Piccolo® AmLyte 13

CE 2797

For In Vitro Diagnostic Use and Healthcare Professional Use
Only Customer and Technical Service: 1-800-822-2947
Customers outside the US: +49 6155 780 210
AB-PiccoloTechSupport@zoetis.com

Abaxis, Inc.
3240 Whipple Rd.
Union City, CA 94587
USA

CH REP
Tao of Excellence GmbH
Vorstadt 26,
8200 Schaffhausen
Switzerland

EC REP
ABAXIS Europe GmbH
Bunsenstr. 9-11
64347 Griesheim
Germany

1. Intended Use

The Piccolo® AmLyte 13 , used with the Piccolo Xpress® chemistry analyzer, is intended to be used for the *in vitro* quantitative determination of alanine aminotransferase (ALT), albumin, amylase, aspartate aminotransferase (AST), calcium, c-reactive protein (CRP), creatine kinase, creatinine, glucose, potassium, sodium, total bilirubin and blood urea nitrogen (BUN) in lithium heparinized whole blood, lithium heparinized plasma, or serum in a clinical laboratory setting or point-of-care location/ near-patient testing. The Abaxis CRP method is not intended for high sensitivity CRP measurement.

2. Summary and Explanation of Tests

The Piccolo AmLyte 13 and the Piccolo Xpress chemistry analyzer comprise an *in vitro* diagnostic system that aids the physician in diagnosing the following disorders in adults:

Alanine aminotransferase (ALT):	Liver diseases, for example, viral hepatitis and cirrhosis.
Albumin:	Liver and kidney diseases.
Amylase:	Pancreatitis.
Aspartate aminotransferase (AST):	Liver disease, for example, hepatitis and viral jaundice, shock liver.
Calcium:	General metabolic disease, for example, Parathyroid, bone and chronic renal diseases; tetany.
C-Reactive Protein (CRP):	General inflammation, for example, Infection, tissue injury, and inflammatory disorders.
Creatine Kinase:	Muscle disease, for example, progressive muscular dystrophy, dermatomyositis, rhabdomyolysis due to drug ingestion, hyperosmolality, autoimmune disease, delirium tremens, convulsions, Crush syndrome, hypothyroidism, surgery, severe exercise, intramuscular injection.
Creatinine:	Renal disease and renal dialysis.
Glucose:	Carbohydrate metabolism disorders, for example, adult and juvenile diabetes mellitus and hypoglycemia.
Potassium:	Renal, endocrine, and metabolic disease as well as iatrogenic causes, for example, Renal glomerular or tubular disease, adrenocortical insufficiency, diabetic ketacidosis, 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 sense of thirst, skin losses, burns, sweating, hyperaldosteronism, CNS disorders, dilutional, depletional and delusional hyponatremia and syndrome of inappropriate ADH secretion.
Total bilirubin:	Liver disorders, for example, hepatitis and biliary obstruction; jaundice.
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

Alanine Aminotransferase (ALT)
Alanine aminotransferase (ALT) has been measured by three methods. Two of these methods—the colorimetric dinitrophenylhydrazine coupling technique [1,2] and the fluorescent enzymatic assay—are rarely used.[3] An enzymatic method based on the work of Wróblewski and LaDue[4] is the most common technique for determining ALT concentrations in serum. A modified Wróblewski and LaDue procedure has been proposed as the recommended procedure of the International Federation of Clinical Chemistry (IFCC).[5]

The method developed for use on the Piccolo analyzers is a modification of the IFCC-recommended procedure. In this reaction, ALT catalyzes the transfer of an amino group from L-alanine to α-ketoglutarate to form L-glutamate and pyruvate. Lactate dehydrogenase catalyzes the conversion of pyruvate to lactate. Concomitantly, NADH is oxidized to NAD^+, as illustrated in the following reaction scheme.

$$\text{L-Alanine} + \alpha\text{-Ketoglutarate} \xrightarrow{\text{ALT}} \text{L-Glutamate} + \text{Pyruvate}$$

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

The rate of change of the 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 ALT present in the sample.

