

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound/Analyte: Alpha-Pinene/Alpha-Pinene
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

Male Blood

Treatment Group (ppm)

50 ^a

100 ^a

Cmax_pred (ng/mL)	867 ± 87.2	3310 ± 437
Alpha (hour ⁻¹)	0.492 ± 0.136	1.07 ± 0.382
Alpha_Half-life (hour)	1.41 ± 0.389	0.647 ± 0.231
Beta (hour ⁻¹)	0.0522 ± 0.127	0.112 ± 0.146
Beta_Half-life (hour)	13.3 ± 32.3	6.18 ± 8.07
k10 (hour ⁻¹)	0.409 ± 0.110	0.735 ± 0.201
k10_Half-life (hour)	1.70 ± 0.456	0.943 ± 0.258
k12 (hour ⁻¹)	0.0729 ± 0.0981	0.285 ± 0.286
k21 (hour ⁻¹)	0.0628 ± 0.153	0.163 ± 0.239
Cl1 (mL/h/kg)	55200 ± 13400	517000 ± 12900
Cl2 (mL/h/kg)	9840 ± 13000	20100 ± 19100
V1 (mL/kg)	135000 ± 13600	70400 ± 9310
V2 (mL/kg)	157000 ± 457000	123000 ± 165000
Vss (mL/kg)	292000 ± 458000	193000 ± 166000
AUC_0-T (h*ng/mL)	2120 ± 516	4510 ± 1120

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound/Analyte: Alpha-Pinene/Alpha-Pinene
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

Female Blood

Treatment Group (ppm)

50^a

100^a

Cmax_pred (ng/mL)	765 ± 73.4	3000 ± 273
Alpha (hour ⁻¹)	0.484 ± 0.130	0.932 ± 0.217
Alpha_Half-life (hour)	1.43 ± 0.386	0.744 ± 0.173
Beta (hour ⁻¹)	0.0358 ± 0.0768	0.0764 ± 0.0685
Beta_Half-life (hour)	19.4 ± 41.6	9.08 ± 8.13
k10 (hour ⁻¹)	0.343 ± 0.130	0.578 ± 0.133
k10_Half-life (hour)	2.02 ± 0.770	1.20 ± 0.275
k12 (hour ⁻¹)	0.126 ± 0.124	0.307 ± 0.171
k21 (hour ⁻¹)	0.0505 ± 0.102	0.123 ± 0.113
Cl1 (mL/h/kg)	51900 ± 18700	4500 ± 9350
Cl2 (mL/h/kg)	19100 ± 18500	23700 ± 12400
V1 (mL/kg)	152000 ± 14600	77000 ± 7020
V2 (mL/kg)	379000 ± 957000	192000 ± 186000
Vss (mL/kg)	531000 ± 959000	269000 ± 187000
AUC_0-T (h*ng/mL)	2230 ± 805	5190 ± 1090

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound/Analyte: Alpha-Pinene/Alpha-Pinene Oxide
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

Male Blood

Treatment Group (ppm)

50^b

100^b

C_0min_pred (ng/mL)	73.4	107
Cmax_obs (ng/mL)	52.4	97.7
Tmax_obs (hour)	0.0830	0.0830
Lambda_z (hour ⁻¹)	0.0900	0.0876
Half-life	7.70	7.91
AUCinf_pred (h*ng/mL)	222	207

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound/Analyte: Alpha-Pinene/Alpha-Pinene oxide
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

Female Blood

Treatment Group (ppm)

50^b

100^b

C_0min_pred (ng/mL)	93.9	136
Cmax_obs (ng/mL)	65.8	102
Tmax_obs (hour)	0.0830	0.0830
Lambda_z (hour ⁻¹)	0.531	0.293
Half-Life (hour)	1.31	2.37
AUCinf_pred (h*ng/mL)	79.8	170

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound/Analyte: Alpha-Pinene/Alpha-Pinene
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

Male Mammary

Treatment Group (ppm)

50 °

100 °

Cmax_obs (ng/g)	131000	345000
Tmax_obs (hour)	0.333	0.500
Lambda_z (hour ⁻¹)	0.0890	0.131
Half_life (hour)	7.79	5.28
Cl (g/h/kg)	255	177
AUC_inf(h*ng/g)	459000	1320000

