TransDetect® Alkaline Phosphatase Live Detection Kit

Please read the instructions carefully before use

Cat. No. MA101

Storage: Store at -20℃ in a dark place. Avoid repeated freezing and thawing.

Description

Alkaline phosphatase (AP) is highly expressed in pluripotent stem cells, including embryonic stem cells (ESCs) and induced pluripotent stem cells (iPSCs). In the iPSCs induction process, the activation of AP is also necessary for successful reprogramming. In traditional AP detection methods, cells are fixed and cannot be used for further studies. This kit provides a fast, simple and sensitive method for AP detection without fixing the cells. The reagent is nontoxic to the cells, and the fluorescent signal will be discharged from the cells by exocytosis completely within two hours after removal of the staining solution, allowing the cell being used for following studies.

Kit Contents

Component	MA101-01	MA101-02
AP Live Solution (500×)	20 µl	50 µl

Procedures

The amount of reagents used in this protocol is based on a 24-well plate, for other plates and dishes, please make the corresponding adjustments.

- Remove the growth medium from the cultures.
- Wash with 500 µl of 37℃ DMEM/F12 for 3 minutes , repeat two times. (DMEM/F12 should be pre heated to 37℃ before use)
- Dilute the 500×AP Live Solution with DMEM/F12 to make a 1×working solution.
- Add 200 µl of working solution directly to the cells and incubate in a 37℃ CO_2 incubator for 30 minutes.
- Remove the working solution. Wash with 500 µl DMEM/F12 for 3 minutes, repeat two times.
- Add 500 µl DMEM/F12 to the cells and observe the results under a fluorescence microscope. Cells express AP are GFP positive and cells do not express AP are GFP negative ones.

Notes

- Do not use pluripotent stem cell culture medium to dilute the 500×AP Live Solution. Do not observe the result in pluripotent stem cell culture medium.
- Since AP is ubiquitously expressed in most cell types and highest in pluripotent stem cells. It is normal that dim staining can be observed in some somatic cells such as CHO cells, HeLa cells and cells from the placenta, and so on.
- After removal of dye from the media, fluorescently labeled cells lose their signal by exocytosis in 1.5-2 hours, please observe the results within 1 hour after staining.
- Please avoid repeated freezing and thawing. When necessary, the working concentration can be increased.

For research use only, not for clinical diagnosis.

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