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NB400-134 Protocol

Immunocytochemistry/Immunofluorescence protocol for SR-BI/SR-BII Antibody (NB400-134)

[[URL:https://www.novusbio.com/products/sr-bi-sr-bii-antibody_nb400-134]][[Caption:SR-BI/SR-BII Antibody]] Immunocytochemistry Protocol

Culture cells to appropriate density on suitable glass coverslips in 35 mm culture dishes or 6-well plates.

- 1. Remove culture medium and add 10% formalin to the dish. Fix at room temperature for 5-10 minutes.
- 2. Remove the formalin and add 0.5% Triton-X 100 in TBS to permeabilize the cells. Incubate for 5-10 minutes.
- 3. Remove the permeabilization buffer and add wash buffer (i.e. PBS or PBS with 0.1% Tween-20). Be sure to not let the specimen dry out. Gently wash three times for 10 minutes.
- 4. Alternatively, cells can be fixed with -20C methanol for 10 min at room temperature. Remove the methanol and rehydrate in PBS for 10 min before proceeding.
- 5. To block nonspecific antibody binding incubate in 10% normal goat serum for 1 hour at room temperature.
- 6. Add primary antibody at appropriate dilution and incubate at room temperature for 1 hour or at 4 degrees C overnight.
- 7. Remove primary antibody and replace with wash buffer. Gently wash three times for 10 minutes.
- 8. Add secondary antibody at the appropriate dilution. Incubate for 1 hour at room temperature.
- 9. Remove antibody and replace with wash buffer. Gently wash three times for 10 minutes.
- 10. Nuclei can be staining with 4',6' diamino phenylindole (DAPI) at 0.1 ug/ml, or coverslips can be directly mounted in media containing DAPI.
- 11. Cells can now be viewed with a fluorescence microscope.

*The above information is only intended as a guide. The researcher should determine what protocol best meets their needs. Please follow proper laboratory procedures for the disposal of formalin.