Experiment Number: S0874
Route: Gavage, IV
Species/Strain: Mouse/CD-1

Toxicokinetics Data Summary
Compound: 3'-Azido-3'-Deoxythymidine/ **Analyte:** 3'-Azido-3'-Deoxythymidine
CAS Number: 30516-87-1

Request Date: 7/11/2023
Request Time: 10:03:16
Lab: RTI

Female

	Treatment Group (mg/kg)	
	50 IV Plasma[a,b]	**100 IV Plasma[a,b]**
Cmax_obs (ug/mL)	84.1	143
Tmax_obs (minute)	5	10
Cl (mL/min)	0.014	0.015
V1 (mL)	7.42 ± 0.014	2.61 ± 0.015
MRT (minute)	98.9	63.3
AUCinf_pred (ug min/mL)	4199	7360

Experiment Number: S0874 | **Toxicokinetics Data Summary** | **Request Date:** 7/11/2023

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Species/Strain: Mouse/CD-1 | **CAS Number:** 30516-87-1 | **Lab:** RTI

Female

	Treatment Group (mg/kg)			
	50 Gavage Plasma[a,g]	**100 Gavage Plasma[a,h]**	**200 Gavage Plasma[a,i]**	**300 Gavage Plasma[a,j]**
Cmax_obs (ug/mL)	20.8	38.3	67.5	131.7
Tmax_obs (minute)	5	20	20	20
MRT (minute)	272	250	253	263
AUCinf_pred (ug min/mL)	4029	5858	13586	21141
F	1.16	0.84	0.98	1.02

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Female

	Treatment Group (mg/kg)	
	50 IV Fetal Litter[a]	**100 IV Fetal Litter[a]**
Cmax_obs (ug/mL)	31.4	53.3
Tmax_obs (minute)	15	15
MRT (minute)	129	104
AUCinf_pred (ug min/mL)	4316	5543

Experiment Number: S0874 | **Toxicokinetics Data Summary** | **Request Date:** 7/11/2023

Route: Gavage, IV | **Compound:** 3'-Azido-3'-Deoxythymidine/ **Analyte:** 3'-Azido-3'-Deoxythymidine | **Request Time:** 10:03:16

Species/Strain: Mouse/CD-1 | **CAS Number:** 30516-87-1 | **Lab:** RTI

Female

	Treatment Group (mg/kg)			
	50 Gavage Fetal Litter[a,c]	**100 Gavage Fetal Litter[a,d]**	**200 Gavage Fetal Litter[a,e]**	**300 Gavage Fetal Litter[a,f]**
Cmax_obs (ug/mL)	11.4	23.2	50.5	75.9
Tmax_obs (minute)	30	30	60	20
MRT (minute)	314	288	267	297
AUCinf_pred (ug min/mL)	3966	5130	11139	17891

Experiment Number: S0874
Route: Gavage, IV
Species/Strain: Mouse/CD-1

Toxicokinetics Data Summary
Compound: 3’-Azido-3’-Deoxythymidine
Analyte: 3'-Azido-3'-deoxythymidine beta-D-glucuronide, sodium salt
CAS Number: 30516-87-1

Request Date: 7/11/2023
Request Time: 10:03:16
Lab: RTI

Female

	Treatment Group (mg/kg)	
	50 IV Plasma[a]	**100 IV Plasma[a]**
Cmax_obs (ug/mL)	0.56	1.2
Tmax_obs (minute)	20	20
MRT (minute)	121	248
AUCinf_pred (ug min/mL)	91.9	167

Experiment Number: S0874
Route: Gavage, IV
Species/Strain: Mouse/CD-1

Toxicokinetics Data Summary
Compound: 3’-Azido-3’-Deoxythymidine
Analyte: 3'-Azido-3'-deoxythymidine beta-D-glucuronide, sodium salt
CAS Number: 30516-87-1

Request Date: 7/11/2023
Request Time: 10:03:16
Lab: RTI

Female

	Treatment Group (mg/kg)			
	50 Gavage Plasma[a,k]	**100 Gavage Plasma[a,l]**	**200 Gavage Plasma[a,m]**	**300 Gavage Plasma[a,n]**
Cmax_obs (ug/mL)	0.26	0.48	0.83	1.3
Tmax_obs (minute)	30	20	30	20
MRT (minute)	408	323	465	235
AUCinf_pred (ug min/mL)	102	120	378	247

