

Alkaline Phosphatase Assay Kit (Colorimetric) ab83369

[273 References](#) [8 Images](#)

Overview

Product name	Alkaline Phosphatase Assay Kit (Colorimetric)
Detection method	Colorimetric
Sample type	Urine, Serum, Plasma, Other biological fluids, Tissue Extracts, Cell Lysate, Cell culture media
Assay type	Enzyme activity (quantitative)
Sensitivity	> 10 μ U
Range	10 μ U - 250 μ U
Assay time	1h 10m
Species reactivity	Reacts with: Mammals, Other species
Product overview	Alkaline Phosphatase Assay Kit (Colorimetric) ab83369 is a highly sensitive, simple, direct and HTS-ready colorimetric assay designed to measure alkaline phosphatase (ALP) activity in serum and other mammalian samples.

The kit is a pNPP assay; it uses p-nitrophenyl phosphate (pNPP) as a phosphatase substrate which turns yellow (λ_{\max} = 405 nm) when dephosphorylated by ALP.

Alkaline phosphatase assay protocol summary:

- add samples and standards to wells
- add pNPP solution to sample wells (not to standards)
- add ALP enzyme solution to standard wells (not to samples)
- incubate for 60 min at room temp
- add stop solution
- analyze with microplate reader

Notes This product is manufactured by BioVision, an Abcam company and was previously called K412 Alkaline Phosphatase Activity Colorimetric Assay Kit. K412-500 is the same size as the 500 test size of ab83369.

This kit contains 10 substrate tablets providing convenience for multiple usages.

pNPP Assays

pNPP assays are a class of phosphatase assays using p-nitrophenyl phosphate (pNPP) as a phosphatase substrate. They can be used to measure alkaline phosphatase, neutral phosphatase, and acid phosphatase activity. These different types of phosphatases are active respectively in alkaline, neutral, and acid assay buffers. pNPP assays include Alkaline Phosphatase Assay Kit ab83369 and **Acid Phosphatase Assay Kit ab83367**.

Platform

Microplate reader

Properties

Storage instructions

Store at -20°C. Please refer to protocols.

Components	500 tests	10000 tests
ALP Assay Buffer I	1 x 100ml	20 x 100ml
Enzyme Mix II	1 vial	20 vials
pNPP Substrate	1 vial	20 vials
Stop Solution I	1 x 10ml	20 x 10ml

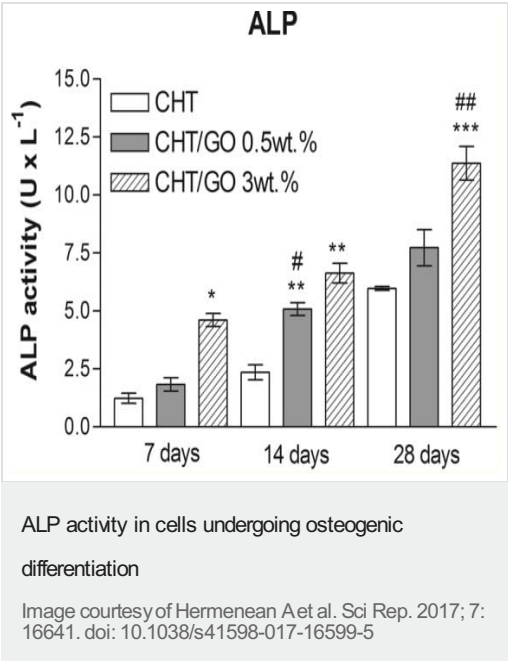
Relevance

Alkaline phosphatase (ALP) catalyzes the hydrolysis of phosphate esters in alkaline buffer and produces an organic radical and inorganic phosphate. Changes in alkaline phosphatase level and activity are associated with various disease states in the liver and bone.

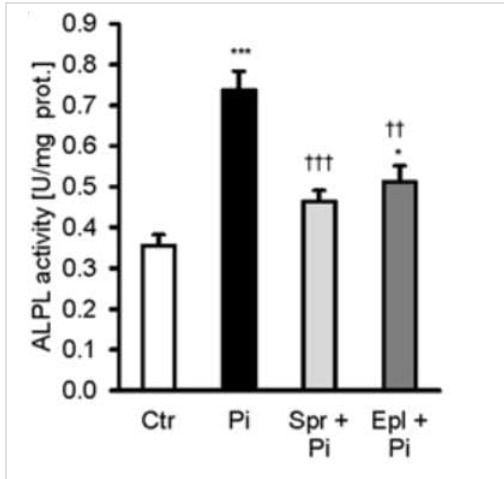
Cellular localization

Cell membrane; Lipid-anchor, GPI-anchor.

