EURx® MOLECULAR BIOLOGY PRODUCTS

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GeneMAGNET PCR / DNA Clean-Up Purification Kit

Kit for purification of PCR products / DNA after enzymatic reactions

Cat. no. E3420

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Table of Contents

Content	96 preps E3420-01	Storage/Stability
Orange DX	48 ml	15-25°C
Wash DX1	60 ml	15-25°C
Wash DX2	80 ml	15-25°C
Elution	20 ml	15-25°C
Magnetic Beads	1000 µl	2-8°C
Protocol	1	

Equipment and reagents to be supplied by the user

1. Magnetic stand E0361 for 16 tubes, E0362 for 24 tubes, E0363 for 96-well plate. To be purchased separately.
2. Disposable gloves, pipettes, sterile pipette tips, sterile 1.5-2 ml tubes or 96-well plates (well volume at least 800 µl), a heating block capable of incubation at 56°C, vortex and laboratory rack for the tubes.

Introductory Notes

NOTE 1 · Kit Specification. The kit is suitable for fast cleanup of DNA fragments from PCR and other enzymatic reactions. This kit selectively removes primers below 40 nt and double-stranded DNA below 20 bp. However, common short by-products of not optimal or problematic PCR, known as primer-dimers, also consist of double-stranded DNA. They are produced from self-annealed and extended primers and co-migrate on a gel along with unincorporated single-stranded DNA primers. These double-stranded DNA artefacts co-purify with an expected PCR product, if their length exceeds 20 bp. If the removal of primer-dimers is necessary, we recommend PCR reaction optimization and/or agarose gel electrophoresis followed by isolation of PCR product using our spin-column GeneMATRIX Agarose-Out Purification Kit E3540.

NOTE 2 · Sample amount and buffers volume. In order to achieve satisfying results follow the manual provided on page 4. Maximal Sample Amount for one prep is 150 µl. Sample volume can be increased, but the amount of DNA binding buffers needs to be scaled up. For example: to 300 µl of sample add 900 µl Orange DX and 20 µl magnetic beads. The amount of washes and elution buffer can be used according to the protocol on page 4. Note the limit of the vial maximal volume.

The protocol adaptation to the specific needs is possible with the method optimization. For example: it is possible to carry out the PCR clean-up in PCR reaction plates with the working volume of 200 µl. Use not more than 50 µl PCR volume, add 3 volumes Orange DX, 10 µl magnetic beads and use 200 µl of Wash DX1 and Wash DX2. Before the use of elution buffer remove the remaining drops of washes for the bottom and walls of the wells. The amount of elution buffer can be used according to the protocol on page 4. Do not vortex the plate, mix instead by pipetting to avoid cross-contamination.

NOTE 3 · Kit Compounds Storage. Once the kit is unpacked, store components at room temperature except Magnetic Beads that should be store in 2-8°C. In case of buffer Orange DX and Wash DX1 precipitation, simply warm up in 37°C water bath, until clarified.

NOTE 4 · Maintaining Good Working Practice. All solutions should be kept tightly closed to avoid evaporation and resulting concentration changes of buffer components. To obtain high quality DNA, stick carefully to the protocol provided below.

NOTE 5 · Elution buffer is a low salt solution, that contains no metal ion chealators (e.g. EDTA) that can inhibit subsequent enzymatic reactions. Elution buffer composition is suitable for downstream applications such as digestion with restriction enzymes, phosphorylation, ligation, Sanger sequencing, NGS etc. It is also possible to elute the DNA with Tris-HCl, water or TE.

Protocol

1. Add 3 volumes of orange-coloured **Orange DX** buffer to 1 volume of the DNA sample and mix.
   - *For example, add 300 µl of Orange DX buffer to 100 µl DNA sample.*
   - *Maximal Sample Amount for one prep is 150 µl.*
2. Resuspend **Magnetic Beads** before removing them from the storage tube by vortexing. Add 10 µl of resuspended **Magnetic Beads** to the sample.
3. Mix by vortexing or pipetting for 1 min.
4. Separate the **Magnetic Beads** against the side of the tubes/wells. Wait until all the beads have been attached to the magnets.
5. Remove and discard the supernatant by pipetting.
6. Remove the magnetic stand/transfer tubes to the laboratory rack, add 500 µl of **Wash DX1** and mix by pipetting or vortexing for 10 s.
7. Separate the **Magnetic Beads** against the side of the tubes/wells. Wait until all the beads have been attached to the magnets. Remove and discard the supernatant by pipetting.
8. Remove the magnetic stand/transfer tubes to the laboratory rack, add 300 µl of **Wash DX2** and mix by pipetting or vortexing for 10 s.
9. Repeat steps 7-8.
10. Separate the **Magnetic Beads** against the side of the tubes/wells. Wait until all the beads have been attached to the magnets. Remove and discard the supernatant by pipetting.
    - *Remove all the remaining Wash DX2 solution from the buttom of the tube/well.*
11. Leave the open tubes/plate in magnetic stand and air dry the beads for 15 min.
    - *Wash DX2 contains alcohol, make sure all the solution evaporates before proceding to step 12.*
12. Add 50-100 µl of **Elution** buffer to the tube/well and mix thoroughly by pipetting or vortexing and incubate 2-5 min.
13. Separate the **Magnetic Beads** against the side of the wells. After all the beads have been attached to the magnets transfer the supernatant containing the purified DNA to a suitable tube/plate. DNA is ready for analysis/manipulations. Isolated DNA can be stored either at 2-8°C or at -20°C.

