For a dog, there was a similar tendency. Biliary excretion continued up to 48 hours after IHL-305 dosing, whereas it was completed at 24 hours after CPT-11 dosing in rats.
Conclusions: After the administration of IHL-305, irinotecan was cleared very slowly from plasma, and most irinotecan in plasma existed as the lactone form. IHL-305 dosing also retained more plasma SN-38 longer than CPT-11 dosing in rats. These pharmacokinetic profiles of IHL-305 were considered to explain its superior antitumor activity other than enhanced permeability and retention (EPR) effect.

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Novel prodrugs of SN38 generated by Multi-Arm Poly(ethylene glycol)

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Background: SN38 (10-hydroxy-7-ethyl-camptothecin) is the active metabolite of CPT-11 (Camptosar®). SN38 has not been used directly as an anticancer drug due to its poor solubility in any pharmaceutically acceptable excipients. Using multi-arm high molecular weight PEG, we have successfully generated novel water soluble prodrugs of SN38.
Material and Methods: In order to increase drug payload, multi-arm PEG was used. In particular, 40k 4-arm-PEG-OH was first converted to PEG acid, then conjugated with a properly protected SN38 intermediate with different amino acid linkers attached to the 20-hydroxyl position to give the PEG-SN38 conjugates. The aqueous stability and hydrolysis property in rat and human plasma were monitored using UV based HPLC methods. The *in vitro* cytotoxicity of all the PEG conjugates was tested in several different tumor cell lines. The *in vitro* metabolism study of PEG-SN38 conjugates was examined in rat hepatocytes.
Results: Using proper protecting and de-protecting strategies, two different chemistries have been developed to synthesize the PEG-SN38 conjugates in high yields. The process was readily adaptable for scale up development. All four PEG-SN38 conjugates had good solubility in water, with up to 4 mg/mL equivalent solubility of SN38 achieved. All compounds showed good stability in saline and other aqueous medium for up to 24 hrs at room temperature. All conjugates demonstrated potent *in vitro* cytotoxicity against a panel of cancer cell lines. The sensitivity of cells to PEG-SN38 was in the order: COLO205 > HT29 = OVCAR-3 > A549. PEG-SN38 conjugates were equipotent to native SN38 and about 10 to 600 fold more potent than CPT-11. PEG-SN38 conjugates were 8 to16 fold more sensitive than Pegamotecan (a PEGylated prodrug of camptothecin) in COLO 205, HT-29 and OVCAR-3 cells. In human plasma, SN38 was steadily released from the PEG conjugates with a doubling time of 22 to 52 minutes and the release appeared to be pH and concentration dependent. Metabolic study using rat hepatocytes showed SN38 released from conjugates formed a phase II SN38-glucuronide metabolite.
Conclusions: Using multi-arm high molecular PEG, we have successfully prepared several water soluble prodrugs of SN38 for direct parental applications. The payload of the parent drug was almost doubled compared to the traditional straight chain PEGylation. High water solubility was achieved. All PEG-SN38 conjugates showed potent *in vitro* anti-tumor activities which are much more potent than the small molecule prodrug CPT-11 and Pegamotecan. These results warrant further study of these conjugates in animals. PEGylation appears to be a promising approach to deliver SN38, a potent but insoluble cytotoxic agent.

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Mass balance, pharmacokinetics and metabolism of [14C] BMS-354825 in healthy male subjects

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Background: Dasatinib (DAS) – a potent, orally active inhibitor of several oncogenic kinases – has demonstrated clinical efficacy in CML and Ph+ ALL. This study assessed the mass balance, PK, metabolism, and routes/extent of elimination of a single oral dose of 100 mg (120 μCi) [^{14}C] DAS in healthy male subjects.
Methods: This was an open-label, non-randomized, single-dose study involving 8 subjects (21−41 y.o.). All received a single oral dose of 100 mg of [^{14}C] DAS solution containing 120 μCi of total radioactivity (TRA). Vital signs, physical exams, ECGs, clinical labs, and adverse events were conducted/monitored. Blood, urine, and feces were collected to measure DAS, the piperazine N-oxide metabolite of DAS (M5) and TRA, and for biotransformation analyses.

