SA Scientific™

SAS™ Strep*Alert* FAQ

1. **What are the most common causes of False Negatives?**
   - Specimen antigen concentrations below the minimal detectable threshold of the SAS™ Strep*Alert* test
   - Inadequate specimen collection.
   - Improper specimen handling or transport
   - Specimens transported in unqualified transport media

   A negative test result does not rule out the presence of Strep A. The results from the SAS™ Strep*Alert* test should be used in conjunction with other clinical findings to establish diagnosis.

2. **What are the common causes of Faint Lines?**
   - The addition of too much sample or buffers
   - Sample is near the cutoff value
   - Sample has a matrix affect

3. **I put the test strip in the sample tube and its not running.**
   The test may not be running because a) there is not enough liquid or b) the strip was not sufficiently stirred after it was inserted in the sample tube (as directed in the PI). To ensure there is enough liquid in the sample tube, squeeze the swab thoroughly with the tube.

4. **Will the SAS™ Strep*Alert* Test detect other viruses (i.e. Influenza, RSV, etc.) that are associated with respiratory infections?**
   No, the SAS™ Strep*Alert* Test is not intended for confirmation of a respiratory infection caused by etiological agents other than Strep A.

5. **The Strep A EIA I am using is positive for Strep A and the SAS™ Strep*Alert* test is negative, which result is correct?**
   - It is recommended that all negative SAS™ Strep*Alert* results be confirmed by cell culture or equivalent method. Cell culture is the currect Gold Standard for Strep A detection.
   - A direct comparison of an EIA and SAS™ Strep*Alert* test cannot be made due to following:
     - EIA uses washing to remove non-specific binding antigens, longer incubation times and enzyme kinetics. Enzyme kinetics enhances detection of low-level antigen concentrations.
     - SAS™ Strep*Alert* test is a particle based assay. In low-level antigen concentrations, insufficient antibody antigen sandwich binding may occur making a line undetectable by the human eye.

6. **The PI recommends that all negatives should be confirmed by cell culture. We do not currently use any additional means to confirm negatives; can I still use the test?**
   This is a recommendation that is given by the FDA. Each lab should follow its own established guidelines to determine if negatives should be confirmed by cell culture.

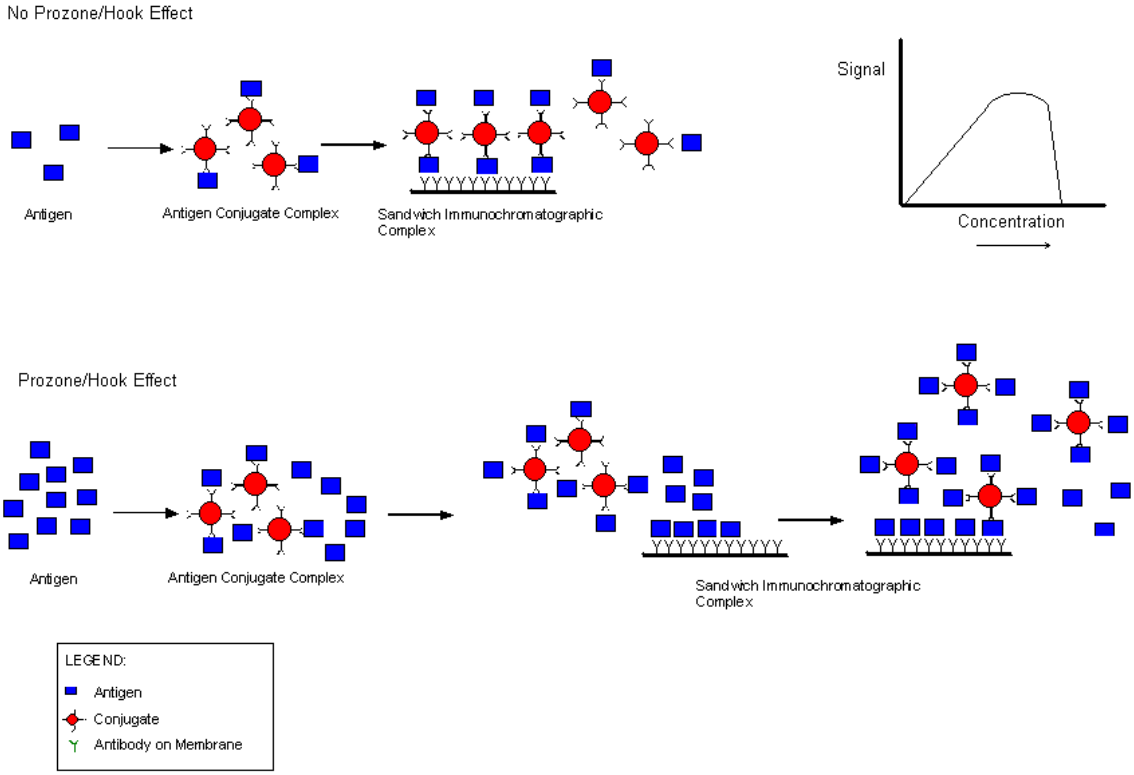
7. **What is the Prozone or Hook Effect?**
   Prozone or Hook Effect is the condition by which large quantities of antigen in an immunoassay system impair antigen-antibody binding, resulting in low antigen determination.

Prozone/Hook Effect FAQ

1. **What is a Prozone/Hook Effect?**

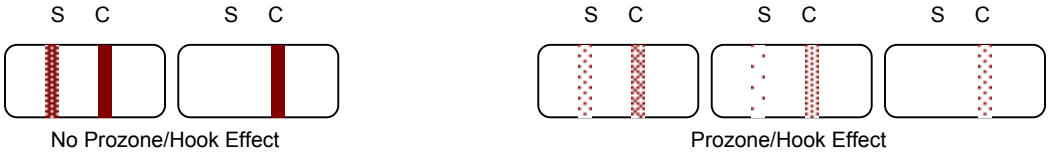
   Prozone/Hook Effect occurs when excess antigen binds the antibody at the test line, therefore not allowing the binding of the conjugate-antigen colored complex to the antibody on the test line (See Drawing Below). Light specimen and control lines characterize an assay having a Prozone/Hook Effect.

This phenomenon is mainly found in immunoassays in which three components (antigen, conjugate, and capture antibody) are incubated together. Dilution of the antigen sample with saline or PBS will enhance the signal intensity.



2. How do you recognize a Prozone/Hook Effect in an immunochromatographic assay?

Prozone/Hook Effect is characterized by either a light or non-existent test (specimen) line and a light control line. See illustration below.



3. Can you correct for Hook Effect?

Yes, simply dilute the sample 1:2 or 1:4 with PBS or Saline, and perform the assay once again.

4. Are references available to read more on Prozone/Hook Effect?

Yes, please see list below.

1. Butch AW. [Letter]. Clin Chem 2000: 1720-1721.
2. Dahlmann N et al. "Hook Effect" In a Patient with a Gonadotropin-Secreting Tumor [Letter]. Clin Chem 1990; 36: 168.
3. Diamandis EP and Christopoulos TK. Immunassay. New York, pp. 230-231
4. Ermens AM, et al. Dilution Protocols for Detection of Hook Effects/Prozone Phenomenon [Letter]. Clin Chem 2000; 46: 1719-1720.
5. Hussa RO. The Clinical Marker hCG. West Port, Conn. , 1987.
6. Wu JT, Christensen SE. Effect of different test designs of immunoassays on "hook effect" of CA 19-9 measurement. J. Clin Lab Anal 1191; 5:228-32.