

Figure S4. Localisation of CARM1 in the mouse preimplantation embryo: differential distribution of CARM1 in 4-cell stage blastomeres.

(a)CARM1 exhibits both nuclear and cytoplasmic localisation in dots throughout all the stages analysed. Embryos were collected following hormonal induction at the indicated stages, fixed and processed for immunostaining with a CARM1 antibody (Upstate, green). DNA is shown in blue. Images were captured using a 60x oil objective in a BioRad Radiance Upright Confocal Laser Microscope. Shown are representative single optical sections of 10 embryos examined per stage.

(b) Levels of CARM1 are different in the blastomeres of the 4-cell stage embryo

Freshly collected tetrahedral four cell stage embryos were processed for immunostaining with a CARM1 antibody. The cells of individual embryos were then disaggregated and scanned under confocal microscopy separately. Shown are projections of the 4 blastomeres of a representative embryo (n=8). Sections were taken every 0.8 μ m. Scale bar 10 μ m.

(c)Total levels (nuclear and cytoplasmic) of CARM1 fluorescence were quantified through Z-planes using the Volocity software and normalised against the blastomere showing the highest level which was set at 100%. Decreasing values of fluorescence were calculated, normalised in each embryo and averaged accordingly (* p=0.0002). Each bar represents the relative fluorescence level of each of the 4-cell stage blastomeres.

(d) Comparison of the distribution of CARM1 (blue bars) and H3R26me2 (red bars) in tetrahedral (EM and ME) 4-cell stage embryos.