

Exploration of Multiple Treatments on the Metabolic Efficiency of the Mitochondria

Dom Fenoglio, Brian Kim, Kathleen Zhang

1 Background

2 Exploratory Data Analysis

3 Modeling

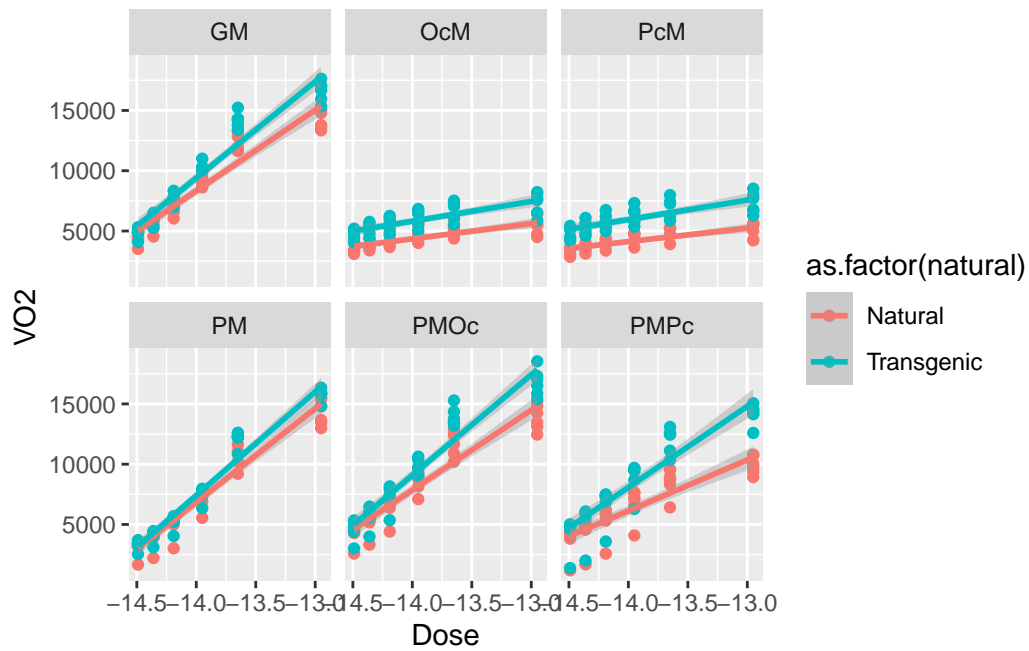
4 Analysis

5 Conclusion and Future Work

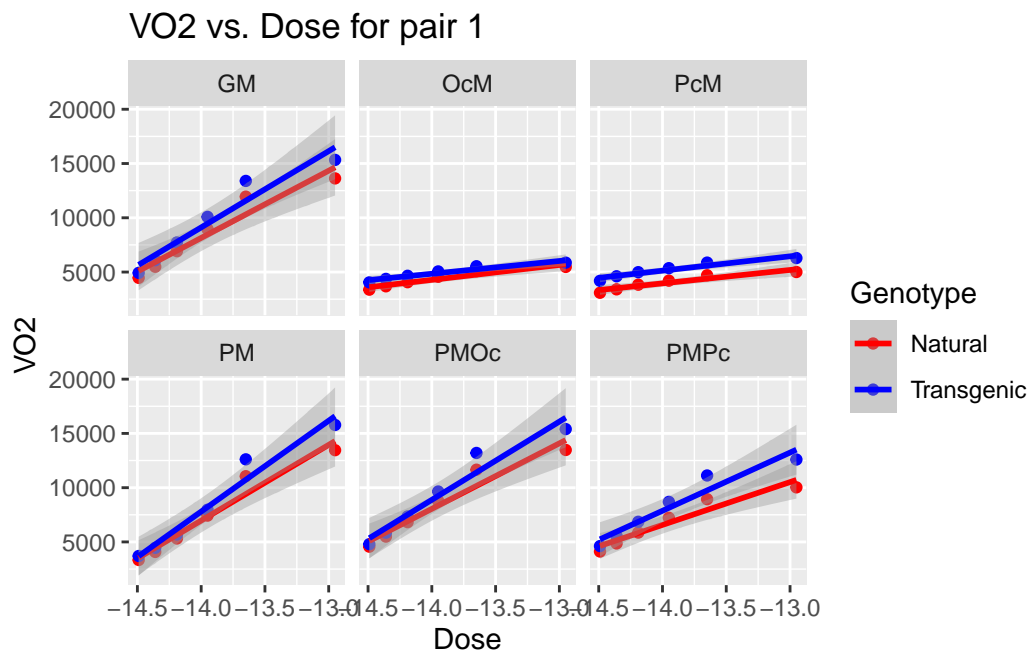
6 Appendix

Exploratory Data Analysis

```
`geom_smooth()` using formula = 'y ~ x'
```

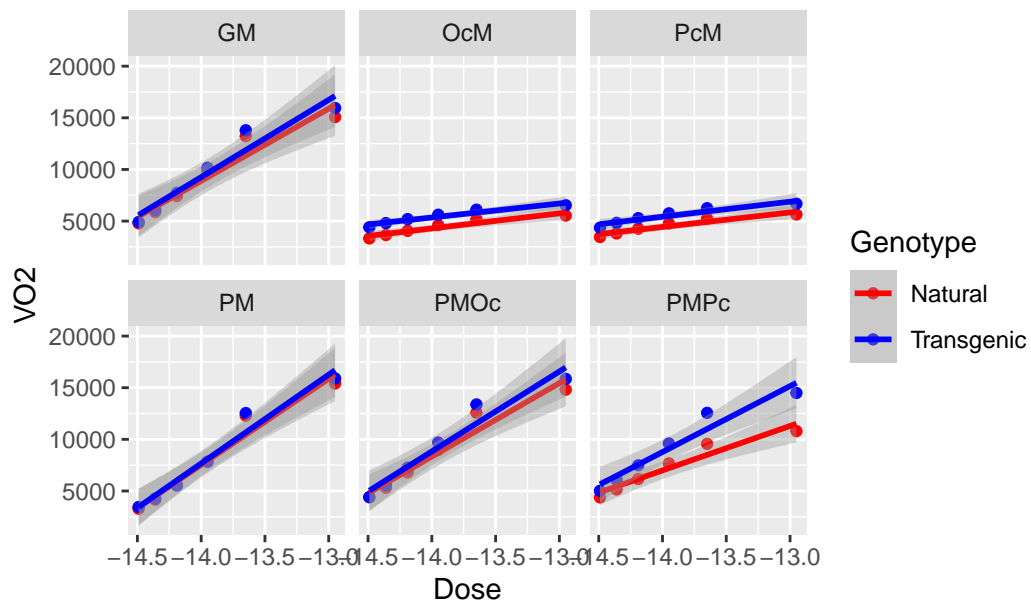


``geom_smooth()`` using formula = `'y ~ x'`



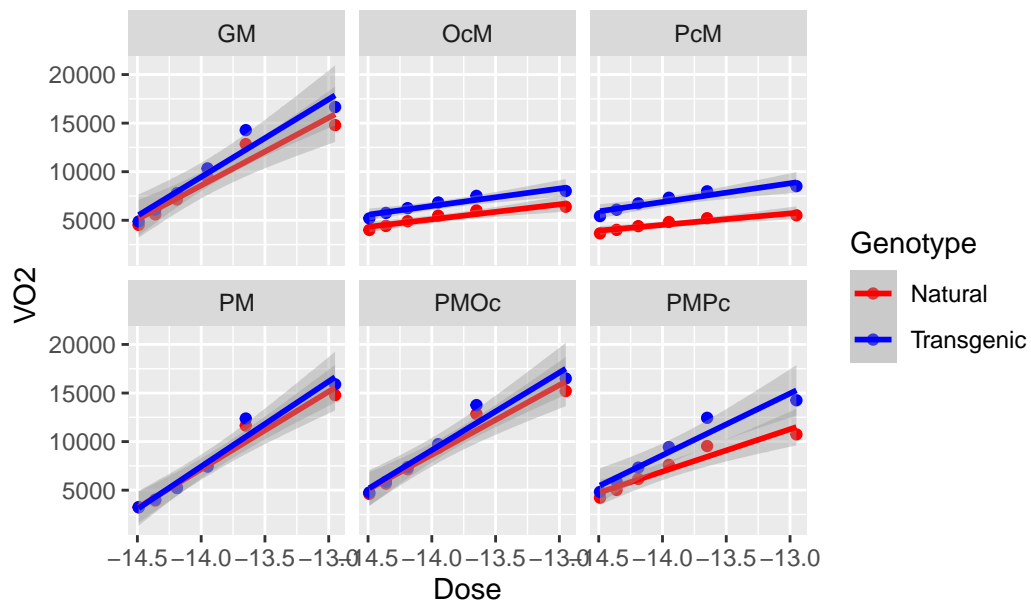
``geom_smooth()`` using formula = `'y ~ x'`

