Supplemental Materials and Methods

Correction of fluorescence brightness in assembled fraction

To correct the ratio of assembled fraction by taking consideration of the difference in the brightness of fluorescence between monomer and assembly, the following equations were used (*38*)

$$ {}_{r}F_{assembly} = \frac{F_{assembly}}{F_{assembly} + \alpha^{2} \cdot \left(1 - F_{assembly}\right)} \tag{S1} $$

with

$$ \alpha = \frac{cpm_{assembly}}{cpm_{monomer}} \tag{S2} $$

where $F_{assembly}$ is an assembled fraction estimated from autocorrelation amplitude, ${}_{r}F_{assembly}$ is a real fraction of assembly in a sample, and α is a correction factor that is the ratio of counts per molecule (*cpm*) of monomic rod fragment to *cpm* of assembled particles. The data of assembled fraction shown in Figs. 2 E and 2F was corrected with equations (S1) and (S2) (Figure S2).

Supplemental Figures

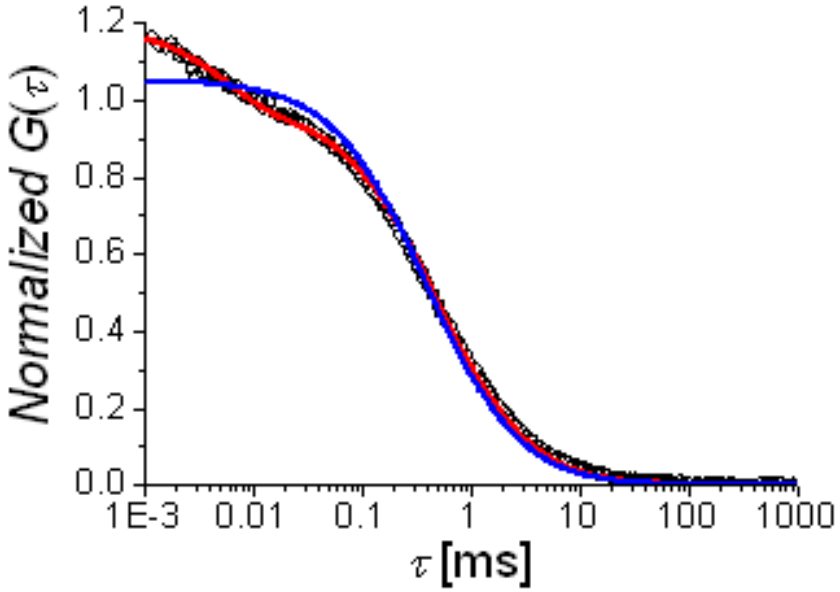

Fig. S1. Fitting analysis of autocorrelation curve

The autocorrelation curve of 633-ARF at 500 mM NaCl shown in Figure 1A was fitted with one-component model with or without triplet component (i = 1 in equation 2). The fitting curve without the triplet component (blue line) showed an unsatisfactory fit especially between 10^{-3} and 10^{0} (ms). The diffusion time was 404 μs. The fitting analysis with triplet component (red line) showed a good agreement with the data. The obtained diffusion time was 493 μs and the triplet time was 5.6 μs. Since the diffusion time of Alexa Fluor 633 was 54 μs, the estimated triplet time is too fast as a translation diffusion of free dye in this FCS apparatus.

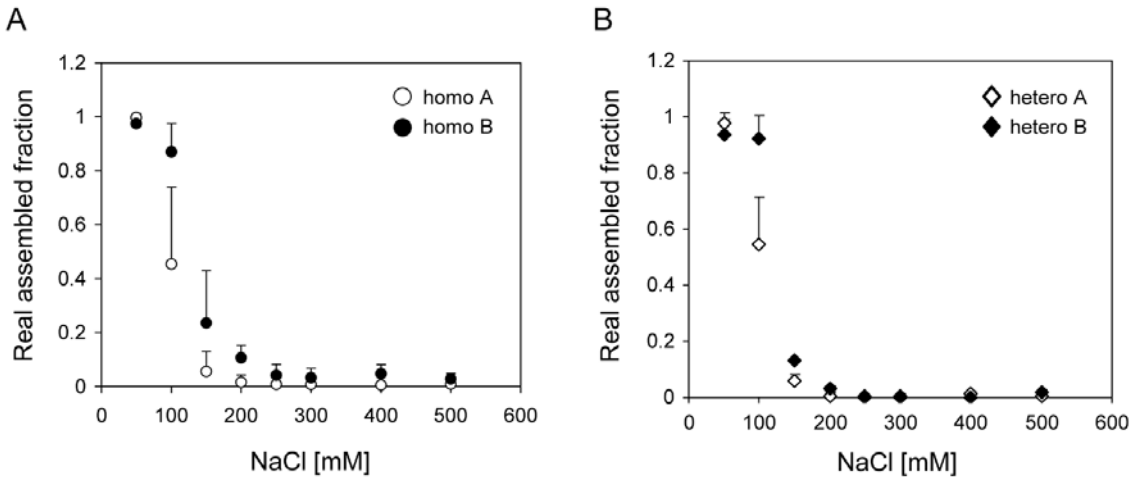

Fig. S2 Corrected fraction of assembly in the homo- and hetero-assembling conditions

The assembled fractions shown in Figure 2E and 2F were corrected by using equation (S1) and (S2) with the correct factor of each data set. (A) ${}_rF_{assembly}$ of 633-ARF (open circle) in the homo-mixture of 633-ARF and 488-ARF (open circle), 488-BRF (closed circle) in the homo-mixture of 633-BRF and 488-BRF as a function of NaCl concentration. (B) ${}_rF_{assembly}$ of 633-ARF (open diamond) and 488-BRF (closed diamond) in the hetero-mixture of 633-ARF and 488-BRF as a function of NaCl concentration. Data represent mean + S. D. of three independent experiments. The tendency of assembling properties of ARF and BRF was not changed from the original results, that is, ARF or BRF assembled in an isoform-dependent manner, regardless of the experimental conditions.