ALP

cobas®

Alkaline phosphatase liquid acc. to IFCC

Order information

REF	CONTENT		Analyzer(s) on which kit(s) can be used
12173107 122	ALP ([1] 6 x 66 mL, [2] 6 x 16 mL)		Roche/Hitachi MODULAR P
12173158 122	ALP ([1] 6 x 267 mL)		Roche/Hitachi MODULAR P, MODULAR D
12173174 122	ALP ([2] 6 x 71 mL)		
10759350 190	Calibrator f.a.s. (12 x 3 mL)	Code 401	
10759350 360	Calibrator f.a.s. (12 x 3 mL, for USA)	Code 401	
10171743 122	Precinorm U (20 x 5 mL)	Code 300	
12149435 122	Precinorm U plus (10 x 3 mL)	Code 300	
12149435 160	Precinorm U plus (10 x 3 mL, for USA)	Code 300	
10171778 122	Precipath U (20 x 5 mL)	Code 301	
12149443 122	Precipath U plus (10 x 3 mL)	Code 301	
12149443 160	Precipath U plus (10 x 3 mL, for USA)	Code 301	
05117003 190	PreciControl ClinChem Multi 1 (20 x 5 mL)	Code 391	
05947626 190	PreciControl ClinChem Multi 1 (4 x 5 mL)	Code 391	
05947626 160	PreciControl ClinChem Multi 1 (4 x 5 mL, for USA)	Code 391	
05117216 190	PreciControl ClinChem Multi 2 (20 x 5 mL)	Code 392	
05947774 190	PreciControl ClinChem Multi 2 (4 x 5 mL)	Code 392	
05947774 160	PreciControl ClinChem Multi 2 (4 x 5 mL, for USA)	Code 392	
11930630 001	Chimneys		

Some analyzers and kits shown may not be available in all countries. For additional system applications, contact your local Roche Diagnostics representative.

English

System information

For Roche/Hitachi MODULAR P/D analyzers: ACN 158

Intended use

In vitro test for the quantitative determination of alkaline phosphatase (ALP; E.C. 3.1.3.1) in human serum and plasma on Roche automated clinical chemistry analyzers.

Summary[1,2,3,4,5,6]

Alkaline phosphatase in serum consists of four structural genotypes: the liver-bone-kidney type, the intestinal type, the placental type and the variant from the germ cells. It occurs in osteoblasts, hepatocytes, leukocytes, the kidneys, spleen, placenta, prostate and the small intestine. The liver-bone-kidney type is particularly important.

A rise in the alkaline phosphatase occurs with all forms of cholestasis, particularly with obstructive jaundice. It is also elevated in diseases of the skeletal system, such as Paget's disease, hyperparathyroidism, rickets and osteomalacia, as well as with fractures and malignant tumors. A considerable rise in the alkaline phosphatase activity is sometimes seen in children and juveniles. It is caused by increased osteoblast activity following accelerated bone growth.

The assay method was first described by King and Armstrong, modified by Ohmori, Bessey, Lowry and Brock and later improved by Hausamen et al. In 2011 the International Federation of Clinical Chemistry and Laboratory Medicine (IFCC) Scientific Division, Committee on Reference Systems of Enzymes (C-RSE) recommended a reference procedure for the determination of alkaline phosphatase using an optimized substrate concentration and 2-amino-2-methyl-1-propanol as buffer plus the cations magnesium and zinc at 37 °C. This assay follows the recommendations of the IFCC, but was optimized for performance and stability.

Test principle[6]

Colorimetric assay in accordance with a standardized method

- Sample and addition of R1
- Addition of R2 and start of reaction:

$$\text{p-nitrophenyl phosphate} + H_2O \xrightarrow{\text{ALP}} \text{phosphate} + \text{p-nitrophenol}$$

In the presence of magnesium and zinc ions, p-nitrophenyl phosphate is cleaved by phosphatases into phosphate and p-nitrophenol. The p-nitrophenol released is proportional to the ALP activity and is measured photometrically.

