

AN ASSESSMENT OF STUART'S TRANSPORT MEDIUM IN THE DIAGNOSIS OF GONORRHOEA

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SUMMARY

Genital discharge from patients with smear-positive gonorrhoea was transported from the clinic to the laboratory in Stuart's transport medium (Oxoid CM 111). Within six hours of transit time the recovery rate of gonococci was 94%. When compared with "bedside" inoculation onto Modified Thayer Martin medium, there was no significant difference in recovery rates up to 6 hours of transportation in Stuart's transport medium. However, the rate of isolation of gonococci was significantly reduced after 20 to 30 hours of transportation. It is concluded that Stuart's transport medium is an acceptable transport medium for specimens containing gonococci when specimens reach the laboratory within 6 hours of collection.

INTRODUCTION

The diagnosis of gonorrhoea can often be made by direct microscopy of Gram-stained smears alone. However the laboratory identification of the gonococcus is essential to make a medicolegal diagnosis and is often necessary for the diagnosis of asymptomatic or complicated gonorrhoea especially in the female patient for whom a smear examination is much less sensitive. Recovery of the gonococcus also enables us to monitor its antibiotic susceptibility patterns which form a basis for therapeutic recommendations.

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For the isolation of gonococci the ideal practice is direct plating of the specimen collected onto a culture medium followed by immediate incubation.¹ When this is not feasible, a holding medium must be used to preserve the viability of the organism during its transportation from the clinic where the specimen is taken to the laboratory where the specimen is inoculated onto suitable culture media. Many methods have been devised to overcome the problems of loss of viability and overgrowth by contaminants during transit.^{2,3,4} Stuart *et al*⁵ described a semi-solid, non-nutrient transport medium which has been modified by increasing its agar concentration^{6,7} and incorporating carbon particles⁸ to increase its efficacy. This medium and its modifications have been used by many laboratories in the world. The University Hospital, Kuala Lumpur has been using a modified Stuart's Transport Medium (Oxoid CM 111) for the past 5 years. This paper attempts to show the usefulness and limitations of this medium in the laboratory diagnosis of gonorrhoea.

MATERIALS AND METHODS

Endocervical or urethral specimens were collected on charcoal-impregnated cotton wool swabs from patients seen at the Kuala Lumpur Clinic, Jalan Pudu, Kuala Lumpur.

STUDY I

Recovery of gonococci after transportation in Stuart's Transport Medium (STM).

In the first study, each specimen collected was used to make a Gram-stained smear and then was broken off into a bottle of STM (Oxoid CM 111) which was transported to the STD laboratory, University Hospital, Kuala Lumpur after varying periods of time. In the laboratory, specimens were

