

High-throughput, automated, magnetic bead-based purification of high quality, endotoxin-free DNA directly from culture.

Introduction

The Zyppy™-96 Plasmid MagBead Miniprep is the fastest and simplest high-throughput method available for automated isolation of plasmid DNA from *E. coli*. It is the only fully automated method available and requires no centrifugation or pelleting of cells common to all other conventional procedures. The kit features a modified alkaline lysis system that allows for the direct lysis of *E.coli* in the growth medium. With the pellet-free procedure you can grow, lyse, and process samples in the same plate.

Automation Equipment

- Tecan Freedom EVO®
- Freedom EVOware®
- 8 channel Liquid Handling Arm (LiHa), configured for Disposable Tips (DiTis)
- Robotic Manipulation Arm (RoMa)
- Te-Shake™ Shaker
- 96-well Magnetic Stand

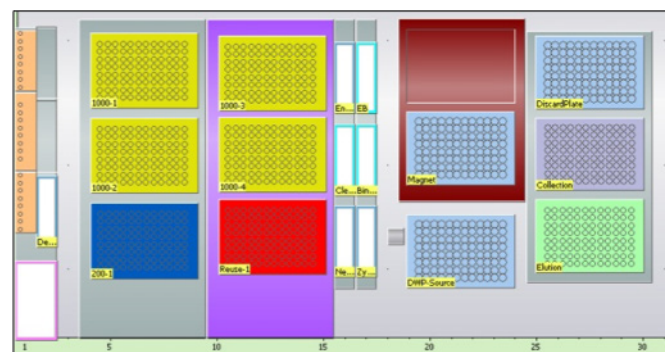
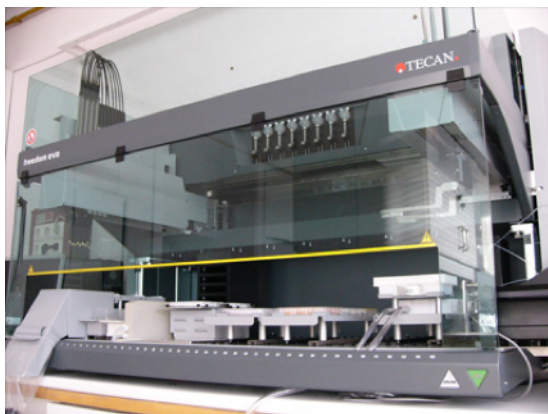
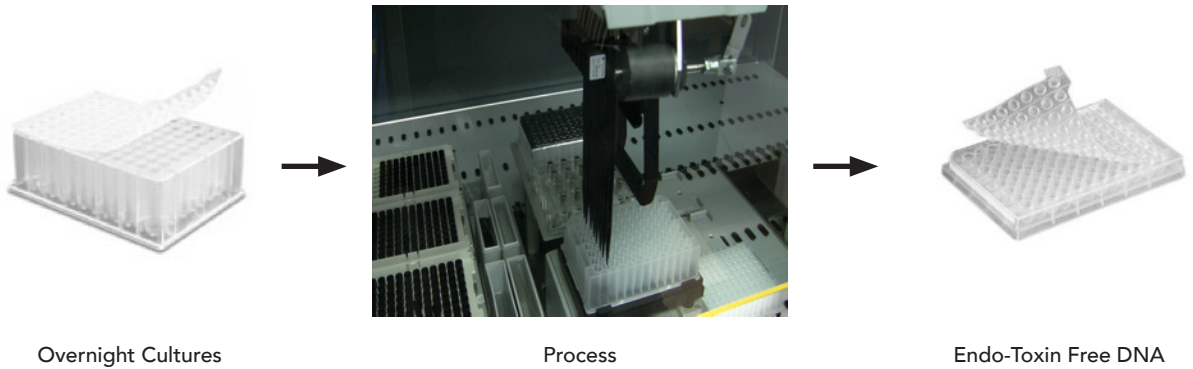


Figure 1. Example Deck Layout

Overview of Procedure

Cultures grown overnight in a 96-Well Block are transferred to the Freedom Evo®. The uniquely formulated Deep Blue Lysis Buffer is added directly to bacterial cultures in each well. After neutralization, lysate is cleared using MagClearing Beads. MagBinding Beads are then added to the cleared lysate and the DNA-bound beads are washed and dried. Once eluted, plasmid DNA is ready for immediate use, or can be stored at -20°C for later use.



