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Evagreen Dye Fluorescence Verification

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This protocol is to establish the relationship between Evagreen Dye fluorescent intensity and DNA concentration.

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Preparation

1 Take **30 µL** of 10µM ssDNA (T4 ligation primer) into a new eppendorf

2 Dilute 10µM ssDNA into 1µM :
Take **3 µL** of 10µM ssDNA and **27 µL** RNase-free water into a new eppendorf

3 Dilute 1µM ssDNA into 100nM :
Take **3 µL** of 1µM ssDNA and **27 µL** RNase-free water into a new eppendorf

4 Dilute 100nM ssDNA into 10nM :
Take **3 µL** of 100nM ssDNA and **27 µL** RNase-free water into a new eppendorf

5 Dilute 10nM ssDNA into 1nM :
Take **3 µL** of 10nM ssDNA and **27 µL** RNase-free water into a new eppendorf

6 Dilute 1nM ssDNA into 100pM :
Take **3 µL** of 1nM ssDNA and **27 µL** RNase-free water into a new eppendorf

7 Dilute 100pM ssDNA into 10pM :
Take **3 µL** of 100pM ssDNA and **27 µL** RNase-free water into a new eppendorf

8 Add **1.5 µL** of 20X evagree dye into each eppendorf.

Measure

9 Respectively load **20 µL** of 10µM,1µM, 100nM, 10nM, 1nM, 100pM and 10pM ssDNA into each well

10 Measure the fluorescence excitation and emission intensity

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