	Cmax (ng/mL), GM[a] (CV)	AUC_{∞} (ng h/mL), GM[a] (CV)	AUC_{0-T} (ng h/mL), GM[a] (CV)	Tmax (h), Med[b] (Min, Max)	T-half (h), Mean (SD)	CLR (mL/h), Mean (SD)	UR (%), Mean (SD)
DAS	104.47 (29)	313.97 (42)	298.8 (44)	0.5 (0.25, 1.5)	3.59 (1.01)	404.83 (168.73)	0.12 (0.05)
M5	2.96 (55)	15.28 (53)	8.88 (74)	1.5 (0.75, 3)	3.15 (1.2)	–	1.2 (0.49)
TRA	224.61 (23)	1231.34 (53)	400.43 (41)	0.5 (0.25, 1.5)	3.96 (2.63)	8636.92 (2651.12)	–

[a]GM, geometric mean. [b]Med, median.

In plasma, DAS AUC(INF) accounted for ~29% of the AUC(INF) of TRA. Multiple metabolites were identified with DAS as the major component. In feces, DAS was a prominent component accounting for 19% of the dose. Metabolites M20 (4-hydroxy-chloromethylphenyl DAS) and M6 (the carboxylic acid derivative of DAS) were detected in significant amounts. No conjugated metabolites were detected in feces.
Conclusions: (1) Radioactivity was primarily eliminated in feces. Mean total recoveries through 9 days post dose were 85% in feces and 4% in urine (total mean = 89%). (2) Negligible amounts of DAS and M5 were excreted in the urine, ~1% of dose. (3) The parent drug was an important drug-related component and M5 a minor metabolite in plasma. (4) A single 100 mg dose of [^{14}C] DAS was safe and tolerable.

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Antitumor activity of IHL-305, a novel PEGylated liposome containing irinotecan, in human xenograft models

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Background: Irinotecan hydrochloride (CPT-11) is an antitumor agent that acts by inhibiting DNA topoisomerase I. CPT-11 is widely used in clinic due to its confirmed evidence of antitumor efficacy. IHL-305 is a preparation of irinotecan encapsulated in PEGylated liposome. Liposome preparations are known to be selectively transported to tumor tissues due to the effect of enhanced permeability and retention (EPR). In this study, antitumor efficacy profiles of IHL-305 were evaluated in comparison with CPT-11 using nude mice subcutaneously transplanted with various human cancer cell lines.
Materials and Methods: After transplanting human cancer cell lines (colon, non-small cell lung, small cell lung, prostate, ovarian, and gastric cancer cells) subcutaneously to the inguinal region of nude mice, the animals were grouped on the day when the estimated tumor volume reached about 60–180 mm^3 (Day 0). IHL-305 or CPT-11 was administered intravenously (i.v.) 1–3 times at 4–14 days intervals (total dose 16.875–135 mg/kg or 18.75–270 mg/kg as irinotecan). Physiologic saline or empty liposomes were administered as negative controls with the same administration schedule. Tumors were excised on Day 21, and tumor growth inhibition (TGI) rates (%) were calculated from tumor weights.
Results: The TGI rates for IHL-305 doses (16.875–135 mg/kg) versus CPT-11 doses (18.75–270 mg/kg) tested were 99.2−99.5% vs 35.5−67.2% on QG-56 (NSCLC), 34.7−93.1% vs 4.8−45.8% on NCI-H460 (NSCLC), 86.7−99.8% vs 74.1−88.0% on NCI-H82 (SCLC), 97.9−99.0% vs 48.0−62.3% on PC-3 (prostate), 24.0−89.9% vs 7.7−42.5% on HT-29 (colon), 62.1−91.9% vs 39.0−87.7% on HCT116 (colon), 77.1−80.8% vs 57.3−69.2% on MKN45 (gastric), and 69.1−97.7% 20.2−64.3% on ES-2 (ovarian) cancer xenografts. In all tested xenograft models, IHL-305 demonstrated superior TGI rates to CPT-11 even in HT-29 colon cancer cell line, which has shown intrinsic resistance to CPT-11. No significant changes of body weight were noted in IHL-305 treated groups.
Conclusions: IHL-305 demonstrated stronger tumor growth inhibition effect than CPT-11 on various human cancer xenografts.

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Combination therapy for liver tumor growth and metastasis by low dose rapamycin and FTY720

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Background: Our previous studies demonstrated that the new immunomodulator FTY720 could suppress liver tumor growth and metastasis through down-regulation of cell survival and invasion pathways. On the