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🌐 SABER-FISH – Signal amplification for multiplexed fluorescence *in situ* hybridization assays

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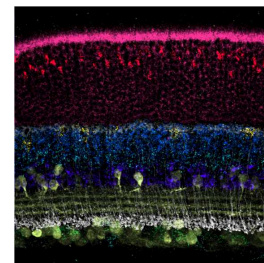
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We are still developing and optimizing this collection

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Abstract

The SABER Technology

The Signal Amplification By Exchange Reaction (SABER) method is used for amplifying signal from multiplexed *in situ* fluorescence staining experiments. Developed by the Yin and Cepko labs at Harvard University and the Wyss Institute, the technique uses **Primer Exchange Reactions (PERs)** to generate three-letter (A, T, C) concatemeric sequences in bulk *in vitro* reactions. These concatemers can then be *in situ* hybridized to fixed cells and tissues and act as scaffolds that localize fluorescent 'imager' strands. The method can further be paired with **DNA-Exchange Imaging (DEI)** to increase multiplexing via rapid stripping of old imager strands and hybridization of new imager strands (imager exchange) and/or cell segmentation with puncta counting on a per cell basis for quantitative analyses.

See the following references and resources below for further information. SABER provides a scalable and cost-effective way to amplify multiplexed *in situ* stainings for RNA/DNA (**SABER-FISH**) and protein targets (**Immuno-SABER**, [website](#), [collection on protocols.io](#)).

SABER-FISH

Multiplexed signal amplification enables rapid tissue mapping by increasing the number of targets that can be visualized per sample and reducing the exposure times required to see signals. Signal amplification in fluorescence *in situ* hybridization (FISH) assays can also increase sensitivity, potentially allowing smaller genomic and transcriptomic loci to be imaged, fewer probes to be applied, and/or less expensive microscopy setups to be utilized. SABER-FISH uses *in vitro* **Primer Exchange Reactions (PERs)** to synthesize long repetitive 'concatemer' sequences onto the 3' end of probes designed to be complementary to DNA and RNA targets of interest. After concatemer extension, probes are *in situ* hybridized to fixed cell and tissue samples, followed by a short secondary fluorescent hybridization that binds 20nt fluorophore-conjugated strands ('imagers') to the concatemers.

Multiplexing with SABER-FISH is achieved through the use of orthogonal concatemer sequences appended to probe sets, which can be read out to spectrally separated fluorophores on complementary imagers. Imager sequences can also be stripped from one set of concatemers without disrupting the underlying probe binding, which allows new sets of imagers targeting different loci to be imaged in iterative rounds of fluorescence imaging. This process of stripping and hybridization is referred to as **DNA-Exchange Imaging (DEI)**. SABER-FISH signal can further be enhanced with a branching strategy, where multiple rounds of concatemerized probe binding create branched structures *in situ*. Below are a number of resources intended to help adoption of the technology, including references, animations, and protocols.

Attachments



[SABER amplifies FISH...](#)

3.3MB

Guidelines

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Safety warnings

 For hazard information and safety warnings, please refer to the SDS (Safety Data Sheet).



Attachments



SABER
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3.3MB

Files

 SEARCH

Protocol



NAME

1: User-friendly protocol: Probe set design (SABER-FISH)

VERSION 1

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NAME

2: User-friendly protocol: Oligo ordering and preparation (SABER-FISH)

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3: 30mer branch melting temperatures (SABER-FISH)

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NAME

4: User-friendly protocol: Cost-efficient Primer Exchange Reaction (PER) concatemerization (SABER-FISH)

VERSION 1

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Protocol



NAME

5: User-friendly protocol: SABER RNA FISH in cells

VERSION 1

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Protocol



NAME

6: Protocol optimization for SABER-FISH in tissues

VERSION 1

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NAME

7: User-friendly protocol: Retina Tissue Sections RNA FISH

VERSION 1

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