

Conversion of Reference Intervals From Hitachi 911 to Cobas 501c

March 2008

The new reference intervals for the serum chemistry analytes or calculated values were determined by the following process.

For dogs and horses:

Thanks to the efforts of clinicians, we have attempted to establish new reference intervals for the Hitachi 911 assays. For horses, most samples were collected through the efforts of Dr. Shane DeWitt. For dogs, most samples were collected through the efforts of clinicians in the Pet Health Center (Drs. Nelson, Dixon-Jimenez, Akers, and Riegel). Based on historical and physical findings, the animals were considered to be clinically healthy. The goal for the reference population was include at least 60 adult, healthy animals of each species.

None of the acceptable samples had either evidence of hemolysis or lipemia of sufficient degree to interfere with analysis. After samples were analyzed, the data were evaluated for obviously abnormal values and a few animals were excluded because data indicated that a subclinical disease was present. Then the preliminary reference values were tabulated and analyzed with Analyse-it[®] for outlier values using non-parametric analysis. If outlier values were found (usually recognized by being greater than three interquartile-ranges from the median value), the other analyte concentrations for that sample were assessed to determine if it reflected a subclinical disorder. If it did, that animal was removed from the reference population. If it did not appear to reflect a disorder, then the outlier was excluded from the reference values but the other values for that sample were kept.

The remaining data were assessed by using Analyse-it[®] to calculate the lower reference limit (2.5-percentile value) and the upper reference limit (97.5-percentile value) for the Hitachi 911 and its reagents. In the attached tables, these results are in the column labeled “New Hitachi 911 RI.” The number of reference values used for the reference intervals are within parentheses. The previous Hitachi reference intervals are included in the “Old Hitachi RI” columns in the attached tables. Those intervals were mostly established by Cornell University for the Hitachi 911; a few were modified in the past 12 years when changes in reagents justified modifications.

For cats and cattle:

Clinicians in the Pet Health Center also collected samples from clinically healthy cats. However, we did not have sufficient number of samples to attempt to calculate new Hitachi feline reference intervals. We did not have bovine samples for calculating new Hitachi bovine reference intervals. The previous feline and bovine Hitachi reference intervals are included in the “Old Hitachi RI” columns in the attached tables. Those intervals were mostly established by Cornell University for the Hitachi 911; a few were modified past 12 years when changes in reagents justified modifications.

Conversion of Reference Intervals

After our new chemistry analyzer (Cobas 501c) was installed, clinical samples were analyzed by both the Hitachi 911 and the Cobas 501c and data were compared and analyzed by Deming regression analysis and bias plots. For most analytes, at least 70 samples were analyzed and included samples that had abnormal analyte concentrations. For some analytes, there was not evidence of either constant or proportional biases. For other analytes, there was evidence of constant, proportional, or both constant and proportional biases.

The formulas for the fitted regression line from Deming regression analysis were used to convert Hitachi reference limits to preliminary Cobas reference limits and the resultant preliminary Cobas reference intervals in the attached tables. Table 5 provides the formula and the fitted regression line data (slope and y-intercept) used for the conversions.

Confirmation of Calculated Serum Chemistry Reference Intervals

During the next few months, we will be analyzing samples from at least 20 clinically healthy animals for an assessment of the calculated Cobas reference intervals. If the preliminary reference intervals are appropriate, at least 19 of the 20 new Cobas values should be within the calculated reference intervals.

Establishment of New Cobas Serum Chemistry Reference Intervals

As part of our continued efforts to obtain reference intervals for all current laboratory methods, we will be analyzing samples from clinically healthy animals for determination of new Cobas reference intervals. When we have at least 60 acceptable reference values for a given species, new reference intervals will be determined.

Conversion of Clinical Data

If there is a need to convert clinical data measured by the Hitachi 911 to values that would have been obtained if the same sample had been analyzed by the Cobas 501c, use the conversion information provided in Table 5. Please note that the calculated values will be approximate values because they do not consider factors which effect analytical precision, specificity, or accuracy. The amount of difference between the original measured value and the calculated Cobas value could be clinically insignificant for some analytes, but clinically significant for others.

