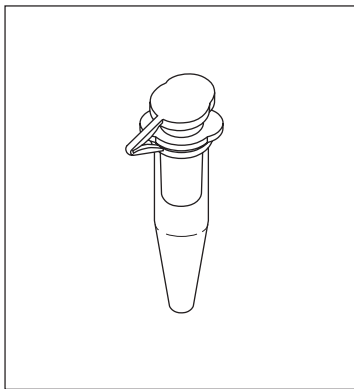


Micropure[®]-EZ

Enzyme Removers



MILLIPORE

Notice

The information in this document is subject to change without notice and should not be construed as commitment by Millipore Corporation. Millipore Corporation assumes no responsibility for any errors that may appear in this document. This manual is believed to be complete and accurate at the time of publication. In no event shall Millipore corporation be liable for incidental or consequential damages in connection with or arising from the use of this manual.



©2006 MILLIPORE CORPORATION. PRINTED IN THE UNITED STATES OF AMERICA. ALL RIGHTS RESERVED.

Millipore, Micropure, Microcon, and Ultracel are registered trademarks of Millipore Corporation. Vent is a registered trademark of New England Biolabs, Inc. or its subsidiaries.

Product Overview

Micropure-EZ enzyme removers provide a simple, rapid and phenol-free method of removing restriction and other enzymes from double-stranded DNA. The membrane employed in the device has a high affinity for protein, but not dsDNA. Protein removal is as simple as adding the enzyme reaction mix to the Micropure-EZ device and spinning for 60 seconds to recover undiluted DNA. If sample concentration or desalting is also required, Microcon[®] microconcentrators may be incorporated for DNA purification, concentration and desalting in a single unit.

Operating Modes

Micropure-EZ devices can be operated in two modes:

- With vial, for enzyme removal from sample.
- With Millipore's Microcon concentrators, for enzyme removal, DNA concentration and buffer exchange or desalting.

Materials Supplied

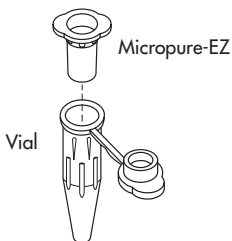
Micropure-EZ enzyme removers are offered in packages of 8, 24 or 100 units and may be ordered either with or without vials. The product selection guide under "Ordering Information" provides recommendations on device combinations for the applications selected.

Equipment Required

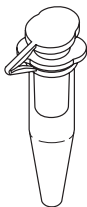
Any centrifuge that can accommodate 1.5 mL microcentrifuge tubes.

Using Micropure-EZ Device with Vial

Place Micropure-EZ device into vial; add sample.



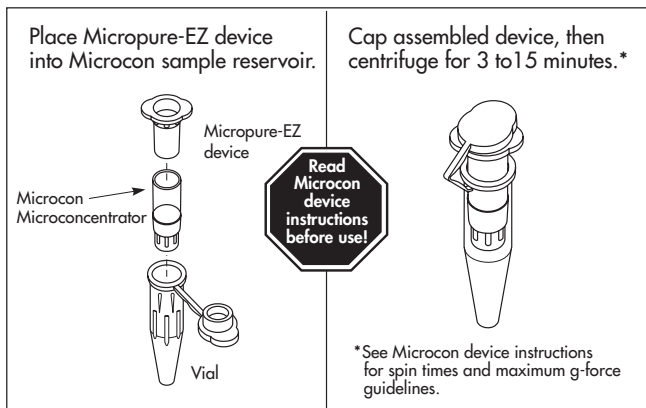
Cap assembled device, then centrifuge for 60 seconds.



Procedure:

1. Place Micropure-EZ device into vial; pipette the sample (up to 200 μL) into the Micropure-EZ reservoir. Close cap.
2. Place assembled device in a microcentrifuge and spin at full speed (12,000–14,000 $\times g$) for 60 seconds. Spin time will depend on the sample volume. For example, a 50 μL sample will pass in less than 30 seconds when spun at 14,000 $\times g$.
3. Filtrate contains purified DNA. To maximize DNA recovery add 5–10 μL of TE buffer or water to Micropure-EZ reservoir and spin for 30 seconds. The Micropure-EZ reservoir may now be discarded.
4. Cap vial to store sample.

Using Micropure-EZ Device with Microcon Device



Procedure:

1. Place Micropure-EZ reservoir into a Microcon microconcentrator device and then into the Micropure-EZ vial.
2. Add sample (up to 200 μL) to Micropure-EZ reservoir. Cap assembly and centrifuge 3–15 minutes (spin time and g-force depend on initial sample volume and NMWL of the membrane in the Microcon device).
3. Remove the Micropure-EZ device and discard. If buffer exchange is required, add wash buffer to Microcon sample reservoir and spin again according to the Microcon device operating instructions (publication 99394). If only sample concentration is desired, proceed to step 4.
4. Invert Microcon device into a new filtrate vial; spin briefly to recover sample. If the Microcon membrane appears dry, add 10–20 μL of an appropriate buffer to sample reservoir and vortex briefly.

Storage

Store Micropure-EZ devices at room temperature.

