02_04_SYBR-Gold-staining

FREITAG, 14.5.2021

Goal-Setting

- Post-electrophoresis staining of a gel to visualize DNA bands

Terms / abbreviations

- PAA = Polyacrylamide
- TAE = Tris-acetat-EDTA
- TBE = Tris-borate-EDTA

Risk areas

Hazard symbols

Required materials and / or information

- Chemicals:
  - 1x TBE for PAA gels
  - 1x TAE for agarose gels
    Update 26.05.21: TBE-Buffer (1x)
  - Gels
  - SYBR Gold Nucleic Acid Gel Stain (10,000x concentrate in DMSO), ThermoFischer
- Material:
  - Aluminium foil
  - Green spatula
  - Schottflask 100 mL
  - Staining container

Templates, devices, software

- Platform rocker, Heidolph DUOMAX 1030

Preliminary work

- 00_02_10x-TBE-buffer-recipe
- 00_03_1x-TAE-buffer-recipe
- 02_01_PAA-gel-preparation (native)
- 02_02_Performing-PAA-gel-electrophoresis

or

- 01_01_Agarose-gel-preparation
- 01_03_Performing-agarose-gel-electrophoresis

Operation

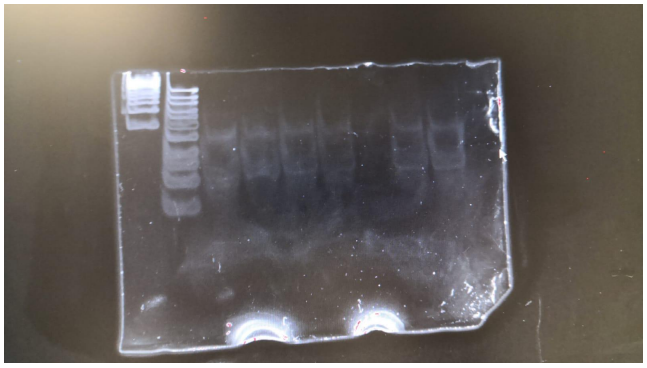
Preparation of SYBR Gold Solution

- Mix 5 µL SYBR Gold with 50 mL buffer to create new stock solution for staining
    - Use the same buffer (TBE or TAE) which was used for the electrophoresis
- The stock may be used 3-4 times before replacement, please mark the number of uses on the Schottflask!
  Update 08.06.21: Always use fresh staining solution!

Staining Procedure

1. Pour all 50 mL of the staining solution into the staining container
2. Take the gels out of the glas slides by carefully lifting the top slide with the green spatula
    a. For later identification of proper orientation of the gel, cut a small peace off the gel at the bottom right corner (see image)

Example: Gel with a cut corner



1. Let the gel carefully slide into the staining container with the staining solution
2. Put on the aluminium cover and place the container on the shaker
3. Let incubate for 20 min
4. When finished, pour the staining solution back into the Schottflask, mark the number of uses, and put it back into the fridge
   Update 08.06.21: discard staining solution

Disposal

- Black canister on ground next to trash cans in gel electrophoresis room

Troubleshooting

- Make sure the stain is not too old
- Always cover the stain from light with aluminium foil

Follow-up work

- 04_07_Documentation-with-BioRad-Photographer