

Evaluation of Sigma Transwab® in Liquid Amies Transport Medium for *Neisseria gonorrhoeae* Culture

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Introduction

The use of selective culture agar for near patient inoculation of samples for the detection of *Neisseria gonorrhoeae* has been the 'gold standard' for diagnosis and treatment of gonorrhoea for many years. This method can produce high sensitivity and specificity with the additional benefit of the provision of antimicrobial susceptibility antibiogram allowing for early detection of antibiotic resistance. However, the organism's fastidious nature makes it intolerant of delays in transportation to laboratory facilities where the culture plates can be incubated under carefully controlled conditions (37°C at 5%CO₂). Any delay in processing, transport, and incubation can significantly reduce the sensitivity of the test resulting in false negative results.

Aim

Amies transport medium would result in comparable recovery of *Neisseria gonorrhoeae* when compared with traditional near patient inoculation of specimens to Vancomycin, Colistin sulphate, Amphotericin B, Trimethoprim (VCAT) agar culture plates.

Materials

- MW177S Mini tip swab, orange capped, for urethral sampling.
- MW176S Standard tipped swab, purple capped, for cervical, rectal, throat and eye sampling.
- VCAT agar culture plate

The Sigma Transwab® have a capture mechanism whereby the swab is placed in the conical based tube containing 1ml of liquid Amies transport medium, and snapped at its breakpoint. The cap is screwed home and the swab is captured within the cap securely, allowing the swab to be removed safely for sub culture.



Method

The Sigma Transwab® was obtained in parallel with the direct inoculation/culture on VCAT plates in three Sexual health clinics over a twelve month period. Patients were informed that the trial swabs would be obtained.

Procedure for direct culture to VCAT agar plates

10µl disposable loops for urethral sampling were used to prepare a smear slide and inoculate a single VCAT culture plate.

For all other anatomical sites (cervical, throat, rectal and eye) a dry cotton swab was used to collect and inoculate a single VCAT agar culture plate.

Inoculated plates placed in gas jar with enhanced CO₂.

If direct transport to lab not available gas jars kept overnight in 37°C incubator at clinic site

Gas jars transported to laboratory.

Procedure for Sigma Transwab® sample collection

The Sigma Transwab® was obtained after all other samples for direct culture plate inoculation and NAAT testing had taken place.

On receipt in the laboratory the Sigma Transwab® were inoculated on to individual VCAT plates and streaked out to obtain single colonies.

Both the direct culture plates and the Sigma Transwab® culture plates were then incubated in parallel at 37 +/-2 °C in an atmosphere of 5% CO₂ for up to 48 hours.

Culture plates were examined at 24 & 48 hours. Bacteria were presumptively identified on the basis of colony morphology, oxidase (positive), Gram stain (Gram negative diplococci). Identification was confirmed using the Vitek MS (MALDI-ToF). Sensitivity testing was performed on all isolated confirmed to be *Neisseria gonorrhoeae*.

Results

Specimen Number	Specimen Type	Direct Culture on VCAT Agar		Sigma Transwab® Culture	
		24hrs	48hrs	24hrs	48hrs
1	Urethral	Neg	Pos	Pos	Pos
2	Throat	Neg	Pos	Pos	Pos
3	Urethral	Neg	Pos	Neg	Pos
4	Urethral	Neg	Pos	Neg	Neg
5	Urethral	Neg	Pos	Neg	Pos
6	Urethral	Neg	Pos	Neg	Neg
7	Cervical	Neg	Pos	Pos	Pos
8	Urethral	Pos	Pos	Pos	Pos
9	Rectal	Neg	Pos	Neg	Pos
10	Urethral	Neg	Neg	Neg	Pos
11	Cervical	Neg	Neg	Neg	Pos
12	Urethral	Neg	Neg	Neg	Pos
13	Swab (site not specified)	Neg	Neg	Neg	Pos
14	Urethral	Neg	Neg	Neg	Pos
15	Urethral	Neg	Neg	Neg	Pos
16	Urethral	Neg	Neg	Neg	Pos
17	Rectal	Neg	Pos	Neg	Pos
18	Rectal	Neg	Pos	Neg	Pos

Pos = *Neisseria gonorrhoeae* isolated

Neg = *Neisseria gonorrhoeae* NOT isolated

Sigma Transwab® VCAT culture

Sensitivity: 90%

Specificity: 100%

Positive Predictive value: 100%

Negative Predictive value: 98%

Direct near patient culture

Sensitivity: 72%

Specificity: 100%

Positive Predictive value: 100%

Negative Predictive value: 96%

Discussion

Confirmed *Neisseria gonorrhoeae* was isolated from 18 samples. The colonies of *Neisseria gonorrhoeae* obtained from the Sigma Transwab® plates were larger and growth was heavier. It is also worth noting that Sigma Transwab® VCAT culture produced a reduction in time to detection of positivity by 24hrs for three patient samples.

Two sample sets produced a positive result from the near patient inoculated culture VCAT plate, but a negative result on the Sigma Transwab® VCAT culture plate. Both of these samples were urethral swabs. The discrepant results may be due to sampling protocol and small sample volume (the sampling protocol was to obtain material for the microscope slide for Gram staining slide and to prepare the near patient inoculation onto VCAT media prior to obtaining the Sigma Transwab®).

Seven sample sets produced a negative result from the near patient inoculated culture VCAT plate, but a positive result on the Sigma Transwab® VCAT culture plate. The current method of direct inoculation would have produced false negative results in all seven of these cases.

Delays in incubation of direct culture plates along with use of media below optimum conditions directly contributes to failures of the current detection system. Culture plates require refrigerated storage prior to use and have a short shelf life (approximately two weeks). Sigma Transwab® stock can be stored at room temperature and has a shelf life of two years.

Conclusion

Sigma Transwab® in liquid Amies transport medium provides a superior recovery method for *Neisseria gonorrhoeae* from clinical material. This methodology will not only provide significant quality improvements but also service provision improvements for our patients.

This study has resulted in a significant change in procedure at all YorSexual health clinics. York Teaching Hospital NHS Foundation Trust will be adopting the use of Sigma Transwab® in liquid Amies transport medium to obtain the relevant anatomical samples for transportation to the Microbiology laboratory for culture and identification of *Neisseria gonorrhoeae* in July 2017.

Acknowledgments

Thank you to all of the staff at YorSexual health clinics, the Biomedical Scientist team at the York Microbiology laboratory at York Teaching Hospital NHS Foundation Trust, and Claire Hill, Clinical Product Manager Medical Wire.