
Emerging Imaging Techniques with One nm Accuracy

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Abstract: *A single dye or a single group of dyes can be localised to within 1-nm accuracy using the fluorescence imaging with one-nanometer accuracy (FIONA) approach. Total internal reflection fluorescence microscopy, oxygen removal techniques, and a high quantum yield, low-noise detector are used to attain this high level of precision. FIONA comes in a variety of forms, some of which have resolutions better than 10 nm. One of these alternatives is single- molecule high-resolution imaging with photo bleaching (SHRIMP), which only needs one kind of dye, such as two green fluorescent proteins (GFPs) or two rhodamines. SHRIMP can only produce high resolution on static systems, though. On the other hand, single-molecule high-resolution colocalization (SHREC) is a FIONA variation that can achieve high resolution in dynamic systems. DOPI, which stands for Defocused Orientation and Positional Imaging, allows the 3-D Orientation.*

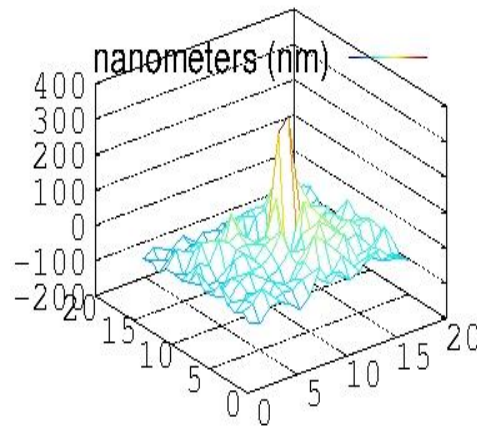
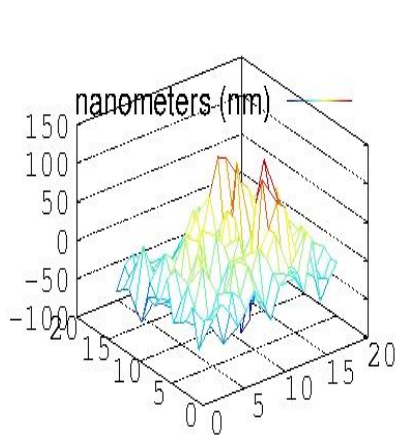
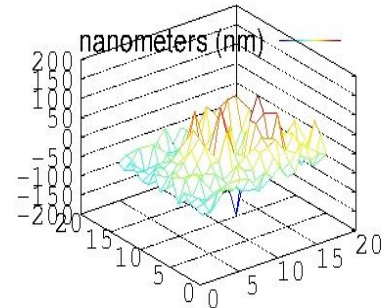
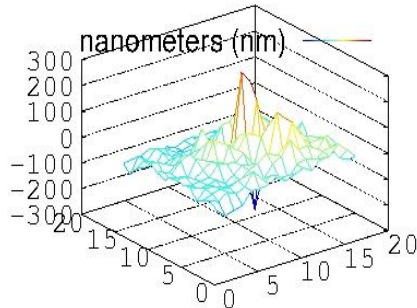
Keywords: *1nm Resolution Accuracy, High level Precision, SHREC.*

1. INTRODUCTION

The microscope enables researchers to look inside tiny objects. The microscope has expanded our capacity to decode biological information from selectively labelled proteins, DNA, and organelles using novel chemical dyes and fluorescent proteins.

2. METHODOLOGY

Although it is nearly impossible to see objects smaller than the diffraction limit, which is approximately 250 nm, due to a physical property of light known as diffraction. However, a lot of information can be insightful at a smaller scale. FIONA is a method that enables this without exceeding the diffraction limit. With FIONA, a diffraction-limited spot's position can be pinpointed on a time scale longer than a millisecond to within a few nano meters. In this chapter, we go over some fundamental ideas that underpin FIONA and its use



Sl.no	Pixel size nm	Background Noise
1	106.7	22
2	260	20
3	270	30
4	280	40
5	290	50

Sl.NO	Exposing Time Sec	Photon Conversion Factor
1	0.1	0.449
2	1	0.5
3	2	1.0
4	3	1.5
5	4	2.0

3. RESULTS

A 50 mm × 50 mm area is covered by the Molecular Measuring Machine (MMM), which aims to carry out 2D point-to-point measurements with one nanometer accuracy. With sub-nanometer precision, the equipment combines a scanning tunneling microscope (STM) to probe the surface with a Michelson interferometer system to quantify the probe movement. The instrument also has seismic and acoustic vibration isolation, millidegree temperature control at 20 degrees Celsius, and an ultra-high vacuum environment with a base pressure below 10(-5) Pa. On 1D gratings, high-accuracy pitch measurements have been made. In one experiment, a 5 micron by 1 millimeter region was covered by the MMM STM probe's images of a collection of laser-focused, atomically formed chromium lines. The data analysis produced an average line spacing of 212.69 nm with a Chromium lines.

The computer, data processing, and social media-related global business drivers keep pushing the standards for semiconductor manufacturing to heights never before reached. The development of manufacturing methods with the accuracy levels required to build the next-generation chip is being worked on by individual manufacturers as well as technological consortiums all across the world.

4. CONCLUSION

Identifying the technology gaps necessary to attain next generation performance presents issues in a number of different areas. Precision has been highlighted as a key requirement for motion control, namely to produce structural stability and position stability at the single nanometer level. To close the gaps, material metrology must boost processing speed and accuracy while preserving position stability. For instance, image resolution at the 100nm level is anticipated.

5. REFERENCES

1. R Roy, S Hohng, T Ha, A practical guide to single-molecule FRET. *Nat Methods* 5, 507–516 (2008).
2. T Ha, Single-molecule methods leap ahead. *Nat Methods* 11, 1015–1018 (2014).
3. LS Churchman, Z Okten, RS Rock, JF Dawson, JA Spudich, Single molecule high-resolution colocalization of Cy3 and Cy5 attached to macromolecules measures intramolecular distances through time. *Proc Natl Acad Sci USA* 102, 1419–1423 (2005).
4. MJ Rust, M Bates, X Zhuang, Sub-diffraction-limit imaging by stochastic optical reconstruction microscopy (STORM). *Nat Methods* 3, 793–795 (2006)