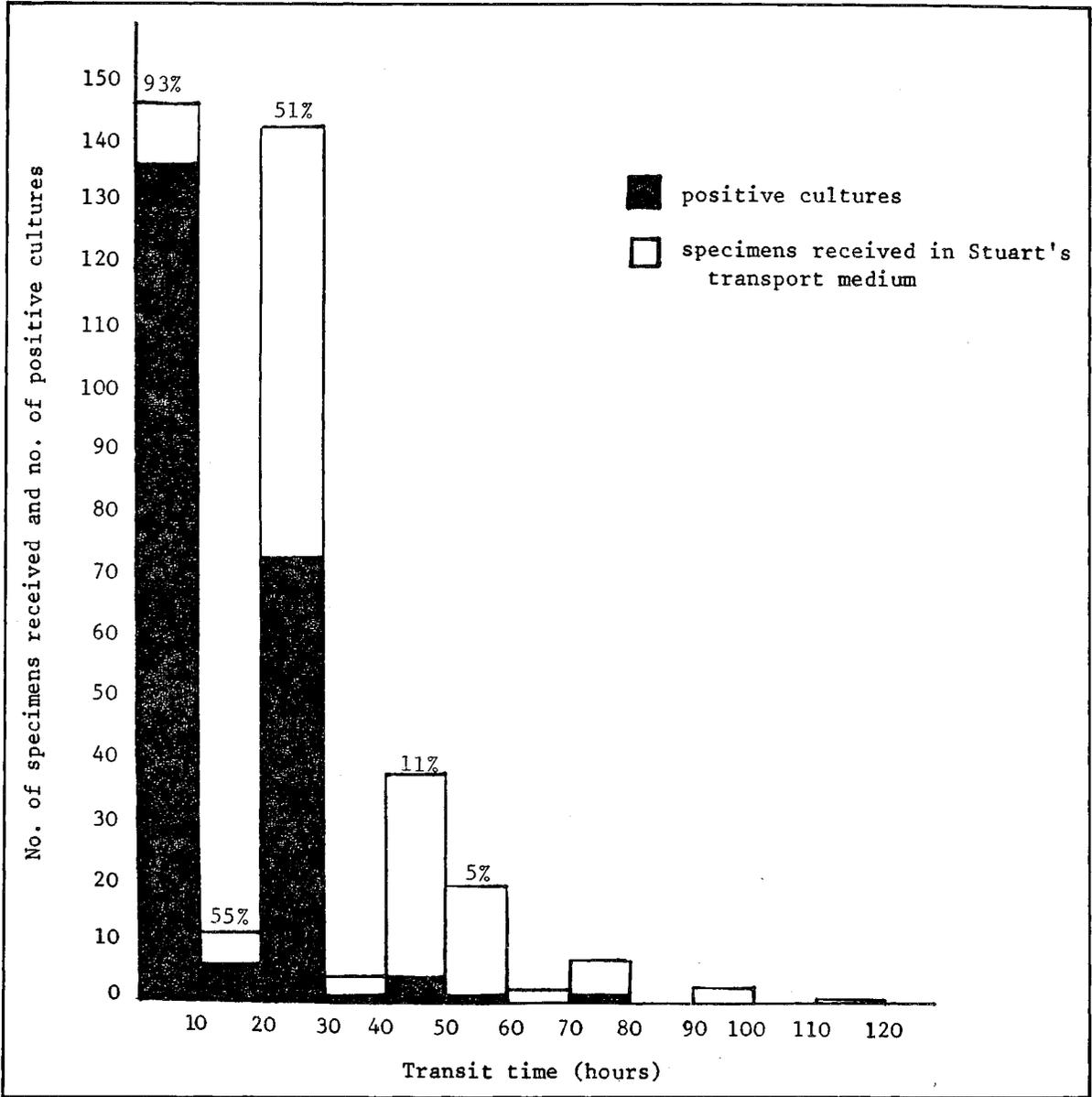


Fig. 1 Recovery of gonococci in relation to transit time.

cultured on a Modified Thayer-Martin (MTM) plate^{9,10} and a chocolate agar plate (10% ox blood in Oxoid Blood Agar Base). The plates were incubated for 2 days at 36°C in 7% CO₂ incubation. Colonies of *Neisseria gonorrhoeae* were identified by the Gram stain, oxidase reaction and fluorescent antibody staining. For each specimen, the time of specimen collection and the time when the specimen was received in the laboratory were noted. The difference between the former and the

latter was recorded as the transit time. Only specimens which were smear-positive and with known transit times were included for analysis.

STUDY 2

Comparing recovery of gonococci by immediate inoculation in the clinic ("bedside" inoculation) and after transportation in STM.

In the second study, two specimens were

TABLE I
RECOVERY OF GONOCOCCI WITHIN 6 HOURS OF
TRANSIT TIME

Transit time (hours)	No. of specimens	No. (%) of positive cultures
0 - 3	57	57 (100)
4 - 6	87	78 (98)
Total	144	135 (94)

obtained from each patient diagnosed as having gonorrhoea by a positive Gram smear. One specimen was placed in STM which was transported to the STD laboratory after varying time intervals. The other specimen was immediately inoculated onto an MTM plate which was then sent to the laboratory in a candle extinction jar. The isolation and identification of *N. gonorrhoeae* from these specimens were carried as described above.

RESULTS

STUDY 1

A total of 371 smear-positive specimens with recorded transit times were available for analysis. Out of this, 221 were culture positive and 150 were culture negative giving an apparently very low overall diagnostic yield of 60%. However, when the figures were broken down by transit time, it became obvious that loss of viability was due to prolonged transportation (Fig. 1). Within 10 hours of transportation 136 out of 146 (93 percent) specimens received yielded gonococci. After 20 to 30 hours in STM, only half of the specimens received were still culture positive. Thereafter 66 of the remaining 73 specimens received (90 percent) did not contain viable gonococci.

Table I shows that there was little loss of viability up to 6 hours in STM. A total of 144 specimens yielded 135 positives (94 percent). Moreover, all specimens received within 3 hours were positive.

STUDY 2

A total of 45 pairs of specimens were received out of which 43 (96 percent) positive cultures were obtained by "bedside" inoculation onto MTM agar while only 37 (82 percent) positive cultures were obtained after transportation in STM (Table II). As in study 1, loss of viability in STM increased with transit time and again there was 100 percent recovery of gonococci within 3 hours of

TABLE II
RATES OF RECOVERY OF GONOCOCCI AFTER
"BEDSIDE" INOCULATION COMPARED WITH
THAT AFTER TRANSPORTATION IN STM

Transit time (hours)	Total no. of specimens	No. (%) of isolates after "bedside" inoculation	No. (%) of isolates after transportation
0 - 3	6	6 (100)	6 (100)
4 - 6	20	19 (95)	19 (95)
7 - 19	1	1	1
20 - 30	13	13 (100)	9 (69)
30 - 96	5	4 (80)	2 (40)
Total	45	43 (96)	37 (82)

transportation. The rate of recovery after 4 to 6 hours transportation was even better at 95 percent. Two specimens which were culture-negative after "bedside" inoculation were positive after transportation in STM, one after 4 hours of transportation and the other after 48 hours.

