U-CHCF365

Introduction

The U-CHCF365 cell line was developed by Kevin Mellert and Peter Moller at the University of Ulm from the CF365 PDX model established from a scalp metastatic lesion presenting in a <20 year old male with clival chordoma. It is one of the faster growing and easier to maintain chordoma cell lines.

Media formulation

Materials

- IMDM (Gibco, Cat# 12440079)
- RPMI 1640 (Gibco, Cat# 11875085)
- FBS, Certified, US (Gibco, Cat# 16000044) or equivalent quality FBS
- MEM NEAA (Corning, Cat# 25-025-CIR)
- GlutaMAX (Gibco, Cat#35050061)
- Nalgene Rapid-Flow Vacuum Filter (Thermo Scientific, Cat# 5660010)

(4:1) IMDM/RPMI 1640 + 10% FBS + 1% NEAA + 1% GlutaMAX media preparation protocol

- 1. Prepare media in the bio-safety cabinet using aseptic technique. This media is made without antibiotics so extra precautions need to be taken.
- 2. Douse items in 70% Ethanol and place in bio-safety cabinet. Wait at least 10 minutes so ethanol has a chance to disinfect. Check for liquid at base of cap, if present, aspirate.
- 3. Create 500mL of media using volumes to the right by pipetting the media components into the upper compartment of a 500mL Nalgene vacuum filter, filter the components, and then store media at 4°C.

Component	Volume (mL)
IMDM	352
RPMI	88
FBS	50
NEAA	5
Glutamax	5
Total	500

Culturing characteristics

This cell line is thawed using CF Labs general thawing protocol, <u>found here</u>. This line can be grown on standard tissue culture treated, non-collagen coated vessel surfaces as this line's viability and growth are not dependent on collagen. Using the above media formulation and grown in the absence of collagen, U-CHCF365 cells exhibit a doubling time of approximately three and a half days and exhibits a heterogeneous physaliferous morphology. **(A)** U-CHCF365 representative image. The cells have a cobblestone morphology and exhibit a dispersed growth pattern, spreading uniformly across the plate, possibly due to increased mobility. However, this claim needs further investigation. Around 10% - 20% of the cells are in G2/M and will appear detached, spherical, and refractive, as indicated by the blue arrow **(B)** In a 96-well plate, plating 3,000 cells/well (9,375 cells/cm²) will yield 15% confluence 36 hours after plating, labeled as day 0 where 36 hours is the time allowed for cells to adhere and spread out on the plate. The cells reach 90% confluence after nine days. This information can be extrapolated to other vessel types.



