

# PureLink™<sup>TM</sup> 96

## PureLink™<sup>TM</sup> 96 HQ Mini Plasmid Purification Kit

Automated, high throughput purification of plasmid DNA

The ever-increasing demand for high throughput (HTP) applications for molecular techniques, particularly plasmid DNA purification methods, has led to a number of new automated purification procedures in life sciences laboratories. To meet the needs of these automated methods, Invitrogen offers you the PureLink™96 HQ (High Quality) Mini Plasmid Purification Kit. This kit provides a universal 96-well plate design that not only is compatible with most automated liquid handling platforms but also has the ability to deliver the high quality plasmid DNA necessary for today's stringent applications. Use the PureLink™96 HQ Mini Plasmid Purification Kit with your robotic system to quickly

generate high mini-prep yields with low genomic DNA contamination and non-detectable RNA contamination, without compromising quality.

### Workhorse for plasmid prep purification

The PureLink™96 HQ Mini Plasmid Purification Kit is based on an alkaline/SDS cell lysis procedure, followed by neutralization of the lysate and selective plasmid DNA binding to glass microfiber filters. The bound DNA is then washed to remove impurities from the original sample (such as genomic DNA and RNA), and the clean DNA is eluted in a low-salt buffer. This DNA isolation method is typically faster and easier to perform than anion exchange methods. There's no need

for alcohol precipitation or desalting, making it ideal for HTP studies.

### Validated HTP procedure using Tecan automated robot

To get you started on your HTP applications, we have validated a robust procedure to isolate plasmid DNA from bacteria in a 96-well format using the PureLink™96 HQ Mini Plasmid Purification Kit and a Tecan Freedom EVO® automated liquid handling platform with Freedom EVOware® software. The Freedom EVO separation system with the Te-VacS™ vacuum separation module allows for walkaway automated processing. Pipetting is performed by the liquid handling (LiHa) arm, while the robotic manipulator (RoMa) arm performs all transports of the plates and assembling steps of the Te-VacS vacuum manifold.

Briefly, the PureLink™96 HQ Mini Plasmid Purification Kit script opens with the deck layout as shown in Figures 1 and 2. The pelleted cell culture plate is placed on the Te-Shake™ orbital mixer. Cell resuspension, lysis, and neutralization buffers are added sequentially, with shaking between each buffer addition. The binding and clarification plates are

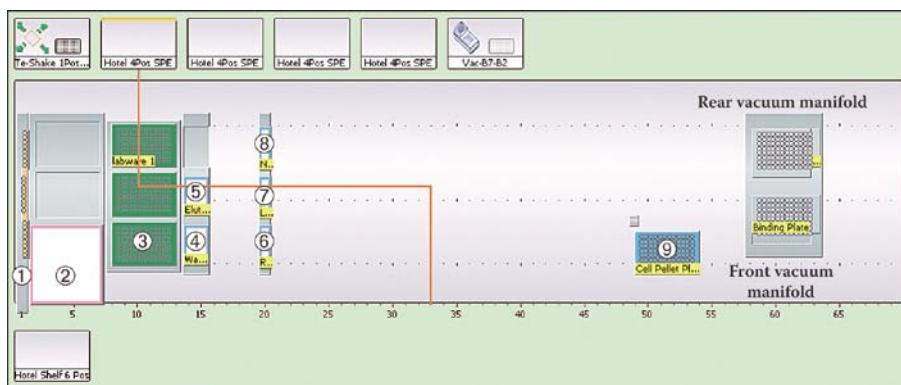


Figure 1:

1. Wash station
2. Disposable tip (DiTi) waste
3. Disposable tip carrier
4. Wash Buffer
5. Elution Buffer
6. Resuspension Buffer
7. Lysis Buffer
8. Neutralization/Binding Buffer
9. Te-Shake shaker with deep-well plate