Reagents - working solutions

R1 2-Amino-2-methyl-1-propanol: 1.12 mol/L, pH 10.44 (30 °C); magnesium acetate: 2.49 mmol/L; zinc sulfate: 0.50 mmol/L; N-(2-hydroxyethyl)-ethylenediamine triacetic acid: 2.49 mmol/L

R2 p-Nitrophenyl phosphate: 99.5 mmol/L, pH 8.50 (25 °C); preservatives.

Precautions and warnings

For in vitro diagnostic use.
Exercise the normal precautions required for handling all laboratory reagents.
Disposal of all waste material should be in accordance with local guidelines.
Safety data sheet available for professional user on request.

For USA: Caution: Federal law restricts this device to sale by or on the order of a physician.

This kit contains components classified as follows in accordance with the Regulation (EC) No. 1272/2008:

Warning

H315	Causes skin irritation.
H319	Causes serious eye irritation.
H412	Harmful to aquatic life with long lasting effects.

Prevention:

P264	Wash skin thoroughly after handling.
P273	Avoid release to the environment.
P280	Wear protective gloves/ eye protection/ face protection.

Response:

P302 + P352 IF ON SKIN: Wash with plenty of soap and water.

P332 + P313 If skin irritation occurs: Get medical advice/attention.

P337 + P313 If eye irritation persists: Get medical advice/attention.

P362 + P364 Take off contaminated clothing and wash it before reuse.

Disposal:

P501 Dispose of contents/container to an approved waste disposal plant.

Product safety labeling follows EU GHS guidance.

Contact phone: all countries: +49-621-7590, USA: 1-800-428-2336

Reagent handling

R1: Ready for use

R2: Ready for use

Absorption of the atmospheric CO_2 by the opened reagent bottle R1 leads to impaired reagent stability. This kit therefore requires the use of color-coded chimneys which reduce the uptake of CO_2 by the reagent. The chimneys should be placed directly into the apporpriate reagent: white for R1. (Black chimneys are not required). The chimneys can be reused for reagent bottles with the same kit. However, to avoid contamination of the reagent with detergent or dilution of the reagent with water it is not permitted to wash the chimneys before reuse. Chimneys are used on all systems.

Storage and stability

Unopened kit components: Up to the expiration date at 2-8 °C

R1: 2 weeks opened and refrigerated on the analyzer when chimney used.

R2: 4 weeks opened and refrigerated on the analyzer.

Please note: A yellow coloration of reagent R2 does not impair the performance of the assay.

Specimen collection and preparation

For specimen collection and preparation only use suitable tubes or collection containers.

Only the specimens listed below were tested and found acceptable.

Serum

Plasma: Heparin (Li-, Na-, NH_4^+-) plasma.

The sample types listed were tested with a selection of sample collection tubes that were commercially available at the time of testing, i.e. not all available tubes of all manufacturers were tested. Sample collection systems from various manufacturers may contain differing materials which could affect the test results in some cases. When processing samples in primary tubes (sample collection systems), follow the instructions of the tube manufacturer.

Stability:[7] 7 days at 20-25 °C
7 days at 4-8 °C
2 months at -20 °C

Centrifuge samples containing precipitates before performing the assay.

Materials provided

See “Reagents – working solutions” section for reagents.

Materials required (but not provided)

- See “Order information” section
- 0.9 % NaCl
- General laboratory equipment

Assay

For optimum performance of the assay follow the directions given in this document for the analyzer concerned. Refer to the appropriate operator’s manual for analyzer-specific assay instructions.

The performance of applications not validated by Roche is not warranted and must be defined by the user.

Calibration

S1: 0.9 % NaCl

S2: C.f.a.s. (Calibrator for automated systems)

Calibration frequency

2-point calibration is recommended:

- after reagent lot change
- as required following quality control procedures

Calibration interval may be extended based on acceptable verification of calibration by the laboratory.

Traceability: This method has been standardized against the IFCC procedure (2011).[6]

Quality control

For quality control, use control materials as listed in the "Order information" section.

In addition, other suitable control material can be used.

The control intervals and limits should be adapted to each laboratory’s individual requirements. Values obtained should fall within the defined limits. Each laboratory should establish corrective measures to be taken if values fall outside the defined limits.

Follow the applicable government regulations and local guidelines for quality control.

