

PicoGreen Dye (200× in DMSO)

Cat. No.: P1522560 | Pack size: 100 µL; 1 mL | Storage: 2-8°C; protected from light

Product Information

Attribute	Value
Pack Size(s)	100 µL; 1 mL
CAS Number	P1522560
Molecular Weight	588.26
Specifications & Purity	BioReagent, Biological Stain, for fluorescence analysis, for microscopy, ≥95%
Stability And Storage	Store at 2-8°C long term (24 months). Store in the dark.
Storage Conditions	Store at 2-8°C, Protected from light
Shipped In	Wet ice. This product requires cold chain shipping. Ground and other economy services are not available.
Grade	Biological Stain, BioReagent, for fluorescence analysis, for microscopy

Product Description

PicoGreen is a saturating fluorescent dye that specifically binds to double-stranded DNA (dsDNA). It has an excitation wavelength of 480 nm and an emission wavelength of 520 nm, and is compatible with most fluorometers and fluorescent microplate readers. PicoGreen only fluoresces upon binding to dsDNA, and is not affected by nucleotides or single-stranded nucleic acids. The resulting fluorescence intensity is proportional to the dsDNA concentration in a sequence-independent manner, making it highly suitable for quantitative detection of dsDNA. It can detect dsDNA in the concentration range of 25 pg/ml to 1 µg/ml.

Due to its high sensitivity, strong specificity, good tolerance, and no requirement for DNA purification, PicoGreen has been listed as one of the standard methods for residual DNA detection in biological products since the 2010 edition of the Chinese Pharmacopoeia. According to the pharmacopoeia, the linearity is optimal ($R^2 > 0.99$) when the dsDNA concentration ranges from 1.25 ng/ml to 80 ng/ml.

Protocol

1. Preparation of Dye Working Solution

PicoGreen stock solution is supplied as a concentrate in anhydrous DMSO (Dimethyl Sulfoxide). For experiments, the stock solution is generally diluted 1:100 with 1× TE buffer to prepare a 2× dye working solution, which is then mixed with samples at a 1:1 ratio to achieve a final 1× PicoGreen concentration. As this reagent readily adsorbs to glass surfaces, preparation should be performed in plastic containers with protection from light as much as possible. The dye working solution is best used within several hours of preparation for optimal results.

2. Experimental Procedure

① Preparation of Standard Solution

Weigh 1 mg of calf thymus DNA powder (with standardized concentrations of Tris, NaCl, etc.), add 1 ml of double-distilled water to prepare a 1 mg/ml standard solution.

② Preparation of Dye Working Solution

Take 10 µl of PicoGreen stock solution and add 990 µl of 1× TE buffer to prepare a 2× dye working solution.

③ Preparation of Standard Working Solutions

a. Stock dilution: Take 10 µl of 1 mg/ml standard solution and add to 990 µl of 1× TE buffer to dilute to a 10 µg/ml standard working solution. Then take 10 µl of the 10 µg/ml standard working solution and add to 990 µl of 1× TE buffer to dilute to a 100 ng/ml standard working solution.

b. Serial dilution: Take 800 µl of 100 ng/ml standard working solution and add to 200 µl of 1× TE buffer to obtain 80 ng/ml. Then take 500 µl of 80 ng/ml standard working solution and add to 500 µl of 1× TE buffer to dilute to 40 ng/ml. Continue serial dilutions to prepare standard working solutions of 20 ng/ml, 10 ng/ml, 5.0 ng/ml, 2.5 ng/ml, and 1.25 ng/ml.

④ Standard Curve Construction

Mix the serially diluted standard working solutions (1.25 ng/ml to 80 ng/ml) with the 2× dye working solution at a 1:1 volume ratio (adjust volume according to fluorometer and container requirements). Incubate at room temperature for 5 min protected from light. Measure the fluorescence using a fluorometer: add the mixed solution to a microcuvette, avoid introducing air bubbles, and gently tap the outside of the cuvette to remove bubbles. Use 1× TE buffer as blank control, set excitation wavelength to 480 nm and emission wavelength to 520 nm, and measure fluorescence of samples and blank at a constant temperature. Alternatively, detection can be performed in a 96-well microplate using a fluorescent microplate reader. After detection, subtract blank fluorescence from each sample reading, perform linear regression against standard solution concentrations (ng/ml), and construct a standard curve.

⑤ Determination of Sample Concentration

Mix the test sample solution with the 2× dye working solution at a 1:1 volume ratio. Measure the fluorescence intensity under the same temperature and conditions as in step ④ (dilute samples with TE buffer into the linear range of PicoGreen if necessary). Calculate the sample concentration using the linear regression equation of the standard curve.

Precautions

1. All fluorescent dyes are subject to quenching; minimize exposure to light to reduce fluorescence quenching.
2. PicoGreen working solution is best prepared fresh for optimal performance.
3. PicoGreen fluorescence intensity is highly temperature-dependent; ensure standard curve construction and sample measurement are performed at the same constant temperature.
4. For your safety and health, wear a lab coat and disposable gloves during operation.

Purity

≥95%

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