MANUAL

RayBio® DNA Binding Microplate Solution

Catalog #: TF-BIND

ISO 13485:2016

Introduction

The immobilization of both single-stranded and double-stranded DNA onto microplate surfaces is a crucial step in many molecular biology and biochemistry assays, including those focused on DNA-protein interactions and DNA hybridization. Traditionally, this process has required expensive oligo modifications to facilitate DNA binding to plates, such as biotinylation for attachment to streptavidin-coated plates, or the use of activated ELISA plates for covalent binding of DNA. The RayBio® DNA Binding Microplate Solution offers a highly efficient and cost-effective alternative for binding unmodified DNA fragments as short as 50 base pairs to the surfaces of polystyrene 96-well microplates, as well as other polystyrene plastics.

Storage

Buffer should be stored at room temperature for a maximum of 12 months after delivery. Storage below room temperature can cause precipitate formation. If precipitate is observed heat solution to 37ºC with gentle mixing until precipitate is dissolved.

Directions for Use

1. Prepare coating solution by diluting DNA to desired concentration in DNA Binding Microplate Solution; 50 nM (~2 ug/mL) is sufficient for most applications.

   ***Note**: Take care not to dilute DNA Binding Microplate Solution by more than 20% when preparing coating buffer (e.g., 1 part DNA to 4 parts DNA Binding Microplate Solution) as this will result in loss of coating efficiency.*

2. Immediately after mixing, add 100 µL of the DNA solution to each well of the polystyrene 96-well microplate that the DNA is to be bound to. Binding can be observed in as little as 3 hours at 37ºC. However, for best results incubate coated plates overnight at 4ºC with gentle rocking.

3. After incubation remove liquid from the wells and wash with an appropriate buffer such as PBS. Detergents such as Tween can be included in washing, if necessary, although SDS should be avoided.

   ***Note**: It is recommended that polystyrene plastics be used with this product. If using polypropylene substrates, avoid using detergents as this can disrupt the DNA-plastic interaction.*

4. If preparing plate for use in an immunoassay, it is crucial to block the plate with an appropriate coating buffer before use (e.g., 5% bovine serum albumin in PBS).

Example Data

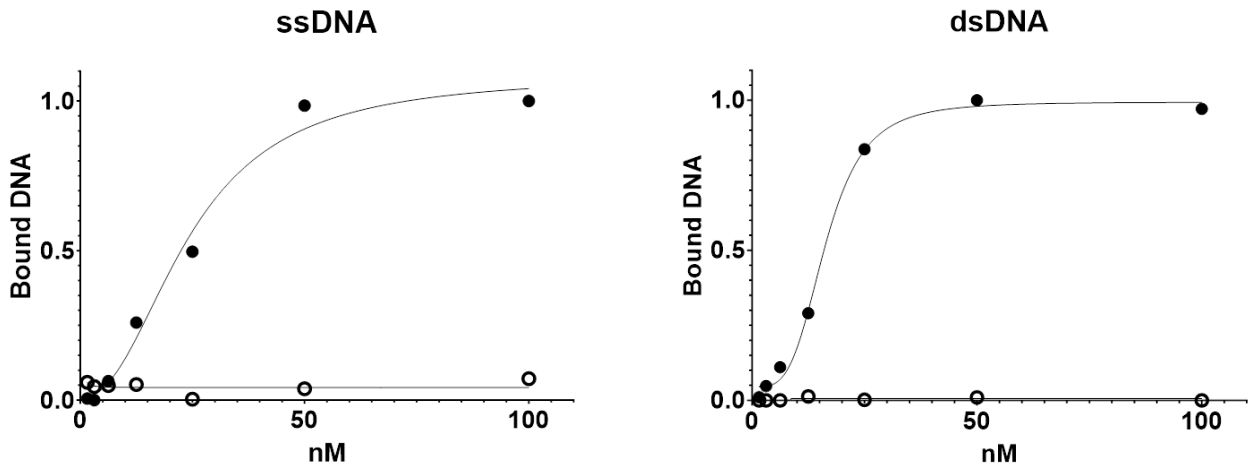

Figure 1. Measurement of bound single stranded (left) and double stranded (right) DNA fragment (70bp length) coated to a polystyrene 96-well microplate plate at increasing concentrations using RayBio® DNA Binding Microplate Solution (solid circles, ●) and Tris-EDTA buffer (hollow circles, ○).

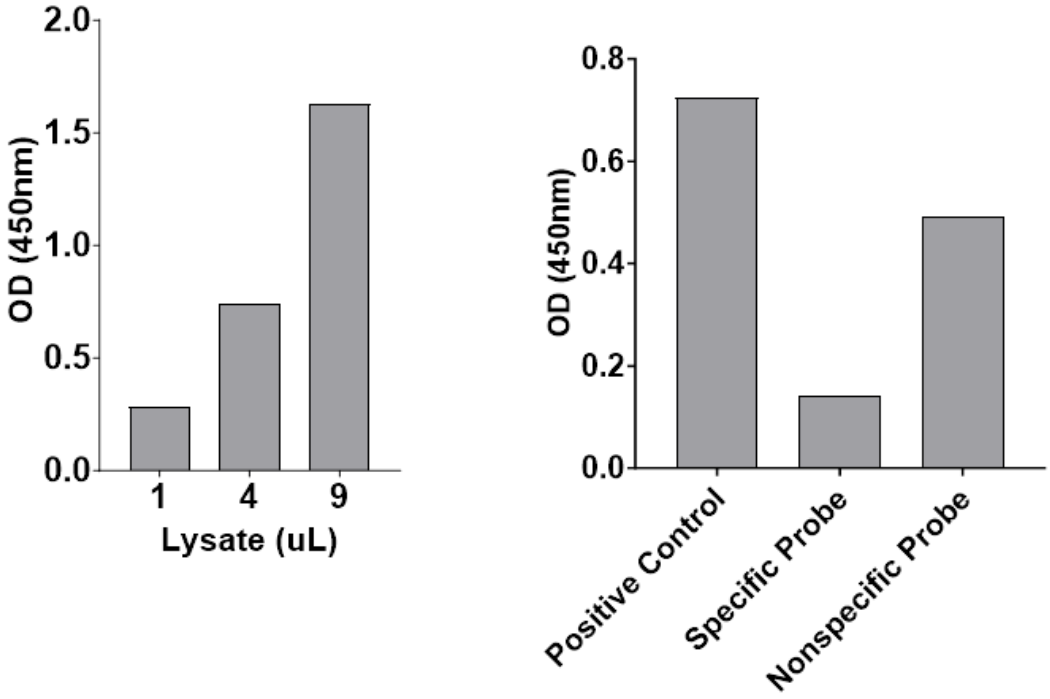

Figure 2. Representative data obtained from the RayBio® Transcription Factor Activity Assay, where the plate was coated using the RayBio® DNA Binding Microplate Solution. (Left) Increasing volumes of nuclear extract produce a corresponding increase in optical density (OD), indicative of greater transcription factor (TF) binding. (Right) A dsDNA probe containing the consensus binding sequence (Specific Probe) competes with coated DNA for binding to the TF (80.5% inhibition). Conversely, a dsDNA with an unrelated sequence (Nonspecific Probe) shows markedly less inhibition (31.9%).