Materials of Construction

Sample reservoir: polypropylene

Sample vial: polypropylene

Specifications

Maximum initial sample volume: 200 μ L

Holdup volume: < 5 μ L

Maximum relative centrifugal force:

Micropure-EZ device: 14,000 \times g

Microcon devices with Ultracel[®] YM membranes:

Ultracel YM-3: 14,000 \times g

Ultracel YM-10: 14,000 \times g

Ultracel YM-30: 14,000 \times g

Ultracel YM-50: 12,000 \times g

Ultracel YM-100: 500 \times g

NOTE: Do not exceed specified centrifugal force!
When spinning Micropure-EZ devices together with Microcon devices, do not exceed specified limits for either device.

Active membrane surface area: 0.17 cm²

Dimensions:

Diameter: 10.7 mm

Length of Micropure-EZ device and filtrate vial: 40.9 mm

Length of Micropure-EZ device, Microcon device, and filtrate vial: 49.2 mm

Enzymes Tested for Use with Micropure-EZ Device

Enzymes Removed	Challenge	Enzymes Removed	Challenge
AMV reverse transcriptase	50 U	<i>EcoR</i> I	100 U
Calf intestinal alkaline phosphatase	10 U	<i>Hae</i> III	100 U
DNase I (bovine pancreas)	10 U	<i>Hinc</i> II	50 U
Exonuclease III (<i>E. coli</i>)	100 U	<i>Hind</i> III	100 U
MMLV reverse transcriptase	600 U	<i>Hpa</i> I	25 U
Mung bean nuclease	50 U	<i>Kpn</i> I	50 U
Proteinase K (Amresco)	5 µg	<i>Mbo</i> I	25 U
T4 DNA ligase	2,000 U*	<i>Mlu</i> I	50 U
T4 DNA polymerase	15 U	<i>Nco</i> I	50 U
T4 polynucleotide kinase [†]	50 U	<i>Nde</i> I	100 U
Toq DNA polymerase	5 U	<i>NgoM</i> I	50 U
Terminal deoxynucleotidyl transferase	45 U	<i>Nbe</i> I	25 U
<i>Acc</i> I	50 U	<i>Not</i> I	50 U
<i>Apa</i> I	100 U	<i>Nru</i> I	50 U
<i>BamH</i> I	100 U	<i>Pst</i> I	100 U
<i>Bcl</i> I	50 U	<i>Sac</i> I	100 U
<i>Bgl</i> I	50 U	<i>Sac</i> II	100 U
<i>BsiW</i> I	70 U	<i>Sal</i> I	100 U
<i>BssH</i> II	20 U	<i>Sca</i> 1	50 U
<i>BstN</i> I	50 U	<i>Spb</i> I	25 U
<i>Dpn</i> I	100 U	<i>Sst</i> I	50 U
		<i>Xbo</i> I	100 U
Enzymes Not Removed	Challenge	Enzymes Not Removed	Challenge
Bacterial alkaline phosphatase	0.6 U	<i>Eae</i> I	15 U
DNA polymerase I (Klenow)	20 U	<i>EcoRV</i>	50 U
Exonuclease I	50 U	<i>Hinf</i> I	50 U
<i>Pfu</i> DNA polymerase	2.5 U	<i>Msp</i> I	100 U
Vent® DNA polymerase	4 U	<i>Pvu</i> I	25 U
Shrimp alkaline phosphatase	1 U	<i>Pvu</i> II	50 U
RNase A (bovine pancreas)	1 µg	<i>Sau3A</i> I	20 U
<i>ApaL</i> I	10 U	<i>Sfi</i> I	50 U
<i>Bgl</i> II	50 U	<i>Sma</i> I	25 U
<i>BsoB</i> I	50 U	<i>Xba</i> I	50 U
<i>Cla</i> I	25 U		

*New England Biolabs unit definition. [†]T4 polynucleotide kinase is not recommended with oligonucleotides, though it works well for dsDNA. Results suggest that this kinase mediates the binding of oligos to the membrane, causing oligo loss. **Note:** T4 RNA ligase is not recommended. This ligase mediates binding of nucleic acids to the membrane in the Micropure-EZ device, causing sample loss.

Ordering Information

Micropure-EZ Enzyme Removers are available in packages of 8, 24 or 100 units. Order devices by catalogue number from the chart below. Microcon Microconcentrators must be purchased separately. They are NOT included with Micropure-EZ Enzyme Removers.

Product	8-Pack	24-Pack	100-Pack
Micropure-EZ device with vials	42528	42529	42530
Microcon devices with Ultracel membrane			
3,000 NMWL	42420	42403	42404
10,000 NMWL	42421	42406	42407
30,000 NMWL	42422	42409	42410
50,000 NMWL	42423	42415	42416
100,000 NMWL	42424	42412	42413

Technical Assistance

For more information, contact the Millipore office nearest you. In the U.S., call **1-800-MILLIPORE** (1-800-645-5476). Outside the U.S., see your Millipore catalogue for the phone number of the office nearest you or go to our web site at www.millipore.com/offices for up-to-date worldwide contact information. You can also visit the tech service page on our web site at www.millipore.com/techservice.

Warranty

This product is covered by Millipore's standard limited warranty, as set forth in Millipore's Bioscience Catalogue.

MILLIPORE

99441, Rev. D, 03/06