

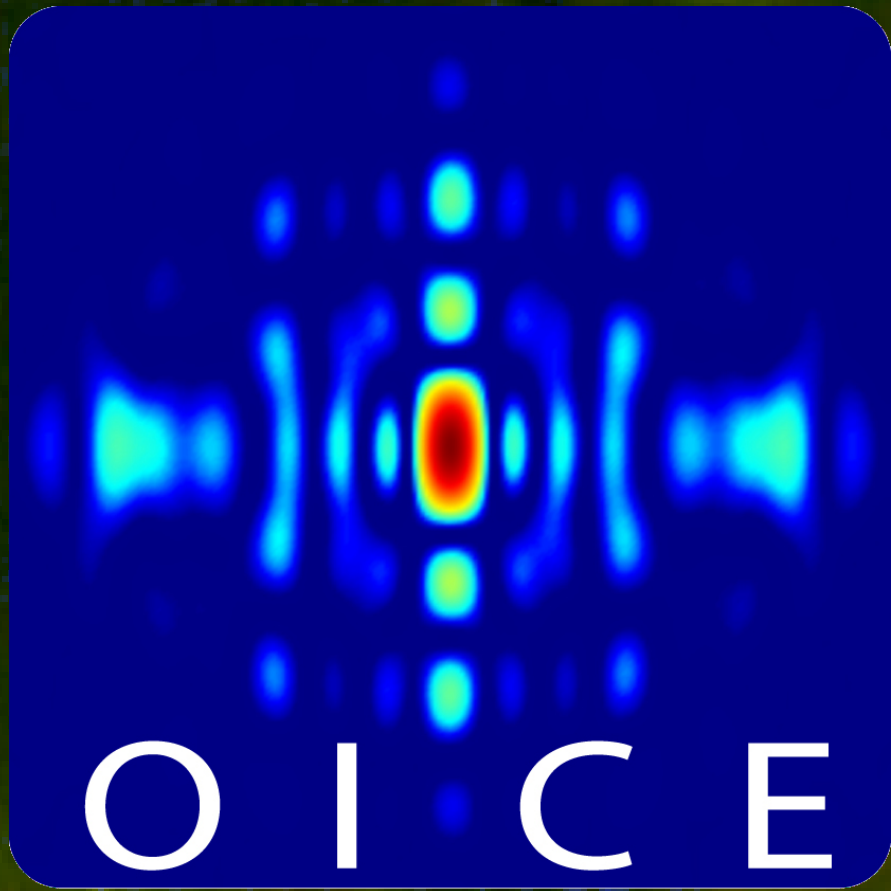


Evaluation of Highlighter Pens as Cheap and Cheerful Samples for Microscope Calibration and Performance Testing

