

**Figure S4** Lethal insertion mutants affect the ability of the Dam1 complex to enhance binding of the Ndc80 complex to microtubules. Using TIRF microscopy, single molecules of GFP-tagged Ndc80 complexes were visualized on taxol-stabilized microtubules. Representative kymographs show the binding and one-dimensional diffusion of 50 pM GFP-tagged Ndc80 complexes on microtubules in the presence or absence of 2.5 nM untagged Dam1 complex.