Piccolo® MetLac 12 Panel

CE 2797

For In Vitro Diagnostic Use and Professional Use Only
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1. Intended Use

The Piccolo® MetLac 12 Panel reagent disc, used with the Piccolo Xpress® chemistry analyzer, is intended to be used for the *in vitro* quantitative determination of albumin, calcium, chloride, creatinine, glucose, lactate, magnesium, phosphorus, potassium, sodium, total carbon dioxide and blood urea nitrogen (BUN) in lithium heparinized whole blood or lithium heparinized plasma in a clinical laboratory setting or point-of-care location/ near-patient testing.

2. Summary and Explanation of Tests

The Piccolo MetLac 12 Panel reagent disc and the Piccolo Xpress chemistry analyzer comprise an *in vitro* diagnostic system that aids the physician in diagnosing the following disorders in adults:

Albumin:	Liver and kidney disease.
Calcium:	General metabolic disease, for example, Parathyroid, bone and chronic renal diseases; tetany.
Chloride:	General metabolic diseases, for example, Dehydration, prolonged diarrhea and vomiting, renal tubular disease, hyperparathyroidism, burns, salt-losing renal diseases, overhydration and thiazide therapy.
Creatinine:	Renal diseases and renal dialysis.
Glucose:	Carbohydrate metabolism disorders, for example, adult and juvenile diabetes mellitus and hypoglycemia.
Lactate:	Lactate measurements are used in the diagnosis and treatment of Metabolic disorders, for example, lactate acidosis, tissue hypoxia, and diagnosis of hyperlactatemia.
Magnesium:	Hypomagnesemia and hypermagnesemia.
Phosphorus:	General metabolic disorders, for example, Dehydration, diabetes, parathyroidism, and renal disease.
Potassium:	Renal, endocrine, and metabolic disease as well as iatrogenic causes, for example, Renal glomerular or tubular disease, adrenocortical insufficiency, diabetic ketoacidosis, excessive intravenous potassium therapy, sepsis, panhypopituitarism, *in vitro* hemolysis, hyperaldosteronism, malnutrition, hyperinsulinism, metabolic alkalosis and gastrointestinal loss.
Sodium:	Metabolic diseases, for example, Dehydration, diabetes insipidus, loss of hypotonic gastrointestinal fluids, salt poisoning, selective depression of the sense of thirst, skin loss, burns, sweating, hyperaldosteronism, CNS disorders, dilutional, depletional and delusional hyponatremia and ADH secretion syndrome.
Total Carbon Dioxide:	Metabolic alkalosis and acidosis and primary respiratory alkalosis and acidosis.
Blood urea nitrogen (BUN):	Renal diseases.

As with any diagnostic test procedure, all other test procedures including the clinical status of the patient, should be considered prior to final diagnosis.

3. Principle of Method

Albumin (ALB)
Early methods used to measure albumin include fractionation techniques[1,2,3] and tryptophan content of globulins.[4,5] These methods were unwieldy to perform and did not have a high specificity. Two immunochemical techniques are considered as reference methods, but are expensive and time consuming.[6] Dye binding techniques are the most frequently used methods for measuring albumin. Bromocresol green (BCG) is the most commonly used of the dye binding methods but may over-estimate albumin concentration, especially at the low end of the normal range.[7] Bromocresol purple (BCP) is the most specific of the dyes in use.[8,9]

Bromcresol purple (BCP), when bound with albumin, changes color from a yellow to blue color. The absorbance maximum changes with the color shift.

$$\text{BCP + Albumin} \xrightarrow[\text{Acid pH}]{\text{Surfactants}} \text{BCP-Albumin Complex}$$

Bound albumin is proportional to the concentration of albumin in the sample. This is an endpoint reaction that is measured as absorbance at 600 nm.

Calcium (CA)
The first methods used to analyze calcium involved precipitating calcium with an excess of anions.[10,11,12] Precipitation methods are laborious and often imprecise. The reference method for calcium is atomic absorption spectroscopy; however, this method is not suited for routine use.[13] Spectrophotometric methods using either *o*-cresolphthalein complexone (CPC) or arsenazo III metallochromic indicators are most commonly used.[14,15,16] Arsenazo III has a high affinity for calcium and is not temperature dependent as is CPC.

Calcium in the patient sample binds with arsenazo III to form a calcium-dye complex.

$$Ca^{2+} + \text{Arsenazo III} \longrightarrow Ca^{2+}\text{-Arsenazo III Complex}$$

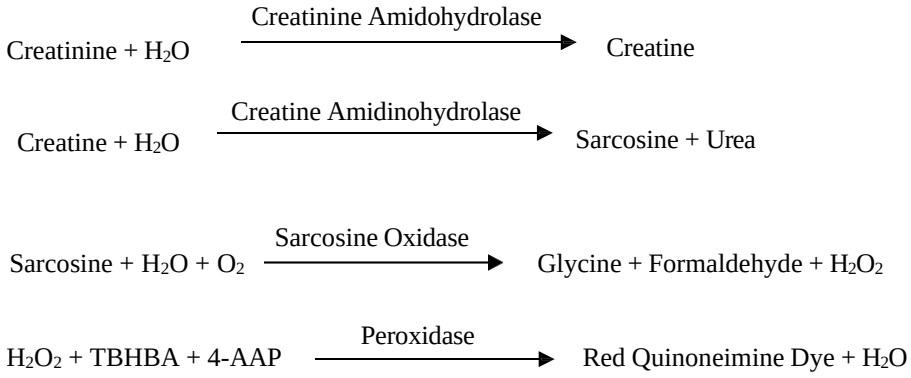
The endpoint reaction is monitored at 405 nm, 467 nm, and 600 nm. The amount of total calcium in the sample is proportional to the absorbance.

Chloride (CL^-)
The Abaxis chloride method is based on the determination of chloride-dependent activation of α-amylase activity. Deactivated α-amylase is reactivated by addition of the chloride ion, allowing the calcium to re-associate with the enzyme. The reactivation of α-amylase activity is proportional to the concentration of chloride ions in the sample. The reactivated α-amylase converts the substrate, 2-chloro-*p*-nitrophenyl-α-D-maltotrioside (CNPG3) to 2-chloro-*p*-nitrophenyl (CNP) producing color and α-maltotriose (G3). The reaction is measured bichromatically and the increase absorbance is directly proportional to the reactivated α-amylase activity and the concentration of chloride in the sample.[17]

$$\text{CNPG3} \xrightarrow[Cl^-,\ Ca^{2+}]{\alpha\text{-Amylase}} \text{CNP + G3}$$

Creatinine (CRE)
The Jaffe method, first introduced in 1886, is still a commonly used method of determining creatinine levels in blood. The current reference method combines the use of Fuller's earth (floridin) with the Jaffe technique to increase the specificity of the reaction.[18,19] Enzymatic methods have been developed that are more specific for creatinine than the various modifications of the Jaffe technique.[20,21,22] Methods using the enzyme creatinine amidohydrolase eliminate the problem of ammonium ion interference found in techniques using creatinine iminohydrolase.[23]

In the coupled enzyme reactions, creatinine amidohydrolase hydrolyzes creatinine to creatine. A second enzyme, creatine amidinohydrolase, catalyzes the formation of sarcosine from creatine. Sarcosine oxidase causes the oxidation of sarcosine to glycine, formaldehyde and hydrogen peroxide (H_2O_2). In a Trinder reaction, peroxidase catalyzes the reaction among the hydrogen peroxide, 2,4,6-tribromo-3-hydroxybenzoic acid (TBHBA) and 4-aminoantipyrine (4-AAP) into a red quinoneimine dye. Potassium ferrocyanide and ascorbate oxidase are added to the reaction mixture to minimize the potential interference of bilirubin and ascorbic acid, respectively.



Two cuvettes are used to determine the concentration of creatinine in the sample. Endogenous creatine is measured in the blank cuvette, which is subtracted from the combined endogenous creatine and the creatine formed from the enzyme reactions in the test cuvette. Once the endogenous creatine is eliminated from the calculations, the concentration of creatinine is proportional to the intensity of the red color produced. The endpoint reaction is measured as the difference in absorbance between 550 nm and 600 nm.

