Supplementary Material 2: Appendices for Chapter 3

S2.2 Linear models and statistical tests for physiological measurements

Fitted linear model for changes in estimated cell densities of *Breviolum minutum per* µg of host protein by pH treatment

Linear model formula: Cell density = 461 + 28 (pH 7.68 no pre-exposure) - 70 (pH 7.68 6-h pre-exposure)

No effect of header tank was detected.

Coefficients	Estimate	Std. Error	t value	*p*-value	Significance
(Intercept)	461.810	88.100	5.242	$1.27e^{-06}$	***
pH 7.68 no pre-exposure	28.360	123.480	0.230	0.819	
pH 7.68 6-h pre-exposure	-69.890	123.480	-0.566	0.573	
Header tank effect	NA	NA	NA	NA	NA

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

Residual standard error: 457.8 on 80 degrees of freedom
Multiple R-squared: 0.008456
Adjusted R-squared: -0.01633
F-statistic: 0.3411 on 2 and 80 df
p-value: 0.712

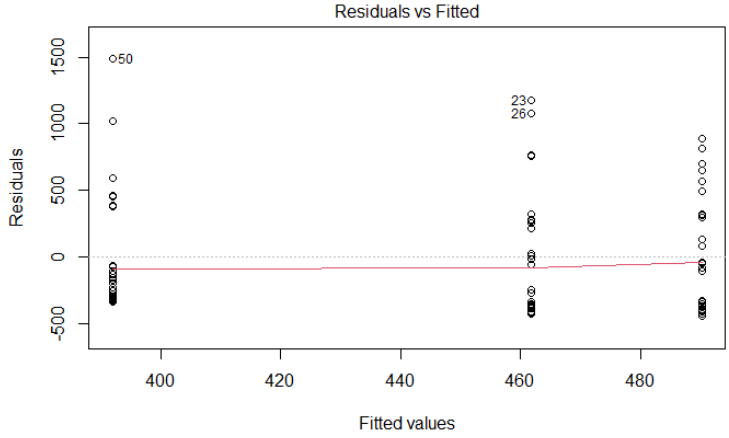
Model ANOVA

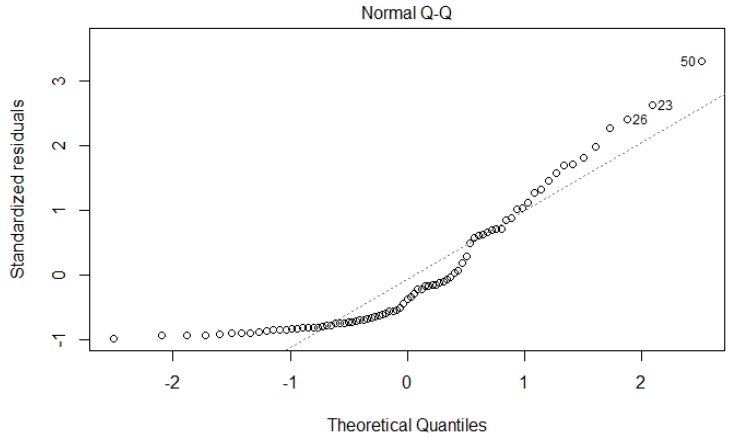
Variation	df	Sum of squares	Mean square	F-value	*p*-value
pH	2	142995	71497	0.341	0.712
Residuals	80	16766874	209586		

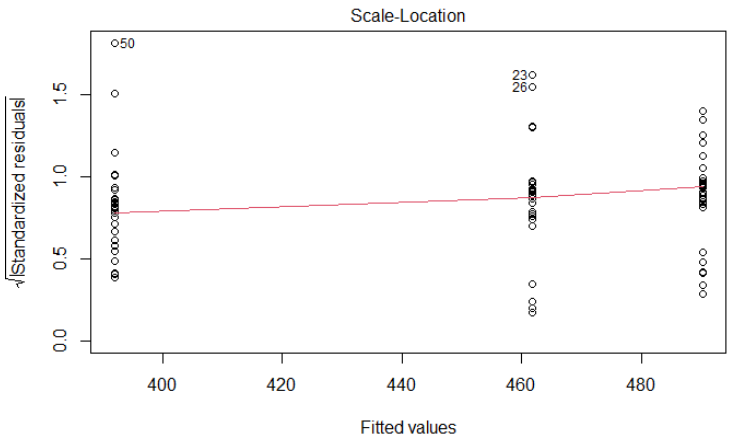
Kruskal-Wallis rank sum test

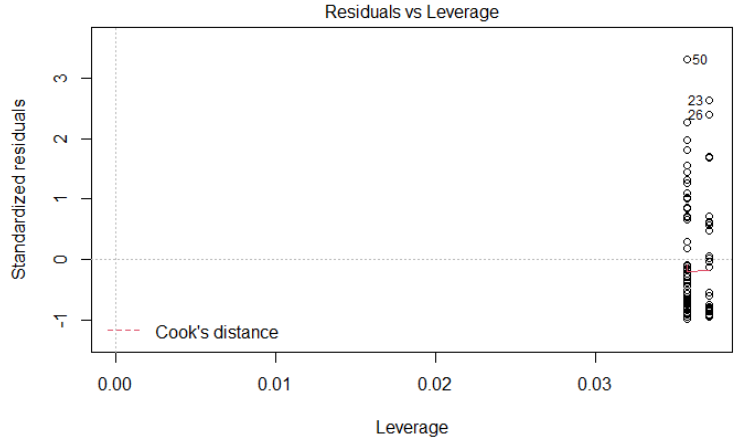
Data	χ^2	df	*p*-value
Cell densities by pH	0.8455	2	0.655

Model plots


Residuals vs Fitted
Residuals
Fitted values
50
23
26


Normal Q-Q
Standardized residuals
Theoretical Quantiles
50
23
26


Scale-Location
√|Standardized residuals|
Fitted values
50
23
26


Residuals vs Leverage
Standardized residuals
Leverage
Cook's distance
50
23
26

Fitted linear model for changes in estimated cell densities of *Breviolum minutum per* µg of host protein over time

Linear model formula: Cell density = 94 + 1032 (time)

No effect of header tank was detected.