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound/Analyte: Alpha-Pinene/Alpha-Pinene
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

Female Mammary

Treatment Group (ppm)

50 °

100 °

Cmax_obs (ng/g)	105000	291000
Tmax_obs (hour)	1.00	0.500
Lambda_z (hour ⁻¹)	0.0886	0.119
Half_life (hour)	7.83	5.83
Cl (g/h/kg)	193	159
AUC_inf(h*ng/g)	601000	1450000

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound/Analyte: Alpha-Pinene/Alpha-Pinene
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

Male Mammary (lipid adjusted)

Treatment Group (ppm)

50^d

100^d

Cmax_obs (ng/g lipid)	665000	2850000
Tmax_obs (hour)	0.500	0.500
Lambda_z (hour ⁻¹)	0.0906	0.138
Half-life (hour)	7.65	5.01
Cl (g/h/kg)	61.7	34.6
AUCinf_pred (h*ng/g)	1900000	6740000

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound/Analyte: Alpha-Pinene/Alpha-Pinene
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

Female Mammary (lipid adjusted)

Treatment Group (ppm)

50^d

100^d

Cmax_obs (ng/g lipid)	643000	1500000
Tmax_obs (hour)	2.00	0.167
Lambda_z (hour ⁻¹)	0.0779	0.120
Half-life (hour)	8.90	5.77
Cl (g/h/kg)	34.7	25.5
AUCinf_pred (h*ng/g)	3340000	9050000

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound/Analyte: Alpha-Pinene/Alpha-Pinene oxide
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

Male Mammary

Treatment Group (ppm)

50^e

100^e

Cmax_obs (ng/g)	2760	4430
Tmax_obs (hour)	0.167	0.0830
Lambda_z (hour ⁻¹)	0.231	0.167
Half-life (hour)	3.00	4.15
AUCinf_pred (h*ng/g)	6770	15100

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound/Analyte: Alpha-Pinene/Alpha-Pinene oxide
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

Female Mammary

Treatment Group (ppm)

50^e

100^e

Cmax_obs (ng/g)	2630	3440
Tmax_obs (hour)	0.250	0.167
Lambda_z (hour ⁻¹)	0.306	0.229
Half-life (hour)	2.27	3.03
AUCinf_pred (h*ng/g)	5860	10100

Toxicokinetics Data Summary

Request Date: 10/27/2020

Route: Inhalation

Compound/Analyte: Alpha-Pinene/Alpha-Pinene oxide

Request Time: 2:30:16

Species/Strain: Mice/B6C3F1/N

CAS Number: 80-56-8

Lab: RTI

Male Mammary (lipid adjusted)

Treatment Group (ppm)

	50^f	100^f
Cmax_obs (ng/g lipid)	8130	9130
Tmax_obs (hour)	0.167	0.0830
Lambda_z (hour ⁻¹)	0.240	0.211
Half-life (hour)	2.89	3.28
AUCinf_pred (h*ng/g)	15500	36200

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound/Analyte: Alpha-Pinene/Alpha-Pinene oxide
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

Female Mammary (lipid adjusted)

Treatment Group (ppm)

50^f

100^f

Cmax_obs (ng/g)	5000	6290
Tmax_obs (hour)	0.0830	0.167
Lambda_z (hour ⁻¹)	0.294	0.229
Half-life (hour)	2.36	3.03
AUCinf_pred (h*ng/g)	12900	19300

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound: Alpha-Pinene
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

