Plasmid purification products from MACHEREY-NAGEL

MN guide to plasmid purification

Find the optimal solution

NucleoSpin®

Superior yields
Outstanding purities
Time-saving procedures

... for reliable downstream applications

MACHEREY-NAGEL

www.mn-net.com



MN technologies for plasmid purification

NucleoBond®

Anion-exchange technology





DNA is bound to the anion-exchanger matrix under low-pH conditions

Interaction between positively charged anion-exchanger group and negatively charged DNA backbone



Stringent washing with increasing salt concentration to remove contaminants



DNA is eluted with high-pH buffer



Desalting / Concentration: Alcohol precipitation of eluted DNA

DNA is collected by centrifugation or by using the NucleoBond® Finalizer

NucleoSpin® Silica-membrane technology





Sample lysis, release of DNA from cells, tissue, etc. in the presence of chaotropic salts



DNA is bound to the silica membrane under high-salt conditions

Interaction between DNA (hydrate shell is reversibly removed by chaotropic salt) and silica membrane

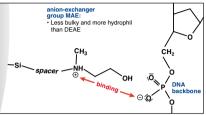


Contaminants are washed away under high-salt and/or ethanolic conditions to keep the DNA bound to the membrane $\,$

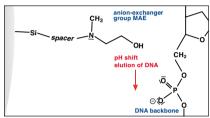


DNA is eluted in low-salt buffer or water, DNA is ready to use for downstream applications





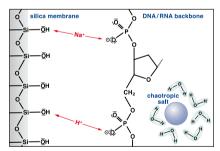
Principle of binding: Ionic bond



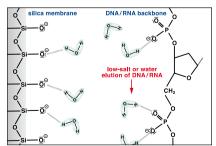
Principle of elution: pH shift

Features/Results

- Ultra-pure, transfection-grade plasmid DNA
- The new generation of anion exchangers
- Xtra fast, Xtra high yield, Xtra convenient
- NucleoSpin® Plasmid / Plasmid (NoLid) _______6
 NucleoSpin® Plasmid QuickPure _______7
 NucleoSpin® 8/96 Plasmid 8



Principle of binding: Removing of hydrate shell Hydrogen bonds/Salt bridges



Principle of elution: Reconstitution of hydrate shell

Features/Results

- · Sequencing and PCR-grade plasmid DNA
- No alcohol precipitation necessary
- Fast and easy procedure

NucleoBond® Xtra Midi • NucleoBond® Xtra Maxi Superior plasmid Midi and Maxi kits – transfection-grade

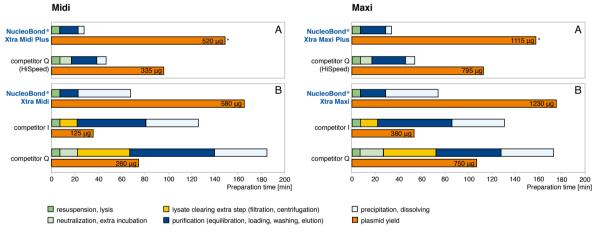
- ► Highest speed 30 min (Midi)/35 min (Maxi)
 Optimal column design and inserted filters avoid inconvenient syringes
- Highest yield typically 250 μg for Midi and 1000 μg for Maxi Improved silica material – patented design!
- High purity transfection-grade plasmid DNA Established anion-exchange technology
- NucleoBond® Xtra Plus kits for super high-speed version
 NucleoBond® Finalizer for omitting time-consuming centrifugation after DNA precipitation



Product at a glance

Sample material	Midi < 200 mL (high cop < 400 mL (low cop		Maxi < 600 mL (high cop < 1200 mL (low cop	
Vector size	< 300 kbp		< 300 kbp	
Typical yield	250 μg		1000 μg	
Precipitation	Midi Centrifugation	Midi Plus NucleoBond® Finalizer	Maxi Centrifugation	Maxi Plus NucleoBond® Finalizer
Preparation time	70 min/prep	30 min/prep	75 min/prep	35 min/prep
Applications	Transfection, cloni	ng, plasmid banks, etc.	Transfection, cloning	ng, plasmid banks, etc.

