

Table 10. Summary of No-Observable-Adverse-Effect-Levels in Repeat-Dose Studies

Study	Description	Test System	Doses (mg/kg); Vehicle	Results	Conclusions
AA-TX-004 (89/007)	Single IV Dose Rat Study	5 rats/sex/dose	0, 4, 8, 16; sterile phosphate buffered saline without Ca ²⁺ or Mg ²⁺ at pH 7.4 (PBS)	Limb edema, coloration of urine, limb, skin at all doses; Subdued behavior at 4 & 8; Subdued behavior with no reaction at 16; No effects on body wt; No macroscopic abnormalities	MTD = 16 mg/kg NOEL could not be determined
AA-TX-005 (89/009)	Single SC Dose Rat Study	5 rats/sex/dose	0, 10, 80, 150; PBS	Limb edema, subdued behavior; unsteady gait, urine coloration at all doses; Skin coloration, tremors, half-closed eyes at 80, 150; Subdued behavior with no reaction, labored breathing at 150; All clinical signs normal at 24 h ↓ body wt gain at ≥80 in ♂ at 1 week; No macroscopic abnormalities	MTD = 150 mg/kg NOEL could not be determined
AA-TX-006 (89/010)	Single SC Dose Mouse Study	5 mice/sex/dose	0, 50, 100, 200 PBS	1/5 ♀ died at 200; Subdued behavior, half-closed eyes, colored ears & urine at ≥50; Tremors, unsteady gait, piloerection, lacrimation at ≥100; Labored breathing & prostration at 200; All clinical signs normal at 24 h; Dose-related ↓ body wt gain in week 1 in ♀; No macroscopic abnormalities	200 mg/kg can be lethal MTD = 100 mg/kg NOEL could not be determined
AA-TX-007 (89/008) AA-TX-007 (continued)	Single IV Dose Mouse Study	5 mice/sex/dose	0, 15, 30, 45; PBS	2/5 ♂ & 2/5 ♀ died at 45; All doses: limb edema, colored urine; Unsteady gait, subdued behavior, piloerection, lacrimation, labored breathing at ≥30; Tremors, hypersalivation at 45; Dose-related ↓ body wt gain in week 1 in ♂; also in week 2 at 30; No macroscopic abnormalities	45 mg/kg can be lethal MTD = 30 mg/kg NOEL could not be determined
AA-TX-003 (034-001)	Single Dose Intrathecal Rat Study	5 rats/sex/dose	80, 160, 320, 640 µg total dose; PBS	Deaths at ≥160 µg; Body tremors/twitching, limb deficiencies, slowness or inability to right, unsteady gait, curled or arched tail & arched back at all doses; Convulsions & prostration at ≥160; Rapid respiration at 640 Most signs normal in survivors by	LD ₅₀ = 250 µg

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				Day 3; No macroscopic abnormalities	
AA-TX-009 (89/006)	7-day SC Cynomolgus Monkey Range-finding Study	1 monkey/sex/dose	0/50; 15, 40, 65, 90; PBS	1/1 ♂ died at 90; Colored skin & urine, ↓BP, HR; ↑bili at all doses; Half-closed eyes, locomotory difficulty, tremors, reduced activity & piloerection at ≥40; Lack of response to stimulation, labored breathing, vomiting at ≥50; ↓ activity, with or without reaction to stimuli, absence of ocular reflex, ↑gluc, urea, creatinine, AST at 65 & 90; Convulsions, ↑ ALT at 90; Dose-related ↓ body wt & food consumption; At necropsy: Colored kidneys, vertebrae, bone marrow, injection site; Colored tissues, distended stomach, clot in heart ventricle in ♂ sacrificed moribund at 90.	90 mg/kg/day can be lethal; MTD = 65 mg/kg/day, but too high for chronic dosing; NOEL could not be determined
AA-TX-008 (89/005)	7-day SC Mouse Range-finding Study	8 mice/sex/dose	0, 40, 80, 120, 160, 200; PBS	1/8 ♂ & 6/8 ♀ died at 200; Colored urine at 40; Subdued behavior, tremors, locomotory difficulties, limb edema, coloration of skin & urine at ≥80; ↑ bili in ♂ at ≥80; ↓ Hb & PCV at 160; ↓ body wt at 24 h from 80 in ♂; dose-related for 7 days ♀ Liver, kidney coloration at all doses--no other macroscopic abnormalities	200 mg/kg/day can be lethal; MTD = 160 mg/kg/day, but too high for chronic dosing NOAEL = 40 mg/kg/day
AA-TX-002 (661965)	7-day IV Continuous Infusion Mouse Range-finding Study	3 mice/sex (controls) 5 mice/sex/dose (treated)	0, 10, 50, 100; PBS	Colored urine at all doses; Ovarian cysts in 3/5 at 50 & 4/5 at 100	NOAEL = 100 mg/kg/day
AA-TX-001 (89/001)	7-day IV Continuous Infusion Cynomolgus Monkey Range-finding Study	1♂, 1♀ alternating escalating doses (24 h) 1/sex/dose for 6 & 7d	6,12, 24, 48, 96, 192, 350, 500 (24 h) 96 (6d) 72 (7d); PBS	350 & 500 lethal (24 h) At 72 (7 days): Coloration of urine & skin, prostration & hypoactivity, ↓ food consumption, body wt, ↓ body temp, arterial BP, ↓ gluc, total chol & ALK-P; ↑ total bili, creatinine, AST, ALT; Microscopic: Slight cortical tubular dilatation in the kidney & lymphoid	Dose levels below 72 mg/kg/day should be used for repeat dosing by this route