LEGEND

MODELING SOFTWARE

WinNonlin, Version 1.0
Microsoft Excel 2000 v9.0.6926

MODELING METHOD & BEST FIT MODEL

[a]WinNonlin (Version 1.0 SCI Software, Apex, NC) or Microsoft Excel 2000 v9.0.6926 (Microsoft Corp., Redmond, WA), Non-compartmental model Mean AZT and GAZT plasma concentration vs. time data were analyzed using noncompartmental modeling techniques (Models 200 and 201, WinNonlin)

EXCEPTIONS

[b]Clearance calculated as Dose/AUC, V1 is Vz
[c]For second peak, Tmax2 is 420 min post fifteenth dose and Cmax2 is 18.8 ug/mL
[d]For second peak, Tmax2 is 380 min post fifteenth dose and Cmax2 is 21.5 ug/mL
[e]For second peak, Tmax2 is 380 min post fifteenth dose and Cmax2 is 46 ug/mL
[f]For second peak, Tmax2 is 380 min post fifteenth dose and Cmax2 is 74.2 ug/mL
[g]For second peak, Tmax2 is 365 min post fifteenth dose and Cmax2 is 21.5 ug/mL
[h]For second peak, Tmax2 is 370 min post fifteenth dose and Cmax2 is 40.3 ug/mL
[i]For second peak, Tmax2 is 380 min post fifteenth dose and Cmax2 is 94.4 ug/mL
[j]For second peak, Tmax2 is 380 min post fifteenth dose and Cmax2 is 151.7 ug/mL
[k]For this second peak, Tmax2 is 420 min post sixteenth dose on day 8, Cmax2 is 0.45 ug/mL
[l]For this second peak, Tmax2 is 380 min post sixteenth dose on day 8, Cmax2 is 0.37 ug/mL
[m]For this second peak, Tmax2 is 420 min post sixteenth dose on day 8, Cmax2 is 0.77 ug/mL
[n]For this second peak,Tmax2 is 420 min post sixteenth dose on day 8, Cmax2 is 0.98 ug/mL

ANALYTE

3’-Azido-3’-Deoxythymidine
3'-Azido-3'-deoxythymidine beta-D-glucuronide, sodium salt

TK PARAMETERS

Cmax_obs = Observed or Predicted Maximum plasma (or tissue) concentration
Tmax_obs = Time at which Cmax predicted or observed occurs
Cl = Clearance, includes total clearance
V1 = Volume of distribution of the central compartment, includes Vd and V volume of distribution, Vz apparent volume of distribution NCA, Vapp apparent volume of distribution for intravenous studies
MRT = Mean residence time
AUCinf_pred = Area under the plasma concentration versus time curve, AUC, extrapolated to time equals infinity
F = Bioavailability, absolute bioavailability

TK PARAMETERS PROTOCOL

ANALYSIS METHOD

The experimental limit of quantitation, ELOQ, for AZT is 0.200 µg/mL for 0.3 mL of plasma extracted. AZT concentration at 0 minutes after dosing (C0) in iv studies was calculated using WinNonlin.

TK_INTRAVENOUS PLASMA

<u>50 mg/kg Female</u> (Analyte: 3'-Azido-3'-deoxythymidine)

Actual mean dose 57.1 mg/kg standard deviation 3.35, n is 48. Pregnant females were administered a single intravenous (IV) dose of AZT on GD17. Blood was obtained from three mice per time point of 16 time points at 0,5,10,15,20,30,45,60,90,120,150,180,300,420,720, and 1440 minutes. Plasma was prepared and analyzed by liquid chromatography using a UV detector.

TK PARAMETERS PROTOCOL (cont’d)

100 mg/kg Female (Analyte: 3'-Azido-3'-deoxythymidine/tissue Plasma)

Actual Mean dose was 109.8, standard deviation 5.85, n is 54. Pregnant females were administered a single intravenous (IV) dose of AZT on GD17. Blood was obtained from four mice per time point of 16 time points at 0,5,10,15,20,30,45,60,90,120,150,180,300,420,720, and 1440 minutes.

ANALYSIS METHOD

The experimental limit of quantitation, ELOQ, for AZT is 0.200 µg/mL for 0.3 mL of plasma extracted. After repeat oral administration, C0 for AZT was the measured value.