Albumin (ALB)
Early methods used to measure albumin include fractionation techniques[6,7,8] and tryptophan content of globulins.[9,10] These methods are unwieldy to perform and do not have a high specificity. Two immunochemical techniques are considered as reference methods, but are expensive and time consuming.[11] Dye binding techniques are the most frequently used methods for measuring albumin. Bromcresol 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.[12] Bromcresol purple (BCP) is the most specific of the dyes in use.[13,14]

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

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

Bound albumin is proportional to the concentration of albumin in the sample. This is an endpoint reaction that is measured as the difference in absorbance between 600 nm and 550 nm.

Amylase (AMY)
About 200 different tests have been developed to measure amylase. Most procedures use a buffered polysaccharide solution but employ different detection techniques. Viscosimetric methods are lacking in precision and accuracy[15], while turbidimetric and iodometric methods are difficult to standardize.[16,17] Commonly used are saccharogenic and chromolytic methods. The "classic" amylase measurement technique is a saccharogenic method [18], but is difficult and time-consuming.[19] Chromolytic methods using *p*-nitrophenyl-glycosides as substrates have been recently developed.[20] These assays have a higher specificity for pancreatic amylase than for salivary amylase and are easily monitored.[20]

In the Piccolo method, the substrate, 2-chloro-*p*-nitrophenyl-α-D-maltotrioside (CNPG3), reacts with α-amylase in the patient sample, releasing 2-chloro-*p*-nitrophenol (CNP). The release of CNP creates a change in color.

$$\text{CNPG3} \xrightarrow{\alpha\text{-Amylase}} \text{CNP} + \text{D-Maltotrioside}$$

The reaction is measured bichromatically at 405 nm and 500 nm. The change in absorbance due to the formation of CNP is directly proportional to α-amylase activity in the sample.

Aspartate Aminotransferase (AST)
The aspartate aminotransferase (AST) test is based on the Karmen rate method[21] as modified by Bergmeyer.[22] The current International Federation of Clinical Chemistry (IFCC) reference method utilizes the Karmen/Bergmeyer technique of coupling malate dehydrogenase (MDH) and reduced nicotinamide dinucleotide (NADH) in the detection of AST in serum.[22,23] Lactate dehydrogenase (LDH) is added to the reaction to decrease interference caused by endogenous pyruvate.

AST catalyzes the reaction of L-aspartate and α-ketoglutarate into oxaloacetate and L-glutamate. Oxaloacetate is converted to malate and NADH is oxidized to NAD^+ by the catalyst MDH.

$$\text{L-aspartate} + \alpha\text{-ketoglutarate} \xrightarrow{\text{AST}} \text{Oxaloacetate} + \text{L-glutamate}$$

$$\text{Oxaloacetate} + \text{NADH} \xrightarrow{\text{MDH}} \text{Malate} + NAD^+$$

The rate of absorbance change at 340 nm/405 nm caused by the conversion of NADH to NAD^+ is directly proportional to the amount of AST present in the sample.

Calcium (CA)

The first methods used to analyze calcium involved precipitating calcium with an excess of anions.[24,25,26] 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.[27] Spectrophotometric methods using either *o*-cresolphthalein complexone or arsenazo III metallochromic indicators are most commonly used.[28,29,30] 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.

C-Reactive Protein (CRP)

Original assays for quantifying CRP were primarily for research and based on ELISA methodology. [31] More recently, latex-enhanced immunonephelometric methods have been used.[32] However; this requires a nephelometer to measure the light scattering. Now, several automated immunoturbidimetric and immunoluminometric assays have been developed that can be run on conventional clinical chemistry analyzers.[33]

The method used by Abaxis is an enhanced latex-agglutination turbidimetric immunoassay. Sample is mixed with a suspension of mouse anti-human CRP monoclonal antibody that is bound to latex. CRP in the sample binds to the antibody-latex particles and agglutinates creating turbidity. Light scattering from the turbidity is used as a measure of CRP. Turbidity is measured as a change in absorbance at 630 nm. This absorbance change is directly proportional to the CRP in the sample.