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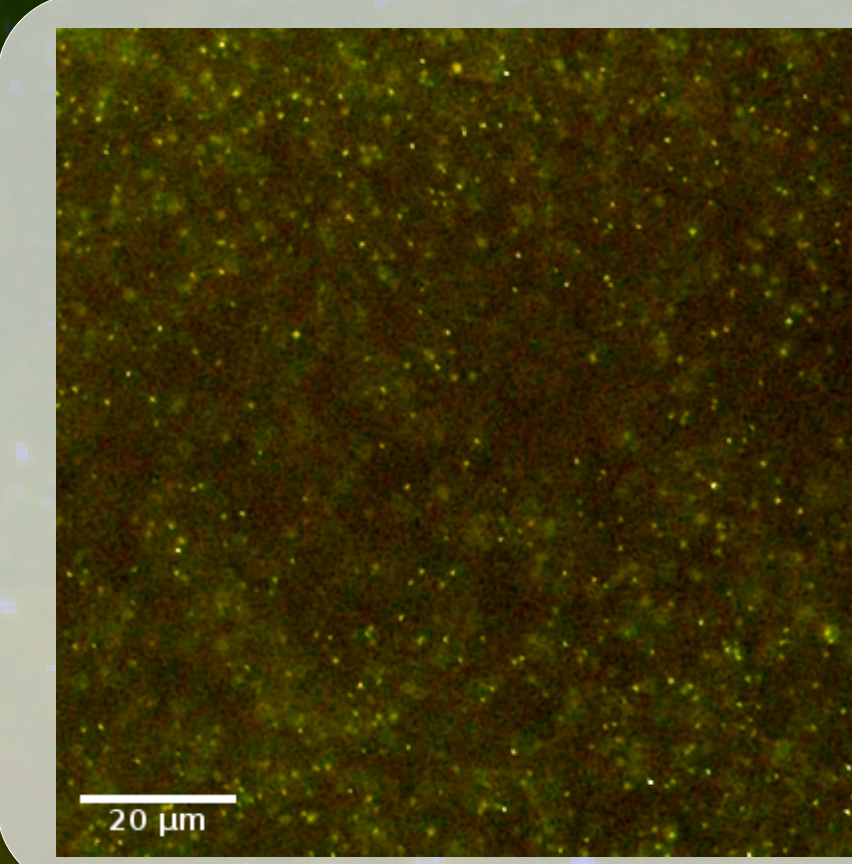
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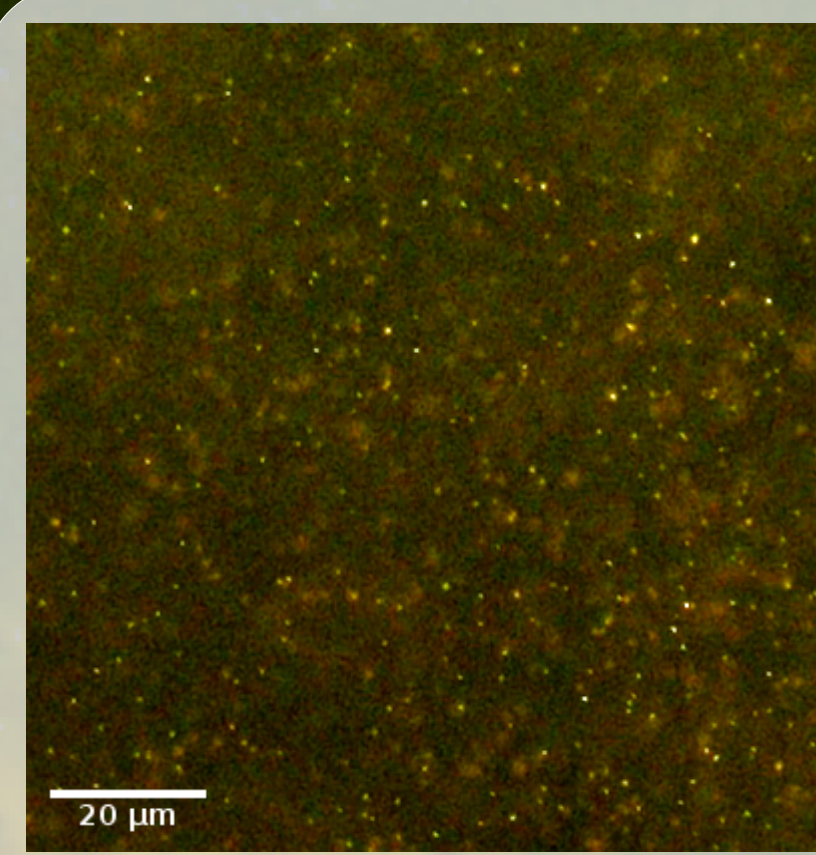
MAX-PLANCK-GESELLSCHAFT

INTRODUCTION:

The bright appearance of highlighter pens is usually connected to the presence of some fluorescent component within the highlighter fluid. Those components can be dissolved fluorophores or a suspension of fluorescent particles. Here we evaluate several highlighter pens and their fluids with regard to their usefulness in the context of microscope performance evaluation.