TK_GAVAGE PLASMA

50 mg/kg Female (Analyte: 3'-Azido-3'-deoxythymidine)

Blood was obtained from three pregnant female mice per time point of 15 time points at 0, 5, 10, 20, 30, 60, 120, 360, 365, 370, 380, 420, 480, 720, and 1440 minutes. PO doses were administered twice daily, approximately 6 h apart, on GD10-GD17. However, on GD17 (Day 8 of dose administration), 1. time 0 dams were euthanized immediately prior to administration of the fifteenth dose, 2. following administration of the fifteenth dose, time 5, 10, 20, 30, 60 and 120 minute dams were euthanized, 3. immediately prior to administration of the sixteenth dose --approximately 6 hours following administration of the fifteenth dose, time 360 dams were euthanized, and 4. following administration of the sixteenth dose, three dams per time point at 365, 370, 380, 420, 480, 720, 1440 min post administration of the fifteenth dose were euthanized.

TK PARAMETERS PROTOCOL (cont’d)

100 mg/kg, 200 mg/kg,300 mg/kg Female (Analyte: 3'-Azido-3'-deoxythymidine)

Three pregnant female mice were dosed per time point of 17 time points at 0,5,10,20,30,60,120,240,360,365,370,380,420,480,600,720, and 1440 minutes. PO doses were administered twice daily, approximately 6 h apart, on GD10-GD17. However, on GD17 (Day 8 of dose administration), 1. time 0 dams were euthanized immediately prior to administration of the fifteenth dose, 2. following administration of the fifteenth dose, time 5, 10, 20, 30, 60, 120 and 240 minute dams were euthanized, 3. immediately prior to administration of the sixteenth dose --approximately 6 hours following administration of the fifteenth dose, time 360 dams were euthanized, and 4. following administration of the sixteenth dose, three dams per time point at 365, 370, 380, 420, 480, 600, 720, 1440 min post administration of the fifteenth dose were euthanized

ANALYSIS METHOD

No fetal samples showed GAZT levels above the experimental limit of quantitation (ELOQ) of 0.1 ug/g. For AZT the ELOQ was 0.10 ug/g. AZT concentration at 0 minutes after dosing (C0) in iv studies was calculated using WinNonlin.

TK_INTRAVENOUS FETAL LITTER

50 mg/kg Female (ANALYTE: 3'-Azido-3'-deoxythymidine)

Actual mean dose 57.1 mg/kg standard deviation 3.35, n is 48. Pregnant animals were administered a single intravenous (IV) dose of AZT on GD17. Three dams per time point --from 0 to 24 hours after dosing-- were sacrificed. (16 time points at 0,5,10,15,20,30,45,60,90,120,150,180,300,420,720, and 1440 minutes.) Fetuses were collected August 2-3, 2004. Immediately following blood collection from the dams, the fetuses were removed from the uterus by C-section and frozen. After thawing, the whole litter was homogenized and an aliquot removed for processing and analysis by liquid chromatography with UV detection.

TK PARAMETERS PROTOCOL (cont’d)

ANALYSIS METHOD

No fetal samples showed GAZT levels above the experimental limit of quantitation (ELOQ) of 0.1 ug/g. For AZT the ELOQ was 0.10 ug/g.

TK_INTRAVENOUS FETAL LITTER

100 mg/kg Female (ANALYTE: 3'-Azido-3'-deoxythymidine)

Actual Mean dose was 109.8, standard deviation 5.85, n is 54. Pregnant animals were administered a single intravenous (IV) dose of AZT on GD17. Four dams per time point --from 0 to 24 hours after dosing-- were sacrificed. (16 time points at 0,5,10,15,20,30,45,60,90,120,150,180,300,420,720, and 1440 minutes.) Fetuses were collected August 19-20, 2004. Immediately following blood collection from the dams, the fetuses were removed from the uterus by C-section and frozen. After thawing, the whole litter was homogenized and an aliquot removed for processing and analysis by liquid chromatography with UV detection.

TK_GAVAGE FETAL LITTER

50 mg/kg,100 mg/kg, 200 mg/kg, 300 mg/kg Female (ANALYTE: 3'-Azido-3'-deoxythymidine)

Pregnant animals were administered a single intravenous (IV) dose of AZT on GD17. Three to four dams per time point --from 0 to 24 hours after dosing-- were sacrificed. (15 time points at 0,5,10,20,30,60,120,360,365,370,380,420,480,720, and 1440 minutes) Fetuses were collected August 12-13, 2004. Immediately following blood collection from the dams, the fetuses were removed from the uterus by C-section and frozen. After thawing, the whole litter was homogenized, and an aliquot removed for processing and analysis by liquid chromatography with UV detection.

