

Experiment Number: K06898

Route: Gavage, IV

Species/Strain: Mice/B6C3F1

Toxicokinetics Data Summary

Compound: alpha/beta-Thujone mixture/ Analyte: alpha-Thujone

CAS Number: 76231-76-0

Request Date: 7/11/2023

Request Time: 10:03:16

Lab: Battelle Columbus

Male

Treatment Group (mg/kg)

6.0 IV Plasma<sup>c,f</sup>

40 Gavage Plasma<sup>b,d,e</sup>

80 Gavage Plasma<sup>b,d,e</sup>

	6.0 IV Plasma <sup>c,f</sup>	40 Gavage Plasma <sup>b,d,e</sup>	80 Gavage Plasma <sup>b,d,e</sup>
C <sub>0min</sub> _pred (ng/mL)	1160 ± 160		
C <sub>max</sub> _pred (ng/mL)		185 ± 48	1060 ± 240
T <sub>max</sub> _pred (minute)		5.32 ± 3.71	7.33 ± 4.59
C <sub>max</sub> _obs (ng/mL)		238 ± 221	2100 ± 1000
T <sub>max</sub> _obs (minute)		5.00 ± 0.00	5.00 ± 0.00
Alpha Half-life (minute)	4.42 ± 0.54		
Beta Half-life (minute)	20.1 ± 3.4		
k <sub>01</sub> (minute <sup>-1</sup> )		0.522 ± 0.590	0.448 ± 0.413
k <sub>01</sub> Half-life (minute)		1.33 ± 1.50	1.55 ± 1.43
k <sub>10</sub> (minute <sup>-1</sup> )	0.130 ± 0.012	0.0403 ± 0.023	0.0194 ± 0.0081
k <sub>10</sub> Half-life (minute)	5.34 ± 0.51	17.2 ± 9.8	35.8 ± 14.9
k <sub>12</sub> (minute <sup>-1</sup> )	0.0199 ± 0.0059		
k <sub>21</sub> (minute <sup>-1</sup> )	0.0416 ± 0.0086		
Cl <sub>1</sub> (mL/min/kg)	671 ± 45		
Cl <sub>2</sub> (mL/min/kg)	103 ± 25		
Cl <sub>1_F</sub> (mL/min/kg)		7020 ± 2570	1270 ± 390
V <sub>1</sub> (mL/kg)	5170 ± 730		
V <sub>2</sub> (mL/kg)	2470 ± 410		
V <sub>1_F</sub> (mL/kg)		174000 ± 71000	65600 ± 19900
MRT (minute)	11.4 ± 0.8		
AUC <sub>0-T</sub> (ng mL <sup>-1</sup> min)		7660 ± .	40200 ± .
AUC <sub>inf</sub> _pred (ng*mL <sup>-1</sup> *min)	8940 ± 600	5700 ± 2100	63000 ± 19000

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Female

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6.0 IV Plasma<sup>c</sup>

40 Gavage Plasma<sup>b,d,e</sup>

80 Gavage Plasma<sup>b,d,e</sup>

	6.0 IV Plasma <sup>c</sup>	40 Gavage Plasma <sup>b,d,e</sup>	80 Gavage Plasma <sup>b,d,e</sup>
C_0min_pred (ng/mL)	837 ± 234		
Cmax_pred (ng/mL)		204 ± 37	683 ± 200
Tmax_pred (minute)		6.21 ± 1.89	6.16 ± 4.20
Cmax_obs (ng/g)		267 ± 76	1350 ± 890
Tmax_obs (minute)		5.00 ± 0.00	5.00 ± 0.00
Alpha Half-life (minute)	4.03 ± 0.97		
Beta Half-life (minute)	26.4 ± 29.9		
k01 (minute <sup>-1</sup> )		0.237 ± 0.239	0.407 ± 0.483
k01 Half-life (minute)		2.92 ± 2.95	1.7 ± 2.02
k10 (minute <sup>-1</sup> )	0.0145 ± 0.031	0.103 ± 0.079	0.0433 ± 0.0288
k10 Half-life (minute)	4.78 ± 1.02	6.72 ± 5.13	16.0 ± 10.6
k12 (minute <sup>-1</sup> )	0.0222 ± 0.0086		
k21 (minute <sup>-1</sup> )	0.0312 ± 0.0363		
Cl1 (mL/min/kg)	1040 ± 140		
Cl2 (mL/min/kg)	159 ± 57		
Cl1_F (mL/min/kg)		10700 ± 2400	3880 ± 1550
V1 (mL/kg)	7170 ± 2000		
V2 (mL/kg)	5100 ± 6160		
V1_F (mL/kg)		103000 ± 73000	89700 ± 44900
MRT (minute)	11.8 ± 7.1		
AUC_0-T (ng mL <sup>-1</sup> min)		3820 ± .	13400 ± .
AUCinf_pred (ng*mL <sup>-1</sup> *min)	5760 ± 780	3750 ± 830	20600 ± 8200

