

Sample name: **M06**
 Assay name: **UV-metric pKa**
 Assay ID: **171-18022**
 Filename: **C:\Sirius_T3\Mehtap\20170918_exp04_uv_M01-M14\171-18022_M06_UV-metric pKa.t3r**

Experiment start time: **9/18/2017 11:06:44 PM**
 Analyst: **Dorothy Levorse**
 Instrument ID: **T311053**

Results

pKa 1 **3.11**
 pKa 2 **11.74**
 RMSD **0.022 0.017 0.009**
 Chi squared **0.0648**
 PCA calculated number of pKas **2**
 Average ionic strength **0.183 M**
 Average temperature **24.9°C**
 Analyte concentration range **86.8 µM to 70.3 µM**

Number of pKas source **Predicted**
 Wavelength clipping **230.0 nm to 450.0 nm**
 pH clipping **1.550 to 13.274**

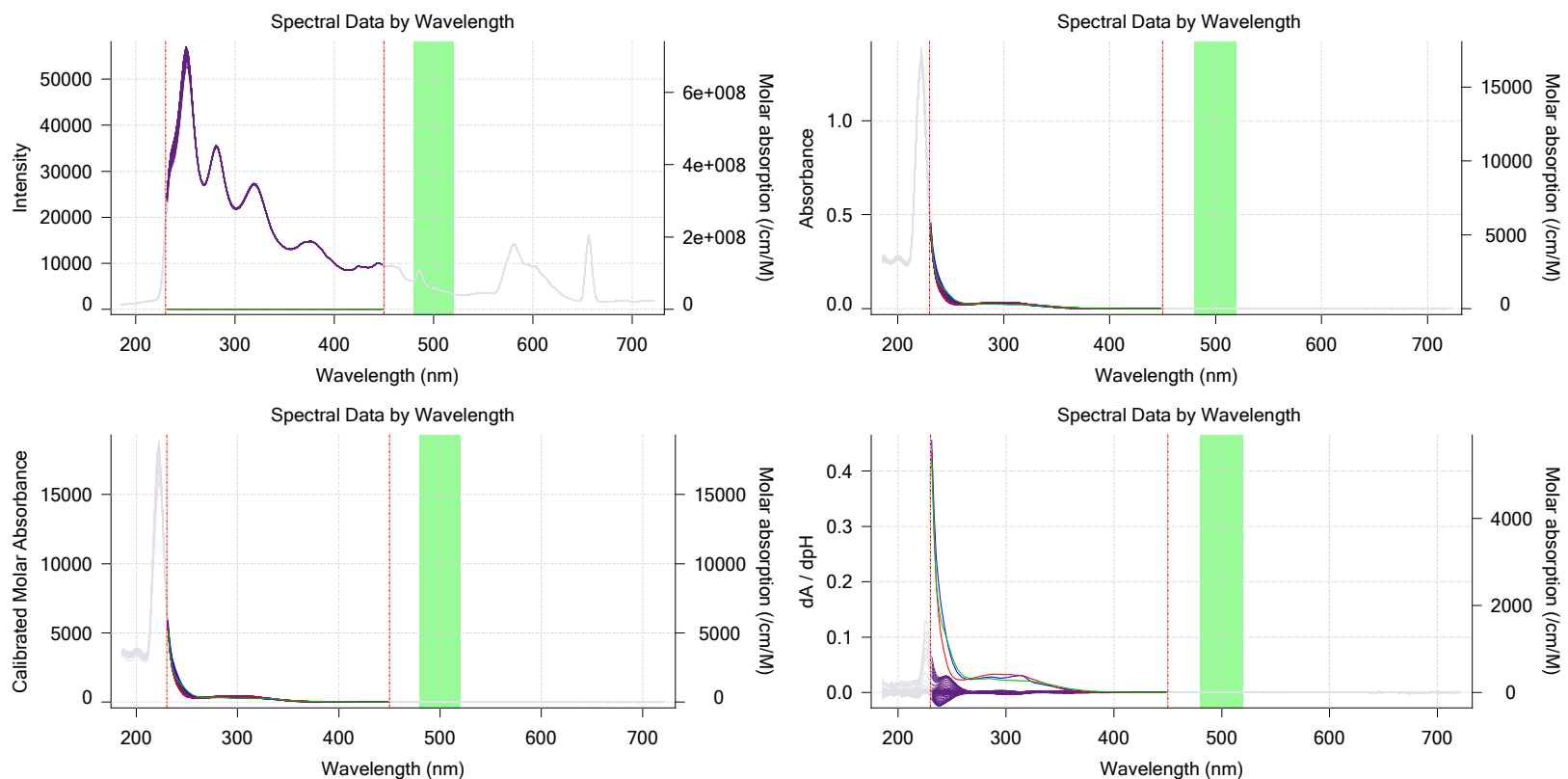
Warnings and errors

Errors None
 Warnings None

Assay Settings

Setting	Value	Original Value	Date/Time changed	Imported from
Buffer in use	Yes			
Buffer type	Phosphate Buffer			
Assay Medium				
Volume of buffer introduced	0.025000 mL			
Add buffer manually	Manual			

