Procedure for preparation

Stock solution
10% (wt/vol) Triton X-100 solution
i) Dissolve 10 g of Triton X-100 (e.g., Nacalai Tesque, Code 355-01 or similar grade) in 80 ml of Milli-Q water by stirring.
ii) Add Milli-Q water to make 100 ml and stir until well mixed.
iii) Store at 4 ºC.

Preparation of Scale solutions

1. ScaleA2 solution (1 liter)
4 M urea
0.1% (wt/vol) Triton X-100
10% (wt/wt) glycerol

i) Dissolve 240.24 g of urea crystals (e.g., Wako Chemical, Code 217-615 or similar grade) in 800 ml of Milli-Q water by stirring.
ii) Add 10 ml of 10% (wt/vol) Triton X-100 solution.
iii) Add 100 g of glycerol (e.g., Sigma, Code 191612 or similar grade).
iv) Mix well by stirring.
v) Add Milli-Q water to make 1,000 ml and stir until well mixed.
vi) Store at room temperature.

2. ScaleB4 solution (1 liter)
8 M urea
0.1% (wt/vol) Triton X-100

i) Dissolve 480.48 g of urea crystals in 800 ml of Milli-Q water by stirring.
ii) Add 10 ml of 10% (wt/vol) Triton X-100 solution.
iii) Add Milli-Q water to make 1,000 ml and stir until well mixed.
iv) Store at room temperature.

Remarks
a. Allow each solution to stand after preparation for at least 1 day prior to use.
b. Sterilization is not required.
c. Addition of preservative is not required.