

## Intended Use

For the quantitative determination of alkaline phosphatase in human serum using the Mindray BS-480 analyzer. *In vitro* diagnostic medical device

## Clinical Significance

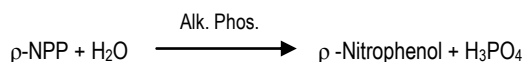
Serum alkaline phosphatase estimations are of interest in the diagnosis of two groups of conditions; hepatobiliary disease and bone disease associated with increased osteoblastic activity.<sup>1</sup>

## Test Summary

Alkaline phosphatase in serum is determined by measuring the rate of hydrolysis of various phosphate esters under specified conditions. *p*-Nitrophenyl Phosphate is one such phosphate ester and was introduced as a substrate by Fujita in 1939.<sup>2</sup>

Bessey, Lowry, and Brock published an endpoint procedure in 1946<sup>3</sup> while Bowers and McComb reported a kinetic procedure in 1966.<sup>4</sup> The kinetic procedure has undergone several modifications and been recommended for routine analysis.<sup>5,6</sup> This liquid reagent is based on the recommended method of the AACC.<sup>7</sup>

## Principle



*p*-Nitrophenyl phosphate is hydrolyzed to *p*-nitrophenol and inorganic phosphate. The rate at which the *p*-NPP is hydrolyzed, measured at 405 nm, is directly proportional to the alkaline phosphatase activity.

## Reagent Composition

After combining R1 and R2 as directed the reagent contains: AMP Buffer (pH 10.45), *p*-NPP ≤16mM, Magnesium ions ≥1.0mM, activators and preservatives.

## Reagent Preparation

The reagents are ready to use.

## Reagent Storage and Stability

Store reagent set at 2-8°C. The reagents are stable until the expiration date if stored as directed. Protect from direct light and avoid microbial contamination. **NOTE:** The R2 reagent is temperature sensitive and can be affected by prolonged exposure to room temperature. Return reagent to 2-8°C as soon as possible after use. Manufacturer studies have shown reagent is stable for 30 days once placed in the refrigerated reagent carousel (2-10°C), however reagent stability may vary based on individual laboratory conditions.

## Precautions and Hazards

1. This reagent set is for *in vitro* diagnostic use only. Do not ingest any material.
2. Do not use if the initial absorbance of the working reagent is greater than 1.0 at 405 nm or if the reagent fails to meet the stated parameters of performance.
3. Reagent should not be used if it fails to recover stated values in control sera or shows evidence of microbial contamination.
4. All specimens and controls should be handled in accordance with good laboratory practices using appropriate precautions as described in the CDC/NIH Manual, "Biosafety in Microbiological and Biomedical Laboratories," 2<sup>nd</sup> Ed., 1988, HHS Publication No. (CDC) 88-8395.

### Hazards:

**R1: Hazard Classifications:** Skin Corrosion/Irritation (Category 1), Eye Damage/Irritation (Category 2), Respiratory Sensitizer (Category 1), Specific Target Organ Toxicity, Single Exposure; Respiratory System (Category 1), Specific Target Organ Toxicity, Repeat Exposure; Respiratory System and Teeth (Category 1)

**Hazard Statements:** H314: Causes severe skin burns and eye damage, H319: Causes serious eye irritation, H334: May cause allergy or asthma symptoms or breathing difficulties if inhaled, H370: Causes damage to organs, H372: Causes damage to organs through prolonged or repeated exposure.

**Precautionary Statements: Prevention:** P260: Do not breathe dust/fume/gas/mist/vapor/spray, P264: Wash skin thoroughly after handling, P270: Do not eat, drink or smoke when using this product, P280: Wear protective gloves/protective clothing/eye protection/face protection, P285: In case of inadequate ventilation wear respiratory protection. **Response:** P310: Immediately call a POISON CENTER or doctor/physician, P314: Get medical advice/attention if you feel unwell, P363: Wash contaminated clothing before reuse. P301 + P330 + P331: IF SWALLOWED: rinse mouth. Do NOT induce vomiting. P303 + P361 + P353: IF ON SKIN (or hair): Remove/Take off immediately all contaminated clothing. Rinse SKIN with water/shower, P304 + P340: IF INHALED: Remove victim to fresh air and Keep at rest in a position comfortable for breathing, P305 + P351 + P338: IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. P337 + P313: IF eye irritation persists: Get medical advice/attention. **Storage:** P404: Store in a closed container.

**Disposal:** P501: Dispose of contents into sewer system after diluting with large volumes of water, if in accordance with local regulations.