The major changes will be found for increased patient values when there is a proportional bias. For example, consider the calculated values if the Hitachi ALT values were 100 U/L or 2,000 U/L using the assay without P5P.

Cobas value = (Slope × Hitachi value) + Y-intercept

If Hitachi value was 100 U/L, then the Cobas value = $(1.119 \times 100) - 5 = 107$ U/L

If Hitachi value was 1000 U/L, then the Cobas value = $(1.119 \times 1000) - 5 = 1114$ U/L

For an individual case, interpretation of a 1000 U/L and 1114 U/L may not be different. However for retrospective studies involving groups of animals, considering data from the Hitachi and the Cobas to be equivalent could result in inappropriate conclusions.

Switching reagent systems for ALT and AST

The ALT and AST activities reported by the Cobas 501c will be using the reagents that contain the cofactor pyridoxal-5'-phosphate (P5P). In doing so, the measured serum activity is not dependent on the amount P5P in the patient sample.

**Table 1: Reference Intervals for Dogs
Serum Chemical Assays
Hitachi 911 to Cobas 501c**

Converted New Hitachi RI with Regression Equations

	Units	Cobas RI	New Hitachi RI (# of values)	Old Hitachi RI
γ-glutamyltransferase	U/L	2-8	3-9 (34)	0-10
Alanine transaminase no PFP*	U/L	14-107	17-100 (46)	13-79
Alanine transaminase with P5P*	U/L	16-142 ¹	15-126 (20)	N/A
Alanine transaminase with P5P*	U/L	28-171 ^{2,3}	17-100 (46)	N/A
Alb:Glob ratio		1.1-3.0	1.0-2.6 (46)	N/A
Albumin	g/dL	3.4-4.2	3.4-4.2 (46)	3.0-4.5
Alkaline phosphatase	U/L	1-142	5-141 (46)	12-122
Amylase	U/L	31--980	329-1054 (43)	454-1380
Anion Gap	mmol/L	16-26	13-23 (46)	N/A
Aspartate transaminase no P5P*	U/L	12-93	20-90 (38)	13-62
Aspartate transaminase with P5P*	U/L	48-74 ¹	32-57 (3)	N/A
Aspartate transaminase with P5P*	U/L	22-102 ^{2,3}	20-90 (38)	N/A
Bicarbonate	mmol/L	18-29	18-29 (46)	16-26
Bile Acid (post)	umol/L	N/A	N/A	0-30
Bile Acid (pre)	umol/L	8-32	7-32 (36)	0-13
Bilirubin (direct)	mg/dL	0.0-0.1	0.0-0.1 (12)	0-0.1
Bilirubin (indirect)	mg/dL	0.0-0.2	0.0-0.2 (7)	N/A
Bilirubin (total)	mg/dL	0.1-0.3	0.1-0.3 (44)	0.1-0.4
Calcium (total)	mg/dL	9.7-12.1	9.9-12.1 (46)	7.2-12.8
Chloride	mmol/L	108-118	108-118 (46)	109-124
Cholesterol	mg/dL	133-394	130-386 (44)	124-335
Creatine kinase	U/L	128-328	45-251 (41)	58-241
Creatinine	mg/dL	0.5-1.5	0.5-1.6 (46)	0.5-1.4
Globulin	g/dL	1.3-3.2	1.5-3.5 (46)	N/A
Glucose	mg/dL	73-113	77-119 (46)	60-120
Hemolytic index		0-137	0-137 (31)	N/A
Icterus index		N/A	N/A	N/A
Lactate dehydrogenase	U/L	30-255	24-250 (21)	31-289
Lipase	U/L	20-152	23-151 (41)	22-216
Lipemia index		0-54	0-54 (33)	N/A
Magnesium (total)	mg/dL	1.8-2.9	1.6-2.9 (43)	1.7-5.0
Na/K ratio		29-42	30-45 (46)	N/A
Phosphorus	mg/dL	2.4-6.4	2.4-6.4 (46)	2.3-6.5
Potassium	mmol/L	3.6-5.3	3.3-5.0 (46)	3.8-5.5
Sodium	mmol/L	147-154	144-152 (46)	141-156
Total Protein	g/dL	5.4-7.6	5.3-7.2 (46)	5.6-7.9
Triglyceride	mg/dL	27-215	26-210 (43)	26-108
Urea nitrogen	mg/dL	9-33	10-33 (45)	8-30
Uric acid	mg/dL	0.0-1.6	0.0-1.0 (9)	0.2-0.9

