

## Supplementary Method 9 Preparation of Binding Buffer (BB)

### Reagents

- Guanidine Hydrochloride (GuHCl)
- MilliQ water
- Isopropanol
- 3M sodium acetate, pH 5.2
- Tween 20

### Equipment

- Weighing scales
- 2L glass beaker
- 3L glass beaker
- 5 L glass beakers
- 2 L graduated cylinder
- Heater/Stirrer

### Procedure

**NOTE: Masks should be worn when making this buffer if there is a possibility of asymptomatic COVID-19 infection causing contamination. This buffer is made in a fumehood**

#### How to make 2 L + 80 mL Binding buffer BB

1. Weigh out **955.2 g** GuHCl and add to a 2 L beaker.

#### **Initials:**

2. Add **500 mL** freshly drawn milliQ. Stir and heat to 60°C to dissolve. Make up to 1200 mL in a 2 L graduated cylinder.

#### **Initials:**

3. Add Isopropanol up to 2 L (**800 mL**).

#### **Initials:**

4. Add **80 mL** 3M sodium acetate, pH 5.2.

#### **Initials:**

5. Add **1mL** Tween-20. Stir well to mix.

#### **Initials:**

6. Vacuum filter into sterile bottles.
7. Label with batch and date.

#### **Initials:**

Store Binding Buffer BB buffer in a flammables cupboard at room temperature (+15°C – +25°C).

## Supplementary Method 9 Preparation of Binding Buffer (BB)

### How to make 4L + 160 mL Binding buffer BB

1. Add **1000 mL** freshly drawn milliQ water which has been heated to 60°C to a 5L beaker.

**Initials:**

2. Weigh out **1.911 Kg GuHCl** in aliquots using a 3 L beaker and carefully add to 5 L beaker Stir and heat to 60 deg C to dissolve A sterile pipette may be required to stir. Once dissolved, make up to 2400 mL with freshly drawn milliQ water using a 2L graduated cylinder and transferring buffer to second 5L beaker.

**Initials:**

3. Add Isopropanol up to 4 L (**1600 mL**) using a 2L graduated cylinder.

**Initials:**

4. Add **160 mL** 3M sodium acetate, pH 5.2.

**Initials:**

5. Add **2 mL** Tween-20. Stir well to mix.

**Initials:**

6. Vacuum filter into sterile bottles.
7. Label with batch and date.

**Initials:**

Store Binding Buffer BB buffer in a flammables cupboard at room temperature (+15°C – +25°C).

### How to make 5L + 200 mL Binding buffer BB

1. Add **1250 mL** freshly drawn milliQ water which has been heated to 60°C to a 5L beaker.

**Initials:**

2. Weigh out **2.388 Kg GuHCl** in aliquots using a 3 L beaker and carefully add to 5 L beaker Stir and heat to 60 deg C to dissolve A sterile pipette may be required to stir. Once dissolved, make up to 3000 mL with freshly drawn milliQ water using a 2L graduated cylinder and transferring buffer to second 5L beaker.

**Initials:**

3. Add Isopropanol up to 5 L (**2000 mL**) using a 2L graduated cylinder.

**Initials:**

4. Add **200 mL** 3M sodium acetate, pH 5.2.

**Initials:**

5. Add **2.5 mL** Tween-20. Stir well to mix.

**Initials:**

6. Vacuum filter into sterile bottles.
7. Label with batch and date.

**Initials:**

Store Binding Buffer BB buffer in a flammables cupboard at room temperature (+15°C – +25°C).