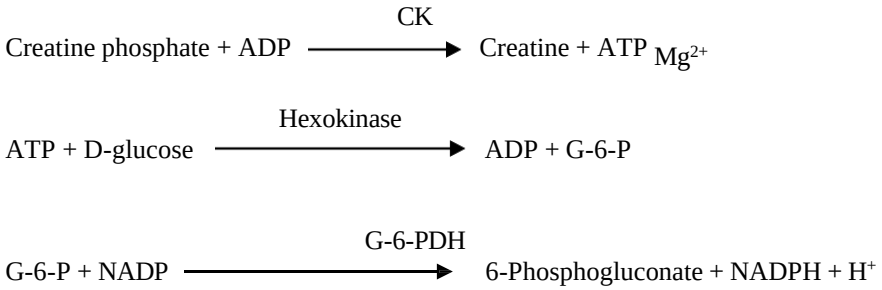
$$\text{Anti-CRP latex particles} + \text{CRP} \longrightarrow \text{Agglutinated CRP-Anti-CRP latex particles}$$

Creatine Kinase (CK)

Creatine kinase catalyzes the reversible phosphorylation of creatine by adenosine triphosphate (ATP). The phosphorylation reaction is favored by alkaline conditions (optimum at pH 9.0) and the dephosphorylation reaction is favored by acidic conditions (optimum at pH 6.5 at 37ºC). Early CK measurement methods were based on the "forward reaction" with creatine phosphate and adenosine diphosphate (ADP) as the products.[34,35,36] The sensitivity of these tests was shown to be low due to problems with interferences. The procedure of choice utilizes the "reverse reaction" coupled with a reaction to produce NADPH, which is directly related to CK levels.[37,38,39]

The CK measurement procedure used by Abaxis is a modified version of the International Federation of Clinical Chemistry (IFCC) method.[40] Key modifications are sample volume fraction, buffer, and temperature. N-acetyl cysteine (NAC) has been added to reactivate the CK.[41] Magnesium is used as a cofactor for both CK and hexokinase. EDTA has been added as a stabilizer for NAC and for the removal of various cations, such as calcium and iron, that inhibit CK. P^1, P^5-di (adenosine- 5')penta phosphate and adenosine monophosphate (AMP) have also been added to inhibit adenylate kinase, another skeletal muscle and erythrocyte enzyme that reacts with the substrates used to measure CK.

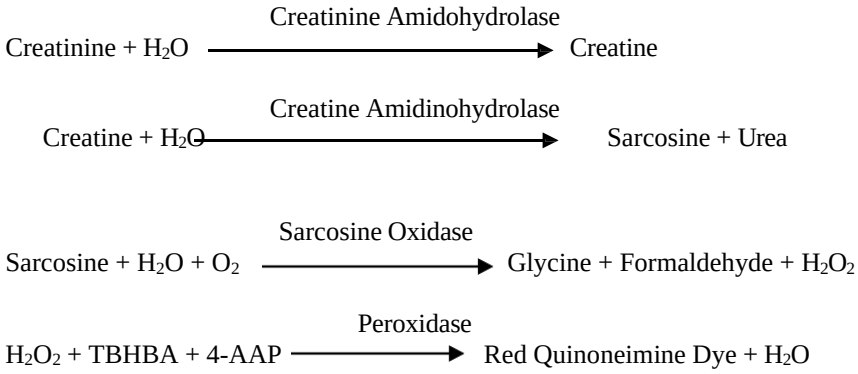
Creatine kinase catalyzes the formation of creatine and ATP from creatine phosphate and ADP at pH 6.7. With hexokinase as a catalyst, ATP reacts with D-glucose to form ADP and D-glucose-6-phosphate (G-6-P), which is reacted with nicotinamide adenine dinucleotide phosphate (NADP) in the presence of glucose-6-phosphate dehydrogenase (G-6-PDH) to produce G-6-P and NADPH.



The formation of NADPH is measured as a change in absorbance at 340 nm relative to 405 nm. This absorbance change is directly proportional to creatine kinase activity in the sample.

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.[42,43] Enzymatic methods have been developed that are more specific for creatinine than the various modifications of the Jaffe technique.[44,45,46] Methods using the enzyme creatinine amidohydrolase eliminate the problem of ammonium ion interference found in techniques using creatinine iminohydrolase.[47]



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.

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.

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

Glucose (GLU)

Measurements of glucose concentration were first performed using copper-reduction methods (such as Folin-Wu[48] and Somogyi-Nelson[49,50]). 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 AmLyte 13 is a modified version of the hexokinase method, which has been proposed as the basis of the glucose reference method.[51]

The reaction of glucose with adenosine triphosphate (ATP), catalyzed by hexokinase (HK), procedures 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.

$$\text{Glucose} + \text{ATP} \xrightarrow{\text{Hexokinase}} \text{G-6-P} + \text{ADP}$$

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

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.[52,53,54] Interference from sodium and ammonium ion are minimized with the addition of Kryptofix and glutamine synthetase respectively.[52]

In the coupled-enzyme reaction, pyruvate kinase (PK) dephosphorylates phosphoenolpyruvate (PEP) 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^+ is directly proportional to the amount of potassium in the sample.

