GDS Green I, 10,000X in DMSO

(Fluorescent Quantitative PCR Grade Dye)

Cat. No. / SPEC.: P6011 / 500 μL

Description

GDS Green I is a fluorescent dye with Green emission that binds to dsDNA double helix groove region and has the same efficiency as Sybr Green I. The fluorescence signal of GDS Green I is enhanced by 800-1000 times after binding with dsDNA, and it is a commonly used qPCR fluorescent dye. The dye is economical, easy to use, has the advantages of high sensitivity and high signal-to-noise ratio, and can be applied to gene expression difference analysis, gene chip and so on.

Components

Component	P6011
GDS Green I, 10,000X in DMSO	500 μL

Storage

Store at 4°C away from light. Repeated freezing and thawing should be avoided when used, and it is recommended to pack into small tubes for freezing storage.

Protocol

1. Preparation of reaction solution

Dilute 10,000X dye with DMSO or sterile ultra-pure water, such as diluting 500 times to get 20X dye, for subsequent experiments.

Refer to the following table to formulate the reaction system:

Component	Amount
10X PCR Buffer（Mg^{2+} free）	5 μL
50 mM $MgCl_2$ [1]	2.5 μL
2 mM dNTP	5 μL
20X GDS Green I	2.5 μL
Taq DNA polymerase	1-5 U
F, R Primers	0.1-0.5 μM each
Template DNA[2]	variable
ddH_2O	To 50 μL

[1] Increasing the concentration of Mg2+ can reduce the inhibitory effect of GDS Green I on PCR reaction. It is suggested that the concentration of Mg2+ in GDS Green I fluorescence PCR reaction should be 0.5~3 mM higher than that in ordinary PCR reaction without GDS Green I.

[2] The amount of DNA template added is usually less than 100 ng. Because different types of DNA templates contain different copy numbers of target genes, gradient dilution can be carried out if necessary to determine the appropriate amount of DNA template addition. The addition of cDNA as a template should not exceed 10% of the total volume of PCR reaction solution.

2. Perform qPCR

Please refer to the following example to set the program according to the experimental needs:

Stage	Temperature	Time	Cycle
Initial denaturation	94°C	3 min	1
Denaturation	94°C	15 sec	40~45
Annealing	55~65°C[1]	15 sec	
Extension[2]	72°C	20 sec[1]	
Dissociation/Melting curve analysis（optional）[4]			

[1] The optimum annealing temperature needs to be explored. The annealing temperature is generally set to the TM-5°C of the primer used, and if it is lower than 55°C, the Tm value is used as the annealing temperature, which is generally not lower than 55°C.

[2] Set up signal acquisition in this step.

[3] Please consider the instrument type when setting the extension time, some instruments need to be set above 30 sec.

[4] Different instruments have different melting curve acquisition procedures, generally according to the default melting curve acquisition procedures of the instrument.

3. Analyze the results

The amplification curve was observed. Adjust baseline, calculate Ct value; The specificity of melting curve was observed. Take relative or absolute quantification.

Notes

① The concentration of GDS Green I is the key factor to ensure the success of fluorescence quantitative PCR experiment. Too low a dye concentration will reduce the change in the fluorescence signal, resulting in a low copy of the sample may not be detectable, and at high concentrations will inhibit PCR reaction. Therefore, in general, when using GDS Green dyes, the concentration should be optimized according to the actual situation, and the final concentration of the reaction is between 1X and 0.2X.

② GDS Green I fluorescent dyes may fade under light over time, resulting in decreased sensitivity, so avoid bright light exposure during storage and use.

③ GDS Green I fluorescent dyes should be stored away from light, and the stock solution should be stored at -20°C; It is recommended to be divided into small tubes and frozen, which can be stored at 4°C for a short time.

④ For your health, please wear a lab coat and disposable gloves.

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. Please refer to the MSDS, available upon request.