VO2 vs. Dose for pair 2



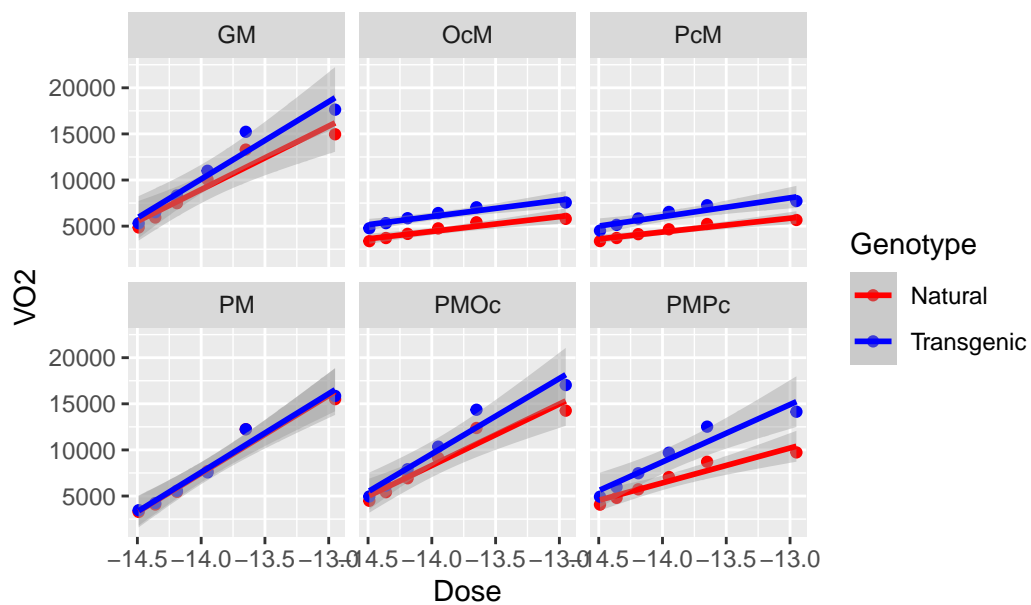
``geom_smooth()`` using formula = `'y ~ x'`

VO2 vs. Dose for pair 3



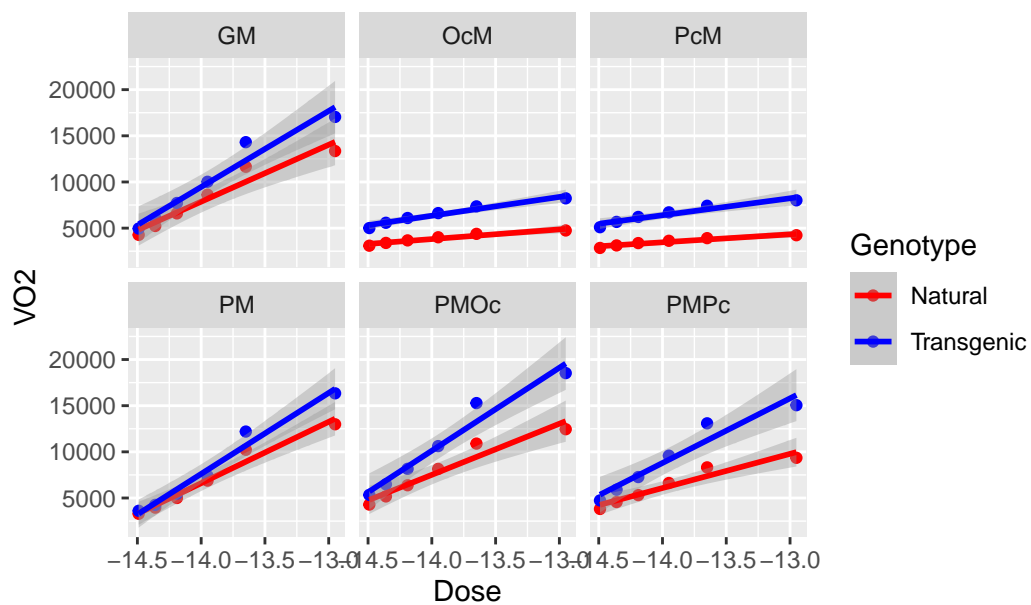
``geom_smooth()`` using formula = `'y ~ x'`

VO2 vs. Dose for pair 4



``geom_smooth()`` using formula = 'y ~ x'

VO2 vs. Dose for pair 5



``geom_smooth()`` using formula = 'y ~ x'

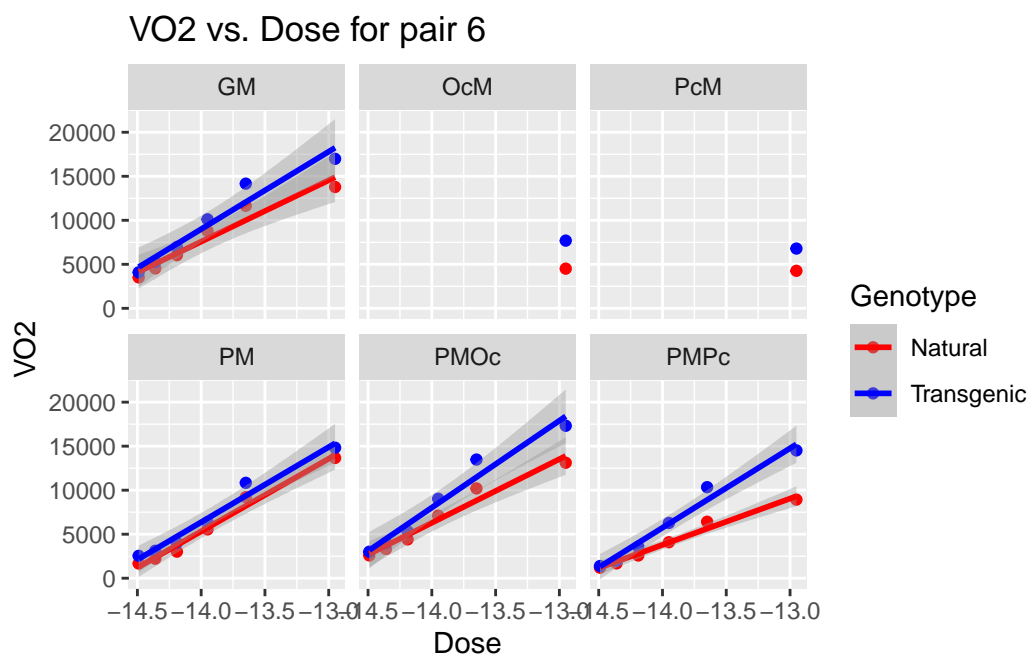


Table 1: Model comparison using AIC and Adjusted R^2

Model	df	AIC	Adjusted_R2
Amino Acids	8	7439.921	0.721
Substrate	9	7386.290	0.755

Modeling

Call:

```
lm(formula = V02 ~ Substrate:natural + Dose:natural:Substrate,
    data = data)
```

Residuals:

Min	1Q	Median	3Q	Max
-3254.1	-370.8	186.2	536.4	1727.3

Coefficients: (1 not defined because of singularities)

	Estimate	Std. Error	t value	Pr(> t)
(Intercept)	138201.0	7230.7	19.113	< 2e-16 ***
SubstrateGM:naturalNatural	5077.4	10225.7	0.497	0.619863
SubstrateOcM:naturalNatural	-104126.4	10724.8	-9.709	< 2e-16 ***

