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Electronic Supplementary Information

New bifunctional ionic liquid-based plant systemic acquired resistance (SAR) inducers with improved environmental hazard profile

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1. Physicochemical properties of SAR inducers

Table S1. Physicochemical properties of compounds included in the study (water solubility, log K_{ow}) as well as their efficacy in inducting SAR activity

No.	Compound	Experimental aqueous solubility [g/L]	Predicted aqueous solubility [g/L]	Log K_{ow}	SAR inducing efficiency [a]
1	BTH	0.007 ref [1]	0.04[b]	2.93 [ref [1]]; 2.38[b]; 2.06[c]	95 ref [2]
2	[BTHMe][LS]	> 10 ref [1]	0.016 [b]	2.95 [ref [1]];3.59[b]	< 10%
3	[BTHMe] [$MeSO_4$]	> 10 ref [1]	85.1 [b]	0.32 [ref [1]]	< 10%
4	[K][BTHCOO]	> 10 ref [2]	154.2 [b]	-2.29[b]; -3.50[c]	85 ref [2]
5	[Chol][BTHCOO]	> 10 ref [2,3]	33.9 [b]	-1.56[b]; -1.08[c]	90 ref [2]
6	[N_{4444}][BTHCOO]	> 10 ref [2]	0.002 [b]	4.90[b]; 4.81[c]	100 ref [2]
7	[N_{11410}][BTHCOO]	1.2 ref [2]	0.005 [b]	4.58[b]; 4.68[c]	95 ref [2]
7a	[N_{11410}][Br]	-	-	7.90[d]; 8.16[e]	
8	[$N_{11010Benz}$] [BTHCOO]	0.08 ref [2]	0.1 x 10^{-6} [c]	10.10[c]	85 ref [2]
8a	[$N_{11010Benz}$][Cl]	-	-	12.75[d]; 13.05[e]	
9	[Py C_{10}][BTHCOO]	0.1 ref [2]	0.005 [b]	3.17[b]; 4.29[c]	75 ref [2]
9a	[Py C_{10}][Cl]	-	-	7.32[d]; 6.70[e]	
10	[Py C_{12}][BTHCOO]	0.08 ref [2]	0.0005 [b]	4.38[b]; 5.23[c]	80 ref [2]
10a	[Py C_{12}][Cl]	-	-	7.75[d]; 8.34[e]	
11	[Terpene C_9] [BTHCOO]	> 0.02 ref [4]	2.3 x 10^{-6} [c]	7.21[c]	95 ref [4]
11a	[Terpene C_9][Cl]	-	-	10.47[d]; 10.28[e]	
12	[Terpene C_{12}] [BTHCOO]	> 0.02 ref [4]	0.7 x 10^{-6} [c]	8.05[c]	93 ref [4]
12a	[Terpene C_{12}][Cl]	-	-	11.93[d]; 11.61[e]	

[a] SAR inducing efficiency measured against tobacco mosaic virus (TMV) in tobacco plants (*Nicotiana tabacum L.* var Xanthi) at concentration of 20mg/L

[b] Predicted using COSMO-RS at TZVP level (in case of ionic compounds calculations are for ion pairs)

[c] Predicted using COSMO-RS at SVP level (in case of ionic compounds calculations are for ion pairs)

[d] Predicted using COSMO-RS at TZVP level for the cation

[e] Predicted using COSMO-RS at SVP level for the cation

2. Correlation analysis in toxicity tests

Two parameters: octanol-water partition coefficient (log K_{ow}) and biological membrane-water partition coefficient (log K_{mw}) were used as descriptors of toxicity of SAR inducers toward IPC-81, *Aliivibrio fischeri* and *Daphnia magna*. The results are shown below.

