Biotage® PhyPrep method library info sheet

High yield via extended runtime

Objective

Improve yields with an extended capture sequence and an optimized plasmid capture strategy

When to use

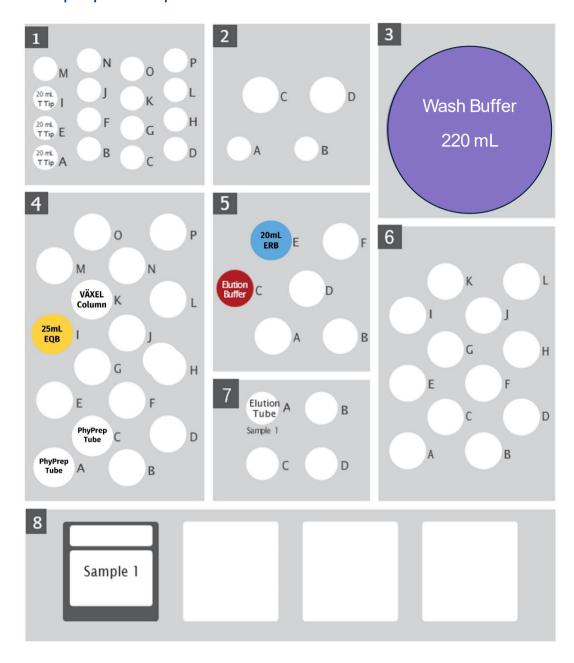
To optimize plasmid recovery through longer runtimes, especially when working with low-copy plasmids, slower-growing cell types, or alternative growth options. Yield improvements are also seen on high-copy plasmids, with the most significant gains observed at the maxiprep scale.

Notes	
Method specifics	Extends the capture step, improves column binding kinetics, optimizes lysate processing
Method version	V132
Additional consumables	Biotage® Phyprep tube [p/n: 417458sp] Maxiprep only Wash buffer [p/n: 900-0092sp]
Deck layout	See below. Also available in the software before executing the method

Prep scale	Number of sample preps	Run time (hour/minutes)	Culture volume per sample	Pellet wet weight per sample	Optimal pellet wet weight	Elution volume
Maxiprep	1	1 h 2 m	150-250 mL	3-5 g	3 g	5 mL
	2	1 h 2 m				
	3	1 h 48 m				
	4	1 h 48 m				
Megaprep	1	2 h 51 m	350-500 mL	6-8 g	7 g	18 mL
	2	2 h 51 m				
	3	5 h 22 m				
	4	5 h 22 m				
Gigaprep	1	3 h 28 m	600-750 mL	14-16 g	15 g	28 mL
	2	3 h 28 m				
	3	6 h 1 m				
	4	6 h 1 m				