*P5P (pyridoxal-5'-phosphate)

¹ Conversion of Hitachi P5P values to Cobas P5P values

² Conversion of Hitachi no-P5P values to Cobas P5P values

³ Using this reference interval for the preliminary Cobas RI

N/A – not available

**Table 2: Reference Intervals for Cats
Serum Chemical Assays
Hitachi 911 to Cobas 501c**

Converted with Regression Equations

	Units	Cobas RI	Old Hitachi RI
γ-glutamyltransferase	U/L	0-1	0-2
Alanine transaminase no PFP*	U/L	25-137	27-127
Alanine transaminase with P5P*	U/L	45-217 ^{2,3}	27-127
Alb:Glob ratio		N/A	N/A
Albumin	g/dL	3.2-4.7	3.2-4.7
Alkaline phosphatase	U/L	11-95	15-96
Amylase	U/L	488-2034	515-2210
Anion Gap	mmol/L	N/A	N/A
Aspartate transaminase no P5P*	U/L	3-45	12-48
Aspartate transaminase with P5P*	U/L	13-54 ²	12-48
Bicarbonate	mmol/L	13-20	13-20
Bile Acid (post)	umol/L	1-25	0-25
Bile Acid (pre)	umol/L	1-14	0-13
Bilirubin (direct)	mg/dL	0.0-0.0	0.0-0.1
Bilirubin (indirect)	mg/dL	0.0-0.4	0.0-0.4
Bilirubin (total)	mg/dL	0.0-0.4	0.0-0.4
Calcium (total)	mg/dL	8.0-11.4	8.2-11.5
Chloride	mmol/L	116-130	116-130
Cholesterol	mg/dL	75-271	73-265
Creatine kinase	U/L	153-572	71-502
Creatinine	mg/dL	0.8-2.1	0.8-2.3
Globulin	g/dL	N/A	N/A
Glucose	mg/dL	60-133	63-140
Hemolytic index	---	N/A	N/A
Icterus index	---	N/A	N/A
Lactate dehydrogenase	U/L	63-455	57-451
Lipase	U/L	6-62	10-64
Lipemia index	---	N/A	N/A
Magnesium (total)	mg/dL	2.1-5.9	2.0-6.5
Na/K ratio	---	N/A	N/A
Phosphorus	mg/dL	2.7-6.5	2.7-6.5
Potassium	mmol/L	3.6-5.5	3.3-5.2
Sodium	mmol/L	150-163	148-161
Total Protein	g/dL	6.3-8.6	6.5-8.9
Triglyceride	mg/dL	26-136	25-133
Urea nitrogen	mg/dL	16-35	17-35
Uric acid	mg/dL	0.0-0.7	0.0-0.5

*P5P (pyridoxal-5'-phosphate)

² Conversion of Hitachi no-P5P values to Cobas P5P values

³ Using this reference interval for the preliminary Cobas RI

N/A – not available

Table 3: Reference Intervals for Horses
Serum Chemical Assays
Hitachi 911 to Cobas 501c