TK PARAMETERS PROTOCOL (cont’d)

ANALYSIS METHOD

The experimental limit of quantitation, ELOQ, is 0.200 ug of 3'-Azido-3'-deoxythymidine beta-D-glucuronide, GAZT, per mL of plasma. No fetal samples showed GAZT levels above the experimental limit of quantitation of 0.1 ug/g. Plasma GAZT concentrations that were less than the limit of detection was set to zero for these analyses. Tmax 2 and Cmax 2 are not applicable for IV groups.

TK_INTRAVENOUS PLASMA

50 mg/kg Female (ANALYTE: 3'-Azido-3'-deoxythymidine beta-D-glucuronide, sodium salt)

Actual mean dose 57.1 mg/kg standard deviation 3.35, n is 48. Pregnant females were administered a single intravenous (IV) dose of AZT on GD17. Blood was obtained from three mice per time point of 16 time points at 0,5,10,15,20,30,45,60,90,120,150,180,300,420,720, and 1440 minutes. Plasma was prepared and analyzed by liquid chromatography using a UV detector.

100 mg/kg Female (ANALYTE: 3'-Azido-3'-deoxythymidine beta-D-glucuronide, sodium salt)

Actual Mean dose was 109.8, standard deviation 5.85, n is 54. Pregnant females were administered a single intravenous (IV) dose of AZT on GD17. Blood was obtained from three to four mice per time point of 16 time points at 0,5,10,15,20,30,45,60,90,120,150,180,300,420,720, and 1440 minutes.

ANALYSIS METHOD

The experimental limit of quantitation, ELOQ, is 0.200 ug of 3'-Azido-3'-deoxythymidine beta-D-glucuronide, GAZT, per mL of plasma. No fetal samples showed GAZT levels above the experimental limit of quantitation of 0.1 ug/g. Plasma GAZT concentrations that were less than the limit of detection was set to zero for these analyses. For the PO groups, Tmax 2 and Cmax 2 are measured relative to second dose in daily BID dosing regimen.

TK PARAMETERS PROTOCOL (cont’d)

TK_GAVAGE PLASMA

50 mg/kg Female (ANALYTE: 3'-Azido-3'-deoxythymidine beta-D-glucuronide, sodium salt)

Blood was obtained from three pregnant female mice per time point of 15 time points at 0, 5, 10, 20, 30, 60, 120, 360, 365, 370, 380, 420, 480, 720, and 1440 minutes. PO doses were administered twice daily, approximately 6 h apart, on GD10-GD17. However, on GD17 (Day 8 of dose administration), 1. time 0 dams were euthanized immediately prior to administration of the fifteenth dose, 2. following administration of the fifteenth dose, time 5, 10, 20, 30, 60 and 120 minute dams were euthanized, 3. immediately prior to administration of the sixteenth dose --approximately 6 hours following administration of the fifteenth dose, time 360 dams were euthanized, and 4. following administration of the sixteenth dose, three dams per time point at 365, 370, 380, 420, 480, 720, 1440 min post administration of the fifteenth dose were euthanized.

100 mg/kg, 200 mg/kg, 300 mg/kg Female (ANALYTE: 3'-Azido-3'-deoxythymidine beta-D-glucuronide, sodium salt)

Three pregnant female mice were dosed per time point of 17 time points at 0,5,10,20,30,60,120,240,360,365,370,380,420,480,600,720, and 1440 minutes. PO doses were administered twice daily, approximately 6 h apart, on GD10-GD17. However, on GD17 (Day 8 of dose administration), 1. time 0 dams were euthanized immediately prior to administration of the fifteenth dose, 2. following administration of the fifteenth dose, time 5, 10, 20, 30, 60, 120 and 240 minute dams were euthanized, 3. immediately prior to administration of the sixteenth dose --approximately 6 hours following administration of the fifteenth dose, time 360 dams were euthanized, and 4. following administration of the sixteenth dose, three dams per time point at 365, 370, 380, 420, 480, 600, 720, 1440 min post administration of the fifteenth dose were euthanized.