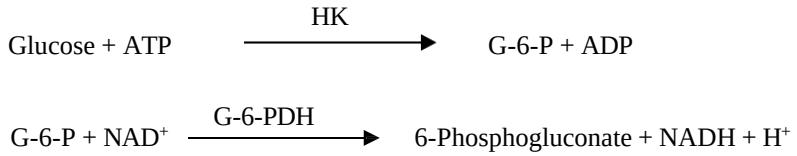
eGFR (calculated)
Serum creatinine is routinely measured as an indicator of renal function. Because creatinine is influenced by age, gender and race, chronic kidney disease (CKD) may not be detected using serum creatinine alone. Thus, the National Kidney Disease Education Program strongly recommends that laboratories routinely report an estimated Glomerular Filtration Rate (eGFR) when serum creatinine is measured for patients 18 and older. Routinely reporting the eGFR with all serum creatinine determinations allows laboratories to help identify individuals with reduced kidney function and help facilitate the detection of CKD. Calculated eGFR values of <60 mL/min are generally associated with increased risk of adverse outcomes of CKD.[24,25,26]

Calculation of the eGFR is performed by the Piccolo using the patient's age, gender and race. The Piccolo method for creatinine is traceable to the IDMS reference method for creatinine so that the following form of the MDRD equation for calculating the eGFR can be used.

GFR (mL/min/1.73 m^2) = 175 x $(S_{cr})^{-1.154}$ x $(Age)^{-0.203}$ x (0.742 if female) x (1.212 if African American)

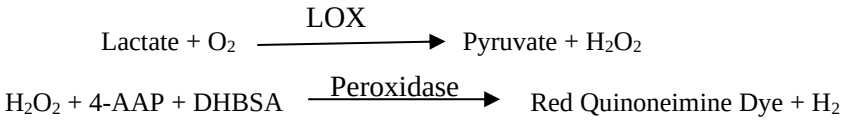
Glucose (GLU)
Measurements of glucose concentration were first performed using copper-reduction methods (such as Folin-Wu[27] and Somogyi-Nelson[28,29]). The lack of specificity in copper-reduction techniques led to the development of quantitative procedures using the enzymes hexokinase and glucose oxidase. The glucose test incorporated into the Piccolo MetLac 12 Panel reagent disc is a modified version of the hexokinase method, which has been proposed as the basis of the glucose reference method.[30]

The reaction of glucose with adenosine triphosphate (ATP), catalyzed by hexokinase (HK), produces glucose-6-phosphate (G-6-P) and adenosine diphosphate (ADP). Glucose-6-phosphate dehydrogenase (G-6-PDH) catalyzes the reaction of G-6-P into 6-phosphogluconate and the reduction of nicotinamide adenine dinucleotide (NAD^+) to NADH.



The absorbance is measured bichromatically at 340 nm and 850 nm. The production of NADH is directly proportional to the amount of glucose present in the sample.

Lactate (LAC)
In the Abaxis method, lactate (LAC) is oxidized by lactate oxidase (LOX) to pyruvate and hydrogen peroxide (H_2O_2). Peroxidase catalyzes the reaction of H_2O_2, 4-aminoantipyrine (4-AAP) and 3,5-dichloro-2-hydroxybenzenesulfonic acid (DHBSA) to form a red quinoneimine dye.


Lactate + O2
LOX
Pyruvate + H2O2
H2O2 + 4-AAP + DHBSA
Peroxidase
Red Quinoneimine Dye + H2

The rate of formation of the red dye is proportional to the LAC concentration in the sample. The reaction is measured bichromatically at 515 nm and 600 nm.

Magnesium (MG)
The hexokinase (HK) activation method for magnesium is the best-fit method for the Piccolo system in terms of sensitivity, precision, and accuracy.[31] The enzymatic magnesium method can be described as :

$$\text{Glucose + ATP} \xrightarrow[Mg^{2+}]{\text{HK}} \text{G-6-P + ADP}$$

$$\text{G-6-P + NADP}^{+} \xrightarrow{\text{G-6-PDH}} \text{6-Phosphogluconate + NADPH + H}^{+}$$

The rate limiting reaction is the HK reaction. Magnesium from the sample activates HK, which in turn catalyzes the break down of glucose to form glucose-6-phosphate (G-6-P) and ADP. G-6-P reacts with nicotinamide adenine dinucleotide phosphate ($NADP^+$) to form reduced nicotinamide adenine dinucleotide phosphate (NADPH) and 6-phosphogluconate in the presence of glucose-6-phosphate-dehydrogenase (G-6-PDH). This is a first-order rate reaction. The rate of production of NADPH is directly proportional to the amount of magnesium present in the sample. Absorbance is measured bichromatically at 340 nm and 405 nm.

Phosphorus (PHOS)
The most applicable enzymatic method for the Abaxis system uses sucrose phosphorylase (SP) coupled through phosphoglucomutase (PGM) and glucose-6-phosphate dehydrogenase (G6PDH).[32,33] Using the enzymatic system for each mole of phosphorus present in the sample, one mole of NADH is formed. The amount of NADH formed can be measured as an endpoint at 340 nm.

$$\text{Sucrose + } P_i \xrightarrow{\text{SP}} \text{Glucose-1-Phosphate (G-1-P) + Fructose}$$

$$\text{G-1-P} \xrightarrow{\text{PGM, } Mg^{2+}} \text{G-6-P}$$

$$\text{G-6-P + NAD}^{+} \xrightarrow{\text{G-6-PDH}} \text{NADH + 6-Phosphogluconate + H}^{+}$$

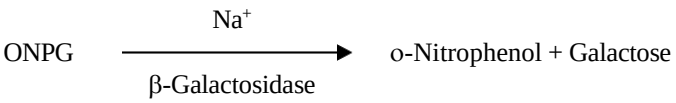
Potassium (K^+)
Spectrophotometric methods have been developed that allow the measurement of potassium concentration on standard clinical chemistry instrumentation. The Abaxis enzymatic method is based on the activation of pyruvate kinase with potassium and shows excellent linearity and negligible susceptibility to endogenous substances.[34,35,36] Interference from sodium and ammonium ions are minimized with the addition of Kryptofix and glutamine synthetase respectively.[34]

In the coupled-enzyme reaction, pyruvate kinase (PK) dephosphorylates phosphoenolpyruvate to form pyruvate. Lactate dehydrogenase (LDH) catalyzes conversion of pyruvate to lactate. Concomitantly, NADH is oxidized to NAD^+. The rate of change in absorbance difference between 340 nm and 405 nm is due to the conversion of NADH to NAD^+ and is directly proportional to the amount of potassium in the sample.

$$\text{ADP + PEP} \xrightarrow{K^+,\ \text{PK}} \text{Pyruvate + ATP}$$

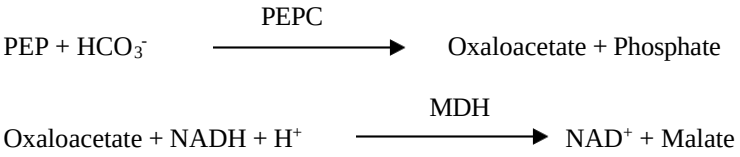
$$\text{Pyruvate + NADH + H}^{+} \xrightarrow{\text{LDH}} \text{Lactate + NAD}^{+}$$

Sodium (NA^+)
Colorimetric and enzymatic methods have been developed that allow the measurement of sodium concentration on standard clinical chemistry instrumentation.[37,38,39] In the Abaxis enzymatic reaction, β-galactosidase is activated by the sodium in the sample. The activated enzyme catalyses the reaction o-nitrophenyl-β-galactopyranoside (ONPG) to o-nitrophenyl and galactose.



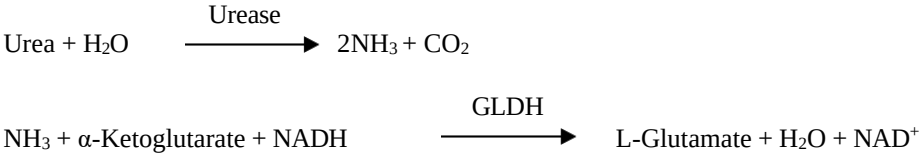
Total Carbon Dioxide (tCO_2)
Total carbon dioxide in serum or plasma exists as dissolved carbon dioxide, carbamino derivatives of proteins, bicarbonate and carbonate ions and carbonic acid. Total carbon dioxide can be measured by pH indicator, CO_2 electrode and spectrophotometric enzymatic methods, which all produce accurate and precise results.[40,41] The enzymatic method is well suited for use on a routine blood chemistry analyzer without adding complexity.

