BLOCKING CLASS A SCAVENGER RECEPTORS

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Updated: June 2012
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BACKGROUND

- This protocol details how to block class A scavenger receptors (CASR) in order to determine whether processes such as binding and phagocytosis is scavenger receptor mediated.

EQUIPMENT

- Dextran sulphate, Sigma #D6001
- Chondroitan Sulfate, Sigma #C9819
- Fucoidan, Sigma #F563
- Polyinosinic acid, Sigma #P4154
- Polycytidylic acid, Sigma #P4903
- Macrophages
- Complete growth media (I.e. X-Vivo for monocyte derived macrophages)

PROTOCOL

1) Prepare Class A Scavenger Receptor blockers and their respective negative controls at the following final concentrations:
    - Dextran Sulfate (+) and Chondroitan Sulfate (-), 10 to 500 µg/ml (100 µg/ml optimal for monocyte-derived macrophages).
    - Polyinosinic (+) and Polycytidylic acid (-), 10 to 500µg/ml (50 µg/ml optimal for monocyte-derived macrophages).
    - Fucoidan (no negative control), (-), 10 to 500 µg/ml (100 µg/ml optimal for monocyte-derived macrophages).

2) Incubate cells with blockers or negative controls for 30 mins at 37°C in complete media prior to stimulations or infections.

3) It is not necessary to wash off blockers prior to stimulation, however it may be warranted depending on the particular research question

LINKS AND REFERENCES

- www.bowdish.ca/lab/protocols