Evaluation of MULTIGENT® Lithium assay on the ABBOTT ARCHITECT® *c*8000 and AEROSET® Clinical Chemistry Systems

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Abstract

Objective: The analytical performance of the MULTIGENT Lithium assay was evaluated on the ABBOTT ARCHITECT c8000 and AEROSET clinical chemistry systems. The goal of the study was to verify whether the analytical performance met the HCFA/CLIA allowed Total Error (<20% or 0.3mmol/L). Acceptance criteria were set to < 5% or 0.075 mmol/L for both Bias (Accuracy) and Total Imprecision to insure that the combined effects of bias and imprecision would meet the HCFA/CLIA requirements.

Materials/instruments: MULTIGENT Lithium, a colorimetric assay manufactured by SENTINEL CH., is based on the use of a chromogenic chelator able to selectively complex lithium ions at alkaline pH. Abbott's ARCHITECT c8000 and AEROSET Systems are random-access clinical chemistry analyzers. ARCHITECT c8000 can be integrated with ARCHITECT $i2000_{SR}$, an immunoassay module, to form the *c*i8200.

Design study: Assay performance was characterized using CLSI derived protocols. Precision was determined at three analyte levels. LOQ was defined as the analyte concentration at which the %CV of the assay is ≤ 20%. Interferences from endogenous substances and from metals were investigated following the Glick protocol. The analytical measuring range (AMR) was defined through linearity. Method comparison consisted of testing patient serum samples on AEROSET or c8000 and the atomic absorption spectrophotometric (AAS) reference method and Hitachi 911.

Results:

Performances	ARCHITECT c8000			AEROSET
Measuring units	mmol/L			mmol/L
AMR	0.10 to 3.51			0.10 to 3.51
Total Imprecision (20 day)	3.23% at 0.883 2.73% at 1.519 1.78% at 2.733			3.98% at 0.839 2.67% at 1.436 1.68% at 2.579
LOQ	< 0.1mmol/l			< 0.1 mmol/L
Method Comparison	*vs.* Hitachi 911	*vs.* AEROSET	*vs.* AAS	*vs.* Hitachi 911
n	60	60	62	60
Range	0.29–2.61	0.29–2.59	0.11–3.10	0.29–2.61
Slope	0.993	0.973	1.062	1.019
Intercept	-0.006	0.024	-0.044	-0.029
n	0.999	0.999	0.996	0.999

Bilirubin (40 mg/dL), Hemoglobin (1000 mg/dL), Intralipid (1500 mg/dL), Calcium (24 mg/dL), Magnesium (6.07 mg/dL), Potassium (13mEq/L), Sodium (350 mEq/L) did not interfere.

Conclusion: The data meet the HCFA/CLIA's imprecision and accuracy requirements on AEROSET and c8000 systems. Interference findings demonstrate the robustness of the assay and its suitability in routine lab use. The MULTIGENT Lithium assay allows consolidation of the therapeutic drug assays with general chemistry on a single high volume chemistry analyzer, which is highly valuable for optimizing workflow and efficiency in today's laboratories.

Introduction

The use of lithium salts as a treatment of bipolar disorder was first discovered by Dr. John Cade, an Australian psychiatrist who published a paper on the use of lithium in 1949. Lithium salts had long been used as a first-line treatment for bipolar disorder. In ancient times, doctors would send their mentally ill patients to drink from "alkali springs" as a treatment. Lithium was present in high concentration in the waters. The therapeutic effect of lithium salts appears to be entirely due to the lithium ion, Li^+. The two lithium salts used for bipolar therapy are lithium carbonate (mostly) and lithium citrate (sometimes). Approved for the treatment of acute mania in 1970 by the Food and Drug Administration (FDA), lithium has been an effective mood-stabilizing medication for many people with bipolar disorder. Lithium is also noted for reducing the risk of suicide.[1] Although lithium is among the most effective mood stabilizers, persons taking it may experience side effects similar to the effects of ingesting too much table salt, such as high blood pressure, water retention, and constipation. Lithium side effects may also include hand trembling and intolerance of hot weather. Cogentin is sometimes used to control trembling, but may cause sedation. The mechanism of lithium salt treatment is believed to work as follows: Some symptoms of bipolar disorder appear to be caused by the action of the enzyme inositol monophosphatase (IMPase), an enzyme that splits inositol monophosphate into free inositol and phosphate. IMPase is involved in signal transduction and is believed to create an imbalance in neurotransmitters in bipolar patients. The lithium ion is believed to produce a mood stabilizing effect by substituting for one of two magnesium ions in the IMPase's active site, inhibiting the enzyme. Lithium has a very narrow therapeutic window. Below that level it has no effect, and above it is toxic. For that reason blood must be sampled frequently to determine if the proper blood level has been achieved.

