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



Required materials and / or information

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

- 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)
- 3. Let the gel carefully slide into the staining container with the staining solution
- 4. Put on the aluminium cover and place the container on the shaker
- 5. Let incubate for 20 min
- 6. 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