In the enzymatic method, the specimen is first made alkaline to convert all forms of carbon dioxide (CO_2) to bicarbonate (HCO_3^-). Phosphoenolpyruvate (PEP) and HCO_3^- then react to form oxaloacetate and phosphate in the presence of phosphoenolpyruvate carboxylase (PEPC). Malate dehydrogenase (MDH) catalyzes the reaction of oxaloacetate and reduced nicotinamide adenine dinucleotide (NADH) to NAD^+ and malate. The rate of change in absorbance due to the conversion of NADH to NAD^+ is directly proportional to the amount of tCO_2 in the sample.



Blood Urea Nitrogen (BUN)
Urea can be measured both directly and indirectly. The diacetyl monoxime reaction, the only direct method to measure urea, is commonly used but employs dangerous reagents.[42] Indirect methods measure ammonia created from the urea; the use of the enzyme urease has increased the specificity of these tests.[43] The ammonia is quantitated by a variety of methods, including nesslerization (acid titration), the Berthelot technique [44,45] and coupled enzymatic reactions.[46,47] Catalyzed Berthelot procedures, however, are erratic when measuring ammonia.[48] Coupled-enzyme reactions are rapid, have a high specificity for ammonia, and are commonly used. One such reaction has been proposed as a candidate reference method.[49]

In the coupled-enzyme reaction, urease hydrolyzes urea into ammonia and carbon dioxide. Upon combining ammonia with α-ketoglutarate and reduced nicotinamide adenine dinucleotide (NADH), the enzyme glutamate dehydrogenase (GLDH) oxidizes NADH to NAD^+.



The rate of change of the absorbance difference between 340 nm and 405 nm is caused by the conversion of NADH to NAD^+ and is directly proportional to the amount of urea present in the sample.

4. Principle of Operation

Refer to the Piccolo Xpress chemistry analyzer Operator's Manual, for the Principles and Limitations of the Procedure.

5. Description of Reagents

Reagents
Each Piccolo MetLac 12 Panel reagent disc contains dry test-specific reagent beads (described below). A dry sample blank reagent (comprised of buffer, surfactants, excipients, and preservatives) is included in each disc for use in calculating concentrations of albumin (ALB), chloride (CL^-), calcium (CA), glucose (GLU), lactate (LAC), magnesium (MG), phosphorus (PHOS), potassium (K+), sodium (NA+), total carbon dioxide (tCO_2) and blood urea nitrogen (BUN). A dedicated sample blank is included in the disc to calculate concentrations of creatinine (CRE). Each disc also contains a diluent consisting of surfactants and preservatives.

Table 1: Reagents

Component	Quantity/Disc
N-Acetyl cysteine	23 µg
Adenosine 5'-diphosphate	36 µg
Adenosine-5'-diphosphate, Lithium salt	4 µg
Adenosine 5'- Triphosphate, Disodium Salt	28 µg
α-Ketoglutaric acid	18 µg
4-Aminoantipyrine	5.4 µg
4-Aminoantipyrine hydrochloride	0 µg
Amylase	0.286 U
Arsenazo III, sodium salt	3.5 µg
Ascorbate oxidase	0.3 U
Brij	39 µg
Bromocresol purple, sodium salt	2.3 µg
Calcium acetate	50 µg
Citric acid	167 µg
Citric acid, trisodium salt	1134 µg
2-Chloro-4-nitrophenyl-α-maltotrioside (CNPG3)	105 µg
Creatine amidinohydrolase	3 U
Creatinine amidohydrolase	1 U
3,5-Dichloro-2-hydroxy-benzenesufonic acid, sodium salt (DHBSA)	66 µg
Ethylenediaminetetraacetic acid (EDTA)	15 µg
Ethylenediaminetetraacetic acid (EDTA), disodium salt	383 µg
Ethylenediaminetetraacetic acid (EDTA), Tetrasodium Salt	180 µg
Ethylene glycol-bis(β-aminoethyl ether)-N,N,N',N'-tetraacetic acid (EGTA)	22 µg
β-Galactosidase	0.009 U
Glucose-1,6-diphosphate	1 µg
L-Glutamic acid	9.2 µg
Glucose-6-Phosphate Dehydrogenase	0.1 U
Glutamate dehydrogenase	0 U
Glutamine synthetase	0.17 U
Hexokinase	0.1 U
Imidazole	57 µg
Lactate dehydrogenase	0.13 U
Lactate oxidase	0.002 U
Lactate dehydrogenase	0 U
Lactate dehydrogenase (LDH)	0.1 U
Lithium hydroxide, monohydrate	102 µg
Magnesium acetate, tetrahydrate	7 µg
Magnesium Chloride, hexahydrate	2 µg
Magnesium sulfate, heptahydrate	122 µg
Malate dehydrogenase	0.1 U
Manganese chloride	10 µg
D-Mannitol	4179 µg
Methylated Cyclodextrin	314 µg
2-Methyl-4-isothizolin-3-one hydrochloride (MIT)	0 µg
β-Nicotinamide Adenine Dinucleotide (NAD)	62 µg
β-Nicotinamide Adenine Dinucleotide Phosphate, Sodium Salt (NADP)	30 µg
ß-Nicotinamide adenine dinucleotide, reduced (NADH)	34 µg
o-Nitrophenyl-β-D-galactopyranoside (ONPG)	44 µg
n-Octylglucoside	31 µg
4,7,13,16,21-Pentaoxa-1,10-diazabicyclo[8.8.5]tricosane	139 µg
Peroxidase	1 U
Phosphoenol pyruvate	14 µg
Phosphoenol pyruvate carboxylase	0.002 U
Phosphoglucomutase	0.141 U
Pluronic F68	18 µg
Polyethylene glycol, 8000	883 µg
Potassium ferrocyanide	1.1 µg
Pyruvate kinase	0.01 U
Sarcosine oxidase	1 U
Sucrose	351 µg

Sucrose phosphorylase	0.28 U
Sodium chloride	784 µg
2,4,6-Tribromo-3-hydroxybenzoic acid (TBHBA)	188 µg
Triethanolamine hydrochloride	314 µg
Triton X-100	281 µg
Urease	0.05 U
Uricase	0 U
Buffers, surfactants, excipients and preservatives	

Warnings and Precautions

- For *in vitro* Diagnostic Use
- The diluent container in the reagent disc is automatically opened when the analyzer drawer closes. A disc with an opened diluent container cannot be re-used. Ensure that the sample or control has been placed into the disc before closing the drawer.
- Used reagent discs contain human body fluids. Follow good laboratory safety practices when handling and disposing of used discs.[50] See the Piccolo Xpress chemistry analyzer Operator's Manual for instructions on cleaning biohazardous spills.
- The reagent discs are plastic and may crack or chip if dropped. Never use a dropped disc as it may spray biohazardous material throughout the interior of the analyzer.
- Reagent beads may contain acids or caustic substances. The operator does not come into contact with the reagent beads when following the recommended procedures. In the event that the beads are handled (e.g., cleaning up after dropping and cracking a reagent disc), avoid ingestion, skin contact, or inhalation of the reagent beads.
- This mixture does not contain substances assessed to be vPvB / PBT according to Regulation (EC) No 1907/2006, Annex XIII. This product contains components considered to have endocrine-disrupting properties for environment, according to REACH Article 57(f), Regulation (EU) 2018/605 or Regulation (EU) 2017/2100. Refer to the Safety Data Sheet (SDS) for specific details.
- Materials incorporated into the device may consist of substances derived from other microbial and animal sourced materials.

Instructions for Reagent Handling

Reagent discs may be used directly from the refrigerator without warming. Do not allow discs to remain at room temperature at 20-25°C (68-77°F) longer than 48 hours prior to use. Open the sealed foil pouch and remove the disc, being careful not to touch the bar code ring located on the top of the disc. Use according to the instructions provided in the Piccolo Xpress chemistry analyzer Operator's Manual. A disc not used within 20 minutes of opening the pouch should be discarded.

Storage

Store reagent discs in their sealed pouches at 2-8 °C (36-46 °F). Do not expose opened or unopened discs to direct sunlight or temperatures above 32 °C (90 °F). Reagent discs may be used until the expiration date included on the package. The expiration date is also encoded in the bar code printed on the bar code ring. An error message will appear on the Piccolo Xpress chemistry analyzer display if the reagents have expired.

Indications of Reagent Disc Instability/Deterioration

A torn or otherwise damaged pouch may allow moisture to reach the unused disc and adversely affect reagent performance. Do not use a disc from a damaged pouch.

6. Instrument

See the Piccolo Xpress chemistry analyzer Operator's Manual for complete information on use of the analyzer.

7. Sample Collection and Preparation

Sample collection techniques are described in the, "Sample Collection" section of the Piccolo Xpress chemistry analyzer Operator's Manual.

