5 µM

-4 log₁₀ mean in vitro iCLIP signal

-2



0

1

В

750 nM

15 µM

-2



Supplemental figure S7, related to Supplemental Material. The iCLIP data approximate a normal distribution on logarithmic scale and indicate an absence of widespread cooperativity in U2AF2 binding. (A) The standard deviation scales with the mean of the iCLIP signal. Scatter plots showing correlation of mean and standard deviation of four biological replicate iCLIP measurements after normalization of each replicate by the total number of sequencing reads. Each dot represents one U2AF2 binding site. The U2AF2 concentration is indicated in each panel. (B) Measurement errors over all U2AF2 binding sites approximately follow a normal distribution. Quantile-quantile (QQ) plot relating the quantiles of the normalized in vitro iCLIP errors to a standard normal distribution. The measurement error of each replicate was calculated by subtracting its value from the mean over all replicates, i.e. [log₁₀(individual replicate) – mean{log., (individual replicate)}] separately for each binding site. The QQ plots resembles straight lines with similar slopes, with no obvious dose-dependent trend, implying that the log-error is constant across U2AF2 concentrations (highlighted by color, see legend). (C) Cooperative U2AF2 binding can be neglected for most binding sites. The goodness-of-fit as measured by a cost function (i.e. -2log(likelihood)) for the non-cooperative one-step binding model (Eq. 5, see Supplemental Material; y-axis) is compared to the fit of a(n) (anti-)cooperative model described by the Hill equation (x-axis) for each binding site. Hill coefficients in the range between 0.5 to 5 were allowed during fitting. Only few binding sites are better described by the (anti-)cooperative model, while on a global scale (considering the aggregate fit of all binding sites), the (anti-)cooperative model can be rejected using model selection criteria (see Supplemental Material).

log₁₀ standard deviation

-2

-3

-2 -3 3 µM

-2