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 °C.

Preparation of Scale solutions

1. <u>Sca*l*eA2 solution (1 liter)</u>

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. <u>ScaleB4 solution (1 liter)</u>

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.