ADME: Permeability- MDCK wild type

Background:

MDCK cells (wild type) are Madin-Darby canine kidney epithelial cells. They endogenously express different transporters, such as canine P-glycoprotein (P-gp). Therefore, it is recommended to analyse test compound on both MDCK and MDCKII-MDR1 cell monolayer and determine the real role of human P-gp on transport of test compound.

Transport of compound through MDCK cell monolayer is used for measuring rate of membrane transport, expressed as apparent (Papp) permeability. Resulting Papp values from both transport directions (apical to basolateral side, and basolateral to apical side) are used for calculation of efflux ratio, as well as of net flux ratio (if MDCKII-MDR1 data are available).

Assay description

Cells

MDCK wild type (wt)

Direction

apical to basolateral (A2B)
basolateral to apical (B2A)
with or without P-gp inhibitor (elacridar)

Compound concentration

10µM (1% DMSO)

Compound requirements

50µl of 10mM stock solution or
1-2 mg of dry matter

Incubation details

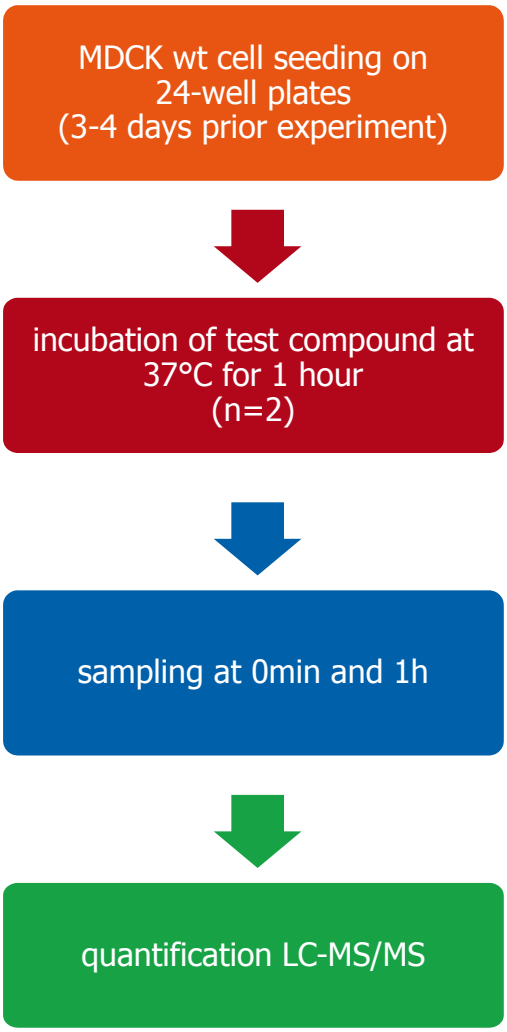
medium: Dulbecco's PBS (pH 7.4)
calibration curve: optional

Detection method

LC-MS/MS with internal standard

Results

P_{app}(A2B), P_{app}(B2A)
efflux ratio, recovery, net flux ratio



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Assay controls

reference compounds: amprenavir and propranolol

cell monolayer integrity control: lucifer yellow

Assay details adjustable to client's and/or project specific requests

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