Supporting Information

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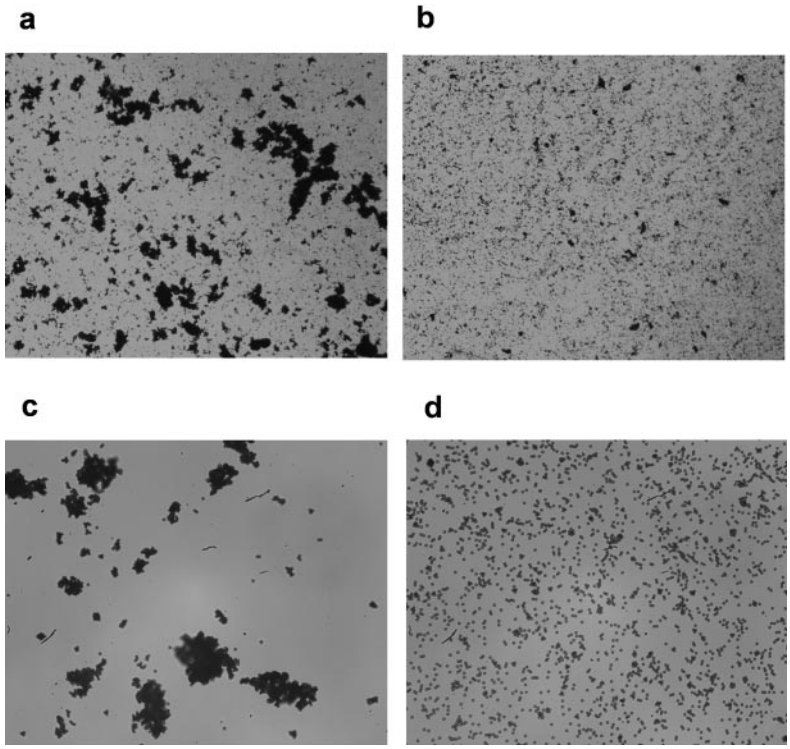

Fig. S1. Biological activity of the cadherin constructs were demonstrated by monitoring the aggregation and disaggregation of cadherin-functionalized beads in the presence or absence of Ca^{2+}, respectively. (*a*) E-cadherin His-tag monomer-coated beads incubated in 1.8 mM Ca^{2+} showed significant bead aggregation. (*b*) E-cadherin His-tag monomer-coated beads incubated in a Ca^{2+} free buffer did not aggregate. This buffer was prepared by passing 50 mM Tris buffer at pH 7.4 containing 100 mM NaCl, 10 mM KCl, and 0.2% (wt/vol) BSA (buffer TB) over a Chelex 100 resin (Bio-Rad) to chelate out Ca^{2+}. (*c*) Cadherin–Fc dimer-coated beads incubated in 1.8 mM Ca^{2+} showed significant bead aggregation. (*d*) Cadherin–Fc dimer-coated beads incubated in 2 mM EGTA (to chelate out free Ca^{2+}) did not aggregate. (Magnification: *a* and *b*, 11.2×; *c* and *d*, 22.4×.)

