



# Automated CosMCPrep Plasmid Preparation Kit on the Biomek i7 Hybrid Workstation

#### Introduction

The CosMCPrep Kit enables plasmid purification procedure based on SPRI paramagnetic bead-based technology. It uses a single protocol to purify a variety of high and low copy number templates while maintaining the flexibility to support both manual and automated processing. This kit enables:

- Isolation of high-quality plasmids
- A single protocol for the purification of all template types
- · Automated procedures to minimize variability in plasmid yield and quality

In this application note, we demonstrate the automation of the CosMCPrep Plasmid Purification Kit on the Biomek i7 dual hybrid Automated Workstation. The automated method has a throughput of 96 samples and reduces hands-on time, potential pipetting errors and the possiblity of cross contamination.

## **Spotlight**

The Biomek i7 Hybrid (Multichannel 96, Span-8) Automated Workstation delivers reliability and efficiency to increase user confidence and walk-away time as compared to manual library preparation.

- 1200 μL Multichannel head with 1-1000 μL pipetting capability
- Span-8 pod with disposable tips
- Enhanced Selective Tip multichannel pipetting to transfer a custom array of samples
- Independent 360° rotating gripper with offset fingers
- High deck capacity with up to 45 positions
- · Shaking, heating/cooling, and tip washing for controlling sample processing
- · Spacious, open platform design to integrate on-deck and off-deck elements (e.g., Automated Thermal Cyclers [ATC])





Figure 1. Top: Biomek i7 Hybrid Automated Workstation with optional enclosure on a Biomek Mobile Workstation. Bottom: Deck layout.

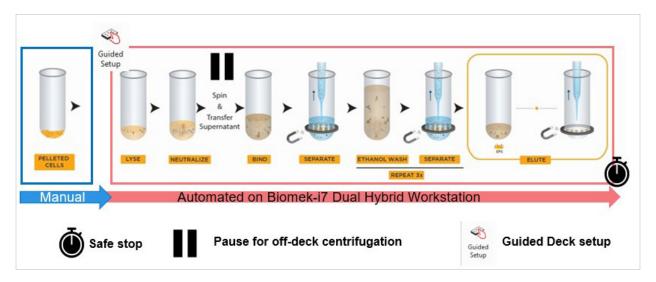


Figure 2. CosMCPrep plasmid purification kit automated workflow.

## Automated method

Automation provides increased efficiency and a reduction in human errors, with minimal hands-on time (Table 1).

| Kit Type                                      | CosMCPrep Plasmid Purification Kit |                |  |  |  |
|---|------------------------------------|----------------|--|--|--|
| Sample Number                                 | 24                                 | 96             |  |  |  |
| Instrument Setup Time                         | 10 mins                            | 10 mins        |  |  |  |
| (a) Pre-centrifugation (until neutralization) | 5 mins                             | 10 mins        |  |  |  |
| (b) Post-neutralization processing            | 19 mins                            | 36 mins        |  |  |  |
| Method Run Time (a+b)                         | 24 mins                            | 46 mins        |  |  |  |
| Total Time* (with off-deck centrifuge)        | 54 mins                            | 1 hour 16 mins |  |  |  |

<sup>\*</sup>Total timing estimates includes 20 min centrifugation at 3000 x g.

Table 1. Estimated run times for purifying 24 or 96 plasmid preps with the CosMCPrep Plasmid Purification kit on the Biomek i7 Hybrid Automated Workstation.

The method can be run using Method Option Selector, which is an interactive user interface that supports modular design and logical start and stop points. Guided Labware Setup simplifies deck setup by providing reagent information, and DeckOptix Final Check software minimizes costly setup errors. An automated method provides flexibility to users in scheduling their workflow and allowing each laboratory to address their individual requirements for sample processing and throughput.

The instrument has a cold plate and a BioShake from QINSTRUMENTS as well as a waste drain for liquid waste removal.

The software provides several user-friendly features such as:

#### 1. Biomek Method Launcher (BML)

BML is a secure interface for method implementation without affecting method integrity. It allows the users to remotely monitor the progress of the run. The manual control options provide the opportunity to interact with the instrument, if needed.

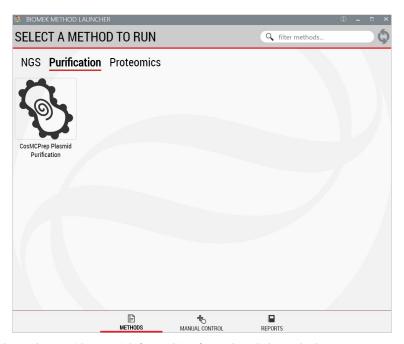


Figure 3. Biomek Method Launcher provides a straightforward interface to launch the method.

#### 2. Method Option Selector (MOS)

MOS enables selection of plate processing and sample number options to maximize user method flexibility, adaptability, and the ease of method execution.

