Acute toxicology: In mice, single IV dosing with DM-CHOC-PEN resulted in a $LD_{10/50}$ of 136/385 mg/kg (sexes combined; with 95% conf. limits); oral − LD_{50} >2 g/kg in mice (sex combined). In rats, single IV doses of DM-CHOC-PEN − 100 to 300 mg/kg were well tolerated but produced a transient DLT of hypercholesterimia − 2nd to release of cholesterol from the carbonyl residue. LDL cholesterol was significantly increased 30 fold in the 200 & 300 mg/kg groups which returned to normal (predominatnt HDL varient) by Day 15. Plasma DM-CHOC-PEN and DM-PEN (a metabolite) were assayed by HPLC. In dogs, single dose IV studies were conducted in adult Beagle dogs @ 10, 20, 30 mg/kg concentrations and no toxiicty or deaths were observed. No microscopic nor macroscopic pathology could be identified in any of the animals. No seizures or CNS toxicity noted in any studies. Total rat and dog brain examinations for cellular necrosis were negative.

Overall, pharmacokinetic studies for DM-CHOC-PEN in rats and dogs revealed the following profile: rats (300 mg/kg) − AUC_{0-t} = 2.95 (mg, h/L), $T_{1/2}\alpha$ = 0.24 (h), $T_{1/2}\beta$ = 2.98 (h) & CL = 34.86 (mL/h) and for dogs (30 mg/kg) − AUC_{0-t} = 1.12 (mg, h/L), $T_{1/2}\alpha$ = 0.63 (h), $T_{1/2}\beta$ = 18.7 (h) & CL = 342.7 (L/h) [a two compartment model]. The AUC was linear for the doses. There were no differences between males & females. DM-CHOC-PEN could be identified in normal rat brain tissue (in 200 μg/g quantities) post single injections with 135 mg/kg, however, neither neurotoxic nor psychological changes were noted in any of the above tudies.

Summary: The release of cholesterol from hydrolysis of the cholesteryl carbonate resulting in elevated levels of LDL-cholesterol, that returns to normal values is the only DLT that could bve identified and does appear to be a limiting toxicity. Neither bone marrow nor hepatic toxicity was noted in the rat or dog models.

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Validation of a cell panel for preclinical evaluation of antitumor efficacy and toxicity of anticancer agents

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Aims: To collect and validate a cell panel for preclinical evaluation of antitumor efficacy and toxicity of anticancer drugs.

Methods: Drug activity of amsacrine, arsenic trioxide, bortezomib, cisplatin, cytarabine, doxorubicin, etoposide, 5-fluorouracil, gefitinib, imatinib, melphalan, PKC412, rapamycin and vincristine were tested in tumor cells prepared from patient samples from eleven different hematological and solid tumor diagnoses. Method validation was performed by comparing drug sensitivity in vitro with established clinical use of the drugs. For in vitro toxicity testing a normal cell panel consisting of lymphocytes (peripheral blood mononuclear cells), renal- (renal proximal tubular epithelial cells), liver- (cell line of tumor origin) and epithelial (telomeras transfected cell line) cells was used. Also, $CD34^+$ umbilical cord blood cells were used for prediction of bone marrow toxicity in the 14 days granuclocyte macrophage (GM14) assay. A GM14 index was calculated by dividing the IC_{50} from the GM14 assay with the median IC_{50} for the most sensitive tumor type for each drug. Drug sensitivity for both tumor and normal cells was measured in the non-clonogenic fluorometric microculture cytotoxicity assay.

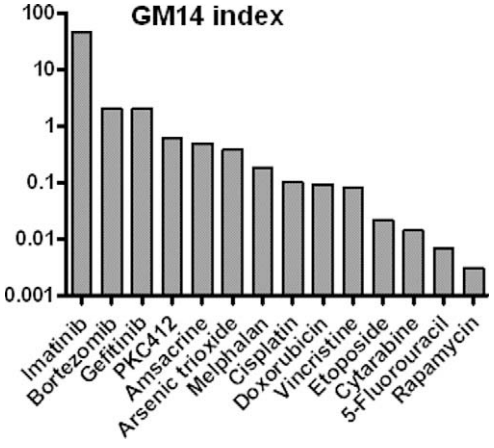

Figure 1. Index of IC_{50} from the GM14 assay and the median IC_{50} from the most sensitive diagnosis in vitro for each drug.

Results: In general, in vitro drug activity in patient tumor cells reflected known clinical activity of the drugs investigated. As an example, CML was the most sensitive tumor type for imatinib, and cisplatin and 5-fluorouracil were the most solid tumour active agents. The toxicity panel was easy to handle in a high throughput manner, and was able to detect differences in therapeutic ratios, e.g. between targeted drugs and classical cytotoxic agents, which is shown in the GM14 assay (figure 1).

Conclusion: In the preclinical stage of drug development, tumor cells from patient samples can be used for prediction of cancer diagnosis specific activity and a normal cell panel may reflect expected toxicity.