ARTICLES

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Identification of spatially associated subpopulations by combining scRNAseq and sequential fluorescence *in situ* hybridization data

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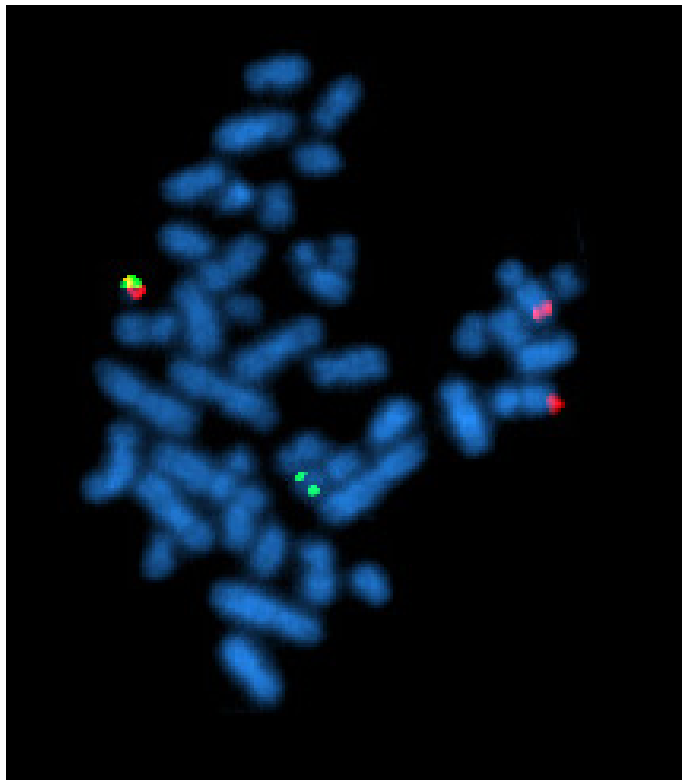
Overview

Background - On the other side of NGS

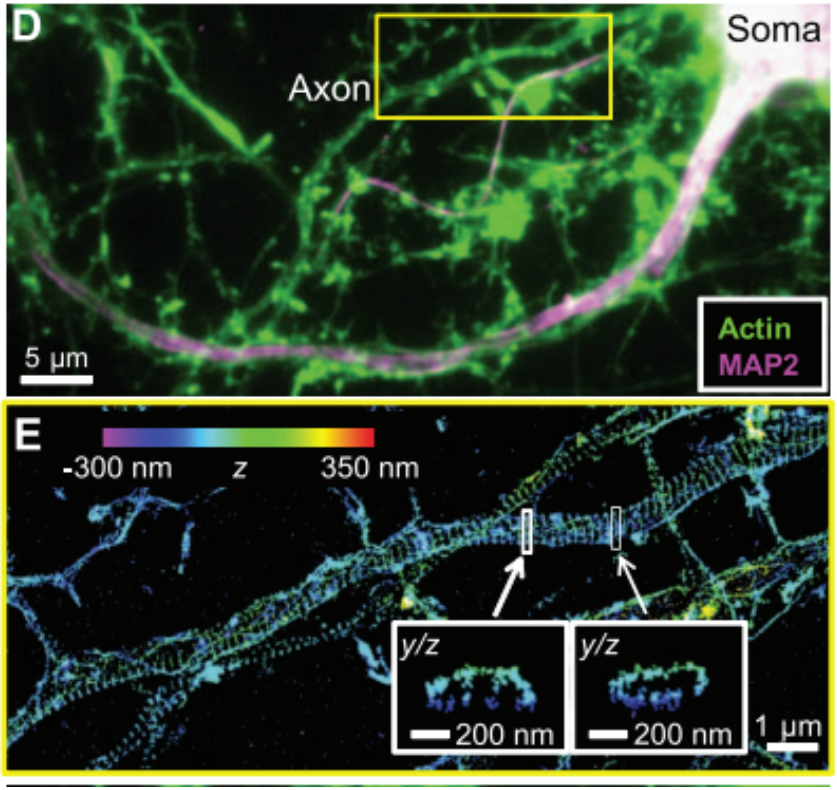
FISH and single molecule FISH



- FISH: what is FISH? [Wikipedia, 2019]
- single molecule FISH: pioneered by [Femino et al., 1998], enhanced version by [Raj et al., 2006]

Super-resolution microscopy

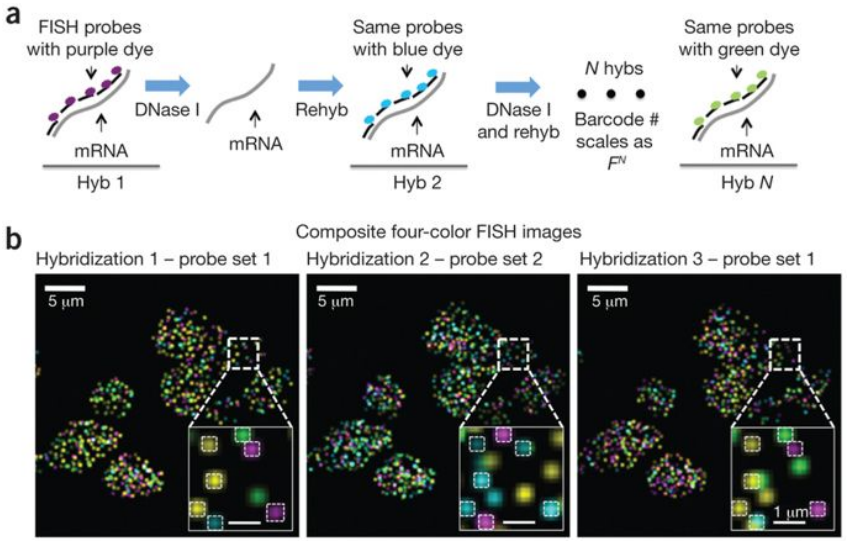
- Resolution 10-20 nm on x, y and ¡50 nm on z direction.
  - It helps to unreal finer structure (under diffraction barrier)



[Xu et al., 2013]

Sequential FISH

- Encode sequencing with combination of color [Lubeck and Cai, 2012]
  - Encode sequencing with order of color



[Lubeck et al., 2014]

- Sequencing by hybridization, *e.g.* two rounds three channels:
  - red- cyan→ Gene 1
  - green- cyan→ Gene 2
  - cyan- red→ Gene 3
- The complexity of the target library: number of hybridization × number of channels/probes

What is good about seqFISH?

- Low drop out rate ($\sim$ 94% per round), very low probability to have two drop outs
- So that it is also easy to correct for this drop out error by one extra round
- 84% efficacy (capture 84% variation presented in gold standard method smHCR, hybridization chain reaction)
- As compared to single-cell RNA-seq, efficacy is 5-20%

[Shah et al., 2016]

What is good about seqFISH? More importantly?

- As an in situ technology, it preserves spatial information which is erased in scRNA-seq

[Shah et al., 2016]

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