| **Intact PTH** |
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| **Purpose** | This procedure provides instructions INTACT PTH on ABBOTT INSTRUMENTATION. The Alinity i Intact PTH assay is a chemiluminescent microparticle immunoassay (CMIA) used for the quantitative determination of intact parathyroid hormone (PTH) in human serum and plasma on the Alinity i analyzer. |
| **Policy Statements** | This procedure applies to all personnel responsible for operating Alinity ci or Architect i1000 at Children’s Minnesota Laboratory in Minneapolis. |
| **Principle** | This assay is a two-step immunoassay for the quantitative determination of intact PTH in human serum and plasma using chemiluminescent microparticle immunoassay (CMIA) technology. Sample, anti-PTH coated paramagnetic microparticles, and assay diluent are combined and incubated. The intact PTH present in the sample binds to the anti-PTH coated microparticles. The mixture is washed. Anti-PTH acridinium-labeled conjugate is added to create a reaction mixture and incubated. Following a wash cycle, Pre-Trigger and Trigger Solutions are added.The resulting chemiluminescent reaction is measured as relative light units (RLUs). There is a direct relationship between the amount of intact PTH in the sample and the RLUs detected by the system optics.The Alinity i Intact PTH assay can be used with both STAT and Routine protocols. The STAT protocol has a shorter incubation time in comparison to the Routine protocol. STAT and Routine protocols require separate calibrations but require only one reagent kit. To account for differences between the STAT and Routine protocols, a correction factor has been added to the STAT protocol. The correction factor ensures that patient specimen results are consistent between the STAT and Routine protocols. As a consequence of the correction factor, controls read differently between the STAT and Routine protocols.For additional information on system and assay technology, refer to the Alinity ci-series Operations Manual, Section 3. |
| **Clinical Significance** | PTH is a single chain polypeptide of 84 amino acids produced by the parathyroid gland. Intact PTH1-84 is secreted into the blood stream and undergoes extensive proteolytic modifications. In contrast to its degradation products, the concentration of intact PTH is relatively independent of glomerular filtration rate and reflects the biologically active portion of the hormone.The primary role of PTH is to regulate the blood calcium level. PTH synthesis and secretion are stimulated within a few minutes by low concentrations of ionized calcium. The biological activity of PTH is to increase absorption of dietary calcium, decrease renal clearance and mobilize skeletal calcium stores. Abnormally high ionized calcium concentrations suppress secretion of PTH.In conjunction with serum calcium levels, the Alinity i Intact PTH assay may be used as an aid in the differential diagnosis of hypercalcemia, hypocalcemia and parathyroid disorders. PTH determination is important in monitoring dialysis patients to manage renal osteodystrophy. |
| **Analyzer** | **Minneapolis: Abbott Alinity c (Sunquest method code: MALIC)****Back up: Abbott Architect i1000SR (Sunquest method code: AI1)** |
| **Sunquest Test Codes** | **PTHB****IPTHC (intraoperative PTH)** |
| **Specimen** | For Intraoperative PTH (IPTHC): Potassium EDTA plasma (lavender) strongly preferred. Lithium Heparin (no gel) also acceptable. For PTH with Calcium PTHB (non-intraoperative): Serum without gel or other additives (RED NO GEL) should be drawn. (Backup method, Mayo Medical Laboratories, accepts serum only.)Add-on testing: Add-on PTH samples can be any of the acceptable sample types if they are marked as such so that subsequent add-ons will not be run in error on the incorrect sample type. (For example, an EDTA plasma sample cannot be used for an add-on testing of a calcium test.) Add-on testing, continued: If a patient has already had a calcium reported on the specimen that a PTH was added to, do one of the following:either 1) order the IPTHC and cancel the added-on PTHB (you may use EDTA plasma, lithium heparin plasma or serum NO GEL sample types if it is PTH only- do not run calcium on EDTA plasma) OR,2) copy the already reported value from the previously run calcium test into the PTHB calcium by looking up the calcium value and manually entering it in MEM. Please do not report two different calcium results from specimens with the same draw times. Minimum Processed Sample Volume: IPTHC (intraoperative): 200 µL of potassium EDTA plasma for PTH on the Abbott Architect i1000SRPTHB: 200 µL of serum (no gel) for PTH on Abbott Architect i1000SR and 200 uL serum for Calcium on the Siemens Vista**Minimum sample volume:** 0.6 mL blood, 0.2 mL serum/plasmaMaximum number of replicates sampled from the same sample cup: 9Priority loaded Sample volume for first test: 200 μL Sample volume for each additional test from same sample cup: 150 μLRoutinely loaded:: Sample volume for first test: 200 μL Sample volume for each additional test from same sample cup: 150 μL**Stability when separated from cells/gel:** **20 to 25°C** if there is any delay in testing, freeze the sample.**2 to 8°C** 2 days**-20°C** 6 monthsPTH is relatively unstable: optimization of pre-analytical conditions, including specimen type, sampling time and storage conditions, is essential. In order to minimize changes in PTH concentration, it is necessary to select the appropriate tube type and size and avoid multiple tube-to-tube transfers**Rejection criteria:** * Unlabeled tube, sample type other than serum or acceptable plasma
* heat-inactivated specimens
* pooled specimens
* grossly hemolyzed specimens
* specimens with obvious microbial contamination