SubstratePcM:naturalNatural	-107896.1	10724.8	-10.060	< 2e-16	***
SubstratePM:naturalNatural	3735.0	10225.7	0.365	0.715163	
SubstratePM0c:naturalNatural	-2949.2	10225.7	-0.288	0.773223	
SubstratePMPc:naturalNatural	-49273.5	10225.7	-4.819	2.25e-06	***
SubstrateGM:naturalTransgenic	27374.7	10225.7	2.677	0.007815	**
SubstrateOcM:naturalTransgenic	-99396.5	10724.8	-9.268	< 2e-16	***
SubstratePcM:naturalTransgenic	-95812.6	10724.8	-8.934	< 2e-16	***
SubstratePM:naturalTransgenic	14409.0	10225.7	1.409	0.159790	
SubstratePM0c:naturalTransgenic	27257.2	10225.7	2.666	0.008081	**
SubstratePMPc:naturalTransgenic	NA	NA	NA	NA	
SubstrateGM:naturalNatural:Dose	9596.5	511.7	18.755	< 2e-16	***
SubstrateOcM:naturalNatural:Dose	2110.8	560.5	3.766	0.000198	***
SubstratePcM:naturalNatural:Dose	1859.4	560.5	3.317	0.001015	**
SubstratePM:naturalNatural:Dose	9622.6	511.7	18.806	< 2e-16	***
SubstratePM0c:naturalNatural:Dose	9064.0	511.7	17.714	< 2e-16	***
SubstratePMPc:naturalNatural:Dose	5889.9	511.7	11.511	< 2e-16	***
SubstrateGM:naturalTransgenic:Dose	11110.1	511.7	21.713	< 2e-16	***
SubstrateOcM:naturalTransgenic:Dose	2346.2	560.5	4.186	3.69e-05	***
SubstratePcM:naturalTransgenic:Dose	2588.6	560.5	4.618	5.64e-06	***
SubstratePM:naturalTransgenic:Dose	10345.9	511.7	20.219	< 2e-16	***
SubstratePM0c:naturalTransgenic:Dose	11128.3	511.7	21.748	< 2e-16	***
SubstratePMPc:naturalTransgenic:Dose	9257.5	511.7	18.092	< 2e-16	***

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 839.7 on 316 degrees of freedom

(20 observations deleted due to missingness)

Multiple R-squared: 0.9233, Adjusted R-squared: 0.9177

F-statistic: 165.4 on 23 and 316 DF, p-value: < 2.2e-16

Call:

lm(formula = V02 ~ natural * Dose * Substrate, data = data)

Residuals:

Min	1Q	Median	3Q	Max
-3254.1	-370.8	186.2	536.4	1727.3

Coefficients:

	Estimate	Std. Error	t value	Pr(> t)
(Intercept)	143278.42	7230.68	19.815	< 2e-16 ***
naturalTransgenic	22297.32	10225.72	2.181	0.0300 *

Dose	9596.47	511.68	18.755	< 2e-16	***
SubstrateOcM	-109203.79	10724.83	-10.182	< 2e-16	***
SubstratePcM	-112973.48	10724.83	-10.534	< 2e-16	***
SubstratePM	-1342.39	10225.72	-0.131	0.8956	
SubstratePMOc	-8026.61	10225.72	-0.785	0.4331	
SubstratePMPc	-54350.93	10225.72	-5.315	2.02e-07	***
naturalTransgenic:Dose	1513.62	723.63	2.092	0.0373	*
naturalTransgenic:SubstrateOcM	-17567.40	15167.20	-1.158	0.2476	
naturalTransgenic:SubstratePcM	-10213.84	15167.20	-0.673	0.5012	
naturalTransgenic:SubstratePM	-11623.31	14461.35	-0.804	0.4221	
naturalTransgenic:SubstratePMOc	7909.05	14461.35	0.547	0.5848	
naturalTransgenic:SubstratePMPc	26976.20	14461.35	1.865	0.0631	.
Dose:SubstrateOcM	-7485.71	758.95	-9.863	< 2e-16	***
Dose:SubstratePcM	-7737.11	758.95	-10.195	< 2e-16	***
Dose:SubstratePM	26.09	723.63	0.036	0.9713	
Dose:SubstratePMOc	-532.46	723.63	-0.736	0.4624	
Dose:SubstratePMPc	-3706.61	723.63	-5.122	5.26e-07	***
naturalTransgenic:Dose:SubstrateOcM	-1278.14	1073.31	-1.191	0.2346	
naturalTransgenic:Dose:SubstratePcM	-784.42	1073.31	-0.731	0.4654	
naturalTransgenic:Dose:SubstratePM	-790.30	1023.37	-0.772	0.4405	
naturalTransgenic:Dose:SubstratePMOc	550.68	1023.37	0.538	0.5909	
naturalTransgenic:Dose:SubstratePMPc	1854.02	1023.37	1.812	0.0710	.

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 839.7 on 316 degrees of freedom

(20 observations deleted due to missingness)

Multiple R-squared: 0.9233, Adjusted R-squared: 0.9177

F-statistic: 165.4 on 23 and 316 DF, p-value: < 2.2e-16

Call:

lm(formula = V02 ~ Substrate - 1, data = data)

Residuals:

Min	1Q	Median	3Q	Max
-5366.1	-1939.5	-445.7	1327.1	7567.6

Coefficients:

	Estimate	Std. Error	t value	Pr(> t)
SubstrateGM	8155.9	346.4	23.55	<2e-16 ***
SubstrateOcM	4955.3	379.4	13.06	<2e-16 ***

SubstratePcM	4926.5	379.4	12.98	<2e-16 ***
SubstratePM	6215.9	346.4	17.95	<2e-16 ***
SubstratePMOc	7716.4	346.4	22.28	<2e-16 ***
SubstratePMPc	6563.2	346.4	18.95	<2e-16 ***

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 2683 on 334 degrees of freedom

(20 observations deleted due to missingness)

Multiple R-squared: 0.8611, Adjusted R-squared: 0.8586

F-statistic: 345.1 on 6 and 334 DF, p-value: < 2.2e-16

Linear mixed model fit by REML. t-tests use Satterthwaite's method [lmerModLmerTest]

Formula: V02 ~ natural * Dose * Substrate + (1 | pair)

Data: data

REML criterion at convergence: 4979.4

Scaled residuals:

Min	1Q	Median	3Q	Max
-3.09075	-0.64782	0.02403	0.63668	3.05598

Random effects:

Groups	Name	Variance	Std.Dev.
pair	(Intercept)	582983	763.5
	Residual	322707	568.1

Number of obs: 340, groups: pair, 6

Fixed effects:

	Estimate	Std. Error	df	t value
(Intercept)	143278.42	4901.42	313.21	29.232
naturalTransgenic	22297.32	6917.62	310.97	3.223
Dose	9596.47	346.15	310.97	27.724
SubstrateOcM	-109503.46	7255.28	310.97	-15.093
SubstratePcM	-113273.15	7255.28	310.97	-15.613
SubstratePM	-1342.39	6917.62	310.97	-0.194
SubstratePMOc	-8026.61	6917.62	310.97	-1.160
SubstratePMPc	-54350.93	6917.62	310.97	-7.857
naturalTransgenic:Dose	1513.62	489.53	310.97	3.092
naturalTransgenic:SubstrateOcM	-17567.40	10260.49	310.97	-1.712
naturalTransgenic:SubstratePcM	-10213.84	10260.49	310.97	-0.995

naturalTransgenic:SubstratePM	-11623.31	9782.99	310.97	-1.188
naturalTransgenic:SubstratePM0c	7909.05	9782.99	310.97	0.808
naturalTransgenic:SubstratePMPc	26976.20	9782.99	310.97	2.757
Dose:Substrate0cM	-7485.71	513.42	310.97	-14.580
Dose:SubstratePcM	-7737.11	513.42	310.97	-15.070
Dose:SubstratePM	26.09	489.53	310.97	0.053
Dose:SubstratePM0c	-532.46	489.53	310.97	-1.088
Dose:SubstratePMPc	-3706.61	489.53	310.97	-7.572
naturalTransgenic:Dose:Substrate0cM	-1278.14	726.09	310.97	-1.760
naturalTransgenic:Dose:SubstratePcM	-784.42	726.09	310.97	-1.080
naturalTransgenic:Dose:SubstratePM	-790.30	692.30	310.97	-1.142
naturalTransgenic:Dose:SubstratePM0c	550.68	692.30	310.97	0.795
naturalTransgenic:Dose:SubstratePMPc	1854.02	692.30	310.97	2.678

Pr(>|t|)

(Intercept)	< 2e-16 ***
naturalTransgenic	0.00140 **
Dose	< 2e-16 ***
Substrate0cM	< 2e-16 ***
SubstratePcM	< 2e-16 ***
SubstratePM	0.84626
SubstratePM0c	0.24681
SubstratePMPc	6.48e-14 ***
naturalTransgenic:Dose	0.00217 **
naturalTransgenic:Substrate0cM	0.08787 .
naturalTransgenic:SubstratePcM	0.32029
naturalTransgenic:SubstratePM	0.23570
naturalTransgenic:SubstratePM0c	0.41945
naturalTransgenic:SubstratePMPc	0.00617 **
Dose:Substrate0cM	< 2e-16 ***
Dose:SubstratePcM	< 2e-16 ***
Dose:SubstratePM	0.95753
Dose:SubstratePM0c	0.27757
Dose:SubstratePMPc	4.23e-13 ***
naturalTransgenic:Dose:Substrate0cM	0.07934 .
naturalTransgenic:Dose:SubstratePcM	0.28083
naturalTransgenic:Dose:SubstratePM	0.25451
naturalTransgenic:Dose:SubstratePM0c	0.42697
naturalTransgenic:Dose:SubstratePMPc	0.00780 **