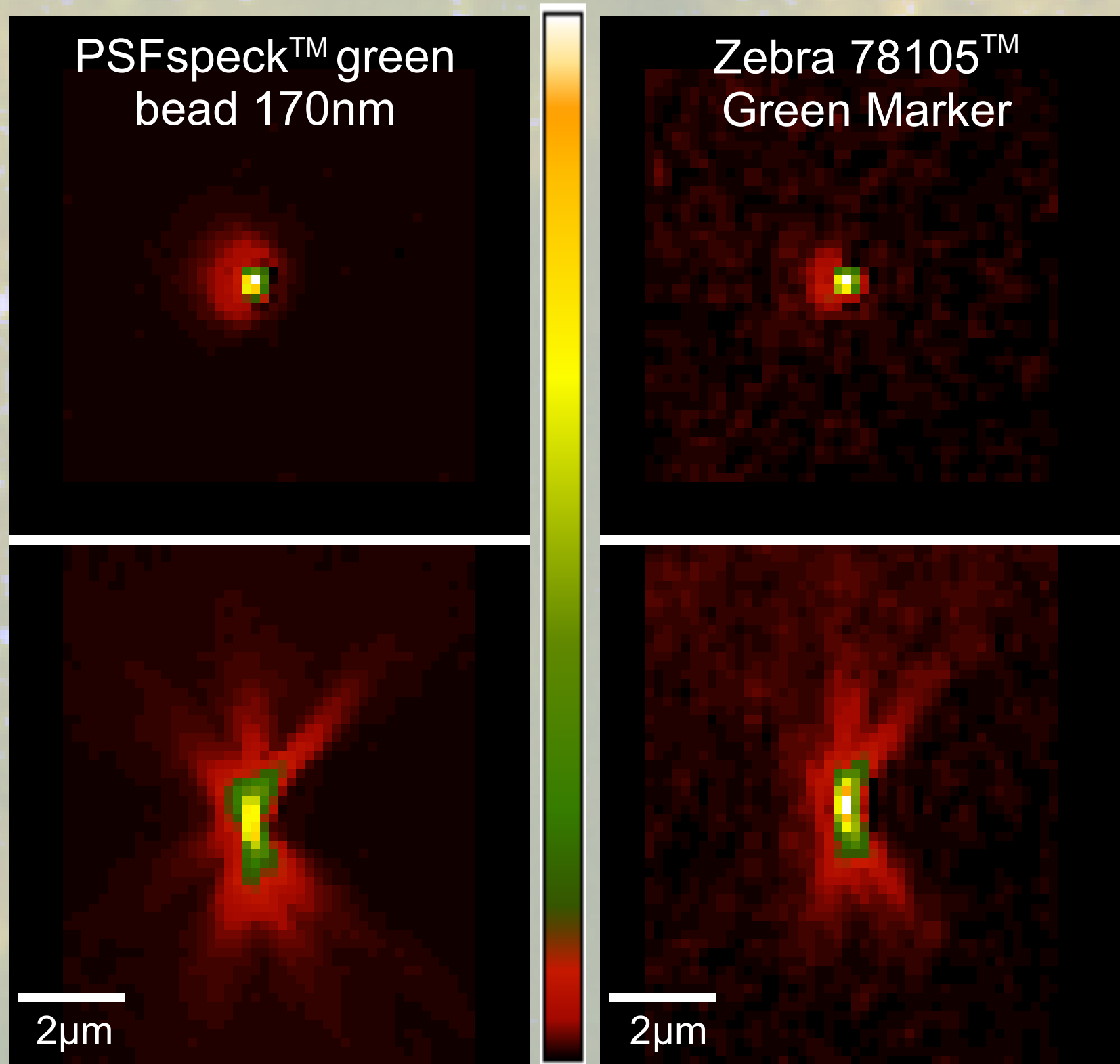


Zebra 78105 (Mildliner)TM pens contain spherical sub-resolution fluorescent particles of uniform size.