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

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

Sodium
(NA^+)

Colorimetric and enzymatic methods have been developed that allow the measurement of sodium concentration on standard clinical chemistry instrumentation.[55,56,57] In the Abaxis enzymatic reaction, β-galactosidase is activated by the sodium in the sample. The activated enzyme catalyzes the reaction of o-nitrophenyl-β-D-galactopyranoside (ONPG) to o-nitrophenol and galactose.

$$\text{ONPG} \xrightarrow[\beta\text{-Galactosidase}]{Na^+} \text{o-Nitrophenol} + \text{Galactose}$$

Total Bilirubin (TBIL)
Total bilirubin levels have been typically measured by tests that employ diazotized sulfanilic acid.[58,59] A newer, more specific method has been developed using the enzyme bilirubin oxidase.[60,61,62] In addition to using the more specific total bilirubin test method, photodegradation of the analyte is minimized on the Piccolo analyzers because the sample can be tested immediately after collection.

In the enzyme procedure, bilirubin is oxidized by bilirubin oxidase into biliverdin.

$$\text{Bilirubin} + O_2 \xrightarrow{\text{Bilirubin Oxidase}} \text{Biliverdin} + H_2O$$

Bilirubin is quantitated as the difference in absorbance between 467 nm and 550 nm. The initial absorbance of this endpoint reaction is determined from the bilirubin blank cuvette and the final absorbance is obtained from the bilirubin test cuvette. The amount of bilirubin in the sample is proportional to the difference between the initial and final absorbance measurements.

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.[63] Indirect methods measure ammonia created from the urea; the use of the enzyme urease has increased the specificity of these tests.[64] The ammonia is quantitated by a variety of methods, including nesslerization (acid titration), the Berthelot technique.[65,66] and coupled enzymatic reactions.[67,68] Catalyzed Berthelot procedures, however, are erratic when measuring ammonia.[69] 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.[70]

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^+.

$$\text{Urea} + H_2O \xrightarrow{\text{Urease}} 2NH_3 + CO_2$$

$$NH_3 + \alpha\text{-Ketoglutarate} + \text{NADH} \xrightarrow{\text{GLDH}} \text{L-Glutamate} + H_2O + NAD^+$$

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 AmLyte 13 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 alanine aminotransferase (ALT), albumin (ALB), amylase (AMY), aspartate aminotransferase (AST), calcium (CA), c-reactive protein (CRP), creatine kinase (CK), glucose (GLU), potassium (K+), sodium (NA+), and blood urea nitrogen (BUN). A dedicated sample blank is included in the disc to calculate concentrations of creatinine (CRE), and total bilirubin (TBIL). Each disc also contains a diluent consisting of surfactants and preservatives.

Table 1: Reagents

Component	Quantity/Disc
2, 4, 6-Tribomo-3-hydroxybenzoic acid (TBHBA)	188 µg
2-Chloro-4-nitrophenyl-α-D-maltotrioside (CNPG3)	35.6 µg
4,7,13,16,21-Pentaoxa-1,10-diazabicyclo[8.8.5]trisocosane	139µg
4-Aminoantipyrine hydrochloride	0 µg
Adenosine-5'-diphosphate	35 µg
Adenosine-5'-monophosphate	33 µg
Adenosine 5'-diphosphate, trilithium salt	3.7 µg
Adenosine 5'-triphosphate, disodium salt	12 µg
4-Aminoantipyrine	5.4 µg
Anti-human CRP coated latex	268.8 µg
Anti-human CRP	0.5 µg
Arsenazo III, disodium salt	3.5 µg
Ascorbate oxidase	0.3 U
Bilirubin oxidase	0.1 U
Bromocresol purple, sodium salt	2.3 µg
Calcium acetate	15.2 µg
Creatine amidinohydrolase	3 U
Creatine phosphate	122 µg
Creatinine amidohydrolase	1 U
3,5-Dichloro-2-hydroxybenzenesulfonic acid , Sodium salt (DHBSA)	34.8 µg
Ethylene glyco-bis(b-aminoethyl ether)-N,N,N',N'-tetraacetic acid (EGTA)	7 µg
Ethylenediaminetetraacetic acid (EDTA)	7.4 µg
Ethylenediaminetetraacetic acid (EDTA), disodium salt	44 µg
Ethylenediaminetetraacetic acid (EDTA), Tetrasodium Salt	215 µg
Glucose	58 µg
Glucose-6-phosphate dehydrogenase (G6PDH)	0.1 U
Glutamate dehydrogenase	0 U
Glutamine synthetase	0.2 U
Hexokinase	0.2 U
Imidazole	83 µg