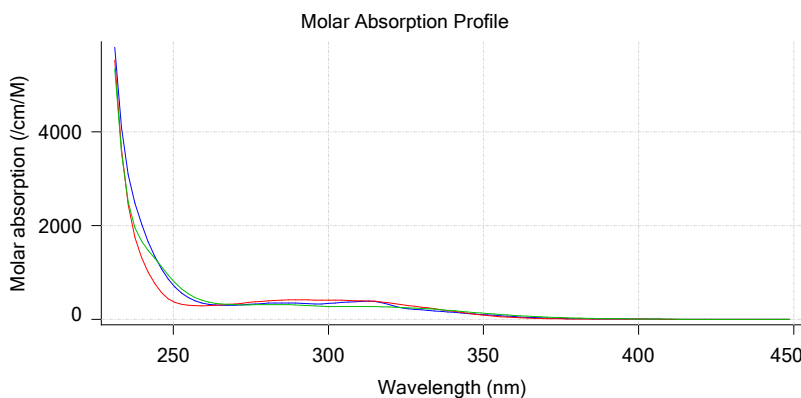
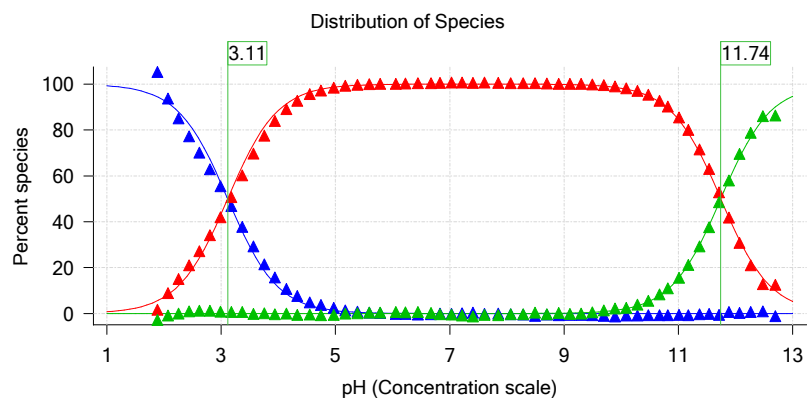
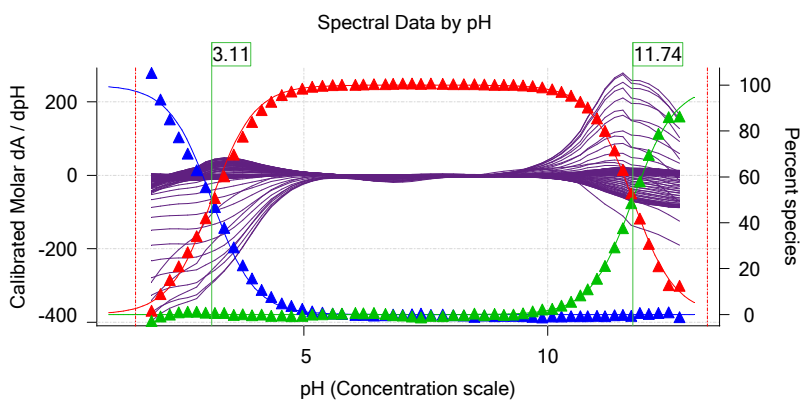
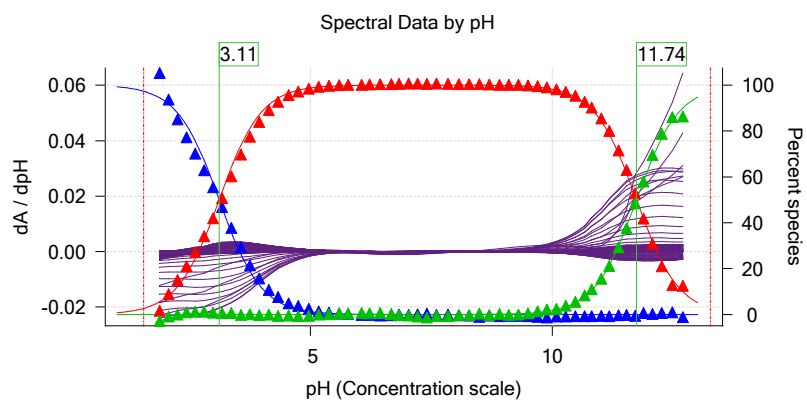
