

Crystallization Solution C Protocol

Goal: Preparation of a Crystallization Solution.

- Created from one or more chemicals with a buffer
- The pH is adjusted by the addition of a counter acid or base

Step 1: Identification of the crystallization solution characteristics.

The required component concentration(s) and the counter acid/base used to adjust the pH can be found in the production report (see example in Table 1).

Step 2: Calculation of the amount of each chemical required to prepare 1 liter of stock solution.

Amount required in g = molarity (M) x FW x liters of solution

Using the example in Table 1:

Amount of chemical L required in g = 2.4 x 70.00 x 1 = 168.00 g

Amount of buffer M required in g = 0.1 x 160.00 x 1 = 16.00 g

Step 3: Crystallization solution preparation (1 liter).

1. Weigh the required amount(s) of chemical(s) into a beaker.
2. Add water to 75% of the final volume (~750 ml).
3. Bring the pH near to the desired value with the appropriate counter-acid or base.
4. Add water to 95% of the final volume (~ 950 ml).
5. Store the crystallization solution at room temperature for 24 hours*.
6. Adjust the pH to the desired value.
7. Add water to give a final volume of 1 liter.
8. Filter the crystallization solution using a 0.22 µm filter.
9. Transfer the solution to the desired format (e.g., 24- or 96-well plate) in a sterile environment.

* Since the acid/base reaction is usually exothermic, this step will bring the solution back to room temperature.

Table 1. Identification of the Crystallization Solution characteristics.

Formulation	Chemical FW
2.4 M Chemical L	70.00
0.1 M Buffer M	160.00
Final pH targeted: 6.0	
Counter acid/base used: Sodium hydroxide (NaOH)	

The value of a chemical's formula weight (FW) is the sum of the atomic weights of the atoms found in one formula unit (including its hydration state) of an ionic compound. For other information about each chemical, please refer to the production report specific for each solution.

Things to remember:

- All chemical information can be found in the production report for each solution.
- Amount of chemical required for each solution is calculated thus:

If concentration is given as X M:

$$\text{Amount in grams} = X \times \text{FW} \times \text{FV (liters)}$$

If the concentration is given as X% v/v:

$$\text{Volume required} = X \text{ ml per } 100 \text{ ml}$$

If the concentration is X% w/v:

$$\text{Amount in grams required} = X \text{ g}/100 \text{ ml}$$

Units Definition

M: Molarity of chemical

FV: Final volume of solution

FW: Formula weight of chemical