Study	Description	Test System	Doses (mg/kg); Vehicle	Results	Conclusions
AA-TX-010 (89/011) AA-TX-010 (continued)	4-week SC Mouse Study with 2-week Recovery Period	Main 16/sex/dose Recovery 8/sex/dose TK 9/sex/dose	0, 10, 20, 40, 80; PBS	cell infiltration in the liver 1/16 sacrificed at 80 (self-mutilation of tail); Clinical: Colored urine, dorsal skin, injection sites, ears & tail at all doses, all return to normal within days except skin; Tremor, unsteady gait, subdued behavior at 80 on first day of dosing; ↓ Hb, PCV at ≥20 in ♀; ↑ monocytes in ♂ at 20 & 80; ↑ K ⁺ in ♂ at ≥20, & in ♀ at ≥40; ↑ gluc ♂ at ≥10; Chemistries normal after 2-week recovery; At necropsy: dose-related ↑ in dark liver, kidney, & epididymides, stained injection sites, stained skin/subcutis. At end of recovery period: stained injection sites, epididymides, ↓ incidence of dark liver & kidney; Microscopic: dose related pigmented Kupffer cells, pigmented fibroblasts in skin/subcutis & macrophages at injection site; some pigment accumulation in proximal tubules of nephron; After 2-week recovery similar microscopic findings.	NOAEL = 20 mg/kg/day At 20mg/kg/day: Cmax = 9501 ng/mL AUC = 15057 hr*ng/mL
AA-TX-011 (89/012) AA-TX-011 (continued)	4-week SC Cynomolgus Monkey Study	4 monkeys/sex/dose	0, 5, 15, 40; PBS	Colored urine, hair, injection sites at all doses; Half-closed eyes, tremor at ≥15; Locomotory difficulties, salivation at 40; ↓ BP, HR, body temp at all doses; Prolonged QT & QT _c associated with ↓HR at all doses; ↓ body wt at all doses in ♀, at 40 in ♂; ↓ RBC, Hb, PCV at ≥15 ↑ total bili, indirect bili in ♀ at 40; At necropsy: ↑ adrenal & ↓ thymus wt at 40; ↑ kidney wt in ♀ at 40; Colored lymph nodes, injection site; Skin/subcutis staining at ≥15; Microscopic: Dose-related eosinophilic droplets in kidney proximal tubule, liver Kupffer cells	NOAEL = 5 mg/kg/day, except for a transitory effect on the rectal temperature & some individual changes in cardiovascular parameters At 5 mg/kg/day: Cmax = 9241 ng/mL AUC = 22752 hr*ng/mL

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				& in macrophages at injection sites & in lymph nodes (probably test item or metabolites); ↑ Bowman's space around kidney glomeruli at 40	
AA-TX-014 (89/003)	Reverse Mutation Assay in Bacteria	<i>S. typhimurium</i>	52-5000 µg/plate; water	Dose/effect ↑ in revertants in TA100 & TA102 with S9	Bacterial mutagen <i>in vitro</i>
AA-TX-015 (89/004)	Mouse lymphoma Mammalian Cell Mutation	L5178Y Mouse lymphoma cells TK ^{+/-}	1.7-5000 µg/mL; water	Mutagenic effects in absence but not in presence of metabolic activation (S9)	Mammalian cell mutagen <i>in vitro</i>
AA-TX-017 (89/015)	Mouse Bone Marrow Erythrocyte Micronucleus	5 OF1(IOPS Caw) mice/sex/dose	11, 20, 35; saline	No increase in MNPCE	Not mutagenic in this <i>in vivo</i> system
AA-TX-018 (1797/AEO/03)	hERG	HERG-transfected CHO cells	13.5, 45, 135, 450, 1350 µM;	No effect at 13.5 µM; 40-54% inhibition at ≥45 µM	Inhibits hERG <i>in vitro</i> by 40-54%
AA-TX-019 (1798/AEO/03)	Action potential duration	Rabbit Purkinje fibers	13.5, 45, 135 µM; Tyrodes	No effect on action potential duration	No effect
AA-TX-016 (1034-001)	CV/Pulmonary Cynomolgus monkey	4 ♂, 4 ♀ monkeys, ascending dose	CV: 0, 0.9, 2.7, 9, 22.5, 45; Pulm: 0, 2.7, 9, 22.5; PBS	↓ BP, HR, body temp, at ≥9, no significant change on ECG no effect on pulmonary	NOAEL = 2.7 mg/kg
AA-TX-013 (89/013)	CV/Pulmonary Rat	8 ♂ S-D rats	0, 2.5, 5, 12.5, 25; PBS	↓ BP, HR, body temp, at 12.5, 25; dose-related ↓ respiration, biologically significant at ≥12.5	NOAEL = 5 mg/kg
AA-TX-012 (89/014)	Mouse Irwin	5 ♂ IOPS Swiss RjOrl mice	0, 2.5, 25, 100; PBS	↓ activity, severe hypothermia, impaired gait at 100	NOAEL = 25 mg/kg