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Treatment Group (mg/kg)

6.0 IV Brain<sup>a</sup>

40 Gavage Brain<sup>a</sup>

80 Gavage Brain<sup>a</sup>

Cmax_obs (ng/g)	4030 ± 400	976 ± 1080	6180 ± 1450
Tmax_obs (minute)	5.00	8.67	9.67
Half-life (minute)	22.8	28.4	88.8
AUC_0-T (ng*g <sup>-1</sup> *min)	56200		
AUC_0-T (ng/g* min)		24800	103000
AUCinf_pred (ng*g <sup>-1</sup> *min)	56900		
AUCinf_pred (ng*mL <sup>-1</sup> min)		25100	105000

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Female

Treatment Group (mg/kg)

6.0 IV Brain<sup>a</sup>

40 Gavage Brain<sup>a</sup>

80 Gavage Brain<sup>a</sup>

	6.0 IV Brain <sup>a</sup>	40 Gavage Brain <sup>a</sup>	80 Gavage Brain <sup>a</sup>
Cmax_obs (ng/g)	3760 ± 920	1230 ± 470	4160 ± 1150
Tmax_obs (minute)	6.00	10	9.33
Half-life (minute)	6.39	12.9	33
AUC_0-T (ng*g <sup>-1</sup> *min)	128000		
AUC_0-T (ng/g* min)		16900	61600
AUCinf_pred (ng*g*min)		17000	62500
AUCinf_pred (ng*g <sup>-1</sup> * min)	129000		

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LEGEND

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MODELING SOFTWARE

WinNonlin Version 5.0.1

MODELING METHOD & BEST FIT MODEL

<sup>a</sup>WinNonlin, Version 5.0.1, Pharsight Corporation, Mountain View, CA, Noncompartmental Analysis (NCA)

<sup>b</sup>WinNonlin, Version 5.0.1, Pharsight Corporation, Mountain View, CA, one-compartment with first order absorption and elimination with  $1/Y_{hat}^2$  weighting (Model No. 3)

<sup>c</sup>WinNonlin, Version 5.0.1, Pharsight Corporation, Mountain View, CA, Two compartment model with bolus input and first order elimination with  $1/Y_{hat}^2$  weighting (Model No. 8)

<sup>d</sup>When there is no data or value (no attempt to determine a value), there is a blank space. If there is a value below detection, a period is used meaning that it was analyzed for but below detection.

EXCEPTION

<sup>e</sup>AUC 0-T standard error of the mean, SE, was ND, not detected.

<sup>f</sup>The 37.53 ng/mL concentration at 90 minutes was not used in modeling.

ANALYTE

Alpha-Thujone

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TK PARAMETERS

C\_0min\_pred = Fitted plasma concentration at time zero (IV only)

Cmax\_pred = Observed or Predicted Maximum plasma (or tissue) concentration

Tmax\_pred = Time at which Cmax predicted or observed occurs

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

Alpha Half-Life = Half-life for the alpha phase

Beta Half-Life = Half-life for the beta phase

k01 = Absorption rate constant,  $k_a$

k01 Half-life = Half-life of the absorption process to the central compartment

k10 = Elimination rate constant from the central compartment also  $k_e$  or  $k_{elim}$

k10 Half-life = Half-life for the elimination process from the central compartment

k12 = Distribution rate constant from first to second compartment

k21 = Distribution rate constant from second to first compartment

Cl1 = Clearance of central compartment,  $Cl_{app}$  or apparent clearance for intravenous groups