Application data



^{*} Yield of plasmid DNA is slightly lower due to residual DNA remaining on the desalting tool (compared to kits without desalting tool).

Highest yield in less time - comparison to anion-exchange competitor kits

Plasmid DNA was isolated following each manufacturer's protocol using maximum culture volume. Plasmid DNA yield (III) was determined after precipitation. The comparison shows the results of the kits with NucleoBond® Finalizer as desalting tool (Fig. A) and without desalting tool (Fig. B).

Ordering information

Product	Preps	REF
NucleoBond® Xtra Midi	10/50/100	740410.10/.50/.100
NucleoBond® Xtra Midi Plus (incl. Finalizer)	10/50	740412.10/.50
NucleoBond® Xtra Maxi	10/50/100	740414.10/.50/.100
NucleoBond® Xtra Maxi Plus (incl. Finalizer)	10/50	740416.10/.50
NucleoBond® Xtra Combi Rack	-	740415

www.mn-net.com — 3 →



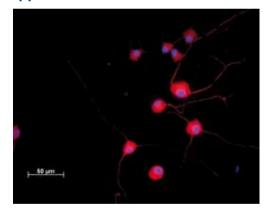
NucleoBond® Xtra Midi EF • NucleoBond® Xtra Maxi EF Superior plasmid kit for transfection of sensitive cell lines – endotoxin-free

- Highest purity endotoxin-free transfection of sensitive cell lines Patented endotoxin removal
- Highest speed 45 min (Midi)/50 min (Maxi)
 No extra incubation for endotoxin removal
 Optimal column design and inserted filters avoid inconvenient syringes
- Highest yield typically 250 μg for Midi and 1000 μg for Maxi Improved silica material patented design!
- NucleoBond® Xtra Plus EF kits for super high speed version
 NucleoBond® Finalizer for omitting time-consuming centrifugation after DNA precipitation

Product at a glance

Sample material	Midi EF < 200 mL (high cop < 400 mL (low cop		Maxi EF < 600 mL (high cop < 1200 mL (low co	-,
Vector size	< 300 kbp		< 300 kbp	
Typical yield	250 μg		1000 μg	
Precipitation	Midi EF Centrifugation	Midi Plus EF NucleoBond® Finalizer	Maxi EF Centrifugation	Maxi Plus EF NucleoBond® Finalizer
Preparation time	85 min/prep	45 min/prep	90 min/prep	50 min/prep
Applications	Transfection, clonic	ng, plasmid banks, etc.	Transfection, cloning	ng, plasmid banks, etc.

Application data



Transfection of PC12 cells with plasmid DNA purified using NucleoBond® Xtra EF

Plasmid purification: 9.2 kbp plasmid, bacterial strain: E. coli DH5α, purification with NucleoBond® Xtra Midi EF

Transfection experiment: highly sensitive PC12 cells, in vitro cultivation, transfection with 0.5 μ g plasmid DNA, transfection method: Lipofectamine™ LTX Reagent (1 μ L) + 100 μ L DMEM, cells were imaged by fluorescence microscopy 24 h post-transfection

Results: Cells show transient expression of Phogrin (PTP NP, Protein Tyrosine Phosphatase), localized by red fluorescent antibody, nuclei are localized by DAPI staining (violet)

Data kindly provided by AG Knauer, Duisburg-Essen University, Institute for Molecular Biology II, Essen, Germany

Ordering information

Product	Preps	REF
NucleoBond® Xtra Midi EF	10/50	740420.10/.50
NucleoBond® Xtra Midi Plus EF (incl. Finalizer)	10/50	740422.10 / .50
NucleoBond® Xtra Maxi EF	10/50	740424.10 / .50
NucleoBond® Xtra Maxi Plus EF (incl. Finalizer)	10/50	740426.10 / .50
NucleoBond® Xtra Combi Rack	-	740415



