Technical Note

QIAprep® Spin Miniprep Kit – Competition

Introduction

The QIAprep Spin Miniprep Kit is designed to isolate up to 20 µg of highly pure plasmid DNA for use in routine molecular biology applications, including fluorescent and radioactive sequencing and cloning. Higher yields can be achieved using the *High-Yield Supplementary Protocol*, which is specially optimized for increasing yield purification of high-copy plasmid DNA from bacterial cultures grown to a high cell density (e.g., culture grown in 5 ml 2xYT medium).

The following analysis was run to show consistent performance of the high-yield (HY) protocol using different *E. coli* strains in comparison to QIAprep standard protocol and standard protocols of other suppliers.

Results

A comparison of the plasmid DNA yield from *E. coli* host strains Top10, HB101, JIM109 and DH5a harboring pUC19 or pBS plasmid showed that QIAprep Spin Miniprep Kit HY protocol consistently produces higher yields of plasmid DNA compared to standard QIAprep protocols and those of other suppliers (figure 1).

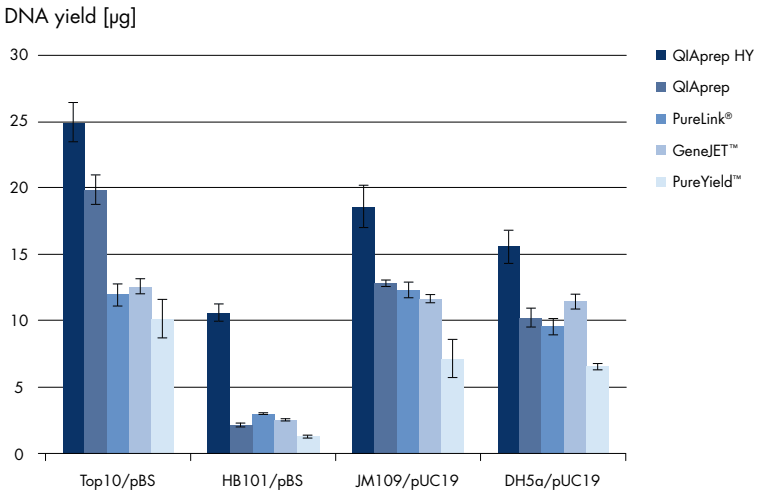
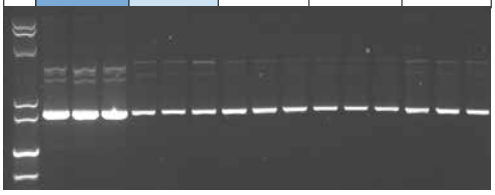


Figure 1.
Comparison of plasmid DNA yield (total yield per prep) isolated using QIAprep Spin Miniprep Kit *High-Yield (HY) Supplementary Protocol* versus standard QIAprep protocols and those of other suppliers.

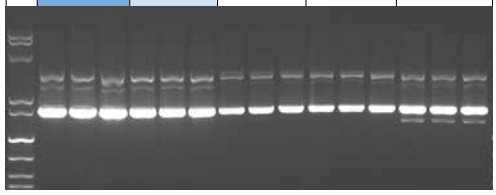
The plasmid DNA is isolated using the QIAprep Spin Miniprep Kit HY protocol versus standard QIAprep protocols and those of other suppliers. It also shows equivalent A_{260}/A_{280} ratios and purity in the isolated samples (figure 2).

	M	QIAprep HY	QIAprep	PureLink	GeneJET	PureYield
Total yield (µg)		10.6	2.1	3.0	2.5	1.3
Conc. (ng/µl)		177	42	60	50	43
A_{260}/A_{280} ratio		1.9	1.9	1.9	1.9	1.8



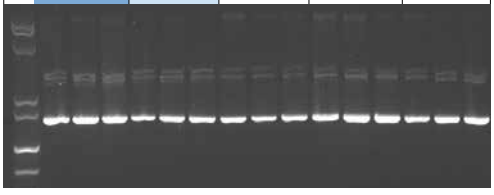
HB101-pBS

	M	QIAprep HY	QIAprep	PureLink	GeneJET	PureYield
Total yield (µg)		24.9	19.9	12.0	12.6	10.1
Conc. (ng/µl)		415	398	240	252	337
A_{260}/A_{280} ratio		1.9	1.9	1.9	1.9	1.9