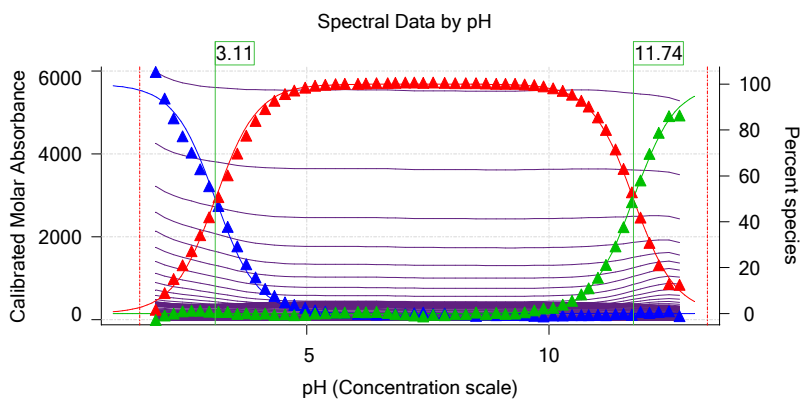
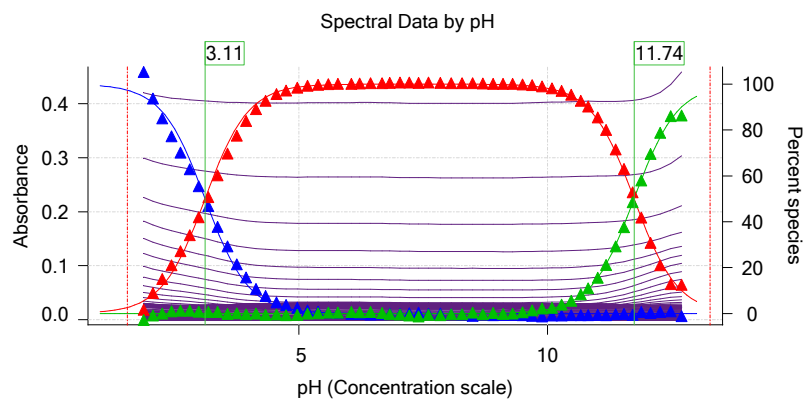
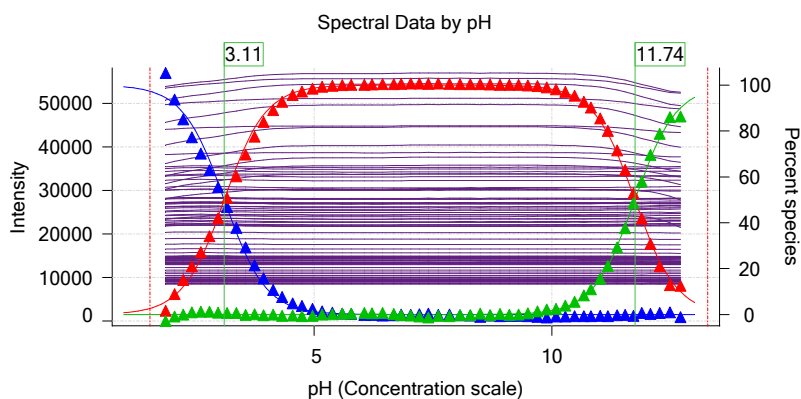
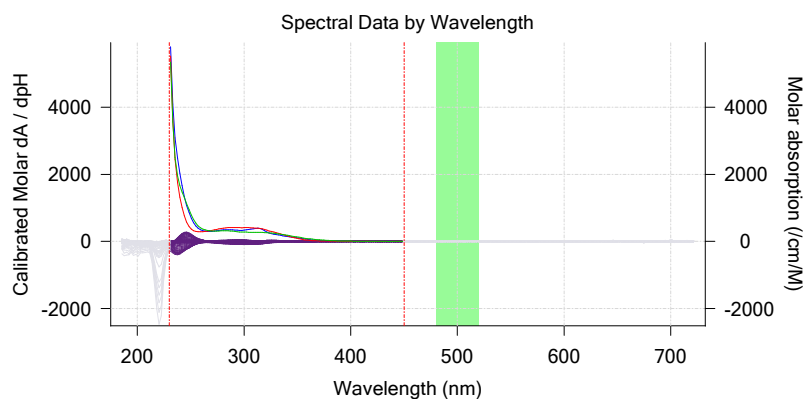
Graphs