Table 1: Reagents (continued)

Component	Quantity/Disc
Lactate dehydrogenase	0 U
Lactate dehydrogenase (LDH)	0.1 U
L-Aspartic acid	426 µg
L-glutamic acid	9 µg
Magnesium acetate, tetrahydrate	75 µg
Magnesium sulfate, heptahydrate	122 µg
Malate dehydrogenase	0 U
Manganese chloride	10 µg
N-Acetyl cysteine	68µg
o-Nitrophenyl-ß-D galactopyranoside (ONPG)	44µg
P1, P5di(adenosine-5') pentaphosphate	0.2 µg
Peroxidase	1 U
Phosphoenol pyruvate	14 µg
Potassium ferrocyanide	0.6 µg
Pyruvate kinase	0.01 U
Sarcosine oxidase	1 U
Sodium Cholate	58 µg
Sodium lauryl sulfate	145 µg
Sulfhydryl blocked BSA	420 µg
ß-Nicotinamide adenine dinucleotide, (NAD)	20 µg
ß-Nicotinamide adenine dinucleotide, reduced (NADH)	32 µg
b-Nicotinamide adenine dinucleotide phosphate (NADP+), sodium salt	104 µg
Urease	0.05 U
Uricase	0 U
α-Ketoglutaric acid	96 µg
alpha-Ketoglutaratic acid, disodium salt	37 µg
β-Galactosidase	0.009 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 can not 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.[39] 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 sealed in their foil pouches to remain at room temperature at 20-25°C (68-77°F) longer than 48 hours prior to use. Open the sealed foil pouch, remove the disc and 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 lithium heparinized whole blood, lithium heparinized plasma, serum 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 potassium leakage.[71]

- Whole blood venipuncture samples should be run within 60 minutes of collection.[72, 84] **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.[73]

- Refrigerating whole blood samples can cause significant changes in concentration of **aspartate aminotransferase**, **creatinine** and **glucose**.[74] The sample may be separated into plasma or serum and stored in capped sample tubes at 2-8 °C (36-46 °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.

- **Total bilirubin** results may be adversely affected by photodegradation.[75] Whole blood samples not run immediately should be stored in the dark for no longer than 60 minutes. If the sample cannot be analyzed within that period, it should be separated into plasma or serum and stored in a capped sample tube in the dark at 2-8°C (36-46°F)..)..[76]

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

8. Procedure

Materials Provided
- One Piccolo AmLyte 13 PN: 400-1041 (a box of discs PN 400-0041)

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 32 °C (59-90 °F). The analysis time for each Piccolo AmLyte 13 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 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 AmLyte 13 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
ALT	IFCC	Colorimetric
ALB (BCP)	Correlation to Beckman LX-20 / DX-20	Dye Binding Bromocresol Purple
AMY	IFCC	Colorimetric
AST	IFCC	Colorimetric
CA	Correlation to Beckman LX-20 / DX-20	Arsenazo III Dye
CRP	CRM 470 and Correlation to Beckman LX-20 / DX-20	Latex Agglutination Turbidimetric
CK	IFCC	Enzymatic
CRE	NIST SRM #967	Enzymatic

GLU	NIST SRM #909	Enzymatic
K+	NIST SRM #909	Enzymatic
NA+	Correlation to Beckman LX-20 / DX-20	Enzymatic
TBIL	NIST SRM #916A	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 Abaxis 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 Abaxis 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 Operator's Manual. Results are printed onto paper rolls available from Abaxis. The paper rolls have 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 Xpress chemistry analyzer system 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 AmLyte 13. 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 then re-run in a new reagent disc.