- The minimum required sample size is ~100 µL of heparinized whole blood, heparinized plasma, or control material. The reagent disc sample chamber can contain up to 120 µL of sample.

- Whole blood samples obtained by venipuncture must be homogeneous before transferring a sample to the reagent disc. Gently invert the collection tube several times just prior to sample transfer. Do not shake the collection tube; shaking may cause hemolysis.

- Whole blood samples should only be obtained via venipuncture, not from capillary blood.

- Hemolysis may cause erroneously high results in **potassium** assays. This problem may go undetected when analyzing whole blood (release of potassium from as few as 0.5% of the erythrocytes can increase the potassium serum level by 0.5 mmol/L). In addition, even unhemolyzed specimens that are not promptly processed may have increased potassium levels due to intracellular leakage.[51]

- Whole blood venipuncture samples for **lactate** determination require that the patient should be at rest for 2 hours, avoid any forearm exercise, and the blood should be obtained either without a tourniquet or immediately after the tourniquet is applied. Lactate should be analyzed or blood cells separated from the plasma as soon as possible after collection. Lactate in whole blood increases rapidly because of glycolysis.[52] Blood lactate increases by 0.01 to 0.02 mmol/L/minute at room temperature in the absence of antiglycolytic agents.[53]

- For all other methods, whole blood venipuncture samples should be run within 60 minutes of collection.[54,55] **Glucose** concentrations are affected by the length of time since the patient has eaten and by the type of sample collected from the patient. To accurately determine glucose results, samples should be obtained from a patient who has been fasting for at least 12 hours. The glucose concentration decreases approximately 0.28-0.67 mmol/L in 1 hour in uncentrifuged samples stored at room temperature.[56]

- Refrigerating whole blood samples can cause significant changes in concentrations of **creatinine** and **glucose**.[57] The sample may be separated into plasma or serum and stored in capped sample tubes at 2-8 °C (36-4 6°F) if the sample cannot be run within 60 minutes.

- Use only lithium heparin evacuated specimen collection tubes for whole blood or plasma samples. Use no-additive evacuated specimen collection tubes or serum separator tubes for serum samples.

- The concentration of **total carbon dioxide** is most accurately determined when the assay is done immediately after opening the tube and as promptly as possible after collection and processing of the blood in the unopened tube. Ambient air contains far less carbon dioxide than does plasma and gaseous dissolved carbon dioxide will escape from the specimen into the air, with a consequent decrease in carbon dioxide value of up to 6 mmol/L in the course of 1 hour.[58]

- Start the test within 10 minutes of transferring the sample into the reagent disc.

8. Procedure

Materials Provided
- One Piccolo MetLac 12 Panel PN: 400-1037 (a box of discs PN 400-0037)

Materials Required but Not Provided
- Piccolo Xpress chemistry analyzer
- Sample transfer pipettes (fixed volume approximately 100 µL) and tips are provided with each Piccolo Xpress chemistry analyzer and may be reordered from Abaxis.
- Commercially available control reagents recommended by Abaxis (contact Abaxis Technical Support for approved control materials and expected values).
- Timer

Test Parameters
The Piccolo Xpress chemistry analyzer operates at ambient temperatures between 15 °C and 3 2°C (59-90 °F). The analysis time for each Piccolo MetLac 12 Panel reagent disc is less than 14 minutes. The analyzer maintains the reagent disc at a temperature of 37°C (98.6°F) over the measurement interval.

Test Procedure
The complete sample collection and step-by-step operating procedures are detailed in the Piccolo Xpress chemistry analyzer Operator's Manual.

Calibration

The Piccolo Xpress chemistry analyzer is calibrated by the manufacturer before shipment. The bar code printed on the reagent disc bar code ring provides the analyzer with disc-specific calibration data. See the Piccolo Xpress chemistry analyzer Operator's Manual.

The Piccolo Xpress chemistry analyzer is calibrated using internal calibrators or reference materials. The methods and materials used in the procedures for the calibration and control value assignments are traceable either to the comparative method and/or to the standards listed below:

- IFCC – International Federation of Clinical Chemistry
- NIST – National Institute of Standards and Technology
- CRMLN – Cholesterol Reference Method Laboratory Network
- SRM – Standard Reference Material
- CRM – Certified Reference Material

The calibration and value assignment processes are in compliance with EN ISO 17511, Metrological Traceability of Values Assigned to Calibrators and Control Materials.

The measured analytes in the MetLac 12 are traceable to the following reference materials or methods. The Piccolo Xpress Analysis System controls and calibration verification materials are validated for use only with the Piccolo Xpress Analysis System and assigned values may not be commutable with other methods. For specific analyte reference standard methods, refer to Section 3. Principles of Method.

Analyte	Calibration Standard	Method
ALB (BCP)	Correlation to Beckman LX-20 / DX-20	Dye Binding Bromocresol Purple
CA	Correlation to Beckman LX-20 / DX-20	Arsenazo III Dye
CL	Correlation to Beckman LX-20 / DX-20	Enzymatic
CRE	NIST SRM #967	Enzymatic
GLU	NIST SRM #909	Enzymatic
LAC	Correlation to i-Stat CG4+	Enzymatic
Mg	NIST SRM #909B	Enzymatic
PHOS	SRM #3186	Enzymatic
K+	NIST SRM #909	Enzymatic
NA+	Correlation to Beckman LX-20 / DX-20	Enzymatic
tCO2	Correlation to Beckman LX-20 / DX-20	Enzymatic
BUN	NIST SRM #912A	Enzymatic

Quality Control

See Section 6 (Calibration and Quality Control) of the Piccolo Xpress chemistry analyzer Operator's Manual. Performance of the Piccolo Xpress chemistry analyzer can be verified by running controls. For a list of approved quality control materials with acceptance ranges, please contact Abaxis Technical Support. Other human serum or plasma-based controls may not be compatible. Quality control materials should be stored as per the package-insert included with the controls.

If control results are out of range, repeat one time. If still out of range, call Technical Support. Do not report results if controls are outside their labeled limits. See the Piccolo Xpress chemistry analyzer Operator's Manual for a detailed discussion on running, recording, interpreting, and plotting control results.

Waived Laboratories (US only): Abaxis recommends control testing as follows:

- at least every 30 days
- whenever the laboratory conditions have changed significantly, e.g. Piccolo moved to a new location or changes in temperature control
- when training or retraining of personnel is indicated

- with each new lot (CLIA waived tests in waived status labs)

Non-Waived Laboratories (US only): Abaxis recommends control testing to follow federal, state, and local guidelines.

And the non-waived ones will use the below:

- Performance of the Piccolo Xpress chemistry analyzer can be verified by running controls. For a list of approved quality control materials with acceptance ranges, please contact Abaxis Technical Support. Other human serum or plasma-based controls may not be compatible. Quality control materials should be stored as per the package-insert included with the controls.

- If control results are out of range, repeat one time. If still out of range, call Technical Support. Do not report results if controls are outside their labeled limits. See the Piccolo Xpress chemistry analyzer Operator's Manual for a detailed discussion on running, recording, interpreting, and plotting control results.

9. Results

The Piccolo Xpress chemistry analyzer automatically calculates and prints the analyte concentrations in the sample. Details of the endpoint and rate reaction calculations are found in the Piccolo Xpress chemistry analyzer Operator's Manual.

Interpretation of results is detailed in the analyzer Operator's Manual. Results are printed onto paper rolls supplied by Abaxis. The paper roll has an adhesive backing for easy placement in the patient's files.

The reaction for each analyte occurs at 37°C (98.6°F).

10. Limitations of Procedure

General procedural limitations are discussed in the Piccolo Xpress chemistry analyzer Operator's Manual.

- The only anticoagulant **recommended for use** with the Piccolo blood chemistry analyzer or the Piccolo Xpress chemistry analyzer is **lithium heparin**. Abaxis has performed studies demonstrating that EDTA, fluoride, oxalate, and any anticoagulant containing ammonium ions will interfere with at least one chemistry contained in the Piccolo MetLac 12 Panel reagent disc. Do not use sodium heparin.

- Samples with hematocrits in excess of 62% packed red cell volume (a volume fraction of 0.62) may give inaccurate results. Samples with high hematocrits may be reported as hemolyzed. These samples may be spun down to get plasma. The plasma can then be re-run in a new reagent disc.

- **Any result for a particular test that exceeds the assay range should be analyzed by another approved test method or sent to a referral laboratory. Do not dilute the sample and run it again on** the Piccolo Xpress chemistry analyzer.