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Correlation matrix not shown by default, as $p = 24 > 12$.
 Use `print(x, correlation=TRUE)` or
`vcov(x)` if you need it

Type III Analysis of Variance Table with Satterthwaite's method

	Sum Sq	Mean Sq	NumDF	DenDF	F value	Pr(>F)
natural	17606539	17606539	1	310.97	54.5589	1.397e-12
Dose	1517357360	1517357360	1	310.97	4701.9667	< 2.2e-16
Substrate	380517369	76103474	5	310.97	235.8284	< 2.2e-16
natural:Dose	15683854	15683854	1	310.97	48.6009	1.886e-11
natural:Substrate	8560933	1712187	5	310.97	5.3057	0.0001077
Dose:Substrate	362539320	72507864	5	310.97	224.6864	< 2.2e-16
natural:Dose:Substrate	8399558	1679912	5	310.97	5.2057	0.0001323

```

natural      ***
Dose         ***
Substrate    ***
natural:Dose ***
natural:Substrate ***
Dose:Substrate ***
natural:Dose:Substrate ***

```

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

refitting model(s) with ML (instead of REML)

Data: data

Models:

lmm2: V02 ~ Dose * Substrate + (1 | pair)

lmm1: V02 ~ natural * Dose * Substrate + (1 | pair)

	npar	AIC	BIC	logLik	deviance	Chisq	Df	Pr(>Chisq)
lmm2	14	5632.8	5686.4	-2802.4	5604.8			
lmm1	26	5332.5	5432.0	-2640.2	5280.5	324.39	12	< 2.2e-16 ***

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Likelihood ratio test

Model 1: V02 ~ natural * Dose * Substrate + (1 | pair)

Model 2: V02 ~ Dose * Substrate + (1 | pair)

#Df	LogLik	Df	Chisq	Pr(>Chisq)
-----	--------	----	-------	------------

```

1 26 -2489.7
2 14 -2724.9 -12 470.45 < 2.2e-16 ***
---
Signif. codes:  0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

boundary (singular) fit: see help('isSingular')

# R2 for Mixed Models

Conditional R2: 0.965
Marginal R2: 0.901

boundary (singular) fit: see help('isSingular')

# Intraclass Correlation Coefficient

Adjusted ICC: 0.644
Unadjusted ICC: 0.064

Call:
lm(formula = V02 ~ natural * Dose * Substrate, data = data)

Residuals:
    Min       1Q   Median       3Q      Max
-3254.1  -370.8   186.2   536.4  1727.3

Coefficients:
                                Estimate Std. Error t value Pr(>|t|)
(Intercept)                   143278.42    7230.68  19.815 < 2e-16 ***
naturalTransgenic              22297.32   10225.72   2.181  0.0300 *
Dose                          9596.47     511.68  18.755 < 2e-16 ***
SubstrateOcM                  -109203.79   10724.83 -10.182 < 2e-16 ***
SubstratePcM                  -112973.48   10724.83 -10.534 < 2e-16 ***
SubstratePM                   -1342.39    10225.72  -0.131  0.8956
SubstratePMOc                 -8026.61    10225.72  -0.785  0.4331
SubstratePMPc                 -54350.93   10225.72  -5.315 2.02e-07 ***
naturalTransgenic:Dose         1513.62     723.63   2.092  0.0373 *
naturalTransgenic:SubstrateOcM -17567.40   15167.20  -1.158  0.2476
naturalTransgenic:SubstratePcM -10213.84   15167.20  -0.673  0.5012
naturalTransgenic:SubstratePM  -11623.31   14461.35  -0.804  0.4221

```

naturalTransgenic:SubstratePM0c	7909.05	14461.35	0.547	0.5848	
naturalTransgenic:SubstratePMPc	26976.20	14461.35	1.865	0.0631	.
Dose:Substrate0cM	-7485.71	758.95	-9.863	< 2e-16	***
Dose:SubstratePcM	-7737.11	758.95	-10.195	< 2e-16	***
Dose:SubstratePM	26.09	723.63	0.036	0.9713	
Dose:SubstratePM0c	-532.46	723.63	-0.736	0.4624	
Dose:SubstratePMPc	-3706.61	723.63	-5.122	5.26e-07	***
naturalTransgenic:Dose:Substrate0cM	-1278.14	1073.31	-1.191	0.2346	
naturalTransgenic:Dose:SubstratePcM	-784.42	1073.31	-0.731	0.4654	
naturalTransgenic:Dose:SubstratePM	-790.30	1023.37	-0.772	0.4405	
naturalTransgenic:Dose:SubstratePM0c	550.68	1023.37	0.538	0.5909	
naturalTransgenic:Dose:SubstratePMPc	1854.02	1023.37	1.812	0.0710	.

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 839.7 on 316 degrees of freedom

(20 observations deleted due to missingness)

Multiple R-squared: 0.9233, Adjusted R-squared: 0.9177

F-statistic: 165.4 on 23 and 316 DF, p-value: < 2.2e-16

Warning in modelUpdate(objects[[i - 1]], objects[[i]]): original model was of class "lmerModLmerTest", updated model is of class "lm"

Likelihood ratio test

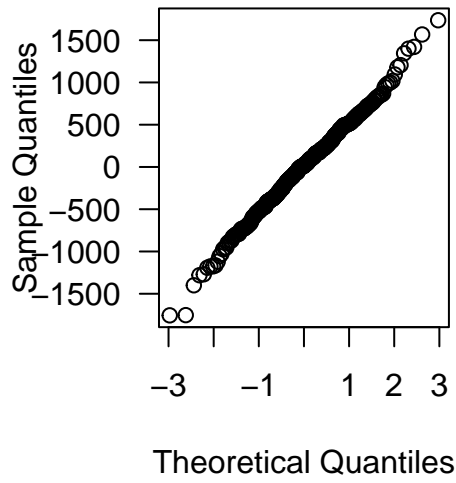
Model 1: V02 ~ natural * Dose * Substrate + (1 | pair)

Model 2: V02 ~ natural * Dose * Substrate

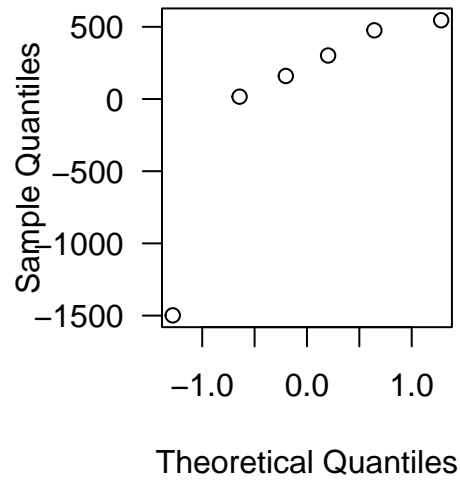
	#Df	LogLik	Df	Chisq	Pr(>Chisq)
1	26	-2489.7			
2	25	-2759.2	-1	539.1	< 2.2e-16 ***

