

Sulfide and GSSH solution

GSSG solution. Tris-HCl-buffer (0,1M) mixed with GSSG:

1. Dissolve Tris Base (1.21 g) in deionised water (100 mL) and stir with a magnetic stirrer.
2. Adjust the pH-value with HCl until the pH reaches pH 7.4.
3. Dissolve 0.392 g GSSG in Tris-HCl buffer (100 mL) and stir with the magnetic stirrer.
 - The solution is now ready to use with sodium sulfide in step 4.

Sulfide solution:

All work is done in a fume hood.

1. Prepare the work space in the fume hood with magnetic stirrer, weight, containers and all the necessary equipment.
2. Make the master stock solution by dissolving Na_2S (0.05 g) in deionised water (0.5 L) and close the container as tightly as possible. Stir with the magnetic stirrer until the salt is dissolved.
 - When the next step is done, loosen the cap to avoid an explosion due to H_2S -gas production.
 - The concentration in this solution is now $1.28 \cdot 10^{-3} \text{ mol/L}$
3. Pipette master stock solution (5 mL) to a falcon tube (50 mL), and close the cap as tightly as possible.
4. Pipette GSSG solution (5 mL) to the falcon tube. Close tightly and mix by gently shaking the tube.
5. Let the falcon tube with Na_2S and GSSG react on a heat block in the fume hood for 15 minutes. GSSH will now be produced. The solution is diluted further in step 6.
6. Dilute the GSSH-solution the way it is described in Table 1. Use 14 mL tubes. When performed the dilutions are ready to be applied to the bacteria solution.

Table 1: Dilution of GSSH-solution. Enough for 4 parallels of each concentration.

Concentration [$\mu\text{g/L}$]	2000	500	100	25
Volume GSSH-solution [μL]	2198	550	110	27
Volume deionised water [μL]	1802	3450	3890	3973