Images



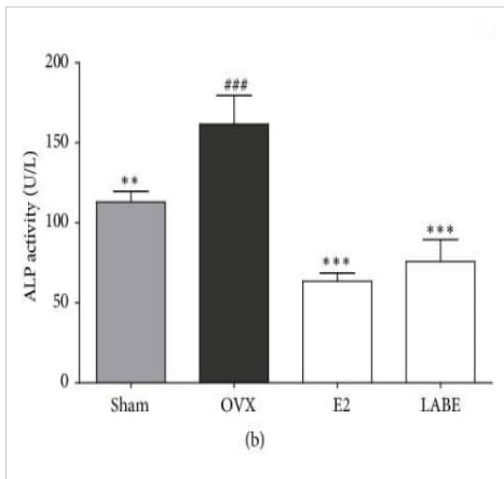
Hermenean A et al. (2017) measured alkaline phosphatase activity in cells cultured in CHT/GO (3D chitosan scaffolds enriched with graphene oxide) biomaterials for up to 28 days using ab83369.



Effects of mineralocorticoid receptor blockade on phosphate-induced vascular calcification in vitro

Image courtesy of Alesutan I et al. Sci Rep. 2017; 7: 2059. doi: 10.1038/s41598-017-01882-2. Reproduced under the creative commons license <http://creativecommons.org/licenses/by/4.0/>.

Alesutan I et al. (2017) measured alkaline phosphatase activity in vitro in primary human aortic smooth muscle cells using ab83369. Cells were treated with phosphate (pi) or without (Ctr, and with or without additional treatment of spironolactone (Spr) or 10 μ M eplerenone (Epl).

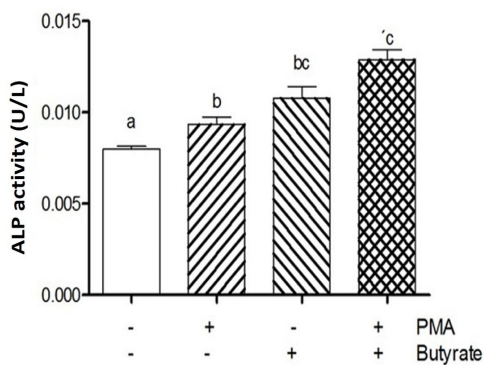


Alkaline phosphatase activity measured in the study of osteoporosis

Image from Lee YM et al., Biomed Res Int. 2019 Feb 19;2019:5073085. Fig 6b doi: 10.1155/2019/5073085. Reproduced under the Creative Commons license <http://creativecommons.org/licenses/by/4.0/>

Lee YM et al. (2019) used alkaline phosphatase assay kit ab83369 to measure the serum levels of alkaline phosphatase as a marker of bone turnover. Rat groups include sham-operated (Sham), OVX-control (OVX), E2-treated OVX groups (E2), and LBE 500 mg/kg treated OVX groups (LBE) (N=10/group).

Serum ALP levels in the OVX group were significantly increased compared to those in Sham group, and oral administration of LBE (500 mg/kg) significantly reduced the serum ALP levels as an osteoporosis-related serum marker.



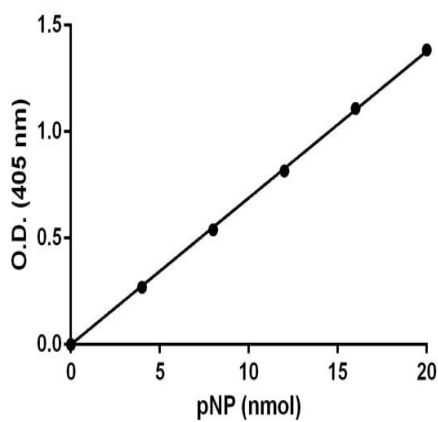
Nepelska M et al., PLoS One 7:e52869 (2012).

