CE IVD DIALAB

**Liquid Reagents – ready to use**

# ALKALINE PHOSPHATASE

**mod. IFCC**
2 Reagents

**Diagnostic reagent for quantitative in vitro determination of alkaline phosphatase (ALP) in human serum or plasma on photometric systems**

| REF | Cont. | | |
|---|---|---|---|
| **D03101B** | **1 x 1.2 L** | 1 x 1 L | Reagent 1 |
| | | 1 x 250 mL | Reagent 2 |
| **D95564** | **5 x 100 mL** | 4 x 100 mL | Reagent 1 |
| | | 1 x 100 mL | Reagent 2 |
| **D95565** | **5 x 50 mL** | 4 x 50 mL | Reagent 1 |
| | | 1 x 50 mL | Reagent 2 |
| **D00523** | **5 x 25 mL** | 4 x 25 mL | Reagent 1 |
| | | 1 x 25 mL | Reagent 2 |
| **D00524** | **5 x 10 mL** | 4 x 10 mL | Reagent 1 |
| | | 1 x 10 mL | Reagent 2 |
| **DA0803*** | **5 x 50 mL** | 5 x 40 mL | Reagent 1 |
| | | 5 x 10 mL | Reagent 2 |

Additionally offered:

| | | | |
|---|---|---|---|
| D98485 | 5 x 3 mL | Calibrator | Diacal Auto |
| D98481 | 12 x 5 mL | Control normal | Diacon N |
| D98482 | 12 x 5 mL | Control abnormal | Diacon P |

* Autolyser System Pack

## TEST PARAMETERS

| | |
|---|---|
| Method: | Colorimetric, Kinetic, Increasing Reaction, mod. IFCC |
| Wavelength: | 405 nm (400 – 420 nm) |
| Temperature: | 37°C |
| Sample: | Serum, heparin plasma |
| Linearity: | up to 4000 U/L (on Hitachi 911) |
| Sensitivity: | The lower limit of detection is 2 U/L |

## REAGENT COMPOSITION

| COMPONENTS | FINAL CONCENTRATION |
|---|---|
| **Reagent 1:** | |
| 2-Amino-2-Methyl-1-Propanol pH 10.4 | 0.90 mol/L |
| Magnesium Acetate | 1.6 mmol/L |
| Zinc sulphate | 0.4 mmol/L |
| HEDTA | 2.0 mmol/L |
| **Reagent 2:** | |
| p-Nitrophenylphosphate | 16.0 mmol/L |

## REAGENT PREPARATION

**Substrate Start:**
Reagents are ready for use.

**Sample Start:**
Mix 4 parts of Reagent 1 with 1 part of Reagent 2.
( = Working Reagent)

## REAGENT STABILITY AND STORAGE

Conditions: protect from light (Reagent 2!)
close immediately after use
do not freeze the reagents!

**Substrate Start:**
Storage: at 2 – 8°C
Stability: up to the expiration date

**Sample Start (Working Reagent):**
Stability: at 2 – 8 °C 4 weeks
at 15 – 25°C 5 days
Maximum allowable absorbance of the Working Reagent measured at 405 nm against water as reference is 1.0.

## SAMPLE STABILITY AND STORAGE

Stability: at 4 – 8 °C 7 days
at - 20 °C 2 months
Loss of activity: at 15 – 25 °C within 2 – 3 days < 10%
Discard contaminated specimens.

## INTERFERING SUBSTANCES

no interference up to:

| | |
|---|---|
| ascorbic acid | 30 mg/dl |
| conjugated bilirubin | 60 mg/dl |
| unconjugated bilirubin | 25 mg/dl |
| triglycerides | 2000 mg/dl |

## MANUAL TEST PROCEDURE

Bring reagents and samples to room temperature.