Converted New Hitachi RI with Regression Equations

	Units	Cobas RI	New Hitachi RI	Old Hitachi RI
γ-glutamyltransferase	U/L	6-24	7-24 (53)	8-33
Alanine transaminase no P5P*	U/L	0-8	4-12 (35)	2-12
Alanine transaminase with P5P*	U/L	5-16 ¹	4-14 (11)	N/A
Alanine transaminase with P5P*	U/L	6-20 ^{2,3}	4-12 (35)	N/A
Alb:Glob ratio	---	0.5-1.5	0.5-1.3 (56)	N/A
Albumin	g/dL	2.7-3.7	2.6-3.7 (56)	3.0-3.7
Alkaline phosphatase	U/L	76-249	78-245 (54)	75-220
Amylase	U/L	21-27	3-9 (51)	5-10
Anion Gap	mmol/L	10-17	7-15 (56)	N/A
Aspartate transaminase no P5P*	U/L	182-457	167-406 (56)	199-413
Aspartate transaminase with P5P*	U/L	244-543 ^{1,3}	214-484 (54)	N/A
Aspartate transaminase with P5P*	U/L	189-461 ²	167-406 (56)	N/A
Bicarbonate	mmol/L	26-36	26-37 (56)	22-34
Bile Acid	umol/L	4-16	3-15 (52)	0-11
Bilirubin (direct)	mg/dL	0.0-0.3	0.1-0.3 (56)	0.1-0.3
Bilirubin (indirect)	mg/dL	0.3-2.6	0.3-2.5 (48)	N/A
Bilirubin (total)	mg/dL	0.5-2.7	0.5-2.6 (56)	0.7-2.6
Calcium (total)	mg/dL	11.7-14.8	11.8-14.7 (56)	11.2-13.0
Chloride	mmol/L	95-105	95-105 (56)	95-113
Cholesterol	mg/dL	60-134	58-131 (56)	63-118
Creatine kinase	U/L	192-565	111-495 (55)	93-348
Creatinine	mg/dL	0.8-1.8	0.8-2.0 (56)	0.9-1.7
Globulin	g/dL	2.5-4.7	2.8-5.0 (56)	N/A
Glucose	mg/dL	70-118	74-124 (54)	75-117
Hemolytic index	---	0-25	0-25 (28)	N/A
Icterus index	---	1-5	1-5 (29)	N/A
Lactate dehydrogenase	U/L	179-576	173-572 (51)	100-453
Lipase	U/L	5-15	9-18 (56)	12-39
Lipemia index	---	0-25	0-25 (29)	N/A
Magnesium (total)	mg/dL	1.6-2.4	1.4-5.4 (55)	1.3-5.6
Na/K ratio	---	31-58	30-56 (54)	N/A
Phosphorus	mg/dL	1.3-5.1	1.3-5.1 (56)	2.3-4.4
Potassium	mmol/L	2.2-4.8	1.9-4.5 (56)	2.4-5.6
Sodium	mmol/L	135-144	132-141 (56)	131-143
Total Protein	g/dL	5.8-7.5	6.0-7.8 (56)	6.1-7.4
Triglyceride	mg/dL	11-68	11-66 (56)	14-77
Urea nitrogen	mg/dL	9-22	10-23 (56)	9-20
Uric acid	mg/dL	0.2-0.3	0.2-0.3 (8)	0.1-0.4

*P5P (pyridoxal-5'-phosphate)

¹ Conversion of Hitachi P5P values to Cobas P5P values

² Conversion of Hitachi no-P5P values to Cobas P5P values

³ Using this reference interval for the preliminary Cobas RI

N/A – not available

Table 4: Reference Intervals for Cattle
Serum Chemical Assays
Hitachi 911 to Cobas 501c
 Converted with Regression Equations