4

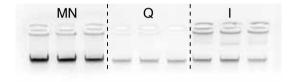
NucleoBond® Xtra BAC Kit for large construct DNA

- High yield up to 150 μg BAC DNA Optimal silica material for large constructs
- High speed 75 minutes per prep
 Optimal column design and inserted filters for high flow rates and parallel lysate clearing and loading
- High purity transfection-grade BAC DNA Established anion-exchange technology



Sample material250–750 mL E. coli cultureVector size< 300 kbp</td>Typical yield10–150 μgPreparation time75 min/prepApplicationsTransfection, cloning, restriction analyses, sequencing

Application data





Superior yield in less time - comparison to competitor kits

BAC DNA (300 kbp) was isolated in triplicate from 500 mL E.~coli DH5 α using NucleoBond® Xtra BAC and competitor products (Q and I). After precipitation, BAC DNA was reconstituted in 1000 μ L TE buffer and 5 μ L of each sample was used for analysis on a 1 % TAE-agarose gel.

Obtained yields: MN: 150 μg, Q: 44 μg, I: 75 μg

Suitable for restriction digestion

Restriction digestion of BAC DNA isolated with NucleoBond® Xtra BAC. BAC DNA samples were purified from overnight cultures of *E. coli* DH5α transformed with a pBAC10 clone. Approximately 3 μg of DNA from each sample was digested with 3 units of *Mspl*, *Hin*dIII, or *Eco*RI at 37 °C for 2 hours. Digestions were analysed on a 1 % TAE agarose gel. Lane 1: undigested BAC DNA, lane 2: *Mspl* digested, lane 3: *Hin*dIII digested, lane 4: *Eco*RI digested, lane M: marker.

Ordering information

Product	Preps	REF
NucleoBond® Xtra BAC	10/25	740436.10/.25
NucleoBond® Xtra Combi Rack	-	740415



NucleoSpin® Plasmid · NucleoSpin® Plasmid (NoLid) High-yield plasmid Mini prep – molecular biology-/sequencing-grade

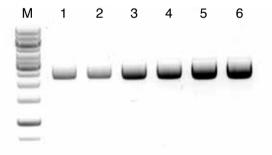
- Highest yield
 Up to 40 µg plasmid DNA
- Highest quality Phred score of 20 (up to 1000 bp)
- Fast procedure
 18 Mini preps in 25 min
- ▶ Choose between columns with or without a lid



Product at a glance

Sample material	1–5 mL <i>E. coli</i> cultures (standard protocol) 5–10 mL <i>E. coli</i> cultures*
Vector size	< 15 kbp
Typical yield	< 25 μg (from 1–5 mL) < 40 μg (from 5–10 mL)*
Elution volume	50 μL
Binding capacity	60 μg
Applications	Sequencing, cloning, PCR, restriction analysis, transformation, enzymatic modifications

Application data



High-yield plasmid Mini prep

Plasmid DNA isolation (pUC18) from *E. coli* DH5 α using NucleoSpin® Plasmid. For the isolation 2 mL (lane 1–2), 5 mL (lane 3–4) and 8 mL (lane 5–6) LB cultures were used and analyzed on an agarose gel (2 μ L of each eluate). Lane M: marker.

