## Electronic Supplementary Information for:

## Microarray-based fluorescence assay of endonuclease functionality and inhibition

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Additional figures S1-S3

Additional scheme S1



**Fig. S1** Fluorescence images and corresponding data analysis upon different concentration of E1 in spotting solution. Column 1-4: 1  $\mu$ M, 10  $\mu$ M, 30  $\mu$ M and 50  $\mu$ M, respectively. The microarray is blocked with 0.8 mg/mL PEG-NH<sub>2</sub> and incubated with 500 U/mL EcoRI for 10 h.



**Fig. S2** Fluorescence images and corresponding relative fluorescence intensity upon different blocking conditions: (a) 0.1 M ethanolamine, (b) 100 mg/mL PEG-NH<sub>2</sub>, (c) 20 mg/mL PEG-NH<sub>2</sub>, (d) 4 mg/mL PEG-NH<sub>2</sub>, (e) 0.8 mg/mL PEG-NH<sub>2</sub>, and (f) 0.16 mg/mL PEG-NH<sub>2</sub>, respectively. The concentration of E-1 is 30  $\mu$ M in spotting solution. The microarray is incubated with 500 U/mL EcoRI for 10 h.



**Fig. S3** Fluorescence images (EcoRI (A), BamHI (B), mixture of EcoRI and BamHI (C)) and corresponding relative fluorescence intensity under different endonucleases (EcoRI (a), BamHI (b), and the mixture of EcoRI (c) and BamHI (d)) and different incubation times, respectively. EcoRI-specific substrate in the left column of fluorescence image, and BamHI-specific substrate in the right column of fluorescence image, respectively. The concentration of endonuclease is 500 U/mL.



**Scheme S1** Chemical structures of actinomycin D (ACTD), doxorubicin hydrochloride (DOX), 5-fluorouracil (5-FU) and ethidium bromide (EB), respectively.