

A COMPARATIVE STUDY OF ENRICHED CULTURE MEDIA AND SELECTIVE CULTURE MEDIA IN THE DIAGNOSIS OF GONORRHOEA

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The Laboratory aids the Clinician in his diagnostic work. A diagnosis of gonorrhoea can only be made with certainty by the isolation of *N. gonorrhoeae*. Moreover, cultural methods yield a higher proportion of positive results especially in chronic cases (Chacko and Nair,^{1,2} Eldering and Palser,³ Juhlin and Crook,⁴ and Scott and Stome,⁵.) In female patients due to chronicity of infection, significantly higher number of positive results can be obtained by culture alone than by smear examination and higher still, if both methods are employed (Platts-Personal communication,⁶ and Chacko and Nair,²)

The difficulties of the techniques of culture are reflected in the variety of new media and modifications of old media which have been recommended and used from time to time. Consequently there has been constant search for media which could easily be prepared and would give consistent results and raise the standard of cultural work outside certain highly specialized laboratories. The medium which has been most widely used and has stood the test of time is chocolate agar medium devised by McLeod et al,⁷. McLeod's⁸ results suggested that blood added to the medium might protect from toxic substance present in the medium for the

gonococcus, heated blood seemed to give better results than unheated blood. Chacko and Nair⁶ from the Institute of Venereology, Madras has introduced a Trypsin digest beef extract agar base medium which has been enriched with the contents of hen's egg. This medium can be prepared conveniently in the Laboratory and is found to be effective in the primary cultural isolation and diagnosis of gonorrhoea (Chacko and Nair,^{1,2}).

Isolation of gonococci from infected secretions is often made difficult because of over growth by other organisms like Staphylococci, Diphtheroids and Coliform bacilli (Amies and Garabedian,⁹ Chacko and Nair,² Martin et al,¹⁰ Roepstorff and Hammarstron¹¹, Scott and Stone,⁵ Thayer and Martin,^{12,13} and Wilkinson,¹⁴.) A further considerable advance was made by Thayer and Martin^{12,13} at the Venereal Diseases Research Laboratory of the U.S. Public Health Service, Atlanta, Georgia, who incorporated antibiotics, Polymyxin B/Colistimethate sodium to inhibit Gram-negative contaminants, Ristocetin/Vancomycin to inhibit gram-positive contaminants and Nystatin to inhibit the fungal contaminants, in the Chocolate agar base.

Material and Method

It was proposed to evaluate different culture methods including the accepted and recently introduced culture media for their scope and utility in the diagnosis of gonorrhoea in both sexes. Two

groups of culture methods namely Enriched media which includes Chocolate agar medium (McLeods et al⁷) and Chacko-Nair medium (Chacko and Nair¹) and the second group comprises of Selective culture media which are Chacko-Nair medium with antibiotics and Thayer-Martin Medium (Thayer and Martin,^{12,13}).

The antibiotics which were incorporated in the Selective media were as follows;

Ristocetin	10/ugm/ml of the medium
Colistimethate Sodium	7.5/ugm/ml of the medium
Nystatin	12.5 units/ml of the medium

A total of 125 cases suspected to be suffering from gonorrhoea were taken for this study. Out of these 100 were male and 25 female cases. Collection of pus material was made by routine method. In male cases it was obtained from urethra and in female cases two specimens were taken, one from urethra and another from the cervix. In cases of unmarried females the collection of material was done from vagina instead of cervix.

It is to be noted that in the present study, from three cases, gonococci failed to grow on the Selective media, though the growth was positive on the Enriched media.

The inoculum on all the four culture plates was done and colonies of gonococci were identified by oxidase test and culture smear examination. These results were confirmed by sugar fermentation test.

Results and Observations

Male cases: Culture was done on Enriched media as well as on Selective media from all the 100 male cases. A comparison of positive results thus obtained is given in table I.

TABLE I
Number of Positive Cultures on Different Mediae
(Series of 100 Male Patients)

Culture Mediae	Number
A. Enriched Media:	
(i) Chocolate agar medium	62
(ii) Chacko-Nair medium	71
B. Selective Media:	
(i) Thayer-Martin medium	83
(ii) Chacko-Nair medium (With antibiotics)	83

Female cases: Culture was done on Enriched media as well as Selective media from all the 25 cases. A comparison of positive results thus obtained is given in Table II.

TABLE II
Number of Positive Cultures on Different Mediae
(Series of 25 Female Patients)

Culture Mediae	Cervix or Vagina No. percent		Urethra No. percent		Total No. percent	
A. Enriched Media:						
(i) Chocolate agar Medium	7	28	1	4	7	28
(ii) Chacko-Nair Medium	9	36	2	8	9	36
B. Selective Media:						
(i) Thayer-Martin Medium	13	52	4	16	13	52
(ii) Chacko-Nair Medium	13	52	4	16	13	52

Discussion

Efficacy in detecting the gonococcus is an important factor in treating and controlling the spread of such infections; but even at this time the facilities for diagnosing and controlling the disease are not universally available. We can say that we are 'long' on these items but are 'short' on skilled laboratory facilities.

Thayer and Martin^{12, 13} and Chacko and Nair^{1, 2} described the uses of a Selective medium which allowed the growth of gonococci but suppressed the growth of the other organisms. This seems to offer a distinctive advantage in the diagnosis of gonorrhoea specially in female cases.

From male cases we have observed that only 62 to 71 percent positive cultures could be obtained on Enriched medium and 83 percent on Selective medium. This shows that by using Selective media a 12 to 21 percent more cases were confirmed. Wilkinson¹⁴ has also compared the Enriched medium with Selective medium. In his study 72 percent were positive on Enriched medium and 82 percent on Selective medium. Thus in his study there was an increase of positive results by 10 percent having used the Selective medium.

From female patients 28 to 36 percent cases yielded gonococci on Enriched media and 54 percent on the Selective media. Thus 16 to 24 percent additional cases were diagnosed by the Selective medium. We have observed that in male cases only 12 to 21 percent more cases were confirmed on Selective medium as compared to 16 to 24 percent in the female group. This shows some-

what increased efficacy of Selective medium in the diagnosis of female patients.

It is to be noted that in our series out of 99 positive cultures from 125 cases, when both the Enriched and Selective media were used 3 were positive on Enriched media only and they failed to grow on the Selective media. It may be due to as Reyn¹⁵ has attributed it to the marked sensitivity of these strains towards the antibiotics which were used in the Selective media.

It can be concluded that Selective procedures to inhibit the growth of contaminants on media for the primary isolation of *N. gonorrhoeae* have proved successful. However, it should be borne in mind that strains extremely sensitive to the antibiotics may be encountered. When the Selective medium is used alone, infection with very susceptible strains may remain undiagnosed.

Summary

A total of 125 suspected cases of gonorrhoea were taken for the study. Out of these 100 were male cases and 25 female cases. Cultures were done on Chocolate agar and Chacko-Nair enriched media as well as on Thayer-Martin medium and Chacko-Nair medium (with antibiotics) Selective media. Positive results were 62%, 71%, 83% and 83% respectively in male series and 28%, 36%, 52% and 52% respectively in female series. Selective media has given better results as compared to enriched media, specially in female cases. It is to be noted that 3.03% strain were encountered which were isolated on the Enriched media but failed to grow on Selective media.

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False

Brown Falco and associates in 1966 reported that abnormality in hair roots was detected in 49% of a series of patients whom he studied. Others have observed changes in hair shaft. Orfanos et al have shown hair shaft changes in the form of furrows, grooves and severe deformation of the hair surface with probably profound damage to the cortical structures.