- CRP is an "acute-phase" protein and rises non-specifically in response to inflammation. Intra-individual variation of c-reactive protein are significant (30 to 60%) and should be taken into account when interpreting values[77]. Serial measurements may be required to estimate the true mean of c-reactive protein in any specific individual.

- HAMA (human anti-mouse antibodies) up to 115 ng/mL do not interfere. Specimens from patients who have received preparations of mouse monoclonal antibodies for diagnosis or therapy may contain HAMA.

- Rheumatoid factor up to a concentration of 644 U/mL do not interfere.

- No high dose hook effect (prozone effect) was observed in this assay at CRP concentrations up to 1,000 mg/L.

- **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 system 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. Human serum pools were prepared. The concentration at which each potential interferent was tested was based on the testing levels in CLSI EP7-A.[78]

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 card 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.

- For the CRP method, the levels of endogenous substances that trip the HEM, LIP, or ICT suppressions are 750 mg/dL for hemoglobin, 750 mg/dL for lipemia, and 35 mg/dL for bilirubin.

- 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
Thirty-five exogenous and therapeutic substances were selected as potential interferents for Abaxis test methods based on recommendations by Young. [79] Significant interference is defined as greater than ± 10% shift in the result for a normal range specimen. Human serum pools were supplemented with known concentrations of the drugs or chemicals and then analyzed. **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 (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
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
Sulfasalaazine	150
Sulfanilamide	50
Theophylline	20

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

	Concentration Which Produces > 10% Interference
Alanine Aminotransferase (ALT)	
Ascorbic acid	20
Oxaloacetate	132
Albumin (ALB)	
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
C-Reactive Protein	
Glutathione	30
Isoniazide	4
L-dopa	5
Oxaloacetate	132

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

	Concentration Which Produces > 10% Interference
Creatine Kinase	
Cephalothin	400
Dopamine	15
L-dopa	5
Methotextrate	0.5
Nitrofurantoin	20
Creatinine	
Ascorbic acid	20
Dopamine	19
L-dopa	5
Epinephrine	1
Glutathione	30
Glucose	
Oxaloacetate	132
Pyruvate	44
Potassium	
Penicillin G	100
Sulfasalazine	150
Sodium	
Cephalothin	400
Methotrexate	0.5
Penicillin G	100
Alanine Aminotransferase (ALT)	
Ascorbic acid	20
Oxaloacetate	132
Total Bilirubin (TBIL)	
Dopamine	19
L-dopa	5
Sulfasalazine	10

11. Expected Values

Samples from a total of 193 adult males and females were used to determine the reference ranges for ALT, albumin, amylase, calcium, creatinine, glucose, total bilirubin, and BUN. Samples from a total of 186 adult males and females were used to determine the reference range for AST . Samples from 125-150 adult males and females were analyzed for electrolytes on the Piccolo blood chemistry analyzer and 69 adult males and females were analyzed on the Piccolo Xpress chemistry analyzer for CRP to determine the reference intervals. The electrolyte ranges were calculated based on the 95% reference interval estimated from the combined (overall) values obtained from the reference subjects and the CRP range was based on demonstrated transferability of the reference intervals from the Beckman.[80] These intervals are provided as a guideline only. It is recommended that your office or institution establish normal ranges for your particular patient population.

Table 4: Piccolo Reference Intervals*

Analyte	Common Units	SI Units
Alanine Aminotransferase (ALT)	10-47 U/L	10-47 U/L
Albumin (ALB)	3.3-5.5 g/dL	33-55 g/L
Amylase (AMY)	14-97 U/L	14-97 U/L
Aspartate Aminotransferase (AST)	11-38 U/L	11-38 U/L
Calcium (CA)	8.0-10.3 mg/dL	2.00-2.58 mmol/L
C-Reactive Protein	< 7.5 mg/L	< 7.5 mg/L
Creatine Kinase (Female)	30-190 U/L	30-190 U/L
Creatine Kinase (Male)	39-380 U/L	39-380 U/L
Creatinine	0.6-1.2 mg/dL	53-106 µmol/L
Glucose	73-118 mg/dL	4.1-6.6 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 Bilirubin (TBIL)	0.2-1.6 mg/dL	3.4-27.4 µmol/L
Blood Urea Nitrogen (BUN)	7-22 mg/dL	2.5-7.9 mmol/L

*Data was generated using equivalence device Piccolo Blood Chemistry Analyzer.