DISCUSSION

The final identification of pathogens depends very much on the condition of the specimens received by the laboratory. Hence, it is important that suitable specimens carefully collected by clinicians should reach the laboratory, containing viable organisms and free from gross contamination. For general practitioners whose clinics are far from laboratories the transportation of specimens to the laboratory can be a real problem especially when the pathogen to be identified is a fastidious bacteria like *N. gonorrhoea*.

In Peninsular Malaysia the use of commercial transport-culture systems like the Jembec/Neigon plates has not been popular owing to various reasons such as cost, supply and storage problems. Stuart's transport medium being cheap and easily prepared in the laboratory should be a suitable medium for distribution to peripheral hospitals or clinics from a central laboratory with facilities for gonococcal culture. However, several field studies on the efficacy of this medium for the recovery of gonococci have shown varying results. Stuart *et al*⁵ showed that it was well over 90 percent effective under a transport time of 24 hours. Danielsson and Johannisson (1973)¹ using Ringertz's modified STM obtained a diagnostic yield of 95 percent in males and 97 percent in females after 18-20 hours'

transportation. In contrast, Hosty *et al*¹¹ had a 44 percent loss of viability in STM held for 24 hours before culturing. Taylor and Phillips¹² compared the holding ability of 3 semi-solid transport media using gonococcal suspensions prepared in the laboratory. They found that with a modification of STM containing more agar (Oxoid R 22), 41 percent of cultures kept at 4°C were lost after 24 hours and the corresponding failure rate for cultures kept at room temperature was 79 percent.

Our first study showed a very good recovery rate (94 percent) for gonococci transported in STM when the transit time is \leq 6 hours. The second study showed no difference in the recovery rates after "bedside" inoculation onto MTM agar and after \leq 6 hours of transportation in STM. In both studies, all specimens received in the laboratory within 3 hours of collection contained viable gonococci; after 20 to 30 hours transportation, recovery rates dropped to 51 to 69 percent and after 30 hours there was a rapid loss of viability, although on a few occasions, it was still possible to recover the organism after 72 hours in STM. It was shown in a separate laboratory experiment that an inoculum of 10^4 gonococci/ml survived 120 hours in STM kept at 4°C after inoculation. These results suggest that STM can be a useful gonococcal transport medium for our clinicians as in most instances it is possible to transport specimens from the clinic to the laboratory in less than 6 hours. When delay is unavoidable, specimens in STM should be kept refrigerated at 4°C until they are transported to the laboratory.

REFERENCES

- ¹ Danielsson D & Johannisson G (1973) Culture diagnosis of gonorrhoea. A comparison of the yield with selective and non-selective gonococcal culture media inoculated in the clinic and after transport of specimens. *Acta Dermatovenar (Stockholm)* 53, 75-80.
- ² Martin J E and Lester B A (1971) Transgrow, a medium for transport and growth of *Neisseria gonorrhoeae* and *Neisseria meningitidis*. HSMHA Health Reports, 86, 30-33.
- ³ Jephcott A E, Bhattacharyya M N and Jackson D H (1976) Improved transport and culture system for the rapid diagnosis of gonorrhoea. *Br. J. Vener. Dis.*, 52, 250-252.
- ⁴ Willcox R R and John J (1976) Simplified method for the cultural diagnosis of gonorrhoea. *Br. J. Vener. Dis.*, 52, 256.
- ⁵ Stuart R D, Toshack S R and Patsula T M (1954) The problem of transport of specimens for culture of gonococci. *Can. J. Publ. Health*, 45, 73-83.
- ⁶ Ringertz O (1960) A modified Stuart medium for the transport of gonococcal specimens. *Acta Pathol. Microbiol. Scand.* 48, 105-112.
- ⁷ Gastrin B and Kallings L O (1968) Improved methods for gonococcal sampling and examination on a large scale. *Acta Pathol. Microbiol. Scand.* 74, 362-370.
- ⁸ Amies C R (1967) A modified formula for the preparation of Stuart's transport medium. *Can. J. Publ. Health*, 58, 296-300.
- ⁹ Thayer J D and Martin J E (1964) A selective medium of the cultivation of *Neisseria gonorrhoeae* and *Neisseria meningitidis*. *Public Health Rep.* 79, 49-57.
- ¹⁰ Seth A D (1970) Use of trimethoprim to prevent overgrowth by *Proteus* in the cultivation of *N. gonorrhoeae*. *Br. J. Vener. Dis.*, 46, 201-202.
- ¹¹ Hosty T S, Freear M A, Baker C and Holston J (1974) Comparison of Transportation Media for the culturing of *N. gonorrhoeae*. *Am. J. Clin. Path.*, 62, 435-437.
- ¹² Taylor E and Phillips I (1980) Assessment of transport and isolation methods for gonococci. *Br. J. Vener. Dis.*, 56, 390-393.