THE VIRAL FLORA OF VILLAGE CHILDREN FROM BIRTH TO TWO AND A HALF YEARS OF AGE[@]

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A child born into a crowded room in a Calcutta bustee or in a mud hut in the nearby village of Singur, or in the totally rural economically deprived village of Jhalda (West Bengal) is exposed to a veritable host of viruses carried by his siblings from the day he is born. ⁽¹⁾, ⁽²⁾, ⁽³⁾ (Table 1)

- (1) How many of these viruses establish infection and what part of the new born child's system is affected ?
- (2) How many persist and for how long ?
- (3) How many cause disease, and do they interact with resident or introduced bacteria ?

These were some of the primary questions posed in a study of thirty Singur village children who were examined continuously from birth to the age of two and a half years. While the area of study (Singur) which surrounds a bazaar street has increasingly assumed the character of a suburb of Greater Calcutta, it maintains many traditional rural activities such as farming and village industries. The houses and the village are somewhat different from their equivalents in rural or suburban Kathmandu, for most of the houses are one-storey

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[@] This is a preliminary description of the results of a study which extended from 1966-69. It was the joint effort of M. Cooper-Dastur, A.B. Choudhuri, A.K. Dey, T.K. Dey, A.K. Ghosh, W.D. Hillis, W. Kloene, J. Thomas and F.B. Bang.

thatched dwellings built of mud; they may be scattered through the village proper or clustered along well-travelled paths radiating from the town. Nearly all are close to a natural pond, or to a "tank", a pond formed when earth is dug in order to make the mud for building the dwellings. Tube wells are present throughout the village, so that either relatively clean tube well water or contaminated pond water is always available. Otherwise the similarities with villages near Kathmandu are great. Contact with a variety of animals is common, sewage disposal is inadequate and crowding together is the way of life. The results obtained in our study in Singur may therefore have some general interest here.

Briefly, the study was initiated by arranging with the prospective mother through Dr. Joyce Biswas* to follow the child at weekly intervals after birth. Fetal cord serum was collected for future immunologic studies. Throat and rectal swabs for virological and bacteriological studies were taken at birth and at weekly intervals subsequently. At the same time the child was examined by a resident pediatrician several times a week. Growth measurements were made at one month intervals throughout the study, and illness was promptly treated whenever indicated. Complete records were kept and were later transferred to cards for statistical evaluation.

Viruses were isolated by the inoculation of washings from separate oropharyngeal and rectal swabs onto tissue cultures. Two kinds of cells were used: monkey kidney primary cultures, and Hep-2 (a human epithelial cell line), prepared and maintained at the laboratory at the S. S. K. M. Hospital, Calcutta. As a routine procedure the cultures were inoculated within one or two hours after the swabs were obtained. Using these methods it was possible to identify a host of viruses, but the method is not suitable for the isolation of measles, cytomegalic viruses, and various rhinoviruses since these agents do not produce destruction of these cells. Thus, while it was not possible to test for all of the viruses that are of significance in human disease, commonly prevalent agents which are liable such as parainfluenza 1, 2 and 3 and R.S. virus (4) were recovered in good numbers. We believe that this was related to the prompt inoculation of tissue cultures with the throat swabs, a procedure that can be readily done, if the cultures are brought to the field, instead of waiting for some hours before inoculation at a central laboratory.

The data have had only preliminary analysis. After the remaining ten percent of the viruses are identified and specific symptoms have been correlated with particular agents final analysis will be possible.

There are three major points that we wish to emphasize in this paper. The first is that the major load of viruses imposed by the community is promptly reflected in high rates of infection in the infants, starting within a few weeks of birth and rising to 20% for respiratory and 50% for enteric infection at 3 months. The second is that there is a relatively poor correlation between infection and individual illness, and the third is that persistent infection

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in individual children occurs not only with adenoviruses and herpes virus, but with several different enteroviruses as well.

