Structure	MeCP2
PDB Code	6OGJ
Entry clone accession	BC011612.1
Entry clone source	AU51-B9
Tag	C-terminal hexahistidine tag: AAHHHHHH
Construct sequence	ASASPKQRRSIIRDRGPMYDDPTLPEGWTRKLKQRKSGRSAGKYDVYL INPQGKAFRSKVELIAYFEKVGDTSLDPNDFDFTVTGRGSPSAHHHHHH
Vector	pNIC-CH
Expression host	*Escherichia coli BL21 (DE3)-RIL* strain
Growth medium	TB
Growth method	A fresh transformation was used to inoculate 50 mL LB media containing 50 µg/mL kanamycin and 30 µg/mL Chloramphenicol. The culture was grown overnight at 37ºC with shaking. The next day this starter culture was used to inoculate 2L of TB growth medium. The culture was grown in LEX at 37ºC to OD_{600} of 1.0. IPTG-based induction was carried out according to the manufacturer's protocol (final concentration for IPTG is 0.35 mM). The temperature was reduced to 16ºC and the culture was incubated for a further 18 hours before harvesting the cells.
Extraction buffers	**Lysis buffer**: 20 mM Tris-HCl, pH 7.5, 500 mM NaCl, 0.5 mM PMSF and 5% glycerol
Extraction procedure	Cells were harvested by centrifugation and pellets were stored in -80ºC. Prior to purification, the cell pellet was resuspended in lysis buffer. Cells were disrupted by sonication (10 minutes) and samples were centrifuged for 60 min at 70000 g.
Purification buffers	**NiNTA Elution buffer (EB)**: 20 mM Tris-HCl, pH 7.5, 500 mM NaCl and 300 mM imidazole **Gel Filtration buffer**: 20 mM Tris-HCl, pH 7.5 and 150 mM NaCl, 1mM DTT.
Purification procedure	Column 1: Affinity purification, open Ni-NTA column Procedure: The supernatant was incubated with 4mL of 50% slurry Ni-NTA beads by rocking. After 30 min incubation at 4ºC, the beads were washed with 50 mL of lysis buffer with additional 10 mM imidazole. The protein was eluted using ~20 mL EB. The Column 2: Gel Filtration (Superdex S75 16/60 Hi-Load, GE Healthcare). Then the fractions containing protein were identified on a SDS-PAGE gel.
Protein stock concentration	10mg/ml.
Crystallization	30% PEG 2000 MME, 0.2 M potassium bromide
Data collection	