Coefficients	**Estimate**	**Std. Error**	**t value**	***p*-value**	**Significance**
(Intercept)	94.71	42.370	2.235	0.0282	*
Day 7	37.19	59.920	0.621	0.5366	
Day 14	375.90	59.920	6.274	$1.76e^{-08}$	***
Day 28	1031.59	60.660	17.005	$2.00e^{-16}$	***
Header tank effect	NA	NA	NA	NA	NA

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

Residual standard error: 194.2 on 79 degrees of freedom
Multiple R-squared: 0.8239
Adjusted R-squared: 0.8172
F-statistic: 123.2 on 3 and 79 df
p-value: $2.20e^{-16}$

Model ANOVA

Variation	**df**	**Sum of squares**	**Mean square**	**F-value**	***p*-value**
pH	3	13931826	4643942	123.2	$2e^{-16}$
Residuals	79	2978043	37697		

Kruskal-Wallis rank sum test

Data	**χ^2**	**df**	***p*-value**
Cell densities by Day	0.6699	3	0.1874

Fitted linear model for changes in estimated cell densities of *Breviolum minutum per* µg of host protein by pH and over time

Coefficients	Estimate	Std. Error	t value	*p*-value	Significance
(Intercept)	86.143	70.593	1.220	0.2264	
pH 7.68 no pre-exposure	19.714	99.833	0.197	0.844	
pH 7.68 6-h pre-exposure	6.000	99.833	0.060	0.9522	
Day 7	24.000	99.833	0.240	0.8107	
Day 14	478.857	99.833	4.797	8.63E-06	***
Day 28	1103.857	103.909	10.623	2.56E-16	***
pH 7.68 no pre-exposure:Day 7	19.143	141.185	0.136	0.8925	
pH 7.68 6-h pre-exposure:Day 7	20.429	141.185	0.145	0.8854	
pH 7.68 no pre-exposure:Day 14	-1.286	141.185	-0.009	0.9928	
pH 7.68 6-h pre-exposure:Day 14	-307.571	141.185	-2.178	0.0327	*
pH 7.68 no pre-exposure:Day 28	-87.286	144.096	-0.606	0.5466	
pH 7.68 6-h pre-exposure:Day 28	-120.429	144.096	-0.836	0.4061	
Header tank effect	NA	NA	NA	NA	NA

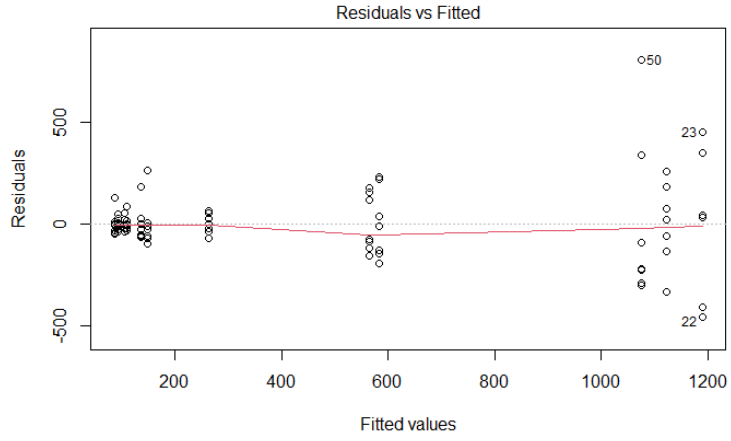
Significance. codes: 0 ‘***’ 0.001 ‘**’ 0.01 ‘*’ 0.05 ‘.’ 0.1 ‘ ’ 1

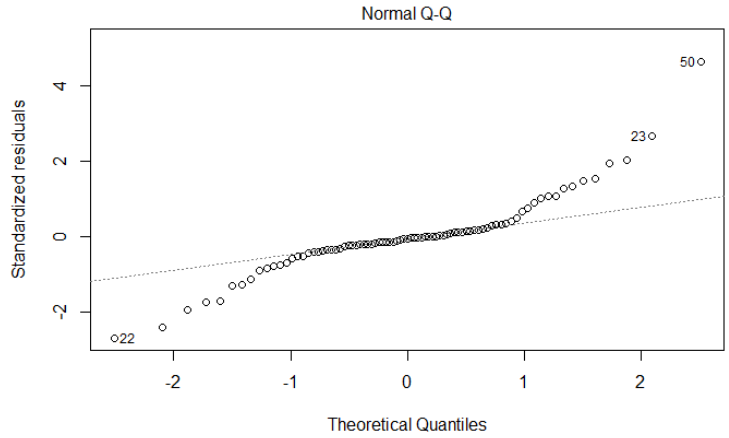
Residual standard error: 186.8 on 71 degrees of freedom
Multiple R-squared: 0.8535
Adjusted R-squared: 0.8308
F-statistic: 37.61 on 11 and 71 df
p-value: $2.20e^{-16}$

