Jun 29, 2022

Concentrated DNA Loading dye stock: User protocol

In 1 collection

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1 *Works for me* Share

This protocol is published without a DOI.

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ABSTRACT

Beneficial Bio 6X DNA Loading Dye is a premixed loading buffer used to prepare samples for
loading onto agarose and polyacrylamide gels. It contains bromophenol blue and xylene
cyanol dyes for visual tracking of DNA during electrophoresis. The trehalose ensures that
DNA forms a layer at the bottom of the well and facilitates sample loading. The EDTA chelates metal ions and stops metal-dependent enzymatic reactions. The solution contains
SDS which results in sharper bands by removing restriction enzymes from DNA.

Features:

- Double tracking: Xylene cyanol, Bromophenol blue
- No interaction with bands during gel exposure to UV or Blue light
- Inhibits metal-dependent nucleases (EDTA)

Applications:

Preparation of DNA ladders, markers, and samples for loading on agarose or polyacrylamide
DNA electrophoresis.

PROTOCOL CITATION

Stephane Fadanka, Nadine Mowoh 2022. Concentrated DNA Loading dye stock: User protocol. **protocols.io** https://protocols.io/view/concentrated-dna-loading-dye-stock-user-protocol-cbtvsnn6

COLLECTIONS

Beneficial Bio Products: User Protocol

CREATED

Jun 22, 2022

LAST MODIFIED

Jun 29, 2022

PROTOCOL INTEGER ID

65109

PARENT PROTOCOLS

Part of collection

Beneficial Bio Products: User Protocol

GUIDELINES

Follow good laboratory practices when carrying out experiment although all reagents used are generally safe.

MATERIALS TEXT

- Concentrated DNA loading dye stock
- 20% Trehalose
- DNA samples
- Micro Pipette and tips
- Agarose gel electrophoresis system
- Gel visualization system

BEFORE STARTING

Ensure all materials, equipment and chemicals to be used for this experiment are all in place before starting.

User Protocol

1

Our DNA loading dye stock is usually prepared in a concentrated form that is stored in the fridge and diluted to 6x working stock before it is mixed with DNA for loading.

Preparing 1ml of 6x working stock:

1. Vortex or shake the concentrated stock tube for 10 seconds prior to use.
2. Dilute 1 part of concentrated DNA Loading Dye stock with 9 parts of 20% Trehalose (pipette **100 µL** from the concentrated dye stock and add **900 µL** of **20 % (v/v)** Trehalose to make a 1ml 6x working stock).
3. Cork the tube firmly and mix by inverting the tube several times.
4. Store the working solution at room temperature or **4 °C** for up to 12months, for longer periods, store at **-20 °C** .
5. To use, dilute the nucleic acid solutions 1:5 to give a 1x solution e.g. add **1 µL** 6x DNA loading dye for every **5 µL** DNA.
6. Load the sample into a precast agarose gel well, run to finish and visualize the gel with the help of a UV transilluminator or blue gel system.

Notes and troubleshooting

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- BenBio 6X DNA Loading Dye enables easy tracking and sample migration and follow up during the run without creating background noise during visualization.
- If you notice any interference of the tracking dye with the DNA bands, check the working concentration and try further dilutions to find the one that works for you.

Also the storage temperature is important as the trehalose in the dye might loose its efficacy over time as noticed if not stored properly, hence DNA loading dye may not be able to cause samples to sink into the gel wells (may cause the samples to leak or just spread out instead).