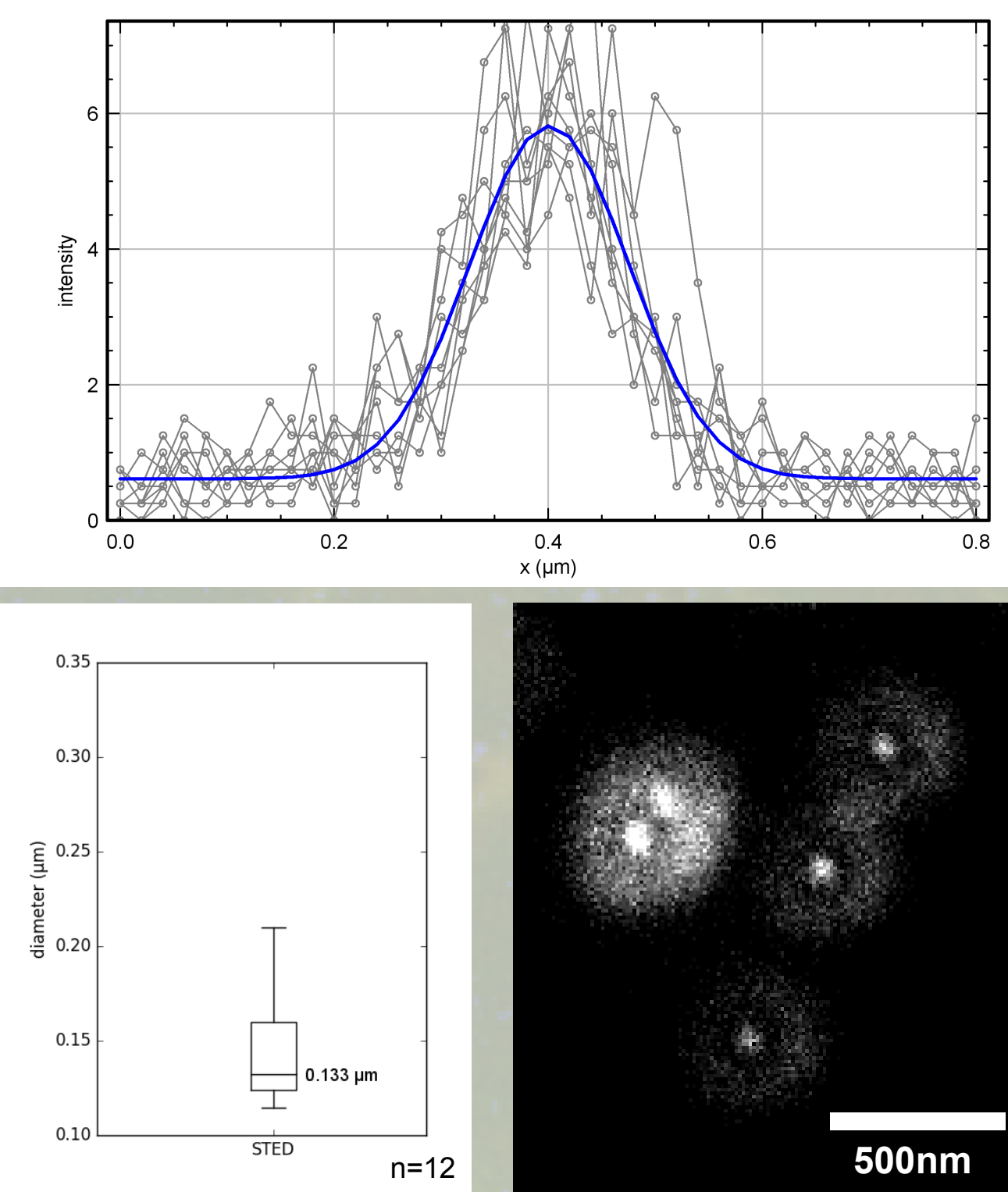
Stabilo Pen 68 neonTM pens contain spherical particles which are quite bright and of non-uniform size. Apart from fluorescence also darkfield can be used to visualise the particles.

Zebra 78105TM particles cross sections show same features as commercial beads used for microscope performance control.

