernatant without disturbing the pellet of beads by angling the pipette tip away from the pellet (see **Handling Magnetic Beads**).
- 5. Repeat Steps 1–4 once.
- 6. Proceed immediately to **Eluting DNA**.

Purifying PCR Products, continued

Eluting DNA

- 1. Remove the tube containing the pelleted magnetic beads from the MagnaRack[™] base station (step 5, **Washing DNA**).
- 2. Add 25–50 µL Elution Buffer (E5; 10 mM Tris-HCl, pH 8.5) to the tube, and pipet up and down gently to mix the sample without forming bubbles.
- 3. Incubate at room temperature for 1 minute.
- 4. Place the sample on the MagnaRack[™] base station for 1 minute or until the beads form a tight pellet.
- 5. Without removing the tube from the MagnaRack[™] base station, carefully transfer the **supernatant containing the purified PCR product** to a sterile microcentrifuge tube without disturbing the pellet of beads.
- 6. Store the purified PCR product at -20°C or use the PCR product in the downstream application of choice.

Troubleshooting Guide

Observation	Cause	Solution
Low DNA yield	PCR conditions not optimal	Check amplicon on gel to verify the PCR product prior to purification.
	Incorrect binding conditions	Use 25–50 µL Purification Buffer (N5) for sample preparation.
	Incorrect elution conditions	After adding Elution Buffer (E5) to the sample, pipet up and down to resuspend the magnetic beads before incubation.

Accessory Products

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

Product	Amount	Catalog No.
MagnaRack [™] Magnetic Rack	1 each	CS15000
ChargeSwitch [®] PCR Clean-Up Kit	960 purifications	CS12000-10
Quant-iT™ DNA Assay Kit, High Sensitivity	1000 assays	Q33120
Quant-iT™ dsDNA Assay Kit, Broad Range	1000 assays	Q33130
Quant-iT [™] PicoGreen [®] dsDNA Assay Kit	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 percentages and well formats. A large variety of DNA ladders is available from Life Technologies for sizing DNA.

For more information on these products, visit www.lifetechnologies.com 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.

Limited Product Warranty

Life Technologies Corporation and/or its affiliate(s) warrant their products as set forth in the Life Technologies' General Terms and Conditions of Sale found on Life Technologies' website at **www.lifetechnologies.com/termsandconditions**. If you have any questions, please contact Life Technologies at **www.lifetechnologies.com/support**.

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