

CHRONIC CERIODAPHNIA SURVIVAL AND REPRODUCTION BIOASSAY

DATE: 10 January - 2018

STANDARD TOXICANT: Copper Chloride

ENDPOINT: SURVIVAL

NOEC = 10.00 ug/l

EC25 = 11.25 ug/l

EC50 = 17.50 ug/l

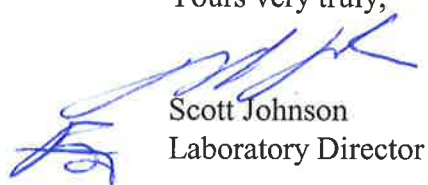
ENDPOINT: REPRODUCTION

NOEC = 10.00 ug/l

IC25 = 13.58 ug/l

IC50 = 19.14 ug/l

Yours very truly,



Scott Johnson
Laboratory Director

CETIS Summary Report

Report Date: 12 Feb-18 11:04 (p 1 of 2)
Test Code: CER011018 | 08-2407-5029

Ceriodaphnia 7-d Survival and Reproduction Test

Aquatic Bioassay & Consulting Labs, Inc.

Batch ID: 15-9098-8963	Test Type: Reproduction-Survival (7d)	Analyst:
Start Date: 10 Jan-18 15:15	Protocol: EPA/821/R-02-013 (2002)	Diluent: Laboratory Water
Ending Date: 17 Jan-18 14:10	Species: Ceriodaphnia dubia	Brine: Not Applicable
Duration: 6d 23h	Source: Aquatic Biosystems, CO	Age:
Sample ID: 02-6252-8216	Code: CER011018c	Client: Internal Lab
Sample Date: 10 Jan-18 15:15	Material: Copper chloride	Project:
Receipt Date:	Source: Reference Toxicant	
Sample Age: n/a	Station: REF TOX	

Multiple Comparison Summary

Analysis ID	Endpoint	Comparison Method	NOEL	LOEL	TOEL	TU	PMSD	✓
08-4111-7370	7d Survival Rate	Fisher Exact/Bonferroni-Holm Test	10	30	17.32		n/a	✓
20-1627-3979	Reproduction	Steel Many-One Rank Sum Test	10	30	17.32		41.0%	✓

Point Estimate Summary

Analysis ID	Endpoint	Point Estimate Method	Level	µg/L	95% LCL	95% UCL	TU	✓
21-1689-8833	7d Survival Rate	Linear Interpolation (ICPIN)	EC5	4	3.4	11		✓
			EC10	5	3.8	12		✓
			EC15	7.5	4.2	13		✓
			EC20	10	4.6	14		✓
			EC25	11.25	5.833	15		✓
			EC40	15	8.75	18		✓
06-1579-6172	Reproduction	Linear Interpolation (ICPIN)	EC50	17.5	10	20		✓
			IC5	7.807	5.531	11.03		
			IC10	10.24	6.062	12.05		
			IC15	11.36	6.597	13.08		
			IC20	12.47	7.129	14.1		
			IC25	13.58	7.662	15.13		
			IC40	16.92	9.258	18.2		
			IC50	19.14	11.2	20.25		

Test Acceptability

Analysis ID	Endpoint	Attribute	Test Stat	TAC Limits			Overlap	Decision
				Lower	Upper			
08-4111-7370	7d Survival Rate	Control Resp	1	0.8	>>		Yes	Passes Criteria
21-1689-8833	7d Survival Rate	Control Resp	1	0.8	>>		Yes	Passes Criteria
06-1579-6172	Reproduction	Control Resp	20.9	15	>>		Yes	Passes Criteria
20-1627-3979	Reproduction	Control Resp	20.9	15	>>		Yes	Passes Criteria
20-1627-3979	Reproduction	PMSD	0.4098	0.13	0.47		Yes	Passes Criteria

7d Survival Rate Summary

Conc-µg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	10	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	0.00%
3		10	1.0000	1.0000	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	0.00%
5		10	0.9000	0.6738	1.0000	0.0000	1.0000	0.1000	0.3162	35.14%	10.00%
10		10	0.8000	0.4984	1.0000	0.0000	1.0000	0.1333	0.4216	52.70%	20.00%
30		10	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000		100.00%
50		10	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000		100.00%