Calculation

The analyzer automatically calculates the analyte activity of each sample.

Conversion factor: U/L x 0.0167 = µkat/L

Limitations – interference

Criterion: Recovery within ± 10 % of initial value.

Icterus:[8] No significant interference up to an I index of 70 for conjugated and unconjugated bilirubin (approximate conjugated and unconjugated bilirubin concentration: 1197 µmol/L or 70 mg/dL).

Hemolysis:[8] No significant interference up to an H index of 500 (approximate hemoglobin concentration: 310.5 µmol/L or 500 mg/dL).

Lipemia (Intralipid):[8] No significant interference up to an L index of 2000. There is poor correlation between the L index (corresponds to turbidity) and triglycerides concentration.

In very rare cases, gammopathy, in particular type IgM (Waldenström’s macroglobulinemia), may cause unreliable results.[9]

For diagnostic purposes, the results should always be assessed in conjunction with the patient’s medical history, clinical examination and other findings.

ACTION REQUIRED

Special Wash Programming: The use of special wash steps is mandatory when certain test combinations are run together on Roche/Hitachi analyzers. Refer to the latest version of the carry-over evasion lists and the operator’s manual for further instructions. US users refer to the Special Wash Programming document, available at usdiagnostics.roche.com, and the operator’s manual for special wash instructions.

Where required, special wash/carry-over evasion programming must be implemented prior to reporting results with this test.

Limits and ranges

Measuring range

1-1200 U/L (0.02-20.00 µkat/L)

Determine samples having higher activities via the rerun function. Dilution of samples via the rerun function is a 1:4 dilution. Results from samples diluted using the rerun function are automatically multiplied by a factor of 4.

Lower limits of measurement

Lower detection limit of the test

0.67 U/L (0.011 µkat/L)

The lower detection limit represents the lowest measurable analyte level that can be distinguished from zero. It is calculated as the value lying 3 standard deviations above that of the lowest standard (standard 1 + 3 SD, repeatability, n = 21).

Expected values

(measured at 37 °C)

Adults[10]	U/L	µkat/L
Men (n = 221)	40–129	0.67–2.15
Women (n = 229)	35–104	0.58–1.74

Children[11]		
Males		
Age	U/L	µkat/L
0 – 14 days	83–248	1.39–4.14
15 days – < 1 year	122–469	2.04–7.83
1 – < 10 years	142–335	2.37–5.59
10 – < 13 years	129–417	2.15–6.96
13 – < 15 years	116–468	1.94–7.82
15 – < 17 years	82–331	1.37–5.53
17 – < 19 years	55–149	0.92–2.49
Females		
Age	U/L	µkat/L
0 – 14 days	83–248	1.39–4.14
15 days – < 1 year	122–469	2.04–7.83
1 – < 10 years	142–335	2.37–5.59
10 – < 13 years	129–417	2.15–6.96
13 – < 15 years	57–254	0.95–4.24
15 – < 17 years	50–117	0.84–1.95
17 – < 19 years	45–87	0.75–1.45

Roche has not evaluated reference ranges in a pediatric population.

Each laboratory should investigate the transferability of the expected values to its own patient population and if necessary determine its own reference ranges.

Specific performance data

Representative performance data on the analyzers are given below. Results obtained in individual laboratories may differ.

Precision

Precision was determined using human samples and controls in an internal protocol with repeatability (n = 21) and intermediate precision (3 aliquots per run, 1 run per day, 21 days). The following results were obtained:

	Repeatability			Intermediate precision		
Sample	Mean		CV	Mean		CV
	U/L	µkat/L	%	U/L	µkat/L	%
Human serum low	63.3	1.06	0.7	62.0	1.03	0.7
Human serum high	305	5.08	0.6	298	4.97	0.7
Precinorm U	84.1	1.40	0.5	85.6	1.43	0.3
Precipath U	215	3.59	0.5	211	3.51	0.5

Method comparison

A comparison of the alkaline phosphatase determination using the Roche ALP IFCC liquid assay (y) versus the Roche ALP IFCC granulate assay (x) on the Roche/Hitachi 917 analyzer gave the following correlation (U/L):

Passing/Bablok[12]	Linear regression
y = 0.981x + 0.812	y = 0.981x + 1.36
τ = 0.990	r = 1.00

Number of samples measured: 66

The activities of the samples were between 46.2 and 1050 U/L (0.77-17.5 µkat/L).