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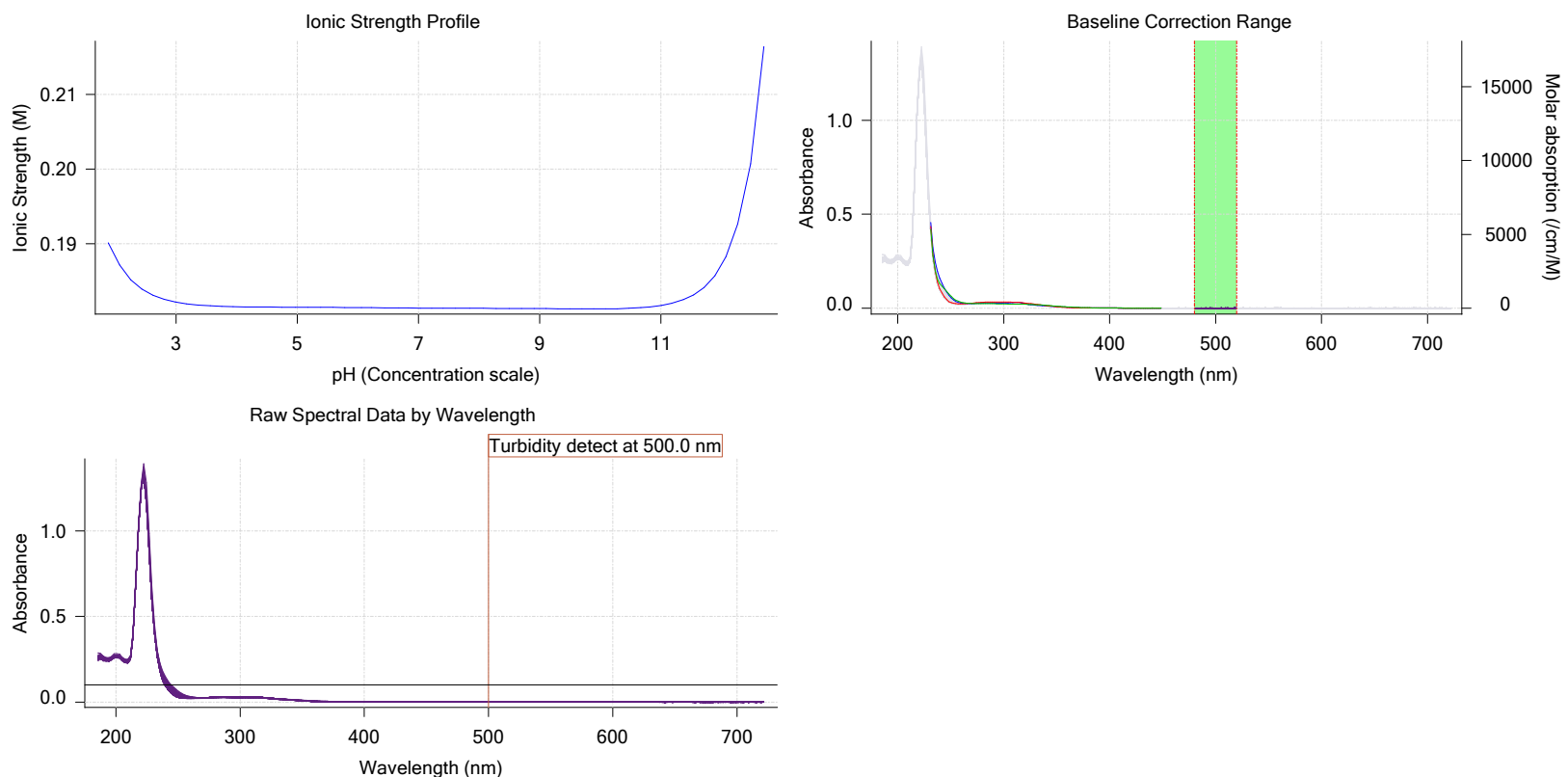
Graphs (continued)