Instruments and materials

AEROSET, ARCHITECT c8000 and c16000 clinical chemistry analyzers. ARCHITECT cSystems can be integrated with ARCHITECT $i2000_{SR}$, an immunoassay module, to form the *c*i8200 or *c*i16200.

Multigent Lithium assay, manufactured by SENTINEL CH and distributed by Abbott Laboratories, is a colorimetric assay based on the use of a chromogenic chelator able to selectively complex lithium ions at alkaline pH.

Methodology/Acceptance criteria

The acceptance criteria were based on CLIA criteria for acceptable performance in proficiency testing events in the form of ***total allowable error, TE_a***.
CLIA Acceptable Performance: the greatest of 0.30 mmol/L or 20%
Evaluation study acceptance criteria were set to meet the target TE_a <= 20.0% based on formula TE_a = Accuracy + 1.65 x Imprecision.
Imprecision specifications: <= the greatest of 5% or SD 0.075 mmol/L
Accuracy specifications: <= the greatest of 5% or SD 0.075 mmol/L
Allowed total analytical error: <= the greatest of 13 % or SD 0.20 mmol/L

Functional Sensitivity (Limit of quantitation): Sensitivity was evaluated according NCCLS/ CLSI document EP17-A. A sample containing a low lithium concentration was serially diluted with physiological saline and with analyte-free human serum. Five replicates of each dilution level were tested on AEROSET, and 10 replicates were tested on the ARCHITECT c8000. Additional testing was performed on the ARCHITECT c8000 using Analyte-free human serum as the diluent. The limit of quantitation is the lowest concentration of analyte which has imprecision ≤ 20% CV, and % Recovery within 5% or ± 0.075 mmol/L of the theoretic concentration.

Total imprecision: Two commercially available human sera controls containing lithium and a human serum pool spiked with lithium lactate to approximately 1.5 mmol/L were tested on an AEROSET and an ARCHITECT c8000 in replicates of two, twice per day (separated by a minimum of two hours), for twenty days. Precision was reported as the total %CV. These studies were in accordance with CLSI protocol EP5-A. Precision was considered acceptable if the total %CV was ≤ 5% CV (conc. ≤ 1.00 mmol/L) or ≤ 4%CV (conc. > 1.00 mmol/L).

Interferences: Endogenous substance interferences were evaluated according the Glick model. A pool of human sera was spiked with the potentially interfering substances and then serially diluted with unspiked human serum pool. Metals interferences were evaluated according to the Glick model. A pool of human sera was spiked with concentrated solution of the metals under investigation and then serially diluted with unspiked human serum pool. Acceptance criterion were the greater of +/- 0.075 mmol/L or +/- 5%.

Linearity: Linearity and analytical measuring range (AMR) were evaluated on at least ten test dilutions at Lithium concentrations spanning the desired linearity range for each assay. Data were analyzed according to NCCLS (CLSI) document EP6-P2. Acceptance criteria were the greater of +/- 5% or +/- 0.075 mmol/L bias.

Methods comparison: Method comparison was performed using a protocol based on NCCLS (CLSI) document EP9-A2. This testing was performed on the Abbott ARCHITECT c8000 and AEROSET Systems in comparison to a colorimetric assay on Hitachi 911 and to atomic absorption spectrophotometry (AAS). The testing was performed on the Abbott ARCHITECT c16000 in comparison to the AEROSET.

Conclusions

1. The precision and accuracy data on the AEROSET and c8000 systems meet the CLIA requirements. Interference findings demonstrate the robustness of the assay and its suitability in routine lab use.

