Poster #1513
Abstract #64977

Absorption, Distribution, and Excretion of [^{14}C]-APX001 after Single-Dose Administration to Rats and Monkeys

Robert S. Mansbach, PhD [1], Karen J. Shaw, PhD [1], Michael R. Hodges, MBBS, BSc [1*], Samantha Coleman, PhD [2] and Michael E. Fitzsimmons, PhD [2]
[1] Amplyx Pharmaceuticals Inc., San Diego, CA, U.S.A. [2] Covance Laboratories, Madison, WI, U.S.A.

Presenting author:
*Michael R. Hodges MBBS, BSc
Amplyx Pharmaceuticals Inc.
mhodges@amplyx.com

ABSTRACT

Background: APX001 is a first-in-class, intravenous (IV) and oral (PO) broad-spectrum antifungal agent in clinical development for the treatment of invasive fungal infections (IFIs) due to *Candida*, *Aspergillus* and rare molds.

Methods: The absorption, distribution and excretion profiles of [^{14}C]APX001-derived radioactivity were determined in rats (albino and pigmented) and monkeys. Rats (some implanted with bile duct cannulae) were administered a single 100 mg/kg oral dose or a 30 mg/kg intravenous (IV) dose. Monkeys were administered a single 6 mg/kg IV dose. Samples of blood, urine, feces and bile, as well as carcasses, were collected through 168 hours after dosing. Samples were analyzed for total radioactivity content by liquid scintillation counting, and carcasses were analyzed by quantitative whole-body autoradiography (QWBA).

Results: [^{14}C]APX001-derived radioactivity was rapidly and extensively absorbed and extensively distributed to most tissues for both routes of administration in both species. In rats, tissues with the highest radioactivity C_{max} values included bile, abdominal fat, reproductive fat, subcutaneous fat, and liver, but radioactivity was also detected in tissues associated with IFI, including lung, brain and eye. In monkeys, the highest C_{max} values were in bile, urine, uveal tract, bone marrow, abdominal fat, liver, and kidney cortex. Liver and kidney were the tissues with highest radioactivity, but as in the rat, radioactivity was also detected in lung, brain and eye tissues. In pigmented rats, radiocarbon was densely distributed into pigmented tissue and more slowly cleared than from other tissues. Mean recovery of radioactivity in rats was approximately 95-100%. In bile duct-intact rats, >90% of radioactivity was recovered in feces. In cannulated rats, biliary excretion of radioactivity was the major route of elimination and accounted for 88.8% of the dose, whereas urinary and fecal excretion of radioactivity was minor and accounted for 2.56% and 5.42% of the dose, respectively. In monkeys, the overall recovery of radioactivity was 87.6%, and was eliminated in feces (49.8% of dose) and to a lesser extent in urine (20.6% of dose).

Conclusion: The results indicate that APX001-related radioactivity is extensively distributed to major tissues (including tissues relevant to IFI) in both rats and monkeys and cleared primarily by biliary/fecal excretion.

METHODS

Male and female Sprague Dawley rats were administered a single 100 mg/kg oral dose or a 3-hour 30 mg/kg IV infusion of [^{14}C]APX001. Some rats were equipped with bile duct cannulae. Distribution and excretion was also assessed in pigmented (Long Evans) rats. Samples of blood were collected at approximately up to 120 hours post-dose after oral dosing and up to 120 hours after the end of IV infusions. Urine was collected for up to 168 hours post-dose; feces was collected for up to168 hours post-dose; and bile was collected for up to 120 hours post-dose. Blood and carcasses for assessment of tissue distribution were sampled for up to 72 hours post-dose. All blood, plasma, and excreta were analyzed for total radioactivity content by liquid scintillation counting, and carcasses were analyzed by QWBA.

Male cynomolgus monkeys were administered a 3-hour intravenous infusion (6 mg/kg) of [^{14}C]APX001. Blood, urine, feces and carcasses were collected for up to 168 hours after the conclusion of the infusion. As in the rat study, blood, plasma, urine, and feces were analyzed for radioactivity by liquid scintillation counting, and carcasses were analyzed by QWBA.

For QWBA analyses, sections were collected at five to six levels of interest in the sagittal plane. All major tissues, organs, and biological fluids were represented. Collected sections were dried and mounted. Mounted sections were exposed on phosphorimaging screens along with fortified blood standards for subsequent calibration of the image analysis software. Screens were exposed for four days. The autoradiographic standard image data were sampled using image analysis software to create a calibrated standard curve. Specified tissues, organs, and fluids were analyzed. Tissue concentrations were interpolated from each standard curve as nanocuries/g and then converted to ng equivalents/g on the basis of the test article specific activity.

RESULTS

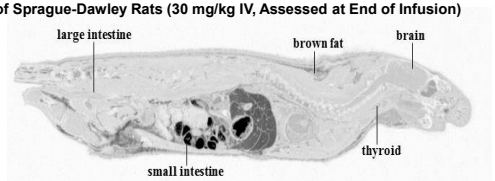
In rats, highest radioactivity C_{max} values were recorded in bile, abdominal fat, reproductive fat, subcutaneous fat, and liver, but radioactivity was also detected in tissues associated with IFI. In pigmented rats, radioactivity was most abundant in the melanin-containing uveal tract of the eye as well as the non-pigmented tissues noted in Sprague Dawley rats after both oral and IV doses.

