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Photo-Activated Localization Microscopy of Single Carbohydrate Binding Modules on Cellulose Nanofibers[1] AMY HOR, DARYL DAGEL, QUOCANH LUU, MADHUSUDAN SAVAIKAR, South Dakota School of Mines and Technology, SHI-YOU DING, Michigan State University, STEVE SMITH, South Dakota School of Mines and Technology — Photo Activated Localization Microscopy (PALM) is used to conduct an in vivo study of the binding affinity of polysaccharide-specific Carbohydrate Binding Modules (CBMs) to insoluble cellulose substrates. Two families of CBMs, namely *Tr*CBM1 and *Ct*CBM3, were modified to incorporate photo-activatable mCherry fluorescent protein (PAmCherry), and exposed to highly crystalline *Valonia* cellulose nano-fibrils. The resulting PALM images show CBMs binding along the nano-fibril long axis in a punctuated linear array, localized with, on average, 10 nm precision. Statistical analysis of the binding events results in nearest neighbor distributions between CBMs. A comparison between *Tr*CBM1 and *Ct*CBM3 reveals a similarity in the nearest neighbor distribution peaks but differences in the overall binding density. The former is attributed to steric hindrance among the CBMs on the nano-fibril whereas the latter is attributed to differences in the CBMs' binding strength. These results are compared to similar distributions derived from TEM measurements of dried samples of *Ct*CBM3-CdSs quantum dot bioconjugates and AFM images of *Ct*CBM3-GFP bound to similar *Valonia* nano-fibrils.

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