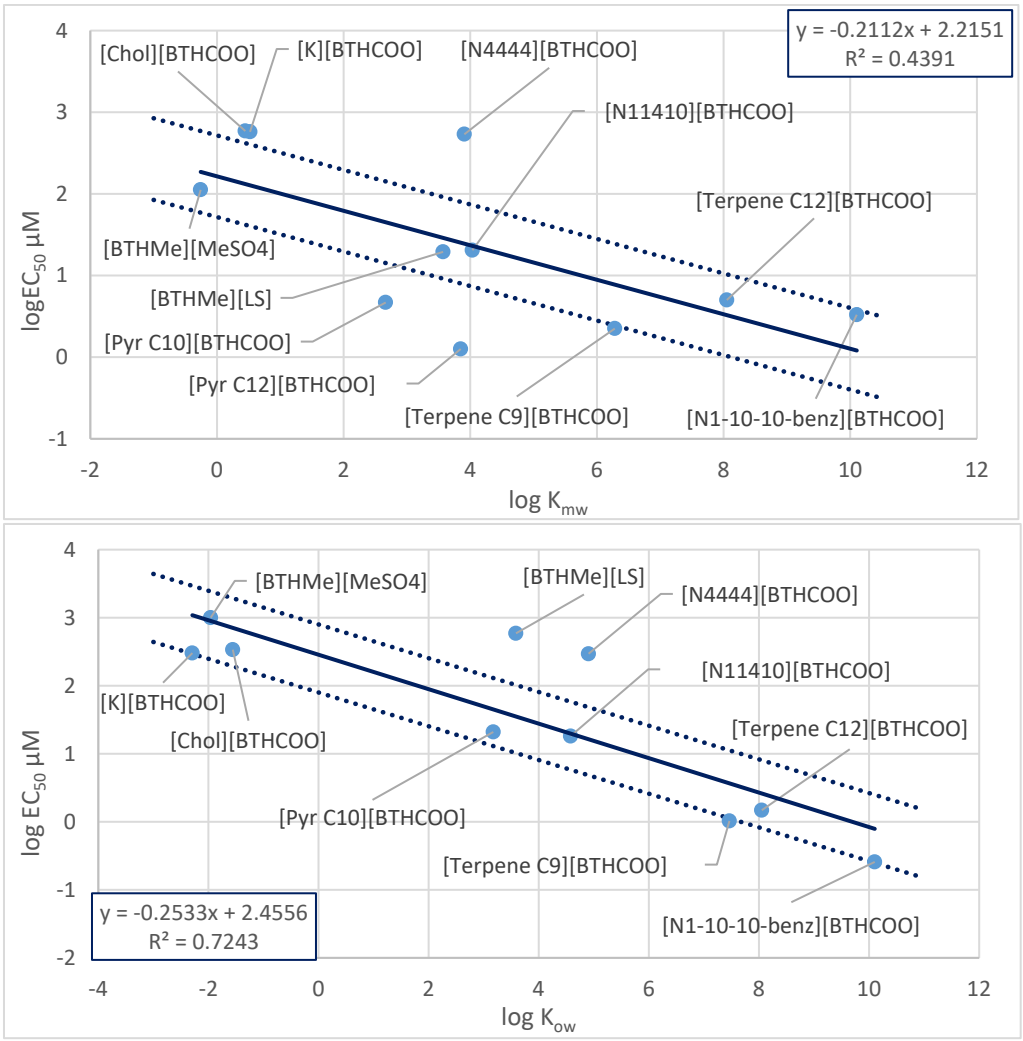
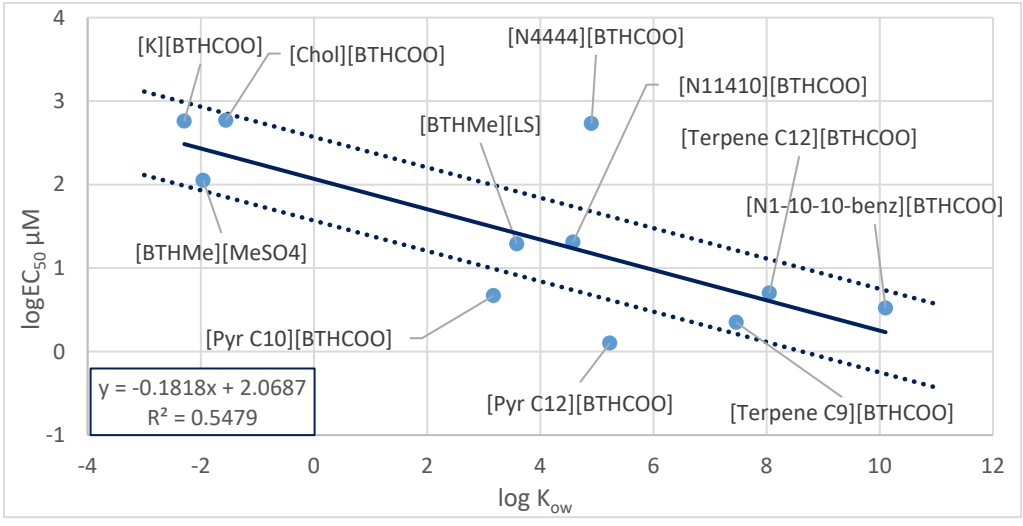


Figure S1. Correlation of effective concentration in IPC-81 test system and (a) log Kow or (b) log Kmw. Solid line shows the correlation and dashed line the +/- 0.5 log unit from the correlation line.



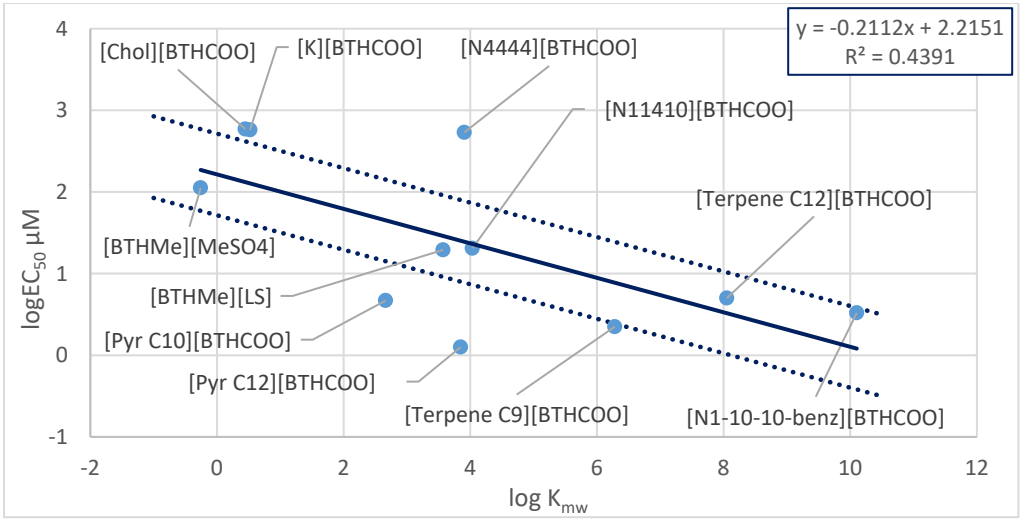


Figure S2. Correlation of effective concentration in *Aliivibrio fischeri* test system and (a) log Kow or (b) log Kmw. Solid line shows the correlation and dashed line the +/- 0.5 log unit from the correlation line.