**R2: Hazard Classifications:** Skin Corrosion/Irritation (Category 2), Eye Damage/Irritation (Category 2), Specific Target Organ Toxicity, Single Exposure; Liver, Kidney, Central Nervous System, Eyes (Category 2), Specific Target Organ Toxicity, Repeated Exposure; Liver, Kidney, Central Nervous System (Category 2), Reproductive Toxicity (Category 2), Germ Cell Mutagenicity (Category 2), Carcinogenicity (Category 2)

**Hazard Statements:** H315: Causes skin irritation, H319: Causes serious eye irritation, H341: Suspected of causing genetic defects, H351: Suspected of causing cancer, H361: Suspected of damaging fertility or the unborn child, H371: May cause damage to organs, H373: Causes damage to organs through prolonged or repeated exposure

**Precautionary Statements: Prevention:** P202: Do not handle until all safety precautions have been read and understood, P260: Do not breathe dust/fume/gas/mist/vapors/spray, P264: Wash skin thoroughly after handling, P270: Do not eat, drink or smoke when using this product, P280: Wear protective gloves/protective clothing/eye protection/face protection. **Response:** P314: Get medical advice/attention if you feel unwell, P362: Take off contaminated clothing and wash before use, P302 + P352: IF ON SKIN: wash with plenty of soap and water. P305 + P351 + P338: IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. P309 + P311: IF exposed or if you feel unwell: call a POISON CENTER or doctor/physician, P332 + P313: IF SKIN irritation occurs: Get medical advice/attention, P337 + P313: IF eye irritation persists: Get medical advice/attention. **Storage:** P404: Store in a closed container. **Disposal:** P501: Dispose of contents to an approved waste disposal plant. **Refer to the Safety Data Sheet for this product (SDS-ALP600) available at [www.medtestdx.com](http://www.medtestdx.com).**

## Specimen Collection and Storage

1. Use non-hemolyzed serum (plasma should not be used since anticoagulant agents inhibit alkaline phosphatase activity).<sup>8,9</sup>
2. Serum samples should be stored at 2-8°C and run within two days.<sup>10</sup>
3. Specimen collection should be carried out in accordance with NCCLS M29-T2.<sup>11</sup> No method can offer complete assurance that human blood samples will not transmit infection. Therefore, all blood samples should be considered potentially infectious.



**Signal Word: Danger**

# Alkaline Phosphatase Reagent Set

## Interferences

1. Young, et al<sup>8</sup> provide a list of drugs and other substances that interfere with the determination of ALP activity.

## Materials Provided

Alkaline Phosphatase, catalog number: ALP480

## Materials Required but not Provided

1. Mindray BS-480 Analyzer.
2. BS-480 Operation manual.
3. Chemistry Control, catalog number CHEQ480

## Limitations

1. This methodology measures total alkaline phosphatase irrespective of tissue or organ of origin. Further tests may be necessary to assist in differential diagnosis.
2. Samples with values exceeding 1000 IU/L should be diluted with an equal volume of saline and re-assayed multiplying the results by two.

## Calibration

The procedure is standardized by means of the millimolar absorptivity of p-nitrophenol (18.75 at 405nm) under the specified conditions. Results are based on the change in absorbance per unit of time; all parameters must be known and controlled.

## Quality Control

The validity of the reaction should be monitored using control sera with known normal and abnormal ALP activities and should be run with every working shift in which ALP assays are performed. It is recommended that each laboratory establish its own frequency of control determination. Quality control requirements should be performed in conformance with local, state, and/or Federal regulations or accreditation requirements.

## Expected Values

Adults 35-123 IU/L at 37°C. This reference range is based on a study performed by the manufacturer using samples from 783 apparently healthy adults. Children have a higher normal value. It is strongly suggested that each laboratory establish its own normal range.

## Performance

1. Assay Range: 2-1000 IU/L
2. Comparison: A study was performed between the Mindray BS-480 and a similar analyzer and method, resulted in the following:

Method	Alkaline Phosphatase
N	81
Mean Alk. Phos. (IU/L)	126.5
Range (IU/L)	19-833
Standard Deviation	147.7
Regression Analysis	$y = 1.063x - 9.7$
Correlation Coefficient	0.9987

3. Precision: Precision studies were performed using the Mindray BS-480 analyzer following a modification of the guidelines which are contained in NCCLS document EP5-T2.<sup>13</sup>

Sample	Within Day			Total		
	LOW	MID	HIGH	LOW	MID	HIGH
N	20	20	20	40	40	40
Mean	88.6	302.1	887.3	89.5	311.2	870.7
Standard Deviation	1.1	1.2	3.6	4.2	12.5	23.0
Coefficient of Variation (%)	1.3%	0.4%	0.4%	4.7%	4.0%	2.6%

