GUS Solution

Greg Reeves

Abstract

The **GUS reporter system** (*GUS*: β-glucuronidase) is a reporter gene system, particularly useful in plant molecular biology[1] and microbiology.[2]Several kinds of GUS reporter gene assay are available, depending on the substrate used. The term **GUS staining** refers to the most common of these, a histochemical technique. (Source: https://en.wikipedia.org/wiki/GUS_reporter_system)

Citation: Greg Reeves GUS Solution. **protocols.io**
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Guidelines

- This GUS staining solution recipe is for a volume of 500mL.
- The solution can be partitioned into smaller tubes and stored long term at -20°C in the dark, as the GUS staining solution is light-sensitive.

Protocol

Step 1.

0.5M EDTA (pH 8.0)

AMOUNT
1 ml Additional info:

NOTES
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EDTA will need to be pH adjusted with NaOH (Sodium hydroxide) until it reaches a pH of 8.0 in order to dissolve fully in deionised water. *To make a 0.5M solution of EDTA, add 93.3g into 500mL of deionised water. Adjust pH to 8.0 with NaOH.*

Step 2.

0.1M Potassium ferrocyanide

AMOUNT

10 ml Additional info:

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Both Potassium ferrocyanide (II) and Potassium ferricyanide (III) solutions should be made fresh each time a GUS staining solution is made. A higher concentration of Potassium ferro/ferricyanide (usually between 0.5mM and 10mM) constricts the movement of GUS between cells.

To make a 0.1M solution of Potassium ferrocyanide (II) add 0.42g into 10mL of deionised water.

To make a 0.1M solution of Potassium ferricyanide (III) add 0.33g into 10mL of deionised water.

Step 3.

0.1M Potassium ferricyanide

AMOUNT
10 ml Additional info:

Step 4.

1M Sodium phosphate buffer (pH 7.0)

AMOUNT
50 ml Additional info:

NOTES
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The sodium phosphate buffer is formed from two sodium compounds (NaH_2PO_4, MW=137.99g/mol; and Na_2HPO_4, MW=141.96g/mol). These two compounds should be made up as two separate stock solutions and added together when making the sodium phosphate buffer for the GUS solution in this manner:

--To make a 1M solution of NaH_2PO_4, add 13.8g into 100mL of deionised water.

--To make a 1M solution of Na_2HPO_4, add 28.4g into 200mL of deionised water.

-To make 50mL of a 1M solution of sodium phosphate buffer, add 19.5mL of the 1M NaH_2PO_4 solution and 30.5mL of the 1M Na_2HPO_4 solution together. The pH of this 50mL solution should be at 7.0, so adjusting the pH should not be necessary.

Step 5.

X-Gluc

AMOUNT
500 mg Additional info:

Step 6.

dH_20 up to 500mL

NOTES

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After completion filter sterilze with 0.22um membrane. Aliquots can be frozen at -20oC prior to use.

Warnings

Note: The staining solution contains toxic chemicals.

https://en.wikipedia.org/wiki/Potassium_ferrocyanide

https://en.wikipedia.org/wiki/Potassium_ferricyanide