Checking DNA Concentration with Agarose Gel

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Abstract

Citation: Matthew Sullivan Checking DNA Concentration with Agarose Gel. **protocols.io**
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Guidelines

This protocol comes from a group of other protocols. This protocol is (4) of (4):
1. 'Large Volume Marine Cyanophage Phage Protocols'
2. 'DNA Extraction Protocol'
3. 'DNA Precipitation Protocol'
4. 'Checking DNA Concentration with Agarose Gel'

Needed:

- 0.7% Agarose
- (Agarose)
- (TAE Buffer)
- (HINDIII (lambda) DNA)
- Nanodrop

Materials

- SYBR Gold Nucleic Acid Gel Stain S-11494 by Contributed by users
- Lambda DNA/HindIII Marker SM0101 by Contributed by users

Protocol

Step 1.
Prepare 0.7% agarose

PROTOCOL

. 0.7% Agarose

CONTACT: VERVE Team
Preparation of 0.7% Agarose
Step 1.1.

Mix 0.35 g agarose in 50 ml TAE running buffer

AMOUNT

0 g Additional info:

Step 2.

Post-stain with SYBR Gold

Step 3.

While the gel is cooling, prepare samples:
1µl of 10x loading buffer per 10µl of sample

Step 4.

Prepare the appropriate dilutions of the appropriate marker (commonly 10ng of HINDIII [lambda] DNA)

AMOUNT

10 ng Additional info:

REAGENTS

Lambda DNA/HindIII Marker SM0101 by Contributed by users

Step 5.

Add running buffer to cover gel

Step 6.

Gently rock the comb before pulling it out

Step 7.

Run at 50V about 2-4 hours to get clean band separations

DURATION

03:00:00

NOTES

VERVE Team 17 Jun 2015

Note: (if in hurry, watch for bromophenol blue marker to run about 3-4 cm)

Step 8.

Post-stain the gel in SYBR Gold 'bath' (20ul SYBR: 200ml TAE for 30 minutes)

AMOUNT

20 µl Additional info:

DURATION

00:30:00

Step 9.

Compare gel quantification to that obtained using Nanodrop to get a feeling for how 'clean' the DNA prep is

NOTES

VERVE Team 21 Jan 2016

2µl volume needed; only good down to the ~4ng/µl concentrations; A260/A280 estimates are very approximate in that range.