Table 1. [^{14}C]APX001-Related Radioactivity in Tissues of Sprague-Dawley Rats

Concentration (ng Equivalents [^{14}C]APX001/g)					
Tissue	Sacrifice Time				
	0.5 h	4 h	8 h	24 h	72 h
Bile	242000	434000	82500	39600	ND
Fat (abdominal)	21900	223000	77700	3850	ND
Fat (reproductive)	17900	181000	80700	3880	ND
Fat (subcutaneous)	16900	166000	72600	3490	ND
Liver	98400	138000	46000	9690	BLQ
Lung(s)	8060	24200	8770	1560	ND
Brain cerebellum	9550	21000	7380	1340	ND
Brain cerebrum	9460	20500	7490	1330	ND
Brain medulla	10200	22800	8500	1300	ND

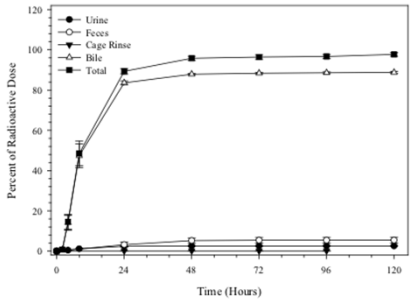
ND=Not detectable (sample shape not discernible from background or surrounding tissue): BLQ=below quantification limit: h=hours

Figure 1. Autoradiographic Distribution of [^{14}C]APX001-Related Radioactivity in Tissues of Sprague-Dawley Rats (30 mg/kg IV, Assessed at End of Infusion)



Nearly all (95-100%) radioactivity was recovered from rats. In rats without bile duct cannulae, >90% of radioactivity was recovered in feces. In cannulated rats, biliary excretion of radioactivity was the major route of elimination and accounted for 88.8% of the dose, whereas urinary and fecal excretion of radioactivity was minor and accounted for 2.56% and 5.42% of the dose, respectively.

Figure 2. Recovery of [^{14}C]APX001-Related Radioactivity from Male Sprague-Dawley Rats with Implanted Bile Duct Cannulae (30 mg/kg IV)



RESULTS (cont'd)

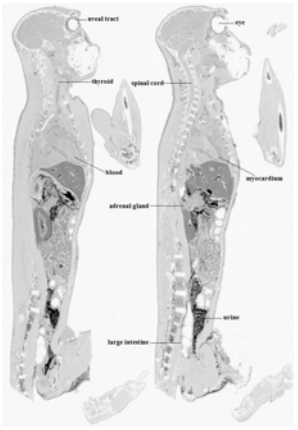
In monkeys, the highest C_{max} values were in bile, urine, uveal tract, bone marrow, abdominal fat, liver, and kidney cortex. Liver and kidney were the tissues with highest radioactivity, but as in the rat, radioactivity was also detected in lung, brain and eye tissues.

Table 2. [^{14}C]APX001-Related Radioactivity in Tissues of Cynomolgus Monkeys

Concentration (ng Equivalents [^{14}C]APX001/g)						
Tissue	Sacrifice Time					
	0 h	1 h	8 h	24 h	72 h	168 h
Bile	230000	257000	286000	153000	4080	125
Bone marrow	15200	8170	2080	184	ND	ND
Eye uveal tract	25100	24000	27100	15200	5450	3730
Fat (abdominal)	12800	14100	NR	192	ND	ND
Kidney cortex	12100	10400	4770	1440	299	ND
Liver	12400	9670	4680	911	233	BLQ
Urine	47900	89500	16700	7930	ND	ND
Lung(s)	3320	3000	1160	354	ND	ND
Brain cerebellum	3090	1800	1130	ND	ND	ND
Brain cerebrum	3090	1860	1010	ND	ND	ND
Brain medulla	3180	1800	1050	ND	ND	ND

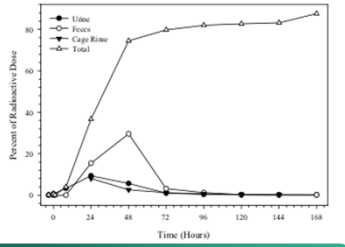
ND=Not detectable (sample shape not discernible from background or surrounding tissue); NR=Not represented (tissue not present in section); BLQ=below quantification limit; h=hours

Figure 3. Autoradiographic Distribution of [^{14}C]APX001-Related Radioactivity in Monkey (6 mg/kg IV, Assessed at End of Infusion)



The overall recovery of radioactivity was 87.6%, and was eliminated in feces (49.8% of dose) and to a lesser extent in urine (20.6% of dose). Tissue concentrations of radioactivity declined below quantifiable levels in all matrices by 168 hours post infusion except gall bladder and melanin-containing tissues (uveal tract, eye, and pigmented skin).

Figure 4. Recovery of [^{14}C]APX001-Related Radioactivity from Monkey (6 mg/kg IV)



CONCLUSIONS

APX001-related radioactivity is extensively distributed to major tissues relevant to IFI (including lung, brain and eye) in both rats and monkeys. The major route of clearance of radioactivity appears to be by biliary/fecal excretion. These properties suggest APX001 has the potential to treat deep-seated invasive fungal infections in a wide variety of tissues including lung, liver, kidney, and brain.