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# **Small** Micro

### Supporting Information

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Characterization of Membrane Patch-Ion Channel Probes for Scanning Ion Conductance Microscopy

Wenqing Shi, Yuhan Zeng, Cheng Zhu, Yucheng Xiao, Theodore R. Cummins, Jianghui Hou, and Lane A. Baker\* Supporting Information

## Characterization of Membrane patch (MP) - Ion Channel Probes (ICPs) for Scanning Ion Conductance Microscopy

Wenqing Shi,<sup>1†</sup> Yuhan Zeng,<sup>1†</sup> Cheng Zhu,<sup>1</sup> Yucheng Xiao,<sup>2</sup> Theodore R. Cummins,\* Jianghui Hou\* and Lane A. Baker\*<sup>1</sup>

<sup>1</sup>Department of Chemistry Indiana University 800 E. Kirkwood Avenue Bloomington, Indiana 47405

<sup>2</sup>Department of Biology Indiana University-Purdue University Indianapolis Stark Neurosciences Research Institute 723 West Michigan Street, SL-306 Indianapolis, Indiana 46202

<sup>3</sup>Department of Internal Medicine – Renal Division Division of Biological and Biochemical Sciences Washington University School of Medicine 660 South Euclid Avenue, Campus Box 8126 St. Louis, Missouri 63110

\*Authors to whom correspondence should be addressed.

<sup>1</sup>E-mail: <u>lanbaker@indiana.edu</u>; Phone: (812) 856-1873; Fax: (812) 856-8300

<sup>†</sup> These authors contributed equally to this work.

#### Approach curve

The MP-ICP probe was approached to the PI membrane surface after the fixed-position measurements, and the current response of the SICM barrel was recorded as a function of probe-sample distance to obtain the approach curve. The zero probe-sample distance was determined by taking the first derivative of the current curve, and a maximum derivative is expected at zero probe-sample distance.<sup>1</sup> Ideally, current drops to zero when the probe first makes contact with the surface, but due to any unequal height of the two barrels or tilt between the probe tip and sample, a zero current was not reached at contact between probe and sample and probe was further approached into the PI membrane substrate. This causes the approach curve to show "negative" distance. Moreover, the approach curve was obtained with the SICM barrel, and the probe-sample distances determined from this approach curve were used as distances of the ICP barrel in molecule/ion flux measurements, errors exist due to any unequal height of the two barrels.

#### FIGURE CAPTIONS

**Figure S1.** Current-voltage (I-V) relationship of a single stably-transfected BK channel derived from I-T measurements from an inside-out membrane patch.

Figure S2. Approach curve of the MP-ICP probe used for measuring local  $Ca^{2+}$  flux.

The dashed line indicates a zero probe-sample distance.

FIGURES



Figure S1. Shi. et al.



Figure S2.

Shi et al.

#### REFERENCES

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