

Version: 1.0

PicoGreen dsDNA Quantitation Reagent

Cat. No. / Spec.: P6021 / 1 mL

Description

PicoGreen is a highly sensitive fluorescence detection reagent specifically designed for the quantitation analysis of double-stranded DNA (dsDNA). It is widely used in molecular biology research and bioproduct testing due to its excellent sensitivity, specificity, and convenience. PicoGreen only emits fluorescence upon binding to dsDNA, and the fluorescence intensity is proportional to the DNA concentration, making it an ideal choice for quantifying dsDNA.

Components

Component	P6021
PicoGreen dsDNA Quantitation Reagent	1 mL

Storage

Store at 4°C away from light.

Parameters

Excitation/Emission Wavelength: Ex/Em = 480/520 nm (when bound to dsDNA)

Application

Quantitation of PCR products, genomic DNA quantitation, determination of dsDNA in complex mixtures, viral DNA quantitation, etc.

Features

High Sensitivity: Can detect dsDNA in the range of 25 pg/mL to 1000 ng/mL with a good linear relationship (R²>0.99).

Easy Operation: The quantitation method is simple and quick, easy to perform in the laboratory.

Strong Anti-interference Ability: Basically unaffected by single-stranded DNA (ssDNA) and RNA, can tolerate certain concentrations of salts, urea, ethanol, chloroform, detergents, proteins, and other interferences.

Wide Linear Range: The fluorescence response has a broad linear range, covering multiple orders of magnitude.

Protocol

1. Reagent Preparation

PicoGreen dsDNA Quantitation Reagent is supplied as a 1 mL concentrate in anhydrous DMSO (dimethyl sulfoxide).

For the experiment, prepare a 2X PicoGreen working solution by diluting the concentrate with 1X TE (10 mM Tris-HCl, 1 mM EDTA, pH 7.5) at a ratio of 1:200. For a final volume of 200 μ L detection system, if you need to prepare enough working solution for 20 sample measurements, add 10 μ L PicoGreen to 1.99 mL 1X TE; for a final volume of 2 mL detection system, if you need to prepare enough working solution for 20 sample measurements, add 100 μ L PicoGreen concentrate to 19.9 mL 1X TE.

Since the reagent can adsorb to glass surfaces, prepare it in plastic containers.

PicoGreen is photosensitive and degrades upon exposure to light, so it should be protected from light.

It is best to use the solution within a few hours after preparation to ensure the best results.

2. Experimental Method

2.1 Standard Working Solution Preparation

Prepare a standard solution by dissolving 1 mg of calf thymus DNA powder (with a standard system of Tris, NaCl, etc.) in 1 mL of double-distilled water to make a 1 mg/mL solution.

2.2 Dye Working Solution Preparation

Add 5 μ L PicoGreen to 0.995 mL TE (Note: Dilute PicoGreen 200 times with 1X TE, prepare as needed, and protect from light).

2.3 Standard Solution Dilution

- (1) Master Dilution: Take 10 μ L (1 mg/mL) of the standard solution and add it to 990 μ L of TE solution to dilute to 10 μ g/mL, then take 10 μ L (10 μ g/mL) of the standard solution and add it to 990 μ L of TE solution to dilute to 100 ng/mL.
- (2) Serial Dilution: Take 800 μ L (100 ng/mL) of the standard solution and add it to 200 μ L of TE solution for a concentration of 80 ng/mL, then take 500 μ L (80 ng/mL) of the standard solution and add it to 500 μ L of TE solution to dilute to 40 ng/mL; continue with serial dilutions to prepare 20 ng/mL, 10 ng/mL, 5.0 ng/mL, 2.5 ng/mL.

2.4 Standard Curve Preparation



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After serial dilution, mix 100 µL of each gradient standard solution with 100 µL of dye working solution, let it stand in the dark at room temperature for 5 minutes. Use a fluorometer to detect the fluorescence values of the samples: add the mixed solution to a microcuvette, being careful not to introduce bubbles into the sample, and tap the outside of the microcuvette to disperse any bubbles. Use 1X TE buffer as a blank control, and measure the fluorescence values of the samples and the blank control; or directly use a 96-well plate for fluorescence detection, with an excitation wavelength of 480 nm and an emission wavelength of 520 nm, and prepare a standard curve using the fluorescence intensity corresponding to the concentration of the standard solution (ng/mL).

2.5 Measurement of Fluorescence Values for Samples

Calculate the concentration of the samples to be measured based on the prepared DNA concentration standard curve.

Precautions

- 1. Before use, please centrifuge the product briefly to the bottom of the tube, then proceed with the subsequent experiments.
- 2. Fluorescent dyes are subject to quenching; please try to protect from light to slow down fluorescence quenching.
- 3. It is best to prepare and use the PicoGreen working solution immediately to ensure the best results.
- 4. For your safety and health, please wear a lab coat and disposable gloves when operating.

Product Use Limitations

This product is sold exclusively for research purposes and in vitro use. Neither the product, nor any individual components, was tested for use in diagnostic applications or for drug development, nor is it suitable for administration to humans or animals.