# Plasmid DNA Purification from small to large scale

## **NucleoBond® Plasmid Purification Kits**

1. NucleoBond® PC Kits Five column sizes are available for purification of up to 10 mg

of ultra-pure high- and low copy plasmid DNA

2. NucleoBond® PC EF Kits Four column sizes are available for purification of up to 100 mg

of endotoxin-free plasmid DNA for transfection of endotoxin-

sensitive cells, gene therapy, and vaccination

3. NucleoBond® BAC 100 Kit Increased volumes of lysis buffers and RNase A for purification

of large constructs, e.g. BACs, PACs, P1s, cosmids

## 1. NucleoBond® PC Kits

NucleoBond® AX is a silica-based anion exchanger developed and manufactured by MACHEREY-NAGEL for routine separation of different classes of nucleic acids, covered by **European patent E.P. 0496822**. The extraordinary high charge density on its hydrophilic, macroporous surface results in a salt concentration range for binding and elution of nucleic acids which is much larger than on conventional anion exchangers.

All NucleoBond® AX columns are resistant to organic solvents like alcohol, chloroform and phenol. They are free of RNase and DNase.

NucleoBond® AX columns are easy to handle. Solutions are pipetted or just poured into the columns. Their flow through the exchanger bed is by gravity. The columns do not run dry.

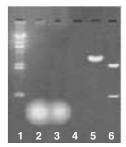


#### **Features**

- anion-exchange chromatography
- ✓ fast and versatile separation system
- working range from nanograms to (milli) grams DNA
- ✓ recovery of plasmid DNA > 90%
- NucleoBond® folded filters eliminate the centrifugation step – no shearing even for large constructs
- ✓ approved and easy procedures
- ✓ structural integrity > 92% ccc monomer

# Purification of plasmid DNA (pUC19) using NucleoBond® AX 100

As can be seen in the figure, this procedure yields high pure plasmid DNA that is free of contaminants like genomic DNA or RNA.



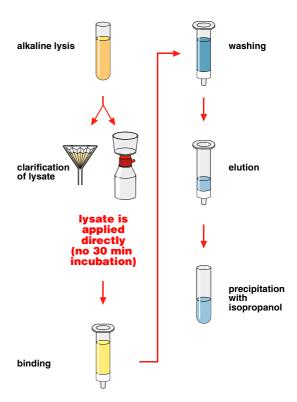
- 1 % agarose gel (TAE)
- 1: marker
- 2: flow-through
- 3: 1st washing step
- 4: 2<sup>nd</sup> washing step
- 5: elution
- 6: purified plasmid digested with EcoRI / Sspl

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## 2. NucleoBond® PC EF Kits

#### **Procedure**



#### **Features**

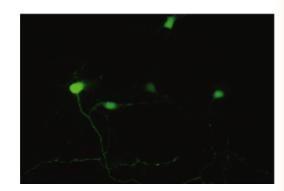
- ✓ covered by US patent # US 6, 428703 B1
- no additional steps required minimal hands-on-time
- endotoxin-free plasmids for transfection, gene therapy, and vaccination
- ✓ DNA with less than 0,05 EU/µg DNA
- columns available for up to 500 μg, 2000 μg, 10 mg, and 100 mg
- folded filters/bottle top filters included for clarification of bacterial lysate

## **Application Data**

# Endotoxin-free DNA for transfection of primary neurons

Expression of GFP (Green Fluorescent Protein) in mature hippocampal neurons (>7 days in culture) grown at low density on poly-L-lysine coated glass coverslips.

The neurons were transiently transfected with a cDNA (EF-free plasmid DNA was purified with NucleoBond® PC 10000 EF) encoding GFP using a modified calcium phosphate protocol (Koehrmann et al, 1999) and the expression pattern was viewed 16 hours post-transfection.



### 3. NucleoBond® BAC 100 Kit

#### **Customer Testimonial**

"Only one of the kits we tried worked; that was the NucleoBond® kit. The vector DNA purified with this kit gave us a transformation efficiency that was comparable to that reported in the literature using BAC vector DNA prepared by two or three rounds of CsCl gradient centrifugation."

Teh-hui Kao, Ph.D. and Andrew G. McCubbin, Ph.D., Department of Biochemistry and Molecular Biology, Eberly College of Science Penn State University (USA)

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