Reproduction Summary

Conc-µg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	10	20.9	19.66	22.14	18	24	0.5467	1.729	8.27%	0.00%
3		10	25.4	18.67	32.13	7	37	2.975	9.407	37.03%	-21.53%
5		10	27.8	24.4	31.2	19	34	1.504	4.756	17.11%	-33.01%
10		10	22.5	11.05	33.95	0	46	5.062	16.01	71.15%	-7.66%
30		10	0.3	-0.3786	0.9786	0	3	0.3	0.9487	316.23%	98.56%
50		10	0	0	0	0	0	0	0		100.00%

CETIS Analytical Report

Report Date: 12 Feb-18 11:04 (p 1 of 2)
Test Code: CER011018 | 08-2407-5029

Ceriodaphnia 7-d Survival and Reproduction Test

Aquatic Bioassay & Consulting Labs, Inc.

Analysis ID: 20-1627-3979	Endpoint: Reproduction	CETIS Version: CETISv1.9.2
Analyzed: 31 Jan-18 13:52	Analysis: Nonparametric-Control vs Treatments	Official Results: Yes
Batch ID: 15-9098-8963	Test Type: Reproduction-Survival (7d)	Analyst:
Start Date: 10 Jan-18 15:15	Protocol: EPA/821/R-02-013 (2002)	Diluent: Laboratory Water
Ending Date: 17 Jan-18 14:10	Species: Ceriodaphnia dubia	Brine: Not Applicable
Duration: 6d 23h	Source: Aquatic Biosystems, CO	Age:
Sample ID: 02-6252-8216	Code: CER011018c	Client: Internal Lab
Sample Date: 10 Jan-18 15:15	Material: Copper chloride	Project:
Receipt Date:	Source: Reference Toxicant	
Sample Age: n/a	Station: REF TOX	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU	PMSD
Untransformed	C > T	10	30	17.32		40.98%

Steel Many-One Rank Sum Test

Control	vs	Conc-µg/L	Test Stat	Critical	Ties	DF	P-Type	P-Value	Decision(α:5%)
Negative Control		3	125	76	2	18	Asymp	0.9958	Non-Significant Effect
		5	145	76	2	18	Asymp	1.0000	Non-Significant Effect
		10	105	76	2	18	Asymp	0.8000	Non-Significant Effect
		30*	55	76	0	18	Asymp	3.1E-04	Significant Effect

Test Acceptability Criteria

TAC Limits

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	20.9	15	>>	Yes	Passes Criteria
PMSD	0.4098	0.13	0.47	Yes	Passes Criteria

ANOVA Table

Source	Sum Squares	Mean Square	DF	F Stat	P-Value	Decision(α:5%)
Between	4832.28	1208.07	4	16.27	<1.0E-37	Significant Effect
Error	3341.5	74.2556	45			
Total	8173.78		49			

Distributional Tests

Attribute	Test	Test Stat	Critical	P-Value	Decision(α:1%)
Variances	Bartlett Equality of Variance Test	63.77	13.28	<1.0E-37	Unequal Variances
Variances	Levene Equality of Variance Test	14.39	3.767	1.2E-07	Unequal Variances
Variances	Mod Levene Equality of Variance Test	11.7	3.767	1.3E-06	Unequal Variances
Distribution	Anderson-Darling A2 Normality Test	2.077	3.878	<1.0E-37	Non-Normal Distribution
Distribution	D'Agostino Kurtosis Test	2.367	2.576	0.0179	Normal Distribution
Distribution	D'Agostino Skewness Test	0.9951	2.576	0.3197	Normal Distribution
Distribution	D'Agostino-Pearson K2 Omnibus Test	6.594	9.21	0.0370	Normal Distribution
Distribution	Kolmogorov-Smirnov D Test	0.1766	0.1453	4.8E-04	Non-Normal Distribution
Distribution	Shapiro-Wilk W Normality Test	0.9116	0.9367	0.0012	Non-Normal Distribution

Reproduction Summary

Conc-µg/L	Code	Count	Mean	95% LCL	95% UCL	Median	Min	Max	Std Err	CV%	%Effect
0	N	10	20.9	19.66	22.14	21	18	24	0.5467	8.27%	0.00%
3		10	25.4	18.67	32.13	28.5	7	37	2.975	37.03%	-21.53%
5		10	27.8	24.4	31.2	28	19	34	1.504	17.11%	-33.01%
10		10	22.5	11.05	33.95	21	0	46	5.062	71.15%	-7.66%
30		10	0.3	-0.3786	0.9786	0	0	3	0.3	316.23%	98.56%
50		10	0	0	0	0	0	0	0		100.00%

CETIS Analytical Report

Report Date: 12 Feb-18 11:04 (p 1 of 4)
Test Code: CER011018 | 08-2407-5029

Ceriodaphnia 7-d Survival and Reproduction Test

Aquatic Bioassay & Consulting Labs, Inc.

