



Customized solutions for cell cultures.

Protocol

Production of **liquid medium** from HEK TF powder

Please note, this document may be periodically updated in order to ensure the most current practices are in place. It is the user's responsibility to ensure the latest release of this protocol is applied. Valid versions are made available via Xell's webshop.

Production of liquid medium from HEK TF powder

Guideline:

We recommend preparing the whole powder container in a single batch. For that, please adjust the amounts/volumes per liter given in this protocol according to your batch size.

Material:

- HEK TF powder (use at 20.72 g/L); Xell Cat. 861-XXXXDPM
- H₂O (WFI or equivalent quality)
- 2.10 g/L NaHCO₃ Ph. Eur.
- 1 - 2 mL/L Growth hormone supplement (recommended 1.6 mL/L; e.g. Xell Cat. No. 1005-XXXX); alternatively: 0.05 - 0.1 mL/L LONG® R3 IGF-I (Cat. No. 1006-XXXX; recommended 0.08 mL/L)
The values are corresponding to the use of 5 - 10 mg/L rInsulin (recommended 8 mg/L); alternatively: 0.05 - 0.10 mg/L LONG® R3 IGF-I (recommended 0.08 mg/L)



We recommend wearing a dust mask during preparation.

Visual control:

Check:

- A. Container **Sealed and without any damage.**
- B. Appearance **Free flowing powder** (record color).











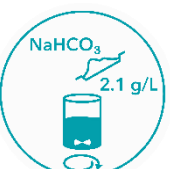

Color: _____






Procedure:

Check:

1.		Fill 0.94 L per 1 L (94% v/v) final medium solution 15-35°C water (WFI or equivalent quality) into the stirred tank/blending vessel. Note: Deviating temperature may alter dissolution rate. An adaption of time for solubilization might be necessary.	
2.		Start the stirrer of the system. Due to foam formation during medium production, the vortex should not reach the stirrer.	

3.		<p>Add 20.72 g/L of HEK TF powder slowly to the stirred water. Avoid clumping.</p> <p>Note: We recommend preparing the whole powder container at once.</p>	
4.		<p>Rinse the emptied powder container with 0.05 L per 1 L final medium solution (5% v/v) of water (WFI or equivalent quality) and pour liquid into the stirred tank.</p>	
5.		<p>Stir for 45 minutes with lid closed.</p> <p>Note: The powder should be completely dissolved and the solution should be clear.</p>	
6.		<p>Add 1 - 2 mL/L Growth hormone supplement (recommended 1.6 mL/L; alternatively: 0.05 - 0.1 mL/L LONG® R3 IGF-I (recommended 0.08 mL/L) from Xell stock solutions.</p> <p>The values are corresponding to the use of 5 to 10 mg/L rInsulin (recommended 8 mg/L) alternatively: 0.05 - 0.1 mg/L LONG® R3 IGF-I (recommended 0.08 mg/L) directly from powder or an appropriate stock solution.</p> <p>Note: Adjustment of growth hormone concentration for optimization is possible, but depends on used cell line and application.</p>	
7.		<p>Stir for 10 minutes with lid closed.</p>	
8.		<p>Add 2.1 g/L NaHCO₃ Ph. Eur. to the stirred tank.</p>	

9.		<p>Stir for 10 minutes with lid closed.</p> <p>Note: <i>The solution should be clear, without precipitates. If not, stepwise increase mixing time by up to further 10 min.</i></p>	<input type="radio"/>
10.		<p>Measure pH (7.0 - 7.5) and osmolality (300 ± 15 mOsmol/kg) of the medium.</p>	<input type="radio"/>
11.		<p>The medium can now be sterile filtered (0.45 µm + 0.2 µm or 0.45 µm + 0.1 µm) and bottled.</p>	<input type="radio"/>

Change History:

Revision	Date	Author	Comment/Description
01-03	n/a	n/a	Initial version
04	03.12.2021	SST	Addition of change history

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