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Abstract, Please use no more than 300 words
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Observation of tubulin incorporation events into growing microtubules using single molecule fluorescence.

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Microtubules are hollow tubes consisting of typically 13 parallel protofilaments that *in vitro* and *in vivo* grow by the addition of tubulin dimers to their growing ends. It is generally believed that the growing ends of microtubules consist of several individual protofilaments that after assembly first bind to each other laterally in a slightly outward-curved sheet and then close into a straight hollow tube. The kinetic details of the tubulin assembly process itself as well as the conformational changes of the sheet-like structures at the growing ends remain poorly understood. Fluorescence microscopy can be used to monitor the incorporation of fluorescently labeled tubulin subunits into single microtubules *in vitro* or *in vivo*. When low labeling ratios are used, so-called speckles are observed that result from spatial fluctuations in the distance between individual fluorescent labels. These speckles are a powerful tool in monitoring microtubule dynamics, transport and treadmilling in living cells at the normal resolution of fluorescence microscopy. Here we push the observation of microtubule speckles to the single fluorophore limit. Using TIRF microscopy and a very low labeling ratio we demonstrate the observation of single label incorporation into microtubules growing from pure tubulin *in vitro*. With improved time resolution, the ability to observe tubulin addition at the single molecule level should allow for the detection of individual attachment as well as detachment events, which will help settle existing controversies about the off-rate for tubulin in growing microtubules. Also, tracking the lateral position of newly added tubulin subunits at nanometer resolution, should allow for the detection of straightening events of putative outward-curved sheets at the ends of growing microtubules.