Linearity Study Report of the

L-Pet Veterinary Lactate System

Approved By: _____   Date: 23rd August 2019

Linearity Study Report of the L-Pet Veterinary Lactate Screening System

Scope and Objective

Woodley Equipment Company Ltd intends to determine the linear limits of the L-Pet Veterinary Lactate Screening System Meter to be 5-225 mg/dL. This protocol used NCCLS EP-6A Evaluation of the Linearity of Quantitative Measurement Procedures as guidelines. The protocol allowed Woodley Equipment Company Ltd to demonstrate that the L-Pet Veterinary Lactate Screening System Meter meets these specifications.

The accuracy of the L-Pet Veterinary Lactate Screening System Meter was verified prior to the start of each test set. The Beckman Coulter AU480 Clinical Chemistry System Analyser was calibrated prior to each test using the Coulter Calibrator DR0070-2 Level 2 Set.

Testing was performed using venous whole blood and adjusted blood lactate value to provide samples using the L-Pet Meter at each blood lactate range. After the lactate test using the L-Pet Meter, a blood lactate test using the same blood sample was performed on the Beckman Coulter AU480 Clinical Chemistry System Analyser as the reference in this study.

Three lots of the L-Pet Test Strips were used in the linearity study. Five samples at different blood lactate concentrations, ranging from 10-215 mg/dL, were tested on three different lots of test strips. The mean (mg/dL), standard deviation (SD, mg/dL) and coefficient of variation (CV, %) were calculated at the blood lactate range of three lots of test strips. System accuracy was evaluated by the mean coefficient of variation (mCV). Linear regression was used, the slope, y-intercept and r value were calculated. The results were also presented as the percentage of values falling within the ±20% intervals. The L-Pet Meter without the haemoglobin cut-off limits set had been used to detect blood lactate concentration <5 mg/dL and >225 mg/dL in the linearity study.

Materials and Equipment

- L-Pet Meter
- L-Pet Test Strips (3 Lots)
- Beckman Coulter AU480 Clinical Chemistry System Analyser
- Coulter DR0070-2 Level 2 Calibrator Set

Test Protocol

1. Sample Preparation
   a. Prepare venous whole blood samples and use a centrifuge to separate the plasma and red blood cells.
   b. Remove the plasma or red blood cells to adjust the haemoglobin concentration to 43% (14.3 mg/dL).

c. Add lithium lactate into the blood to adjust the blood lactate concentration:
   - C1: 10-45 mg/dL
   - C2: 50-85 mg/dL
   - C3: 90-125 mg/dL
   - C4: 130-165 mg/dL
   - C5: 170-215 mg/dL
d. Using a calibrated Beckman Coulter AU480 Analyser, determine the blood lactate level of all samples.
e. Label the blood samples C1 through C5.

2. Testing
   a. Obtain the required supplies and equipment.
   b. Record the serial number of the L-Pet Meter, Beckman Coulter AU480 Analyser and the Lot No. of the test strip, as shown below:
      - L-Pet Meter 1: A004393
      - L-Pet Meter 2: A004398
      - L-Pet Meter 3: A004395
      - L-Pet Meter 4: A004390
      - L-Pet Meter 5: A004392
   c. The test conditions will be as follows:
      - Temperature: 18-30°C
      - Humidity: Less than 80%

      Check conditions prior to testing. If conditions are within specification, continue testing. If test conditions are not within specification, stop testing and repeat all tests since last approved condition check.
   d. Prior to the start of each test set, verify that the meters are operating properly using the appropriate method per the appropriate operator's manual.
   e. Select one of the three lots of L-Pet Test Strips.
   f. Assign one vial of test strips to each meter.
   g. Remove one strip from the assigned vial and insert it into the meter. The meter will turn on automatically.
   h. Select one of the 5 blood samples. Apply a drop of blood from the sample to the test area of the L-Pet Test Strip.
   i. In approximately 45 seconds, the blood lactate value will be displayed. Record the results. Remove the test strip and discard.
   j. Using the same sample and the same meter, repeat sections 2g-2i.
   k. Using the same sample, repeat sections 2f-2j with each of the four remaining meters and vials. This results in a total of ten measurements per sample.
   l. Using the same sample, repeat sections 2e-2k with the two remaining lots of L-Pet Test Strips.
   m. Take the next sample and repeat section 2e-2i.

n. Calculate the mean value, the standard deviation and the coefficient of variation from the twenty measurements of each sample.

Acceptance Criteria

The mCV (≤5%, 80%) of the individual results shall fall within ±20% of the Beckman Coulter AU480 reference.

The slope should be within 1±0.1 and the intercept should be within 0±1.