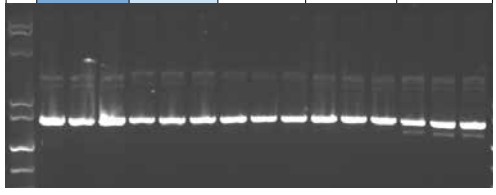
Top10-pBS

	M	QIAprep HY	QIAprep	PureLink	GeneJET	PureYield
Total yield (µg)		15.6	10.2	9.6	11.5	6.5
Conc. (ng/µl)		260	204	192	230	216
A_{260}/A_{280} ratio		1.9	1.9	1.9	1.9	1.9



DH5a-pUC19

	M	QIAprep HY	QIAprep	PureLink	GeneJET	PureYield
Total yield (µg)		18.6	12.8	12.3	11.6	7.1
Conc. (ng/µl)		310	256	246	232	236
A_{260}/A_{280} ratio		1.9	1.9	1.9	1.9	1.9



JM109-pUC19

Figure 2. Comparison of quality of plasmid DNA isolated using QIAprep Spin Miniprep Kit High-Yield (HY) Supplementary Protocol against standard QIAGEN protocols and those of other suppliers.

Conclusion

QIAprep Spin Miniprep Kit HY protocol consistently produces higher yields and more concentrated plasmid DNA with equivalent purity as compared to standard QIAGEN plasmid prep protocols and those from other suppliers.

Materials and methods

Plasmid harboring *E. coli* strains were plated on LB agar plates with the appropriate antibiotics and incubated overnight at 37°C. Starter cultures in flasks with the appropriate antibiotic were made from each plate using a single colony. The starter cultures in flasks were used to inoculate the main cultures to OD600 = 0.05. Main cultures were incubated for 22 h at 37°C and 220 rpm. The cells were harvested by centrifugation, the medium was decanted and the obtained cell pellets were processed using the recommended protocols. All plasmid preps except supplier P were performed according to recommended protocols using 5 ml of the same overnight culture of *E. coli* strains HB101, Top10, DH5α and JM109 cultured in LB medium. Plasmid preps of supplier P were performed with recommended 3 ml culture volume. Plasmid DNA using the HY protocol was isolated from cultures grown to a high cell density in 5 ml 2xYT medium.

The plasmid DNA yields, concentration and A_{260}/A_{280} ratio were determined on the QIAxpert, an innovative high-speed microfluidic UV/VIS spectrophotometer for accelerated DNA, RNA and protein quantification and quality control. The DNA integrity was analyzed on a 0.8% agarose gel.

Kit	Supplier	Protocol
QIAprep Spin Miniprep Kit	QIAGEN	Standard protocol (5 ml LB) HY protocol (5 ml 2xYT)
PureLink Quick Plasmid DNA Miniprep Kit	Invitrogen	Standard protocol (5 ml LB)
GeneJet Plasmid Miniprep Kit	Thermo Fisher Scientific	Standard protocol (5 ml LB)
PureYield Plasmid Miniprep Kit	Promega	Standard protocol (3 ml LB)

E. coli strain	Genotype
HB101	F- mcrB mrr hsdS20 (rB- mB-) recA13 leuB6 ara-14 proA2 lacY1 galK2 xyl-5 mtl-1 rpsL20 (SmR) glnV44 λ-
Top10	IF- mcrA Δ (mrr-hsdRMS-mcrBC) ϕ80lacZΔM15 ΔlacX74 nupG recA1 araD139 Δ (ara-leu) 7697 galE15 galK16 rpsL (StrR) endA1 λ-
DH5a	F- endA1 glnV44 thi-1 recA1 relA1 gyrA96 deoR nupG Φ80dlacZΔM15 Δ (lacZYA-argF) U169, hsdR17(rK- mK+), λ–
JM109	PendA1 glnV44 thi-1 relA1 gyrA96 recA1 mcrB+ Δ (lac-proAB) e14- [F' traD36 proAB+ lacIq lacZΔM15] hsdR17 (rK-mK+)

Plasmid	Background
pUC19	pMB1 ORI, high copy, 2686 bp
pBluescript (pBS)	colE1 ORI, high copy, 3204 bp