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 Dynamic Ranges

Analyte	Common Units	SI Units
Alanine Aminotransferase (ALT)	5-2000 U/L	5-2000 U/L
Albumin (ALB)	1-6.5 g/dL	10-65 g/L
Amylase (AMY)	5-4000 U/L	5-4000 U/L
Aspartate Aminotransferase (AST)	5-2000 U/L	5-2000 U/L
Calcium	4.0-16.0 mg/dL	1.0-4.0 mmol/L
C-Reactive Protein	5.0-200.0 mg/L	5.0-200.0 mg/L
Creatine Kinase	5-5,000 U/L	5-5,000 U/L
Creatinine	0.2-20 mg/dL	18-1768 µmol/L
Glucose	10-700 mg/dL	0.6-38.9 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 Bilirubin (TBIL)	0.1-30 mg/dL	1.7-513 µmol/L
Blood Urea Nitrogen (BUN)	2-180 mg/dL	0.7-64.3 mmol/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. GLU >700* U/L. If lower than the dynamic range, a

"<" will be printed with an asterisk, e.g. GLU <10* 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 Technical Support.

Sensitivity (Limits of Detection)
The lower limit of the reportable (dynamic) range for each analyte is: alanine aminotransferase 5 U/L; albumin 1 g/dL (10 g/L); amylase 5 U/L; aspartate aminotransferase 5 U/L; calcium 4.0 mg/dL (1.0 mmol/L); c-reactive protein 5.0 mg/L; creatine kinase 5 U/L; creatinine 0.2 mg/dL (18 µmol/L); glucose 10 mg/dL (0.6 mmol/L); potassium 1.5 mmol/L; sodium 110 mmol/L; total bilirubin 0.1 mg/dL (1.7 µmol/L); and blood urea nitrogen 2.0 mg/dL (0.7 mmol/L).

Precision
Precision studies were conducted using CLSI EP5-A guidelines[81] with modifications based on CLSI EP18-A[82] for unit-use devices. Results for within-run and total precision were determined using two levels of commercially available control materials and in the case of potassium two levels of plasma pools. The studies made use of multiple instruments. Two reagent disc lots were used for the electrolytes and one for c- reactive protein. Creatine kinase, creatinine, glucose, sodium and urea nitrogen testing was performed at one site; potassium testing was performed at two sites over 20 days; c-reactive protein (serum 1, control 1 and control 2) testing was done at two sites over a period of five days. C-reactive protein serums 2 and 3 and plasmas 1 and 2 were done at one site over a period of five days. 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 precision studies are shown in Table 6.

Table 6: Precision

Analyte	Sample Size	Within-Run	Total
Alanine Aminotransferase (U/L)	N = 80		
Control Level 1			
Mean		21	21
SD		2.76	2.79
%CV		13.4	13.5
Control Level 2			
Mean		52	52
SD		2.70	3.25
%CV		5.2	6.2
Albumin (g/dL)	N = 80		
Control Level 1			
Mean		5.6	5.6
SD		0.09	0.11
%CV		1.7	2.1
Control Level 2			
Mean		3.7	3.7
SD		0.07	0.11
%CV		2.0	2.9
Amylase (U/L)	N = 80		
Control Level 1			
Mean		46	46
SD		2.40	2.63
%CV		5.2	5.7

Control Level 2			
Mean		300	300
SD		11.15	11.50
%CV		3.7	3.8
Aspartate Aminotransferase (U/L)	N = 80		
Control Level 1			
Mean		47	47
SD		0.98	1.84
%CV		2.1	3.9
Control Level 2			
Mean		145	145
SD		1.83	4.62
%CV		1.3	3.2
Calcium (mg/dL)	N = 80		
Control Level 1			
Mean		8.6	8.6
SD		0.21	0.25
%CV		2.4	2.9
Control Level 2			
Mean		11.8	11.8
SD		0.39	0.40
%CV		3.3	3.4
C-Reactive Protein (mg/L)			
Serum 1	N = 80		
Mean		8.3	8.3
SD		0.70	0.81
%CV		8.4	9.8
Serum 2	N = 40		
Mean		8.1	8.1
SD		0.49	0.51
%CV		6.1	6.3
Serum 3	N = 40		
Mean		8.8	8.8
SD		0.54	0.54
%CV		6.2	6.2
C-Reactive Protein (mg/L)			
Plasma 1	N = 40		
Mean		34.5	34.5
SD		1.04	1.09
%CV		3.0	3.2