4. Sensitivity: 2SD Limit of Detection (95% Conf) = 2 IU/L

## References

1. Tietz, N.W., Fundamentals of Clinical Chemistry, W.B. Saunders co., p 603 (1982).
2. Fujita, H., J. Biochem, (Japan) 30:69 (1969).
3. Bessey, O.A., Lowry, O.H., Brock, M.J., J. Biol. Chem. 164:321 (1964).
4. Bowers, G.N., Jr., McComb, R.B., Clin. Chem. 12:70 (1966).
5. The Committee on Enzymes of the Scandinavian Society for Clinical Chemistry and Clinical Physiology, Scand. J. Clin. Lab. Invest 32:291 (1974).
6. Wilkinson, J.H., et al, Clin. Chem. 15:487 (1969).
7. Tietz, N.W., et al, Clin. Chem. 29:751 (1983).
8. Young, D.S., et al, Clin. Chem. 21:1D (1975).
9. Demetriou, J.A., Drewes, P.A., Gin, J.B., Clinical Chemistry: Principles and Technics, 2<sup>nd</sup> Ed., Hagerstown (MD), Harper & Row, p. 927 (1974).
10. Rej, R., Clin. Chem. 23:1903 (1977).
11. NCCLS document "Protection of Laboratory Workers from Infectious Disease Transmitted by Blood, Body Fluids, and Tissue", 2<sup>nd</sup> Ed. (1991).
12. NCCLS document "Interference testing in Clinical Chemistry", 2<sup>nd</sup> Ed. (1992).
13. NCCLS document "Evaluation of Precision Performance of Clinical Chemistry Devices", 2<sup>nd</sup> Ed. (1992).

**CHEMISTRY PARAMETERS**

Chem:	ALKP	No.:		Sample Type:	Serum
Chemistry:	Alkaline Phosphatase			Print Name:	ALKP
Reaction Type:	Kinetic			Reaction Direction:	Positive
Pri Wave:	412			Sec Wave:	660
Unit:	U/L			Decimal:	0
Blank Time:	0	0		Reaction Time:	56 71
	Sample Vol.	Aspirated	Diluent	Reagent Vol.	Diluent
Standard:	3.0 ul	-- ul	-- ul	R1:	120 ul -- ul
Decreased:	-- ul	-- ul	-- ul	R2:	30 ul -- ul
Increased:	-- ul	-- ul	-- ul	R3:	-- ul -- ul
	<input type="checkbox"/> Sample Blank	<input checked="" type="checkbox"/> Auto Rerun		R4:	-- ul -- ul
<b><u>Slope/Offset Adjustment</u></b>					
Slope: 1		Offset: 0			

Linearity Range (Standard)	2	1000	Linearity Limit:	0.2
Linearity Range (Decreased)	___	___	Substrate Depletion:	25000
Linearity Range (Increased)	___	___	Mixed Blank Abs:	
R1 Blank Abs:	___	___	Uncapping Time	
Blank Response:	___	___	Reagent Alarm Limit:	
Twin Chemistry:			<input type="checkbox"/> Enzyme Linear Extension	
<input type="checkbox"/> Prozone Check		<input type="radio"/> Rate Check	<input type="radio"/> Antigen Addition	
Q1:	Q2:	Q3:	Q4:	
PC:	ABS:			

# Alkaline Phosphatase Reagent Set

## CALIBRATION PARAMETERS

<b>Calibrator Definition</b>						
Calibrator:	*	Lot No.:	*			
Exp Date:	*					
<b>Carousel</b>		<b>Pos</b>				
Sample Carousel 1	*					
Sample Carousel 2						
Sample Carousel 3						
<b><u>Reagent/Calibration</u></b>						
<u>Calibrator</u>	<u>Pos</u>	<u>Lot No</u>	<u>Exp Date</u>	<u>Chem</u>	<u>Conc</u>	<u>Unit</u>
Water	W	*	*	ALKP	0	U/L
<b><u>Calibration Setup</u></b>						
Chem:	ALKP					
<b><u>Calibration Settings</u></b>						
Math Model:	K Factor					
Factor:	2708	Replicates:	1			
<b><u>Acceptance Limits</u></b>						
Cal Time:	*	Hour				
Slope Diff:	---	SD:	---			
Sensitivity :	---	Repeatability:	---			
Deter Coeff:	---					
<b><u>Auto Calib.</u></b>						
<input type="checkbox"/> Bottle Changed	<input type="checkbox"/> Lot Changed	<input type="checkbox"/> Cal Time				

It is recommended that two levels of control material be assayed daily.

\* Indicates user defined parameter.

**REF** ALP480



Manufactured for MedTest DX  
5449 Research Drive Canton, MI 48188



**IVD**

### Symbol Key

Use by (YYYY-MM-DD)	<b>LOT</b> Lot and batch code	<b>REF</b> Catalog number	Manufacturer
Temperature limitation	Consult instructions for use	<b>IVD</b> <i>In vitro</i> diagnostic medical device	