



## Alkaline Phosphatase (AP) Detection Kit (Ready-to-Use) - Blue

**Catalog Number: ST01001**

**Size: 50 tests**

Alkaline Phosphatase (AP) Detection Kit is used to detect alkaline phosphatase activity in all types of pluripotent stem cells.

### Product Description

Stemmera™ Alkaline Phosphatase Detection Kit is used to detect alkaline phosphatase activity in all types of pluripotent stem cells including embryonic stem cells, embryonic germ cells and induced pluripotent stem cells.

Unlike other immunostaining methods, which require specific equipment, Alkaline Phosphatase staining is the fastest and simplest method in characterizing the pluripotency of stem cells *in vitro*. Stemmera™ Alkaline Phosphatase Detection Kit (Ready-to-Use) - Blue provides an efficient system in distinguishing between undifferentiated and differentiated stem cells by staining undifferentiated stem cells.

### Product Component

Component	Size	Storage	Cat #
Pre-wash Buffer	30 ml	2-8°C	ST01001-S1
Fix Solution	30 ml	2-8°C	ST01001-S2
Wash Buffer I	30 ml	2-8°C	ST01001-S3
Wash Buffer II	30 ml	2-8°C	ST01001-S4
Staining Solution I	12 ml	2-8°C	ST01001-S5
Staining Solution II	12 ml	2-8°C	ST01001-S6
Staining Solution III	12 ml	2-8°C	ST01001-S7
Stop Solution (optional)	30 ml	2-8°C	ST01001-S8

### Storage and Handling

Stemmera™ Alkaline Phosphatase Detection Kit (Ready-to-Use) - Blue is shipped with blue ice pack. Store all components of the kit at 2-8°C upon arrival and until the expiration dates indicated on the product label. Do not freeze. Protect from light for Staining Solution II and Staining Solution III. These reagents should remain stable for at least 6 months when stored as directed.

### Additional Materials Required But Not Supplied

- 15 ml conical tubes
- Aluminum Foil
- Light microscope

### Product Use

This product is intended for *in vitro* and research use only. Not intended for human or animal diagnostic or therapeutic uses.

### Protocol

#### Preparation of substrate solution

For one well of a 6-well plate, add 500 µl of Staining Solution I and 500 µl of Staining Solution II in a 15 ml conical tube. Mix well and incubate at room temperature for 1-2 minutes. Add 500 µl Staining Solution III into the mixture and mix well. Use substrate solution within an hour after preparation.

*Note: As the Staining Solutions II and III are light sensitive, mixture should be prepared in either 15 ml amber conical tubes or 15 ml conical tubes wrapped with foil. Prepare sufficient fresh Alkaline Phosphatase Substrate Solution for the experiment only. Discard remaining mixture solutions.*

#### AP staining for cultured cells

1. Aspirate the culture medium and wash the cells with 1 ml of Pre-wash Buffer.
2. Aspirate Pre-wash Buffer and fix cells with 1 ml of Fix Solution at room temperature for 2 minutes.
3. Aspirate the Fix Solution and wash cells with 1 ml of Wash Buffer I.
4. Aspirate Wash Buffer I, add 1 ml of Wash Buffer II and incubate at room temperature for 5 minutes.
5. Aspirate Wash Buffer II, add 1.5 ml of freshly prepared AP substrate solution and incubate the cells in the dark at room temperature for 30-60 minutes.

*Note: Wrap plate with foil or place plate in dark room or dark container.*

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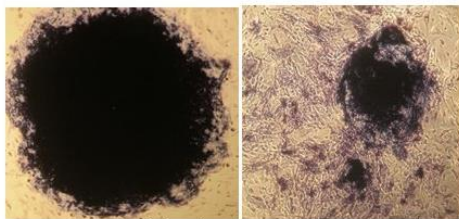
6. Stop the reaction by gently aspirating the substrate solution, rinse the cells once with 1 ml of Stop Solution.

*Note: Rinsing of cells with Stop Solution is optional.*

7. Add 1 ml of Stop Solution into well, observe and count the Blue stained cell colonies (undifferentiated stem cells) versus colorless colonies (differentiated stem cells) using a microscope and take the pictures.
8. Store the plate at 2-8°C or discard the plate after taking the pictures.

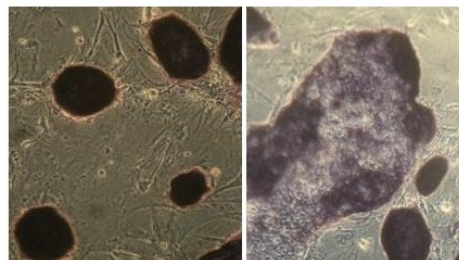
### Quality Control

This product is used to detect the pluripotency in both human and mouse ESCs/iPSCs, as well as other pluripotent stem cells. To ensure the quality, the following images are representative results of staining for human and mouse ESCs/iPSCs detection.



Positive staining in human ES cell H9

Negative staining in differentiated H9



Positive staining in mouse ESC colonies

Negative staining in differentiated mouse ESC colony

### Warranty and Limited Liability

Stemmera™ will not be liable for any damage caused by misuse, improper handling and storage of the product, non-compliance with precautions and procedures, and damages caused by events occurring after the product is released.

### References

James A. Thomson\*, Joseph Itskovitz-Eldor, Sander S. Shapiro, Michelle A. Waknitz, Jennifer J. Swiergiel, Vivienne S. Marshall, Jeffrey M. Jones. Embryonic Stem Cell Lines Derived from Human Blastocysts. Science 6 November 1998: Vol. 282 no. 5391 pp. 1145-1147.

Chunli Zhao, Ruqiang Yao, Jie Hao, etc.

Establishment of customized mouse stem cell lines by sequential nuclear transfer. Cell Research (2007) 17: 80-87.

### Related Products

Product	Cat #	Size
Alkaline Phosphatase Detection Kit (Ready-to-Use) - Red	ST01002	50 tests
Human ESCs/iPSCs Serum-/Feeder-Free Medium (hStemSFM)	ST02001	500 ml
Human ESCs/iPSCs Xeno-Free Medium (hStemXFM)	ST02002	500 ml
Non-Enzymatic Cell Dissociation Solution (1X)	ST03001	100 ml
Serum-Free Cryopreservation Solution for Human ESCs/iPSCs	ST03002	50 ml

### Technical Support

Table 1 Suggested Volume per Well

Culture Vessel	24-well plate	12-well plate	6-well plate
Pre-Wash Buffer	0.5 ml	1 ml	1 ml
Fix Solution	0.5 ml	0.5 ml	1 ml
Wash Buffer I	0.5 ml	1 ml	1 ml
Wash Buffer II	0.5 ml	1 ml	1 ml
AP Staining Solution	0.5 ml	1 ml	1.5 ml
Stop Solution	0.5-1 ml	0.5-1 ml	1-2 ml
Reactions per Kit	30-72	30-60	15-30

For more product and technical information, please visit our website at [www.stemmera.com](http://www.stemmera.com).

For further assistance, email your inquiries to our Technical Support team at [techsupport@stemmera.com](mailto:techsupport@stemmera.com).

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