Ordering information

Product	Preps	REF
NucleoSpin® Plasmid	10/50/250	740588.10/.50/.250
NucleoSpin® Plasmid (NoLid)	50/250	740499.50 / .250
NucleoSpin® Plasmid Buffer Set*	300	740953

^{*} If larger culture volumes are frequently used, increased buffer volumes are required. Please see "ordering information"



6

NucleoSpin® Plasmid QuickPure

High-speed plasmid Mini prep – molecular biology-/sequencing-grade

Highest speed

18 Mini preps in 11 min

▶ Highest quality

Phred score of 20 (up to 1000 bp)

Reliable yield

Up to 15 µg plasmid DNA

Product at a glance

Sample material

Vector size

< 15 kbp

Typical yield

< 15 μg

Elution volume

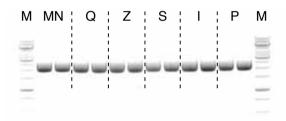
50 μL

Binding capacity

Applications

Sequencing, cloning, PCR, restriction analysis, transformation, enzymatic modifications

Application data



Comparison of plasmid yield

Plasmid DNA isolation (pUC18) from a 3 mL LB *E. coli* DH5 α culture. Two isolations were each performed according to the manufacturer's protocol. For the comparison 4 μ L of each 50 μ L eluate were loaded on a 1 % TAE agarose gel. The yield of NucleoSpin® Plasmid QuickPure is higher or equal to competitors Q, Z, S, I, P. Lane M: marker.



Just three steps = Less than 10 min per prep

Ordering information

Product	Preps	REF
NucleoSpin® Plasmid QuickPure	10/50/250	740615.10/.50/.250

www.mn-net.com — 7 —



NucleoSpin® 8/96 Plasmid · NucleoSpin® 8/96 Plasmid Core Kit

High-throughput plasmid Mini prep - molecular biology-/sequencing-grade

- ▶ Suitable for up to 5 mL bacterial culture
- Convenient filtration of bacterial lysates NucleoSpin® 8 Filter Strips/96 Filter Plate Optimized filter material minimizes the risk of filter clogging
- ▶ Minimizes risk of cross-contamination MN Wash Plate technology
- Core Kit versions available Cost savings - kits with basic contents*



Product at a glance

Format	8-well strips/96-well plates
Processing	Manual and automated, vacuum or centrifugation**
Lysate clarification	8-well filter strips / 96-well filter plates
Sample material	1–5 mL <i>E. coli</i> culture
Vector size	<15 kbp
Typical yield	4–6 μg/mL <i>E. coli</i> culture
Elution volume	75–150 μL
Preparation time	45 min/6 strips or 1 plate
Binding capacity	20 μg
Applications	Sequencing, cloning, PCR, restriction analysis, transformation, enzymatic modifications

Ordering information

Product	Preps	REF
NucleoSpin® 8 Plasmid	12 x 8/60 x 8	740621/.5
NucleoSpin® 8 Plasmid Core Kit*	48 x 8	740461.4
NucleoSpin® 96 Plasmid	1 x 96/4 x 96/24 x 96	740625.1/.4/.24
NucleoSpin® 96 Plasmid Core Kit*	4 x 96	740616.4

- Core kit contents are focused on automation platforms. Additional accessories can be combined as needed. Please check www.mn-net.com or contact our Technical Support.
- ** Support protocol available at Technical Support (tech-bio@mn-net.com)

Your local distributor

Trademarks

MACHEREY-NAGEL: NucleoSpin®, NucleoBond® Other companies: Lipofectamin™ [Invitrogen]

www.mn-net.com

MACHEREY-NAGEL



Tel.: +49 24 21 969-0 Fax: +49 24 21 969-199 E-mail: info@mn-net.com

Switzerland: **MACHEREY-NAGEL AG** Tel.: +41 62 388 55 00 Fax: +41 62 388 55 05 E-mail: sales-ch@mn-net.com

MACHEREY-NAGEL GmbH & Co. KG · Neumann-Neander-Str. 6-8 · 52355 Düren · Germany

MACHEREY-NAGEL EURL Tel.: +33 388 68 22 68 +33 388 51 76 88 Fax: E-mail: sales-fr@mn-net.com MACHEREY-NAGEL Inc.

Tel.: +1 484 821 0984 Fax: +1 484 821 1272 E-mail: sales-us@mn-net.com