Plasma 2	N = 40		
Mean		105.5	105.5
SD		2.06	2.30
%CV		1.9	2.2
Control 1	N = 80		
Mean		33.0	33.0
SD		1.21	2.12
%CV		3.7	6.4
Control 2	N = 80		
Mean		108.0	108.0
SD		1.88	3.14
%CV		1.7	2.9
Creatine Kinase (U/L)	N = 120		
Control 1			
Mean		134	134
SD		2.7	2.7
%CV		2.0	2.0
Control 2			
Mean		526	526
SD		7.7	7.7
%CV		1.5	1.5
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)	N = 80		
Control 1			
Mean		66	66
SD		0.76	1.03
%CV		1.1	1.6
Control 2			
Mean		278	278
SD		2.47	3.84
%CV		0.9	1.4
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 Bilirubin (mg/dL)	N = 80		
Control Level 1			
Mean		0.8	0.8
SD		0.06	0.07
%CV		8.0	9.3
Control Level 2			
Mean		5.2	5.2
SD		0.09	0.15
%CV		1.7	2.8
Blood 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: 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

Lithium heparinized whole blood and serum samples were collected and assayed by comparative methods for chloride, creatine kinase, creatinine, glucose, potassium, sodium, total carbon dioxide and urea nitrogen. The whole blood samples were analyzed at the field sites and the serum samples were analyzed by the comparative methods. In some cases, high and low supplemented samples were used to cover the dynamic range. For CRP, lithium heparinized plasma samples were tested by the Piccolo Xpress chemistry analyzer and a comparative method.

Representative correlation statistics are shown in Table 8.

Table 8: Correlation with Comparative Method(s)**

	Correlation Coefficient	Slope	Intercept	SEE	N	Sample Range	Comparative Method
Alanine	0.981	0.905	1.3	3.21	86	10-174	Paramax®
Aminotransferase (U/L)	0.985	0.946	- 2.5	2.84	67	10-174	Technicon
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
Amylase (U/L)	0.979	0.692	- 4.7	3.11	99	11-92	Paramax
	0.963	1.065	- 4.1	3.47	80	19-118	Technicon
Aspartate	0.93	0.87	5.3	2.76	159	13-111	Paramax
Aminotransferase (U/L)	1.0	0.97	3.0	1.9	46	13-252	DAX™
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
C-Reactive Protein (mg/L)	0.998	0.990	- 0.4	4.6	113	5.4-198.6	Beckman
Creatine Kinase (U/L)	0.967	1.194	- 25	9.05	47	6-813	Cobas Fara®
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
Potassium (mmol/L)	0.984	0.99	0.13	0.10	130	1.3 – 9.5	Siemens
Whole Blood (waived laboratory)							VISTA Plasma
Potassium (mmol/L)	0.984	0.98	0.12	0.18	178	1.5 – 8.6	Siemens
Whole Blood (moderately complex laboratory)							VISTA Plasma
Potassium (mmol/L)	0.99	0.98	0.06	0.14	178	1.4-8.5	Siemens
Serum (moderately complex laboratory)							VISTA Serum
Sodium (mmol/L)	0.937	0.782	27.7	3.79	113	116-154	Radiometer KNA 2
Total Bilirubin (mg/dL)	0.974	0.901	0.0	0.07	250	0.2-3.7	Paramax
	0.980	1.113	- 0.4	0.09	91	0.1-6.4	Beckman
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.
**Data was generated using equivalence device Piccolo Blood Chemistry Analyzer.

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 and is dependent on a number of factors. The primary effect is dependent upon the number of blood cells present in the patient sample.[83]

Table 9: Sample-type Correlation for CRP
For the CRP test method the Deming regression analysis for sample-type gave the following results for n = 21.

Y Axis	X Axis	R^2	Slope	Intercept
Lithium Heparinized Plasma	Lithium Heparinized Whole Blood	1.000	0.995	0.2
Serum	Lithium Heparinized Whole Blood	0.999	1.005	0.5
Serum	Lithium Heparinized Plasma	0.999	1.010	0.3

No significant differences between lithium heparinized whole blood, lithium heparinized plasma, and serum results were observed for CRP.

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

denotes conformity to specified European directives

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