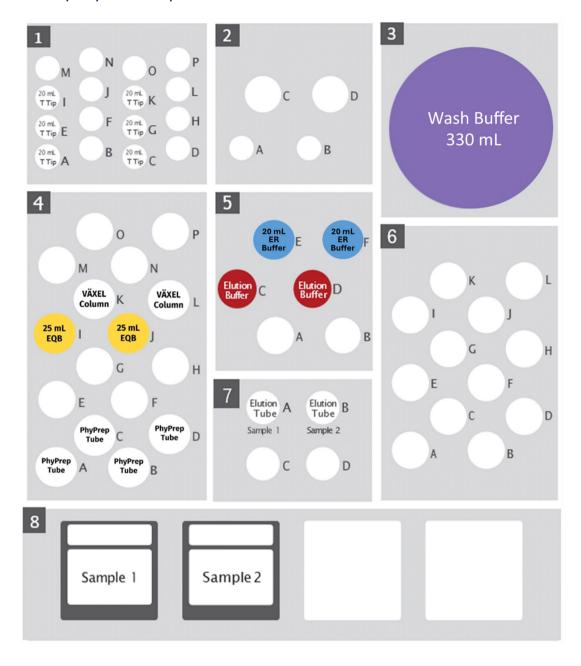


Maxiprep 1 Sample Method



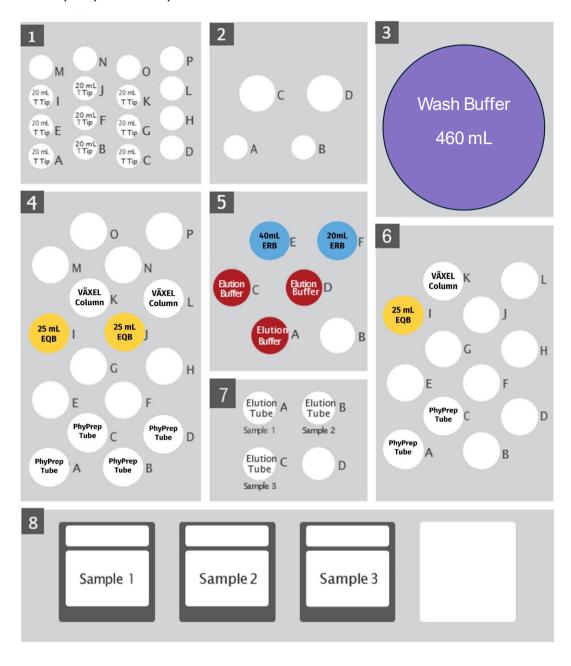


Maxiprep 2 Sample Method



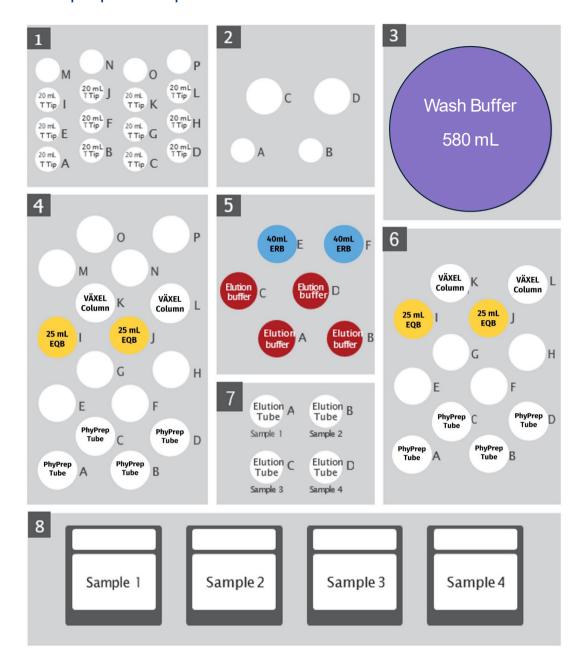


Maxiprep 3 Sample Method



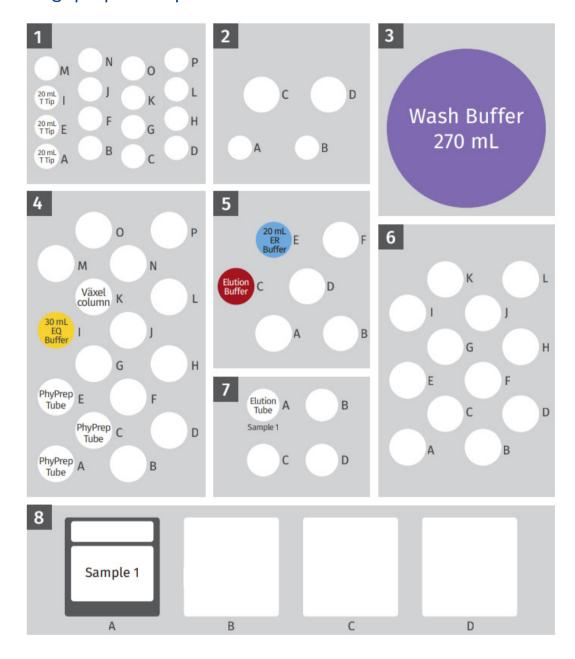


Maxiprep 4 Sample Method



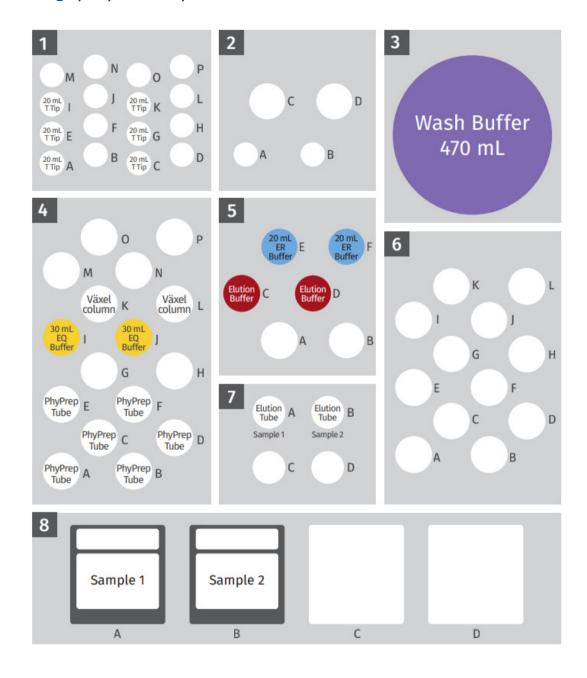


Megaprep 1 Sample Method



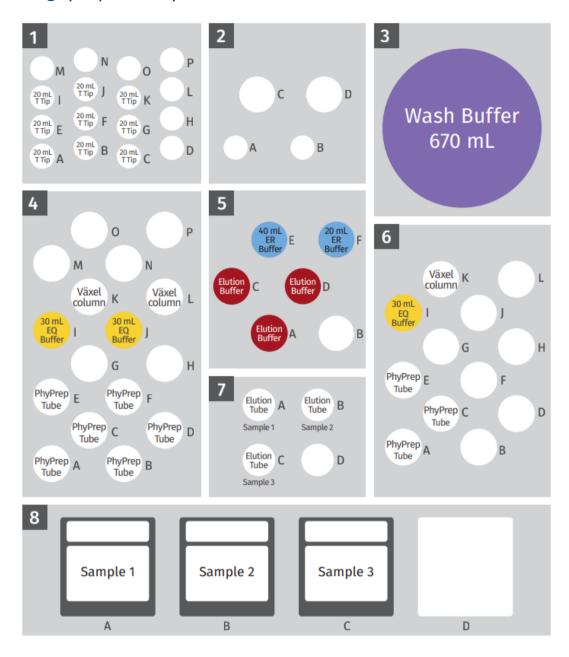


Megaprep 2 Sample Method



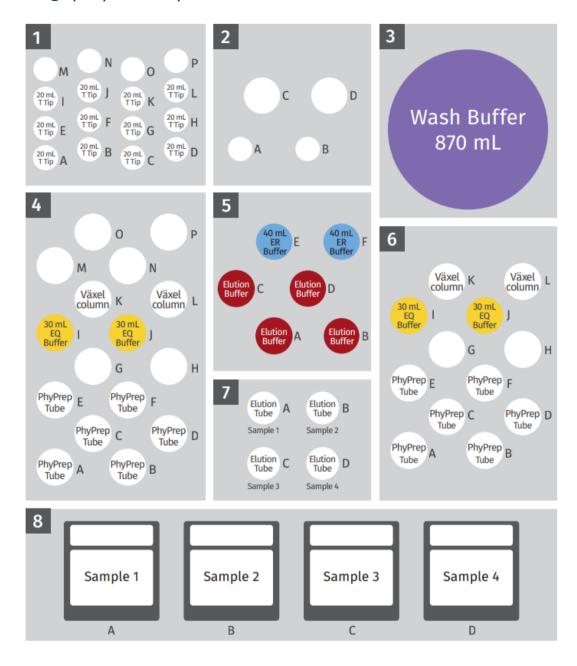


Megaprep 3 Sample Method



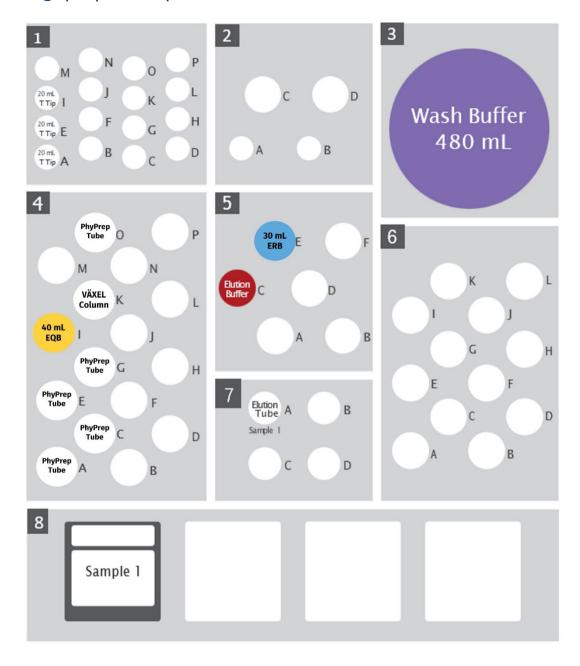


Megaprep 4 Sample Method



