THE POLYMERASE CHAIN REACTION (PCR) TECHNIQUE FOR THE SPECIFIC DETECTION OF *CHLAMYDIA* SPP. AND ITS APPLICATION TO DIAGNOSIS AND EPIDEMIOLOGICAL STUDIES

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The polymerase Chain Reaction (PCR) technique was applied to the diagnosis and epidemiological study of Chlamydiosis.

The results were summarized as follows.

1) Two sets of primers that annealed to the upstream or downstream region of the chlamydial major outer membrane protein gene (ompl) were prepared. Each primer set was used for the amplification of DNA from *Chlamydia psittaci* or *Chlamydia trachomatis*.

2) PCR Products were recognized as single bands of about 1.2 kilo base pairs on agarose gel stained with ethidium bromide. By theoretical calculation, a single copy of chlamydial DNA was detected.

3) The specific bands were detected from all chlamydial strains tested. No signal was detected when bacterial or Hela cell DNA were used as templates.

4) A chlamydia-specific fragment was detected from the rectal contents of chicks infected with *C. psittaci* strain IZAWA–1. The absence of a specific band was noted from the PCR products of the non-infected group.

5) Out of 7 samples of conjunctiva from patients with conjunctivitis positive for antigen with Immuno-fluorescence assay, 6 were positive upon PCR. Out of 7 samples of conjunctiva negative for antigen, 1 was positive upon PCR. The samples from persons who had a history of trachoma, but now recovered, all were negative for both antigen and upon PCR. Out of 3 samples from children with pneumonia, 1 was positive upon PCR.

6) For cloning of ompl, PCR products derived from chlamydial strains or clinical specimens were ligated into an *E. coli* plasmid vector followed by restriction endonuclease analysis and DNA sequencing. The restriction sites of EcoRI and SacI were present in *C. psittaci* mammalian strains but not in the avian strains. The comparison of the sequence of approximately 70 bases around Variable Domain (VD) II with other strains revealed that amino acid homologies were 75 %–100 % in the constant region and 20–90 % in the VDII.