Analysis ID: 21-1689-8833	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.2
Analyzed: 31 Jan-18 13:52	Analysis: Linear Interpolation (ICPIN)	Official Results: Yes
Batch ID: 15-9098-8963	Test Type: Reproduction-Survival (7d)	Analyst:
Start Date: 10 Jan-18 15:15	Protocol: EPA/821/R-02-013 (2002)	Diluent: Laboratory Water
Ending Date: 17 Jan-18 14:10	Species: Ceriodaphnia dubia	Brine: Not Applicable
Duration: 6d 23h	Source: Aquatic Biosystems, CO	Age:
Sample ID: 02-6252-8216	Code: CER011018c	Client: Internal Lab
Sample Date: 10 Jan-18 15:15	Material: Copper chloride	Project:
Receipt Date:	Source: Reference Toxicant	
Sample Age: n/a	Station: REF TOX	

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	0	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

TAC Limits

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	1	0.8	>>	Yes	Passes Criteria

Point Estimates

Level	µg/L	95% LCL	95% UCL
EC5	4	3.4	11
EC10	5	3.8	12
EC15	7.5	4.2	13
EC20	10	4.6	14
EC25	11.25	5.833	15
EC40	15	8.75	18
EC50	17.5	10	20

7d Survival Rate Summary

Calculated Variate(A/B)

Conc-µg/L	Code	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect	A	B
0	N	10	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	0.0%	10	10
3		10	1.0000	1.0000	1.0000	0.0000	0.0000	0.00%	0.0%	10	10
5		10	0.9000	0.0000	1.0000	0.1000	0.3162	35.14%	10.0%	9	10
10		10	0.8000	0.0000	1.0000	0.1333	0.4216	52.70%	20.0%	8	10
30		10	0.0000	0.0000	0.0000	0.0000	0.0000		100.0%	0	10
50		10	0.0000	0.0000	0.0000	0.0000	0.0000		100.0%	0	10

7d Survival Rate Detail

Conc-µg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	N	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
5		1.0000	1.0000	1.0000	0.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
10		1.0000	1.0000	0.0000	1.0000	1.0000	0.0000	1.0000	1.0000	1.0000	1.0000
30		0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
50		0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000

7d Survival Rate Binomials

Conc-µg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	N	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
3		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
5		1/1	1/1	1/1	0/1	1/1	1/1	1/1	1/1	1/1	1/1
10		1/1	1/1	0/1	1/1	1/1	0/1	1/1	1/1	1/1	1/1
30		0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1
50		0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1

CETIS Analytical Report

Report Date: 12 Feb-18 11:04 (p 3 of 4)
Test Code: CER011018 | 08-2407-5029

Ceriodaphnia 7-d Survival and Reproduction Test

Aquatic Bioassay & Consulting Labs, Inc.

Analysis ID: 06-1579-6172	Endpoint: Reproduction	CETIS Version: CETISv1.9.2
Analyzed: 31 Jan-18 13:52	Analysis: Linear Interpolation (ICPIN)	Official Results: Yes
Batch ID: 15-9098-8963	Test Type: Reproduction-Survival (7d)	Analyst:
Start Date: 10 Jan-18 15:15	Protocol: EPA/821/R-02-013 (2002)	Diluent: Laboratory Water
Ending Date: 17 Jan-18 14:10	Species: Ceriodaphnia dubia	Brine: Not Applicable
Duration: 6d 23h	Source: Aquatic Biosystems, CO	Age:
Sample ID: 02-6252-8216	Code: CER011018c	Client: Internal Lab
Sample Date: 10 Jan-18 15:15	Material: Copper chloride	Project:
Receipt Date:	Source: Reference Toxicant	
Sample Age: n/a	Station: REF TOX	