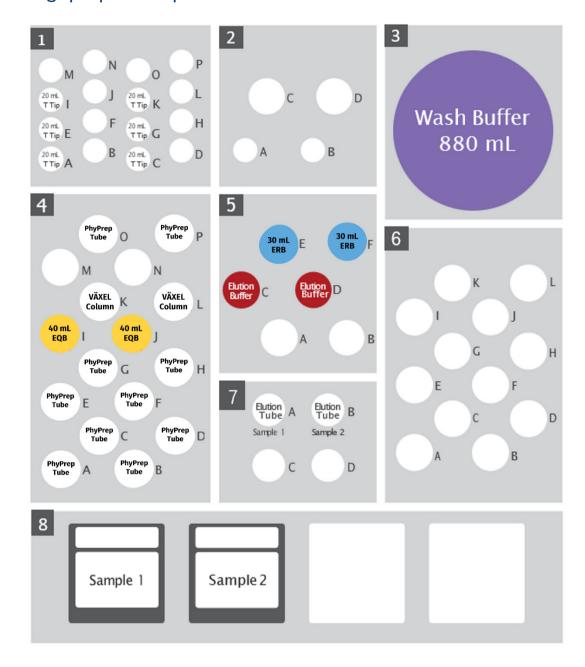


Gigaprep 1 Sample Method



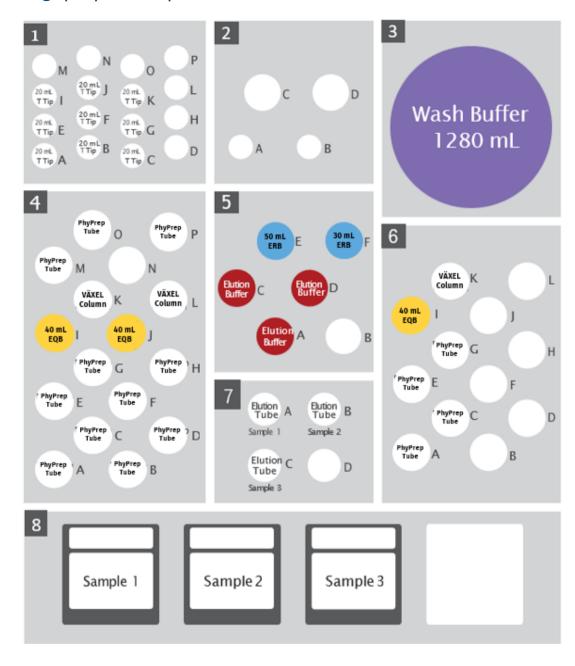


Gigaprep 2 Sample Method



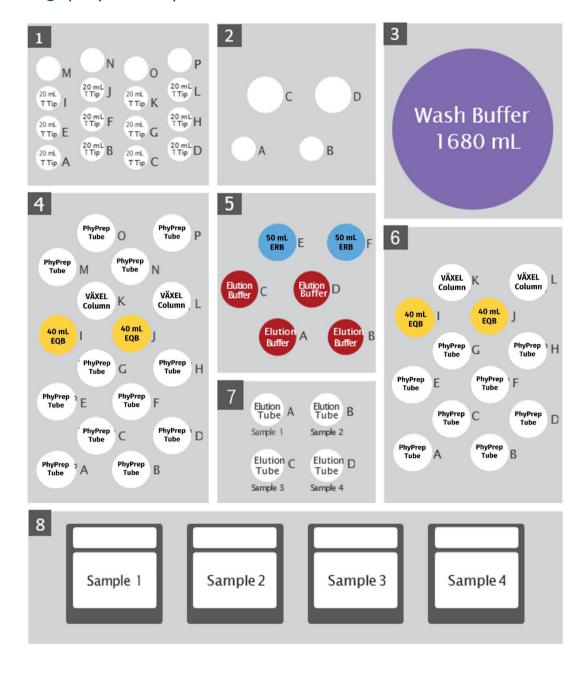


Gigaprep 3 Sample Method





Gigaprep 4 Sample Method





Method demonstration

Materials/Method

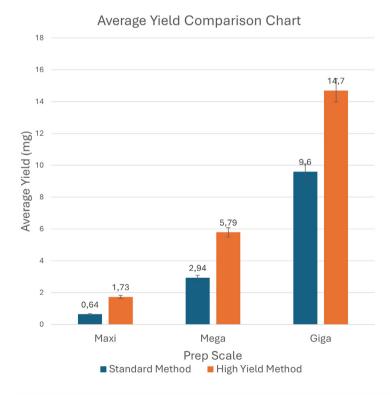
Notes	Maxi	Mega	Giga
Pellet size	3g	7g	15g
Kit lot	021713	020197	021531
Plasmid	BT-02 (medium copy)	BT-02 (medium copy)	BT-02 (medium copy)

Culture growth