2. The MULTIGENT Lithium assay allows consolidation of the therapeutic drug assays with general chemistry on a single high volume chemistry analyzer, which is highly valuable for optimizing workflow and efficiency in today's laboratories.

References

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Results: Sensitivity

Limit of Quantitation (Imprecision *vs* Concentration) - Human serum diluted with saline

AEROSET		Limit of Quantitation (according to CLSI- EP17-A) (Bias <= 0.075 mmol/L and CV% <= 20%)					
	% Factor	0	6.25	12.5	25	50	100
Found Values	Rep 1	0.001	0.092	0.164	0.323	0.589	1.293
	Rep 2	0.008	0.082	0.173	0.316	0.592	1.263
	Rep 3	0.016	0.098	0.179	0.350	0.580	1.261
	Rep 4	0.016	0.085	0.163	0.317	0.620	1.297
	Rep 5	-0.003	0.078	0.174	0.340	0.615	1.266
	Mean	0.008	0.087	0.171	0.329	0.599	1.276
	Dev. Std.	0.009	0.008	0.007	0.015	0.017	0.017
	CV%	113%	9%	4%	5%	3%	1%
	Calculated mmol/L	0.006	0.085	0.164	0.321	0.636	1.266
	Absolute Bias mmol/L	0.002	0.002	0.007	0.008	-0.037	0.010
	Relative Bias %	27%	3%	4%	3%	-6%	1%

ARCHITECT c8000		Limit of Quantitation (according to CLSI- EP17-A) (Bias <= 0.075 mmol/L and CV% <= 20%)						
	% Factor	10	20	30	40	60	80	100
Found Values	Rep 1	0.06	0.12	0.18	0.22	0.33	0.43	0.53
	Rep 2	0.06	0.13	0.17	0.22	0.33	0.41	0.55
	Rep 3	0.08	0.11	0.18	0.24	0.34	0.43	0.53
	Rep 4	0.07	0.12	0.16	0.21	0.32	0.44	0.54
	Rep 5	0.06	0.13	0.16	0.23	0.32	0.44	0.54
	Rep 6	0.05	0.13	0.15	0.23	0.33	0.45	0.54
	Rep 7	0.08	0.14	0.17	0.22	0.33	0.43	0.52
	Rep 8	0.07	0.10	0.17	0.21	0.33	0.43	0.51
	Rep 9	0.07	0.11	0.17	0.23	0.33	0.43	0.52
	Rep 10	0.05	0.12	0.17	0.23	0.32	0.44	0.53
	Mean	0.065	0.121	0.168	0.224	0.328	0.433	0.531
	Dev. Std.	0.011	0.012	0.009	0.010	0.006	0.011	0.012
	CV%	17	10	5	4	2	2	2
	Calculated mmol/L	0.067	0.119	0.171	0.223	0.326	0.430	0.534
	Absolute Bias mmol/L	-0.002	0.002	-0.003	0.001	0.002	0.003	-0.003
	Relative Bias %	-3%	2%	-2%	1%	0%	1%	-1%

Limit of Quantitation (Imprecision *vs* Concentration)- Human serum diluted with analyte-free human serum.

ARCHITECT c8000		Limit of Quantitation (according to CLSI- EP17-A) (Bias <= 0.075 mmol/L and CV% <= 20%)						
	% Factor	10	20	30	40	60	80	100
Found Values	Rep 1	0.07	0.10	0.16	0.21	0.33	0.43	0.55
	Rep 2	0.05	0.10	0.18	0.22	0.31	0.42	0.53
	Rep 3	0.06	0.10	0.18	0.23	0.32	0.44	0.59
	Rep 4	0.06	0.11	0.16	0.22	0.33	0.42	0.54
	Rep 5	0.05	0.11	0.16	0.21	0.31	0.41	0.55
	Rep 6	0.05	0.11	0.17	0.21	0.33	0.43	0.52
	Rep 7	0.05	0.12	0.18	0.22	0.33	0.41	0.57
	Rep 8	0.05	0.11	0.18	0.22	0.30	0.42	0.51
	Rep 9	0.05	0.11	0.17	0.21	0.33	0.46	0.53
	Rep 10	0.05	0.12	0.17	0.18	0.30	0.41	0.53
	Mean	0.054	0.109	0.171	0.213	0.319	0.425	0.542
	STD. Dev.	0.007	0.007	0.009	0.013	0.013	0.016	0.024
	CV%	12.9%	6.8%	5.1%	6.3%	4.0%	3.7%	4.4%
	Calculated mmol/L	0.056	0.109	0.163	0.216	0.323	0.430	0.537
	Absolute Bias mmol/L	-0.002	0.000	0.008	-0.003	-0.004	-0.005	0.005
	relative Bias %	-3%	-0%	5%	-1%	-1%	-1%	1%