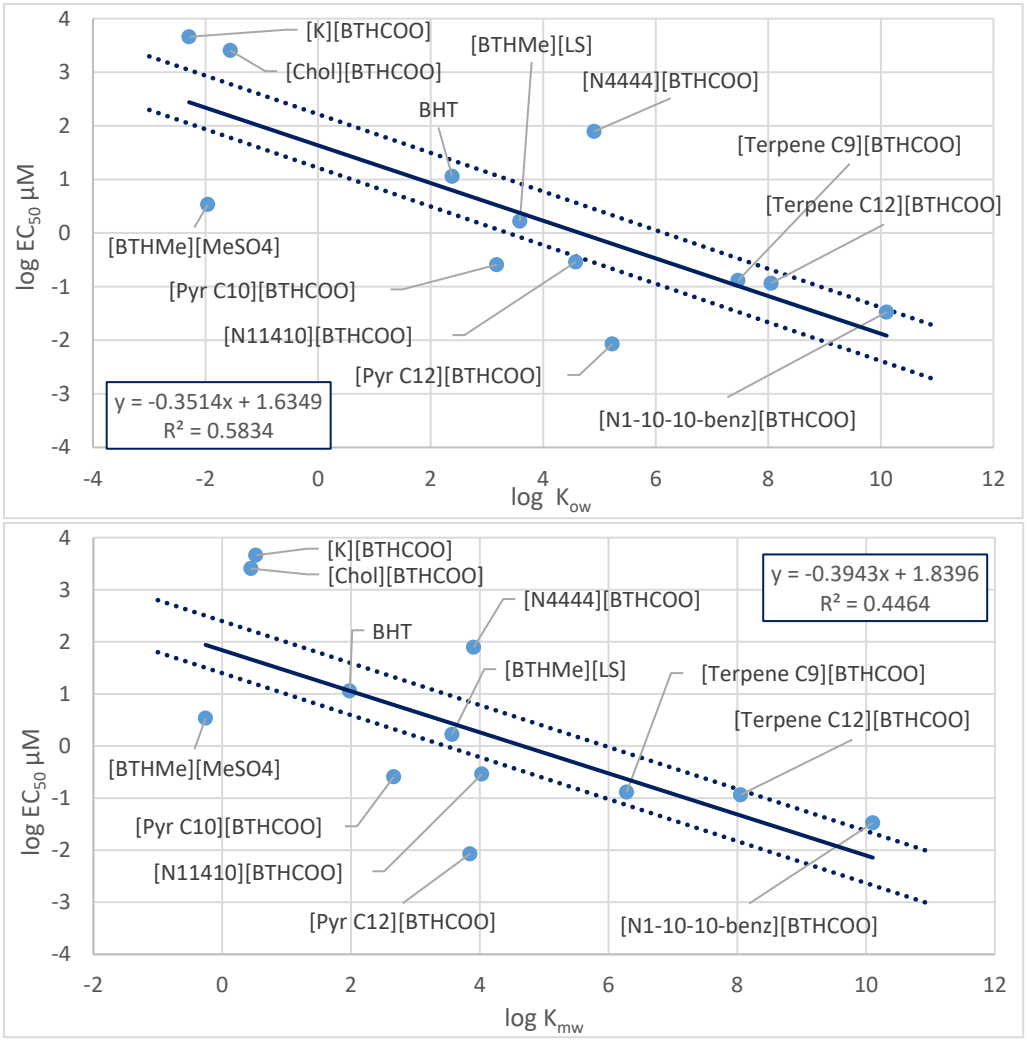


Figure S3. Correlation of effective concentration in *Daphnia magna* test system and (a) log Kow or (b) log Kmw. Solid lien shows the correlation and dashed line the +/- 0.5 log unit from the correlation line.

3. Details of data analysis

In almost all test a monotonic dose-response curve was observed and EC_{50} was estimated using one of two models: logit or linlogit – Table S2 presents the details of model fitting including type of model and residual sum of squares (RSS). However in several cases EC_{50} could not be calculated:

- For BTH in IPC-81 test system and for [K][BTHCOO] in Daphnia magna test system there was no observable effect in any of the doses tested
- For [PyrC_{12}][BTHCOO] a monotonic dose-response curve was observed but EC_{50} could not be estimated

For the former two cases we tested the significance of the difference between the treatments and controls using Mann-Whitney tests. The details are shown in Table S3.

Table S2. Details of dose-response curve fitting in toxicity assessment. Model used to calculate effective concentrations and residual sum of squares are given for each compound and test systems except cases when no test was conducted or no dose-response curve was fitted. Below the table the dose-response curves are shown.

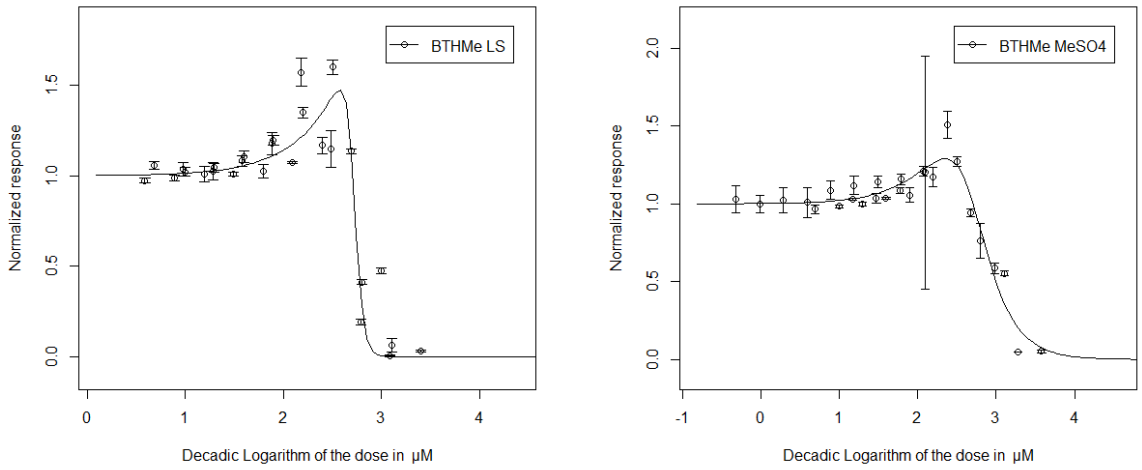
No.	Compound	IPC-81	*Aliivibrio fischeri*	*Daphnia magna*
1	BTH	No effect	Not tested	Not tested
2	[BTHMe][LS]	linlogit, RSS=0.1537	linlogit, RSS=0.0134	logit RSS= 0.0925
3	[BTHMe][$MeSO_4$]	linlogit RSS=0.1516	linlogit, RSS= 0.0473	linlogit RSS= 0.0608
4	[K][BTHCOO]	linlogit, RSS=0.0724	linlogit, RSS=0.0281	No effect
5	[Chol][BTHCOO]	linlogit RSS=0.0854	linlogit, RSS=0.0204	linlogit RSS=0.1902
6	[N_{4444}][BTHCOO]	linlogit RSS=0.1406	linlogit, RRS= 0.0296	linlogit, RSS=0.1769
7	[N_{11410}][BTHCOO]	linlogit, RSS=0.3152	linlogit, RSS=0.0183	linlogit, RSS=0.2517
8	[$N_{11010Benz}$][BTHCOO]	linlogit, RRS=0.1136	linlogit, RSS=0.0389	linlogit, RSS=0.1852
9	[PyC_{10}][BTHCOO]	linlogit, RRS=0.1004	linlogit, RSS=0.0384	linlogit, RSS=0.1598
10	[PyC_{12}][BTHCOO]	No fit	linlogit RSS=0.0187	linlogit, RSS=0.2700
11	[TerpeneC_9][BTHCOO]	linlogit, RSS=0.1325	linlogit, RSS=0.0241	probit, RSS=0.0947
12	[TerpeneC_{12}] [BTHCOO]	linlogit, RSS=0.1495	linlogit, RSS=0.0530	linlogit, RSS=0.1306

