Reviewer's report

Title: Membrane Transport of Camptothecin is Facilitated by Human P-glycoprotein (ABCB1) and Multidrug Resistance Protein 2 (ABCC2)

Version: 1 Date: 31 December 2003

Reviewer: James Polli

Reviewer's report:

General

This manuscript concerns the membrane transport mechanisms of camptothecin. Transport of camptothecin has been the subject of a few previous investigations, since camptothecin oral absorption is low, variable. The authors of the present study have previously observed active camptothecin transport [Gupta et al. The intestinal absorption of camptothecin, a highly lipophilic drug, across Caco-2 cells is mediated by active transporter(s). Anticancer Research. 20(2A):1013-6, 2000], and the present manuscript further characterizes this active transport, particularly in terms of contributions by P-gp and MPR2.

Discretionary Revisions (which the author can choose to ignore)

none

Minor Essential Revisions (such as missing labels on figures, or the wrong use of a term, which the author can be trusted to correct)

A few points should be added to the manuscript:

1) While the focus of this manuscript is the identification and quantification of P-gp and MRP2 roles in camptothecin transport, a point that appears evident from the data is the active apical-to-basolateral (A-B) influx of camptothecin. In all panels in Fig 1, the A-B influx profiles (filled symbols) decrease with camptothecin concentration. Qualitatively, these profiles suggest A-B absorptive influx (in addition to the B-A secretory efflux), although the extent of this influx is typically very modest (Fig 1B, C, and D). Since the scope of the present report concerns the identification of transport mechanism of camptothecin, it would seem that this point merits clarification. The manuscript does make reference to some preliminary MRP1 data that the authors have completed (page 13). Do any result in hand more definitively identify an influx transporter?

2) It would also seem notable that, in Fig 1, MDCKII/P-gp and MDCKII/MRP2 provided efflux ratios greater than 1 (i.e. B-A permeability greater than A-B permeability), suggesting that these models are “better” than Caco-2 or MDCKII wt. On page 13 (bottom), the text suggests MDCKII wt are a more preferable model than Caco-2. It would seem worth emphasizing that [based upon Fig 1B, C, and D] MDCKII/P-gp and MDCKII/MRP2 have definitive advantages over MDCKII wt, which only provided an efflux ratio of about 1 for all concentrations.

3) It would also seem important to note that for Caco-2 monolayers, camptothecin A-B permeability was generally larger than its B-A permeability (Fig 1A). This pattern, along with the decrease in camptothecin permeability with camptothecin concentration, suggests active camptothecin influx for A-B studies. While this absorptive transporter is not identified, this observation is supportive of the less pronounce net influx in Fig 1C and 1D, and 1B. With regard to Fig 1, a suggestion is to show/discuss the more relevant plots first (i.e Fig 1C and 1D), and then show/discuss the supporting [but more complicated] plots (i.e. Fig 1B and 1A), which better emphasis the active A-B absorptive
influx. This observation is of qualitative interest and relates to the manuscript’s general interest, even this observation is not a focus of the stated objective.

Other Minor Essential Revisions:
4) In abstract, rather than or, … MDCK II wt, and MDCKII P-gp and MRP2 … (three substitutions of and for or)
5) Page 7. …300 (space) - 350 ohm cm^2
6) Top page 8. Active and passive transport were evaluated (not paracellular or transcellular).
7) The symbols in Fig 4 and 5 were not all readable in the text (Figure Legends). Perhaps the lower two curve in Fig 5A are switched?
8) In the Figure Legends, it would be helpful to always indicate the direction (and cell model) of transport.
9) The y-axis on Fig 5A needs attention.
10) In Fig 4B, one datum indicates a negative etoposide concentration.
11) Number of significant figures in Fig 4B?

Major Compulsory Revisions (that the author must respond to before a decision on publication can be reached)
none

What next?: Accept after minor essential revisions

Level of interest: An article of importance in its field

Quality of written English: Acceptable

Statistical review: No

Declaration of competing interests:
None