**Substrate Start** 37°C

| Pipette into test tube: | Blank: | Sample: |
|---|---|---|
| Reagent 1 | 1000 µl | 1000 µl |
| Sample | | 20 µl |
| Dist. water | 20 µl | |
| Mix. Incubate for approximately 1 minute. Then add: | | |
| Reagent 2 | 250 µl | 250 µl |
| Mix. Read initial absorbance after 1 minute and start a timer. Read absorbance again after exactly 1, 2 and 3 min. | | |

**Sample Start** 37°C

| Pipette into test tubes | Blank: | Sample: |
|---|---|---|
| Working Reagent | 1000 µl | 1000 µl |
| Sample | | 20 µl |
| Dist. water | 20 µl | |
| Mix. Read initial absorbance after 1 minute and start a timer. Read absorbance again after exactly 1, 2 and 3 min. | | |

## CALCULATION (light path 1 cm)

$\Delta A/min = [\Delta A/min\ sample] - [\Delta A/min\ blank]$

Alkaline Phosphatase (U/L) = $\Delta A/min \times Factor$

**Factors (37°C):**

**Substrate Start:** 3433
**Sample Start:** 2757

## UNIT CONVERSION

U/L x 0.01667 = µkat/L

## REFERENCE RANGE*(U/L)

| **Adults:** | Years | 37 °C |
|---|---|---|
| Females | 20-50 | 42–98 |
| Females | > 60 | 53–141 |
| Males | 20-50 | 53-128 |
| Males | > 60 | 56–119 |

| **Children:** | Age | 37 °C | |
|---|---|---|---|
| | | Females | Males |
| | 1–30 days | 48–406 | 75–319 |
| | 1 month–1year | 124–341 | 82–383 |
| | 1–3 year(s) | 108-317 | 104–345 |
| | 4–6 years | 96–297 | 93–309 |
| | 7–9 years | 69–325 | 86–315 |
| | 10–12 years | 51–332 | 42–362 |
| | 13–15 years | 50–162 | 74–390 |
| | 16–18 years | 47-119 | 52–171 |

* It is recommended that each laboratory establishes its own normal range.

## TEST PRINCIPLE

p-Nitro-phenylphosphate + $H_2O$ $\xrightarrow{\text{Alkaline phosphatase}}$ p-Nitrophenol + Phosphate

Under alkaline condition, colorless p-nitrophenol is converted to 4-nitrophenoxide, which develops a very intense yellow color.

Its intensity is proportional to the activity of alkaline phosphatase in the sample.

## PERFORMANCE CHARACTERISTICS

### LINEARITY

The test has been developed to determine alkaline phosphatase activities which correspond to a maximal ΔA/min of 0.25.
If the value is exceeded the sample should be diluted 1+9 with NaCl solution (9 g/L sodium chloride in dist. water) and results multiplied by 10.

### PRECISION

| Intra-assay n = 20 | Mean [U/L] | SD [U/L] | CV [%] |
|---|---|---|---|
| Sample 1 | 68.6 | 0.58 | 0.85 |
| Sample 2 | 107 | 0.71 | 0.67 |
| Sample 3 | 243 | 0.97 | 0.40 |

| Inter-assay n = 20 | Mean [U/L] | SD [U/L] | CV [%] |
|---|---|---|---|
| Sample 1 | 69.2 | 1.37 | 1.99 |
| Sample 2 | 104 | 1.22 | 1.08 |
| Sample 3 | 238 | 2.40 | 1.01 |

### METHOD COMPARISON

A comparison between Dialab Alkaline Phosphatase (y) and a commercially available test (x) using 104 samples gave following results: y = 1.01 x + 1.51 U/l; r= 0.999.

## QUALITY CONTROL

All control sera with ALP values determined by this method can be used.
We recommend:

REF Cont.

| | | | |
|---|---|---|---|
| **D98481** | 12 x 5 ml | **DIACON N** | Assayed Control Serum Normal |
| **D98482** | 12 x 5 ml | **DIACON P** | Assayed Control Serum Abnormal |

## CALIBRATION

The use of an ALP Calibrator is optional.
We recommend:

REF Cont.

| | | | |
|---|---|---|---|
| **D98485** | 5 x 3 ml | **DIACAL AUTO** | Assayed Multi Calibration Serum |

## AUTOMATION

Special adaptations for automated analyzers can be made on request.

## WARNINGS AND PRECAUTIONS

1. The reagents contain sodium azide (0.95 g/L) as preservative. Do not swallow! Avoid contact with skin and mucous membranes.
2. During reaction p-nitrophenol is produced which is poisonous when inhaled, swallowed or absorbed through skin. If the reaction mixture comes in contact with skin or mucous membranes wash copiously with water!
3. Take the necessary precautions for the use of laboratory reagents.

## WASTE MANAGEMENT

Please refer to local legal requirements.

## REFERENCES


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2°C 8°C IVD


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