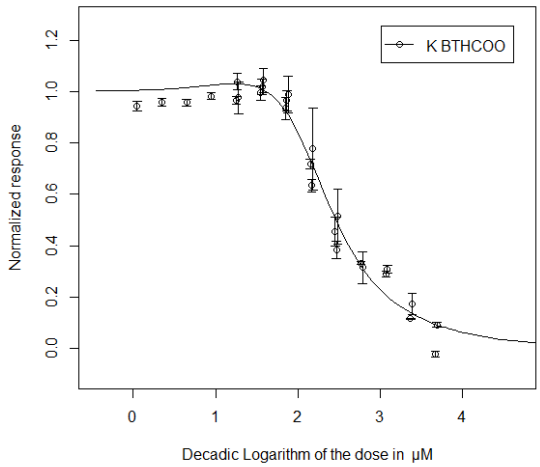
Table S3. Results of statistical significance testing for the compounds/test for which no dose-response curve was obtained based on Mann-Whitney test.

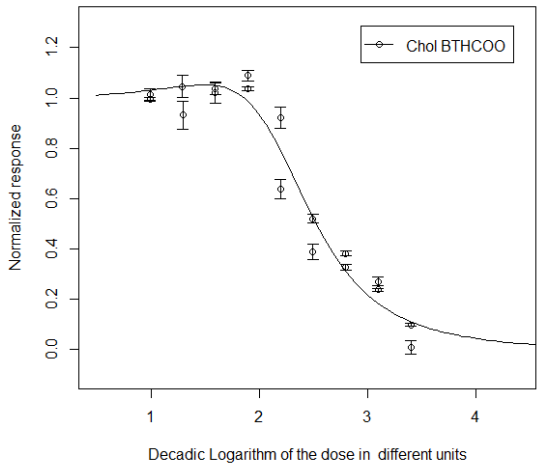
BTH in IPC-81 test	
Dose [µmol/L]	**p-value (α=0.05) and result of hypothesis testing**
0.074	0.3733, not significantly different from control
0.297	0.2975, not significantly different from control
0.149	0.0575, not significantly different from control
0.594	0.3891, not significantly different from control
1.189	0.3848, not significantly different from control
2.378	0.0220, not significantly different from control
4.755	0.0506, not significantly different from control
9.51	0.2021, not significantly different from control
19.2	0.1793, not significantly different from control
[K][BTHCOO] in *Daphnia magna* test	
Dose [mg/L]	**p-value (α=0.05) and result of hypothesis testing**
1.0	not significantly different from control
4.0	p=0.4237, not significantly different from control
21	not significantly different from control
40	not significantly different from control
100	NA
125	p=0.4237, not significantly different from control
200	NA
250	not significantly different from control
500	not significantly different from control
1000	p=0.4237, not significantly different from control