  Warning: Extensive testing of the Piccolo Xpress chemistry analyzer has shown that, in very rare instances, sample dispensed into the reagent disc may not flow smoothly into the sample chamber. Due to the uneven flow, an inadequate quantity of sample may be analyzed and several results may fall outside the reference ranges. The sample may be re-run using a new reagent disc.

  Note: Operators shall report any serious incident that has occurred in relation to the device to the manufacturer.

Interference
Substances were tested as interferents with the analytes. For lactate human plasma pools (for all other analytes human serum pools) were prepared. The concentration at which each potential interferent was tested was based on the testing levels in CLSI (formerly NCCLS) EP7-P[59] and CLSI EP7-A2.[60]

Effects of Endogenous Substances
- Physiological interferents (hemolysis, icterus and lipemia) cause changes in the reported concentrations of some analytes. The sample indices are printed on the bottom of each result tape to inform the operator about the levels of interferents present in each sample.

- The Piccolo Xpress chemistry analyzer suppresses any results that are affected by >10% interference from hemolysis, lipemia or icterus. "HEM", "LIP", or "ICT" respectively, is printed on the printout in place of the result.

- Extremely elevated amylase levels (>9,000 U/L) will have a significant effect, >10% increase, on the chloride result. The

concentration of amylase is not evaluated by the Piccolo system for each specimen.

- The potassium assay in the Piccolo system is a coupled pyruvate kinase (PK) / lactate dehydrogenase (LDH) assay. Therefore, in cases of extreme muscle trauma or highly elevated levels of creatine kinase (CK), the Piccolo may recover a falsely elevated potassium (K+) value. In such cases, unexpected high potassium recoveries need to be confirmed utilizing a different methodology.

- For maximum levels of endogenous substances contact Abaxis Technical Support.

Effects of Exogenous and Therapeutic Substances

For lactate, 41 exogenous and therapeutic substances were selected as potential interferents for Abaxis test methods based on recommendations by Young.[61] For all other methods, 35 exogenous and therapeutic substances were selected and tested. Significant interference is defined as greater than ± 10% shift in the result for a normal range specimen. For lactate, human plasma pools were supplemented with known concentrations of the drugs or chemicals and then analyzed. For all other methods, human serum pools were used. **Please see Table 2 for a list of exogenous and therapeutic substances evaluated**. **Please see Table 3 for a list of analytes where interference was observed.**

Table 2: Exogenous and Therapeutic Substances Evaluated

Potential Interferent	Highest Concentration Tested[A] (mg/dL unless otherwise specified)
Acetaminophen	100
Acetoacetate	102
Acetylsalicylic Acid	50
Ampicillin	30
Ascorbic acid	20
Bromide*	30
Caffeine	10
Calcium Chloride	20
Cephalothin (Keflin)	400
Chloramphenicol	100
Cimetidine	16
Digoxin*	5
D-lactate*	45
Dopamine	19
Epinephrine	1
Erythromycin	10
Glucose*	700
Glutathione	30
Glycolic acid*	7.6
Hydrochlorothiazide	7.5
Hydroxyurea*	0.7
Ibuprofen	50
Isoniazide	4
α-Ketoglutarate	5
Ketoprofen	50
L-dopa	5
Lidocaine	1
Lithium Lactate**	84
Metformin*	500
Methicillin	100
Methotrexate	0.5
Metronidazole	5
Nafcillin	1
Nitrofurantoin	20
Oxacillin	1
Oxaloacetate	132
Penicillin G	100
Phenytoin (5,5-Diphenylhydantion)	3
Proline	4
Pyruvate*	44
Rifampin	0.5

Salicylic Acid	50
Sulfasalazine	150
Sulfanilamide	50
Theophyline	20
Uric Acid*	30

*Tested for lactate only
**Omitted for lactate only

Please see Table 3 for a list of analytes where interference was observed.

Table 3: The following substances showed greater than ± 10 % shift in the result for a normal range specimen

	Concentration Which Produces > 10% Interference
Albumin	
Acetoacetate	*102*
Ampicillin	30
Caffeine	*10*
Calcium chloride	20
Cephalothin (Keflin)	400
Ibuprofen	50
α-Ketoglutarate	5
Nitrofurantoin	20
Proline	4
Sulfasalazine	10
Sulfanilamide	50
Theophylline	20
Creatinine	
Ascorbic acid	20
Dopamine	19
L-dopa	5
Epinephrine	1
Glutathione	30
Glucose	
Oxaloacetate	*132*
Pyruvate	44
Lactate	
Dopamine	13
Dopamine	0.52
L-dopa	5
L-dopa	0.50
Magnesium	None
Phosphorus	
Nitrofurantoin	20
Oxaloacetate	132
Potassium	
Penicillin G	100
Sulfasalazine	150
Sodium	
Cephalothin	400
Methotrexate	0.5
Penicillin G	100

Total Carbon Dioxide	
Acetaminophen	100
Ascorbic Acid	20
Cephalothin	400
Cimetidine	16
Erythromycin	10
Lidocaine	1
Methotrexate	0.5
Nitrofurantoin	20
Salicylic Acid	50
Sulfasalazine	150

- For the Chloride assay, bromide at toxic levels (≥ 15 mmol/L) can cause a significant effect (> 10% increase), on the chloride result. Iodide at very high concentrations (30 mmol/L, highest level tested) has no effect. Normal physiological levels of bromide and iodide do not interfere with the Piccolo Chloride Test System.

11. Expected Values

Samples from approximately 90 – 140 adult males and females were analyzed on the Piccolo Xpress chemistry analyzer to determine the reference intervals for the following assays. These intervals are provided as a guideline only. It is recommended that your office or institution establish normal ranges for your particular patient population.[62,63]
Intervals

Table 4: Piccolo Reference Intervals

Analyte	Common Units	SI Units
Albumin	3.3-5.5 g/dL	33-55 g/L
Calcium	8.0-10.3 mg/dL	2.0-2.58 mmol/L
Chloride	98-108 mmol/L	98-108 mmol/L
Creatinine	0.6-1.2 mg/dL	53-106 µmol/L
Glucose	73-118 mg/dL	4.1-6.6 mmol/L
Lactate	4.8 – 18.9 mg/dL*	0.53 – 2.10 mmol/L
Magnesium	1.6 – 2.3 mg/dL	0.66 – 0.95 mmol/L
Phosphorus (plasma)	2.2-4.1 mg/dL	0.71-1.32 mmol/L
Phosphorus (serum)	2.5-4.4 mg/dL**	0.81-1.42 mmol/L
Potassium	3.6-5.1 mmol/L	3.6-5.1 mmol/L
Sodium	128-145 mmol/L	128-145 mmol/L
Total Carbon Dioxide	18-33 mmol/L	18-33 mmol/L
Blood Urea Nitrogen (BUN)	7-22 mg/dL	2.5-7.9 mmol urea/L

* In the United States, the SI units are used for reporting lactate. To convert values in "mmol/L" to "mg/dL" multiply the value in mmol/L by the factor 9.009.
** There is no observed difference between the concentration of Phosphorus measured in heparinized whole blood and heparinized plasma. However, a small increase (0.3 mg/dL) was observed in serum when compared to heparinized whole blood and heparinized plasma. This increase is consistent with the difference between Phosphorus in serum and plasma as described in the literature.[64, 65, 66, 67]

12. Performance Characteristics

Linearity
The chemistry for each analyte is linear over the dynamic range listed below when the Piccolo Xpress chemistry analyzer is operated according to the recommended procedure (refer to the Piccolo Xpress chemistry analyzer Operator's Manual).

Table 5: Piccolo Xpress chemistry analyzer Dynamic Ranges

Analyte	Common Units	SI Units
Albumin	1-6.5 g/dL	10-65 g/L
Calcium	4.0-16.0 mg/dL	1.0-4.0 mmol/L
Chloride	80-135 mmol/L	80-135 mmol/L
Creatinine	0.2-20 mg/dL	18-1768 µmol/L

Glucose	10-700 mg/dL	0.6-38.9 mmol/L
Lactate	2.7 – 90.0 mg/dL	0.30 – 9.99 mmol/L
Magnesium	0.1 – 8.0 mg/dL	0.04 – 3.3 mmol/L
Phosphorus	0.2-20 mg/dL	0.06-6.5 mmol/L
Potassium	1.5-8.5 mmol/L	1.5-8.5 mmol/L
Sodium	110-170 mmol/L	110-170 mmol/L
Total Carbon Dioxide	5-40 mmol/L	5-40 mmol/L
Blood Urea Nitrogen (BUN)	2-180 mg/dL	0.7-64.3 mmol urea/L

If the analyte concentration is above the measuring range (dynamic range), but less than the system range, the printout will indicate a ">" sign at the upper limit and an asterisk after the number, e.g. ALT >2000* U/L. If lower than the dynamic range, a "<" will be printed with an asterisk, e.g. ALT <5* U/L. For values that are grossly beyond the measurement range (system range), "~~~" will be printed instead of a result. Any time "~~~" appears on a printout, collect a new sample and rerun the test. If results for the second sample are suppressed again, please call Abaxis Customer Service.