**Preparation:** 1. Whole blood specimens should be centrifuged following complete clot formation, according to Specimen Processing procedures prior to analysis.
2. Serum or plasma should be physically separated from cells as soon as possible with a maximum limit of two hours from the time of collection.
3. Specimens should be free of particulate matter.
4. Transfer serum or plasma directly to a properly labeled pilot tube.
5. Architect and Alinity systems utilize a specimen level detect mechanism, so special racks specific to tube-type are not required.
6. Minimum labeling includes sample accession ID, and/ or patient name, medical record number, collection date and time.
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| **Reagents** | **Reagent Handling** Upon receipt, gently invert the unopened reagent kit by rotating it over and back for a full 180 degrees, 5 times with green label stripe facing up and then 5 times with green label stripe facing down. This ensures that liquid covers all sides of the bottles within the cartridges. During reagent shipment, microparticles can settle on the reagent septum.**Place a check in the square on the reagent kit to indicate to others that the inversions have been completed.**Upon receipt, gently invert cartridges 5 times, then place reagent cartridges in an upright position for 1 hour before use to allow bubbles that may have formed to dissipate. If a reagent cartridge is dropped, place in an upright position for 1 hour before use to allow bubbles that may have formed to dissipate. Reagents are susceptible to the formation of foam and bubbles. Bubbles may interfere with the detection of the reagent level in the cartridge and cause insufficient reagent aspiration that may adversely affect results. Use a pipette to remove all bubbles prior to loading on the Alinity or Architect system.* Do not use reagents beyond the expiration date.
* Do not pool reagents within a kit or between kits.
* Do not use components from one lot with components from another lot.
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|  | **Alinity c** and **Architect i1000**

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| ***Product Description*** | ***Product Code*** | ***Stability*** |
| Alinity i Intact PTH Reagent Kit | 08P3121 | **Store at:** 2 to 8°C**Unopened:** Until expiration DateStore in upright position. If cartridge does not remain upright, gently invert the cartridge 10 times and place in an upright position for 8 hours before use.Reagents may be stored on or off the system. If removed from the system, store reagents with new replacement caps in an upright position at 2 to 8°C. For reagents stored off the system, it is recommended that they be stored in their original trays or boxes to ensure they remain upright.**On-board**: 30 daysStore in upright position. If cartridge does not remain upright during storage, discard the cartridge.Do not reuse original reagent caps or replacement caps due to the risk of contamination and potential to compromise reagent performance. |
| Alinity i Intact PTH Calibrator Kit | 08P3101 | **Store at:** 2 to 8°C**Unopened:** Until expiration Date**Opened expiration:** 60 days |
| Architect Intact PTH Package Insert | 08K2527 | **Store at:** 2 to 8°C**Unopened:** May be used immediately after removal from 2-8°C storage.Store in upright position.**Opened expiration: 30 days.** **Discard after 30 days.**For information on tracking onboard time, refer to the ARCHITECT System Operations Manual, Section 5.Reagents may be stored on or off the ARCHITECT iSystem. If reagents are removed from the system, store them at 2-8°C (with septums and replacement caps) in an upright position. For reagents stored off the system, it is recommended that they be stored in their original trays and boxes to ensure they remain upright. If the microparticle bottle does not remain upright (with a septum installed) while in refrigerated storage off the system, the reagent kit must be discarded. |
| Architect Intact PTH Package Insert | 08K2503 | **Store at:** 2 to 8°C**Unopened:** manufacturer’s expiration date**Opened expiration 30 days.** Store at -20°C or colder. Prior to use, thaw completely at room temperature (15-30°C) for 30 to 60 minutes and mix THOROUGHLY by gentle inversion (5-10 times). After each use, tightly close the caps, return the thawed calibrators to the carton and store at 2-8°C.Do not refreeze. After thawing, it is suggested to record the thaw date on the carton or the bottles to aid in tracking the expiration date. Discard calibrators 30 days after thaw date or if expired. |

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| **Risk and Safety** | **CAUTION:** This product requires the handling of human specimens. It is recommended that all human-sourced materials be considered potentially infectious and handled in accordance with the OSHA Standard on Bloodborne Pathogens. Biosafety Level 2 or other appropriate biosafety practices should be used for materials that contain or are suspected of containing infectious agents**The following warnings and precautions apply to:** *Conjugate* **WARNING** Contains polyethylene glycol octylphenyl ether and sodium azide.Causes serious eye irritation.Contact with acids liberates very toxic gas.Safety data sheets (MSDS/SDS) available on [Children’s Intranet](https://starnet.childrenshc.org/emergency-and-safety/) |
| **Calibration** | **Alinity c and Architect i1000:**

