UNRAVELING THE MECHANISMS OF CENTRIOLE FORMATION IN HUMAN CELLS USING ADVANCED MICROSCOPY TECHNIQUES

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Centrioles are minute barrel-shaped organelles of ~130 nm inner diameter and exhibiting a conserved nine-fold radial symmetric arrangement of microtubules. SAS-6 proteins are thought to impart the nine-fold symmetry of newly formed centrioles [1], but the mechanisms by which this occurs within cells remain elusive. We revealed the ring-like organization of the centriolar protein HsSAS-6 in cells using 3D-STORM with optimized buffer conditions, enabling significantly increased dye brightness and near-isotropic resolution of ~30 nanometer [2]. Moreover, we found that the angular distribution of signal intensities exhibits an average quasi-period of ~42 degrees, strongly supporting the notion that HsSAS-6 proteins exhibit a nine-fold symmetric distribution in vivo.

Furthermore, using FCS, we uncover that HsSAS-6 is present in the cytoplasm primarily as a homodimer, and that its oligomerization into a nine-fold symmetrical ring occurs at centrioles. Finally, FRAP analysis demonstrates that HsSAS-6 is immobilized progressively at centrosomes during cell cycle progression, and that this is accompanied by an increased size of the underlying structure, as shown by 3D-STORM imaging.

These findings lead us to propose a mechanism whereby HsSAS-6 homodimers are targeted to centrosomes where the local environment and high concentration of HsSAS-6 promote oligomerization, thus initiating procentriole formation [3].