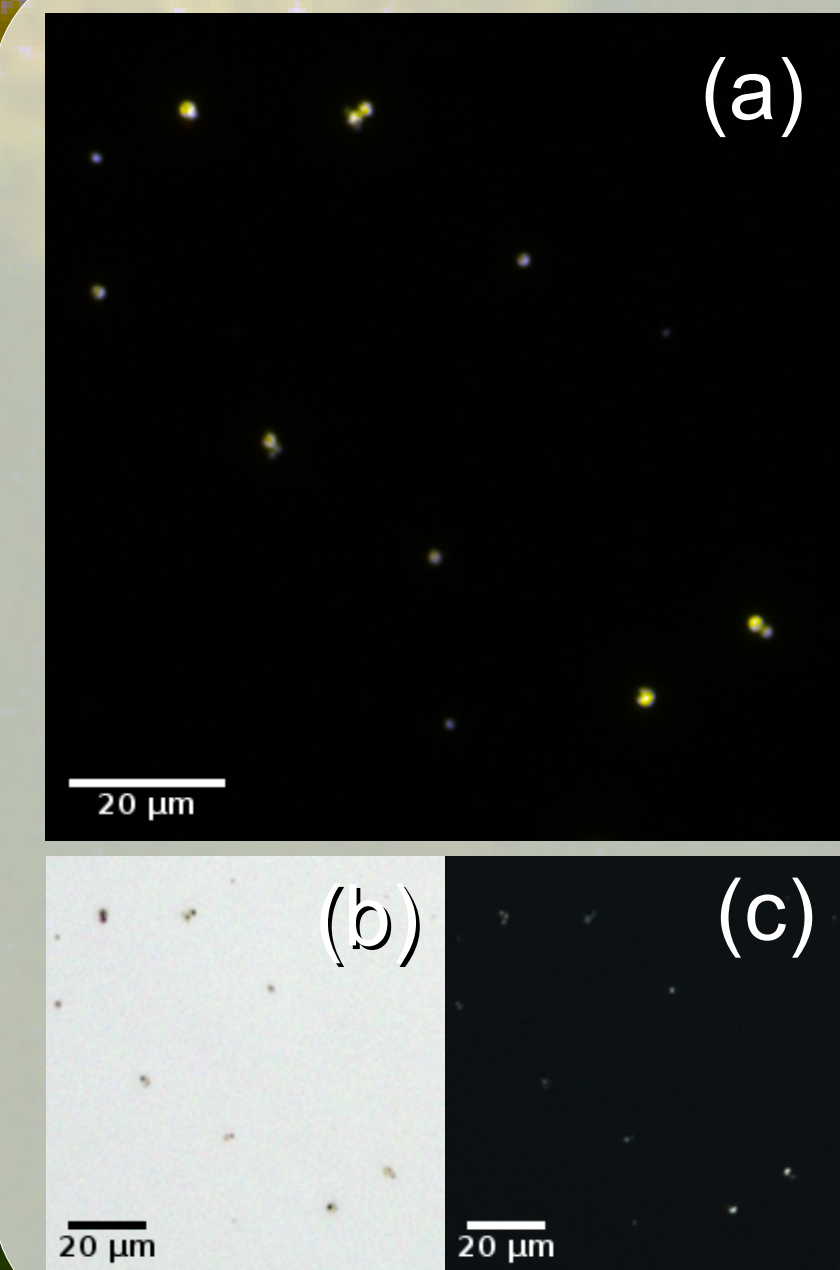


Widefield, Plan-Apochromat 63x/1,40 Oil filter 450-490/495/500-550

Zebra 78105TM orange beads are stable enough to withstand STED conditions and exhibit a size of ~130nm in diameter.