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| Assay Range: | 4.0 to 2500.0 pg/mL |
| Reference Material: | Abbott Alinity i or Architect i1000 Calibrator Kit |
| Suggested Calibration Levels: | A – 0.0 pg/mLB – 4.8 pg/mLC – 24.0 pg/mLD – 120.0 pg/mLE – 600.0 pg/mLF – 3000 pg/mL |
| Calibration Scheme: | 6 Levels |
| Calibration Frequency: | • For each new lot of reagent • After major maintenance or service, if indicated by quality control results• As indicated in laboratory quality control procedures |
| AMR | AMR is verified with every calibration and at least every 6 months |

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| **Quality Control** | **Both analyzers:****Qc Material: Bio-Rad Lyphochek Specialty Immunoassay Levels 1, 2 and 3****Frequency:** Three levels each day of use.**Stability:** 3 Days at 2° to 8°C.**Preparation:** Reconstitute with exactly 2.0 mL of DI water. Let vials sit for 15 minutes and gently swirl to ensure homogeneity. **Acceptable ranges:** * Non-Bio-Rad controls will utilize manufacturer ranges and 2 SD Westgard rules.
* New lots of Bio-Rad controls should be run for 20 days in parallel with the current lot whenever possible prior to switching to the new lot.
* Refer to the Westgard Rules in Chemistry procedure for current Westgard rules in place for each analyte.
* **Acceptable ranges are current in Unity Real Time only.** Quality Control results must be rejected in Sunquest when the results cross the interface.
* In the event of a QC failure, refer to the [Unity Real Time QC Review, General User](https://starnet.childrenshc.org/References/labsop/chem/quality/ch-2.17-unity-real-time-qc-review-general-user.pdf) and navigate to the QC Troubleshooting section.
* Do not load or release patients until QC is acceptable in Unity Real Time.
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| **Interferences** | Potential interference in the ARCHITECT Intact PTH assay from hemoglobin, bilirubin, triglycerides, and protein at the levels indicated below was demonstrated by a study based on guidance from the NCCLS Protocol EP7A. There was no significant interference observed (>10%) at the following levels:Hemoglobin 500 mg/dLBilirubin 20 mg/dLTriglycerides 5000 mg/dLProtein Low 4 g/dLProtein High 9.5 g/dL |
| **Reference Intervals** |

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| **Age** | **Lower Limit** | **Upper Limit** |
| Birth to < 1 year | 6 pg/mL | 88 pg/mL |
| 1 year to < 9 years | 15 pg/mL | 65 pg/mL |
| 9 years to <17 years | 22 pg/mL | 88 pg/mL |
| 17 years to < 19 years | 15 pg/mL | 65 pg/mL |

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| **Critical Values** | None specified |
| **Limitations** | • Results should be used in conjunction with other data; e.g., symptoms, results of other tests, and clinical impressions.• If the Intact PTH results are inconsistent with clinical evidence, additional testing is suggested to confirm the result.• Heterophilic antibodies in human serum can react with reagent immunoglobulins, interfering with in vitro immunoassays. Patients routinely exposed to animals or to animal serum products can be prone to this interference, and anomalous values may be observed. Additional information may be required for diagnosis.• This assay has not been clinically validated for intraoperative use. |
| **Dilutions** | Do not dilute. |
| **Result Reporting** | **Alinity c and Architect i1000:*** Results between 4.0 and 2500.0 pg/mL without error messages are released
* Results below 4.0 without error messages are reported as < 4.0 pg/mL.
* Results > 2500.0 should be reported as > 2500 pg/mL.
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| **Specimen Storage** | Promptly stopper tested specimen and store upright in a specimen rack. Every 8 hours remove specimens to refrigerator/freezer storage. Samples are retained 14 days in specimen storage freezer. |
| **References** | 1. Abbott Architect Intact PTH reagent cartridge insert sheet Abbott Laboratories, Abbott Park, IL, 60064. Revised Date January 2016
2. Abbott Architect Intact PTH calibrator insert sheet Abbott Laboratories, Abbott Park, IL 60064. Revised November 2015.
3. Abbott Architect Safety Data Sheet, Abbott Diagnostics, Abbott Park, IL 60064. Revised 2016-04-09.
4. Abbott Alinity i Intact PTH Reagent Kit Instructions for Use, Abbott Diagnostics, Abbott Park IL 60064. Revised March 2018
5. Abbott Alinity i Intact PTH Calibrator Kit Instructions for Use, Abbott Diagnostics, Abbott Park IL 60064. Revised March 2018
6. Bio-Rad Lyphochek Specialty Immunoassay Control Product Insert, Bio-Rad Laboratories, Irvine, CA 92618 January 2018
7. Caliper Paediatric Reference Intervals. The Hospital for Sick Children, Toronto, Ontario
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| **Historical Record** | **Version** | **Written/Revised by:** | **Effective Date:** | **Summary of Revisions** |
|  | Elauteria Earnhardt | May 18, 2020 | New Procedure for Abbott analyzers |
| 1 | Erin Bartos | October 28, 2020 | Updated for Alinity i analyzer.  |