



Mouse Bcrp1 Vesicular Transport Assay

The BCRP transporter (ABCG2/MXR) belongs to the family of ABC transporters. In addition to its role in cancer resistance, BCRP can influence the ADME/Tox properties of drugs and nutrients. BCRP is localized on the apical membrane of polarized cells and can be found at the major barriers in the body, including the intestine, the BBB and the liver. BCRP has wide substrate specificity, transporting both hydrophilic and hydrophobic compounds.

Mouse Bcrp1 is the mouse orthologue of BCRP (ABCG2). SOLVO's mouse Bcrp1 vesicular transport assay uses isolated membrane vesicles from mouse Bcrp1 expressing MDCKII cells, and operates with ³H-estrone-sulfate (E3S) as the probe substrate. Mouse Bcrp1 transports E3S efficiently while there is negligible background transport in control vesicles (Fig.1.).

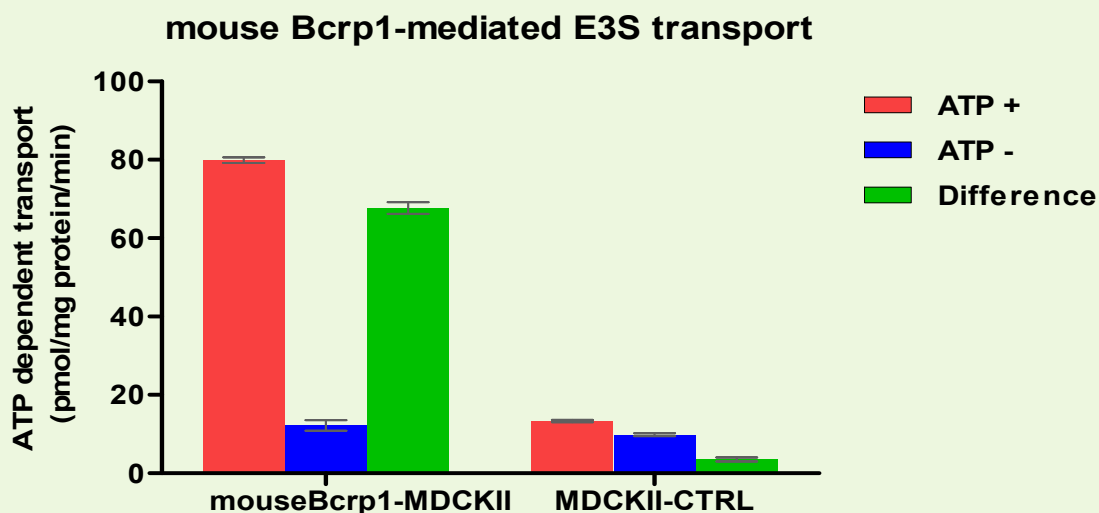


Figure 1 – Uptake of E3S by mouse Bcrp1 containing and control MDCKII vesicles. The experiment was performed in the presence of 1 μ M E3S.



The transport is saturable and fits well to the Michaelis-Menten kinetics (Fig.2.). K_M was found to be around 40 μM and V_{max} was 1400 pmol/mg protein/min for mouseBcrp1. Control vesicles showed no concentration and ATP dependent accumulation of E3S.

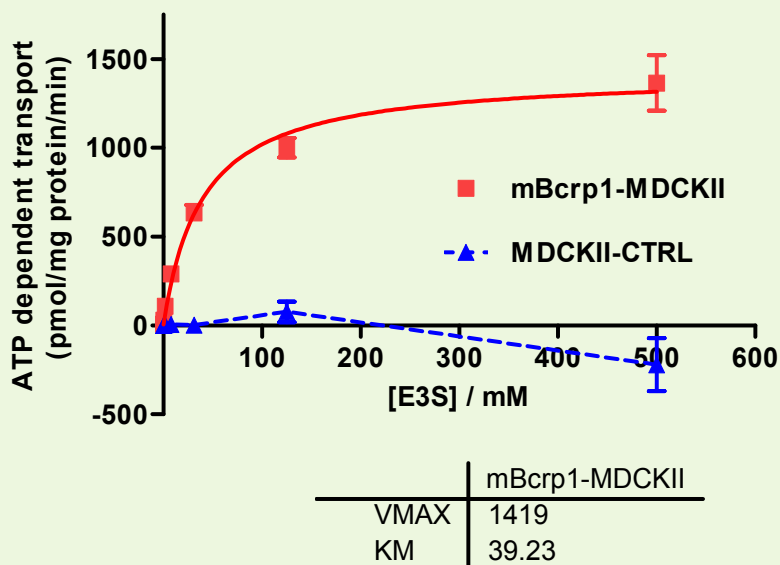


Figure 2. – mouseBcrp1 mediated uptake of E3S at various substrate concentrations.

The transport is time dependent. Saturation can be observed above 5 minutes, while background values are constant up to 15 minutes.

