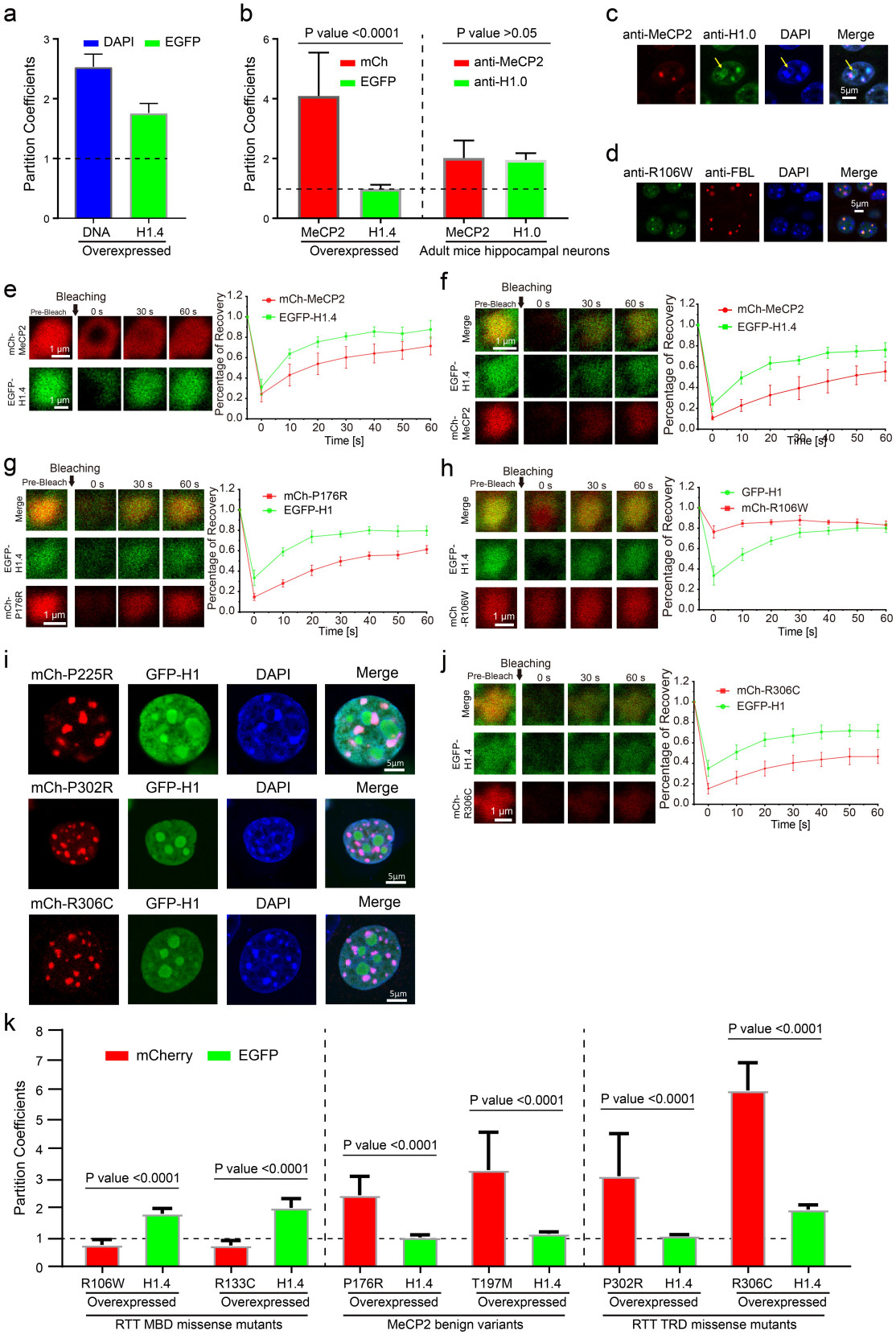

Supplementary information, Fig. S13. Competition between MeCP2 and H1.4 in cells.

a Statistical analysis of the EGFP-H1 on the heterochromatin in Fig. 6c. **b** Statistical analysis of the H1 and MeCP2 on the heterochromatin in Fig. 6c (P value = 0.8398) and Fig. 6d (P value <0.0001). **c** Immunofluorescence images of MeCP2 and H1.0 in hippocampal neurons from adult mice. H1.0 is partial located in the DAPI-deficient nucleolus region. Scale bar, 5 μm. **d** Immunofluorescence images of MeCP2-R106W is dispersed in the nucleus and also enriched in the nucleolus (labeled with Fibrillarin (FBL)). Scale bar, 5 μm. **e** Left panels, snapshots of puncta formed in NIH 3T3 cells by H1.4 in Fig.6c or WT MeCP2 in Fig.3f during FRAP experiments. Scale bars, 1 μm. Right panel, average fluorescence recovery traces of MeCP2 WT (red curve) and H1.4 (green curve) in puncta (n=6). All data are presented as mean ± SD. **f** Left panels, snapshots of puncta formed in NIH 3T3 cells by H1.4 and WT MeCP2 in Fig.6 c. Scale bar, 1 μm. Right panel, average fluorescence recovery traces of MeCP2 WT(red curve) and H1.4 (green curve) in puncta (n=6). All data are presented as mean ± SD.

g Left panels, snapshots of puncta formed in NIH 3T3 cells by H1.4 and the MeCP2 neutral variant P176R in Fig.6 i during FRAP experiments. Scale bar, 1 μm. Right panel, average fluorescence recovery traces of MeCP2 P176R (red curve) and H1.4 (green curve) in puncta (n=6). All data are represented as mean ± SD. **h** Left panels, snapshots of puncta formed in NIH 3T3 cells by H1.4 and MeCP2 R106W in Fig.6 h during FRAP experiments. Scale bar, 1 μm. Right panel, average fluorescence recovery traces of MeCP2 R106W (red curve) and H1.4 (green curve) in puncta (n=6). All data are presented as mean ± SD. **i** Localizations of overexpressed EGFP-H1.4 with Rett syndrome-related mCherry-MeCP2 TRD mutations in NIH 3T3 cells. Scale bars, 5 μm.

j Left panels, snapshots of puncta formed in NIH 3T3 cells by EGFP-H1.4 and mCherry-MeCP2 R306C in Supplementary information, Fig. S13h during FRAP experiments. Right panel, average fluorescence recovery traces of mCherry-MeCP2 R306C (red curve) and EGFP-H1.4 (green curve) in puncta (n=6). All data are presented as mean ± SD. **k** Statistical analysis the partition coefficients of mCherry-MeCP2 and EGFP-H1.4 (P value (for all 6 groups) <0.0001) on the heterochromatin in Fig. 6h-i and Supplementary information, Fig. S13i. The average fluorescence signals for labeled proteins in DAPI-dense regions were analyzed by using the signals in DAPI-dispersed regions as the background.