xcell

Customized solutions for cell cultures.

Protocol

Production of **TCX6D Medium** from powder

Production of liquid medium with the TCX6D Powder Kit

Material:

- We recommend preparing the whole Powder Kit in a single batch! For that, please adjust the amounts/volumes per L given in this protocol according to your container/batch size!

- TCX6D Powder (27.01 g/L; Cat.No. 1070-XXXXDPM)
- ESK-Supplement (2.00 mL/L)
- approx. 1 L H_2O per L medium (WFI or equivalent quality)
- 7.50 mL/L 1 M NaOH Ph. Eur.
- 2.10 g/L $NaHCO_3$ Ph. Eur.
- 0.10 mL/L LONG® R3 IGF-I (Cat. No. 1006-XXXX);
  Alternatively, 2 mL/L Growth hormone supplement (Cat. No. 1005-XXXX)
  The values are corresponding to the use of 0.10 mg/L LONG® R3 IGF-I; alternatively: 10 mg/L rInsulin.
- We recommend wearing a dust mask during preparation!

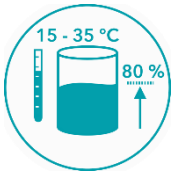
Visual control:

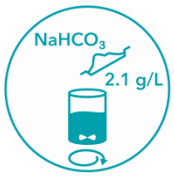
Check:

A.	Container	**Sealed** and **without any damage**.	◯
B.	Appearance	**Free flowing powder** (record color).	◯ Color:_____

Procedure:

Check:

1.		Fill **0.8 L** per 1 L (80% v/v) final medium **15-35°C water (WFI or equivalent quality)** into the stirred tank/blending vessel.	◯
2.		Start the stirrer of the system. Due to foam formation during medium production, the vortex should not reach the stirrer.	◯
3.		Add **27.01 g/L of TCX6D powder** slowly to the stirred water. Avoid clumping. ***Note:*** *We recommend preparing the whole powder kit at once.*	◯

4.		Rinse the empty medium container with a suitable amount of **water** (WFI or equivalent quality) and pour liquid into the stirred tank.	◯
5.		Stir for **30 minutes**. ***Note:*** *The powder will not be completely dissolved at this stage.*	◯
6.		Add **7.50 mL/L of 1 M NaOH** to the stirred tank. *Adjust volume according to batch size.*	◯
7.		Stir for **30 minutes**. ***Note:*** *The powder should be completely dissolved and the solution should be clear.*	◯
8.		Add **2.10 g/L $NaHCO_3$** to the stirred tank. *Adjust volume according to batch size.*	◯
9.		Add an appropriate volume of **water** (WFI or equivalent quality) to reach the final volume. *Final volume depends on batch/container size!*	◯
10.		Stir for ~ **10 minutes** with lid closed. ***Note:*** *The solution should be clear, without precipitates. If not, stepwise increase mixing time by up to further 10 min.*	◯
11.		Add **2.00 mL/L ESK Supplement** to the stirred tank. *Adjust volume according to batch size.*	◯

12.		Add **0.10 mL/L LONG® R3 IGF-I** (alternatively: 2.00 mL/L Growth hormone supplement ***Note:*** *Adjustment of growth hormone concentration for optimization is possible, but depends on used cell line and application.*	◯
13.		Stir for **~ 10 minutes** with lid closed.	◯
14.		Check pH (**pH 7.1 – pH 7.6**) and osmolality (**283 - 343 mOsmol/kg**).	◯ ◯
15.		The Medium can now be **sterile filtered** (0.45 µm + 0.1µm) and **bottled**.	◯

Change History:

Revision	Date	Author	Comment/Description
01	31.03.2022	AWU	Initial version

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