Nexxo-Prep PCR Clean-Up mini

DNA isolation, by spin-column system, for PCR products, restriction digestion or cDNA synthesis purification.

PCR products, restriction digestion or cDNA synthesis purification can be achieved in approx. 5 minutes, with an effective removal of primers, enzymes, unincorporated nucleotides, dyes or other impurities, with a yield of up to 95 %. (DNA size: 80 bp – 30 kb, max. 100 µl)

Eluted DNA is ready for down-stream applications and can directly be used for sequencing, cloning, ligation, enzymatic digestion, hybridization, labelling, etc., or can be stored for future use.

I. Kit components

	10 preps[1]	50 preps	250 preps
Binding Solution S1 (Solution de fixation S1)	X	12 ml (final volume: 32 ml)	63 ml (final volume: 163 ml)
Elution Buffer (Tampon d'élution)	X	3 x 2 ml	30 ml
Spin Filter (Filtres de centrifugation)	X	50	5 x 50
1,5 ml Receiver Tubes (Tubes receveurs 1,5 ml)	X	50	5 x 50
2,0 ml Receiver Tubes (Tubes receveurs 2,0 ml)	X	50	5 x 50
User guide	X	1	1
Art. No.	2037.10	2037.50	2037.250

[1] The kit Nexxo-Prep PCR Clean-Up mini 10 preps is supplied in the form of the Nexxo-Prep duo, Gel Extraction & PCR Clean-Up 10 preps kit. The Nexxo-Prep duo, Gel Extraction & PCR Clean-Up kit comprises the components of the Nexxo-Prep PCR Clean-Up mini kit and the Nexxo-Prep Gel Extraction DNA mini kit. This kit allows to carry out the indicated number of isolation (10, 50 or 250) without isolation type limitations (gel/PCR).

Required material and equipment not included in this kit

- Isopropanol >99.7 % (propanol-2 >99.7 %)
- Microtubes (1.5 ml and 2.0 ml)
- Microcentrifuge (min. 11100 x g)
- Pipettes with corresponding tips
- Disposable gloves

Some components are delivered in concentrated form and have to be diluted appropriately (see chapter « Reagents and buffer solutions preparation », page 2).

II. Storage and stability

All kit components should be stored at room temperature (15-30 °C).

Isopropanol is a volatile compound. Keep **Binding Solution S1** tightly closed.

Note: all kit components are stable for at least 12 months.

Check solutions for absence of precipitates before use. If necessary redisolve precipitates by heating (< 30 °C).

III. Reagents and buffer solutions preparation

1. Kit 10 extractions:

- Add 7 ml of >99.7 % isopropanol to the **Binding Solution S1**.

2. Kit 50 extractions:

- Add 20 ml of >99.7 % isopropanol to the **Binding Solution S1.**

3. Kit 250 extractions:

- Add 100 ml of >99.7 % isopropanol to the **Binding Solution S1.**

IV. Protocol: purification of PCR products, restriction digestions or cDNA synthesis products

Before starting

- Ensure that reagents/buffers preparation has been done (see chapter "Reagents and buffer solutions preparation", page 2).
- Ensure that all components are at room temperature and check solutions for absence of precipitates before use. If necessary resuspend precipitates by heating (< 30 °C).

Note: To prevent contamination, use new pipet tip for each pipetting step.

Depending on sample characteristics/volume, start with step 1a, 1b or 1c

1a	**DNA adsorption to the Spin filter** (sample volume: max. 50 µl)
	• Add 250 µl of **Binding Solution S1** to the sample (max. 50 µl) *Note: if the sample contains mineral oil (e.g. PCR samples …), start with step 1b instead of step 1a.* • Mix completely by pipetting or vortex • Put a **Spin Filter** into a **2.0 ml Receiver tube** • Transfer the whole sample mixture, containing **Binding Solution S1,** into the **Spin Filter** • Centrifuge 2 min. at 11000 x g • Proceed with step 2 « **DNA elution** »

1b	**DNA adsorption to the Spin filter** (sample volume: 50 - 100 µl)
	• Add 500 µl of **Binding Solution S1** to the sample (50 - 100 µl) • Mix completely by pipetting or vortex • Put a **Spin Filter** into a **2.0 ml Receiver tube** • Transfer the whole sample mixture, containing **Binding Solution S1,** into the **Spin Filter** • Centrifuge 2 min. at 11000 x g • Discard the flow-through and put the **Spin Filter** back into the **2.0 ml Receiver tube** • Centrifuge 3 min. at 11000 x g • Proceed with step 2 « **DNA elution** »

Steps 1c and 2 →

1c	**DNA adsorption to the Spin filter** (sample volume: max. 200 µl)
	• Add 1000 µl of **Binding Solution S1** to the sample (max. 200 µl) *Note: if the sample contains mineral oil (e.g. PCR samples …), increase volume of **Binding Solution S1** by 500 µl.* • Mix completely by pipetting or vortex • Put a **Spin Filter** into a **2.0 ml Receiver tube** • Transfer approx. half of the sample mixture, containing the **Binding Solution S1,** into the **Spin Filter** • Centrifuge 2 min. at 11000 x g • Discard the flow-through and put the **Spin Filter** back into the **2.0 ml Receiver tube** • Transfer the remaining half of the sample mixture, containing the **Binding Solution S1,** into the **Spin Filter** • Centrifuge 2 min. at 11000 x g • Discard the flow-through and put the **Spin Filter** back into the **2.0 ml Receiver tube** • Centrifuge 3 min. at 11000 x g • Proceed with step 2 « **DNA elution** »

2	**DNA elution**
	• Put the **Spin filter** into a new **1.5 ml Receiver tube** • Add, at least, 10 µl of **Elution Buffer** (or ddH_2O, or Tris buffer) into the center of the filter • Incubate 1 min. at room temperature • Centrifuge 1 min. at 11000 x g *Note: increasing the incubation time by 5 min. enhances slightly the yield.*

Eluted DNA is ready for down-stream applications and can directly be used for sequencing, cloning, ligation, enzymatic digestion, hybridization, transformation, labelling, etc., or can be stored at 4 °C for several weeks.

For long-time storage, store eluted DNA at -20 °C.

Note: this kit has been calculated for samples up to 100 µl. The protocol for 200 µl samples (1c) reduces the total number of purification.