Sensitivity (Limits of Detection)

The lower limit of the reportable (dynamic) range for each analyte is: albumin 1 g/dL (10 g/L); calcium 4.0 mg/dL (1.0 mmol/L); chloride 80 mmol/L; creatinine 0.2 mg/dL (18 µmol/L); glucose 10 mg/dL (0.56 mmol/L); lactate 0.07 mmol/L (0.6 mg/dL)); magnesium 0.1 mg/dL (0.04 mmol/L); phosphorus 0.2 mg/dL (0.06 mmol/L); potassium 1.5 mmol/L; sodium 110 mmol/L; total carbon dioxide 5 mmol/L; and blood urea nitrogen 2.0 mg/dL (0.7 mmol urea/L).

Precision

Precision studies were conducted using CLSI (formerly NCCLS) EP5-A and CLSI EP5-A2 guidelines[68,69], with modifications based on CLSI (formerly NCCLS) EP18-A and CLSI EP18-A2 for unit-use devices. [70, 71] Results for within-run and total precision were determined by testing levels of commercially available control materials and in the case of potassium two levels of plasma pools. The studies made use of multiple instruments. Precision for albumin, calcium, creatinine, glucose, sodium and urea nitrogen was performed at one site; potassium and total carbon dioxide testing was performed at two sites over 20 days; and chloride, magnesium and phosphorus testing was conducted at two sites over a period of five days. Precision for lactate was performed on controls 1, 2, and 3 at two sites using two lots of disc over a period of five days; on plasma pools at Abaxis over a period of five days; and on whole blood at three point-of-care sites by two operators, each testing 10 replicates on 10 Abaxis analyzers. Potassium testing was conducted at a CLIA waived site making use of three analyzers, one lot of reagent discs, and two operators over five days.

Results of the precision studies are shown in Table 6.

Table 6: Precision

Analyte	Sample Size	Within-Run	Total
Albumin (g/dL)	N=80		
Control 1			
Mean		5.6	5.6
SD		0.09	0.11
%CV		1.7	2.1
Control 2			
Mean		3.7	3.7
SD		0.07	0.11
%CV		2.0	2.9
Calcium (mg/dL)	N=80		
Control 1			
Mean		8.6	8.6
SD		0.21	0.25
%CV		2.4	2.9
Control 2			
Mean		11.8	11.8
SD		0.39	0.40
%CV		3.3	3.4
Chloride (mmol/L)	N=160		
Control 1			
Mean		97.8	97.8
SD		1.63	1.74

%CV		1.7	1.7
Control 2			
Mean		113.6	113.6
SD		1.97	2.22
%CV		1.7	2.0
Creatinine (mg/dL)	N=80		
Control 1			
Mean		1.1	1.1
SD		0.14	0.14
%CV		12.5	13.1
Control 2			
Mean		5.2	5.2
SD		0.23	0.27
%CV		4.4	5.2
Glucose (mg/dL)			
Control 1	N=80		
Mean		66	66
SD		0.76	1.03
%CV		1.1	1.6
Control 2			
Mean		278	278
SD		2.47	3.84
%CV	N =80	0.9	1.4
Lactate (mmol/L)			
Control 1			
Mean		1.62	1.62
SD		0.03	0.04
%CV	N =80	1.8	2.2
Control 2			
Mean		3.63	3.63
SD		0.05	0.08
%CV	N =80	1.5	2.3
Control 3			
Mean		6.99	6.99
SD		0.18	0.36
%CV	N =40	2.6	5.2
Pooled Plasma 1			
Mean		0.86	0.86
SD		0.02	0.02
%CV	N =40	1.9	1.9
Pooled Plasma 2			
Mean		6.22	6.22
SD		0.20	0.20
%CV		3.2	3.2

	Operator 1 (N = 10)	**Operator 2 (N = 10)**	**Combined (N = 20)**
Whole Blood 1 - Site 1*			
Mean	1.52	1.50	1.51
SD	0.03	0.02	0.03
%CV	2.1	1.7	2.0
Whole Blood 2 - Site 1			
Mean	0.72	0.70	0.71
SD	0.02	0.02	0.02
%CV	2.2	2.5	2.7
Whole Blood 3 - Site 1			

Mean		4.67	4.59	4.63
SD		0.17	0.10	0.15
%CV		3.7	2.3	3.1
Whole Blood 4 - Site 1				
Mean		4.13	4.17	4.15
SD		0.12	0.17	0.15
%CV		3.0	4.1	3.5
Whole Blood 5 - Site 2*				
Mean		1.00	1.02	1.01
SD		0.03	0.02	0.03
%CV		2.8	1.7	2.5
Whole Blood 6 - Site 2				
Mean		1.09	1.12	1.10
SD		0.03	0.03	0.03
%CV		2.6	2.3	2.7
Whole Blood 7 - Site 2				
Mean		6.18	6.38	6.28
SD		0.21	0.25	0.24
%CV		3.4	3.9	3.9
Whole Blood 8 - Site 2	N =20			
Mean		5.88	5.90	5.89
SD		0.32	0.17	0.25
%CV		5.5	2.9	4.2
Whole Blood 9 – Site 3*	N =20			
Mean		0.88	0.88	0.88
SD		0.03	0.04	0.03
%CV		3.3	4.3	3.8
Whole Blood 10 – Site 3	N =20			
Mean		1.09	1.06	1.08
SD		0.03	0.03	0.03
%CV		3.0	2.9	3.2
Whole Blood 11 – Site 3	N =20			
Mean		5.93	5.86	5.89
SD		0.24	0.14	0.20
%CV		4.1	2.5	3.3
Whole Blood 12 – Site 3	N =20			
Mean		7.76	7.76	7.76
SD		0.28	0.28	0.27
%CV		3.7	3.5	3.5

* Whole Blood testing performed at three point-of-care sites by two operators with 10 repetitions per operator. Two normal samples and two elevated samples were tested by each operator.

Magnesium (mg/dL)	N = 80		
Control 1			
Mean		1.9	1.9
SD		0.03	0.06
%CV		1.7	3.4
Control 2			
Mean		3.9	3.9
SD		0.04	0.10
%CV		1.0	2.6
Phosphorus (mg/dL)	N = 80		
Control 1			
Mean		3.1	3.1

SD		0.12	0.14
%CV		3.7	4.7
Control 2			
Mean		7.3	7.3
SD		0.09	0.15
%CV		1.3	2.0
Potassium (mmol/L)	N = 150		
Control 1			
Mean		3.2	3.2
SD		0.09	0.11
%CV		2.8	3.3
Control 2	N = 149		
Mean		6.2	6.2
SD		0.09	0.10
%CV		1.4	1.7
Plasma Pool 1	N = 150		
Mean		3.2	3.2
SD		0.07	0.09
CV		2.3	2.9
Plasma Pool 2	N = 150		
Mean		5.4	5.4
SD		0.09	0.10
CV		1.6	1.9
Sodium (mmol/L)	N = 80		
Control 1			
Mean		143.5	143.5
SD		2.28	2.28
%CV		1.6	1.6
Control 2			
Mean		120.0	120.0
SD		2.13	2.13
%CV		1.8	1.8
Total Carbon Dioxide (mmol/L)	N=120		
Control 1			
Mean		21.4	21.4
SD		2.29	2.29
%CV		10.7	10.7
Control 2			
Mean		10.5	10.5
SD		0.90	0.90
%CV		8.6	8.6
Urea Nitrogen (mg/dL)	N = 80		
Control 1			
Mean		19	19
SD		0.35	0.40
%CV		1.9	2.1
Control 2			
Mean		65	65
SD		1.06	1.18
%CV		1.6	1.8

Whole Blood Precision for Potassium

Whole blood precision was tested at a CLIA waived site by two CLIA waiver operators. The study used four Piccolo Xpress Analyzers with 16 replicates per sample for four (4) fresh, lithium heparin whole blood samples.

