

SpeeDNA Plasmid Miniprep Kit (SDPM)

Catalog #MB6928-100 100 Preps or Catalog #MB6928-100 100 Preps

Description

ScienCell's SpeeDNA Plasmid Miniprep Kit (SDPM) provides a quick and easy way to purify plasmid DNA from bacterial cells in ≤ 15 minutes. With ScienCell's optimized buffer system, SPDM does not require use of RNase. All items in the kit can be stored at room temperature. SDPM uses a spin-column based method to recover up to $25~\mu g$ high-quality plasmid DNA that is ready for use in downstream applications, such as restriction enzyme digestion, ligation and transformation, transfection, PCR amplification, and DNA sequencing.

Kit Components

Catalog #MB6928-100

Cat #	Item	Volume
MB6928a-1	Buffer PM1	30 mL
MB6928b-1	Buffer PM2	30 mL
MB6928c-1	Buffer PM3	40 mL
MB6928d	Buffer DW	20 mL
MB6928e-1	Buffer DE	8 mL
MB6928f	DNA spin columns	100 pieces
MB6928g	Wash tubes	100 pieces

or

Catalog #MB6928-250

Cat #	Item	Volume
MB6928a-2	Buffer PM1	70 mL
MB6928b-2	Buffer PM2	70 mL
MB6928c-2	Buffer PM3	100 mL
MB6928d	Buffer DW	20 mL x 2
MB6928e-2	Buffer DE	15 mL
MB6928f	DNA spin columns	250 pieces
MB6928g	Wash tubes	250 pieces

Materials Required (Not Provided)

Ethanol (96-100%)

1.7 mL (or 1.5 mL) microcentrifuge tubes (DNase/RNase free)

Quality Control

The yield and purity of purified plasmid from *E. coli* cells using SPGelEx was analyzed by spectrophotometry.

Product Use

SDPM is for research use only. It is not approved for human or animal use, or for application in clinical or *in vitro* diagnostic procedures.

Shipping and Storage

Room temperature. All components of the kit should be stored at room temperature.

Reagent Preparation

Buffer DW (20 mL, Cat # MB6928d) is provided as concentrate. Prior to use for the first time, add **100 mL** ethanol (96-100%) and mix well to make working Buffer DW. Store at room temperature.

Procedures

<u>Note:</u> All steps should be carried out at room temperature. All centrifugations should be carried out at $>12,000 \times g$ (10,000 - 14,000 rpm).

<u>Note</u>: Check Buffer PM2 before use. If there is precipitate, warm the Buffer PM2 up to 37°C until the solution becomes clear.

Note: Buffers PM3 contains guanidine thiocyanate, which can form highly reactive compounds when combined with bleach. If the spilt liquid contains potentially infectious agents, clean the affected area first with laboratory detergent and water, and then with 1% (v/v) sodium hypochlorite.

- 1. Prepare sample by centrifuging bacterial culture (1-5 mL) in a 1.7 mL microcentrifuge tubes at $\geq 8,000$ rpm (6,800 x g) for 1 minutes at room temperature. Pour off supernatant and remove any remaining medium leaving behind the cell pellets.
- 2. Resuspend cell pellet in 250 μl Buffer PM1(Cat #MB6928a) by pipetting up and down until completely mixed.

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- 3. Add 250 µl Buffer PM2 (Cat #MB6928b) to lysis suspension and mix by inverting tube 4-6 times. **Do not vortex to avoid shearing plasmids**. Incubate the mixture at room temperature from 1-4 minutes.
- 4. Add 350 μl of Buffer PM3 (Cat #MB6928c) and mix immediately by inverting tube 4-6 times to neutralize the solution.
- 5. Centrifuge at \geq 15,000 x g for 5 minutes.
- 6. Place DNA spin column (Cat #MB6928f) into a Wash tube (Cat #MB6928g). Carefully transfer the supernatant to the DNA spin column in wash tube, without disturbing the white pellet. Centrifuge for 1 minute. Discard flow-through and return column to the same wash tube.
- 7. Apply 750 µl of working Buffer DW (with ethanol added to Buffer DW), to spin column. Centrifuge for 30-60 seconds. Discard flow-through and return column to the same wash tube.
- 8. Centrifuge empty column (in reused wash tube) for 1 minute to remove any residual wash solution.
- 9. Transfer the spin column to a new 1.7-mL microcentrifuge tube. Add 50 µl of Buffer DE directly to the spin column membrane to elute the plasmid DNA (but do not touch pipet tip to the membrane). Incubate for 2 minutes at room temperature and centrifuge for 2 minutes.
- 10. Discard spin column. Purified plasmid DNA may be used immediately or stored at -20°C.