

Instructions for Use

Life Science Kits & Assays



innuPREP PCRpure Kit

Order No.:

845-KS-5010010 10 reactions

845-KS-5010050 50 reactions

845-KS-5010250 250 reactions

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It needs not necessarily agree with future versions. Subject to change!

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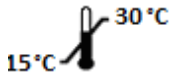






1 Introduction

1.1 Intended use

The innuPREP PCRpure Kit has been designed for the purification and concentration of PCR products from PCR reactions up to 50 μ l. The kit uses Spin Filter technique in combination with a new patented chemistry which requires no washing steps resulting in a very fast and reliable purification of DNA fragments.

1.2 Notes on the use of this manual

For easy reference and orientation, the manual uses the following warning and information symbols as well as the shown methodology:

Symbol	Information
	REF Catalogue number.
	Content Contains sufficient reagents for <N> tests.
	Storage conditions Store at room temperature or shown conditions respectively.
	Consult instructions for use This information must be observed to avoid improper use of the kit and the kit components.
	Expiry date
	Lot number The number of the kit charge.
	Manufactured by Contact information of manufacturer.
	For single use only Do not use components for a second time.
	Note / Attention Observe the notes marked in this way to ensure correct function of the device and to avoid operating errors for obtaining correct results.

The following systematic approach is introduced in the manual:

- The chapters and figures are numbered consecutively.
- A cross reference is indicated with an arrow (e.g. → "Notes on the use of this manual" p. 3).
- Work steps are numbered.

2 Safety precautions

NOTE

Read through this chapter carefully prior to guarantee your own safety and a trouble-free operation.

Follow all the safety instructions explained in the manual, as well as all messages and information, which are shown.

All due care and attention should be exercised in handling the materials and reagents contained in the kit. Always wear gloves while handling these reagents and avoid any skin contact! In case of contact, flush eyes or skin with a large amount of water immediately.



FOR SINGLE USE ONLY!

This kit is made for single use only!

ATTENTION!

Don't eat or drink components of the kit!

The kit shall only be handled by educated personnel in a laboratory environment!

If the buffer bottles are damaged or leaking, wear gloves and protective goggles when discarding the bottles in order to avoid any injuries. This kit could be used with potential infectious samples. Therefore, all liquid waste must be considered as potentially infectious and must be handled and discarded according to local safety regulation.

Please observe the federal, state and local safety and environmental regulations. Follow the usual precautions for applications using extracted nucleic acids. All materials and reagents used for DNA or RNA isolation should be free of DNases or RNases.

ATTENTION!

Do not add bleach or acidic components to the waste after sample preparation!

NOTE

Emergency medical information in English and German can be obtained 24 hours a day from:

Poison Information Center, Freiburg / Germany

Phone: +49 (0)761 19 240.

For more information, please ask for the material safety data sheet (MSDS).

3 Storage conditions

The innuPREP PCRpure Kit should be stored dry at room temperature (15 °C to 30 °C). When stored at room temperature, the kit is stable until the expiration date printed on the label on the kit box.

If there are any precipitates within the provided solutions solve these precipitates by careful warming. Before every use make sure that all components have room temperature.

For further information see chapter "Kit components" (→ p. 7).

4 Functional testing and technical assistance

The Analytik Jena AG guarantees the correct function of the kit for applications as described in the manual. This product has been produced and tested in an ISO 13485 certified facility.

We reserve the right to change or modify our products to enhance their performance and design. If you have any questions or problems regarding any aspects of the innuPREP PCRpure Kit or other Analytik Jena AG products, please do not hesitate to contact us. For technical support or further information in Germany please dial +49 36 41 / 77 94 00. For other countries please contact your local distributor.

5 Product use and warranty

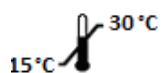
The kit is not designed for the usage of other starting materials or other amounts of starting materials than those, referred to in the manual (→ "Intended use" p. 2) (→ "Product specifications" p. 8). Since the performance characteristics of Analytik Jena AG kits have just been validated for the application described above, the user is responsible for the validation of the performance of Analytik Jena AG kits using other protocols than those described below. Analytik Jena AG kits may be used in clinical diagnostic laboratory systems after the laboratory has validated the complete diagnostic system as required by CLIA' 88 regulations in the U.S. or equivalent regulations required in other countries.

All products sold by the Analytik Jena AG are subjected to extensive quality control procedures and are warranted to perform as described when used correctly. Any problems should be reported immediately.

NOTE




For research use only!

6 Kit components



STORAGE CONDITIONS

All components are stored at room temperature.