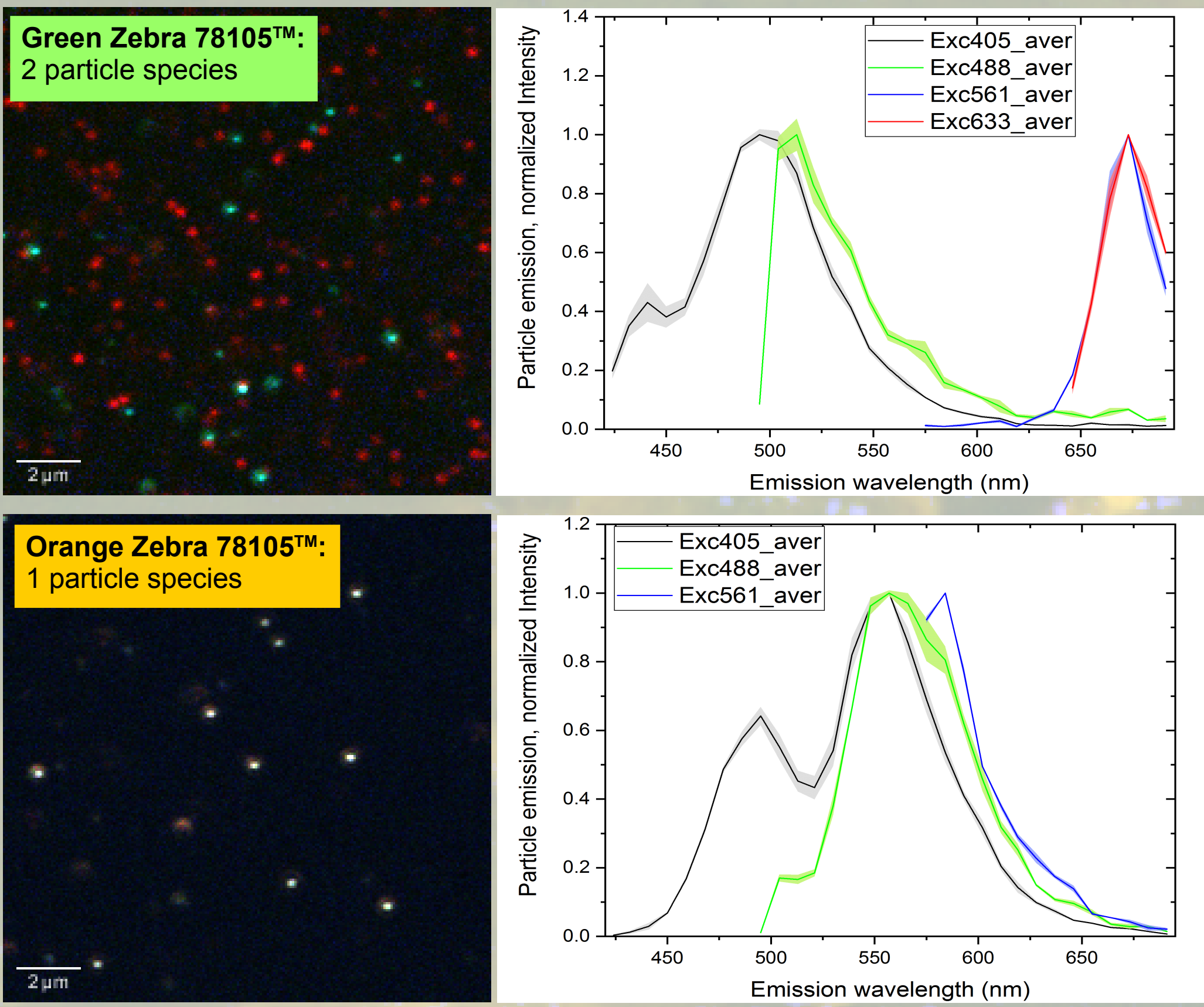


STED, Plan-Apo 100x/1.46 Oil, ex594, depl775 @ 1250mW

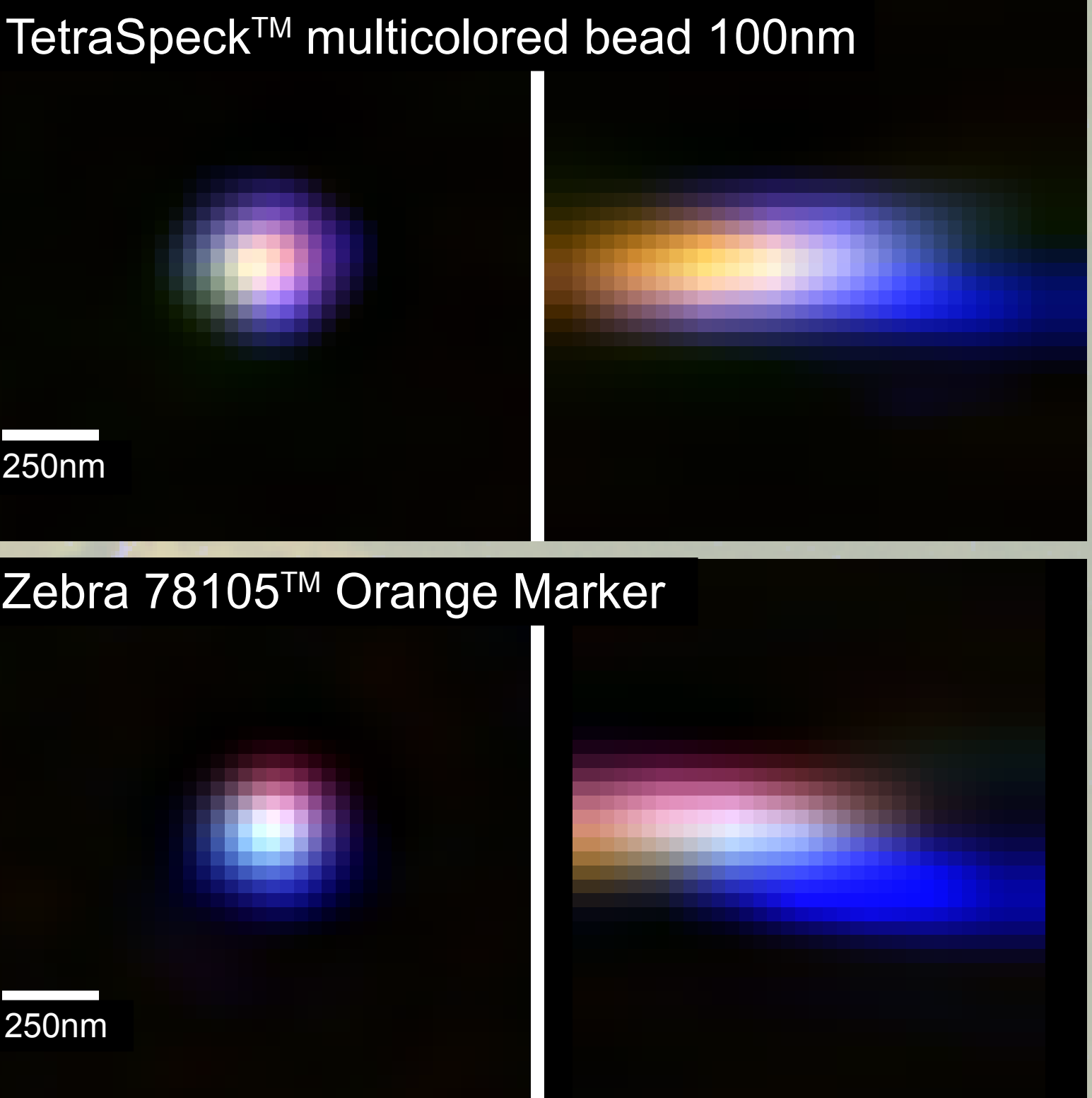


Pilot Frixion light and light softTM pens contain bright, comparably large and quite irregular particles, usually with a mixture of fluorophores on a single particle (a), they are also easily detectable with brightfield (b), DIC (c) and darkfield (not shown).

Spectra of **Zebra 78105TM** show a strong reproducibility and their shape is ideally suited to characterise the precision of spectral detectors.



Detection of chromatic aberrations



Airyscan, Plan-Apochromat 63x/1,40 Oil Ch1 (blue) ex405, Ch2 (green) ex488, Ch3 (red) ex561

CONCLUSIONS:

Fluids of highlighter pens and the particles therein can be used as a cost effective alternative to classic beads for a wide range of quality control tasks and all basic (and some advanced) maintenance measurements can be performed with the aid of these. From the pens used in our study we found the Zebra 78105 (Mildliner)TM pens to be the most versatile one.

please find more information at:
<http://webdav.tuebingen.mpg.de/LM/>



Which brand & model of pen can be used for what

brand/model	type	Parfocality/ Parcentricity/ stage precision	PSF recording	spectral registration	spectral calibration	field flatness	illumination homogeneity
ZebraPen 78105 Mildliner pastel	particle	fluorescence only	yes	yes	yes	sort of	limited
Pilot Frixion light/light soft	particle	yes	no	no	no	sort of	no
Stabilo pen 68 neon	particle	fluorescence only	no	-/-	-/-	sort of	no
Stabilo BOSS neon	solution	no	no	no	no	no	yes
Pelikan 438	solution	no	no	no	no	no	yes