Results

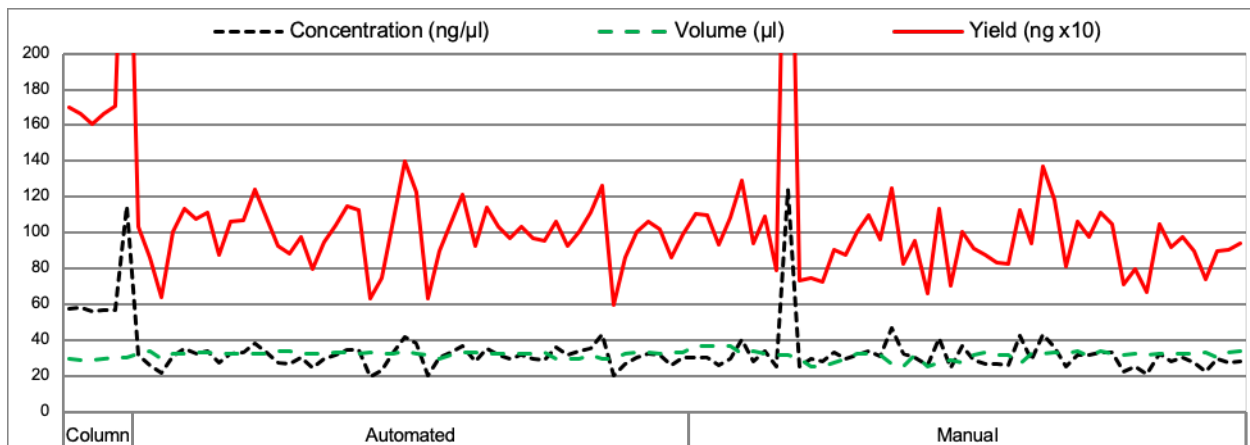


Figure 2. Comparison between manual and automated processing. Data show concentration, recovery volume, and total yield for samples processed across a 96-well plate and spin columns. Half of the plate samples were processed manually, the other half with the Freedom EVO®. Plasmid DNA was purified from *E. coli* grown at 37°C overnight.

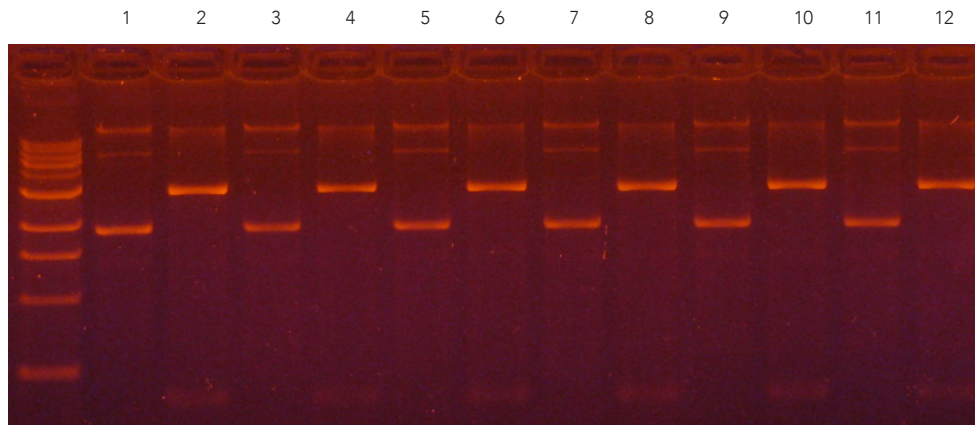


Figure 3. Restriction endonuclease digestion of plasmid DNA. Plasmid DNA (pGEM-3Zf(+)) was purified then digested with BanII for one hour at 37°C. Both undigested (odd lanes) and digested (even lanes) samples were separated in a 1.4% agarose gel. The undigested samples show super-coiled plasmid, while the digested samples show the linearized, 2,850 bp and 350 bp fragments.