Test Results

Test results are presented in Exhibit 1 of this report. Results are summarised in the following:

L-Pet Test Strip Lot No.: LS006K

	C1	C2	C3	C4	C5
Coulter (mg/dL)	33.3	75.2	118	161.8	204
Mean (mg/dL)	34.1	73.7	116.4	160.7	205.7
SD (mg/dL)	1.2	1.3	3.1	4.5	3.3
mCV (%)	2%				
Within Coulter ±20%	10/10	10/10	10/10	10/10	10/10
Overall Within Coulter ±20%	50/50				

L-Pet Test Strip Lot No.: LS006L

	C1	C2	C3	C4	C5
Coulter (mg/dL)	16.7	75.2	118	161.8	198.9
Mean (mg/dL)	16.5	74.1	114.7	160.9	193.8
SD (mg/dL)	0.7	1.9	1.5	2.7	1.7
mCV (%)	2%				
Within Coulter ±20%	10/10	10/10	10/10	10/10	10/10
Overall Within Coulter ±20%	50/50				

L-Pet Test Strip Lot No.: LS006N

	C1	C2	C3	C4	C5
Coulter (mg/dL)	14.7	64.7	106.2	150.4	214.2
Mean (mg/dL)	13.7	64.2	107.1	149.7	208.4
SD (mg/dL)	1.3	1.3	3.7	6.8	9.1
mCV (%)	5%				
Within Coulter ±20%	10/10	10/10	10/10	10/10	10/10
Overall Within Coulter ±20%	50/50				

All the test results meet the requirements of acceptance criteria for system accuracy.

The results of the Bias Analyser indicated that, with the L-Pet System, all of the test results are within ±20% of the Coulter reference.

The resulting scatter grams and associated regression parameters for the haemoglobin values of the L-Pet Meter versus the Coulter AU480 Analyser are shown in Exhibit 2 of this report.

The L-Pet Meters correlated well with the Coulter AU480 Analyser. Following are the results regression parameters:

L-Pet Meter vs Coulter AU480

Lot No.	Slope	y-intercept	R^2 Value
LS006K	1.0052	-0.9599	0.9974
LS006L	0.9792	0.2525	0.9989
LS006N	0.9781	0.9905	0.9936

Conclusion

The results of this linearity study validate the published blood lactate measurement range of 5-225 mg/dL for the L-Pet Meter.

Exhibit 1

L-Pet Test Strip Lot No.: LS006K

Test Date: August 14th, 2018

Measurement Unit: mg/dL   Temperature: 25°C   Humidity: 46%

Coulter	Meter 1	Meter 2	Meter 3	Meter 4	Meter 5
33.3	36	34	35	33	35
	34	33	32	34	35
	Mean	34.1		Ref. +20%	39.9
	SD	1.2		Ref. -20%	26.6
	Within Ref.	10/10 (100%)			
75.2	76	75	73	75	73
	72	74	74	72	73
	Mean	73.7		Ref. +20%	90.24
	SD	1.3		Ref. -20%	60.16
	Within Ref.	10/10 (100%)			
118	121	116	119	119	110
	118	114	115	117	115
	Mean	116.4		Ref. +20%	141.6
	SD	3.3		Ref. -20%	94.4
	Within Ref.	10/10 (100%)			
161.8	166	158	164	163	157
	162	165	161	151	160
	Mean	160.7		Ref. +20%	194.2
	SD	4.5		Ref. -20%	129.44
	Within Ref.	10/10 (100%)			
204	206	207	205	207	211
	209	206	203	204	199
	Mean	205.7		Ref. +20%	163.2
	SD	3.3		Ref. -20%	129.44
	Within Ref.	10/10 (100%)			