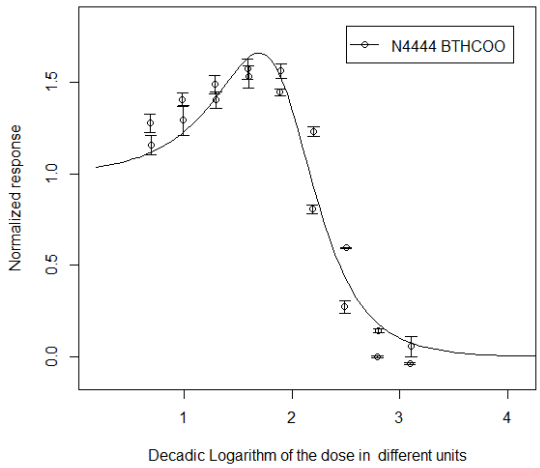
4. Dose-response curves

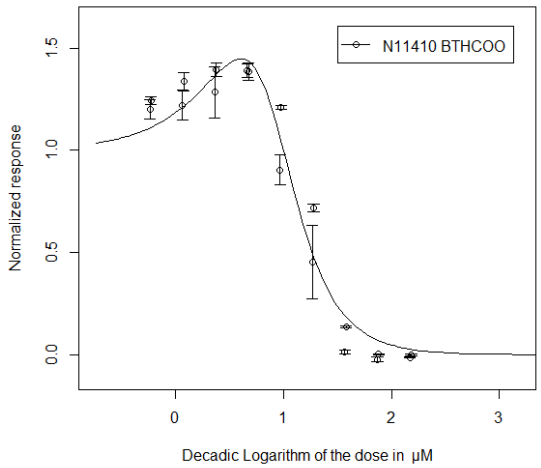
IPC-81

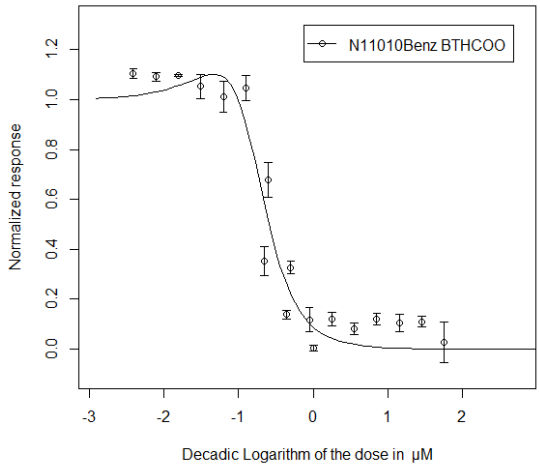


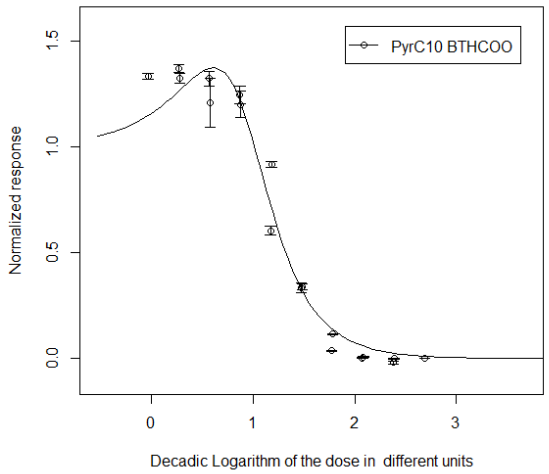

K BTHCOO
Normalized response
Decadic Logarithm of the dose in µM

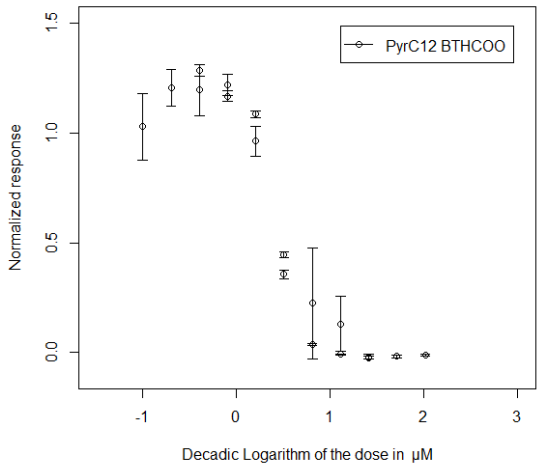

Chol BTHCOO
Normalized response
Decadic Logarithm of the dose in different units

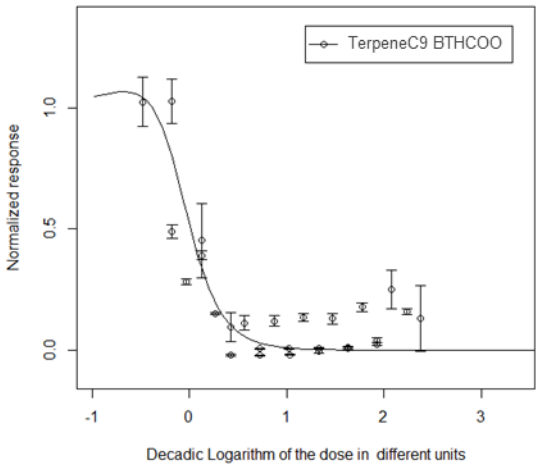

N4444 BTHCOO
Normalized response
Decadic Logarithm of the dose in different units

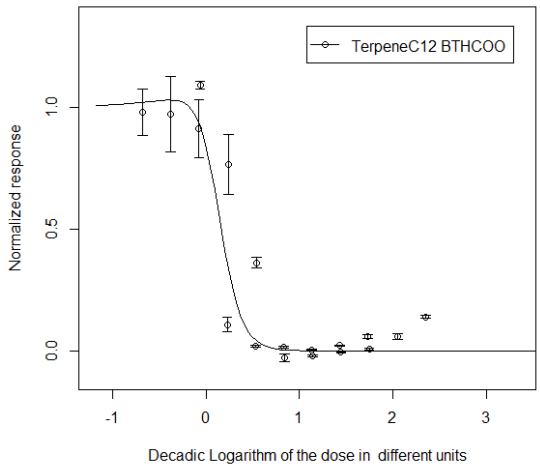

N11410 BTHCOO
Normalized response
Decadic Logarithm of the dose in µM


N11010Benz BTHCOO
Normalized response
Decadic Logarithm of the dose in µM

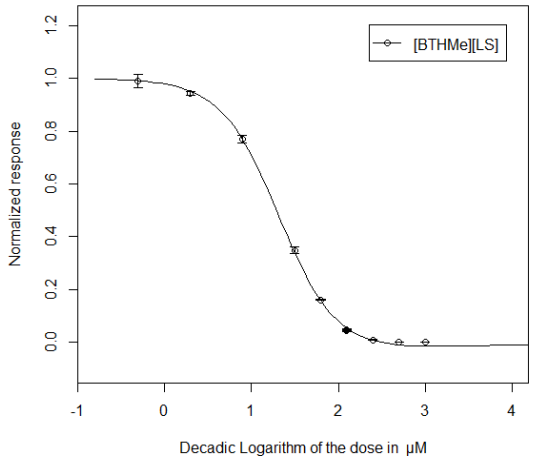

PyrC10 BTHCOO
Normalized response
Decadic Logarithm of the dose in different units

