

NucleoSpin® Plasmid EasyPure

I) Preparations

- Add total content of Liquid RNase A to Buffer A1, mix thoroughly, and store at 2–8 °C.
- Add indicated volume of 96–100 % ethanol to Buffer AQ (Concentrate).
- Before each preparation, check Buffer A2 for precipitated SDS. Dissolve any precipitate for several minutes at 30–40 °C and store buffer at room temperature.

II) Culture volume

Cells can be grown in LB or rich media (e.g., TB, CircleGrow) but make sure not to exceed the recommended culture volumes (see table below). Using too much cell mass will exhaust the lysis capacity of the buffer system and result in poor or even no yield. If nevertheless more cells have to be processed (e.g., for low copy-number plasmids), increase lysis buffers A1, A2, and A3 proportionally.

OD _{600 nm}	1	2	3	4	5	6
Max. volume	15 mL	8 mL	5 mL	4 mL	3 mL	2 mL

For further information, please download the **NucleoSpin® Plasmid EasyPure** user manual at the relating webpage www.mn-net.com/plasmid





III) Protocol overview

1. Pellet cells into a microcentrifuge tube and discard supernatant.
2. Add **150 µL Buffer A1** and vortex to resuspend cells completely.
3. Add **250 µL Buffer A2** and invert 5 times – **DO NOT VORTEX!**
4. Incubate up to **2 min** at **room temperature** to lyse cells.
5. Add **350 µL Buffer A3** and invert **until lysate has turned colorless**.
6. Centrifuge for **3 min** at **full speed** (> 12,000 x g) to pellet precipitate.
7. Put a NucleoSpin® Plasmid EasyPure Column into a Collection Tube (2 mL).
8. Load clear supernatant onto the spin column.
9. Centrifuge for **30 s** at **1,000–2,000 x g** and discard flow-through.
10. Add **450 µL Buffer AQ** to the spin column.
11. Centrifuge for **1 min** at **full speed** (> 12,000 x g) and discard collection tube.
REPEAT IF SPIN COLUMN IS STILL WET!
12. Put spin column into a new 1.5 mL microcentrifuge tube (not provided).
13. Add **50 µL Buffer AE** onto the membrane and **incubate for 1 min***.
14. Centrifuge for **1 min** at **full speed** (> 12,000 x g).

* For larger constructs or large amounts of plasmid, elution efficiency can be increased by incubating spin cup with hot buffer at 70 °C, increasing the elution volume, or eluting several times with fresh buffer.

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IV) Safety instructions

Component	Hazard contents	GHS symbol	Hazard phrases	Precaution phrases
Inhalt	Gefahrstoff	GHS Symbol	H-Sätze	P-Sätze
A2	Sodium hydroxide 0.5–2% Natriumhydroxid 0.5–2%		Warning Achtung	290, 315, 319 234, 280, 302+352, 305+351+338, 332+313, 337+313, 390, 406
A3	Guanidine hydrochloride Guanidinhydrochlorid		Warning Achtung	302, 319 280, 301+312, 305+351+338, 330, 337+313
RNase A	RNase A RNase A	 	Danger Gefahr	317, 334 261, 302+352, 304+340, 333+313, 342+311, 363

Hazard phrases

H 290 May be corrosive to metals. *Kann gegenüber Metallen korrosiv sein.* **H 302** Harmful if swallowed. *Gesundheitsschädlich bei Verschlucken.* **H 315** Causes skin irritation. *Verursacht Hautreizungen.* **H 317** May cause an allergic skin reaction. *Kann allergische Hautreaktionen verursachen.* **H 319** Causes serious eye irritation. *Verursacht schwere Augenreizung.* **H 334** May cause allergy or asthma symptoms or breathing difficulties if inhaled. *Kann bei Einatmen Allergie, asthmaartige Symptome oder Atembeschwerden verursachen.*

Precaution phrases

P 234 Keep only in original container. *Nur im Originalbehälter aufbewahren.* **P 261** Avoid breathing dust. *Einatmen von Staub vermeiden.* **P 280** Wear protective gloves/eye protection. *Schutzhandschuhe/Augenschutz tragen.* **P 301+312** IF SWALLOWED: Call a POISON CENTER/doctor!...*if you feel unwell.* **BEI VERSCHLUCKEN:** Bei Unwohlsein GIFTINFORMATIONSZENTRUM / Arzt /... anrufen. **P 302+352** IF ON SKIN: Wash with plenty of water!... **BEI KONTAKT MIT DER HAUT:** Mit viel Wasser!... waschen. **P 304+340** IF INHALED: Remove victim to fresh air and keep at rest in a position comfortable for breathing. **BEI EINATMEN:** An die frische Luft bringen und in einer Position ruhigstellen, die das Atmen erleichtert. **P 305+351+338** IF IN EYES: Rinse continuously with water for several minutes. Remove contact lenses if present and easy to do – continue rinsing. **BEI KONTAKT MIT DEN AUGEN:** Einige Minuten lang behutsam mit Wasser spülen. Vorhandene Kontaktlinsen nach Möglichkeit entfernen. Weiter spülen. **P 330** Rinse mouth. *Mund ausspülen.* **P 332+313** If skin irritation occurs: Get medical advice / attention. *Bei Hautreizung: Ärztlichen Rat einholen / ärztliche Hilfe hinzuziehen.*

Precaution phrases

P 333+313 If skin irritation or a rash occurs: Get medical advice / attention. *Bei Hautreizung oder -ausschlag: Ärztlichen Rat einholen / ärztliche Hilfe hinzuziehen.* **P 337+313** Get medical advice / attention. *Bei anhaltender Hautreizung: Ärztlichen Rat einholen / ärztliche Hilfe hinzuziehen.* **P 342+311** If experiencing respiratory symptoms: Call a POISON CENTER/ doctor!... *Bei Symptomen der Atemwege: GIFTINFORMATIONSZENTRUM /Arzt!... anrufen.* **P 363** Wash contaminated clothing before reuse. *Kontaminierte Kleidung vor erneutem Tragen waschen.* **P 390** Absorb spillage to prevent material damage. *Verschüttete Menge aufnehmen, um Materialschäden zu vermeiden.* **P 406** Store in a corrosive resistant container with a resistant inner liner. *In korrosionsbeständigem Behälter mit korrosionsbeständiger Auskleidung aufbewahren.*

V) Product use restriction/warranty

NucleoSpin® Plasmid EasyPure is developed, designed, and sold for research purpose only. We offer a one year warranty from date of delivery that our products will conform to applicable specifications set forth in the product specifications if not sold to persons set forth in § 13 of German Civil Code. In such a case, the provisions of the German Civil Code shall be valid. This warranty does not cover defects in or damage to the products which are due to improper installation or maintenance, misuse, neglect or any use other than ordinary commercial application.

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VI) Ordering information

Product	Pack of	REF
NucleoSpin® Plasmid EasyPure	10 / 50 / 250 preps	740727.10 / .50 / .250
NucleoSpin® Plasmid Buffer Set (for the isolation of low copy plasmids)	1	740953
Liquid RNase A	250 mg	740397

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