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Graphs (continued)



Events

Time	Event	Water	Acid	Base	Buffer	pH	dpH/dt	pH R-squared	pH SD
2:36.5	Dark spectrum								
2:37.9	Reference spectrum								
3:05.6	Volume reset due to vial change								
4:22.1	Initial pH = 12.84								
5:32.2	Data point 4	1.20002 mL	0.00000 mL	0.30000 mL	0.02500 mL	12.774	0.02017	0.91537	0.001
6:02.7	Data point 5	1.20002 mL	0.10398 mL	0.30000 mL	0.02500 mL	12.576	0.00713	0.59374	0.000
6:20.9	Data point 6	1.20002 mL	0.17298 mL	0.30000 mL	0.02500 mL	12.373	-0.00132	0.01866	0.000
6:38.3	Data point 7	1.20002 mL	0.21432 mL	0.30000 mL	0.02500 mL	12.185	-0.01001	0.40116	0.000
6:55.5	Data point 8	1.20002 mL	0.24069 mL	0.30000 mL	0.02500 mL	12.004	-0.00876	0.27223	0.000
7:12.4	Data point 9	1.20002 mL	0.25795 mL	0.30000 mL	0.02500 mL	11.830	-0.01985	0.94800	0.001
7:29.3	Data point 10	1.20002 mL	0.26938 mL	0.30000 mL	0.02500 mL	11.657	-0.01482	0.92354	0.000
7:46.2	Data point 11	1.20002 mL	0.27705 mL	0.30000 mL	0.02500 mL	11.492	-0.01872	0.94631	0.000
8:13.3	Data point 12	1.20002 mL	0.28300 mL	0.30000 mL	0.02500 mL	11.296	-0.01281	0.93044	0.000
8:29.9	Data point 13	1.20002 mL	0.28641 mL	0.30000 mL	0.02500 mL	11.131	-0.02353	0.94935	0.001
8:56.7	Data point 14	1.20002 mL	0.28913 mL	0.30000 mL	0.02500 mL	10.937	-0.01663	0.91676	0.000
9:13.2	Data point 15	1.20002 mL	0.29066 mL	0.30000 mL	0.02500 mL	10.789	-0.01188	0.89848	0.000
9:45.2	Data point 16	1.20002 mL	0.29219 mL	0.30000 mL	0.02500 mL	10.597	-0.00796	0.77281	0.000
10:17.1	Data point 17	1.20002 mL	0.29332 mL	0.30000 mL	0.02500 mL	10.401	-0.01325	0.80185	0.000
10:49.3	Data point 18	1.20002 mL	0.29424 mL	0.30000 mL	0.02500 mL	10.207	-0.01302	0.92127	0.000
11:26.2	Data point 19	1.20002 mL	0.29508 mL	0.30000 mL	0.02500 mL	10.008	-0.00532	0.54502	0.000
12:03.1	Data point 20	1.20002 mL	0.29586 mL	0.30000 mL	0.02500 mL	9.812	-0.00735	0.75263	0.000
12:40.0	Data point 21	1.20002 mL	0.29657 mL	0.30000 mL	0.02500 mL	9.614	-0.00774	0.73260	0.000
13:21.8	Data point 22	1.20002 mL	0.29720 mL	0.30000 mL	0.02500 mL	9.417	-0.00945	0.69389	0.000
14:03.9	Data point 23	1.20002 mL	0.29774 mL	0.30000 mL	0.02500 mL	9.220	-0.01080	0.67173	0.000
14:40.8	Data point 24	1.20002 mL	0.29817 mL	0.30000 mL	0.02500 mL	9.024	-0.02801	0.89926	0.001
15:22.6	Data point 25	1.20002 mL	0.29852 mL	0.30000 mL	0.02500 mL	8.815	-0.03901	0.96161	0.001

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Events (continued)