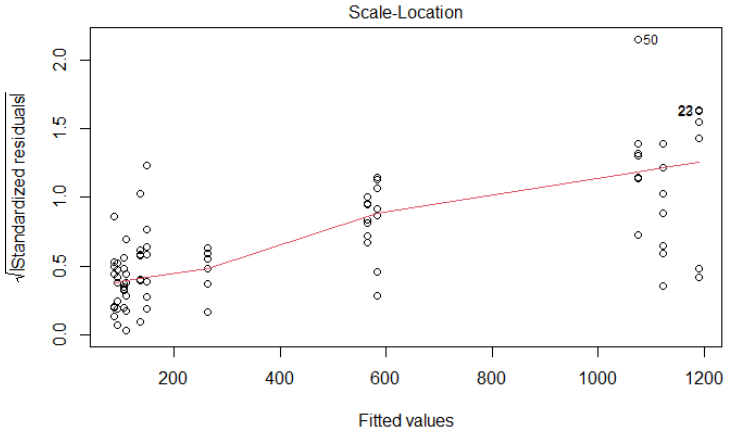
Model ANOVA

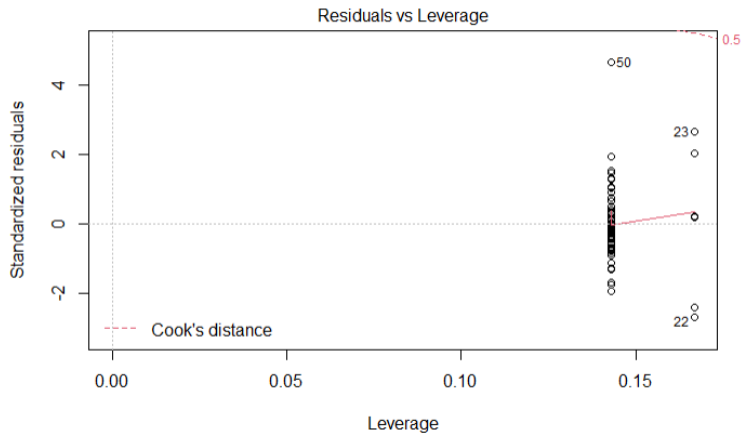
Variation	df	Sum of squares	Mean square	F-value	*p*-value
pH	2	142995	71497	2.05	0.136
Day	3	13961874	4653958	133.416	<2e
pH:Day	6	328296	54716	1.569	0.169
Residuals	71	2476704	34883		

Model plots


Residuals vs Fitted
Residuals
Fitted values
50
23
22


Normal Q-Q
Standardized residuals
Theoretical Quantiles
50
23
22


Scale-Location
√|Standardized residuals|
Fitted values
50
23


Residuals vs Leverage
Standardized residuals
Leverage
Cook's distance
0.5
50
23
22

Fitted linear model for host protein content (µg) by pH

Linear model formula: Protein content = 56.49 + 2.19 (pH 7.68 no pre-exposure) – 3.14 (pH 7.68 6-h pre-exposure)

No effect of header tank was detected.

Coefficients	**Estimate**	**Std. Error**	**t value**	***p*-value**	**Significance**
(Intercept)	56.486	3.366	16.78	$2e^{-16}$	***
pH 7.68 no pre-exposure	2.193	4.761	0.461	0.646	
pH 7.68 6-h pre-exposure	3.136	4.761	0.659	0.512	
Header tank effect	NA	NA	NA	NA	NA

Significance. Codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 17.81 on 81 degrees of freedom
Multiple R-squared: 0.005609
Adjusted R-squared: -0.01894
F-statistic: 0.2284 on 2 and 81 df
p-value: 0.7963

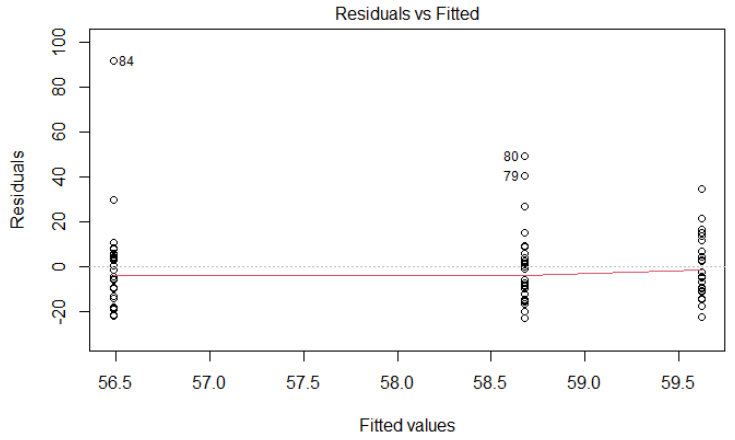
Model ANOVA

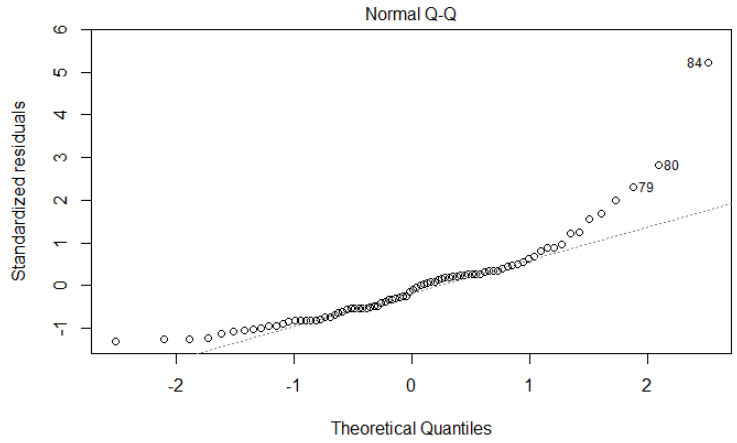
Variation	**df**	**Sum of squares**	**Mean square**	**F-value**	***p*-value**
pH	2	145	72.5	0.228	0.796
Residuals	81	25700	317.3		

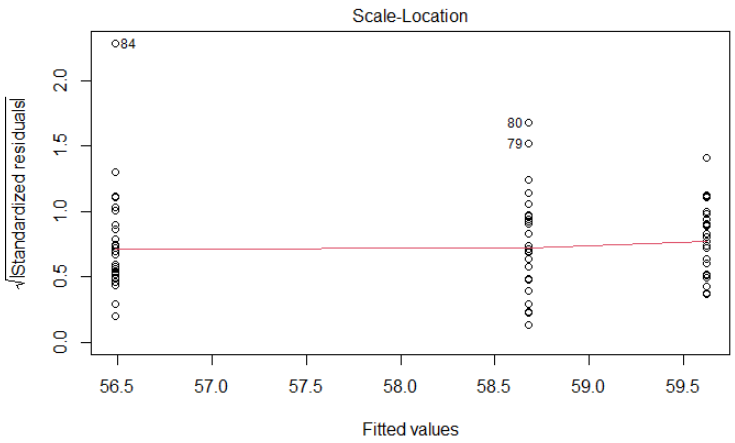
Kruskal-Wallis rank sum test

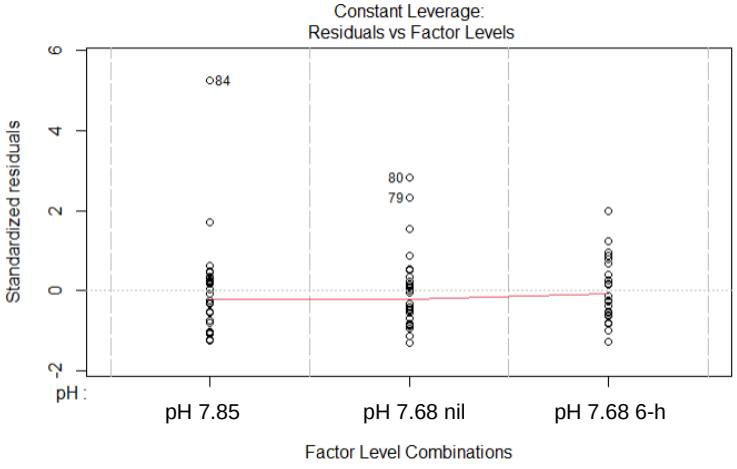
Data	**χ^2**	**df**	***p*-value**
Protein content by pH	1.847	2	0.3971

