

## Blood Cultures In Bacterial Endocarditis\*

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### Summary:

An improved base broth for culturing blood in bacterial endocarditis was compared with routine culture medium in 77 samples, taken from 26 patients of infective endocarditis. On Culture with improved broth 18 patients showed bacteriological presence as compared to 3 patients on routine culture media giving sensitivity of 69.23%. The number of abacteremic patients were 30.76.

### Introduction:

Diagnosis of bacterial endocarditis is confirmed by the isolation of causative agent. However negative blood cultures are quiet common because majority of the blood cultures media fail to support growth of most microorganisms. Because of the high incidence of negative blood cultures an effort was made to evolve an improved base broth and to determine the number of positive and negative blood cultures. The different causes that lead to negative blood cultures such as magnitude of bacteremia, high immunity of the body, delay due to fastidious requirements, anti-microbial therapy and prolongation of lag phase were considered.

### Material & Method:

Three 10. ml blood samples were collected<sup>2,3,4,5</sup> from each patient at an interval of at least 2 hours over a period of 24-48 hours. Blood was inoculated into blood cultures bottles containing base broth and incubated at 37°C<sup>6,7</sup>. Brain Heart Infusion broth was used in which the following ingredients were added per 1000 c.c. of media. Bacto yeast extract 5 gms, PABA 0.05 g<sup>8</sup>, Sucrose 300 g<sup>9</sup>, Thioglycollic acid 3x10<sup>5</sup>M<sup>10</sup>, L-cysteine 10<sup>4</sup>M<sup>11</sup>. Hundred ml of this broth was poured into each blood culture bottle to form a layer of about 8cm deep<sup>12,13</sup>. PABA was used to counter act the effect of sulphonamides. Sucrose

was used as an osmotic stabilizer<sup>9,13,14</sup>. Thioglycollic acid and L-cysteine were used as reducing agents. Hundred ml of the broth was poured into each blood culture bottle to form a layer about 8cm deep.

### BASE BROTH

#### Formula:

Brain Heart Infusion agar in which the following ingredients were added.

Bacto yeast extract	5 gms.
PABA <sup>7</sup>	0.05 gms.
Sucrose <sup>8</sup>	300 gms.
Thioglycollic acid <sup>9</sup>	3x10 <sup>-5</sup> M.
i.e., (0.0272ml) L-cysteine	10 <sup>-4</sup> M. i.e., (1.2 gms.)

All these were diluted with 1000cc of distilled water.

Due to the low order of magnitude of bacteremia the dilution of blood to base broth was 1:10.

### Results:

The age range of patients under study was from 7-41 years. There were 15 females as compared to 11 males the F:M ratio being 1.3:1. Majority of the patients, i.e., 42.3% were in the second decade of life.

The total number of blood specimens drawn from these 26 patients was 77. On these blood specimens

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intermittent culture results were seen. The first and third blood culture gave 22.64% positive results, whereas the second culture gave 9.43% yield only. (Table I).

TABLE I

Percentage of positive and negative specimens cultured in bacteriologically proven cases of infective endocarditis

Total Number of blood cultures = 53.				
Cultures	Positive	%	Sterile	%
First blood specimen	12	22.64	6	11.32
Second blood specimen	5	9.43	13	24.52
Third blood specimen	12	22.64	5	9.4
Total:	29	54.71	24	47.16

In the 18 patients with positive results 13 patients were infected with gamma hemolytic *Streptococcus viridans* (72.22%). *Staphylococcus aureus* was isolated in 4 patients (22.22%) and *Streptococcus faecalis* in 5.55% (Table II). The number of abacteremic patients were 30.76%. Total number of positive blood cultures were 29. Multiple positive cultures were present in 11 patients. When comparing parallel studies on trypticase soya broth the present study showed significant improvement in recovery rate ( $P < 0.01$ ).

#### Discussion:

Different media broths have been used in blood culture. In the present study brain heart infusion broth was used as this supports larger proportion of microorganisms. The addition of reducing agents further facilitates growth from a smaller inoculum. Antimicrobial therapy also affects the results of blood cultures. This was overcome by the simple dilution of blood 1:10 with the base medium. This dilution

renders the concentration of antimicrobials too low to affect bacterial growths. This dilution of 1:10 also dilutes out the antimicrobial powers of the blood and the bacteria has a fair chance of surviving.

The Osmotic shock encountered by the bacteremia in the blood culture broth causes a prolongation of the lag phase. This was prevented by the use of an osmotic stabilizer. It has been stated that a blood culture report cannot be stated as negative unless the blood was incubated in a hypertonic media. Thus addition of sucrose facilitates better yield than ordinary blood culture media.

100cc of broth formed a layer 8cm deep which is capable of supporting facultative anaerobic bacteria also. Due to this change in the formula for blood cultures there were less number of abacteremic cases, i.e., 30.76%, which is comparable to a study done by Cooper et al and less than that in another series. In comparison with parallel studies using trypticase soya broth alone the present results showed significant improvement in recovery rates ( $P < 0.01$ ).

TABLE II

Comparison of present series and other series

	Total No. of cases	No. of +ve cases	%
Present series	26	18	69.23%
Other series on Trypticase soya broth	26	3	11.5%

$P < 0.01$

Sensitivity of the present series: 69.23%

Lerner and Weistein<sup>18</sup> have found that among the factors that may be responsible for failure to recover organisms from the blood of some patients with infective endocarditis are the use of antibiotics in etiologically undefined febrile illness, right sided endocarditis, prolonged duration of subacute bacterial endocarditis and errors in technique of obtaining and culturing blood. Even in patients with endocarditis, however, blood cultures are usually

sterile by the 3rd or 4th day of treatment. This high incidence of bacteriologically negative bacterial endocarditis in countries with easily available, excellent and reliable laboratory facilities should alert us to the alarmingly high incidence of bacterial endocarditis with negative blood culture in Pakistan where many adverse factors dictate against bacterial growth in vitro.

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