Time	Event	Water	Acid	Base	Buffer	pH	dpH/dt	pH R-squared	pH SD	dpH/dt time
16:04.3	Data point 26	1.20002 mL	0.29878 mL	0.30000 mL	0.02500 mL	8.609	-0.05228	0.97200	0.00262	10.0 s
16:41.0	Data point 27	1.20002 mL	0.29896 mL	0.30000 mL	0.02500 mL	8.397	-0.09065	0.97642	0.00455	10.0 s
17:17.7	Data point 28	1.20002 mL	0.29911 mL	0.30000 mL	0.02500 mL	8.180	-0.08983	0.95613	0.00453	10.0 s
17:54.3	Data point 29	1.20002 mL	0.29922 mL	0.30000 mL	0.02500 mL	7.960	-0.09795	0.96242	0.00493	10.0 s
18:31.1	Data point 30	1.20002 mL	0.29934 mL	0.30000 mL	0.02500 mL	7.721	-0.00751	0.21649	0.00080	10.0 s
19:07.9	Data point 31	1.20002 mL	0.29946 mL	0.30000 mL	0.02500 mL	7.516	0.05400	0.95251	0.00273	10.0 s
19:49.7	Data point 32	1.20002 mL	0.29962 mL	0.30000 mL	0.02500 mL	7.328	0.09950	0.99611	0.00497	15.5 s
20:37.3	Data point 33	1.20002 mL	0.29986 mL	0.30000 mL	0.02500 mL	7.142	0.09704	0.99260	0.00487	22.0 s
21:36.4	Data point 34	1.20002 mL	0.30017 mL	0.30000 mL	0.02500 mL	6.940	0.09819	0.98696	0.00488	20.0 s
22:23.3	Data point 35	1.20002 mL	0.30045 mL	0.30000 mL	0.02500 mL	6.784	0.09434	0.98150	0.00476	20.0 s
23:10.5	Data point 36	1.20002 mL	0.30089 mL	0.30000 mL	0.02500 mL	6.550	0.09759	0.98082	0.00493	18.5 s
24:01.3	Data point 37	1.20002 mL	0.30136 mL	0.30000 mL	0.02500 mL	6.331	0.09736	0.96676	0.00491	14.5 s
24:37.7	Data point 38	1.20002 mL	0.30172 mL	0.30000 mL	0.02500 mL	6.167	0.08595	0.94558	0.00442	13.0 s
25:27.9	Data point 39	1.20002 mL	0.30221 mL	0.30000 mL	0.02500 mL	5.891	0.08891	0.82762	0.00487	10.5 s
26:10.4	Data point 40	1.20002 mL	0.30249 mL	0.30000 mL	0.02500 mL	5.699	0.05380	0.95875	0.00271	10.0 s
26:47.1	Data point 41	1.20002 mL	0.30271 mL	0.30000 mL	0.02500 mL	5.491	0.04210	0.84134	0.00227	10.0 s
27:24.0	Data point 42	1.20002 mL	0.30287 mL	0.30000 mL	0.02500 mL	5.278	0.01152	0.56593	0.00077	10.0 s
27:55.8	Data point 43	1.20002 mL	0.30299 mL	0.30000 mL	0.02500 mL	5.075	-0.00400	0.18258	0.00046	10.0 s
28:22.4	Data point 44	1.20002 mL	0.30308 mL	0.30000 mL	0.02500 mL	4.850	-0.02268	0.83812	0.00123	10.0 s
28:49.0	Data point 45	1.20002 mL	0.30317 mL	0.30000 mL	0.02500 mL	4.657	-0.00060	0.00346	0.00051	10.0 s
29:15.6	Data point 46	1.20002 mL	0.30329 mL	0.30000 mL	0.02500 mL	4.437	0.01451	0.73420	0.00084	10.0 s
29:37.2	Data point 47	1.20002 mL	0.30343 mL	0.30000 mL	0.02500 mL	4.243	0.02774	0.95609	0.00140	10.0 s
30:09.1	Data point 48	1.20002 mL	0.30367 mL	0.30000 mL	0.02500 mL	4.043	0.03220	0.97101	0.00161	10.0 s
30:35.7	Data point 49	1.20002 mL	0.30398 mL	0.30000 mL	0.02500 mL	3.855	0.04033	0.98281	0.00201	10.0 s
31:07.6	Data point 50	1.20002 mL	0.30445 mL	0.30000 mL	0.02500 mL	3.666	0.03138	0.97445	0.00158	10.0 s
31:39.5	Data point 51	1.20002 mL	0.30515 mL	0.30000 mL	0.02500 mL	3.468	0.01204	0.90199	0.00063	10.0 s
32:06.1	Data point 52	1.20002 mL	0.30614 mL	0.30000 mL	0.02500 mL	3.278	0.00443	0.53709	0.00030	10.0 s
32:22.8	Data point 53	1.20002 mL	0.30757 mL	0.30000 mL	0.02500 mL	3.089	0.00496	0.51643	0.00034	10.0 s
32:39.6	Data point 54	1.20002 mL	0.30978 mL	0.30000 mL	0.02500 mL	2.904	0.00200	0.29422	0.00018	10.0 s
32:56.4	Data point 55	1.20002 mL	0.31319 mL	0.30000 mL	0.02500 mL	2.722	0.00018	0.00200	0.00020	10.0 s
33:13.1	Data point 56	1.20002 mL	0.31837 mL	0.30000 mL	0.02500 mL	2.539	-0.00101	0.05855	0.00021	10.0 s
33:29.9	Data point 57	1.20002 mL	0.32634 mL	0.30000 mL	0.02500 mL	2.356	-0.00273	0.12512	0.00038	10.0 s
33:46.8	Data point 58	1.20002 mL	0.33862 mL	0.30000 mL	0.02500 mL	2.172	-0.00317	0.26069	0.00031	10.0 s
34:03.9	Data point 59	1.20002 mL	0.35767 mL	0.30000 mL	0.02500 mL	1.990	-0.00943	0.81690	0.00052	10.0 s
36:59.6	Assay volumes	1.45002 mL	0.35767 mL	0.42512 mL	0.02500 mL					