Signif. codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

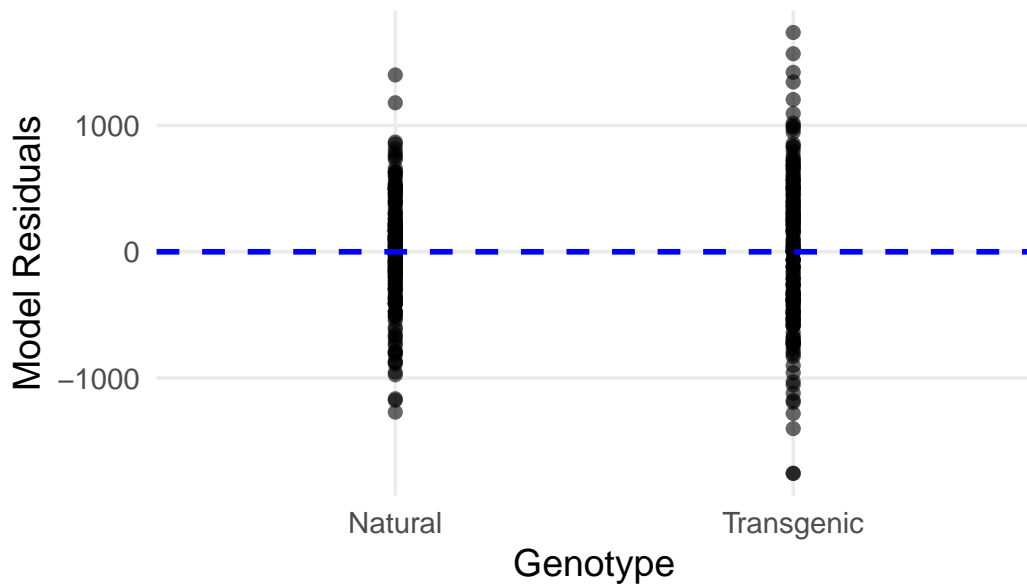
Residuals



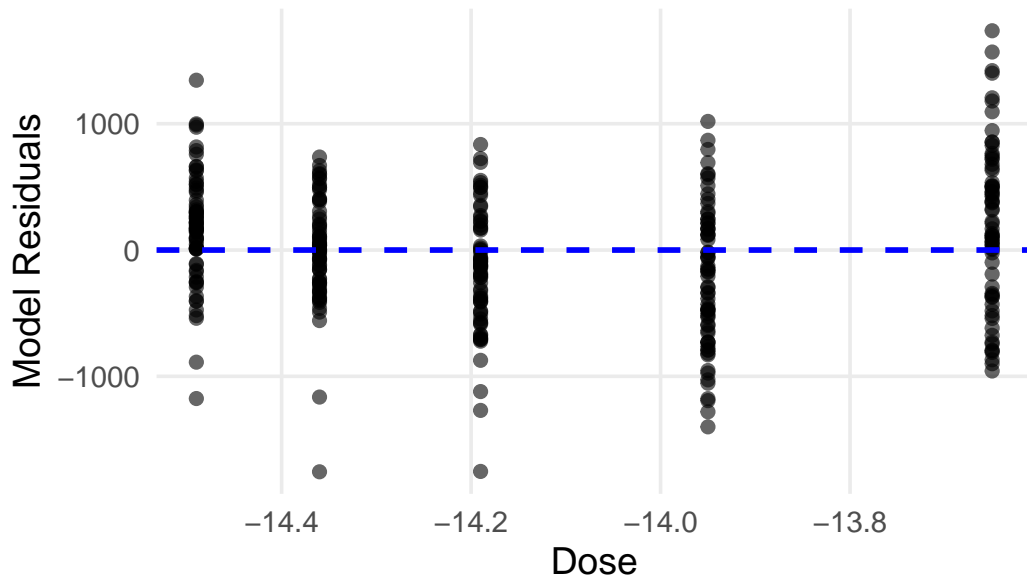
Random Effects



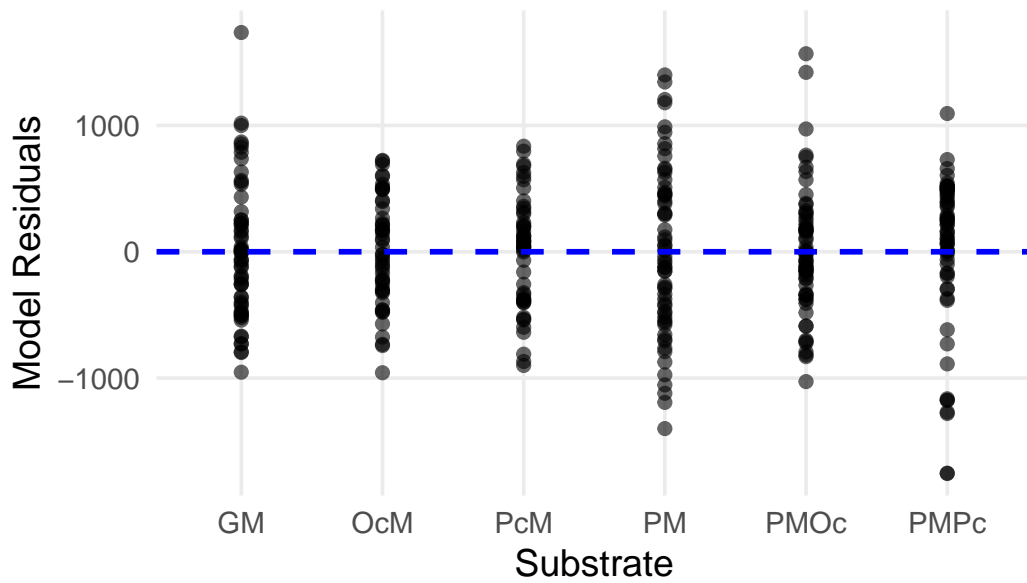
Residuals by Genotype

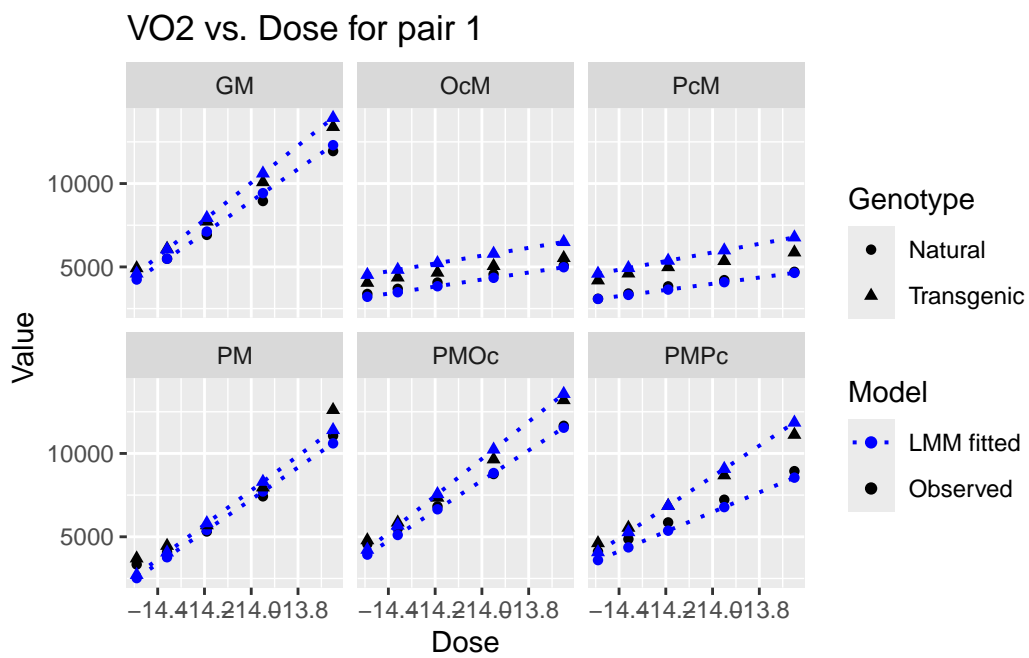
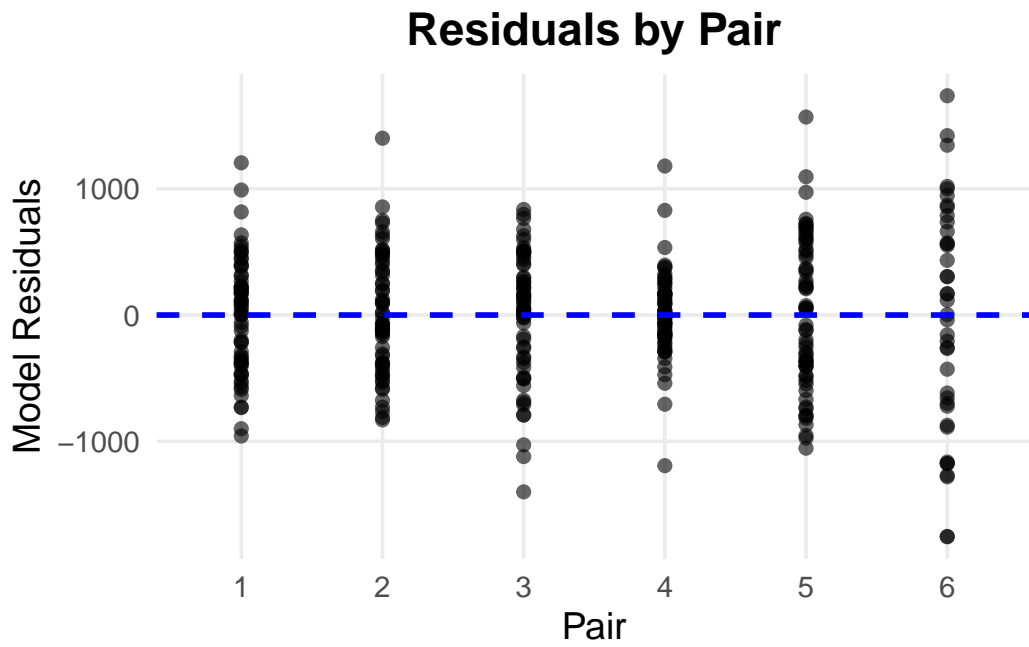


