



May 16,  
2019

Working

## Yale - Alkaline Phosphatase

Gary Cline<sup>1</sup>, John Stack<sup>1</sup>

<sup>1</sup>Yale University

dx.doi.org/10.17504/protocols.io.yz7fx9n

Mouse Metabolic Phenotyping Centers  
Tech. support email: [info@mmpc.org](mailto:info@mmpc.org)

Lili Liang

### ABSTRACT

#### Summary:

Procedure to measure the amount of Alkaline Phosphatase activity. Alkaline Phosphatase (ALP) activity is measured from the hydrolysis of 4-nitrophenylphosphate to 4-nitrophenoxide ion (monitored at 405 nm) and phosphate.

### EXTERNAL LINK

<https://mmpc.org/shared/document.aspx?id=208&docType=Protocol>

### MATERIALS

NAME	CATALOG #	VENDOR
Alkaline Phosphatase Reagent	R85120	Prolabs(cliniqa)
Assayed Control Serum 1	R83082	Prolabs(cliniqa)
Assayed Control Serum 2	R83083	Prolabs(cliniqa)

### MATERIALS TEXT

#### Reagent Preparation:

**Alkaline Phosphatase Reagent:** Add the appropriate amount of water (6.5mL) to the reagent bottle. Invert to mix, allowing 15 minutes for the reagent to settle.

**Assayed Control Serum 1:** Add the appropriate amount of water (6.5mL) to the chemical control bottle. Invert to mix, allowing 15 minutes for the reagent to settle.

**Assayed Control Serum 2:** Add the appropriate amount of water (6.5mL) to the chemical control bottle. Invert to mix, allowing 15 minutes for the reagent to settle.

### BEFORE STARTING

Analysis by automated system Cobas Mira Plus.

- 1 Calibrate Cobas for Alkaline Phosphatase Activity analysis by running two assayed control serum.
- 2 Sample handling as performed by the Cobas Mira Plus.
  - a) Pipette 3  $\mu$ L of sample into a cuvette slot.
  - b) Add 150  $\mu$ L of Alkaline Phosphatase Reagent.
  - c) Mixture is incubated at 37°C and spun for 10 minutes.
  - d) Absorbance is measured at 405 nm.



This is an open access protocol distributed under the terms of the [Creative Commons Attribution License](https://creativecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided the original author and source are credited