Assay Settings

Setting	Value	Original Value	Date/Time changed	Imported from
General Settings				
Analyst name	Dorothy Levorse			
Separate reference vial	Yes			
Standard Experiment Settings				
Number of titrations	1			
Minimum pH	2.000			
pH step between points of	0.200			
Minimum titrant addition	0.00002 mL			
Maximum titrant addition	0.10000 mL			
Argon flow rate	100%			
Start titration using	Cautious pH adjust			
Advanced General Settings				
Detect turbidity using	Spectrometer			
Monitor at a wavelength of	500.0 nm			
Absorbance threshold of	0.100			
Collect turbidity sensor data	No			
Stir after titrant addition for	5 seconds			
For titrant addition, stir at	15%			
Titant Pre-Dose				

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 Instrument ID: **T311053**

Assay Settings (continued)

Setting	Value	Original Value	Date/Time changed	Imported from
Titrant pre-dose	Base Titrant			
Base titrant volume	0.30000 mL			
Allow to stand for	15 seconds			
Assay Medium				
Cosolvent in use	No			
ISA water volume	1.20 mL			
Water added	Automatic			
After water addition, stir for	5 seconds			
At a speed of	15%			
Buffer in use	Yes			
Buffer type	Phosphate Buffer			
Volume of buffer introduced	0.025000 mL			
Add buffer manually	Manual			
After medium addition, stir for	5 seconds			
Sample Sonication				
Sonicate	No			
Sample Dissolution				
Perform a dissolution stage	No			
Carbonate purge				
Perform a carbonate purge	No			
Temperature Control				
Wait for temperature	Yes			
Required start temperature	25.0°C			
Acceptable deviation	0.5°C			
Time to wait	60 seconds			
Stir speed of	15%			
Titration 1				
Titrate from	High to low pH			
Adjust to start pH	No			
Data Point Stability				
Stir during data point collection	Yes			
For point collection, stir at	15%			
Delay before data point collection	0 seconds			
Number of points to average	20 points			
Time interval between points	0.50 seconds			
Required maximum standard deviation	0.00500 dpH/dt			
Stability timeout after	60 seconds			
Experiment cleanup				
Adjust pH to cleanup	To start pH			
And then stir for	60 seconds			
For cleaning, stir at	20%			
Then add water volume	0.25 mL			
And then stir for	30 seconds			

Calibration Settings

Setting	Value	Date/Time changed	Imported from
Four-Plus alpha	0.094	9/18/2017 11:06:44 PM	C:\Sirius_T3\17I-18009_Blank standardisation.t3r
Four-Plus S	1.0023	9/18/2017 11:06:44 PM	C:\Sirius_T3\17I-18009_Blank standardisation.t3r
Four-Plus jH	0.8	9/18/2017 11:06:44 PM	C:\Sirius_T3\17I-18009_Blank standardisation.t3r
Four-Plus jOH	-0.5	9/18/2017 11:06:44 PM	C:\Sirius_T3\17I-18009_Blank standardisation.t3r
Base concentration factor	1.015	9/18/2017 11:06:44 PM	C:\Sirius_T3\KOH17I11.t3r
Acid concentration factor	1.006	9/18/2017 11:06:44 PM	C:\Sirius_T3\17I-18009_Blank standardisation.t3r

Instrument Settings

Setting	Value	Batch Id	Install date
Instrument owner	Merck		

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Instrument Settings (continued)