Residuals by Dose

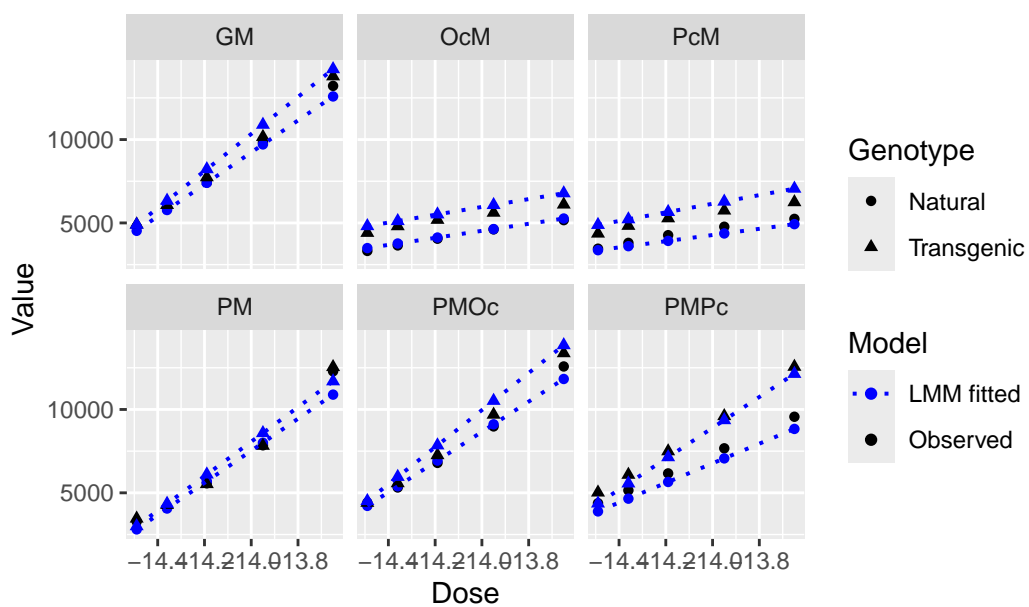


Residuals by Substrate

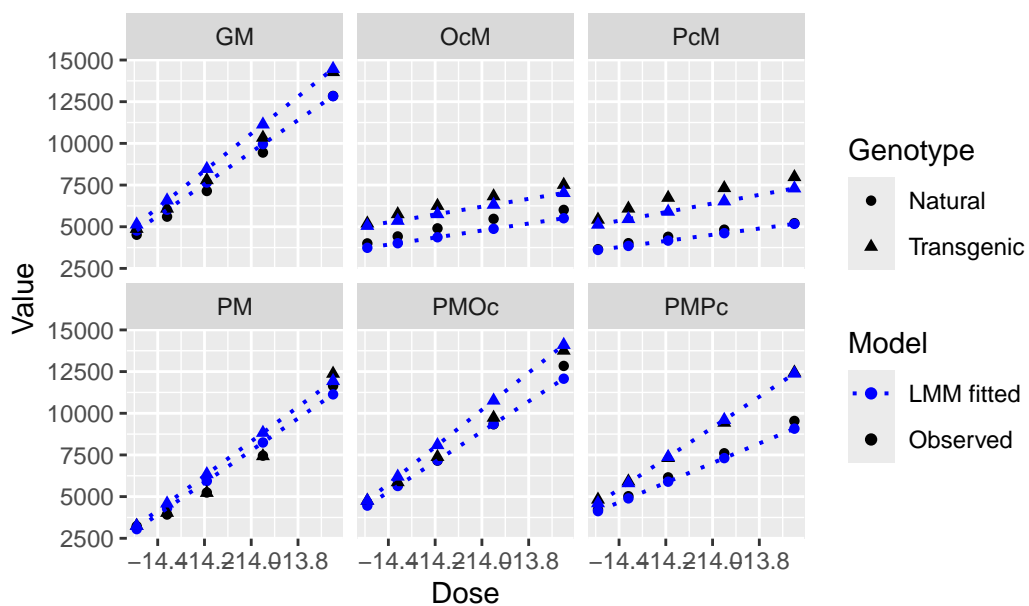




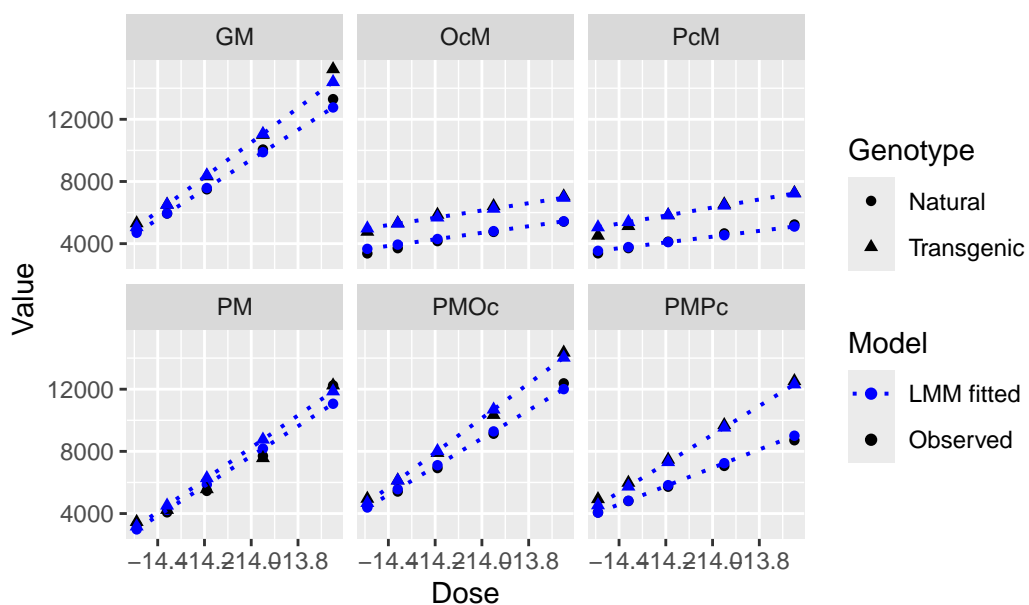
VO2 vs. Dose for pair 2



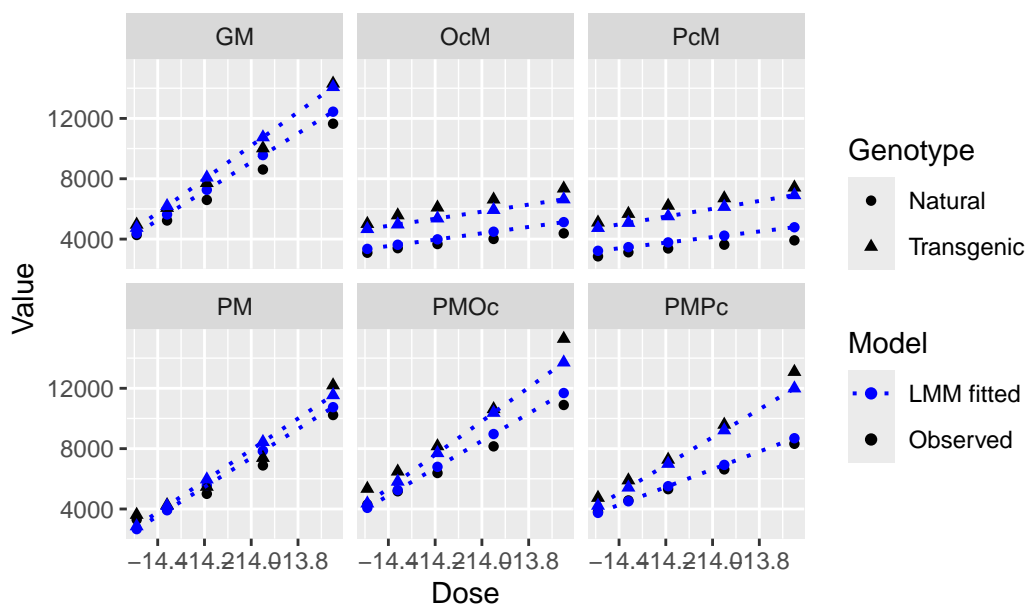
VO2 vs. Dose for pair 3



VO2 vs. Dose for pair 4

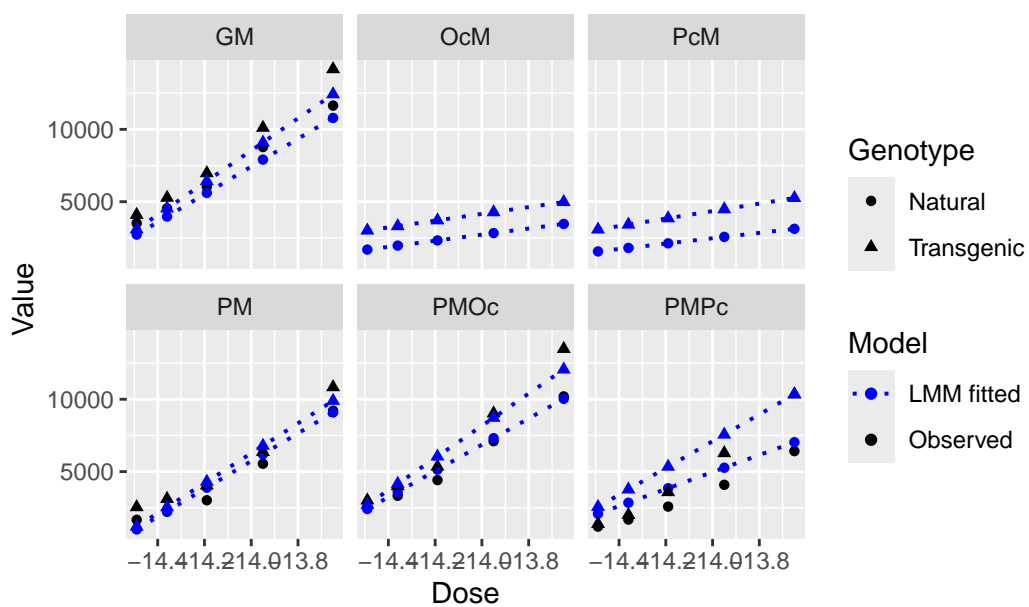


VO2 vs. Dose for pair 5

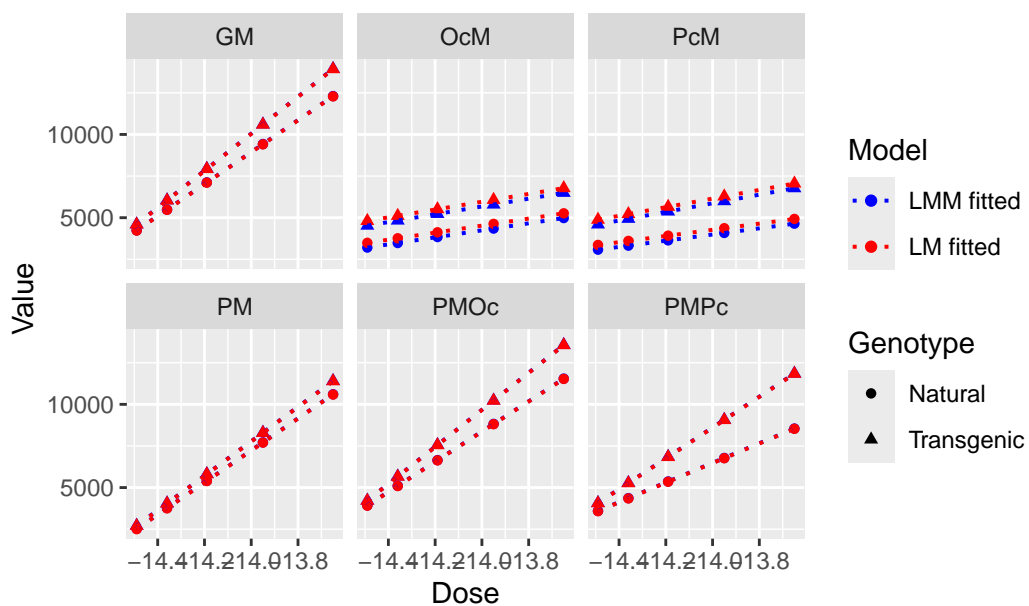


Warning: Removed 20 rows containing missing values or values outside the scale range (``geom_point()``).