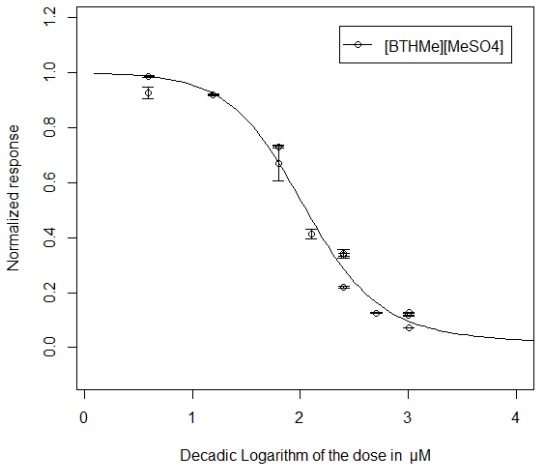


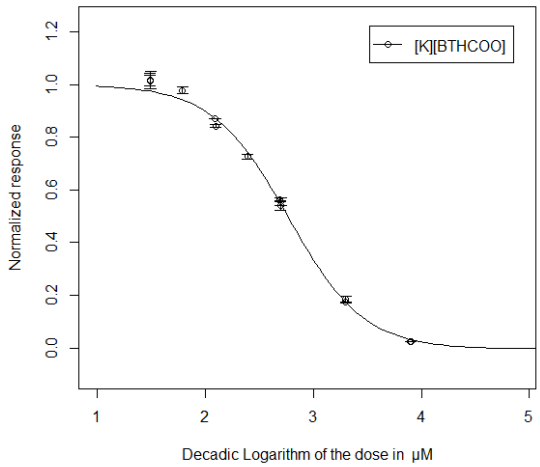


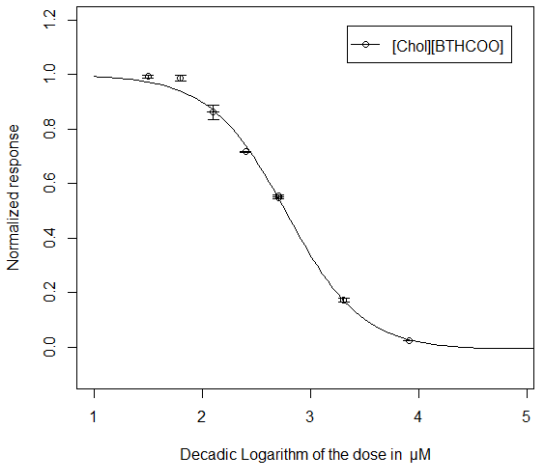


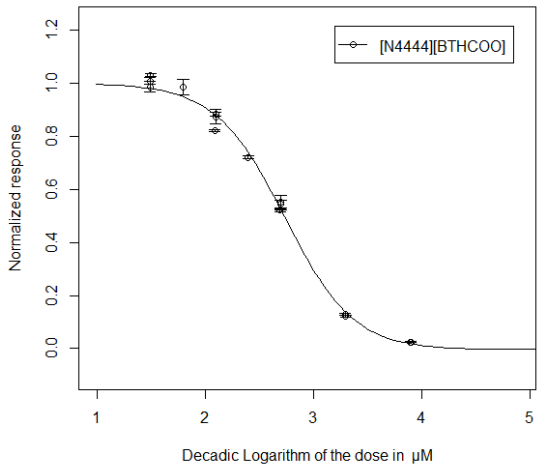
Aliiviibrio fischeri

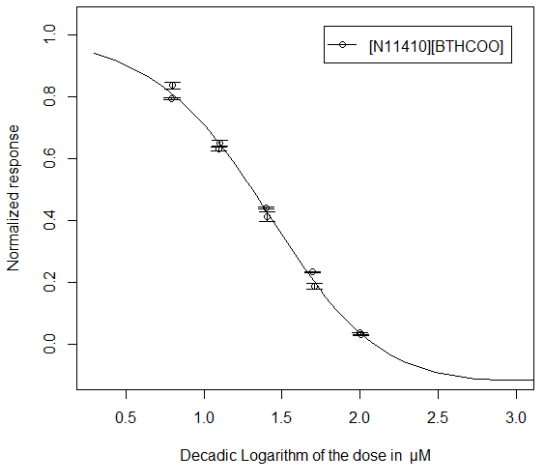


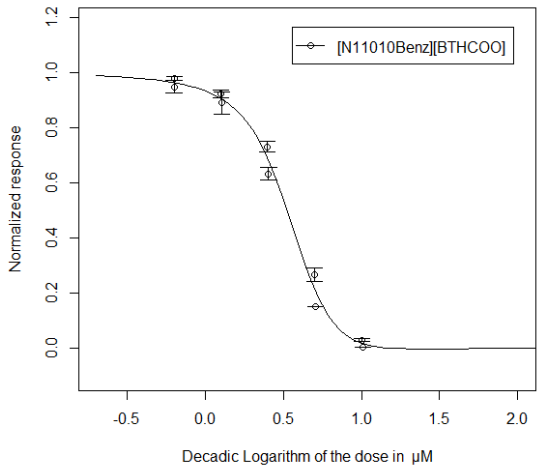


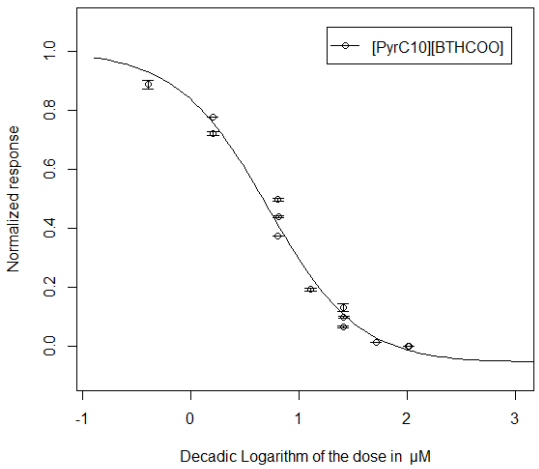

[K][BTHCOO]
Normalized response
Decadic Logarithm of the dose in µM

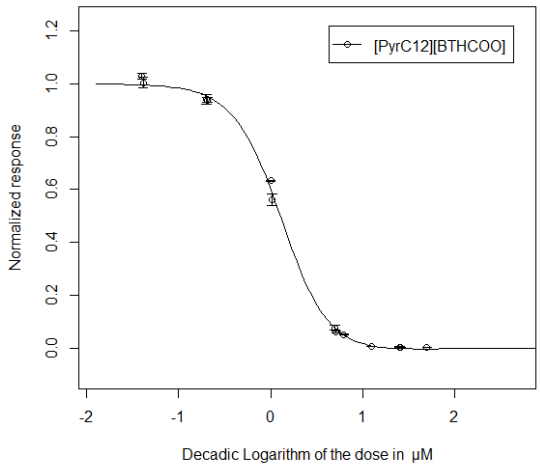

[Chol][BTHCOO]
Normalized response
Decadic Logarithm of the dose in µM

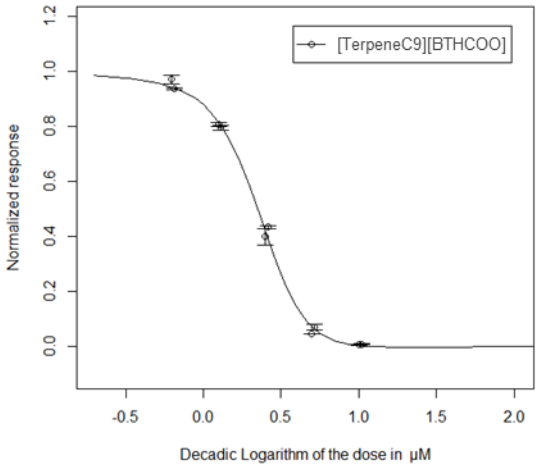

[N4444][BTHCOO]
Normalized response
Decadic Logarithm of the dose in µM

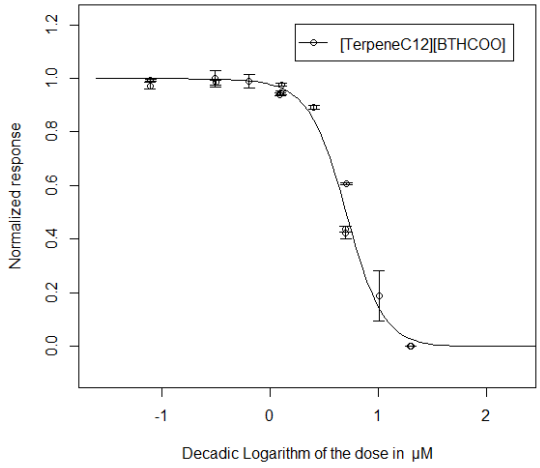

[N11410][BTHCOO]
Normalized response
Decadic Logarithm of the dose in µM