Setting	Value	Batch Id	Install date
Instrument ID	T311053		
Instrument type	T3 Simulator		
Software version	1.1.3.0		
Dispenser module		T3DM1100253	3/31/2009 6:24:52 AM
Dispenser 0	Water		3/31/2009 6:25:05 AM
Syringe volume	2.5 mL		
Firmware version	1.2.1(r2)		
Titrant	Water (0.15 M KCl)	8-18-17	9/18/2017 9:13:04 AM
Dispenser 2	Acid		3/31/2009 6:25:11 AM
Syringe volume	0.5 mL		
Firmware version	1.2.1(r2)		
Titrant	Acid (0.5 M HCl)	166940	9/8/2017 9:21:27 AM
Dispenser 1	Base		3/31/2009 6:25:21 AM
Syringe volume	0.5 mL		
Firmware version	1.2.1(r2)		
Titrant	Base (0.5 M KOH)	01/06/17	9/8/2017 9:20:03 AM
Dispenser 5	Cosolvent		3/31/2009 6:26:24 AM
Syringe volume	2.5 mL		
Firmware version	1.2.1(r2)		
Distribution valve 5	Distribution Valve		3/31/2009 6:28:19 AM
Firmware version	1.1.3		
Port A	Methanol (80%, 0.15 M KCl)	8-15-17	9/13/2017 12:23:11 PM
Dispenser 3	Buffer		8/3/2010 6:05:16 AM
Syringe volume	0.5 mL		
Firmware version	1.2.1(r2)		
Titrant	Phosphate Buffer		9/12/2017 12:32:29 PM
Dispenser 6	Octanol		10/22/2010 11:52:43 AM
Syringe volume	0.5 mL		
Firmware version	1.2.1(r2)		
Titrant	Octanol	9-14-17	9/14/2017 10:30:38 AM
Titrator		T3TM1100153	3/31/2009 6:24:17 AM
Horizontal axis firmware version	1.17 AI1DI2DO2 Stepper 2		
Vertical axis firmware version	1.17 AI1DI2DO2 Stepper 2		
Chassis I/O firmware version	1.11 AI1DI0DO4 Norgren I/O		
Probe I/O firmware version	1.1.1		
Electrode	T3 Electrode	T3E0769	8/15/2017 10:21:54 AM
E0 calibration	-7.21 mV		9/18/2017 11:07:08 PM
Filling solution	3M KCl	KCL095	9/18/2017 9:17:15 AM
Liquids			
Wash 1	50% IPA:50% Water		9/18/2017 9:09:36 AM
Wash 2	0.5% Triton X-100 in H2O		9/18/2017 9:09:39 AM
Buffer position 1	pH7 Wash		9/18/2017 9:09:41 AM
Buffer position 2	pH 7		9/18/2017 9:09:44 AM
Storage position			9/18/2017 9:10:43 AM
Wash water	8.4e+003 mL	9-18-17	9/18/2017 8:54:32 AM
Waste	1.7e+003 mL		9/18/2017 8:54:39 AM
Temperature controller			8/5/2010 7:35:13 AM
Turbidity detector			3/31/2009 6:24:45 AM
Spectrometer		072390	11/23/2010 12:22:28 PM
Dip probe		11086	
Wavelength coefficient A0	185.563		
Wavelength coefficient A1	2.17439		
Wavelength coefficient A2	-0.000285622		
Total lamp lit time	143:53:30		11/23/2010 12:22:28 PM
Calibrated on	9/18/2017 9:35:14 AM		
Integration time	11		
Scans averaged	10		
Autoloader		T3AL1100237	11/10/2015 10:34:13 AM



Assay Settings

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Instrument Settings (continued)

Setting	Value	Batch Id	Install date
Left-right axis firmware version	1.17 AI1DI2DO2 Stepper 2		
Front-back axis firmware version	1.17 AI1DI2DO2 Stepper 2		
Vertical axis firmware version	1.17 AI1DI2DO2 Stepper 2		
Chassis I/O firmware version	1.11 AI1DI0DO4 Norgren I/O		
Configuration			
Alternate titration position	Titration position		
Alternate reference position	Reference position		
Maximum standard vial volume	3.50 mL		
Maximum alternate vial volume	25.00 mL		
Automatic action idle period	5 minute(s)		
Titrant tube volume	1.3 mL		
Syringe flush count	3.50		
Flowing wash pump volume	20.0 mL		
Flowing wash stir duration	5 s		
Flowing wash stir speed	30%		
Solvent wash stir duration	5 s		
Solvent wash stir speed	30%		
Surfactant wash stir duration	5 s		
Surfactant wash stir speed	30%		
E0 calibration minimum number of points	10		
E0 calibration maximum standard deviation	0.01500		
E0 calibration timeout period	60 s		
E0 calibration stir duration	5 s		
E0 calibration preparation stir speed	30%		
E0 calibration buffer wash stir duration	5 s		
E0 calibration buffer wash stir speed	30%		
E0 calibration reading stir speed	0%		
Spectrometer calibration stir duration	5 s		
Spectrometer calibration stir speed	30%		
Spectrometer calibration wash pump volume	20.0 mL		
Spectrometer calibration wash stir duration	5 s		
Spectrometer calibration wash stir speed	30%		
Overhead dispense height	10000		

Refinement Settings

Setting	Value	Default value
Turbidity detection method	Spectrometer	Spectrometer
Turbidity wavelength to assess	500.0 nm	500.0 nm
Turbidity maximum absorbance	0.100	0.100
Turbidity probe threshold	50.00	50.00
Exclude turbid points	Yes	Yes
Low intensity warning threshold	100	100
Minimum absorbance change threshold	0.100	0.100
Eigenvector autocorrelation threshold	0.80	0.80
Maximum RMSD severe warning	0.250	0.250
Maximum RMSD warning	0.050	0.050