



Prussian Blue Staining Kit for Iron Oxide Magnetic Nanoparticles on Cells/Tissues

(Catalog # IPS)

The Prussian Blue staining kit (Catalog # IPS) is designed for qualitative detection of iron oxide magnetic nanoparticles (IOMNPs) on cells and tissues. The kit detects both specific and non-specific uptake and/or adsorption of IOMNPs. Prussian blue staining to detect IONPs is easy to use and the signals can be visualized under a microscope or with a UV-vis spectrophotometer at 680 nm. The staining process is based on the chelation of iron by ferrocyanide, forming $KFe(Fe[CN]_6)$ which is blue in color.

PLEASE READ THE ENTIRE PROTOCOL BEFORE STARTING.

Scope: Prussian blue staining to detect both specific and non-specific uptake and/or adsorption of IOMNPs on cells and tissues

Cell preparation

Prepare the cells following your protocol. Expose the cells to the IOMNPs following your protocol. Adherent cells can be directly used without further treatment. For suspended cells, centrifuge at 3000 to 5000 rpm, collect the pellet and re-suspend in 20 uL of solution C. Suspended cells may be placed in a cuvette or in a 96-well plate depending upon the absorption reader. (The recommended volumes in the protocol need to be properly adjusted depending on the volume required by the absorption reader). If using a microscope for inspection, the IOMNP treated cells must be smeared on a microscope glass slide.

To detect cell uptake, rupture the cell membrane using your fixing solution that you are accustomed to in your laboratory. Use the fixed cells for the Prussian blue staining.

Tissue preparation

Prepare the tissue following your protocol. (The tissue may be taken from an animal treated/injected with the IOMNPs). Homogenize or macerate the tissue with a blade that does not contain iron to avoid contamination from the blade. The tissue homogenate may be placed in a cuvette or in a 96-well plate depending upon the absorption reader. (The recommended volumes in the protocol need to be properly adjusted depending on the volume required by the absorption reader). If using a microscope for inspection, the homogenized/macerated tissue must be smeared on a microscope glass slide.

Contents of the staining kit

- Staining solution A (SSA): yellow salt solution
- Staining solution B (SSB): aqueous acid solution



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- Solution C (SC): storage buffer
- Solution D (SD): Must be freshly prepared 1:1 mixture of SSA and SSB.

Staining Procedure

1. Combine 25 uL each of SSA and SSB in a tube to form SD. Mix well.
2. Put all of SD on the cells or prepared tissue.
3. Incubate at 37^oC for 20 min in a water bath.
5. Wash away excess SD with SC two times.
6. For adherent or smeared cells/tissue, inspect under a microscope.
8. For cells in suspension, pellet the cells at 300-5000 rpm for 5 min. Wash two times with SC.
Re-suspend in SC and smear on a glass slide for inspection under a microscope. For absorption measurement, place in a cuvette or a 96 well plate and scan.

Storage: The staining kit can be stored at RT for 1 year.

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