	Units	Cobas RI	Old Hitachi RI
γ-glutamyltransferase	U/L	10-39	11-39
Alanine transaminase no PFP*	U/L	11-39	12-38
Alanine transaminase with P5P*	U/L	20-64 ^{2,3}	N/A
Alb:Glob ratio	---	N/A	N/A
Albumin	g/dL	3.1-4.3	3.1-4.3
Alkaline phosphatase	U/L	20-76	23-78
Amylase	U/L	28-50	10-34
Anion Gap	mmol/L	N/A	N/A
Aspartate transaminase no P5P*	U/L	43-148	47-138
Aspartate transaminase with P5P*	U/L	53-156 ^{2,3}	N/A
Bicarbonate	mmol/L	21-31	21-31
Bile Acid	umol/L	10-151	9-155
Bilirubin (direct)	mg/dL	0.0-0.1	0.0-0.2
Bilirubin (indirect)	mg/dL	0.0-0.5	0.0-0.4
Bilirubin (total)	mg/dL	0.1-0.5	0.1-0.5
Calcium (total)	mg/dL	8.1-10.3	8.3-10.4
Chloride	mmol/L	92-117	92-117
Cholesterol	mg/dL	75-286	73-280
Creatine kinase	U/L	159-332	77-255
Creatinine	mg/dL	0.5-1.6	0.5-1.7
Globulin	g/dL	N/A	N/A
Glucose	mg/dL	29-73	31-77
Hemolytic index	---	N/A	N/A
Icterus index	---	N/A	N/A
Lactate dehydrogenase	U/L	702-1381	699-1381
Lipase	U/L	3-19	7-22
Lipemia index	---	N/A	N/A
Magnesium (total)	mg/dL	1.9-3.0	1.8-3.1
Na/K ratio	---	N/A	N/A
Phosphorus	mg/dL	4.9-9.0	4.9-9.1
Potassium	mmol/L	4.2-6.3	3.9-6.0
Sodium	mmol/L	138-155	135-153
Total Protein	g/dL	6.0-9.0	6.2-9.3
Triglyceride	mg/dL	7-32	7-32
Urea nitrogen	mg/dL	9-24	10-25
Uric acid	mg/dL	1.0-3.5	0.7-2.1

*P5P (pyridoxal-5'-phosphate)

² Conversion of Hitachi no-P5P values to Cobas P5P values

³ Using this reference interval for the preliminary Cobas RI

N/A – not available

Table 5: Conversion of Hitachi 911 Results to Approximate Cobas 501c Results, March 2008

To convert a Hitachi 911 result to a result that would have approximately been obtained if the same sample had been analyzed by the Cobas 501c, use of following slopes, Y-intercepts, and this formula. The calculated value should be expressed using appropriate significant figures and rounding rules.

$$\text{Cobas value} = (\text{Slope} \times \text{Hitachi value}) + \text{Y-intercept}$$

$$\text{Example for Na}^+: \text{Cobas value} = (\text{Slope} \times \text{Hitachi value}) + \text{Y-intercept} \\ = (0.951 \times 150) + 10 = 153$$

Test	Slope	Y-intercept
Alanine transaminase (ALT) no P5P	1.119	-5
Alanine transaminase with P5P*	1.137	-1
ALT (from no-P5P to P5P reagents)	1.715	-1
Alb:Glob ratio	1.197	-0.1
Albumin	0.986	0.1
Alkaline phosphatase (ALP)	1.033	-4
Amylase (AMS)	0.912	19
Anion Gap	0.981	3.4
Aspartate transaminase (AST) no P5P	1.153	-11
Aspartate transaminase with P5P*	1.055	14
AST (from no-P5P to P5P reagents)	1.137	-1
Bicarbonate	0.977	0.3
Bile Acids (Fasting)	0.966	1
Bile Acids (Post-prandial)	0.966	1
Bilirubin, Direct	1.133	-0.1
Bilirubin, Indirect	1.058	0.0
Bilirubin, Total	1.058	0.0
Calcium (total)	1.054	-0.7
Chloride	0.991	0.7
Cholesterol	1.020	0.6
Creatine kinase (CK)	0.972	84
Creatinine	0.863	0.1
Globulin	0.982	-0.2
Glucose	0.953	0
Lactate dehydrogenase (LD)	0.995	7
Lipase (LPS)	1.031	-4
Magnesium (total)	0.852	0.4
Na:K ratio	0.872	3
Phosphorus	0.991	0.0
Potassium	1.009	0.3
Sodium	0.951	10
Total Protein	0.956	0.1
Triglyceride	1.024	0
Urea Nitrogen	1.022	-1
Uric Acid	1.750	-0.2
γ -glutamyltransferase (GGT)	1.039	-1

*P5P (pyridoxal-5'-phosphate)

Note: Except for the horse conversions, Hitachi 911 ALT and AST activity concentrations were determined with using reagents without P5P. Conversion of enzyme activity without P5P involved comparison of enzyme activity measured by both reagents systems on the Cobas.