Figure 1

Figure 1 shows the rate of virus recovery from throat and rectum charted as to the child at the time of virus recovery and the general class of virus recovered. However since the

thirty children entered the study over a period of one year (21 July 1967 to 11 June 1968). The individual children were exposed to somewhat different viral flora over the year, depending on seasonal variation from family to family. This may have influenced the decrease in virus recovery which appeared at about a year after birth.

Infections were detected earlier in rectal than in throat swabs, and rose to a recovery rate of 37% by six months. This was above the average of 30.9% for the whole study period excluding the first six months of life. Infection of the respiratory tract was low for the first months (about 3%) and then reached a rate of 16% by six months. The average recovery rate from 7 to 30 months of life was 13.7%. Since many of these agents had been demonstrated to be common within the community in the previous study it may be assumed that the infections occurred in the presence of passive immunity acquired from the mother. This introduces the question of active-passive immunization. Passively acquired antibody coming from the mother prevents the extension of many virus infections throughout the tissues of the child but does not prevent infection. Thus the child acquires infection when the dose of the agent is sufficiently high. But he does not usually get ill. At the same time he responds to infection by developing active immunity. This of course is a very useful biological mechanism for preparing the child for the later experience with the large number of agents described earlier. Adenoviruses seem to be able to establish infection and then persist after passive immunity wears off⁵. Herpes virus was not isolated until the child was on years of age, even though it is commonly present in the community².

In studies relating specific infection to disease, it is usually possible to show that the rate of recovery of a given pathogen from the respiratory tract of a sick child is several times greater than that from a well child. Individual children in countries such as England may yield a given suspected pathogen only at the time of illness and therefore the problem of correlation of infection and illness arises only occasionally⁶. When the frequency of infection is increased and signs of respiratory malfunction such as purulent coryza become very frequent, the demonstration of a causal association becomes much more difficult. Thus the first analysis of our results shows a correlation only between the number of weeks of illness and the number of different viruses recovered; i.e., the more illnesses a given child had, the more the agents that were recovered.

Finally, if we examine the situation from the point of view of the virus, there is a continual struggle for existence on the part of each agent. The phenomenon of interference often prevents one virus from invading a child while another one is present. Yet this is clearly an imperfect mechanism since double and sometimes triple infection were found in many of the Singur children. Another mechanism for the survival of an agent is a persistent infection which is sustained long enough to become established in another susceptible individual. It has been known for some time that this is the case with herpes virus, which may recur at various intervals after the original infection. In the present study, adenoviruses 1,2,

5 and 6 were isolated continuously among newly infected children from several weeks to as long as a year after original recovery⁵. More recent analysis of the types of enteroviruses found in the rectal swabs showed that infections with enteroviruses lasting for more than a month were present in at least 19 of the children, and that some of the children carried as many as 4 different viruses and for periods upto 14 weeks. The mechanism whereby persistent agents are able to avoid the usual responses of tissue systems and the immune system is unknown.

Summary

Much can be learned from studies of the initial responses of children from birth through early infancy to the variety of viruses to which these children are exposed in the Indian subcontinent. Many agents establish infection during the first three months of life, especially in the enteric tract, while others are delayed. However, infection during the first month is sufficiently frequent to establish the idea of active-passive immunization. Adenoviruses were not detected for the first six months of life and herpes not for the first year. Infection rates were so high that correlation between specific illness and most virus recovery was poor.

The same virus type was recovered repeatedly from the same child over periods of several months to a year. This occurred with adenoviruses, herpes, and with a variety of enteroviruses.

Table 1

Viruses Isolated from the Pharynx of Children in Singur
1964-70

Influenza A. B
Parainfluenza 1, 2, 3, 4,
Mumps
RS
Adeno 1, 2, 3, 5, 6, 7
Herpes Simplex
Polio 1, 2, 3
Cox A 7, 9
Cox B 1, 2, 3, 4, 5, 6
Echo 1, 2, 6, 7, 9, 11, 12, 14, 19, 20, 24, 25, 26
Reo
Vaccinia

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