References

1 Greiling H, Gressner AM, eds. Lehrbuch der Klinischen Chemie und Pathobiochemie, 3rd ed. Stuttgart/New York: Schattauer Verlag 1995.

2 King EJ, Armstrong AR. Can Med Assoc J 1934;31:376

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4 Bessey OA, Lowry OH, Brock MJ. A method for the rapid determination of alkaline phosphatase with five cubic millimeters of serum. J Biol Chem 1946;164:321-329.

5 Hausamen TU, Helger R, Rick W, et al. Optimal conditions for the determination of serum alkaline phosphatase by a new kinetic method. Clin Chim Acta 1967;15:241-245.

6 Schumann G, Klauke R, Canalias F, et al. IFCC primary reference procedures for the measurement of catalytic activity concentrations of enzymes at 37° C. - Part 9. Reference procedure for the measurement of catalytic concentration of alkaline phosphatase. Clin Chem Lab Med 2011 Sep;49 (9):1439-46.

7 Guder WG, Narayanan S, Wisser H, et al. List of Analytes; Preanalytical Variables. Brochure in: Samples: From the Patient to the Laboratory. Darmstadt: GIT-Verlag 1996.

8 Glick MR, Ryder KW, Jackson SA. Graphical Comparisons of Interferences in Clinical Chemistry Instrumentation. Clin Chem 1986;32:470-475.

9 Bakker AJ, Mücke M. Gammopathy interference in clinical chemistry assays: mechanisms, detection and prevention. Clin Chem Lab Med 2007;45(9):1240-1243.

10 Abicht K, El-Samalouti V, Junge W, et al. Multicenter evaluation of new GGT and ALP reagents with new reference standardization and determination of 37 °C reference intervals. Clin Chem Lab Med 2001;39:Special Supplement pp S 346.

11 Estey MP, Cohen AH, Colantonio DA, et al. CLSI-based transference of the CALIPER database of pediatric reference intervals from Abbott to Beckman, Ortho, Roche and Siemens Clinical Chemistry Assays: Direct validation using reference samples from the CALIPER cohort. Clin Biochem 2013;46:1197–1219.

12 Bablok W, Passing H, Bender R, et al. A general regression procedure for method transformation. Application of linear regression procedures for method comparison studies in clinical chemistry, Part III. J Clin Chem Clin Biochem 1988 Nov;26(11):783-790.

Instrument settings

US users: Refer to the application sheet and Special Wash Programming document, available at usdiagnostics.roche.com, for additional operating information.

Users of MODULAR analyzers: Enter the application parameters via the barcode sheet.

For further information, please refer to the appropriate operator's manual for the analyzer concerned, the respective application sheets and method sheets of all necessary components.

A point (period/stop) is always used in this Method Sheet as the decimal separator to mark the border between the integral and the fractional parts of a decimal numeral. Separators for thousands are not used.

Symbols

Roche Diagnostics uses the following symbols and signs in addition to those listed in the ISO 15223-1 standard (for USA: see https://usdiagnostics.roche.com for definition of symbols used):

CONTENT	Contents of kit
REAGENT	Reagent
CALIBRATOR	Calibrator
→	Volume after reconstitution or mixing
GTIN	Global Trade Item Number

ALP

Alkaline phosphatase liquid acc. to IFCC

FOR US CUSTOMERS ONLY: LIMITED WARRANTY

Roche Diagnostics warrants that this product will meet the specifications stated in the labeling when used in accordance with such labeling and will be free from defects in material and workmanship until the expiration date printed on the label. THIS LIMITED WARRANTY IS IN LIEU OF ANY OTHER WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR PARTICULAR PURPOSE. IN NO EVENT SHALL ROCHE DIAGNOSTICS BE LIABLE FOR INCIDENTAL, INDIRECT, SPECIAL OR CONSEQUENTIAL DAMAGES.

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