[N11010Benz][BTHCOO]
Normalized response
Decadic Logarithm of the dose in µM

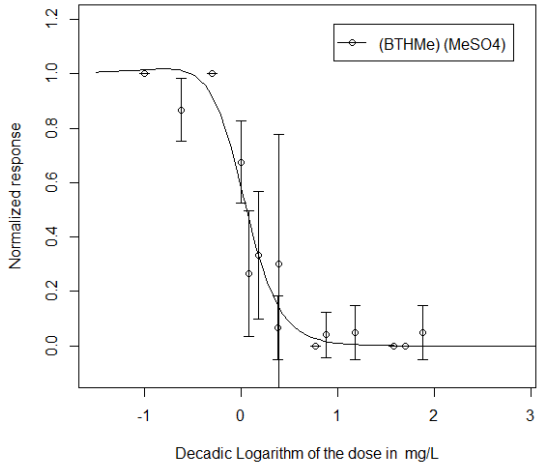

[PyrC10][BTHCOO]
Normalized response
Decadic Logarithm of the dose in µM

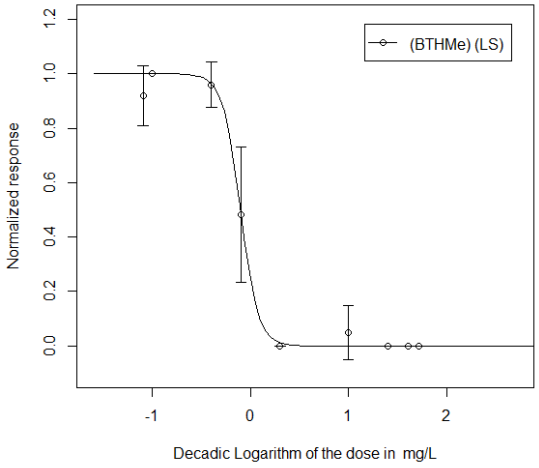


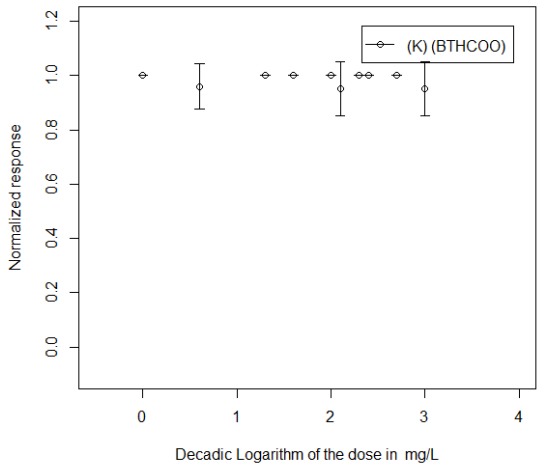


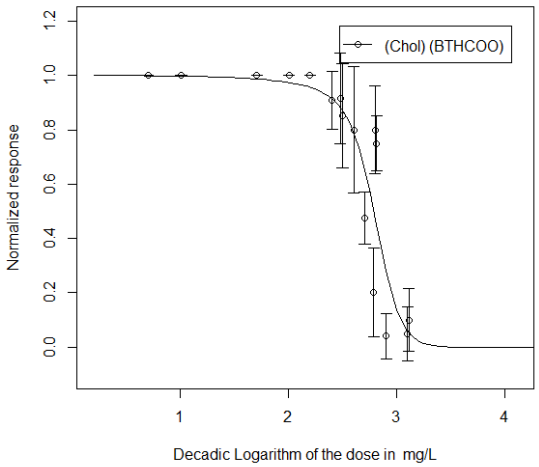


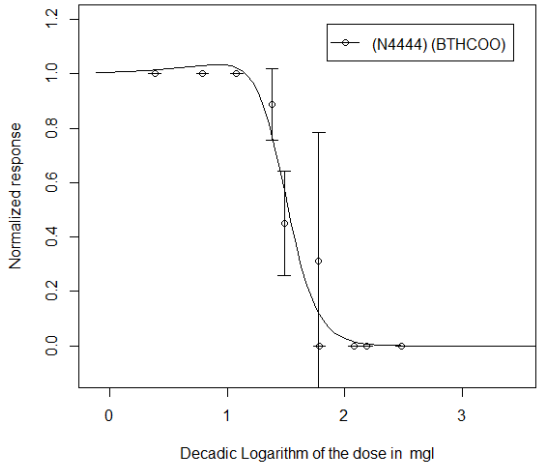
Daphnia manga

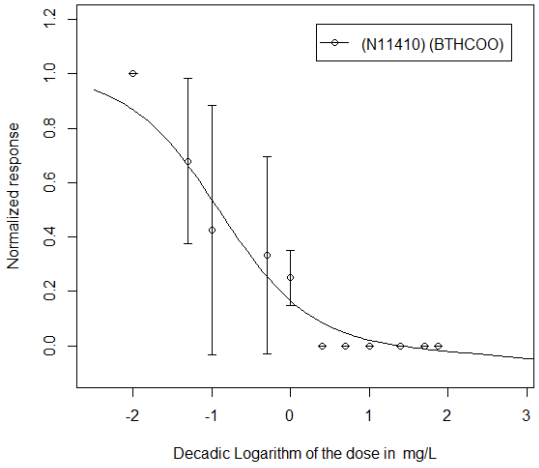


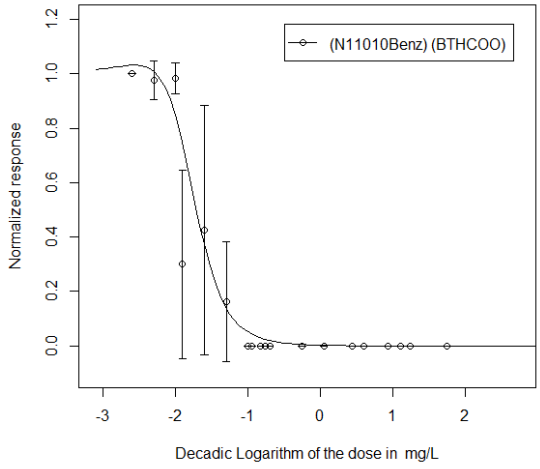


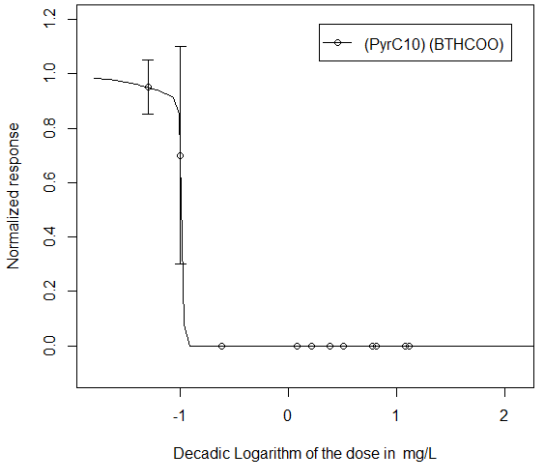

(K) (BTHCOO)
Normalized response
Decadic Logarithm of the dose in mg/L

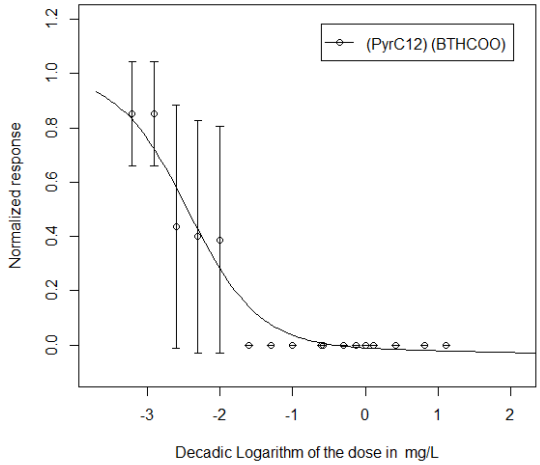

(Chol) (BTHCOO)
Normalized response
Decadic Logarithm of the dose in mg/L

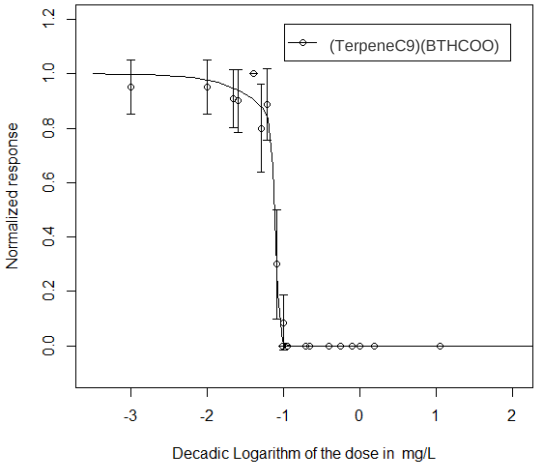