Model Plots


Residuals vs Fitted
Residuals
Fitted values
84
80
79


Normal Q-Q
Standardized residuals
Theoretical Quantiles
84
80
79


Scale-Location
√|Standardized residuals|
Fitted values
84
80
79


Constant Leverage:
Residuals vs Factor Levels
Standardized residuals
pH 7.85
pH 7.68 nil
pH 7.68 6-h
Factor Level Combinations
84
80
79

Fitted linear model for host protein content (µg) over time

Linear model formula: Protein content = 51.02 + 26.83 (time)

No effect of header tank was detected.

Coefficients	Estimate	Std. Error	t value	*p*-value	Significance
(Intercept)	51.019	2.9951	17.034	$2.00e^{-16}$	***
Day 7	0.4381	4.2357	0.103	0.918	
Day 14	1.7048	4.2357	0.402	0.688	
Day 28	26.8286	4.2357	6.334	$1.31e^{-08}$	***
Header tank effect	NA	NA	NA	NA	NA

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

Residual standard error: 13.73 on 80 degrees of freedom
Multiple R-squared: 0.4169
Adjusted R-squared: 0.395
F-statistic: 19.06 on 3 and 80 df
p-value: $2.20e^{-09}$

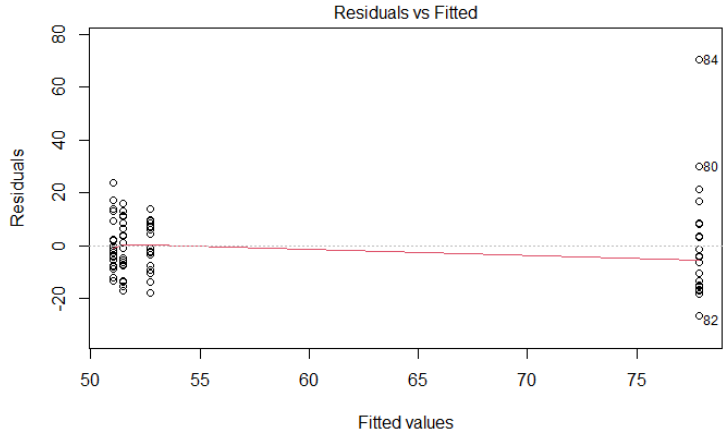
Model ANOVA

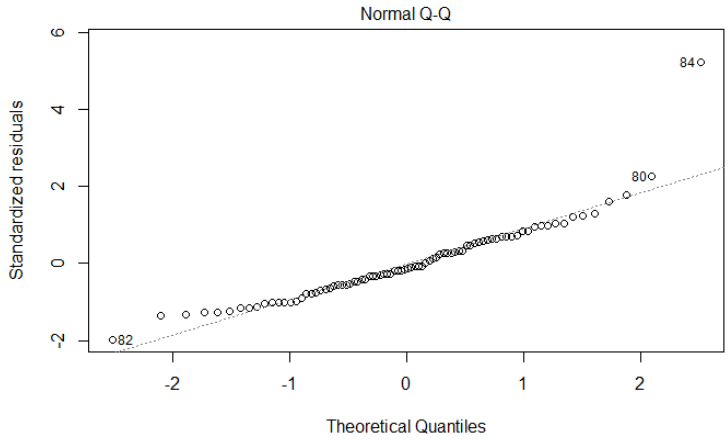
Variation	df	Sum of squares	Mean square	F-value	*p*-value
Day	3	10774	3591	19.06	$2.02e^{-09}$
Residuals	80	15071	188		

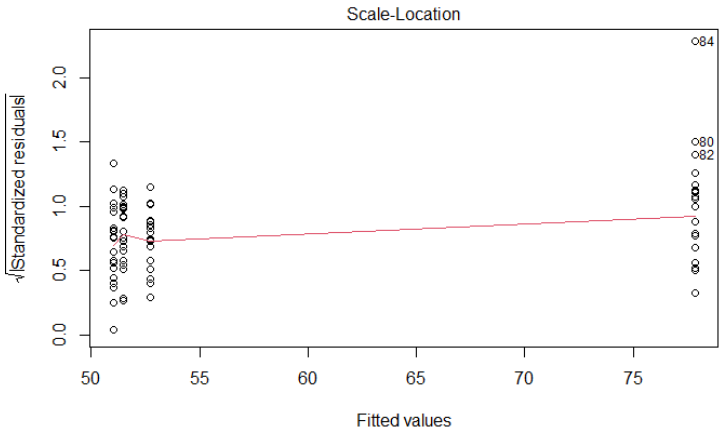
Kruskal-Wallis rank sum test

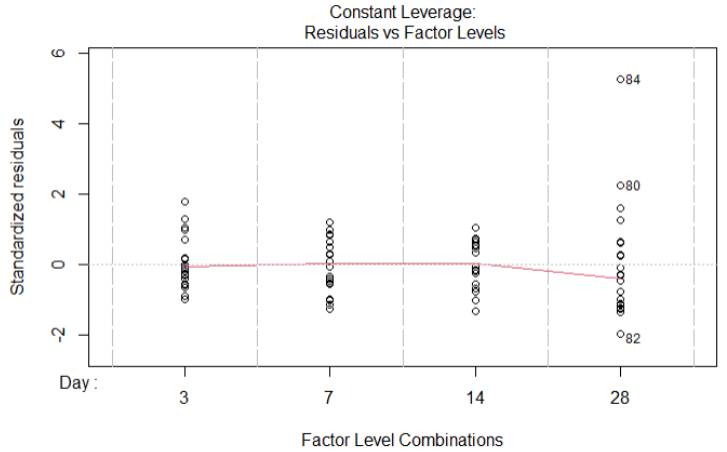
Data	χ^2	df	*p*-value
Protein content by Day	3.239	3	0.4329