LEGEND

MODELING METHOD & BEST FIT MODEL

- ^a Phoenix WinNonlin (Version 6.4), two-compartment
- ^b Phoenix WinNonlin (Version 6.4), noncompartmental, Because APO is a metabolite of AP, no parameters calculated from exposure (i.e., clearance, volume, C_{max}/D, AUC/D) are reported. AUC_{inf}-pred is actually AUC_{inf}_obs
- ^c Phoenix WinNonlin (Version 6.4), noncompartmental, Non-lipid adjusted data, concentration is expressed as ng/g mammary tissue. AUC_{inf}-pred is actually AUC_{inf}_obs
- ^d Phoenix WinNonlin (Version 6.4), noncompartmental, Concentration (ng/g) for lipid adjusted data is expressed as ng/g lipid in mammary tissue. AUC_{inf}-pred is actually AUC_{inf}_obs
- ^e Phoenix WinNonlin (Version 6.4), noncompartmental, Non-lipid adjusted data, concentration is expressed as ng/g mammary tissue. Because APO is a metabolite of AP, no parameters calculated from exposure (i.e., clearance, volume, C_{max}/D, AUC/D) are reported. AUC_{inf}-pred is actually AUC_{inf}_obs.
- ^f Phoenix WinNonlin (Version 6.4), noncompartmental, Concentration (ng/g) for lipid adjusted data is expressed as ng/g lipid in mammary tissue. Because APO is a metabolite of AP, no parameters calculated from exposure (i.e., clearance, volume, C_{max}/D, AUC/D) are reported. AUC_{inf}-pred is actually AUC_{inf}_obs.

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound: Alpha-Pinene
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

ANALYTE

Alpha-Pinene, Alpha-Pinene oxide

TK PARAMETERS

C_{0min_pred} = Fitted plasma concentration at time zero (IV only)
C_{max} = Observed or Predicted Maximum plasma (or tissue) concentration
T_{max} = Time at which C_{max} predicted or observed occurs
Lambda_z = Non-compartmental analysis (NCA) terminal elimination rate constant, NCA ke or kelim
Half_{life} = Lambda z Half life, t_{1/2}, the terminal elimination half-life based on non-compartmental analysis
Alpha = Hybrid rate constant of the alpha phase
Alpha_{Half-life} = Half-life for the alpha phase
Beta = Hybrid rate constant of the beta phase
Beta_{Half-life} = Half-life for the beta phase
k₁₀ = Elimination rate constant from the central compartment also ke or kelim
k_{10 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, Clapp or apparent clearance for intravenous groups
Cl₂ = Clearance of the secondary compartment
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
V_{ss} = Volume of distribution at steady state
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} = Area under the plasma concentration versus time curve, AUC, extrapolated to time equals infinity

Experiment Number: K03014
Route: Inhalation
Species/Strain: Mice/B6C3F1/N

Toxicokinetics Data Summary
Compound: Alpha-Pinene
CAS Number: 80-56-8

Request Date: 10/27/2020
Request Time: 2:30:16
Lab: RTI

TK PARAMETERS PROTOCOL

Mouse Blood and Mammary Tissue

50 ppm Mice Male, 50 ppm Mice Female, 100 ppm Mice Male, 100 ppm Mice Female

Male and female B6C3F1/N mice were exposed by whole body inhalation for 6 hours plus T90 per day for 7 consecutive days. Dose was calculated for individual animals and the average inhaled dose for the groups in mg/kg (117 and 233 mg/kg for male mice at 50 and 100 ppm, respectively; 116 and 231 mg/kg for female mice at 50 and 100 ppm, respectively). The mg/kg doses are the estimated theoretical inhaled doses and not the doses absorbed from the respiratory tract. Both males and females were 62 days old at first exposure. Body weights ranged 22.6 to 27.4 g for males and 18.1-21.0 females at randomization. N = 39 animals/sex/group. Blood and mammary tissues were analyzed for alpha pinene and its metabolite alpha pinene oxide. Animals were fed irradiated NTP-2000 wafer feed available ad libitum, except during exposure. Water was available ad libitum. For the 0 ppm group (n=3), the mice were humanely terminated prior to the initiation of exposure on Day 0 to measure background concentrations of alpha pinene (AP)/alpha pinene oxide (APO) in blood and tissue (prestudy blood and tissue concentrations). Beginning on Day 6, whole blood samples were collected from three animals/sex from the 50 and 100 ppm groups via cardiac puncture while under 70% CO₂/30% O₂ anesthesia at the following time points: pre-exposure (0 minutes) and post-exposure times of 5, 10, 15, 20, and 30 minutes, and 1, 2, 4, 8, 12, 24, and 48 hours. Following terminal blood collection, mammary glands were collected. All samples were stored at ultracold temperatures. Alpha pinene and alpha pinene oxide concentrations in blood and mammary glands were determined using validated analytical methods. All variance listed for the parameters is standard error (\pm SE).