	 10	 50	 250
REF	845-KS-5010010	845-KS-5010050	845-KS-5010250
Binding Buffer	6 ml	30 ml	140 ml
Elution Buffer	2 ml	2 x 2 ml	15 ml
Spin Filter	10	50	5 x 50
Receiver Tubes (2.0 ml)	10	50	5 x 50
Elution Tubes (1.5 ml)	10	50	5 x 50
Manual	1	1	1

7 Product specifications

1. Starting material:
 - PCR reaction mixtures (up to 50 μ l)


2. Time for extraction:
 - 3 minutes
 - Based on a new two-step procedure

3. Fragment length:
 - 60 bp–30 kbp

4. Binding capacity:
 - > 20 μ g DNA

5. Rate of recovery:
 - 60–90 %
 - Depending on the length of PCR fragments

8 GHS classification

Component	Hazard contents	GHS Symbol	Hazard phrases	Precaution phrases
Binding Buffer	Propan-2-ol 50–100 %		225, 319, 336	101, 102, 103, 210, 261, 303+361+353, 305+351+338, 405, 501
	Diammoni- um hydro- gen 2- hydroxy- propane- 1,2,3- tricarbox- ylate 0.1-<2.5 %	Danger		

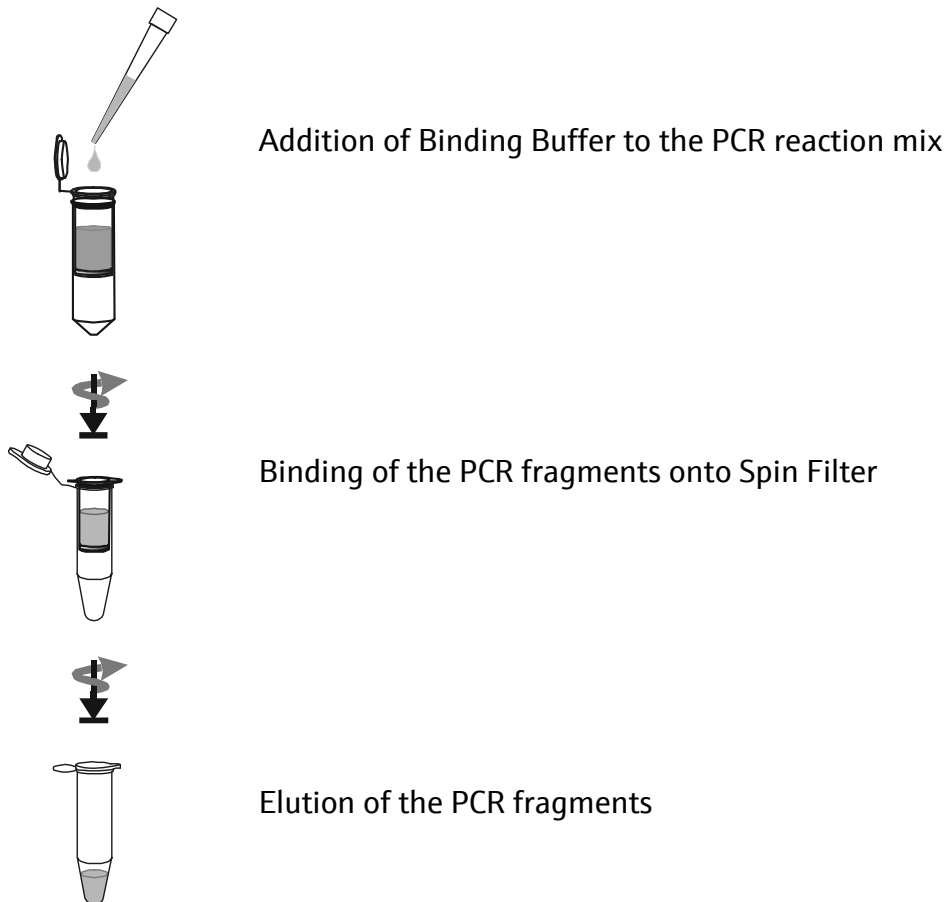
8.1 Hazard phrases

225	Highly flammable liquid and vapor.
319	Causes serious eye irritation.
336	May cause drowsiness or dizziness.

8.2 Precaution phrases

- | | |
|-------------|--|
| 101 | If medical advice is needed, have product container or label at hand. |
| 102 | Keep out of reach of children. |
| 103 | Read label before use. |
| 210 | Keep away from heat, hot surfaces, sparks, open flames and other ignition sources. No smoking. |
| 261 | Avoid breathing dust/fume/gas/mist/vapors/spray. |
| 405 | Store locked up. |
| 501 | Dispose of contents/container in accordance with local/regional/national/international regulations. |
| 303+361+353 | IF ON SKIN (or hair): Take off immediately all contaminated clothing. Rinse skin with water/shower. |
| 305+351+338 | IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. |

9 General procedure for DNA extraction



10 Protocol: Purification and concentration of PCR products from PCR reactions up to 50 μ l

NOTE

Before starting with the purification procedure place a Spin Filter into a 2.0 ml Receiver Tube.

