Product Name: **dsDNA Quantification Reagent (Green)**

Catalog Number: **RA20042**

Basic Information

Product Name	dsDNA Quantification Reagent (Green)
Size	0.1mL/1mL
Storage	4 °C, away from light
Shipping	Shipped with ice pack
Validity	12 months
Ex/Em	480/520 nm

Product Introduction

dsDNA Quantification Reagent (Green) is a product for fluorescent detection and quantification of dsDNA, and this method is highly sensitive. It is commonly used in molecular biology techniques such as the construction of cDNA libraries, purification of subcloned DNA fragments, and applications like DNA quantification, product amplification, and further detection of primers. The conventional method for detecting DNA content is to measure its absorbance at 260 nm. The main drawbacks of this method are that nucleotides, single-stranded nucleic acids, and proteins can significantly affect the signal, and it is also susceptible to interference from contaminants in the nucleic acid preparation process. It cannot distinguish between DNA and RNA, and it is not sensitive (A260 = 0.1 for a 5 μg/mL dsDNA solution). The dsDNA Quantification Reagent (Green) method is simple and convenient, and has become the standard for residual DNA detection in biological products.

dsDNA Quantification Reagent (Green) only emits fluorescence after binding to dsDNA, and the intensity of the fluorescence is proportional to the DNA concentration. dsDNA Quantification Reagent (Green) can detect dsDNA in the range of 25 pg/mL to 1000 ng/mL with a good linear relationship (R2>0.99).

Reagent preparation

dsDNA Quantification Reagent (Green) is stored in anhydrous DMSO (dimethyl sulfoxide) as a 1 mL concentrate. During the experiment, prepare 2×dsDNA Quantification Reagent working solution: dilute the concentrate with 1×TE (10 mM Tris-HCl, 1 mM EDTA, pH 7.5) at a ratio of 1:200. For a final volume of 200 μL, if you need to prepare enough working solution for 20 samples, add 10 μL dsDNA Quantification Reagent to 1.99 mL 1×TE; for a final volume of 2 mL, if you need to prepare enough working solution for 20 samples, add 100 μL dsDNA Quantification Reagent concentrate to 19.9 mL 1×TE. Because the reagent is easily adsorbed to glass surfaces, it should be prepared in a plastic container. dsDNA Quantification Reagent is easily degraded by light, so it should be stored away from light.

Solutions are best used within a few hours of preparation to ensure best results.

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Experimental procedures

1. Preparation of standard working solution:

Sigma calf thymus DNA powder 1 mg (Tris, NaCl and other concentrations have become a standard system), add 1 mL of double distilled water to prepare a 1 mg/mL standard solution.

2. Configuration of dye working solution:

Add 5 μL PicoGreen to 0.995 mL TE (Note: dilute PicoGreen 200 times with 1×TE, prepare before use, and protect from light).

3. Standard solution dilution:

（1）Dilution of stock solution: add 10 μL (1 mg/mL) of standard solution to 990 μL TE solution and dilute to 10 μg/mL; add 10 μL (10 μg/mL) of standard solution to 990 μL TE solution and dilute to 100 ng/mL.

（2）Serial dilution: add 800 μL (100 ng/mL) of the standard solution to 200 μL TE solution to obtain a concentration of 80 ng/mL. Add 500 μL (80 ng/mL) of the standard solution to 500 μL TE solution to dilute to 40 ng/mL. Perform serial dilutions to obtain 20 ng/mL, 10 ng/mL, 5.0 ng/mL, and 2.5 ng/mL.

4. Preparation of standard curve:

Take 100 μL of each gradient standard solution and dye working solution after dilution, mix them and place them at room temperature away from light for 5 min. Use FB-15 portable fluorescence instrument to detect the fluorescence value of the sample: add the mixed solution to the micro-colorimetric dish, be careful not to introduce bubbles into the sample, and tap the outside of the micro-detection dish to disperse the bubbles. Use 1×TE buffer as blank control to measure the fluorescence value of the sample and blank control; or directly use 96-well plate for fluorescence detection, with an excitation wavelength of 480 nm and an emission wavelength of 520 nm, and use the concentration of the standard solution (ng/mL) to make a linear regression of the corresponding fluorescence intensity to prepare the standard curve.

5. Measure the fluorescence value of the sample to be tested. Calculate the concentration of the sample to be tested based on the prepared DNA concentration standard curve.

Note: This reagent is for scientific research use only!