Supplementary Figure 10. Colocalization analysis of HCR-FISH control experiment with initiator probes and without amplifier hairpins. ANME-2b is stained in the Cyp3 channel, Seep-1g in the FITC channel. No dyes fluorescing in the Cy5 channel are present in this experiment. a. Scatterplots of pixel intensities of the FITC, Cy3 and Cy5 channel suggest there is some bleed-through in both the Cy3 and FITC channel. Neither the Pearson’s correlation coefficient, $PC = 0.165$, p-value = 100%, Costes’ method nor the Manders’ correlation coefficients, after thresholding, $M1 = 0.958$, $M2 = 0.39$, p-value = 100%, Costes’ method are high enough to indicate significant cross-correlation.

Correlation between ANME-2b and Seep-1g

Correlation between ANME-2b and nifH

Correlation between Seep-1g and nifH

HCR-FISH control experiment with initiator probes, No amplifier hairpins.