Cl2 = Clearance of the secondary compartment

Cl1\_F = Apparent clearance of the central compartment, also  $Cl_F$  for gavage groups in non-compartmental model

V1 = Volume of distribution of the central compartment, includes  $V_d$  and  $V$  volume of distribution,  $V_z$  apparent volume of distribution NCA,

$V_{app}$  apparent volume of distribution for intravenous studies

V2 = Volume of distribution for the peripheral compartment

V1\_F = Apparent volume of distribution for the central compartment includes  $V_{d,F}$ ,  $V_F$  for oral groups, and  $V_{c,F}$

MRT = Mean Residence Time

AUC\_0-T = Area under the plasma concentration versus time curve, AUC, from time  $t_i$  (initial) to  $t_f$  (final),  $AUC_{last}$

$AUC_{inf\_pred}$  = Area under the plasma concentration versus time curve, AUC, extrapolated to time equals infinity

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TK PARAMETERS PROTOCOL

ANALYSIS METHOD

Target times for blood and brain collection for the intravenous phase of the study were - male rats at 5, 10, 20, and 45 minutes, and 1, 2, 4, 6, 8, and 12 hours; female rats at 5, 10, 15, 30, and 45 minutes, and 1, 1.5, 2, 2.5, and 3 hours; and male and female mice at 2, 5, 7, 10, 15, 20, 30, and 45 minutes, and 1 and 1.5 hours. Target times for blood and brain collection for the gavage phase of the study were: male and female rats at 2, 5, 10, and 30 minutes, and 1.5, 3, 6, and 12 hours; and male and female mice at 2, 5, 10, 20, and 40 minutes, and 1.5 hours, 2 hours (40 mg/kg female mice only), 3 hours, 4 hours (80 mg/kg female mice only), 5 hours (40 mg/kg male mice only), and 6 hours (80 mg/kg male mice only).

TK\_INTRAVENOUS PLASMA

6.0 mg/kg Male and Female

The test article had a purity of 70 percent alpha-Thujone and 11 percent beta-Thujone. Thirty animals/species/sex/compound/dosage group (excluding replacements) were given a single IV injection of a,b-thujone in Cremophor-ethanol-water (1,1,8) using a catheter surgically implanted by the animal supplier into the jugular vein. Dosages were administered at a volume of 2 mL/kg (rats) and 4 mL/kg (mice). Animals were weighed the morning of dosing for calculation of the dosing volume. The dosing volume was administered as a bolus push. Dosed 6/25-27/02.

TK\_GAVAGE PLASMA

40 mg/kg, 80 mg/kg Male and Female

The test article had a purity of 70 percent alpha-Thujone and 11 percent beta-Thujone. Twenty-four animals/species/sex/compound/dosage group (excluding replacements) were given a single oral gavage administration of a,b-thujone in 0.5 percent aqueous methylcellulose. Doses were administered at a volume of 5 mL/kg (rats) and 10 mL/kg (mice). Non-fasted animals were given a single gavage administration. Dosed 12/11-13/02.

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TK PARAMETERS PROTOCOL (cont'd)

TK\_INTRAVENTOUS BRAIN

6.0 mg/kg Male and Female

The test article had a purity of 70 percent alpha-Thujone and 11 percent beta-Thujone. Thirty animals/species/sex/compound/dosage group (excluding replacements) were given a single IV injection of a,b-thujone in Cremophor-ethanol-water (1,1,8) using a catheter surgically implanted by the animal supplier into the jugular vein. Dosages were administered at a volume of 2 mL/kg (rats) and 4 mL/kg (mice). Animals were weighed the morning of dosing for calculation of the dosing volume. The dosing volume was administered as a bolus push. Dosed 6/25-27/02.

TK\_GAVAGE BRAIN

40 mg/kg, 80 mg/kg Male and Female

The test article had a purity of 70 percent alpha-Thujone and 11 percent beta-Thujone. Twenty-four animals/species/sex/compound/dosage group (excluding replacements) were given a single oral gavage administration of a,b-thujone in 0.5 percent aqueous methylcellulose. Doses were administered at a volume of 5 mL/kg (rats) and 10 mL/kg (mice). Non-fasted animals were given a single gavage administration. Dosed 12/11-13/02.