Model plots


Residuals vs Fitted
Residuals
Fitted values
84
80
82


Normal Q-Q
Standardized residuals
Theoretical Quantiles
84
80
82


Scale-Location
√|Standardized residuals|
Fitted values
84
80
82


Constant Leverage:
Residuals vs Factor Levels
Standardized residuals
Day :
3
7
14
28
Factor Level Combinations
84
80
82

Fitted linear model for Φ_{PSII} by pH

Linear model formula: Φ_{PSII} = 0.32 - 0.02 (pH 7.68 no pre-exposure) + 0.01 (pH 7.68 6-h pre-exposure)

No effect of header tank was detected.

Coefficients	Estimate	Std. Error	t value	*p*-value	Significance
(Intercept)	0.31703	0.03084	10.278	<2e-16	***
pH 7.68 no pre-exposure	-0.02471	0.04439	-0.557	0.579	
pH 7.68 6-h pre-exposure	0.00562	0.04439	0.127	0.9	
Header tank effect	NA	NA	NA	NA	NA

Significance. Codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 0.1689 on 83 degrees of freedom
Multiple R-squared: 0.006148
Adjusted R-squared: -0.0178
F-statistic: 0.2567 on 2 and 83 df
p-value: 0.7742

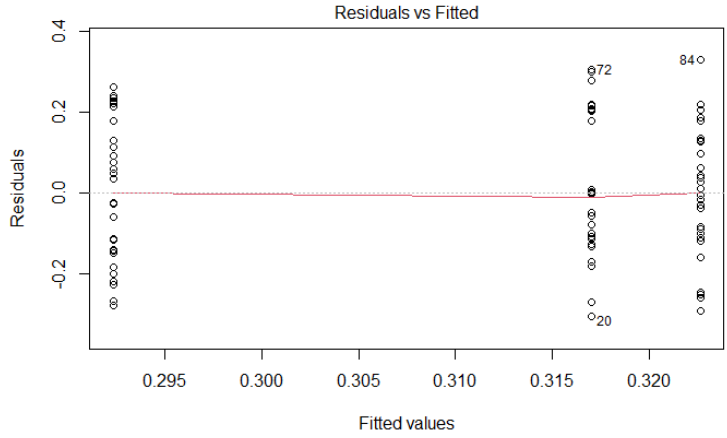
Model ANOVA

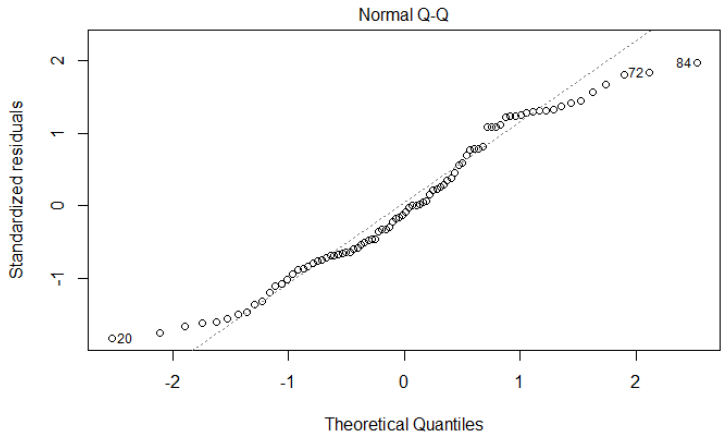
Variation	df	Sum of squares	Mean square	F-value	*p*-value
pH	2	0.0147	0.007327	0.257	0.774
Residuals	83	2.3689	0.028541		

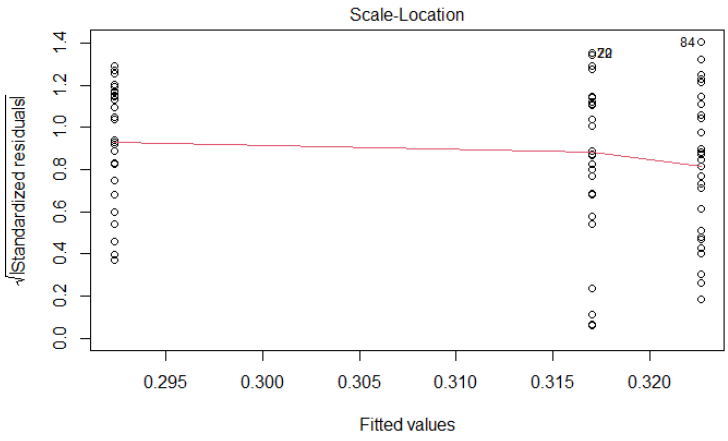
Kruskal-Wallis rank sum test

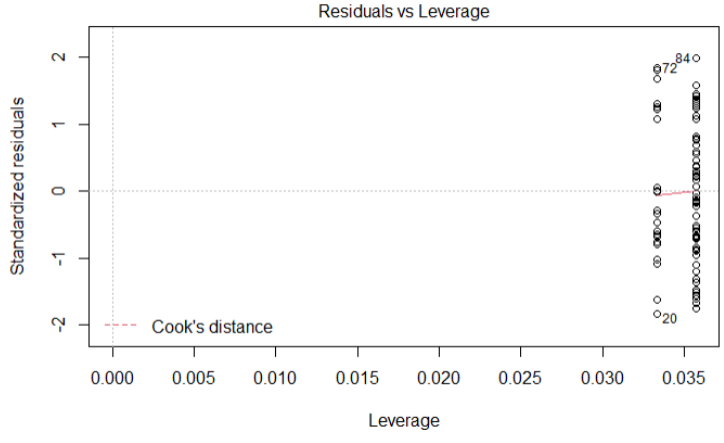
Data	χ^2	df	*p*-value
Φ_{PSII} by pH	0.4614	2	0.794

Model plots


Residuals vs Fitted
Residuals
Fitted values
72
84
20


Normal Q-Q
Standardized residuals
Theoretical Quantiles
72
84
20


Scale-Location
√|Standardized residuals|
Fitted values
84


Residuals vs Leverage
Standardized residuals
Leverage
Cook's distance
72
84
20

Fitted linear model for Φ_{PSII} over time

Linear model formula: $\Phi_{PSII} = 0.17 + 0.35$ (time)

No effect of header tank was detected.

