Establishment and reproducible screening of a large-scale CF patient-derived organoid biobank

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About Hubrecht Organoid Technology (HUB): HUB is a paradigm-shifting platform for drug discovery/development, companion diagnostics, and (pre)clinical patient stratification. HUB exploits the pioneering work of Prof. Dr. Hans Clevers who discovered methods to grow stem cell-derived human epithelial ‘mini-organs’ (organoids) from tissues of patients with various diseases. HUB organoids capture disease relevance and allow for unlimited, genetically and phenotypically stable expansion of the cells from both animals and human. Additionally, HUB has established a patient-derived ‘living biobank’ (HUB Organoid biobank) linking genetic and molecular information to drug responsiveness as well as clinical data of the patient.

HUB Organoid biobank for cystic fibrosis

A biobank of organoids from a total of 498 CF patients has been established at HUB. The most common CF genotypes are captured in the organoid biobank (left). Overview of CFTR nonsense mutations present in the biobank (right).

Drug response testing in the Forskolin-induced swelling assay

Potentiators and correctors

Restoration of CFTR function and organoid swelling in an F508del/F508del (left) and F508del/G551D (right) organoid by VX-809/VX-770 treatment. Normalized swelling of organoids over the timeframe of one hour treated with VX-809 (3 μM; 2 h) and VX-770 (3 μM; acute) at 2 mM forskolin (upper panel) and respective AUC values of indicated forskolin concentrations (lower panel).

CFTF mutation-specific responses towards VX-809/VX-770 treatment. Area under the curve values of the normalized organoid swelling with indicated CFTR mutations treated with VX-809 (3 μM; 2 h) and VX-770 (3 μM; acute) as indicated at 0.8 mM forskolin.

Functional assay for CFTR activity

Forskolin-induced swelling (FIS) assay

Representative images of F508del/F508del organoids pre-treated with VX-809/VX-770 and imaged at t = 0 and t+60 min after forskolin.

Quantification of organoid swelling using image analysis software.

Organoids swelling over time at different forskolin concentrations (left) and area under the curve (AUC) of those swelling curves (right). Here shown for F508del/F508del organoids treated with VX-809/ VX-770.

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Gene therapy

Organoid cultures are suitable models for gene therapy approaches. A CF colon organoid culture was infected with a reporter gene therapy vector at MOIs of 2.5 and 20 for 1 h at 37 °C after infection reporter positive cells were observed in both conditions, here visualized in red in the lower panel and as overlay with the brightfield images in the upper panel.

Read-through agents

Restoration of CFTR function and organoid swelling in a G542X/G542X organoid culture over 2 h treated with G418 (50 μg/mL, 48h), VX-809 (1 μM; 24h) and VX-770 (3 μM; acute) as indicated at 2 μM forskolin.

CFTF mutation-specific responses towards read-through treatment. Area under the curve values of the normalized organoid swelling with indicated nonsense mutations treated with G418 (50 μg/mL, 48h) at 2 μM forskolin.