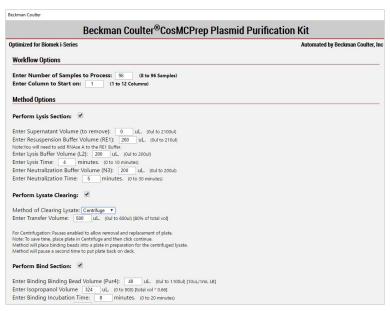
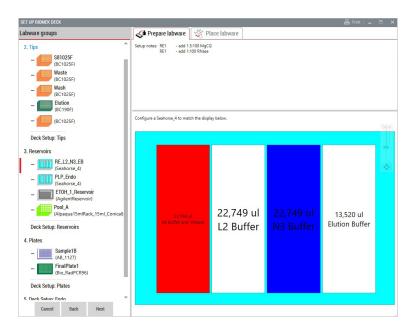


Figure 4. CosMCPrep Plasmid Purification Method Options Selector.

#### 3. Guided Labware Setup (GLS)

GLS is generated based on options selected in the MOS and provides user-specific graphical setup instructions with reagent volume calculations and step-by-step instructions to prepare reagents.



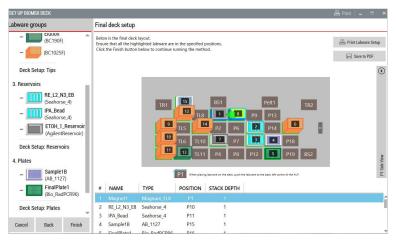


Figure 5. Guided Labware Setup provides recipe notes, indicates reagent volumes (upper image), and guides the user for correct deck setup (lower image).

### 4. DeckOptix Final Check (DFC)

Prior to starting a run, DFC (included with BML) analyzes the deck to reduce setup errors and prevent a failed experiment because of missing or misplaced labware or use of the wrong tip or plate type.

## **Experimental design**

To demonstrate capabilities to perform an automated 96-plasmid preparation using the CosMCPrep kit, 1 mL of bacterial culture was used per well of a 96-deepwell plate. For this demonstration we grew three different types of plasmids (A, B and C) carrying bacteria overnight with shaking at 37°C. Each of the bacterial cultures was split into four columns of the plate as shown in Figure 6.

The yields and purity of the purified plasmids were analyzed using a NanoDrop instrument from Thermo Fisher Scientific (Figure 7). To check for cross contamination, another 96-sample run was performed where 48 samples (32 of plasmid B and 16 of plasmid C) were interleaved with negative control in a checkerboard pattern. After the purification, the yields and purity were analyzed using the NanoDrop instrument.

|   | 1         | 2         | 3         | 4         | 5         | 6         | 7         | 8         | 9         | 10        | 11        | 12        |
|---|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|-----------|
| Α | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C |
| В | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C |
| С | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C |
| D | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C |
| E | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C |
| F | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C |
| G | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C |
| Н | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C | Plasmid A | Plasmid A | Plasmid B | Plasmid B | Plasmid C | Plasmid C |

Figure 6. Schematic representation of sample plate setup.

#### Results

The yields and purity for the automated plasmid prep (Figure 7) were comparable to the manual processing (data not shown). There was no plate-edge effect or column effects observed in any of the runs. In the automated run performed with negative controls, none of the negative samples showed any presence of DNA (Figure 8). This demonstrates thats there is no cross contamination during the automated plasmid purification. For similar plasmid preps the yields vary, but it is not significant and is expected, as the cultures were grown in larger volumes and then split into the wells of plates.

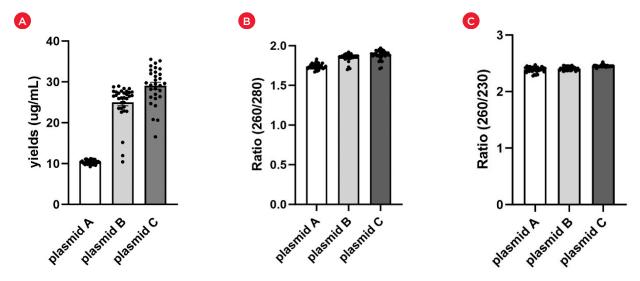


Figure 7. QC for 96 plasmids purified by automation. (A) Yields in µg/mL for the 32 replicates of each Plasmid A, Plasmid B and Plasmid C. (B) 260/280 ratios and (C) 260/230 ratios for the 32 replicates of each Plasmid A, Plasmid B and Plasmid C.

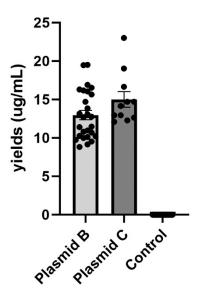


Figure 8. QC for samples purified by automation to check for cross-contamination. (A) Yields in  $\mu$ g/mL for the 32 replicates of Plasmid B, 16 samples of Plasmid C and 48 of the negative control.

# **Summary**

We've demonstrated that automation of the CosMCPrep Plasmid Purification Kit on the Biomek i7 Hybrid Automated Workstation is a semi-walk-away solution that provides an efficient, flexible and scalable solution for any size lab. The automation solution delivers reproducible plasmid preps with good yields and purity and reduces overall hands-on time.

The CosMCPrep Plasmid Purification kit is for Research Use Only. Beckman Coulter makes no warranties of any kind whatsoever express or implied, with respect to this protocol, including but not limited to warranties of fitness for a particular purpose or merchantability or that the protocol is non-infringing. All warranties are expressly disclaimed. Your use of the method is solely at your own risk, without recourse to Beckman Coulter. This protocol is for demonstration only and is not validated by Beckman Coulter.

Biomek Automated Workstations are not intended or validated for use in the diagnosis of disease or other conditions.



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