(N4444) (BTHCOO)
Normalized response
Decadic Logarithm of the dose in mgl

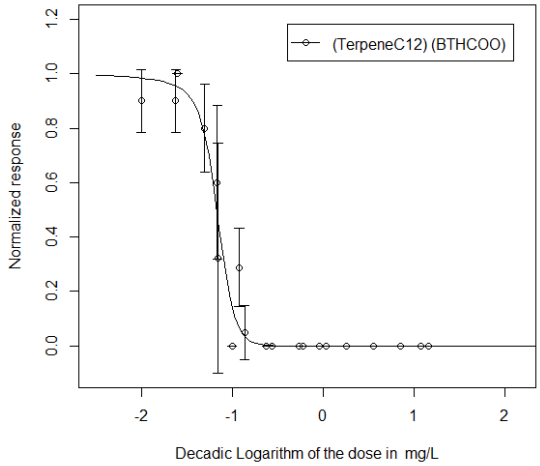

(N11410) (BTHCOO)
Normalized response
Decadic Logarithm of the dose in mg/L


(N11010Benz) (BTHCOO)
Normalized response
Decadic Logarithm of the dose in mg/L


(PyrC10) (BTHCOO)
Normalized response
Decadic Logarithm of the dose in mg/L


(PyrC12) (BTHCOO)
Normalized response
0.0
0.2
0.4
0.6
0.8
1.0
1.2
-3
-2
-1
0
1
2
Decadic Logarithm of the dose in mg/L


(TerpeneC9)(BTHCOO)
Normalized response
0.0
0.2
0.4
0.6
0.8
1.0
1.2
-3
-2
-1
0
1
2
Decadic Logarithm of the dose in mg/L


(TerpeneC12) (BTHCOO)
Normalized response
0.0
0.2
0.4
0.6
0.8
1.0
1.2
-2
-1
0
1
2
Decadic Logarithm of the dose in mg/L