Safety Information

Orange DX

Danger

H302+H332 Harmful if swallowed or if inhaled.
H315 Causes skin irritation.
H319 Causes serious eye irritation.
H334 May cause allergy or asthma symptoms or breathing difficulties if inhaled.
H317 May cause an allergic skin reaction.
P280 Wear protective gloves/protective clothing/eye protection/face protection.
P284 [In case of inadequate ventilation] wear respiratory protection.
P301+P312 If swallowed: call a poison center/ doctor if you feel unwell.
P304+P340 If inhaled: remove person to fresh air and keep comfortable for breathing.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P333+P313 If skin irritation or rash occurs: get medical advice/attention.

Wash DX1

Warning

H226 Flammable liquid and vapour.
H302+H332 Harmful if swallowed or if inhaled.
H315 Causes skin irritation.
H319 Causes serious eye irritation.
P210 Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
P280 Wear protective gloves/protective clothing/eye protection/face protection.
P301+P312 If swallowed: call a poison center/ doctor if you feel unwell.
P302+P352 If on skin: wash with plenty of water.
P304+P340 If inhaled: remove person to fresh air and keep comfortable for breathing.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.

Wash DX2

Danger

H225 Highly flammable liquid and vapour.
H319 Causes serious eye irritation.
P210 Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking.
P280 Wear protective gloves/protective clothing/eye protection/face protection.
P305+P351+P338 If in eyes: rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing.
P403+P235 Store in a well-ventilated place. Keep cool.
P337+P313 If eye irritation persists: get medical advice/ attention.

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SELECTION OF THE KITS DEPENDING ON THE TYPE OF ISOLATED MATERIAL

			ISOLATION OF DNA																			
			E3600	E3585	E3540	E3580	E3510	E3545	E3560	E3555	E3525	E3520	E3595	E3535	E3500	E3565	E3515	E3570	E3575	E3530	E3550	E3551
			MICELLULA DNA[2]	GRAM PLUS & YEAST GENOMIC DNA	AGAROSE – OUT DNA	BACTERIAL & YEAST GENOMIC DNA	BIO – TRACE DNA	BASIC DNA	BONE DNA	CELL CULTURE DNA	FOOD EXTRACT DNA	PCR / DNA CLEAN-UP	PLANT & FUNGI DNA	AGROBACTERIUM PLASMID DNA	PLASMID MINIPREP DNA	QUICK BLOOD DNA	SHORT DNA CLEAN-UP	SOIL DNA	STOOL DNA	SWAB-EXTRACT DNA	TISSUE DNA	TISSUE & BACTERIAL DNA
			AVAILABLE NUMBER OF ISOLATION (PREPS)																			
			50 150	25 100	50 150	50 150	25 100	50 150	25 50	50 150	25 100	50 150	50 150	50 150	50 150	50 150	25 100	50 100	50 100	25 100	50 150	50 150
DNA	GENOMIC	BACTERIA		●		●																●
		YEAST		●		●																
		CELL CULTURE								●											●	●
		PLANT											●									
		FUNGI											●									
		PLANT RICH IN POLYSACCHARIDES[1]											●									
		BLOOD														●						
		SOIL																●				
		STOOL																	●			
		SWAB																		●		
		ANIMAL TISSUES																			●	●
		FFPE TISSUE SECTIONS																			●	●
		RODENT TAILS																			●	●
		HAIR																			●	●
		INSECTS																			●	●
		URINE																			●	●
		BONE							●													
		BIOLOGICAL TRACES					●															
		FOOD									●											
	PLASMID	BACTERIA						●						●	●							
		YEAST				●																
	ISOLATION FROM AGAROSE GELS				●			●														
	PURIFICATION OF PCR PRODUCTS / DNA AFTER ENZYMATIC REACTIONS		●					●				●					●					

All kits contain buffers WASH in ready to use form

1. Additionally required lyse CT buffer (E0324)
2. Kit for creation of emulsions and subsequent DNA purification.

GeneMAGNET PCR / DNA Clean-Up Purification Kit is fast cleanup of DNA fragments from PCR and other enzymatic reactions.

Fragment of sizes from approximately 100 bp to over 15 kb can be obtained in ultrapure form. Contaminants such as: ethidum bromide, primers (below 40 nt), short double-stranded DNA (below 20 bp), RNA, Taq DNA Polymerase, Pfu DNA Polymerase, endo- and exonucleases, DNA-binding and modifying proteins, BSA and other enzymes/proteins, lipids, endotoxins, dyes, detergents, nucleotides, radio- and chemical labels, EDTA, problematic restriction and ligation inhibitors, buffers and salts are effectively removed. Coloured binding buffer is very helpful in simultaneous processing of multiple samples. DNA is then eluted in low salt buffer, e.g.: Tris-HCl, TE or water. Isolated DNA is ready for downstream applications without the need for ethanol precipitation.

- **GeneMAGNET line is based on the use of silica paramagnetic beads (Magnetic Beads) for selective binding of RNA and DNA. The use of specially designed binding and washing buffers enables the efficient purification of highly pure nucleic acids.**

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