Coefficients	Estimate	Std. Error	t value	*p*-value	Significance
(Intercept)	0.1718	0.02007	8.56	$5.41e^{-13}$	***
Week 2	0.03593	0.02838	1.266	0.209	
Week 3	0.15084	0.02806	5.375	$7.03e^{-07}$	***
Week 4	0.35829	0.02806	12.769	$2.00e^{-16}$	***
Header tank effect	NA	NA	NA	NA	NA

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

Residual standard error: 0.09198 on 82 degrees of freedom
Multiple R-squared: 0.709
Adjusted R-squared: 0.6983
F-statistic: 66.58 on 3 and 82 df
p-value: $2.20e^{-16}$

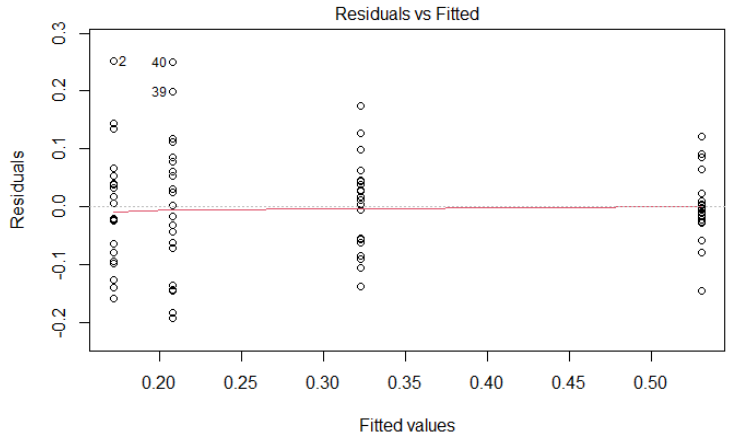
Model ANOVA

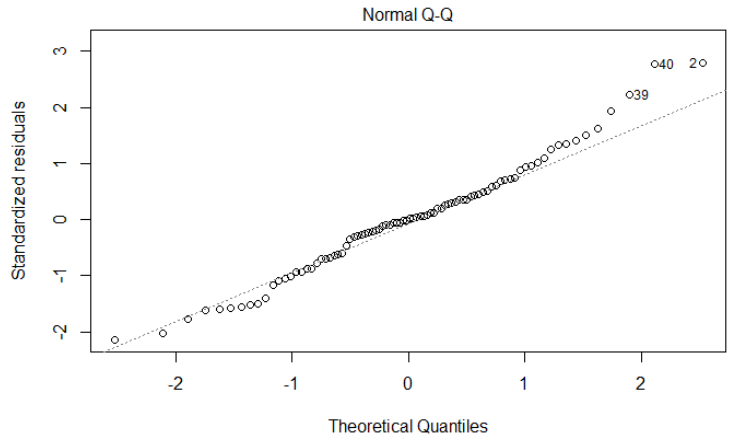
Variation	df	Sum of squares	Mean square	F-value	*p*-value
Week	3	1.6898	0.5633	66.58	$2e^{-16}$
Residuals	82	0.6937	0.0085		

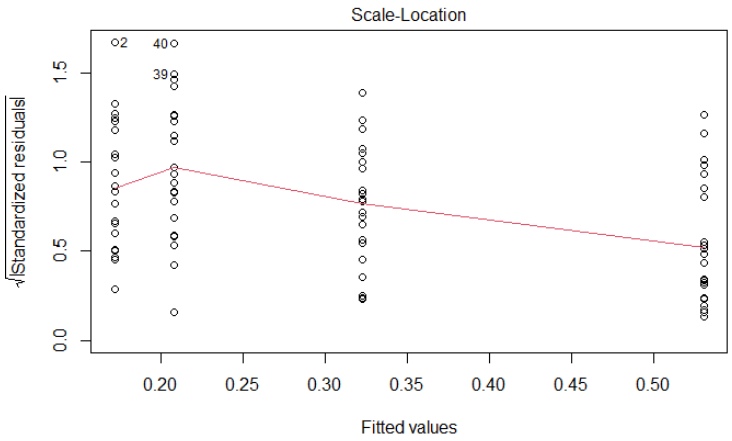
Kruskal-Wallis rank sum test

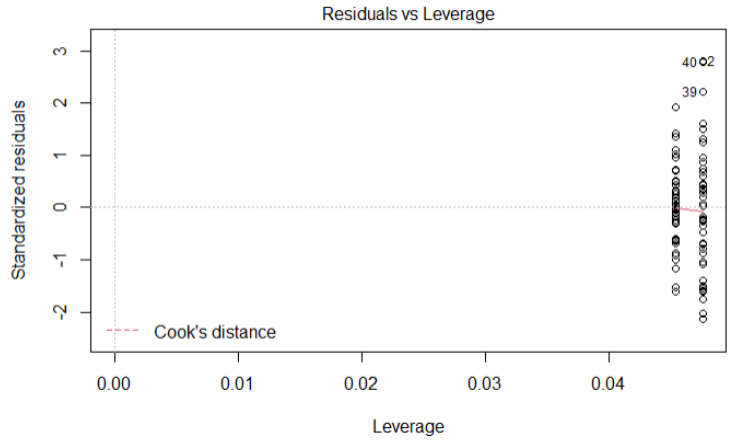
Data	χ^2	df	*p*-value
Φ_{PSII} by Week	1.5932	3	0.821

Model plots


Residuals vs Fitted
Residuals
Fitted values
2
40
39
0.20
0.25
0.30
0.35
0.40
0.45
0.50
-0.2
-0.1
0.0
0.1
0.2
0.3


Normal Q-Q
Standardized residuals
Theoretical Quantiles
40
2
39
-2
-1
0
1
2
3


Scale-Location
√|Standardized residuals|
Fitted values
2
40
39
0.20
0.25
0.30
0.35
0.40
0.45
0.50
0.0
0.5
1.0
1.5


Residuals vs Leverage
Standardized residuals
Leverage
Cook's distance
40
2
39
0.00
0.01
0.02
0.03
0.04
-2
-1
0
1
2
3

Fitted linear model for F_v/F_m by pH

Linear model formula: F_v/F_m = 0.31 + 0.004 (pH 7.68 no pre-exposure) - 0.002 (pH 7.68 6-h pre-exposure)

No effect of header tank was detected.

