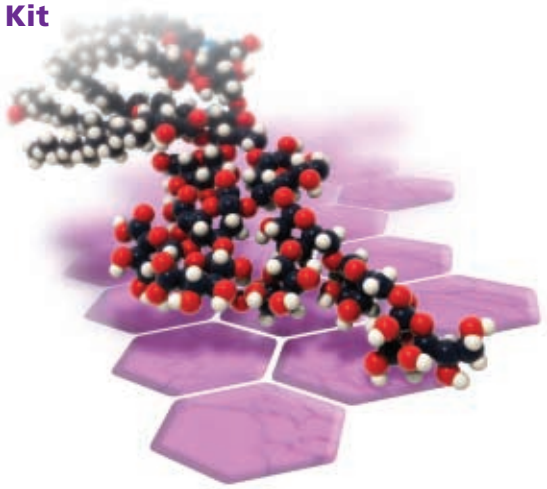
FAQs

As a major Life Science supplier, Sigma-Aldrich has spent years helping customers with numerous application and technique inquiries. By using some of the information gained from these customer contacts, we present some of our Frequently Asked Questions (FAQs) about our feature article. These FAQs will hopefully serve as a guide to help identify and resolve some of your concerns.

GenElute™ HP Endotoxin-Free Plasmid Maxiprep Kit



What size does the kit come in?
The kit is available in 4-, 10- and 25-prep sizes.

Are additional materials needed to use this kit?
Yes. One of the wash solutions included in the kit requires the addition of ethanol.

How does the HP Endotoxin-Free Maxiprep Kit procedure differ from the regular HP Maxiprep Kit?
For the HP Endotoxin-Free Maxiprep Kit, the lysate clearing step is vacuum operated and is performed before the binding solution is added.

Can the solutions from other HP Plasmid Kits be substituted for use in the HP Endotoxin-Free Maxiprep Kit?
No. The neutralization and binding solutions in the Endotoxin-Free Kit have been modified for endotoxin removal.

Can bacterial cultures be grown in rich media, such as TB?
No. Processing bacterial cultures that are grown in rich media may result in endotoxin levels that are higher than 0.1 endotoxin units per µg of plasmid DNA.

What is the best way to store a bacterial culture before processing?
It is recommended to use a fresh overnight culture immediately, but if a culture needs to be stored, then pellet the cells and store at –70 °C until ready to use.

Does the recovered plasmid DNA need to be precipitated?
No. The recovered plasmid is perfectly suitable for use in downstream applications. If a more concentrated sample is desired, the plasmid DNA can be precipitated with the addition of salt and alcohol.

How can the levels of endotoxin be determined in a plasmid sample?
By using a quantitative kit that utilizes the Limulus Amebocyte Lysate and a chromogenic substrate.

Do silica fines interfere in downstream applications? How can you remove them from the final preparation?
No. Silica fines may come through in the final purified plasmid sample but they will have no effect on most downstream applications. Silica fines can be removed by centrifuging the sample and collecting the supernatant containing the plasmid DNA or by filtering the purified sample using a 0.2-micron filter.

What downstream applications will be affected by endotoxins?
Most molecular biology applications will not be affected by the presence of endotoxin. However, data have shown that transfection efficiencies can be reduced when endotoxins are present.

What is the best way to store the recovered plasmid DNA?
For short-term storage, plasmid DNA can be stored at 2-8 °C. For long-term storage it is best to store plasmid DNA at –20 °C or –70 °C.

For advice and information on our products, contact Sigma Technical Services at **1-800-325-5832** *or by email at* **techserv@sial.com**