Results: Total Imprecision

AEROSET 20-days Total Imprecision

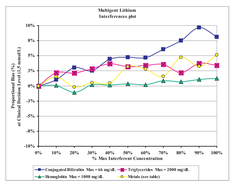
Imprecision (n = 80)	Level 1		Level 2		Level 3	
	SD	% CV	SD	% CV	SD	% CV
Mean (mmol/L)	0.839 mmol/L		2.579 mmol/L		1.436 mmol/L	
Within Run	0.022	2.61	0.034	1.31	0.024	1.69
Between Days	0.020	2.38	0.002	0.08	0.030	2.09
Total	0.033	3.98	0.043	1.68	0.038	2.67

ARCHITECT c8000 20-days Total Imprecision

Imprecision (n = 80)	Level 1		Level 2		Level 3	
	SD	% CV	SD	% CV	SD	% CV
Mean (mmol/L)	0.883 mmol/L		2.733 mmol/L		1.519 mmol/L	
Within Run	0.021	2.41	0.031	1.13	0.023	1.50
Between Days	0.017	1.96	0.037	1.37	0.033	2.19
Total	0.029	3.23	0.049	1.78	0.042	2.73

Results: Interferences

(Accepted bias +/- 0.075 mmol/L or +/- 5% related at clinical decision level 1.5 mmol/L)

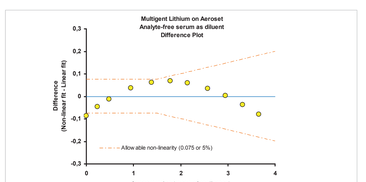


	Metals											
High ions solution	Zn	Cu	Fe	Na	K	Mg	Ca	Lithium mmol/L				
% spiking	umol/L	umol/L	umol/L	mmol/L	mmol/l	mmol/L	mmol/l	1	2	3	mmol/l	mmol/l
0%	8.5	14.4	21.2	142	4.8	0.83	2.87	1.129	1.144	1.139	1.139	0.000
10%	33.5	39.4	41.2	167	5.8	1.03	3..3	1.162	1.187	1.160	1.162	0.023
20%	58.5	64.4	61.2	192	6.8	1.23	3.7	1.136	1.128	1.169	1.136	-0.003
30%	83.5	89.4	81.2	217	7.8	1.43	4.1	1.150	1.147	1.145	1.147	0.008
40%	108.5	114.4	101.2	242	8.8	1.63	4.5	1.146	1.142	1.164	1.146	0.007
50%	133.5	139.4	121.2	267	9.8	1.83	4.9	1.186	1.193	1.187	1.187	0.048
60%	158.5	164.4	141.2	292	10.8	2.03	5.3	1.160	1.181	1.205	1.181	0.042
70%	183.5	189.4	161.2	317	11.8	2.23	5.7	1.156	1.163	1.198	1.163	0.024
80%	208.5	214.4	181.2	342	12.8	2.43	6.1	1.187	1.216	1.210	1.210	0.071
90%	233.5	239.4	201.2	367	13.8	2.63	6.5	1.207	1.158	1.188	1.188	0.049
100%	258.5	264.4	221.2	392	14.8	2.83	6.9	1.216	1.210	1.248	1.216	0.077

Results: Linearity

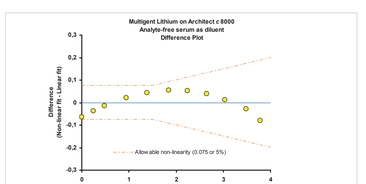
Linearity on Aeroset - Analyte-free serum as diluent
Elaboration according CLSI EP6-A – Analyse-it vs2.04)