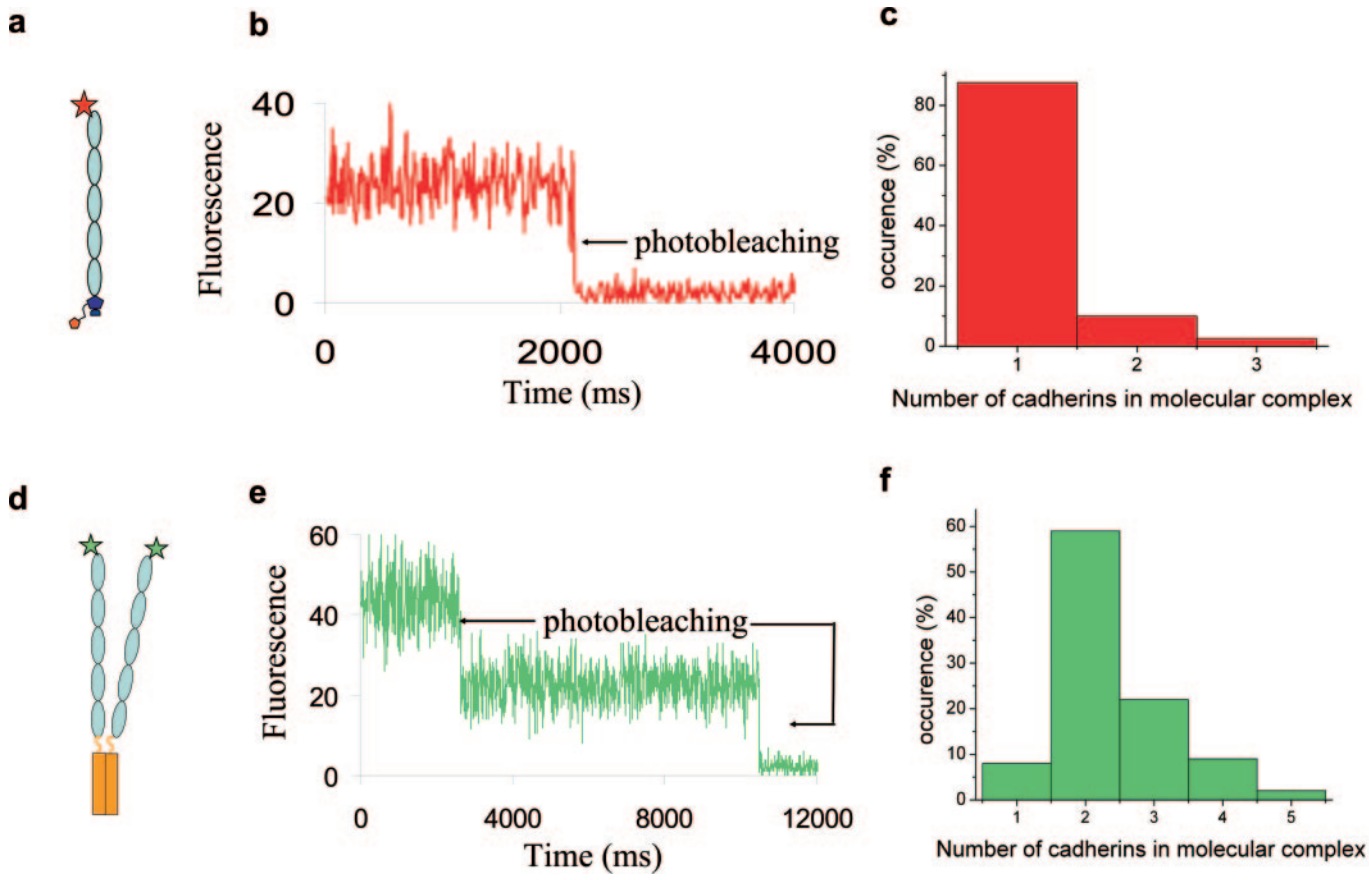

Fig. S2. Monitoring the oligomeric state of cadherin monomers and cadherin-Fc constructs. (*a*) The oligomeric state of the labeled cadherin monomer was measured by immobilizing the molecules on a surface, exciting the fluorophores, and monitoring their photobleaching. (*b*) A typical fluorescence time trace for the photobleaching of a monomer. Because the dye-to-protein labeling ratio was typically between 60% and 90%, monomers photobleach in a single step. (*c*) More than 85% of the cadherin molecules were observed to photobleach in a single step. (*d*) The dimeric state of the labeled cadherin–Fc construct was confirmed by immobilizing the molecules on a surface, exciting the fluorophores, and monitoring their photobleaching. (*e*) A typical fluorescence time trace for the photobleaching of a dimer. (*f*) More than 65% of the cadherin–Fc molecules were dimers.

a

Ca^{2+} Concentration	Molecules crosslinked
0 μM	20%
100 μM	40%
500 μM	32%
1000 μM	49%

b

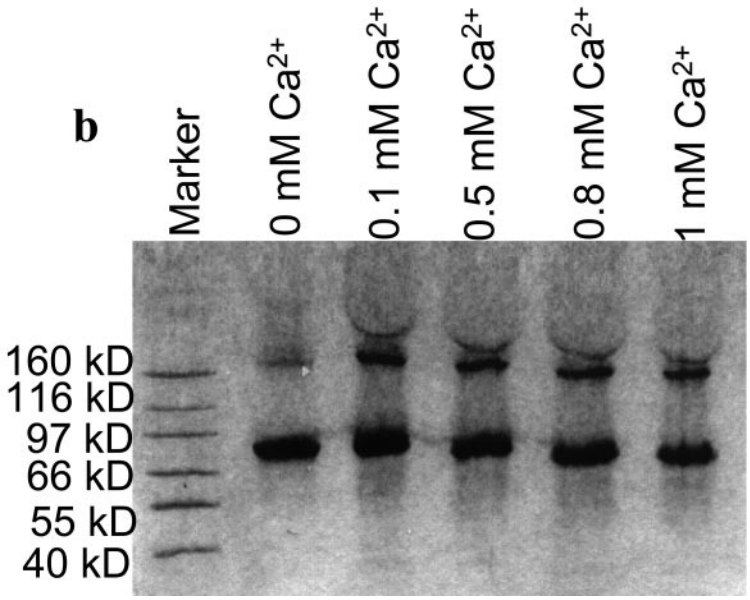

Fig. S3. Cadherin cross-linking yield at different calcium concentrations. (*a*) Donor- and acceptor-labeled cadherin monomers were cross-linked in solution to capture transient interactions between the proteins. Cross-linked dimers and non-cross-linked monomers were separated by using a sizing column, and the cross-linked dimer yield was measured by absorption at 280 nm. Cross-linking reactions were carried out in buffers containing 0, 0.1, 0.5, and 1 mM calcium. The cross-linked dimer yields at these calcium concentrations were 20%, 40%, 32% and 49%, respectively. (*b*) When the product of the cross-linking reaction is run on a denaturing gel and stained with coomasie dye, a cross-linked dimer fraction with molecular mass of ≈150 kDa and a noncross-linked monomer fraction with molecular mass of ≈75 kDa are observed. The yield of dimers cross-linked in 0 mM calcium is significantly lower than dimer yields at higher calcium concentrations.