Table 7: Precision Whole Blood Precision for Potassium

Potassium (mmol/L)	Sample Size	Within-Run	Total
Whole Blood 1	N = 16		
Mean		3.9	3.9
SD		0.06	0.11
CV		1.6	2.8
Whole Blood 2	N = 16		
Mean		4.0	4.0
SD		0.11	0.14
CV		2.9	3.4
Whole Blood 3	N = 16		
Mean		4.0	4.0
SD		0.11	0.15
CV		2.8	3.9
Whole Blood 4	N = 16		
Mean		4.0	4.0
SD		0.11	0.13
CV		2.7	3.4

Correlation

For lactate, lithium heparinized whole blood samples were collected and assayed on the Piccolo Xpress chemistry analyzer and by a comparative method(s). For all other methods, serum samples were collected and tested.

Table 8: Correlation of Piccolo Xpress chemistry analyzer with Comparative Method

	Correlation Coefficient	Slope	Intercept	SEE	N	Sample Range	Comparative Method
Albumin (g/dL)	0.854	1.001	- 0.3	0.22	261	1.1-5.3	Paramax®
	0.896	0.877	- 0.1	0.21	100	1.5-5.0	Beckman
Calcium (mg/dL)	0.991*	0.990	- 0.4	0.17	25	5.2-11.9	Paramax
	0.673	0.742	1.8	0.22	81	8.1-9.9	Beckman
Chloride (mmol/L)	0.978	0.982	- 1.1	1.84	120	71-118	Vitros® 950
Creatinine (mg/dL)	0.993	0.926	0.0	0.15	260	0.4–14.7	Paramax®
	0.987	0.866	0.1	0.16	107	0.4-7.5	Beckman
Glucose (mg/dL)	0.987	1.009	- 2.8	3.89	251	72-422	Paramax®
	0.997	0.943	1.2	4.69	91	56-646	Beckman
Lactate (mmol/L)	0.996	1.02	0.08	0.19	126	0.30–9.88	i-STAT
Magnesium (mg/dL)	0.992	0.990	0.0	0.16	44	0.8 - 6.8	**Inductively Coupled Plasma-Atomic** Optical Emission Spectroscopy (ICP-OES)
Phosphorus (mg/dL)	0.993	1.017	- 0.2	0.236	90	0.8 – 11.7	Vitros® 950
Potassium (mmol/L) **Whole Blood (waived laboratory)**	0.984	0.99	0.13	0.10	130	1.3 – 9.5	Siemens VISTA Plasma
Potassium (mmol/L) **Whole Blood (moderately complex laboratory**	0.984	0.98	0.12	0.18	178	1.5-8.6	Siemens VISTA Plasma

Table 8: Correlation of Piccolo Xpress chemistry analyzer with Comparative Method (continued)

	Correlation Coefficient	Slope	Intercept	SEE	N	Sample Range	Comparative Method
Potassium (mmol/L**)**	0.99	0.98	0.06	0.14	178	1.4-8.5	Siemens
Serum (moderately compex laboratory)							VISTA Serum
Sodium (mmol/L)	0.937	0.782	27.7	3.79	113	116 - 154	Radiometer KNA® 2
Total Carbon Dioxide (mmol/L)	0.947	0.903	2.4	0.84	60	6 – 39	Cobas® Fara
Blood Urea Nitrogen (mg/dL)	0.964	0.923	0.5	1.08	251	6-52	Paramax
	0.983	0.946	0.0	0.66	92	6-38	Beckman

***** Serum samples from hospitalized patients provided a broader, and possibly more useful, sample range than did venous whole blood samples from out-patients. Correlation statistics for the Piccolo calcium test are from these serum samples.

It should be noted that serum will typically give higher results for K+ compared to whole blood or plasma for physiological reasons. The variation can range from approximately 0.2 to 0.9mmol/L ans is dependent on a number of factors. The primary effect is dependent upon the number of blood cells present in the patient sample.[74]

13. Symbols

Use By

Catalog Number

Batch Code

In Vitro Diagnostic Medical Device

Consult Instructions for Use

Manufacturer

Do Not Reuse

X Number of Test Devices in Kit

Manufacturing Sequence

Serial Number

Authorized Representative In the European Community

Temperature Limitation

Caution

PN:
Part Number

CE

denotes conformity to specified European directives

14. Bibliography

1. Howe PE. The use of sodium sulfate as the globulin precipitant in the determination of proteins in blood. J Biol Chem 1921; 49:93-07.
2. Howe PE. The determination of proteins in blood - a micro method. J Biol Chem 1921; 49:109-13.
3. Wolfson WQ, et al. A rapid procedure for the estimation of total protein, true albumin, total globulin, alpha globulin, beta globulin, and gamma globulin in 10 mL of serum. Am J Clin Pathol 1948; 18:723-30.
4. Saifer A, Gerstenfeld S, Vacsler F. Photometric microdetermination of total serum globulins by means of a tryptophan reaction. Clin Chem 1961; 7:626-36.
5. Saifer A, Marven T. The photometric microdetermination of serum total globulins with a tryptophan reaction: a modified procedure. Clin Chem 1966; 12:414-17.
6. Gendler SM. Albumin. *In*: Clinical Chemistry: Theory, Analysis, and Correlation, 2nd Ed. Kaplan LA, Pesce AJ, eds. St Louis: The C.V. Mosby Company. 1989:1029-33.
7. Webster D, Bignell AHC, Attwood EC. An assessment of the suitability of bromocresol green for the determination of serum albumin. Clin Chim Acta 1974; 53:101-8.
8. Louderback A, Mealey EH, Taylor NA. A new dye-binding technic using bromcresol purple for determination of albumin in serum. Clin Chem 1968; 14:793-794. (Abstract)
9. Pinnell AE, Northam BE. New automated dye-binding method for serum albumin determination with bromcresol purple. Clin Chem 1978; 24:80-86.
10. Kramer B, et al. A simple technique for the determination of calcium and magnesium in small amounts of serum. J Biol Chem 1921; 47:475-481.
11. Clark EP, et al. A study of the Tisdall method for the determination of blood serum calcium with suggested modification. J Biol Chem 1925; 63:461-464.
12. Katzman E, et al. The determination of serum calcium by titration with ceric sulfate. J Biol Chem.1937; 118:539-544.
13. Cali JP, et al. A reference method for the determination of total calcium in serum. *In*: Selected Methods of Clinical Chemistry, Vol 8. GR Cooper, ed. Washington, DC: American Association for Clinical Chemistry. 1997:3-8.
14. Kessler G, M Wolfman. An automated procedure for the simultaneous determination of calcium and phosphorus. Clin Chem 1964; 10:686-703.
15. Michaylova V, et al. Photometric determination of micro amounts of calcium with arsenazo III. Anal Chim Acta 1971; 53:194-198.
16. Scarpa A, et al. Metallochromic indicators of ionized calcium. Ann NY Acad Sci 1978; 307:86-112.
17. Ono T, et al. A new enzymatic assay of chloride in serum. Clin Chem 1988; 34:552-3.
18. Knoll VE, et al. Spezifische kreatininbestimmung im serum. Z Klin Chem Klin Biochem 1970; 8:582-587.
19. Haeckel R, et al. Simplified determinations of the "true" creatinine concentration in serum and urine. J Clin Chem Clin Biochem 1980; 18:385-394.
20. Moss GA, et al. Kinetic enzymatic method for determining serum creatinine. Clin Chem 1975; 21:1422-1426.
21. Jaynes PK, et al. An enzymatic, reaction-rate assay for serum creatinine with a centrifugal analyzer. Clin Chem 1982; 28:114-117.
22. Fossati P, et al. Enzymatic creatinine assay: a new colorimetric method based on hydrogen peroxide measurement. Clin Chem 1983; 29:1494-1496.
23. Whelton A, et al. Nitrogen metabolites and renal function. In: Tietz Textbook of Clinical Chemistry, 2nd Ed. Burtis CA, Ashwood ER, eds. Philadelphia: W.B. Saunders Company. 1994; 1513-1575.
24. National Kidney Disease Education Program (NKDEP). Creatinine Standardization. http://www.nkdep.nih.gov/labprofessionals/creatinine_standardization.htm (Accessed September 2010).
25. National Kidney Disease Education Program (NKDEP) Creatinine Standardization Recommendations. IVD Manufacturers. http://www.nkdep.nih.gov/labprofessionals/IVD_Manufacturers.htm (Accessed September 2010).
26. National Kidney Foundation. GFR Calculator. http://www.kidney.org/professionals/kls/gfr_calculator.cfm (Accessed September 2010).
27. Folin O, et al. A system of blood analysis. J Biol Chem 1919; 38:81-110.
28. Somogyi M. A reagent for the copper-iodometric determination of very small amounts of sugar. J Biol Chem 1937; 117:771-776.
29. Nelson N, et al. A photometric adaptation of the Somogyi method for the determination of glucose. J Biol Chem 1944; 153: 375-380.
30. Kaplan LA. Glucose. *In*: Clinical Chemistry: Theory, Analysis, and Correlation, 2nd Ed. Kaplan LA, Pesce AJ, eds. St Louis: The C.V. Mosby Company. 1989: 850-856.
31. Tabata M, et al. Direct Spectrophotometry of magnesium in serum after reaction with hexokinase and glucose-6-phophate dehydrogenase. Clin Chem 1985; 31: 703-705.
32. Schultz DW, Passonneau JV, Lowry OH. An enzymic method for the measurement of inorganic phosphate determination. Anal BioChem 1967; 19:300-314.