Spiked Serum %	Un-spiked Serum %	Found Mean mmol/L	Expected Calculated concentrations mmol/L	Absolute Bias mmol/L	Relative Bias %Bias
0	100	0.002	0.074	-0.072	-97.8%
5	95	0.238	0.281	-0.042	-15.1%
10	90	0.487	0.487	0.000	0.0%
20	80	0.948	0.900	0.048	5.3%
30	70	1.383	1.313	0.070	5.3%
40	60	1.779	1.726	0.053	3.1%
50	50	2.146	2.139	0.007	0.3%
60	40	2.575	2.552	0.023	0.9%
70	30	2.949	2.965	-0.016	-0.5%
80	20	3.309	3.378	-0.069	-2.1%
90	10	3.660	3.791	-0.131	-3.5%
100	0	4.036	4.204	-0.168	-4.0%



Linearity on Architect c8000 - Analyte-free serum as diluent
Elaboration according CLSI EP6-A – Analyse-it vs2.04)

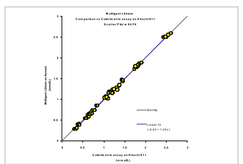
Spiked Serum %	Un-spiked Serum %	Found Mean mmol/L	Expected Calculated concentrations mmol/L	Absolute Bias mmol/L	Relative Bias %Bias
0	100	0.003	0.052	-0.049	-93.6%
5	95	0.240	0.267	-0.027	-10.3%
10	90	0.487	0.483	0.004	0.8%
20	80	0.950	0.913	0.037	4.1%
30	70	1.380	1.343	0.037	2.8%
40	60	1.820	1.773	0.047	2.6%
50	50	2.233	2.203	0.030	1.4%
60	40	2.593	2.634	-0.040	-1.5%
70	30	3.030	3.064	-0.034	-1.1%
80	20	3.490	3.494	-0.004	-0.1%
90	10	3.767	3.924	-0.158	-4.0%
100	0	4.120	4.355	-0.235	-5.4%



Results: Methods comparison

X	Hitachi 911	Hitachi 911	Multigent Lithium (Aeroset)	Multigent Lithium (Aeroset)	AAS	AAS
Y	Multigent Lithium (Aeroset)	Multigent Lithium (c8000)	Multigent Lithium (c8000)	Multigent Lithium (c16000)	Multigent Lithium (Aeroset)	Multigent Lithium (c8000)
N	60	60	60	60	62	62
Range (mg/L)	0.29 – 2.61	0.29 – 2.61	0.29 – 2.59	0.20 – 3.50	0.11 – 3.10	0.11 – 3.10
Slope (95% CI)	1.019 (1.006 to 1.032)	0.993 (0.980 to 1.005)	0.973 (0.963 to 0.984)	0.970 (0.956 to 0.984)	1.071 (1.042 to 1.099)	1.062 (1.039 to 1.085)
	-0.029 (-0.047 to -0.012)	-0.006 (-0.023 to 0.011)	0.024 (0.009 to 0.039)	-0.023 (-0.043 to 0.003)	-0.021 (-0.045 to 0.004)	-0.044 (-0.064 to -0.024)
R	0.999	0.999	0.999	0.999	0.995	0.996
Sy/x	0.031	0.031	0.027	0.036	0.051	0.042
PLOT	1	2	3	4	5	6

PLOT 1



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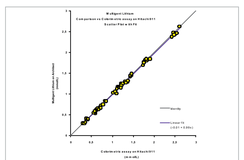

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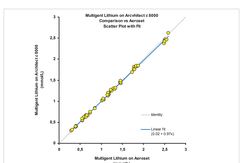

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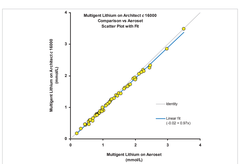

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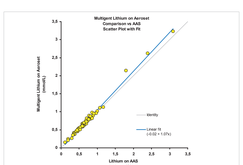
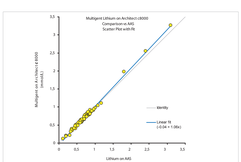
PLOT 6