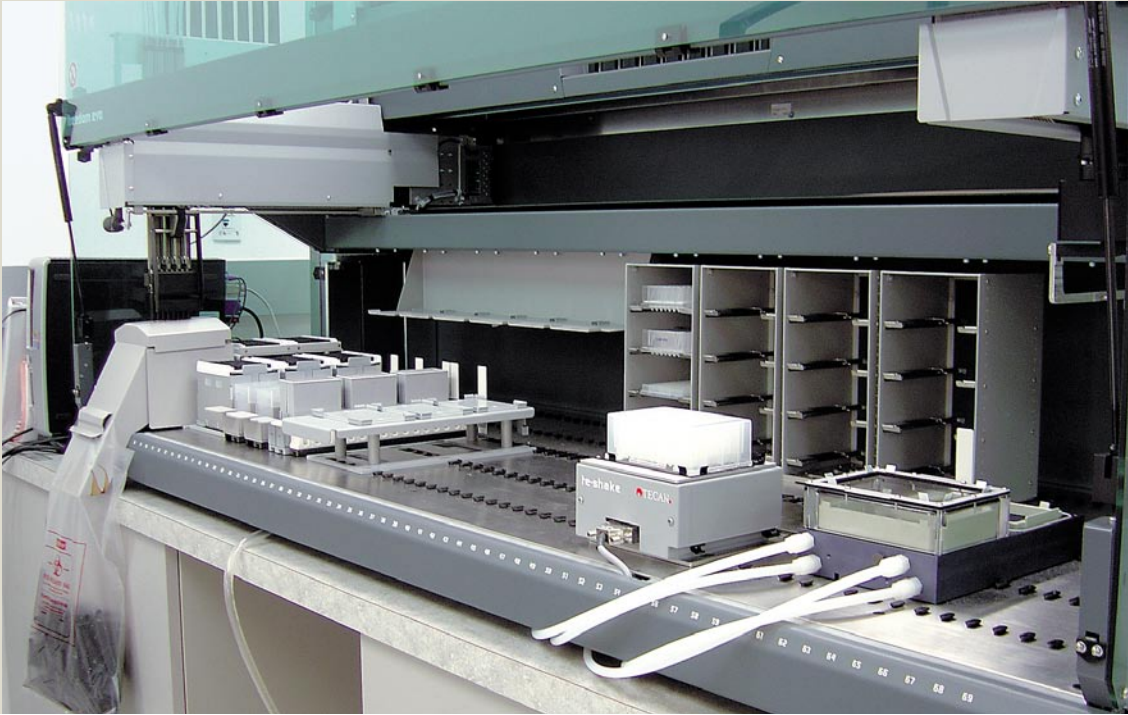


Figure 2: The Freedom EVO workstation layout



Figure 3: High quality plasmid DNA is obtained using the Purelink™96 HQ Mini Plasmid Purification Kit

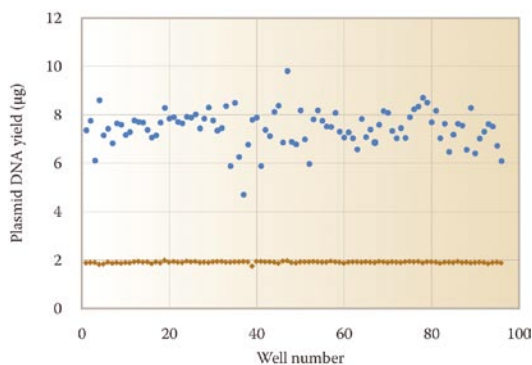


Figure 4: The Freedom EVO provides uniformity and consistency in DNA yield and quality

deployed to the Te-VacS using the RoMa. The neutralized lysate is transferred to the clarification plate, whereupon the vacuum is applied. The cleared lysate is collected in the binding plate and transferred to the second Te-VacS manifold and the vacuum applied to bind the plasmid DNA to the glass fiber matrix.

The filter is then washed with buffer W1 before addition of elution buffer. This script allows for anywhere from 1 to 96 samples to be processed at a time, with up to  $1.5 \times 10^9$  cells per well in <1 hour per 96-well plate.

#### High quality, uniform and consistent results

Plasmid DNA extracted from the PureLink™96 HQ Mini Plasmid Purification Kit via the Tecan robot is of extremely high quality as evidenced by the absence of RNA, extremely low genomic DNA contamination, and high supercoiled to nicked form ratios (Figure 3). In addition, uniformity and consistency in DNA yield and quality among the different wells indicate that the Freedom EVO separation system provides a very reproducible method of automated plasmid DNA isolation (Figure 4). Well-to-well variation was on average <10% for the automated protocol. The resulting plasmid DNA is transfection quality, with low endotoxin levels, and suitable for use in fluorescent sequencing with >98% accuracy of base identity for a 600-base read.

“ The Freedom EVO platform with the Te-VacS vacuum separation module allows walkaway automated processing ”

#### Efficient, quick, reliable and easily automated HTP method

The PureLink™96 HQ Mini Plasmid Purification Kit provides an efficient, quick, reliable, and easily automated HTP method for plasmid DNA sample preparation. Similar scripts have also been written for the PureLink™96 PCR Purification Kit and PureLink™96 RNA Purification Kit on Freedom EVOware software.

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