SUMMARY
I. VIROLOGICAL STUDIES OF MYOCARDITIS/PERICARDITIS IN ADULTS AND CHILDREN.

A comprehensive virological study of 50 cases of myocarditis/pericarditis in adults and children was carried out to find the incidence and role of Coxsackie B1 - B6, Coxsackie A9, ECHO 9, Influenza A and B, mumps and cytomegalovirus more especially Coxsackie B viruses.

The criteria used for establishing association of virus to myocarditis/pericarditis were (a) isolation of virus from the patient with or without four fold rise in antibody titre; (b) demonstration of a similar increase in virus-neutralising, haemagglutination-inhibition or complement-fixing antibody titres and (c) persistent presence of a high antibody titre in all sera collected from the patient.

Twenty-nine (58%) among 50 cases investigated in this study satisfy one or the other of the above criteria and were considered as virologically "positive". It was found that 28 cases had a high "threshold of positivity" according to the criteria of Crist and Bell (1974). In addition, a case of pericarditis in which Coxsackie B1 was isolated without serological support was also considered as "positive" since Lerner and Wilson (1973) include them as of "low order association".
Among the 50 cases of cardiac disease studied, 19 (38%) were associated with one or other of Coxsackie B virus infection and this figure is comparable to several other reports from different parts of the world. Virus types associated with these 19 cases were Coxsackie B4 in 7 cases; Coxsackie B2 in 4; Coxsackie B3 and B6 in 3 cases each and one case each of Coxsackie B1 and B5 virus infections. Thus, the largest group of cardiac illnesses were attributable to Coxsackie B virus infection. Other enterovirus associated with cardiac diseases were Coxsackie A9 and ECHO 9 virus infections in one and two cases respectively.

Apart from the enteroviruses mentioned above, influenza virus infection was associated with 5 cases (10%) among 50 cases of cardiac illnesses studied and they included three cases of myocarditis and two of rheumatic carditis.

Cytomegalovirus infection was found in two cases of myocarditis and since both showed elevated titres of ASLO, this virus was considered to have caused opportunistic secondary infection, with primary disease process being the due to streptococcal infection. Among 14 cases of rheumatic carditis investigated, 7 (50%) were associated with one or other virus infection used in this study. Five cases associated with virus infections showed persistently low titres of ASLO ruling out streptococcal infection. Two cases of rheumatic carditis, both associated with Coxsackie B4 virus infection, persistantly showed elevated titres of ASLO indicating superadded viral infection over streptococcal disease process.
II. CELL-MEDIATED IMMUNITY AGAINST COXSACKIE B3 VIRUS IN GUINEA-PIGS.

Guinea-pigs were immunised with a single intradermal inoculation of $10^2$ TCID$_{50}$ dose of Coxsackie B3 virus along with Freund's adjuvant. The animals developed high cell-mediated immunity (CMI) against the virus as indicated by the presence of delayed hypersensitivity (DH), increase in macrophage migration inhibition (MMI), development of macrophage aggregation (MA) and increased virucidal activity of peritoneal macrophages of immunised animals. Neutralising-antibody titres also developed to titres varying from 1:32 to 1:256.

Under our experimental conditions, CMI lasted in these animals for 12 weeks and was associated with the presence of live virus in the cardiac tissue as had been shown by the isolation of the virus from cardiac tissue monolayer cell cultures upto 12 weeks. It seems likely that the presence of live virus in cardiac tissue is needed for persistence of CMI, since it has been observed that after 16 weeks, CMI is not detectable and virus also could not be isolated from cardiac tissue monolayer cell cultures.

CMI against Coxsackie B3 could be passively transferred to normal guinea-pigs by transfer of immune spleen cells from actively immunised animals. Such passively immunised animals have been shown to possess CMI against the virus on 5th and 7th day after immune spleen cell transfer as indicated by increase of MMI% and high virucidal activity of peritoneal
macrophages specifically activated by heat killed virus antigen. But neutralising-antibody titre against the virus was less than 1:8 in almost all spleen cell recipient animals.

It should be mentioned here that donor spleen cells did not carry live Coxsackie B3 virus and it was shown that the virus was not isolated from MEKC tube cultures in which donor spleen cells were inoculated. Recipient guineapigs, therefore, did not receive any virus from donor guineapig adoptive transfer of spleen cells.

When such passively immunised guinea-pigs were challenged with $10^5$ TCID$_{50}$ dose of virus, they showed viraemia only for 24 hours as against its presence upto 5 days in normal animals challenged with similar dose. Subcutaneous inoculation of $10^5$ TCID$_{50}$ dose of virus produced death in 70% guineapigs between 13 and 27 days after challenge with virus, with the development of myocarditis as shown by typical histological lesions in the cardiac tissue, whereas all animals passively immunised with spleen cells, challenged with similar dose survived beyond 90 days.

It was also found that peritoneal macrophages of both actively and passively immunised guinea-pigs showed significantly high virucidal activity as compared with that of normal animals.

Therefore, it was felt that increased CMI against Coxsackie B3 virus may have a role in the final elimination of the virus from the cardiac tissue even beyond 16 weeks and also prevents establishment of the virus in passively immunised guinea-pigs.