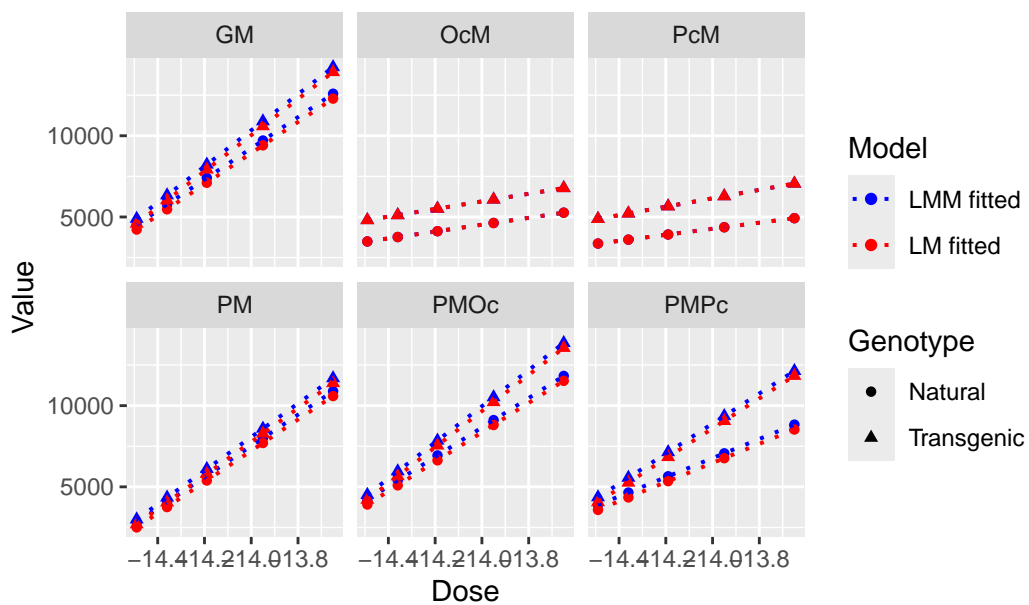
VO2 vs. Dose for pair 6



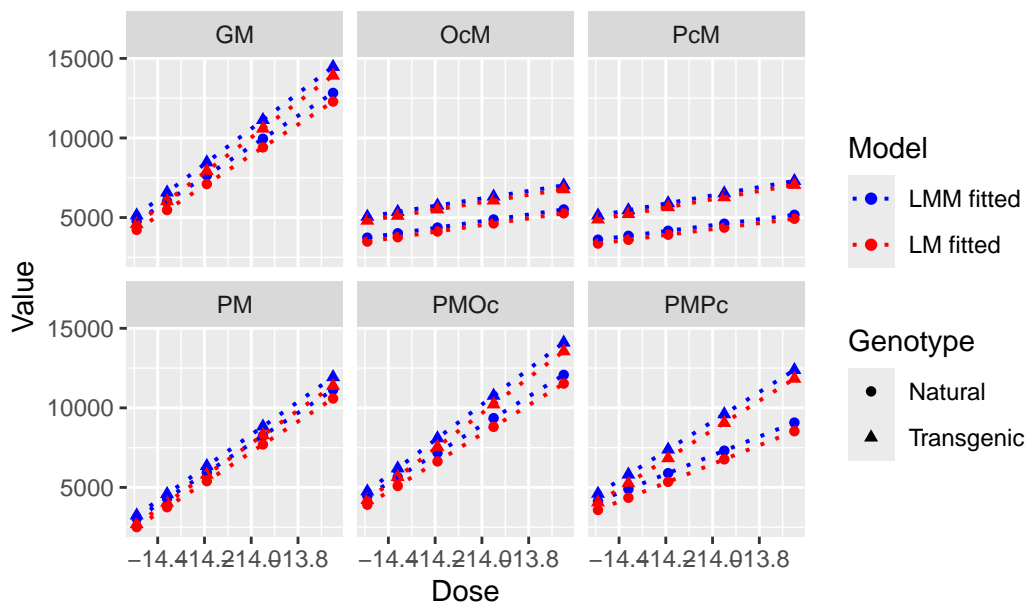
VO2 vs. Dose for pair 1



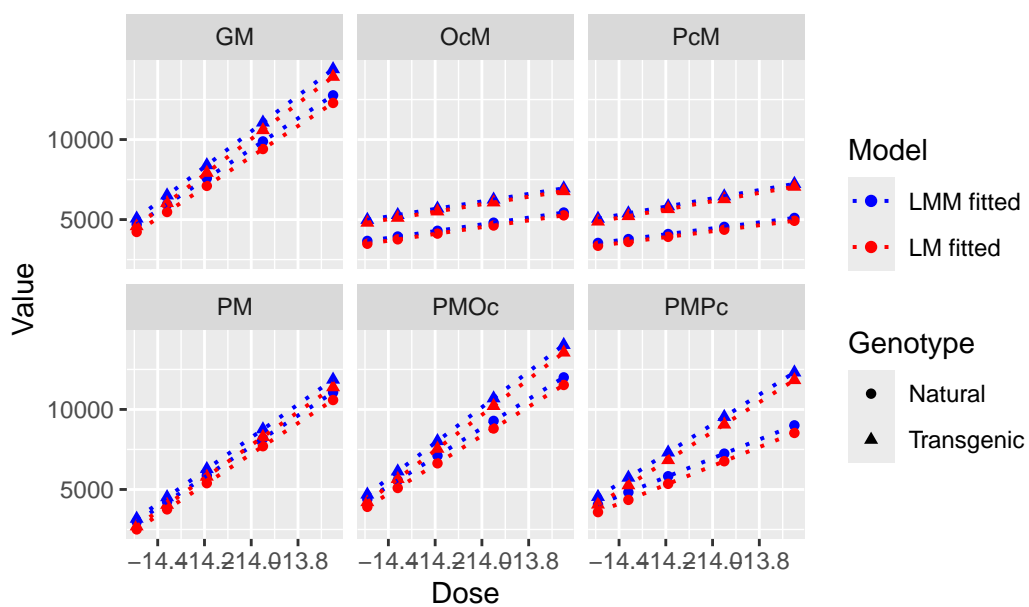
VO2 vs. Dose for pair 2



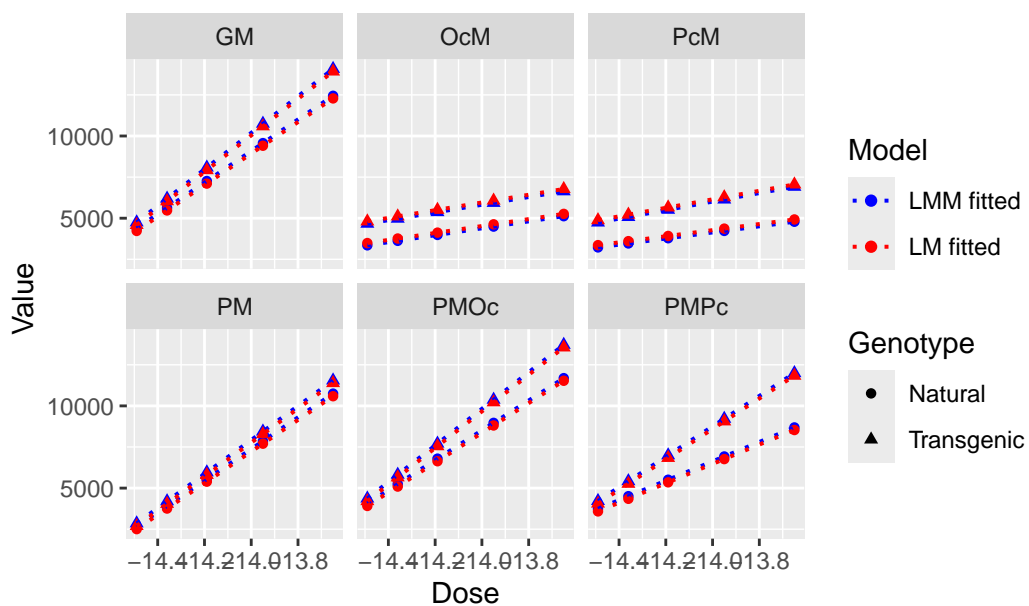
