Experiment Number: S0558	Toxicokinetics Data S	Toxicokinetics Data Summary					
Route: Gavage, IV	Compound: Anthraquinone/	Compound: Anthraquinone/ Analyte: Anthraquinone CAS Number: 84-65-1					
Species/Strain: Rats/Sprague-Dawley	CAS Number						
	Male						
Treatment Group (mg/kg)							
2 IV Plasma	<sup>d</sup> 40 Gavage Plasma <sup>b,e</sup>	100 Gavage Plasma <sup>c,f</sup>	400 Gavage Plasma <sup>c,f</sup>				

Cmax_obs (ug/mL)	2.88 ± 1.12	0.26 ± 0.02	0.70 ± 0.02	2.63 ± 0.28
Tmax_obs (minute)	2			
Tmax_obs (hour)		8	12	18
Half-life (hour)	10-12	12		
AUC_0-T (ug/mL*min)	1.29	3.54	10.7	42.7

Experiment Number: S0558 Route: Gavage, IV Species/Strain: Rats/Sprague-Dawley		Toxicokinetics Data Compound: Anthraquinone CAS Numbe	Request Date: 7/11/2023 Request Time: 10:03:16 Lab: Battelle Columbus			
		Female				
Treatment Group (mg/kg)						
	2 IV Plasma <sup>a,d</sup>	40 Gavage Plasma <sup>b,e</sup>	100 Gavage Plasma <sup>c,f</sup>	400 Gavage Plasma <sup>c,f</sup>		
Cmax_obs (ug/mL)	3.32 ± 1.76	0.25 ± 0.05	0.77 ± 0.14	3.08 ± 0.75		
Tmax_obs (minute)	2					
Tmax_obs (hour)		8	12	18		
Half-life (hour)	10-12	12				
AUC 0-T (ug/mL*min)	1.10	3.73	12.3	51.2		

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

## LEGEND

MODELING SOFTWARE SIGMA PLOT, Version 5.0

## MODELING METHOD & BEST FIT MODEL

<sup>a</sup> Sigma Plot, Version 5.0, was used to calculate the AUC 0-T values by the trapezoidal method. Other reported toxicokinetic parameters are observed values only. Half-life of elimination was "eye-balled" (Half-life). As requested by the NTP, no attempt was made to model the plasma concentration versus time profiles. The plasma concentration time profile was a biphasic curve suggesting that anthraquinone is best described by a two compartment open model, (initial tissue distribution phase (the initial portion of the biphasic curve), and an elimination phase (the terminal linear portion of the biphasic curve).

<sup>b</sup> Sigma Plot, Version 5.0, was used to calculate the AUC 0-T values by the trapezoidal method. Other reported toxicokinetic parameters are observed values only. Half-life of elimination was "eye-balled" (Half-life). As requested by the NTP, no attempt was made to model the plasma concentration versus time profiles. The plasma concentration time profile was characteristic of a two compartmental with first order absorption and elimination. The plasma concentration time profile was characteristic of a two compartmental with first order absorption.

<sup>c</sup>Sigma Plot, Version 5.0, was used to calculate the AUC 0-T values by the trapezoidal method. Other reported toxicokinetic parameters are observed values only. Half-life of elimination was "eye-balled" (Half-life). As requested by the NTP, no attempt was made to model the plasma concentration versus time profiles. The plasma concentration time profile was characteristic of a two compartmental with first order absorption and elimination. The later slow decreasing phase, or terminal linear portion, characterized the elimination phase.

#### **EXCEPTIONS**

<sup>d</sup>AUC from 0-10 hours, Cmax variation unclear could be SD or SEM

<sup>e</sup>Cmax variation unclear could be SD or SEM

<sup>f</sup>The elimination phase was unclear for this dose group and requires extrapolation or additional information to be fully characterized. Half-life values could not be reliably estimated. Cmax variation unclear could be SD or SEM.

Toxicokinetics Data Summary Compound: Anthraquinone/ Analyte: Anthraquinone CAS Number: 84-65-1

#### ANALYTE

Anthraquinone

#### **TK PARAMETERS**

Cmax\_obs = Observed or Predicted Maximum plasma (or tissue) concentration Tmax\_obs = Time at which Cmax predicted or observed occurs Half-Life = Lambda z Half life, t 1/2, the terminal elimination half-life based on non-compartmental analysis AUC\_0-T = Area under the plasma concentration versus time curve, AUC, from time ti (initial) to tf (final), AUClast

## TK PARAMETERS PROTOCOL

#### ANALYSIS METHOD

Plasma was analyzed for anthraquinone concentration by high performance liquid chromatography (HPLC) using an internal standard, a methanol-water mobile phase, and UV detection at 253 nm. The Limit of Quantitation (LOQ) was 0.025 ug/mL.

## TK\_INTRAVENOUS PLASMA

## 2 mg/kg

Mice and Fischer 344 rats were administered a single intravenous or gavage dose. Blood was collected post-dosing from 3 animals/species/route/dose/timepoint for up to 8 timepoints (gavage) or 9 timepoints (intravenous). Mortality was observed in a few mice administered anthraquinone intavenously attributed to rapid rate of delivery and sensitivity to the DMSO formulation. Other rats and mice administered anthraquinone intavenously exhibited clinical signs of toxicity. Group mean and minimum and maximum body weights are for the original group animals before substitutions. Associated with C88036 report date is 10/18/ 1994.

Toxicokinetics Data Summary Compound: Anthraquinone/ Analyte: Anthraquinone CAS Number: 84-65-1 Request Date: 7/11/2023 Request Time: 10:03:16 Lab: Battelle Columbus

## TK PARAMETERS PROTOCOL (cont'd)

TK\_GAVAGE PLASMA

# 40 mg/kg, 100 mg/kg, 400 mg/kg

Mice and Fischer 344 rats were administered a single intravenous or gavage dose. Blood was collected post-dosing from 3 animals/species/route/dose/timepoint for up to 8 timepoints (gavage) or 9 timepoints (intravenous). Mortality was observed in a few mice administered anthraquinone intravenously attributed to rapid rate of delivery and sensitivity to the DMSO formulation. Other rats and mice administered anthraquinone intravenously exhibited clinical signs of toxicity. Group mean and minimum and maximum body weights are for the original group animals before substitutions. Associated with C88036 report date is 10/18/ 1994.