L-Pet Test Strip Lot No.: LS006L

Test Date: August 14th, 2015

Measurement Unit: mg/dL   Temperature: 25°C   Humidity: 46%

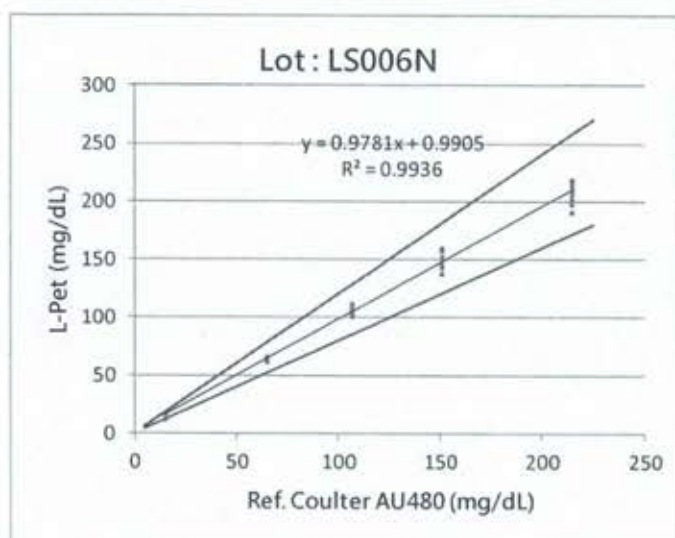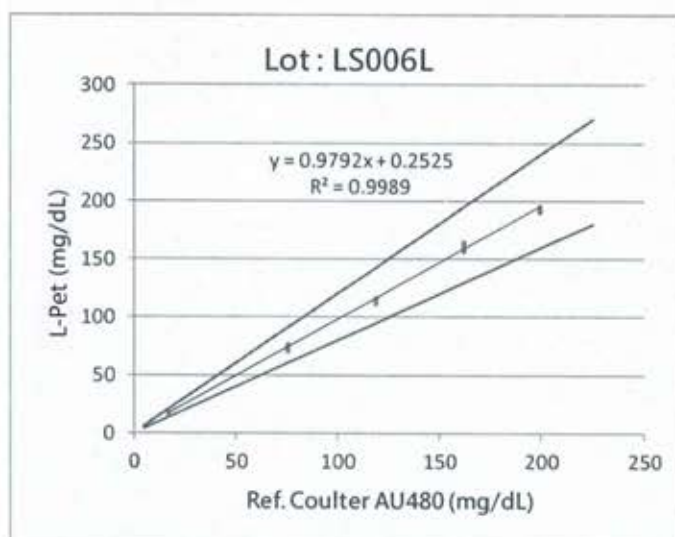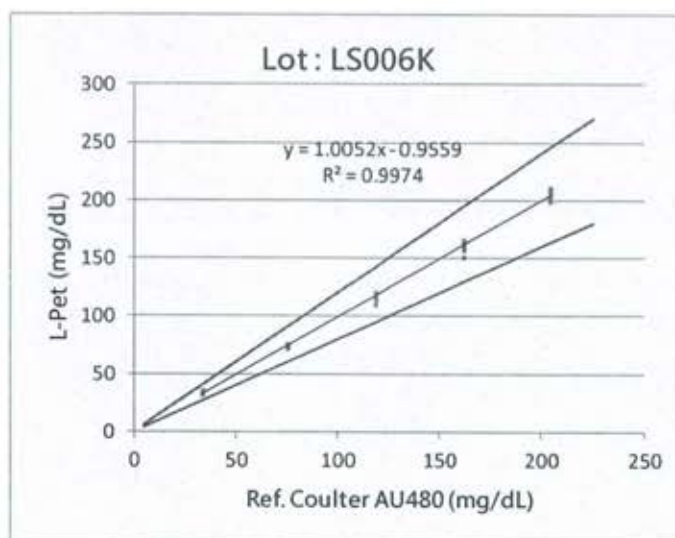| Coulter	Meter 1	Meter 2	Meter 3	Meter 4	Meter 5
16.7	16	16	16	17	16
	18	17	16	16	17
	Mean	16.5		Ref. +20%	20.0
	SD	0.7		Ref. -20%	13.3
	Within Ref.	10/10 (100%)			
75.2	74	76	73	71	74
	72	75	77	76	73
	Mean	74.1		Ref. +20%	90.2
	SD	1.9		Ref. -20%	60.2
	Within Ref.	10/10 (100%)			
118	117	114	115	114	116
	112	116	113	115	115
	Mean	114.7		Ref. +20%	141.6
	SD	1.5		Ref. -20%	94.4
	Within Ref.	10/10 (100%)			
161.8	163	165	162	159	160
	157	159	164	162	158
	Mean	160.9		Ref. +20%	194.2
	SD	2.7		Ref. -20%	129.4
	Within Ref.	10/10 (100%)			
198.9	195	192	195	191	194
	192	193	196	195	195
	Mean	193.8		Ref. +20%	238.7
	SD	1.7		Ref. -20%	159.7
	Within Ref.	10/10 (100%)			

L-Pet Test Strip Lot No.: LS006N

Test Date: August 14th, 2018

Measurement Unit: mg/dL   Temperature: 25°C   Humidity: 46%

Coulter	Meter 1	Meter 2	Meter 3	Meter 4	Meter 5
14.7	12	15	13	14	14
	13	12	16	15	13
	Mean	13.7		Ref. +20%	17.6
	SD	1.3		Ref. -20%	11.8
	Within Ref.	10/10 (100%)			
64.8	65	62	64	64	66
	64	66	63	63	65
	Mean	64.2		Ref. +20%	77.8
	SD	1.3		Ref. -20%	51.8
	Within Ref.	10/10 (100%)			
106.2	110	108	101	112	109
	111	105	103	104	112
	Mean	107.1		Ref. +20%	127.4
	SD	3.7		Ref. -20%	85.0
	Within Ref.	10/10 (100%)			
150.4	153	146	151	160	158
	145	151	143	152	138
	Mean	149.7		Ref. +20%	180.5
	SD	6.8		Ref. -20%	120.3
	Within Ref.	10/10 (100%)			
150.4	210	206	191	215	219
	198	214	211	202	218
	Mean	208.4		Ref. +20%	257.1
	SD	9.1		Ref. -20%	171.4
	Within Ref.	10/10 (100%)			

Exhibit 2

Lot : LS006K

$y = 1.0052x - 0.9559$
$R^2 = 0.9974$

L-Pet (mg/dL) vs Ref. Coulter AU480 (mg/dL)

Lot : LS006L

$y = 0.9792x + 0.2525$
$R^2 = 0.9989$

L-Pet (mg/dL) vs Ref. Coulter AU480 (mg/dL)

Lot : LS006N

$y = 0.9781x + 0.9905$
$R^2 = 0.9936$

L-Pet (mg/dL) vs Ref. Coulter AU480 (mg/dL)