A Binding of the PCR fragments

1. Add 500 μ l Binding Buffer to the Spin Filter.
2. Add up to 50 μ l of your PCR reaction mixture to the Spin Filter which is already pre-filled with the Binding Buffer.
3. Mix Binding Buffer and PCR reaction mixture by pipetting three times up and down. Don't destroy the filter membrane!

Alternatively

Mix 500 μ l Binding Buffer with up to 50 μ l of the PCR reaction mixture very well by pipetting or vortexing outside the Spin Filter in a separate reaction tube.

After this transfer the mixed sample completely onto the Spin Filter.

4. Centrifuge for 3 minutes at 11,000 x g (~11,000 rpm). Discard the Receiver Tube.
-

NOTE

Avoid any contact of the Spin Filter with the flow through.

For maximum purity, discard the flow through after a first centrifugation step for 1 minute at 11,000 x g (~11,000 rpm) and place the Spin Filter back into the Receiver Tube and centrifuge for another 2 minutes at 11,000 x g (~11,000 rpm).

B Elution of the PCR fragments

1. Place the Spin Filter into an Elution Tube.
2. Pipette at least **10 μ l Elution Buffer** or RNase-free water directly onto the center of the Spin Filter.
3. Incubate for 1 minute at room temperature.
4. Centrifuge for 1 minute at 11,000 x g (~11,000 rpm). The Elution Tube now contains the purified PCR fragments.

NOTE

To increase the final DNA yield we recommend an extended incubation time with **Elution Buffer** (up to 5 minutes), which will lead to a slightly higher final yield.

For concentration of PCR fragments it is possible to perform the elution with a lower volume of **Elution Buffer** than the volume of the starting PCR mixture. The minimum volume is 10 μ l.

11 Troubleshooting

Problem / probable cause	Comments and suggestions
Low recovery / poor quality	
Poor elution of DNA	Add the Elution Buffer directly onto the center of the Spin Filter (even if a small elution volume is used).
Poor quality of DNA	<p>After binding of DNA an additional washing step with 80 % ethanol is recommended. Only use pure/non denatured ethanol.</p> <p>Add 600 µl of 80 % ethanol to the Spin Filter and centrifuge for 1 minute at 11,000 x g. Discard the flow through.</p> <p>For removing the ethanol centrifuge for 2 minutes at 11,000 x g.</p> <p>Continue with the elution step as described.</p>
Problems with mineral oil	Increase volume of Binding Buffer .

12 Related products

Name	Amount	Order No.
Products for PCR & Gel Electrophoresis		
innuPREP Gel Extraction Kit	10 rxn	845-KS-5030010
	50 rxn	845-KS-5030050
	250 rxn	845-KS-5030250
innuPREP DOUBLEpure Kit	10 rxn	845-KS-5050010
	50 rxn	845-KS-5050050
	250 rxn	845-KS-5050250
innuTaq DNA Polymerase (5 U/ μ l)	500 U	845-EZ-1000500
50x inNucleotide Mix (12.5 mM)	2x 0.5 ml	845-AS-9000100
inNucleotide Set (100 mM)	4x 0.25 ml	845-AS-1100250
innuMIX rapidPCR MasterMix	100 rxn	845-AS-1600100
	200 rxn	845-AS-1600200
innuMIX Standard PCR MasterMix	100 rxn	845-AS-1700100
	200 rxn	845-AS-1700200
innuMIX Green PCR MasterMix	100 rxn	845-AS-1400100
	200 rxn	845-AS-1400200
innuSTAR 100 bp DNA Ladder Express	500 μ l	845-ST-1010100
	5x 500 μ l	845-ST-1010500
innuSTAR 1 kb DNA Ladder Express	500 μ l	845-ST-1020100
	5x 500 μ l	845-ST-1020500
6x Loading Dye Bromophenol Blue	3x 1.0 ml	845-ST-3010003
	6x 1.0 ml	845-ST-3010006

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Subject to changes in design and scope of delivery as well as further technical development.