14. Bibliography (continued)

33. Tedokon M, et al. Enzymatic assay of inorganic phosphate with use of sucrose phosphorylase and phosphoglucomutase. Clin Chem 1992; 38:512-515.
34. Berry MN, et al. Enzymatic determination of potassium in serum. Clin Chem. 1989; 35:817-820.
35. Van Pelt J. Enzymatic determination of sodium, potassium and chloride in serum compared with determination by flame photometry, coulometry and ion selective electrodes. Clin Chem. 1994; 40:846-847.
36. Hubl W, et al. Enzymatic determination of sodium, potassium and chloride in abnormal (hemolyzed, icteric, lipemic, paraproteinemic, or uremic) serum samples compared with indirect determination with ion selective electrodes. Clin Chem 1994; 40:1528-1531.
37. Helgerson RC, et al. Host-guest complexation. 50. Potassium and sodium ion-selective chromogenic ionophores. J Amer Chem Soc 1989; 111:6339-6350.
38. Kumar A, et al. Chromogenic ionophere-based methods for spectrophotometric assay of sodium and potassium in serum and plasma. Clin Chem 1988; 34:1709-1712.
39. Berry MN, et al. Enzymatic determination of sodium in serum. Clin Chem 1988; 34:2295-2298.
40. Skeggs LT Jr. An automatic method for the determination of carbon dioxide in blood plasma. Am J Clin Pathol 1960; 33:181-185.
41. Korzun WJ, Miller WG. Carbon dioxide. *In*: Clinical Chemistry: Theory, Analysis, and Correlation, 2nd ed. Kaplan LA, Pesce AJ, eds. St Louis: The C.V. Mosby Company. 1989: 869-872.
42. Fales FW. Urea in serum, direct diacetyl monoxime method. *In*: Selected Methods of Clinical Chemistry, Vol 9. Faulkner WR and Meites S, eds. Washington, DC: AACC Press. 1982: 365-373.
43. Van Slyke, et al. A permanent preparation of urease, and its use in the determination of urea. J Biol Chem 1914; 19:211-228.
44. Fawcett JK, et al. A rapid and precise method for the determination of urea. J Clin Pathol 1960; 13:156-159.
45. Chaney, et al. Urea and ammonia determinations. Clin Chem 1962; 8:130-132.
46. Talke H, et al. Enzymatische harnstoffbestimmung in blut and serum im optischen test nach Warburg. Klin Wochensch 1965; 43:174-175.
47. Hallett, et al. Reduced nicotinamide adenine dinucleotide-coupled reaction for emergency blood urea estimation. Clin Chim Acta 1971; 35:33-37.
48. Patton, et al. Spectrophotometric and kinetics investigation of the Berthelot reaction for the determination of ammonia. Anal Chem 1977; 49:464-469.
49. Sampson EJ, et al. A coupled-enzyme equilibrium method for the measuring urea in serum: optimization and evaluation of the AACC study group on urea candidate reference method. Clin Chem 1980; 26:816-826.
50. Clinical and Laboratory Standards Institute (CLSI (formerly NCCLS)). Physician's office laboratory guidelines; Tentative Guideline – Second Edition. NCCLS Document POL1-T2. Wayne, PA: NCCLS, 1992.
51. Scott, M.G. Electrolytes and blood gases. *In:* Tietz Textbook of Clinical Chemistry, 3rd Ed. Burtis CA, Ashwood ER, eds. Philadelphia: WB Saunders Company. 1999: 1058-1059.
52. D.B. Sacks, Carbohydrates, in Tietz Textbook of Clinical Chemistry, 3rd Ed. Burtis CA, Ashwood ER, eds. Philadelphia: WB Saunders Company. 1999.
53. Clinical and Laboratory Standards Institute (*CLSI)*. Bl*oo*d *Ga*s and pH Analysis *a*nd related measurements; Ap*pr*oved Guidelines – Second Edition. Document C46-A2 Vol.29 No. 8. February 2009
54. CLSI GP44-A4 2010:Procedures for the Handling and Processing of Blood Specimens for Common Laboratory Tests; Approved Guideline—Fourth Edition.
55. CLSI GP41 7ED 2017: Collection of Diagnostic Venous Blood Specimens.
56. Overfield CV, et al. Glycolysis: a re-evaluation of the effect on blood glucose. Clin Chim Acta 1972; 39: 35-40.
57. Rehak NN, Chiang BT. Storage of whole blood: effect of temperature on the measured concentration of analytes in serum. Clin Chem 1988; 34-2111-4.
58. Scott, M.G. Electrolytes and blood gases. *In:* Tietz Textbook of Clinical Chemistry, 3rd ed. Burtis CA, Ashwood ER, eds. Philadelphia: WB Saunders Company. 1999: 1065-6.
59. Clinical and Laboratory Standards Institute. Interference testing in clinical chemistry; Proposed Guideline. CLSI Publication EP7-P. Wayne, PA: CLSI, 1986.
60. Clinical and Laboratory Standards Institute. Interference testing in clinical chemistry; Approved Guideline– Second Edition. CLSI Document EP7-A2. Wayne, PA: CLSI, 2005.
61. Young DS. Effects of drugs on clinical laboratory tests, 3rd Ed. Washington, DC: AACC Press, 1990.
62. National Committee for Clinical Laboratory Standards. How to define and determine reference intervals in the clinical laboratory, Approved Guidelines – Second Edition. NCCLS Document C28-A2. Wayne, PA: NCCLS, 2000.
63. Clinical and Laboratory Standards Institute. How to define and determine reference intervals in the clinical laboratory, Approved Guidelines – Third Edition. CLSI Document C28-A3c. Wayne, PA: CLSI, 2010.
64. Lum G, Gambino S. Serum vs plasma determinations in routine chemistry. Clin Chem 1972; 18(7);Abstr 134;710.

14. Bibliography (continued)

65. Lum G, Gambino S. A comparison of serum vs hepirinized plasma for routine chemistry tests. Am J Clin Pathol 1974: 61(1);108-13.
66. Carothers J, Kurtz N, Lehmann J, Jr. Error introduced by specimen handling before determination of inorganic phosphate concentrations in plasma and serum. Clin Chem 1976: 22(11);1909-12.
67. Ladenson J, et al. Serum vs heparinized plasma for routine chemistry tests. Am J Clin Path 1974: 62(4);545-52.
68. Clinical and Laboratory Standards Institute. Evaluation of precision performance of clinical chemistry devices; Approved Guideline. CLSI Document EP5-A. Wayne, PA: CLSI, 1999.
69. Clinical and Laboratory Standards Institute. Evaluation of precision performance of clinical chemistry devices; Approved Guideline– Second Edition. CLSI Document EP5-A2. Wayne, PA: CLSI, 2004.
70. Clinical and Laboratory Standards Institute. Quality management for unit-use testing; Proposed Guideline. CLSI Document EP18-A. Wayne, PA: CLSI, 2002.
71. Clinical and Laboratory Standards Institute. Quality management for unit-use testing; Approved Guideline– Second Edition. CLSI Document EP18-A2. Wayne, PA: CLSI, 2009
72. Clinical and Laboratory Standards Institute. Method comparison and bias estimation using patient samples; Approved Guideline. CLSI Document EP9-A. Wayne, PA: CLSI, 1995.
73. Clinical and Laboratory Standards Institute. Method comparison and bias estimation using patient samples; Approved Guideline– Second Edition. CLSI Document EP09-A2-IR. Wayne, PA: CLSI, 2010.
74. Hartland AJ. Hartland JH, Neary RN. Serum Potassium Is Unreliable as an Estimate of in Vivo Plasma Potassium. Clin Chem 1999;45:1091-1092.