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

In 1 collection

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Human Cell Atlas Method Development Community

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ABSTRACT

This protocol describes the design of 30mer branch sequences.



This protocol is part of the SABER-FISH collection.

EXTERNAL LINK

http://saber.fish/

THIS PROTOCOL ACCOMPANIES THE FOLLOWING PUBLICATION

Kishi, J.Y., Lapan, S.W., Beliveau, B.J. et al. SABER amplifies FISH: enhanced multiplexed imaging of RNA and DNA in cells and tissues. Nat Methods 16, 533–544 (2019). https://doi.org/10.1038/s41592-019-0404-0

ATTACHMENTS

SABER amplifies FISH_enhanced multiplexed imaging of RNA and DNA in cells and tissues.pdf

PROTOCOL CITATION

Jocelyn Y. Kishi, Sylvain W. Lapan, Brian J Beliveau, Emma R. West, Allen Zhu, Hiroshi M. Sasaki, Sinem Saka, Yu Wang, Constance L Cepko, Peng Yin 2020. 3: 30mer branch melting temperatures (SABER-FISH).

protocols.io

https://protocols.io/view/3-30mer-branch-melting-temperatures-saber-fish-bh9hj936

MANUSCRIPT CITATION please remember to cite the following publication along with this protocol

Kishi, J.Y., Lapan, S.W., Beliveau, B.J. et al. SABER amplifies FISH: enhanced multiplexed imaging of RNA and DNA in cells and tissues. Nat Methods 16, 533–544 (2019). https://doi.org/10.1038/s41592-019-0404-0

EXTERNAL LINK

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

KEYWORDS

branch melting temperature, temperature

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PARENT PROTOCOLS

Part of collection

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

SAFETY WARNINGS

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

1 It's recommended to use a temperature at least 1 degree lower than the lowest melting temperature of all branch sequences you plan to use (see plot of melting temperatures Fig. S2).

You can find these melting temperature curves reported for each sequence, as well as those computed for 20mer imagers, 42mer barcode sequences, and an example set of FISH probes, reported in

Supplementary Table 2.xlsx (available on the Nature Methods website).

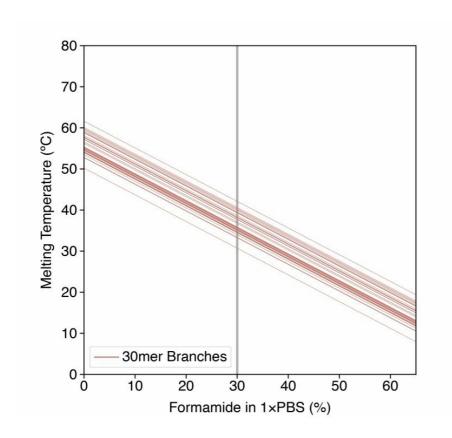


Figure S2: Melting temperatures of branches under different formamide conditions. Melting temperatures of 30mer branch binding sequences are shown for the 50 designed PER primers in $1\times PBS$ with different concentrations of formamide. Modeled with Biopython. 69