Coefficients	Estimate	Std. Error	t value	*p*-value	Significance
(Intercept)	0.305983	0.034034	8.991	$6.20e^{-14}$	***
pH 7.68 no pre-exposure	0.004977	0.049404	0.101	0.92	
pH 7.68 6-h pre-exposure	-0.00198	0.049404	-0.04	0.968	
Header tank effect	NA	NA	NA	NA	NA

Significance. Codes: 0 '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1

Residual standard error: 0.1895 on 84 degrees of freedom
Multiple R-squared: 0.0002395
Adjusted R-squared: -0.02356
F-statistic: 0.01006 on 2 and 84 df
p-value: 0.99

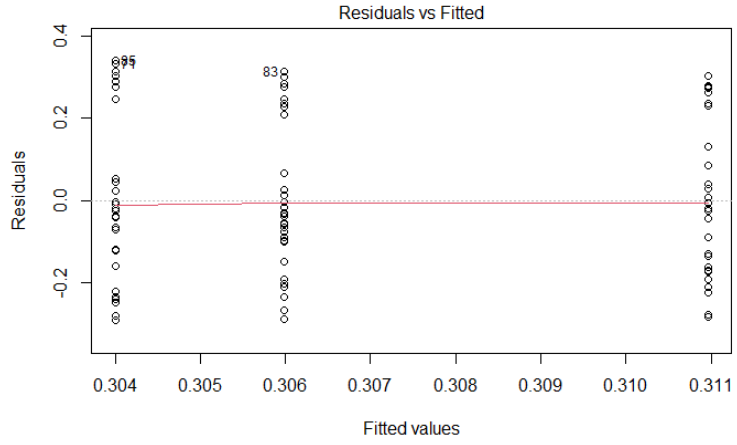
Model ANOVA

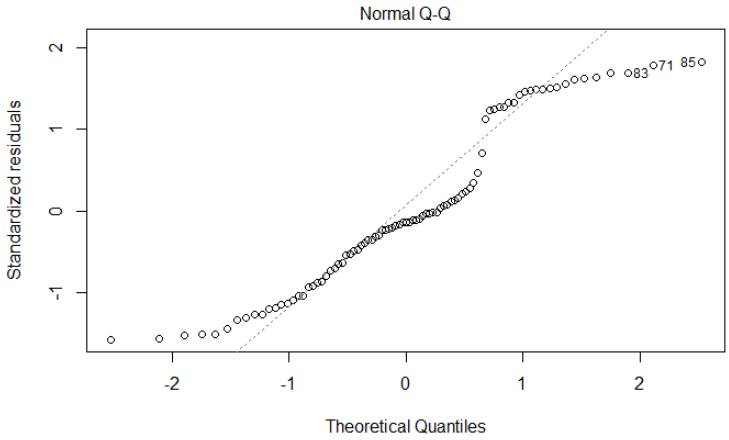
Variation	df	Sum of squares	Mean square	F-value	*p*-value
pH	2	0.0007	0.00036	0.01	0.99
Residuals	84	3.0162	0.03591		

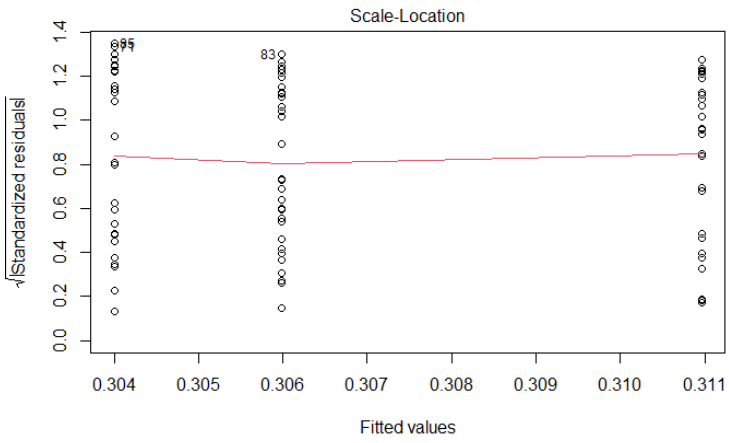
Kruskal-Wallis rank sum test

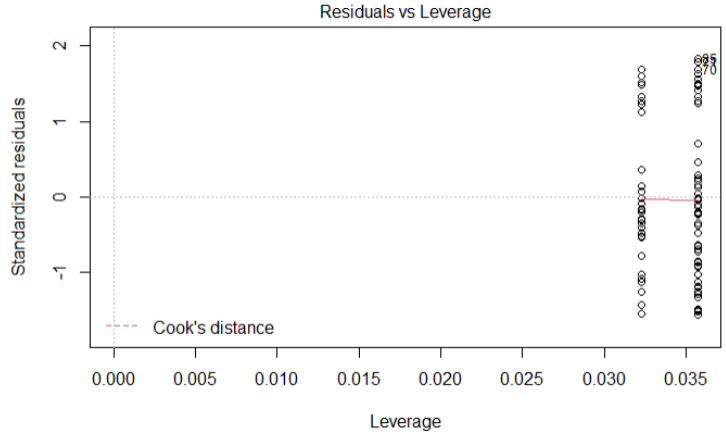
Data	χ^2	df	*p*-value
F_v/F_m by pH	0.044572	2	0.978

Model plots


Residuals vs Fitted
Residuals
Fitted values
0.304
0.305
0.306
0.307
0.308
0.309
0.310
0.311
-0.2
0.0
0.2
0.4
83


Normal Q-Q
Standardized residuals
Theoretical Quantiles
-2
-1
0
1
2
83
71
85


Scale-Location
√|Standardized residuals|
Fitted values
0.304
0.305
0.306
0.307
0.308
0.309
0.310
0.311
0.0
0.2
0.4
0.6
0.8
1.0
1.2
1.4
83


Residuals vs Leverage
Standardized residuals
Leverage
Cook's distance
0.000
0.005
0.010
0.015
0.020
0.025
0.030
0.035
-1
0
1
2

Fitted linear model for F_v/F_m over time

Linear model formula: $F_v/F_m = 0.08 + 0.5$ (time)

No effect of header tank was detected.

