

OPTIMUM GROWTH TEMPERATURE FOR THE ISOLATION OF *PLESIOMONAS SHIGELLOIDES* USING IBB, PDA AND MSS AGARS

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A good satisfactory growth medium for *Plesiomonas shigelloides* is not known as yet. Their identification from a culture plate has always been a problem in presence of other organisms particularly when *Aeromonas* spp. are present. To overcome this problem we investigated various media, such as inositol brilliant green bile salt agar (IBB), *Plesiomonas* differential agar (PDA) and modified Salmonella and Shigella (MSS) agar, in attempting best recovery of these organisms through laboratory experiments. Several strains of clinical and environmental origin of *P. shigelloides* were employed in the investigation. Overnight broth cultures were prepared and plated onto the above mentioned culture media including a few other non-selective plates and incubated at 37, 42 and 44°C. Highest numbers of colony forming units were observed on PDA plates at 42°C, while large and best colonies morphologically were found on the same plates but at 44°C. At 44°C temperature on PDA plates *Aeromonas hydrophila* was found to be completely inhibited followed by *A. caviae* and then *A. sobria*, compared to the growth at 42°C. Also, we observed that colonies of *P. shigelloides* could be recognized after 24 hours of incubation at 42 and 44°C. From this study we conclude that for optimal isolation of *P. shigelloides* PDA medium is the best, and not at 37°C but at 42°C, with reduction of the incubation temperature to 24 hours from the recommended 48 hours.

THE SUICIDE PHENOMENON IN *AEROMONAS* SPECIES AS A CORRELATE TO UNDERSTANDING CONCEPTS OF VIRULENCE AND ENTEROPATHOGENICITY

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Certain strains of mesophilic aeromonads (*Aeromonas hydrophila*, *A. sobria*, *A. caviae*) when grown in broth containing 0.5% glucose undergo growth inhibition concomitant with acetate accumulation. As these strains are non-viable after 24 h, this phenomenon is termed suicide.

We investigated suicidal strains of *Aeromonas* as a correlate to understanding animal virulence and enteropathogenicity. To assess virulence, batches of 5 white mice were inoculated intraperitoneally with washed 10⁷ cells of suicidal and non-suicidal strains of *A. hydrophila* and *A. sobria*, and suicidal strains of *A. caviae*. The three non-suicidal strains of *A. sobria* tested showed lethality as early as 12 h and were uniformly fatal within 36 h post-inoculation. After 36 h the 3 suicidal strains killed only 1 of 15 mice inoculated. Four *A. hydrophila* strains tested which show the suicide phenomenon at 37°C were variably lethal (40-100%). None of 5 suicidal strains of *A. caviae* were lethal. Enteropathogenicity was studied by orally inoculating 3 white mice each with the same *Aeromonas* strains (10⁸ in skim milk), and assessing diarrhoea and intestinal fluid accumulation. Diarrhoea and fluid accumulation were present in all mice inoculated with two non-suicidal *A. sobria*, and in 2/12 mice given four suicidal *A. hydrophila*. Two suicidal strains each of *A. sobria* and *A. caviae* failed to elicit any gastrointestinal disturbances. These data suggest that the suicide phenomenon may explain strain-specific (*A. sobria*, *A. hydrophila*) and species-specific (*A. caviae*) virulence and enteropathogenicity.