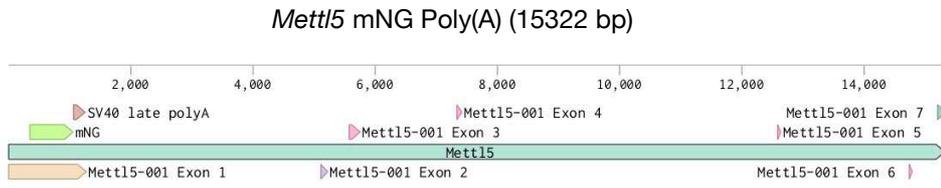
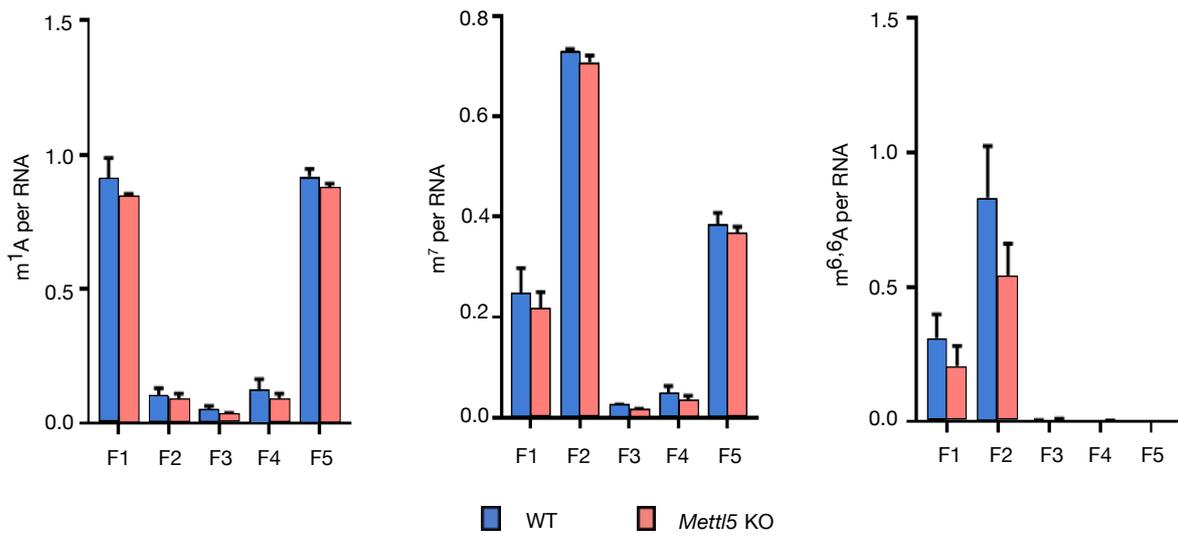


Figure S2

a



b



c

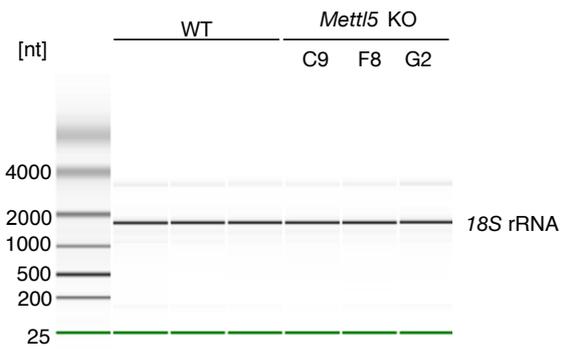


Figure S2. METTL5 methylates 18S rRNA in mESCs.

a. Schematic representation of the CRISPR/Cas9 gene editing strategy to create *Mettl5* KO mESCs. The KO mESCs were created by inserting a mNeonGreen (mNG) PolyA cassette (green) into the first exon of the mouse *Mettl5* gene. See full description in the Material and Methods. The scheme was generated in Benchling (<https://benchling.com>).

b. Quantification of modified nucleosides in indicated RNA fractions (see Figure 2c for fractionation details). Absolute quantification of modified nucleosides per respective RNA for 1-methyladenosine (m¹A), 7-methylguanosine (m⁷G) and N6,N6-dimethyladenosine (m^{6,6}A) (Borland et al. 2019) are plotted. Average of three biological replicates and standard deviations are shown.

c. Bioanalyzer profiling of fraction F2 (from SEC experiment, Figure S2B) from wt and *Mettl5* KO mESCs (clones C9, F8, G2) used as substrates in the *in vitro* MTA with GFP-METTL5.