Reagents used in this study were purchased from VWR unless otherwise stated. The plasmid DNA used in this study is a proprietary medium copy plasmid containing eGFP. The plasmid was transformed into E. coli DH5 (ThermoFisher Scientific, 18265017) to generate strain BT02.

BTo2 was streaked onto LB agar plates containing 100 μ g/mL carbenicillin (Teknova, L1010) from a frozen glycerol stock. The plate was incubated at 37 °C for 24 hours. Single colonies were used to inoculate a starter culture of 10 mL Terrific Broth (Teknova, T7000) and 100 μ g/mL carbenicillin (Teknova, C2135). The starter culture was shaken at 38 °C at 350 RPM incubator shaker for 8 hours. 500 μ L was used to inoculate 500 mL of TB, 100 μ g/mL carbenicillin in 2.5 L baffled flasks (Thomsom, 931136-B). The cultures were shaken at 38 °C for 16 hours. Cells were harvested in 250 mL Fiberlite centrifuge bottles (ThermoFisher Scientific, 001-0303) by centrifugation for 15 minutes at 5,000 RPM. The supernatant was decanted, and cell pellets were weighed to record the pellet wet weight.

Results



Prep scale	Maxi	Mega	Giga
Yield percentage increase from the current standard method	170%	96.94%	53.13%