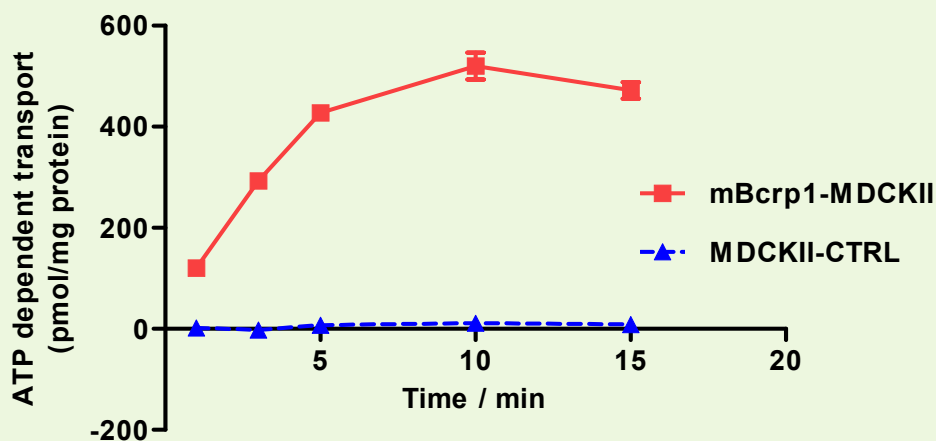


Figure 2 – Uptake of E3S by mBcrp1 containing and control MDCKII vesicles. The experiment was performed in the presence of 1 μM E3S.



The transport is active as it is sensitive to the osmolarity of the buffer (Fig.3.).

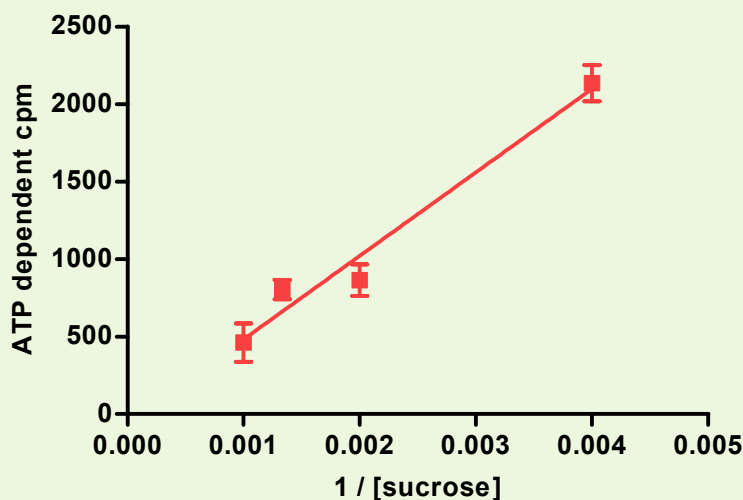


Figure 3 – Sucrose concentration dependence of mouseBcrp1-mediated E3S transport

MouseBcrp1-mediated E3S transport can be inhibited by compounds that interact with the human BCRP transporter (Fig. 4).

Effect of drugs on mBcrp1-mediated E3S transport

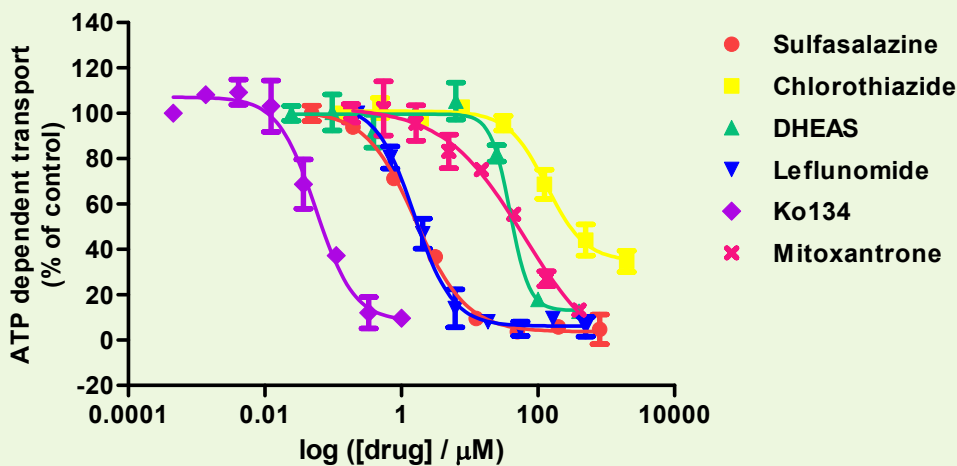


Figure 4 – Effect of known BCRP interactors on the mouseBcrp1 mediated E3S transport.



A comparison of IC₅₀ values obtained for both transporters for the same set of compounds is shown below.

BCRP interactors	IC ₅₀ (μM)	
	mouseBcrp1	BCRP
Leflunomide	1.56	3.86
Chlorothiazide	134	62
Prazosine	2.14	1.80
Dantrolene	0.54	0.25
Mitoxantrone	62.3	104
Ko134	0.056	0.04
Sulfasalazine	1.64	0.31
DHEAS	39.2	43.1