Linear Interpolation Options

X Transform	Y Transform	Seed	Resamples	Exp 95% CL	Method
Linear	Linear	1392	280	Yes	Two-Point Interpolation

Test Acceptability Criteria

TAC Limits

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	20.9	15	>>	Yes	Passes Criteria

Point Estimates

Level	µg/L	95% LCL	95% UCL
IC5	7.807	5.531	11.03
IC10	10.24	6.062	12.05
IC15	11.36	6.597	13.08
IC20	12.47	7.129	14.1
IC25	13.58	7.662	15.13
IC40	16.92	9.258	18.2
IC50	19.14	11.2	20.25

Reproduction Summary

Calculated Variate

Conc-µg/L	Code	Count	Mean	Min	Max	Std Err	Std Dev	CV%	%Effect
0	N	10	20.9	18	24	0.5467	1.729	8.27%	0.0%
3		10	25.4	7	37	2.975	9.407	37.03%	-21.53%
5		10	27.8	19	34	1.504	4.756	17.11%	-33.01%
10		10	22.5	0	46	5.062	16.01	71.15%	-7.66%
30		10	0.3	0	3	0.3	0.9487	316.20%	98.56%
50		10	0	0	0	0	0		100.0%

Reproduction Detail

Conc-µg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	N	19	21	21	18	20	22	20	24	22	22
3		18	15	30	37	24	29	28	32	34	7
5		28	24	34	28	28	23	34	29	31	19
10		36	14	0	18	14	0	37	46	36	24
30		3	0	0	0	0	0	0	0	0	0
50		0	0	0	0	0	0	0	0	0	0

CETIS Analytical Report

Report Date: 12 Feb-18 11:04 (p 1 of 2)
Test Code: CER011018 | 08-2407-5029

Ceriodaphnia 7-d Survival and Reproduction Test

Aquatic Bioassay & Consulting Labs, Inc.

Analysis ID: 08-4111-7370	Endpoint: 7d Survival Rate	CETIS Version: CETISv1.9.2
Analyzed: 31 Jan-18 13:52	Analysis: STP 2xK Contingency Tables	Official Results: Yes
Batch ID: 15-9098-8963	Test Type: Reproduction-Survival (7d)	Analyst:
Start Date: 10 Jan-18 15:15	Protocol: EPA/821/R-02-013 (2002)	Diluent: Laboratory Water
Ending Date: 17 Jan-18 14:10	Species: Ceriodaphnia dubia	Brine: Not Applicable
Duration: 6d 23h	Source: Aquatic Biosystems, CO	Age:
Sample ID: 02-6252-8216	Code: CER011018c	Client: Internal Lab
Sample Date: 10 Jan-18 15:15	Material: Copper chloride	Project:
Receipt Date:	Source: Reference Toxicant	
Sample Age: n/a	Station: REF TOX	

Data Transform	Alt Hyp	NOEL	LOEL	TOEL	TU
Untransformed	C > T	10	30	17.32	

Fisher Exact/Bonferroni-Holm Test

Control	vs	Group	Test Stat	P-Type	P-Value	Decision(α:5%)
Negative Control		3	1.0000	Exact	1.0000	Non-Significant Effect
		5	0.5000	Exact	1.0000	Non-Significant Effect
		10	0.2368	Exact	0.7105	Non-Significant Effect
		30*	0.0000	Exact	2.7E-05	Significant Effect
		50*	0.0000	Exact	2.7E-05	Significant Effect

Test Acceptability Criteria

TAC Limits

Attribute	Test Stat	Lower	Upper	Overlap	Decision
Control Resp	1	0.8	>>	Yes	Passes Criteria

Data Summary

Conc-μg/L	Code	NR	R	NR + R	Prop NR	Prop R	%Effect
0	N	10	0	10	1	0	0.0%
3		10	0	10	1	0	0.0%
5		9	1	10	0.9	0.1	10.0%
10		8	2	10	0.8	0.2	20.0%
30		0	10	10	0	1	100.0%
50		0	10	10	0	1	100.0%

7d Survival Rate Detail

Conc-μg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	N	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
3		1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
5		1.0000	1.0000	1.0000	0.0000	1.0000	1.0000	1.0000	1.0000	1.0000	1.0000
10		1.0000	1.0000	0.0000	1.0000	1.0000	0.0000	1.0000	1.0000	1.0000	1.0000
30		0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
50		0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000

