

Trypan Blue for Viability
(Greg A. Perry, Ph.D.)

Equipment:

Balance
100 ml glass bottle with lid
0.45 μ m or 0.2 μ m syringe top filter
10cc or 60cc syringe

Reagents:

Stock Solution

0.2 gm Trypan Blue (Sigma Catalog # F-7378)
99.8 ml Distilled Water
0.2% Sodium Azide

Working Solution

0.43 gm Sodium Chloride (NaCl)
9.58 ml Distilled Water
2 ml Stock Solution

Methods:

Stock Solution (10x)

- 1) Dissolve the trypan blue in the water.
- 2) Filter with 0.45 μ m or 0.2 μ m filter to remove particles.
- 3) Add 0.2% sodium azide to inhibit bacterial growth.

Working Solution (2x)

- 1) Make a 5x saline solution by dissolving the sodium chloride in 9.58 ml distilled water.
- 2) Add 2ml Trypan Blue Stock Solution to 0.5ml of the 5x Saline Solution and mix well.

To do a viability count: Add 1 part trypan blue working solution to 1 part cell suspension (at 2-5x10⁶ cells/ml), mix and count using a hemacytometer.