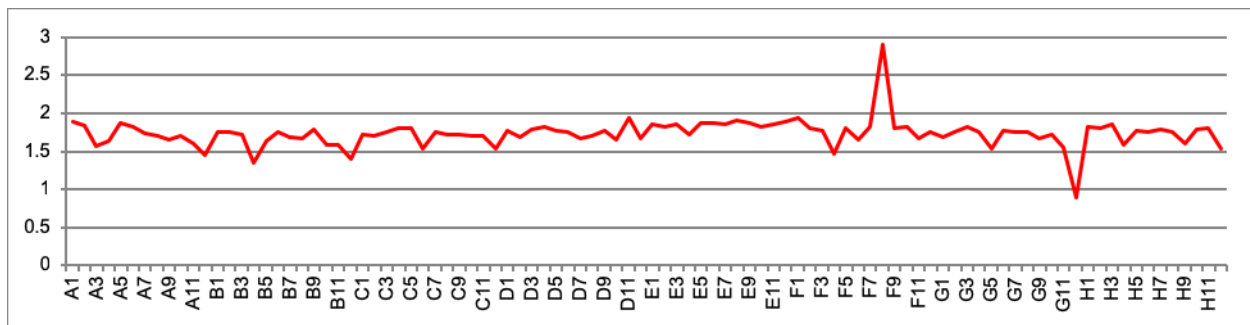


Figure 4. A260/280 ratios across a 96-well plate. Purities of eluted plasmid DNA (A260/280 ratios) are consistently high across an entire 96-well plate. DNA was purified from *E. coli* cultures grown at 37°C overnight and then processed using the Zippy-96 Plasmid MagBead Miniprep on the Freedom EVO®.

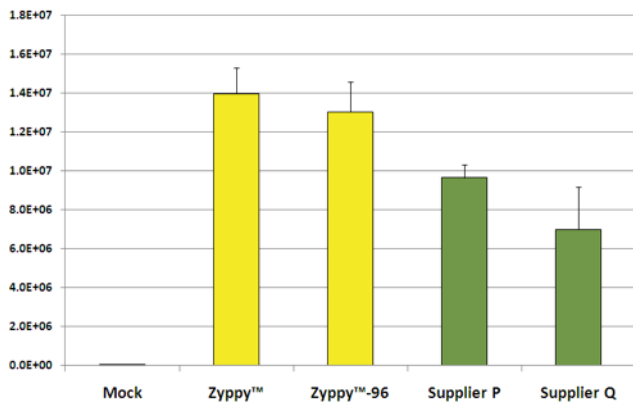


Figure 5. DNA isolated with Zippy™ Plasmid Miniprep technologies show the highest transfection efficiencies. Luciferase activities were determined in lysates from cells transfected with DNA isolated by various methods. Plasmid DNA was purified from *E. coli* using the Pellet-Free Zippy™ Plasmid Miniprep and Zippy™-96 Plasmid MagBead Miniprep or those miniprep products from Suppliers P and Q.

Conclusions

Quality and yield of DNA purified via automated processing was found to be reproducible and consistent. Eluted plasmid DNA was of the highest quality and endotoxin-free, making it ideal for subsequent transfection. DNA is well suited for use in restriction endonuclease digestion, ligation, PCR, transformation, sequencing, and all other sensitive downstream applications.

Specifications

- Sample Sources – *E. coli* cultures in 96-well block.
- DNA Purity – Eluted plasmid DNA is well suited for ligation, sequencing, restriction endonuclease digestion, transfection, in vitro transcription, and other sensitive applications requiring pure DNA. Abs_{260/280} is ≥ 1.8 .
- Plasmid DNA Yield – Up to 10 μg per preparation, depending on the plasmid copy number, culture growth conditions, and strain of *E. coli* processed.
- Plasmid DNA Size – Up to 25 kb.
- Recovery Volume – $\geq 30 \mu\text{l}$ per well.
- Procedure – Performed at room temperature (15-30°C).

Product	Cat. No.	Kit Size
Zyppy™-96 Plasmid MagBead Miniprep	D4100	2 x 96 preps
	D4101	4 x 96 preps
	D4102	8 x 96 preps



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info@zymoresearch.com | www.zymoresearch.com | Toll Free (888) 882-9682