7d Survival Rate Binomials

Conc-μg/L	Code	Rep 1	Rep 2	Rep 3	Rep 4	Rep 5	Rep 6	Rep 7	Rep 8	Rep 9	Rep 10
0	N	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
3		1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1	1/1
5		1/1	1/1	1/1	0/1	1/1	1/1	1/1	1/1	1/1	1/1
10		1/1	1/1	0/1	1/1	1/1	0/1	1/1	1/1	1/1	1/1
30		0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1
50		0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1	0/1

CETIS Measurement Report

Report Date: 12 Feb-18 11:04 (p 1 of 2)
Test Code: CER011018 | 08-2407-5029

Ceriodaphnia 7-d Survival and Reproduction Test

Aquatic Bioassay & Consulting Labs, Inc.

Batch ID: 15-9098-8963	Test Type: Reproduction-Survival (7d)	Analyst:
Start Date: 10 Jan-18 15:15	Protocol: EPA/821/R-02-013 (2002)	Diluent: Laboratory Water
Ending Date: 17 Jan-18 14:10	Species: Ceriodaphnia dubia	Brine: Not Applicable
Duration: 6d 23h	Source: Aquatic Biosystems, CO	Age:
Sample ID: 02-6252-8216	Code: CER011018c	Client: Internal Lab
Sample Date: 10 Jan-18 15:15	Material: Copper chloride	Project:
Receipt Date:	Source: Reference Toxicant	
Sample Age: n/a	Station: REF TOX	

Alkalinity (CaCO3)-mg/L

Conc-µg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	8	66	62.9	69.1	60	68	1.309	3.703	5.61%	0
50		8	64	64	64	64	64	0	0	0.0%	0
Overall		16	65	63.54	66.46	60	68	0.6831	2.733	4.20%	0 (0%)

Conductivity-µmhos

Conc-µg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	8	338.8	333.5	344	330	348	2.226	6.296	1.86%	0
3		8	339.2	332	346.5	327	351	3.069	8.681	2.56%	0
5		8	335	329.8	340.2	323	345	2.196	6.211	1.85%	0
10		8	335.2	333.3	337.2	333	340	0.8183	2.315	0.69%	0
30		4	332.8	330.7	334.8	331	334	0.6292	1.258	0.38%	0
50		2	332	319.3	344.7	331	333	1	1.414	0.43%	0
Overall		38	336.3	334.4	338.3	323	351	0.9786	6.033	1.79%	0 (0%)

Dissolved Oxygen-mg/L

Conc-µg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	8	7.55	7.345	7.755	7.1	7.9	0.0866	0.2449	3.24%	0
3		8	7.7	7.114	8.286	6.1	8.3	0.2478	0.701	9.1%	0
5		8	7.712	7.07	8.355	6	8.4	0.2715	0.768	9.96%	0
10		8	7.712	7.075	8.35	6	8.4	0.2695	0.7624	9.89%	0
30		4	7.925	7.197	8.653	7.4	8.4	0.2287	0.4573	5.77%	0
50		1	8.3			8.3	8.3	0	0	0.0%	0
Overall		37	7.714	7.51	7.917	6	8.4	0.1004	0.6106	7.92%	0 (0%)

Hardness (CaCO3)-mg/L

Conc-µg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	8	97.75	95.81	99.69	94	99	0.8183	2.315	2.37%	0
50		1	97			97	97	0	0	0.0%	0
Overall		9	97.67	95.99	99.34	94	99	0.7265	2.179	2.23%	0 (0%)

pH-Units

Conc-µg/L	Code	Count	Mean	95% LCL	95% UCL	Min	Max	Std Err	Std Dev	CV%	QA Count
0	N	8	8.038	7.871	8.204	7.7	8.4	0.07055	0.1996	2.48%	0
3		8	7.875	7.715	8.035	7.6	8.1	0.06748	0.1909	2.42%	0
5		8	7.963	7.874	8.051	7.8	8.1	0.0375	0.1061	1.33%	0
10		8	7.963	7.886	8.039	7.8	8.1	0.03239	0.09161	1.15%	0
30		4	8	8	8	8	8	0	0	0.0%	0
50		1	8			8	8	0	0	0.0%	0
Overall		37	7.965	7.916	8.014	7.6	8.4	0.02427	0.1476	1.85%	0 (0%)