Functional Studies - Alkaline Phosphatase

Colorimetric Assay Kit (ab83369)

Image from Nepelska Met al., PLoS One 7(12), fig 8c.
 Doi: 10.1371/journal.pone.0052869. Reproduced under
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Nepelska M et al. (2012) assayed alkaline phosphatase activity in Caco-2 cells after 48 hours treatment with PMA and Butyrate using Alkaline Phosphatase assay kit (ab83369).

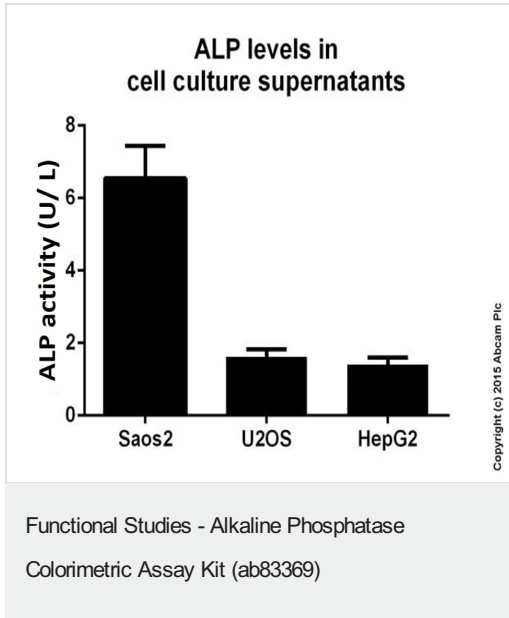


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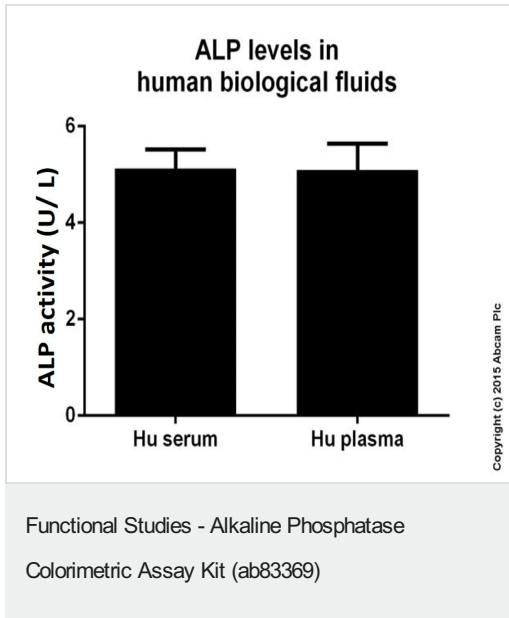
Functional Studies - Alkaline Phosphatase

Colorimetric Assay Kit (ab83369)

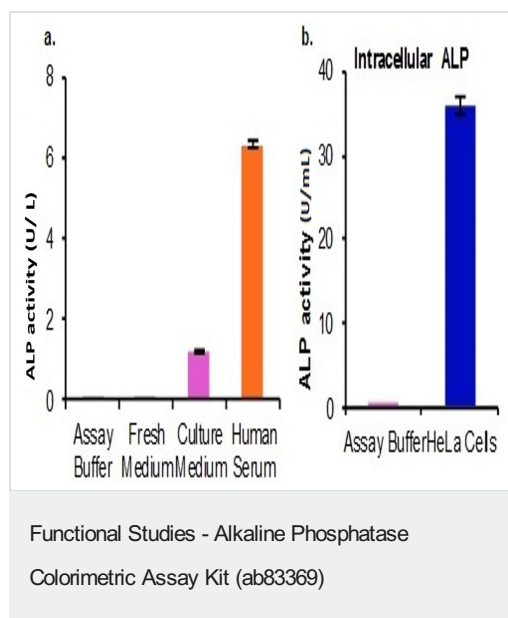
Standard curve: mean of duplicates (+/- SD) with background reads subtracted



ALP measured in cell culture supernatants showing activity (U) per L of tested sample. Samples were diluted 1-16 fold.



ALP measured in biological fluids showing activity (U) per L of tested sample. Samples were diluted 4-64 fold.



Measurement of ALP activity using ab83369. a. ALP activity in fresh medium (80 μ L, without culturing), 3 day old HeLa cell culture medium (80 μ L) and human serum (80 μ L, 1/10 dilution). b. ALP activity in HeLa cells. 5×10^4 HeLa cells were homogenized in 1 mL of Assay Buffer, diluted 1/10 in Assay Buffer, and 80 μ L used for the measurement.

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