Coefficients	Estimate	Std. Error	t value	*p*-value	Significance
(Intercept)	0.08076	0.01145	7.057	$4.74e^{-10}$	***
Week 2	0.18547	0.016	11.591	$2.00e^{-16}$	***
Week 3	0.20728	0.016	12.954	$2.00e^{-16}$	***
Week 4	0.5017	0.016	31.354	$2.00e^{-16}$	***
Header tank effect	NA	NA	NA	NA	NA

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

Residual standard error: 0.05245 on 83 degrees of freedom
Multiple R-squared: 0.9243
Adjusted R-squared: 0.9216
F-statistic: 337.9 on 3 and 83 df
p-value: $2.20e^{-16}$

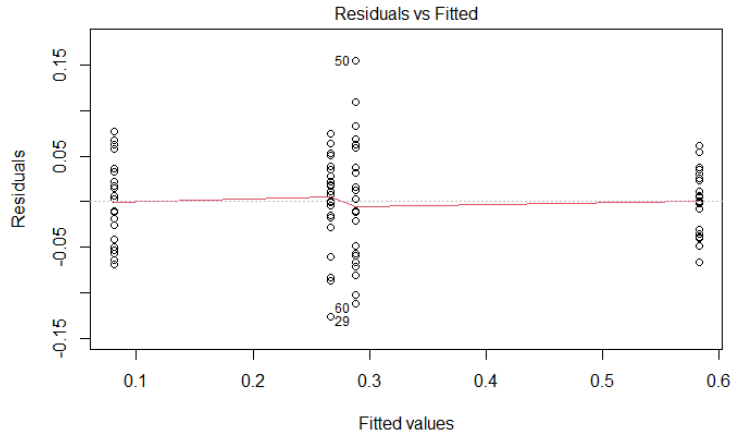
Model ANOVA

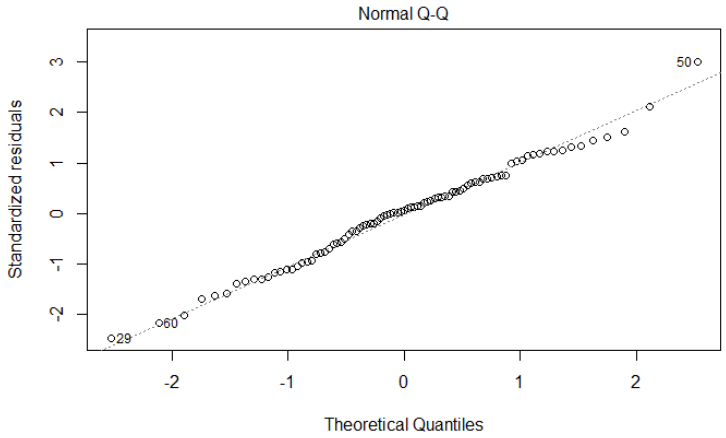
Variation	df	Sum of squares	Mean square	F-value	*p*-value
Week	3	2.7886	0.9295	337.9	$2e^{-16}$
Residuals	83	0.2283	0.0028		

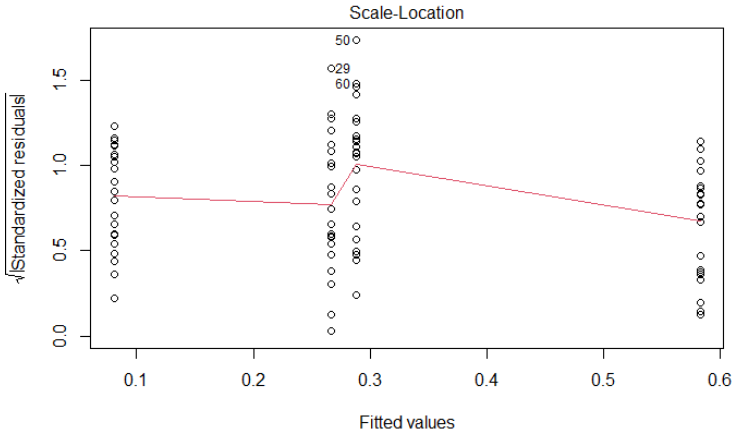
Kruskal-Wallis rank sum test

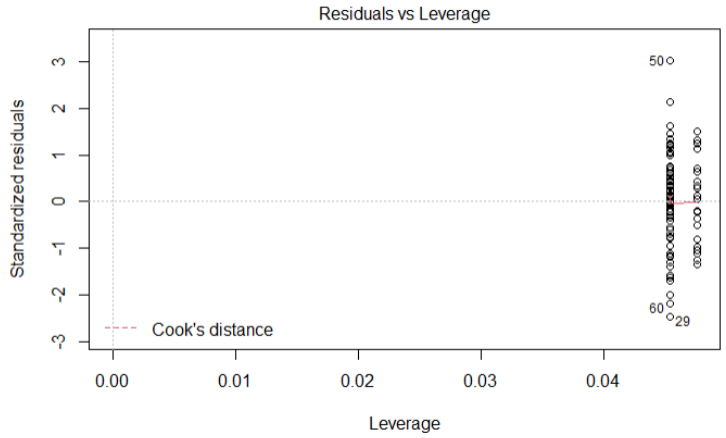
Data	χ^2	df	*p*-value
F_v/F_m by Week	0.7228	3	0.436

Model plots


Residuals vs Fitted
Residuals
Fitted values
50
60
29


Normal Q-Q
Standardized residuals
Theoretical Quantiles
50
60
29


Scale-Location
√|Standardized residuals|
Fitted values
50
29
60


Residuals vs Leverage
Standardized residuals
Leverage
Cook's distance
50
60
29