00_02_10x-TBE-buffer-recipe

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Goal-Setting

Manufacture of 10x Tris-borate-EDTA-buffer

Terms / abbreviations

- EDTA = Ethylenediaminetetraacetic acid
- HCl = Hydrochloric acid
- NaOH = Sodium hydroxide
- PAA = Polyacrylamide
- TBE = Tris-borate-EDTA
- TRIS = Tris(hydroxymethyl)aminomethane

Risk areas



Required materials and / or information

- Chemicals:
 - o Boric acid, Merck
 - o EDTA Lot, Sigma-Aldrich
 - o MilliQ water, Sartorius arium pro VF
 - o Tris Base, AppliChem
- Materials:
 - o Beaker/Schottflask
 - o Stirring fish
- All chemicals are solid substances
- Use TBE buffer only two times for gel electrophoresis

Templates, devices, software

- Analysis balance, Kern ABJ 220-4NM
- pH meter, Knick Digital pH-Meter 646

Preliminary work

None

Operation

- pH meter (ready to use in the chemical room)
 - o If it is used for the first time, ask an experienced person around how to use it
 - o Regularly calibrate the pH meter according to manufacturer
 - o User guide:

- pH meter should measure about 6.5 when the solution is stored (small Falcon Tube)
- Take it out and unlock it (small switch at the top)
- Wash it carefully with some MilliQ water
- Hold into the solution and measure
- Wash again after use, turn off the switch and put it back into the small Falcon Tube

The following recipe is standardized to 1 L

 Weigh the following components into a beaker or schottflask --> Tip: use at least a 1500 mL beaker (easier usage/measurement)

Tris Base: 121,14 g/L
Boric acid: 52,54 g/L
EDTA: 2,92 g/L

- 2. Fill up to 1 L with MilliQ water
- 3. Mix by stirring/shaking/inverting
- 4. Optimal pH = 8.3
 - a. If pH is too low, adjust with NaOH
 - b. If the pH is too high, do not adjust it with HCl, as this would cause errors with the PAA gel, because the Cl⁻ ions will migrate first

Disposal

- Observe all federal, state and local environmental regulations
- Here: Can be discarded in sink with a lot of water

Troubleshooting

None

Follow-up work

- If a chemical runs out or is empty, please fill out the order list on the fridge next to the window
- 01_01_Agarose-gel-preparation
- 01_03_Performing-agarose-gel-electrophoresis
- 02_01_PAA-gel-preparation (native)
- D2_01_PAA-gel-preparation (denaturating)