VO2 vs. Dose for pair 3



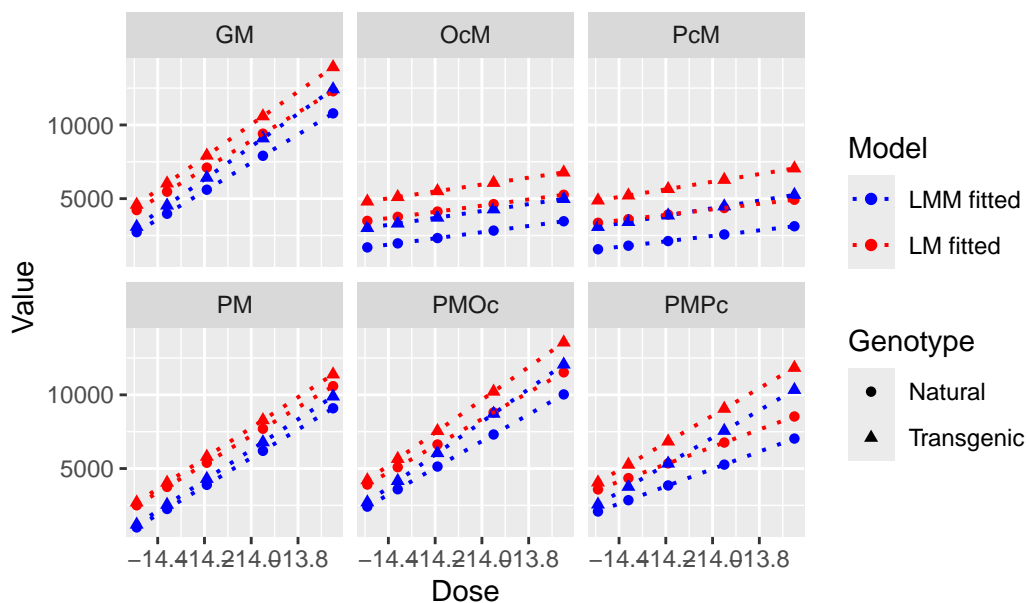
VO2 vs. Dose for pair 4



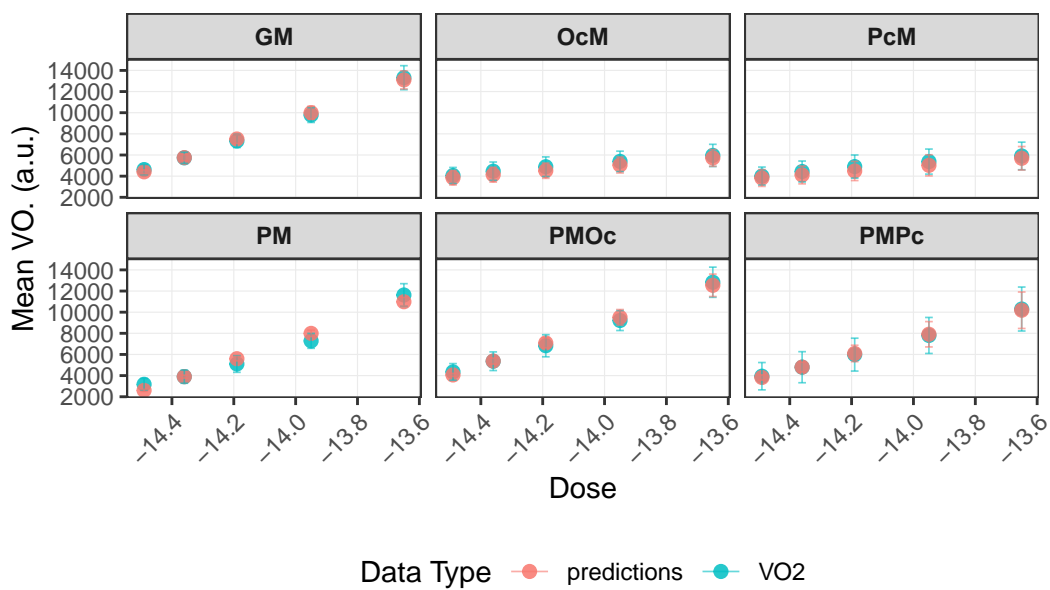
VO2 vs. Dose for pair 5



VO2 vs. Dose for pair 6



Dose vs. Mean VO.



Predictions vs. Observed VO.

